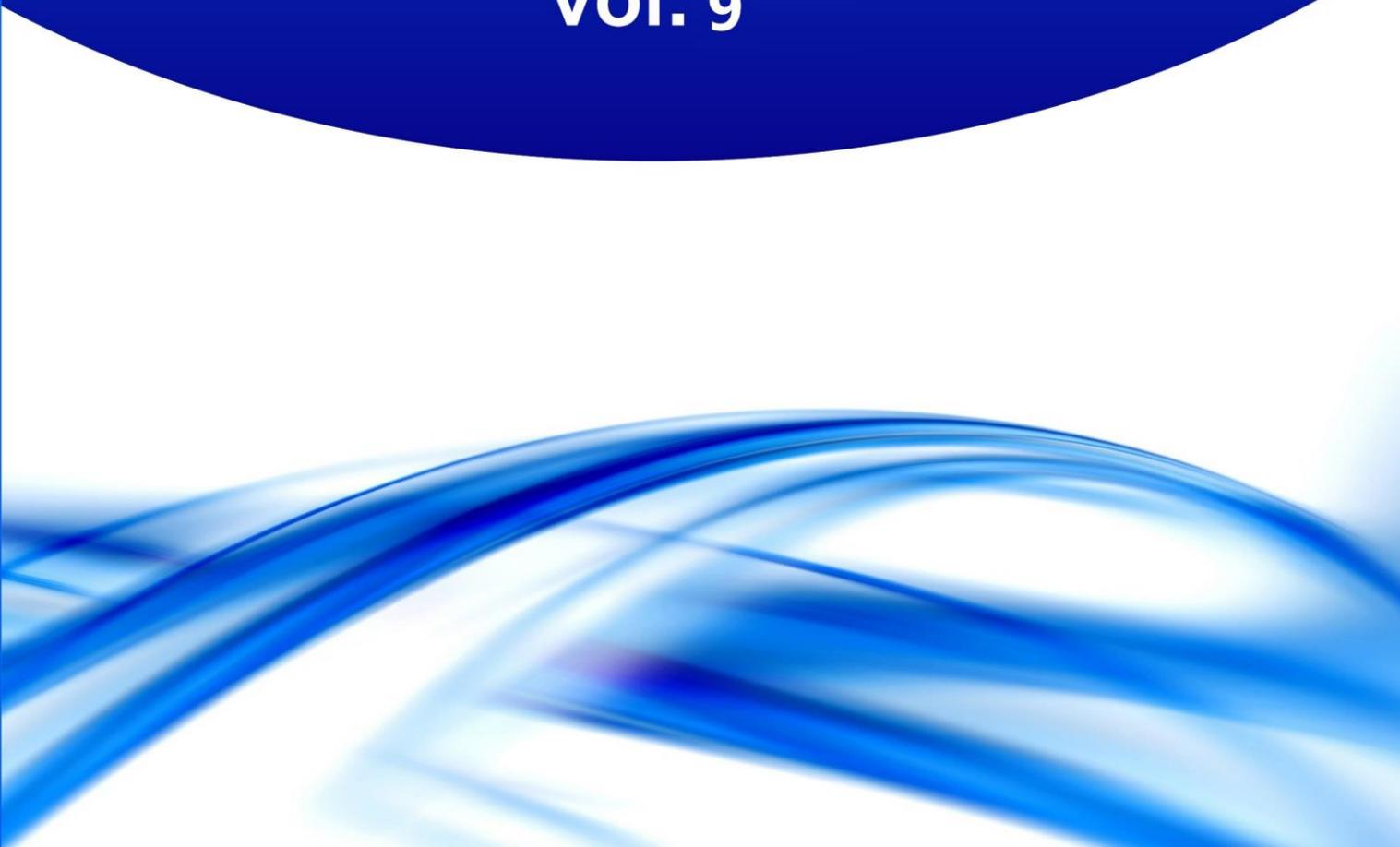


Challenges of the current medicine

Krajewska-Kułak Elżbieta, Kułak Wojciech,
Łukaszuk Cecylia, Lewko Jolanta, Sarnacka Emilia

vol. 9

The bottom half of the cover features a white background with a large, abstract graphic of flowing blue waves. The waves are composed of multiple overlapping, semi-transparent layers, creating a sense of depth and movement. The colors range from light sky blue to deep, vibrant blue. The waves curve across the bottom of the page, mirroring the curved bottom edge of the dark blue upper section.

***Challenges of the current
medicine***

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***Challenges of the
current medicine
Vol. 9***

**Edited by
Elżbieta Krajewska-Kułak
Wojciech Kułak
Cecylia Łukaszuk
Jolanta Lewko
Emilia Sarnacka**

Białystok 2020

Reviewers

Dr hab. Katarzyna Van Damme -Ostapowicz

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Førde, Norway

Ludmila Klimatckaia

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Pedagogical University named after V. P. Astafyev, Russia

Andrei Shpakou

MD, PhD Department of Theory of Physical Culture and Sport Medicine, Yanka Kupala State
University of Grodno, Grodno, Belarus

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Respect health properly, because if you die you will lose a life
Aleksander Fredro

Dear Colleagues

The monograph periodical "*Challenges of the Current Medicine - 9 Edition*" is a collection of works written by authors from many different medical centers.

The leading theme of the monograph applies to the patient. In the particular chapters are discussed various therapeutic care problems: of death and dying, of the old age, selected therapeutic and diagnostic

Health is a priceless gift. Care for one's health is an expression of human responsibility and self-respect. Unfortunately, many people underestimate the condition of their own bodies and count on the fact that various ailments will pass by themselves. Because, as Arthur Schopenhauer claims - "*Nine-tenths of our happiness is about health. A healthy beggar is happier than a sick king* "

Theoretically, health is one of the most valued by human values, but in practice many people neglect it. Losing health is not only a physical problem, but also a psychological one, because with the appearance of illness a person loses the general desire to live, and in the case of serious illnesses, he begins to feel useless.

It is worth taking care of your health and examining yourself regularly. We hope that the presented works will make everyone aware that it is worth taking care of your body because once lost health, it can never be regained again. After all, Democritus claimed that "*People beg God for health. However, no mortal thinks that health is in his own hands* ".

The monograph touched upon various aspects of medicine - it concerns health-related behaviors presented, through the presentation of cases of the use of various methods of treatment, after considering the patient's expectations of the doctor and nurse, because, as Edward Staniek wrote, "*(...) the patient needs a doctor and a nurse. Their vocations and related tasks are different. The doctor is primarily responsible for the disease and aims to cure it or alleviate the suffering that comes from it. The nurse is responsible for the patient as a person struggling with suffering. The patient stays in the doctor's hands for only minutes, because after examinations, diagnosis, and prescribing medication he passes to the next patients. The nurse stays with the patient permanently and helps him whenever possible.* "

Prof. Elżbieta Krajewska-Kułak MD, PhD

Prof. Wojciech Kułak MD, PhD

Cecylia Łukaszuk Ass. Prof. PhD

Jolanta Lewko Ass. Prof. PhD

Emilia Sarnacka PhD

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SELECTED MEDICAL PROBLEMS



Health related behaviour of modern Poles - poll among female residents of Upper Silesian conurbation

Klaudia Mazurek¹, dr n. med. Joanna Wanot²

1. Department of Aesthetic Medicine, Faculty of Pharmaceutical Sciences in Sosnowiec, Medical University of Silesia, Katowice, Poland
2. Department of Anaesthesiology and Nursing-Intensive Care, Faculty of Health Sciences in Katowice, Medical University of Silesia, Katowice, Poland

INTRODUCTION

Are women nowadays taking desirable and preferable action regarding health, or are they likely to make errors that may cause discomfort and even the development of chronic diseases? The matter which raises curiosity is about the preferred by modern women types of healthy behaviour, which may include: physical activity, rational nutrition, taking care of the good appearance and condition of the body, minimizing the stressful factors and situations, avoidance of risky sexual behaviour, lack of tobacco addiction, limiting alcohol consumption or avoidance of psychoactive substances. Some researchers also add more visits to the specialist doctors and self-examination to the long list. The presented health behaviours, the age of women, their conditions of life and work, as well as educational background and skills, and lifestyle, have an unquestionable impact on both the physical and mental health of the population [1-3].

Some scientists disagree with the above theses based on the results of studies showing that the phenomenon of deterioration in the health of women along with longer average length of life is common in all industrialized countries [4].

Detailed analyses revealed the existence of differences in the incidence of particular diseases, dysfunction and adverse mental states among women and men [3]. Women, unfortunately, are more prone for diseases with high morbidity, but also a lower mortality rate, whilst among men the situation is opposite. Thus, although women live longer, in their lives, however, there appear more diseases and disabilities which limit functionalities and significantly lower the quality of life [5]. Ignoring the aspect of the prevalence of diseases in the population of modern women, the fact of growing number of highly responsible roles

imposed on women today should also be paid attention to. Regardless of the generation they represent - women as daughters, wives, mothers or grandmothers – they work every day for their families and their loved ones, undertaking a number of tasks and solving various problems. In addition, they increasingly occupy important, decision-making positions, as well as play an increasingly important role in social life, which is reflected in the growing rate of stress. Congestion of daily duties determines the deficit of time for themselves. Women "do not have the time" for being ill, and worse health and preventive examinations are becoming second category issues [6,A].

Modern perfectionism in relation to women, created primarily by media, introduces kind of a dissonance. On the one hand every woman, regardless of age or social status, must reach the ideal of beauty at any price, which is becoming a disturbing phenomenon. On the other hand the media promote a modern, active lifestyle, in which being ill is simply not fashionable. New trends do not allow the appearance of any defects. In this case, it is perhaps the media who are the kind of stimulus to take measures to promote health and preventive examinations for women?

AIM

Main objective: Analysis of the health related behaviour of modern women.

Specific objectives:

1. Assessment of pain sensations in the female population.
2. Evaluation of the frequency of check-ups and preventive examinations performed by women.
3. Presentation of the issues associated with the consumption of alcohol and cigarette smoking among women.
4. Evaluation of the health related behaviour of modern women.

MATERIALS AND METHODS

In the present study, a diagnostic survey method was used, and the research tool used was a questionnaire voluntarily and anonymously completed by respondents. The questionnaire consisted of 9 questions, open and closed. A percentage analysis of obtained results was carried out. The survey was conducted on a group of 143 female residents of the Upper Silesian conurbation. The majority of the respondents were women 21-30 years old (83 people – 58%),

while the smallest group were women over 50 years old (6 – 4.1%). The vast majority of the respondents were residents of cities – 115 people (80.4%). Most of the respondents declared secondary education – 68 people (47.5%), while 65 women had higher education (45.4%). The majority of the respondents (89 women – 62,3%) were economically active women, unmarried (81 persons – 56.6%), and not having offspring (91 women – 63.6%).

RESULTS

Analysis of the collected research material allowed to obtain the following results: The largest group among the women surveyed were young people 21-30 years old (83 people - 58%). Despite of that, up to 105 women surveyed (73.4%) felt sensations of pain, and only 38 respondents (26.6%) did not complain about this type of dysfunction. The most common ailments, which were the most complained about by the respondents, were: headache (39%), back pain, and bone and joint pain (34.5% and 17.8%). A detailed list of the types of pain reported by women surveyed can be found in the table below (Table 1).

Table 1. Type of pain felt (N=151)

type of pain sensation	number of answers	%
headaches	59	39
back pain	52	34,5
bone and joint pain	27	17,8
stomach ache	9	5,9
other	4	2,8
total	151	100%

An extremely important issue from the point of view of broader health prevention of modern women is regular medical control, as well as performing basic laboratory tests and the necessary diagnostic imaging.

In the study group, only 26 of the women surveyed (18.3%) declared that they went to the dentist in the last six months, and 68 patients (47.6%) in total paid such a visit over the last year. More than half of the respondents – 75 (52.4%) had not visited a dentist's office in the past year.

The statistics on preventive gynaecological examination, which women undergo, are not quite satisfactory as well. Only 37 (25.9%) women surveyed from across the group paid a visit to the gynaecologist in the last six months, and in total over 72 respondents (50.3%) in the last year took this visit. The remaining part – 71 women (49.7%) did not go for the gynaecological consultation clinic in the last twelve months.

Respondents were also asked to which a medical examination (inspection) they attended in the last year. Among the women surveyed the most frequently performed examinations were basic blood and urine tests, as well as cytology (respectively 95 women – 28,9%, 63 - 19.2% and 71 women – 21.5%). Slightly fewer respondents have made an ultrasound of reproductive organs (37 people – 11.2%), as well as breast ultrasound (30 respondents – 9.1%). The results are presented in detail in the following table (Table 2).

Table 2. Kinds of prophylactic examinations performed by the respondents in the last year (N = 329 answers)

kinds of prophylactic examinations performed by the respondents in the last year	number of answers	%
blood test	95	28,9
cytology	71	21,5
urine test	63	19,2
reproductive organs ultrasound	37	11,2
breast ultrasound	30	9,1
stomach ultrasound	16	4,9
lungs roentgen	8	2,5
mammography	7	2,1
other	2	0,6
total	329	100%

Tobacco addiction has an extremely negative effect on women's health. It increases the probability of early menopause and osteoporosis, and accelerates the aging process. It also helps the atherosclerosis process, increases blood clotting and blood vessel wall tension with the ability to induce coronary vasospasm. Chronic exposure to tobacco smoke clearly influences the increase in blood pressure and fast heart rate [7-9].

The conducted study showed that tobacco addiction applies only to a small minority of the population of Polish women. Among the respondents (22.3%) 32 women admitted that they smoked. The fact that the vast majority of smokers do not smoke more than 5 cigarettes a day,

while only 5 women among the 32 smokers reaches an average of 16-20 cigarettes per day is optimistic.

Also, alcohol clearly adversely affects the human body, destroying mainly the nervous system. It interferes with precise thinking, logical reasoning, and proper judgment. It handicaps the so called higher emotionality, existing only in humans, including: ethics, morality, sense of family, interpersonal, and national ties. Alcohol also interferes with the work of the circulatory system, digestive and respiratory systems [10]. Unfortunately, as much as 106 women (74.1%) participating in the survey declared alcohol consumption, and only 37 (25.9%) - abstinence. Among consumed alcoholic beverages, ladies mentioned mainly red and white wine (49.8%), beer (28.5%), vodka (14.3%) and whiskey (4.6%). 57.5% of women declaring the consumption of alcoholic beverages admitted that they reached for it once a week on average. 10.5% declared drinkers consume alcohol even 2-4 times a week, and the remaining 32% on average once a month. It is well known that physical activity which is appropriately tailored to the age and level of mobility, is the best way to preserve the health, longevity, and above all well-being [11].

The results of the research show that the majority of respondents, as many as 102 (71.3%) women, are physically active people. The most common forms of exercise mentioned by respondents are: walking (41.8%), cycling (20.3%) and aerobics (14.6%). The respondents chose swimming and running less frequently, like roller blading or fitness classes. The exact description of the various forms of sport and physical recreation preferred by the respondents is presented in Table 3.

Table 3 Forms of physical activity preferred by the respondents. (N=177 answers)

form of physical activity	number of answers	%
walks	74	41,8
cycling	36	20,3
aerobics	26	14,6
swimming	15	8,5
running	13	7,4
rollerblading	4	2,3
fitness	3	1,7
other	6	3,4
total	177	100

Women participating in the study were also asked about the use of special diets. The majority of respondents, as many as 123 people (86.1%) declared the lack of use of any diet.

DISCUSSION

Long-term health problems, as well as pain and widely understood physical discomfort are the major determinants of chronic diseases. In Polish literature you can find many definitions of chronic disease, but regardless of the source, what distinguishes a chronic disease compared to others are primarily long duration, milder course, and essentially the irreversibility of pathological changes and the need for continuous treatment [12]. In Poland, one woman gets an average of 2.1 chronic diseases. Studies show that diseases of the chronic kind are more common for women than men. Joint, thyroid, liver diseases, hypertension are dominant for women [B].

Among the respondents participating in the survey, up to 73.4% stated that they felt pain. Most often they involve the head, spine, osteoarticular system, stomach, and teeth. The research of Ostrowska show that women significantly more likely than men declare to the existence of long-term health problems, and the pain and physical discomfort felt contributes to the reduction of physical fitness [A].

As defined by the WHO (*World Health Organization*) health is a state of complete well-being, physical, mental and social, and individual or group must be able to define and implement their aspirations, satisfy needs, as well as change the environment or deal with it. Therefore, health is seen as a resource, not a life goal. Health is a positive concept, including personal and social resources and physical abilities, and not merely the absence of objectively existing illness or disability [13,C].

Statistics show that 62.7% of women estimates their health as good or very good, and 12.2% of women as bad and very bad [A]. Whereas the Alejziak examination seems to present much more optimistic conclusions. The majority of Polish women participating in the study assessed their health as good (52%) and very good (32%) [14].

The results of the research deviate a little from the presented above and they show the female residents of the Upper Silesian agglomeration in a slightly worse light. 47.6% of respondents has visited the dentist in the last twelve months, only 21.5% of women has performed cytology, while breast diagnostic imaging as ultrasound or mammography was declared by 11.2% of respondents.

Since several years a program of free cytology for women aged 25-59 has been implemented, but only 25% of those eligible use it. According to research conducted by prof. Łuszczynska it is primarily the matter of a psychosocial barrier impact the low participation of Polish women in preventive tests. One of the reasons for this may be avoidance of talking about health with loved ones, especially in the context of cancer [D]. The survey confirmed the validity of these conclusions. A little over 20% of women surveyed take advantage of cytology, which correlates with the results of prof. Łuszczynska studies.

An exogenous factor of undeniable importance in the pathogenesis of malignant tumours, especially lung cancer, is chronic exposure to harmful components of tobacco smoke. Each year lung cancer is diagnosed in Poland in more than 6 thousand women, and the largest number of deaths induced by the malignant tumour affects women between 55 and 75 years of age. The risk of lung cancer depends mainly on intensifying the active and passive exposure to harmful components of tobacco smoke [E].

The reason for this unfavourable situation in the field of epidemiology is also the relatively late introduction and insufficient effectiveness of population-based programs of early detection of cancers in women without symptoms of the disease (i.e. screening tests). In this particular study, smoking was declared by a minority of respondents, because only 22.3% of women. Only 15.6% of the declared smokers group burns 16-20 cigarettes per day. The results correlate with the results of Kleszczewska and Jaszczuk [15], in which most of the inhabitants of the north-eastern Polish region declares that they belong to non-smokers. 59% of employed and 62% of unemployed women negated the tobacco habit.

One of the disturbing developments in recent decades is the significant reduction in the differences between men and women, both in terms of quantity and frequency of alcohol consumption. This applies to most developed countries as well as highly industrialized ones, including Poland. European statistics show that women consume about 25-30% of commercially available alcohol. The proportion of female addicts is steadily increasing and is currently 1:4 in comparison to men. It is also increasingly stressed that 4-7% of women have problems with alcohol, while the incidence of somatic complications and mortality due to alcohol consumption increases in women more intensively than the incidence of alcohol dependence. The researchers highlight the fact that alcoholism is a disease classified as fatal, and the mortality of women addicts is higher by an average of 50-100% compared to addicted men [F]. Unfortunately, the results related to alcohol consumption, which were obtained in the course of this research confirm the abovementioned statistical data, namely as much as 74.1% of women said they drank alcohol. Among consumed alcoholic beverages women enumerated

mainly red and white wine (49.8%), beer (28.5%), vodka (14.3%) and whiskey (4.6%). The frequency of consumption of alcoholic beverages is also worrisome, because as many as 57.5% of respondents consume alcohol once a week, and 10.5% even several times during the week.

Properly balanced diet combined with physical activity indisputably is a guarantee of health and well-being.

Using diet is of great importance for human health. It should be noted that only properly selected amounts of protein, carbohydrates and fats are conducive to maintaining good health. Ingredients delivered in a reasonably composed meals must provide your body with the nutrients that meet the needs resulting from the occurrence of growth and repair processes of the body. Propagated in the media image of perfect, slim and beautiful woman entails enormous interest in all sorts of diets. Interestingly, obtained in the present study results of dieting completely deny this theory, as 86.1% of women said they did not use any supplements, and only 13.9% of respondents admitted that they fall into trends propagated by media and incline toward their use.

Often discussed issue lately is broadly defined healthy lifestyle, which consists of a number of health behaviours, especially appropriately selected physical activity, coupled with a compatible nutritional program. Alejziak addresses the issue of ways of eating by fitness activities participants [14]. The majority of respondents (69%) declared that they are eating properly, 14% did not know the rules of proper nutrition, or simply does not apply to them, while the remaining 17% of women admit that they eat irrationally. The most common nutritional mistakes that the respondents shared include improper mealtimes (3%), insufficient amount of time to prepare a healthy meal, too fast pace of life (6%) and a weakness for junk food (5%). The vast majority of respondents cared about their diet and considered it as healthy and wholesome [14].

Physical activity, in turn, brings a number of benefits to health in all its dimensions, i.e. biological and psychosocial ones. The best documented correlation is properly selected exercise with frequency of cardiovascular disease. As it is clear from the latest work of the scientists exercise influences on the heart health of women more than those of men. Many previous studies highlighted the fact that regular physical activity induces favourable changes in the concentration of cholesterol in blood, which can contribute to reducing the risk of cardiovascular disease. Presented arguments now seem to take on a special meaning especially towards women [16]. In addition, physical activity has an invaluable influence for the prevention and treatment of obesity, and also reduces the risk of cancer including breast cancer in women. Under the term physical exercise lies not competitive sport, but enough daily chores

or regular walking or jogging. It should be noted and remembered at the same time that the low level of activity along with improper diet causes mainly excessive accumulation of body fat, also among women with normal body weight. According to the WHO an adult should perform weekly at least 150 minutes of exercise of moderate intensity or 75 minutes of vigorous exercise. This type of physical activity is conducive to maintaining good health and prevents the excessive accumulation of body fat with age [17]. CPOR (*Centre of Public Opinion Research*) research conducted in 2013 showed that two-thirds of Poles (66%) did sport over the last year, among them 40% undertook physical activity regularly, and 26% occasionally. By far the most popular sport is cycling (51%) and swimming (28%). Other relatively popular forms of recreation include: running (18%), hiking (16%), football (14%), volleyball (14%), gymnastics, fitness, and aerobics (13%) [G].

The conducted study applied only to women, but in comparison with the above data, the results obtained are presented very positively. Among all women surveyed, as much as 71.3% reported physical activity. As the most attractive form of exercise women acknowledged: walking (41.8%), cycling (20.3%), aerobics (14.6%) and swimming (8.5%). The fact that physical activity is conducted on a regular basis seems to be optimistic: 2-4 times a week - 52% of respondents, 5-7 times a week - 9.8%, once a week - 38.2%.

Taking care of own health goes hand in hand with taking care of your appearance. The body image which we create is an extremely important element determining our successes and failures. Attractive appearance, and more specifically the conviction of an attractive appearance, is a guarantee of success. The body is the basis for shaping own identity and it is an essential link in the process of self-realization, which, according to Goldstein, is the most important theme of human existence [18-21].

Modern women appreciate, therefore, the role of physical activity, they avoid smoking cigarettes and try to use the advice of specialist doctors. More than half of the respondents were women 21-30 years old, which indicates a high public awareness of a healthy lifestyle of the young generation. Health related behaviours are a very important element of human life activity, and knowledge of the types of behaviour and ways of their implementation or modification can largely contribute to improving and strengthening health of society.

CONCLUSIONS

Collecting and developing research material allowed to draw and present the following conclusions:

1. The majority (73.4%) of women complain about the presence of various pain.
2. About 50% of the women reported visiting the gynaecologist and dentist.
3. Among prevention tests in women carried out the most common are: blood and urine tests, cytology, reproductive organs and breasts ultrasound.
4. The majority of women consume alcohol (74.1%) at a frequency of one time per week.
5. The number of women smoking cigarettes and using different types of diets strongly decreases.
6. Most women are physically active, and the most common forms of activities are: walking, cycling, aerobics and swimming.

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Contraception in the opinion of students of the tri-city universities

Lucyna Wójcicka, Agnieszka Czerwińska-Osipiak

Department of Obstetric and Gynaecological Nursing, Faculty of Health Sciences with Institute of Maritime and Tropical Medicine, Medical University of Gdańsk, Poland

INTRODUCTION

According to the definition of the World Health Organization (WHO), reproductive health is an undisturbed state of physical, mental and social, referring to the function of the reproduction system, and as a result, people at every stage of life can lead a safe, responsible and satisfying sexual life, consistent with their individual needs and a personal decision about fertility, including access to effective and acceptable methods of fertility regulation [A].

According to the Public Opinion Research Center (polish CBOS - Centrum Badania Opinii Społecznej) surveys carried out in 1996-2012, almost all respondents in the conceptual age (from 18 to 44 years old), declared their will to have children [B]. At the same time, demographic changes within fertility are observed, such as an increase of the average age of getting married, delaying the birth of the first child, reducing the fertility of young women, increasing childlessness from choice [1]. The reasons for postponing the intentions of starting a family are directly related to changes in the family model, a greater need for education, in order to find a better job and achieve a higher social status. Following this social trend, the need to discuss the methods of planning pregnancy has become clear.

According to the recommendations developed by the Polish Gynecological Society (PTG) in 2014, contraception, being the basic element of birth control should be safe, effective, reversible, easy to use, widely available and cheap [2]. The wide range of contraceptives offered on the market makes education about available methods and forms justified and needed. Lack of knowledge in this area may lead not only to the incorrect use of contraception, but also to not using it at all. As a result psychological, social and economic consequences will increase, for example unscheduled pregnancies, or a high risk of transmission of sexually transmitted diseases (STD).

AIM

The aim of the study was to describe methods of contraception used by students of Tri-city universities, including opinions and knowledge about available methods of birth control.

MATERIALS AND METHODS

The survey was conducted from July 2014 to December 2016. The research material was made up of a group of 750 students from Tri-city universities. All respondents were adult students, and therefore they met the inclusion criteria. The work involved the method of a diagnostic survey using the own authorship tool and questions characterizing general social-demographic data. The study received permission from the Independent Bioethics Commission for Research of the Medical University of Gdańsk.

All statistical calculations were performed using the IBM SPSS 23 statistical package and the Excel 2013 spreadsheet. The qualitative type variables were presented by means of numbers and percentages and the quantitative variables were characterized by means of arithmetic mean and standard deviation. In all calculations, $p < 0.05$ was assumed as the level of significance.

RESULTS

The research covered 750 Tri-city students, mainly from public universities (98.9%). Majority (91.3%) of them declared attending a medical university. Most respondents were in the age range of 18-22 (70.7%) and in the range of 23-26 (25.7%). The smallest respondents groups were from the age range of 27 - 30 (2.5%) and those over 30 years (1.1%). The majority of respondents were women (79.2%). Most of the students lived in large (over 100,000 inhabitants) cities (64.3%). 20.8% lived in cities with a population up to 100,000 inhabitants, while in rural areas - 14.9%. The majority declared single (94.5%), formal relationships were 4.9%, while groups declaring divorce and separation accounted for 0.3% each. 88% students declared themselves as heterosexual, 4.1% - homosexual - 4.1% and 7.9% - bisexual.

Most students (76.4%) have already begun sexual activity, less than $\frac{1}{4}$ (23.6%) declared virginity. According to the statistical calculation, average age of starting sexual intercourse was 18. The lowest indicated age of initiation is 13 years, the highest is 27 years. The current possession of a sexual partner was confirmed by 65.6% of students.

More than a half of the respondents (66.8%) were using contraception. The most frequently indicated method of contraception used in the past or present is a condom (52.2% of indications), the second is an oral hormone pill (25.5%). The percentage share of all methods is presented in Figure 1.

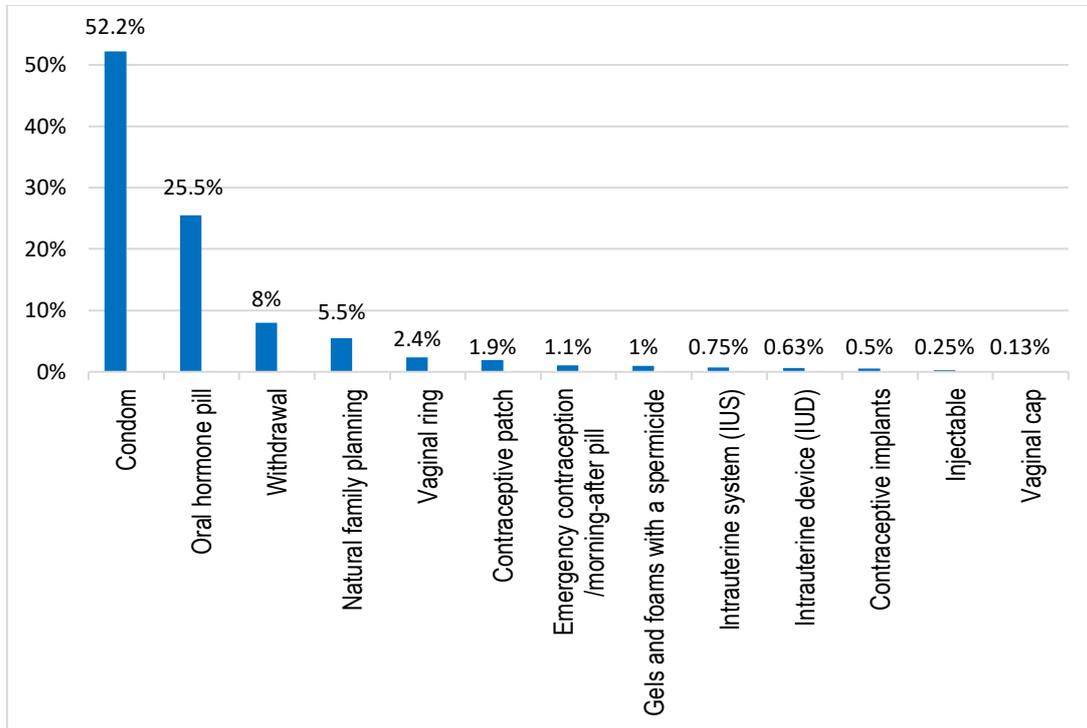


Figure 1. Contraception methods used by students
Source: own studies

The analysis of the collected data shows that 24% of the surveyed students use or used more than one method of contraception in the past.

According to 316 respondents (42.1%), the choice of the method used, was a common decision of both partners, 31.7% declared making a decision on their own, while only 1.5% left the choice of the method used to the sexual partner. As the main factor influencing the choice of a given method of contraception, students indicated negative side effects of other methods (31.3%). More details are presented in Figure 2.

The level of satisfaction with the used method of contraception should be assessed as high (69.7%). Only 2.5% declare dissatisfaction with the method of applied birth control. Among the declared causes of dissatisfaction, the biggest (31.6%) is inconvenient or troublesome application of the chosen method. 26.3% think that the method weakens own of partner's sexual experience, 21.1% is not sure that the method is effective, while 15,8% rejected

the technique because of side effects, and 5.3% thought that the method is incompatible with their internal beliefs.

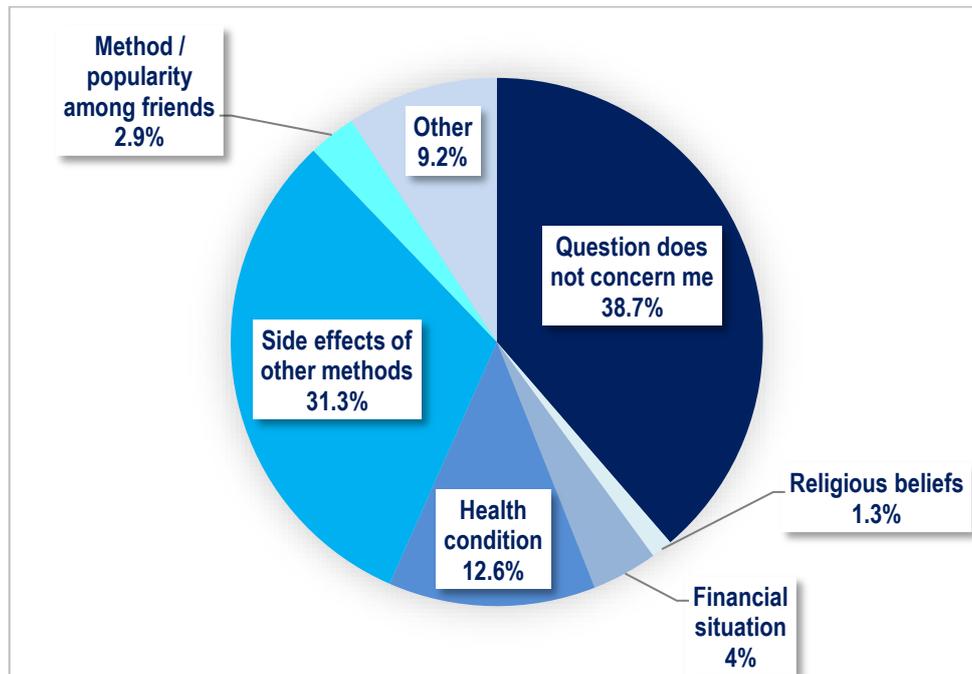


Figure 2. Factors determining the choice of a given method of contraception

Source: own studies

Students obtained the first information on contraception in a variety of ways. The most popular ways was indicated as (22.5% each) a family home and the Internet.

For the question "What is contraception?", 58.4% of respondents answered that these are "methods and techniques preventing fertilization and preventing the implantation of a fertilized egg". Students opinions are summarized in Table 1.

Table 1. Understanding the concept of contraception by students

What is contraceptive?	N	%
Surgery as a result of which the body can't fertilize / become pregnant	1	0.1
Methods and techniques to prevent fertilization	307	40.9
Observation of fertile days symptoms and avoiding intercourse	2	0.3
Methods and techniques to prevent fertilization and to prevent the implantation of a fertilized egg	438	58.4
A procedure resulting in intentional disruption of the fetus's life and removal from the uterus	2	0.3

Source: own studies. N - quantity

For the most effective contraceptive method students chose a surgical sterilization procedure (39.6%). The remaining answers are shown in Figure 3.

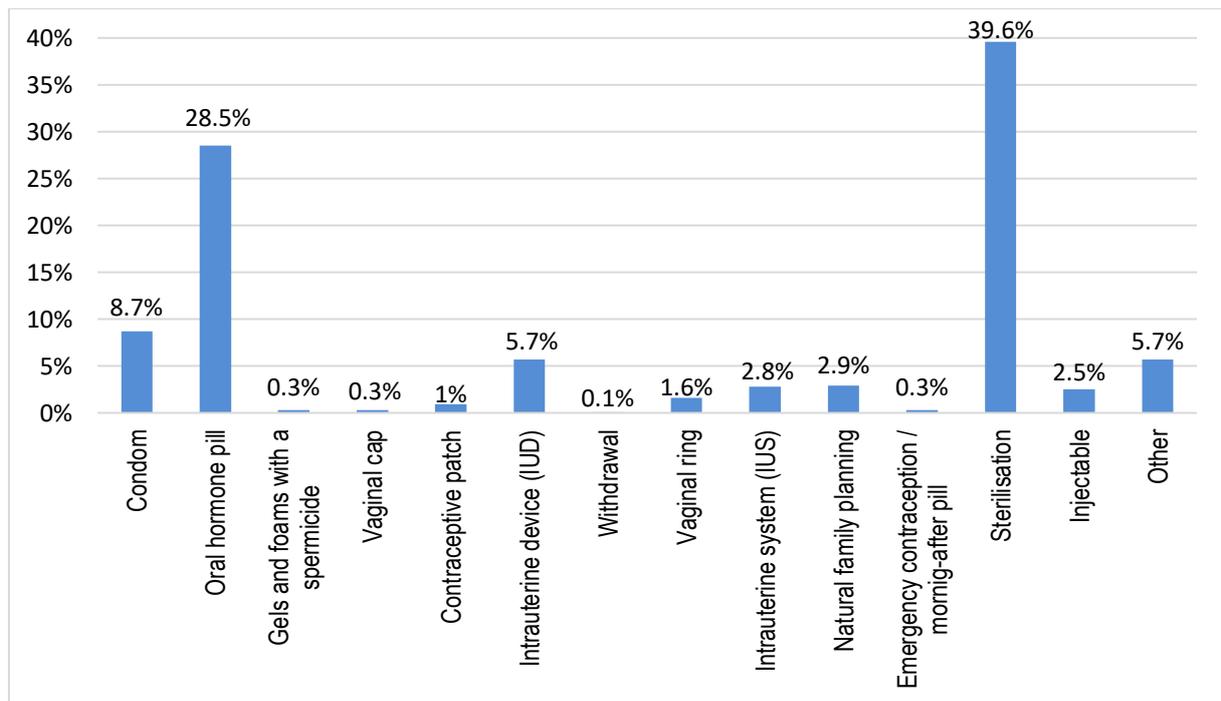


Figure 3. The most effective method of contraception according to students' opinions
Source: own studies

A condom (70.1%) was considered as the healthiest contraceptive. On the next place was natural family planning (19.2%). For 1,7% of responders it was "lack of sexual intercourse", "abandonment of cohabitation", "restraint" or "sexual abstinence".

Students were also asked to group contraceptive methods by type, according to their subjective feeling or knowledge. Table 2 presents the analysis of the percentage share of the indicated responses.

To determine the responders level of knowledge, regarding the classification of contraceptive methods, the number of correct answers to questions was summed up and then the descriptive statistics for the variable thus formed were calculated (normality test of Kolomogorov - Smirnov $K-S_{(750)} = 0,18$). Based on the results (9,2/13 points) the level of knowledge rated as medium.

The study also showed that significantly higher level of knowledge about the types of contraception had students of the medical university than students of technical university (nonparametric significance test Kruskal - Wallis, $H_{(3)} = 11.29$). The method of Bonferroni multiple comparisons was used for checking dependency between students of different universities. Details are summarized in Table 3.

Table 2. Contraceptive methods grouped by students

Methods of contraception (% answers)	Natural	Mechanical	Chemical	Hormonal	Surgical	It's not contraception
Male Condom	0.7	98.0	0.4	0.0	0.0	0.1
Oral hormone pill	0.9	5.9	1.9	91.2	0.0	0.1
Globulks, gels and foams with a spermicide	0.3	8.4	89.2	0.5	0.1	1.5
Vaginal cap	0.1	90.3	1.7	2.0	5.6	0.3
Contraceptive patch	0.9	10.4	12.5	70.5	0.3	5.3
Intrauterine device (IUD)	0.5	55.1	9.6	13.1	21.3	0.4
Withdrawal	42.1	1.6	0.0	0.1	0.3	55.9
Vaginal ring	0.7	51.5	11.6	28.1	7.7	0.4
Intrauterine system (IUS)	0.7	10.4	0.9	85.2	2.5	0.3
Natural family planning	72.9	0.1	0.1	0.1	0.1	26.6
Emergency contraception/ morning -after pill	1.2	6.1	25.3	32.5	0.3	34.5
Sterilisation	0.9	8.9	1.1	1.1	80.1	7.9
Injectable	1.5	6.0	19.5	68.1	1.3	3.6

Source: own studies.

Table 3. Knowledge level vs university

Student status	N	M	SD	H	df	p
Other university	20	9.05	2.09	11.29	3	0.010
University student	26	8.69	2.22			
Medical university student	685	9.27	2.11			
University of Technology student	19	<u>7.53</u>	2.78			

Source: Kruskal-Wallis test $H(3) = 11.29$, N – quantity, M – median, SD - standard deviation
 H - Kruskal-Wallis significance test, df - degrees of freedom, p - level of significance

Analysis of the data also showed that women have a higher level of knowledge about the classification of contraceptive methods than men (U Mann Whitney significance test, $Z = -4,84$). These data are presented in Table 4.

Table 4. Knowledge level vs gender

Knowledge level and gender	N	M	SD	Z	p
Woman	594	9.35	2.15	4.84	0.000
Man	156	8.63	2.03		

Source: Mann-Whitney U test $Z = -4.84$, N – quantity, M – median, SD - standard deviation
 H - Kruskal-Wallis significance test, df - degrees of freedom, p - level of significance

DISCUSSION

Most of the surveyed students (76.4%) have already started the sexual activity. Analyzing the declared time of initiation, it can be assumed that a significant part of the respondents had their first relationship before reaching the age of consent. This trend is also confirmed in other Polish and foreign reports. In their studies from 2013, Bień and Stadnicka indicate that the average age at which teenagers begin sexual intercourse is 17,05 [3]. The report of the National Center for Health Statistics 2019 also defined the average age of sexual initiation, both among boys and girls, for 17 years [C]. In Sweden, in 2015, the declared age at which the first sexual relationship took place was lower among girls (16 years) than among boys (17 years) [4].

The analysis of own research shows, that more than a half of the respondents currently use contraceptives (66.8%). The main method is a condom (52.2%). Similar results were obtained by Izdebski and Wąż, probing in 2011 preferences of 3206 adult Poles aged 15-59. They showed that 72.4% of respondents used at least one method of contraception during the last 12 months, of which 65.9% used the condom [1]. The reason for the popularity of this method is probably the ease of its access and use, and significant efficiency at a low price. In favor of the method is also belief of the respondents about its safety for health. The adverse effects of other methods of contraception were the decision-making factor in the selection of the currently used method. In the studies conducted by Szyper and Gotlib, comparing the knowledge of students of the Warsaw Medical University and the Warsaw University of Technology about oral hormonal contraception, different decisive factor appeared in the first place. There, the respondents inclined the highest effectiveness of a given method of birth control [5]. Despite the fact, that the sterilization of women and men in Poland is prohibited by law, it still was considered the most effective method. Due to decreasing fertility rate of women in Poland, which we can observe in the last 30 years [D], education in the reversibility of contraception is a priority.

Disturbing is high usage of low-effectiveness contraceptives, which are not even included in the current recommendations of the Polish Gynecological Society, such as withdrawal (8%) and natural family planning (5.5%). In studies conducted among eighteen-year-olds, carried out by the Institute for Educational Research (IBE) in 2015, over one-third of respondents considered the intermittent relationship as an effective method of preventing pregnancy. However, as many as 12% of the surveyed teenagers declared regular use of this method [E].

Modern hormonal methods of contraception, like vaginal rings, hormone intrauterine systems, implants, and injections, are not popular or trusted by Tri-city students. The reason may be low knowledge of these methods because, in one of the questions, more than half of the students incorrectly classified the vaginal ring for mechanical contraceptive methods. In studies carried out in 2013-2014 among young Poles aged 18 - 35 years, only 1/4 of the respondents were admitted to the hormonal patch and 10% had heard of the vaginal ring, only 3% of respondents were known to have a contraceptive implant [F]. The situation in other parts of the world is different. In the studies conducted in 2011-2013 in the USA, the percentage of modern methods of hormonal contraception, as the methods of birth control used by women, amounted to 18.7% in total and increased by 10.2% compared to the 2002 study [6]. In England in 2017/18, as many as 41% of women applying for Sexual and Reproductive Health Services (SRHS) used long-acting reversible contraceptives (LARC), while in 2007/08 this percentage was 23%) [G].

In studies conducted among teenagers aged 16-19 by Sieńko and Stokłosa, similarly as in own studies, it was shown that the main source of information on contraception is the Internet [7]. There are concerns about the reliability of information obtained in this way, what definitely indicates an argument for creating structured education for young people and adults regarding reproductive health. The feasibility of these anxieties is confirmed by the Contraception Atlas, published by the European Parliamentary Forum for Sexual & Reproductive Rights (EPF) in February 2019. It is a graphical representation of the level of access to on-line information about modern contraception and access to contraceptives and advice on reproductive health. Poland was ranked in the last place of the accessibility ranking of the aforementioned, just behind Russia (42.8%) and Belarus (44.3%), gaining 31.5%, far behind the leading countries such as France and Belgium (90.1% each), United Britain (87.6%) or the Netherlands (81.1%) [H].

Conducted study shown that the level of knowledge of medical university students on the classification of contraceptives is higher than in students of technical universities. It is consistent with the assumption that a graduate in a field related to medicine should have the ability to make a free conversation with the patient about contraception, sexuality, prevention of sexually transmitted diseases, or the most common sexual dysfunction [8]. This is also confirmed in the aforementioned studies by Szyper and Gotlib [5]. The study also showed that women are equipped with a thorough knowledge of the classification of contraception more often than men. The results of assessing the knowledge of sexual and reproductive health issues conducted by IBE in 2015 are slightly different, where the percentage of correct answers was very different. The vast majority of the respondents knew the principle of condom operation,

however, less than 50% of them properly defined such a principle for hormonal methods, and less than 1/3 of the intrauterine spiral [E].

Analyzing the results of own research as well as other Polish and foreign reports, the deficit in the level of knowledge among tri-city students in the use of contraception and the variety of available forms was highlighted. It becomes appropriate to introduce holistic education and disseminate, especially through widely available, modern information channels, reliable knowledge in the field of reproductive health and family planning, addressed primarily to young people who have not yet taken sexual activity. Such educational activities would help reduce the percentage of people engaging in unprotected sexual activity, thereby affecting the number of unwanted pregnancies or minimizing the risk of STD transmission.

CONCLUSIONS

1. The level of knowledge of students of Tri-city universities on contraception was defined as average. Students of the medical university have more knowledge in this field than students of the technical university. In addition, women are equipped with a thorough knowledge of this subject more often than men. The main source of information on contraception is the family home and the Internet.
2. A significant proportion of students took sexual activity before reaching the age of majority. The most popular method of contraception used by them is the condom. Modern hormonal methods of contraception are not popular, nor trust among students. It is recommended to conduct education in the use of contraception and the variety of available forms, and the promotion, especially through widely available, modern information channels, reliable knowledge on reproductive health and family planning.

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Sexual behaviour of participants of the senior clubs in Gdańsk

Katarzyna Turoń, Jolanta Olszewska, Agnieszka Czerwińska-Osipiak

Department of Obstetrics and Gynaecology Nursing, Institute of Nursing and Midwifery
Faculty of Health Sciences with the Institute of Maritime and Tropical Medicine, Medical
University of Gdansk, Poland.

INTRODUCTION

Ageing is a process of continuous reduction of the body's functional reserves. In Ian Stuart-Hamilton's opinion, this process is influenced by all the phenomena affecting a given person resulting from his development. The author defined the process of old age as achieving measurable attributes of late age. Old age is defined as a phenomenon, life phase or state. According to Puchalska, old age is dynamic because it is related to many variables, e.g. change of living conditions which may have a negative impact on an individual [A,1,2,3].

Precise determination of the moment when the aging process begins is not possible due to individual differences in development. Cieślik indicates that changes occur in every human being, but they differ in intensity, dynamics and duration of the processes, and are influenced by internal (genetic) and external (environmental, lifestyle) factors. Due to a wide range of factors, three age criteria have been distinguished from them, which were presented in Table 1. [A, 1,3,4].

Table 1. Basic criteria for old age according to Cieślik J. [A]

Criterion	Description
Biological	all physiological changes in the body which lead to severe and rapid disease only when there are risk factors, e.g. stress, environmental pollution, genetic stress
Psychological	changes in personality, subjective feelings and mental processes
Socio-economic	human functioning is influenced by institutional, economic and sociological factors such as retirement

The different age definitions presented in Table 2 are also used to determine the starting point of old age.

Table 2. Division and types of age determining old age [2,3]

Age	Description
Calendar/chronological	In western countries, the lower limit of old age is 65 years, which is not reasonable in African countries. The UN has adopted the age limit of 60 years as a contractual limit. The WHO has divided old age into three stages: <ul style="list-style-type: none"> – elderly 60-75 years of age – old age 75-90 years of age – very old age over 90 years of age
Biological	Individual assessment of developmental standards, e.g. dental, physiological and psychological age
Social	Assessment of an individual's ability to adapt to different social situations, considered to be common standards

Women's adaptation to old age can be influenced by modern times, which have brought positive changes for seniors in the sphere of issues previously considered forbidden. One such issue was sex life in late old age and sometimes even in middle adulthood. Currently, on many sites dedicated to seniors it is possible to find papers and surveys concerning the most intimate sphere of life. Also research by Starr and Weiner, conducted on a group of over one thousand older people, show that 97% of 60- and 70-year-olds and 93% of 80-year-olds declared not only a longing for sex but also thinking about it. Professor Izdebski Z. attributes great importance to sex and sexuality in old age. In a study published by him, 35% of women and as much as 60% of men over 50 years of age declared that sex has an important role in their functioning. In his opinion, the differences in perception of sex and sexuality between the group of women and men are caused by the fact that in women the ageing of the body is correlated with a decrease in sexual activity in the perimenopausal period and of course it is partly due to cultural conditions. The processes are typical for both genders, although for men they are more gradual, up to an old age. According to Professor Bogdan Wojciszke, proper sexual functioning in the period of menopause consists of many factors, such as self-esteem, attractiveness, exhaustion or forgetting. Unfortunately, according to research, it is the loss of attractiveness of own body in the eyes of women that is the reason why they avoid intercourses. Moreover, according to James Hillman (2000), the sexual sphere is multidimensional and consists of psychological phenomena and behaviors, e.g. body image, having and relationship with a partner or a desire to risky caresses. However, health, sexual disorders and practical problems (lack of a partner) have a positive or negative impact on sexuality [5-8].

Bretschneider and McCoy recorded a significant percentage of sexually active women in the group of eighty-years-old and older (30%). Data from the report of the *National Opinion*

Research Center in the United States are contradictory – only 3% of women declare sexual activity. Such discrepancies may be caused by different views on the issue of sexuality, under which vaginal, anal, masturbation, peeing, necking and others may be hidden [B,9].

Professor Zbigniew Izdebski in “Wyborcza” (2011) reported that about 51% of Polish people (fifty-year-old and older) are satisfied with the erotic sphere during the ageing period. However, he points out that the rest of them have no clear impressions – almost 1/4 of the respondents did not want to answer this question. The author emphasized that in sexual relations of elderly people the most important thing is the need for closeness. According to the respondents, the couple ages together – they accept changes in appearance. For most of the respondents hugging is more important than the intercourse. Sexually active seniors emphasize the feeling of comfort from sexual life associated with natural lack of pregnancy as a consequence of intercourse [C,D].

MATERIALS AND METHODS

The study covered 100 women who are members of senior clubs in Gdańsk.

The respondents were informed about the aim of the study, its anonymity, and the possibility to refuse to fill in the questionnaire.

The criteria for inclusion in the study were: age 60 and over, understanding of Polish language in speech and writing.

The STATISTICA program and Excel 365 spreadsheet were used for statistical calculations. Spearman’s rank correlation method was used to investigate the correlation between selected variables.

RESULTS

Age

For the purpose of analysis, the collected results were divided into specific areas. The first is age. Respondents were divided into four age categories.

The most numerous group were women aged 66-71 years (24%). The lowest percentage were seniors in the oldest age category (10%). The groups of 60-65 years old and 72-77 years old constituted 24% and 20% respectively.

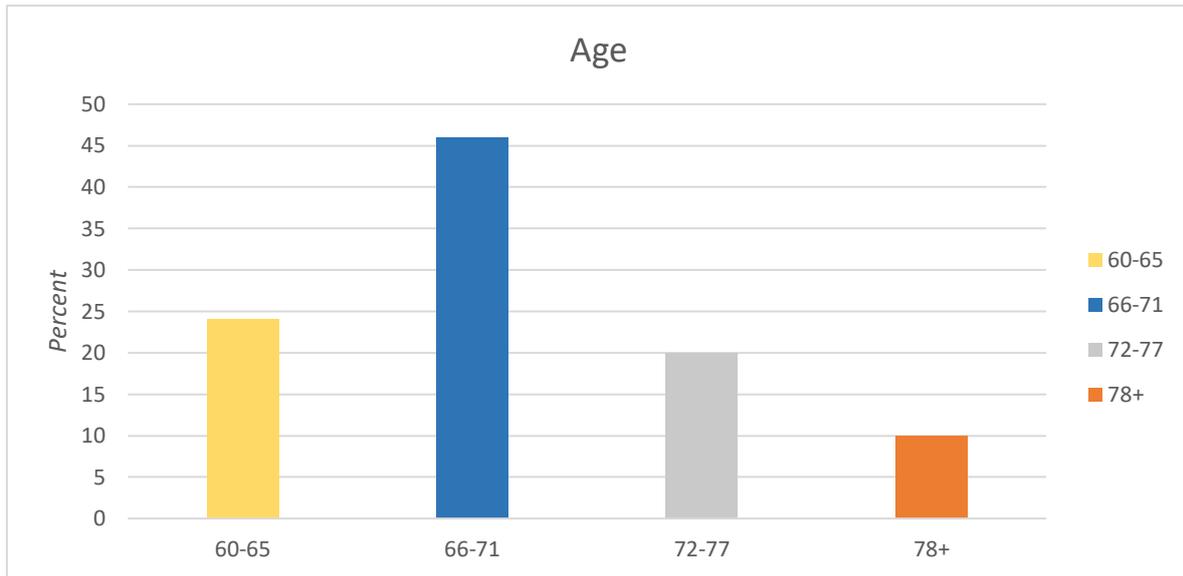


Figure1. Age of seniors
Source: author’s own analysis

Place of residence

Another area used for the analysis is the place of residence. The most numerous group in terms of place of residence were women living in cities with more than 250 thousand inhabitants (55%), while the smallest group of respondents were women living in rural areas (6%). The percentage of female inhabitants of cities up to 100,000 and 200,000 inhabitants was 13% and 26% respectively.

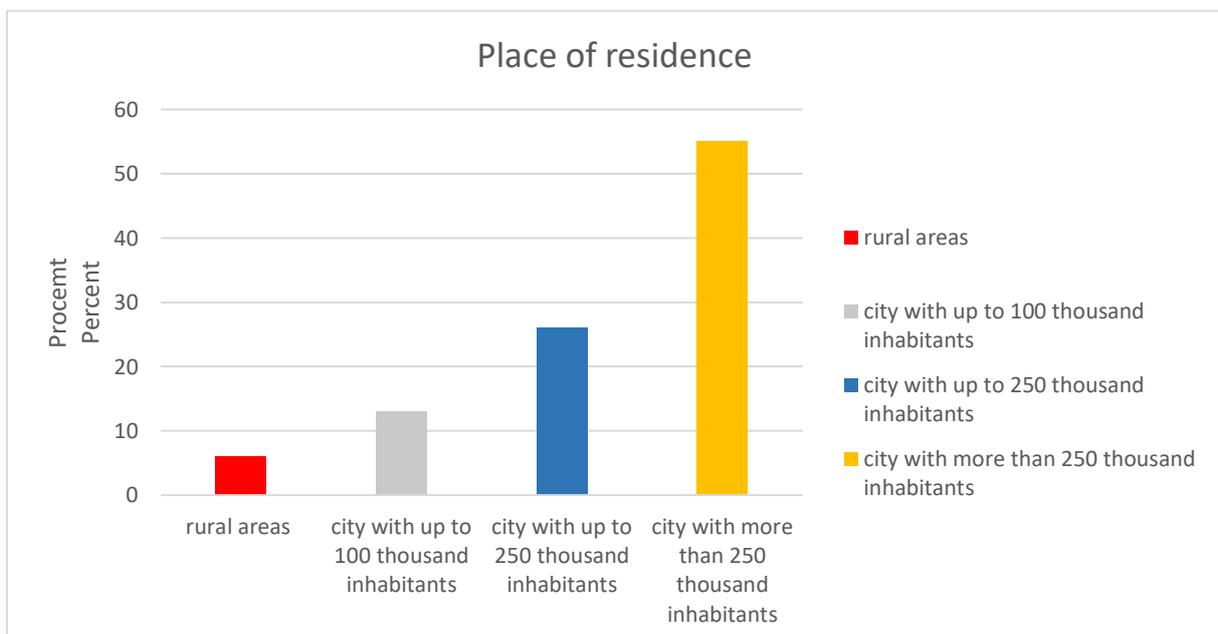


Figure2. Place of residence
Source: author’s own analysis

Education

The next area into which the collected results were divided is the respondents' education. Women with secondary education constituted the most numerous group (44%). Senior women with primary education constituted the smallest percentage of the respondents (5%). 21% of the respondents graduated from vocational schools. 30% of the respondents graduated from higher education (bachelor's degree, master's degree).

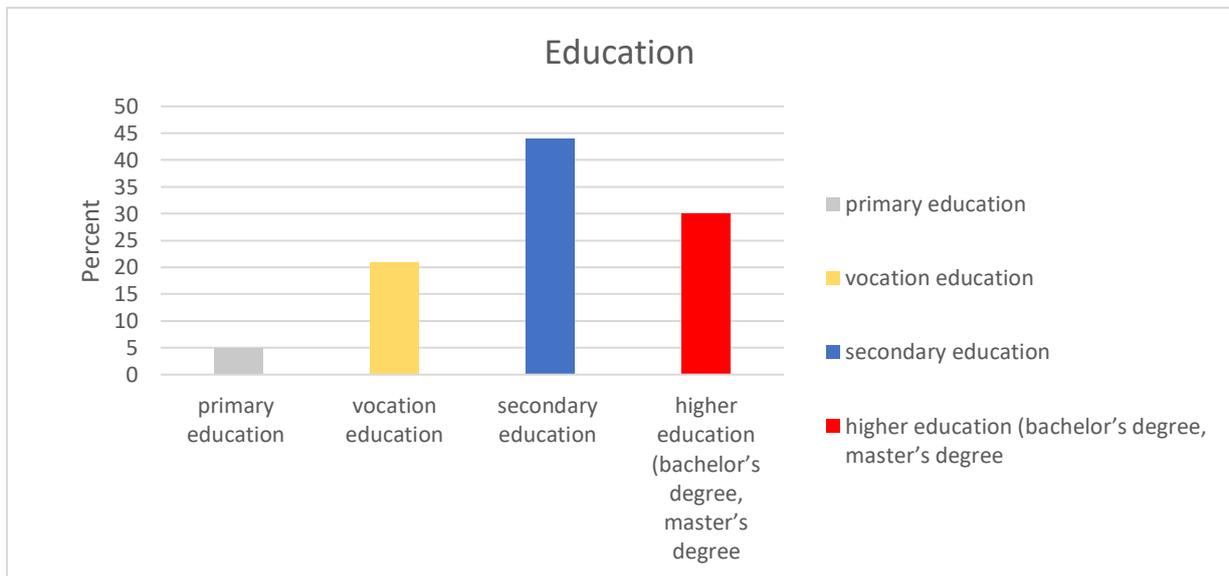


Figure 3. Education of respondents
Source: author's own analysis

Relationship status

Another area is the relationship status. More than 1/5 (22%) of respondents are in a relationship (marriage, partnership) while 78% are single.

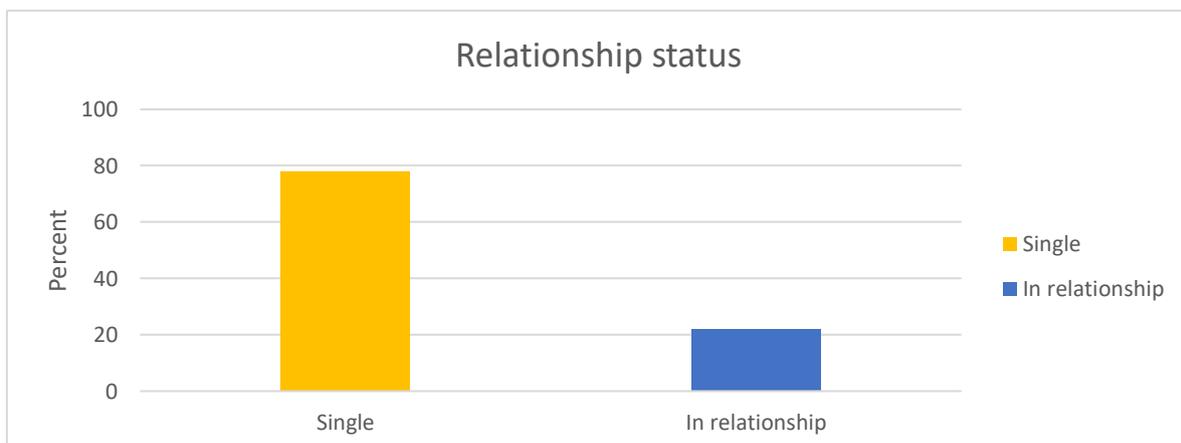


Figure 4. Relationship status
Source: author's own analysis

Sexual behavior

Approximately 2/3 (67%) of respondents lead an active sex life, while 33% did not admit to such activity.

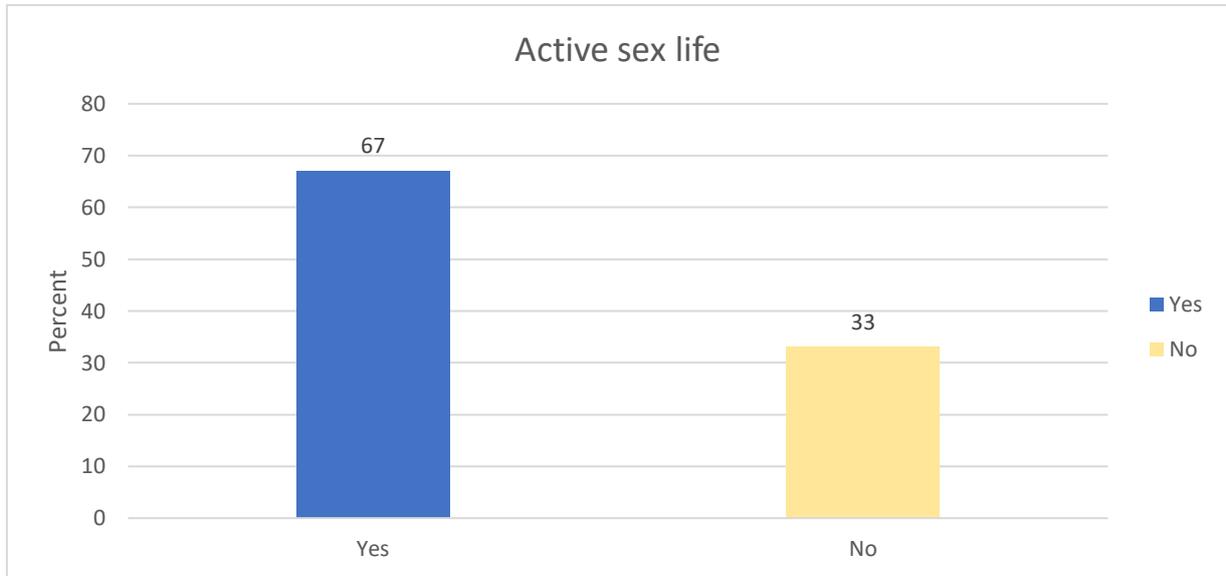


Figure 5. Active sex life

Source: author’s own analysis

Among the respondents leading a sex life (67%), the most popular are: hugging (21%) followed by sexual intercourse (18%), masturbation (18%), holding hands (16%), kissing (14%). The least popular is fondling (12%).

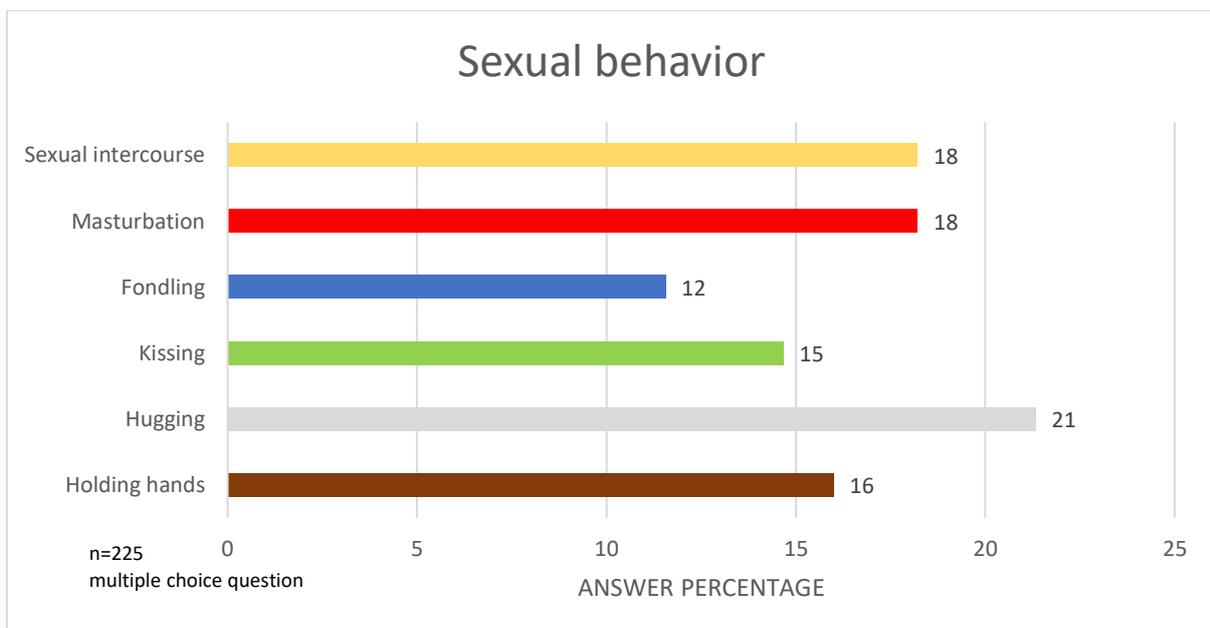


Figure 6. Respondents’ answers – sexual relations

Source: author’s own analysis

The most popular among the respondents is classical sexual intercourse (70% of responses). However, *cunnilingus/fellatio* and anal intercourse was found in 26% and 4% of responses respectively.

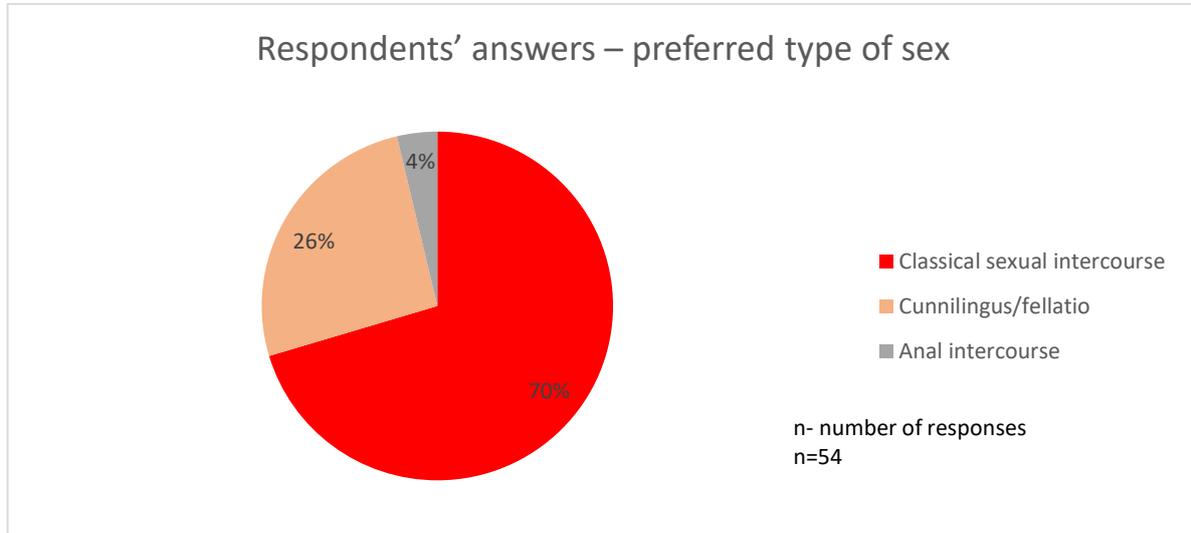


Figure 7. Respondents' answers – preferred type of sex
Source: author's own analysis

The number of sexual intercourses in the last month

Exactly 20% of the respondents stated that had sex 4-6 times in the last month, while 15% of women stated that had sex 1-3 times in the last month. Each 3% of seniors had sex 7 to 9 times and more than 10 times.

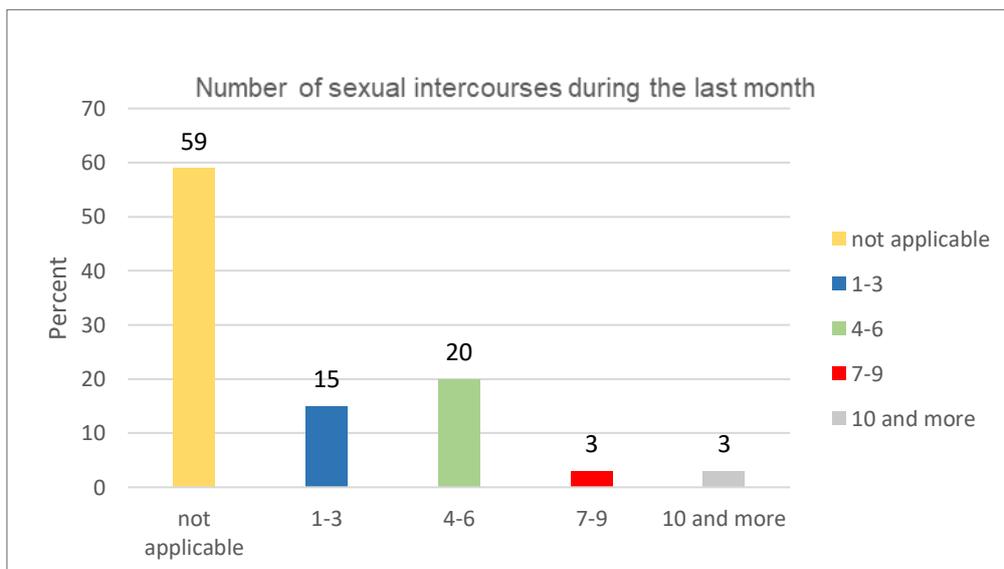


Figure 8. Number of sexual intercourses during the last month
Source: author's own analysis

Analyzing the collected results, several correlations between selected variables were found. Spearman's rank correlation was used in the study, showing the influence of X variable on Y variable. If the values are positive then the value of X variable increases along with the value of Y variable. If the values are negative then the value of X variable increases and the value of Y variable decreases. Qualitative variables are assigned individual ranks.

Correlation coefficient sign (+/-) indicates the direction of correlation (positive/negative). Its absolute value, on the other hand, indicates the correlation strength.

r_s – correlation coefficient

Table 3. Interpretation of Spearman's correlation coefficient

r_s	interpretation
under 0.2	weak correlation (practically no correlation)
0,2-0,4	low correlation (clear correlation)
04,-0,6	moderate correlation (significant correlation)
0,6-0,8	high correlation (significant correlation)
0,8-0,9	very high correlation (very high correlation)
0,9-1,0	correlation almost certain

In order to confirm the "reliability" of calculated Spearman's rank correlation coefficient (r_s), the correlation significance was used. If the result of correlation significance test, i.e. p value is less than 0.05 ($p < 0.05$), it can be assumed that presented correlation is statistically significant.

The analysis of respondents' answers confirmed the existence of some correlations:

- along with the age of respondents, the number of declared sexual intercourses decreased ($r_s=0.35$) (especially classic sex ($r_s=0.32$) and the number of intercourses during the last month ($r_s=0.38$);
- along with the change in marital status the number of sexual intercourses increased ($r_s=0.39$) and the number of intercourses during the last month ($r_s=0.36$);
- respondents declaring holding hands with their partners more often hug ($r_s=0.49$) and kiss ($r_s=0.45$);
- respondents who kiss – more often caress each other with their partner ($r_s=0.55$), more often have sex ($r_s=0.58$), especially the classic ones ($r_s=0.54$), and more often have sex with their partners during the last month;

- along with the increase in the number of fondling, the number of sexual contacts increases ($r_s=0.61$), especially classic intercourses ($r_s=0.57$) and the number of intercourses during the last month ($r_s=0.54$);
- respondents having sex more often have classical sex ($r_s=0.93$) and more often have had sex during the last month ($r_s=0.96$);
- along with the increase in sexual contacts, the number of oral sex increases ($r_s=0.48$);
- along with the increase in the number of classical sex among the respondents, the number of oral intercourses increases ($r_s=0.35$) and the number of intercourses during the last month ($r_s=0.91$);
- along with the choice of oral sex, the number of declared sexual intercourse by respondents increases ($r_s=0.35$) and the number of intercourses during the last month ($r_s=0.49$).

The above correlations are included in the Spearman Rank Correlation Matrix presented below. X variables are in columns and Y variables are in lines of the matrix (Table 4).

Communication with medical personnel

The respondents were also asked if physicians talk to them about sex and sexual problems.

Exactly 8% of the respondents were asked by the physician about problems of a sexual nature. The remaining 92% did not address this topic during medical visits (Fig. 9).

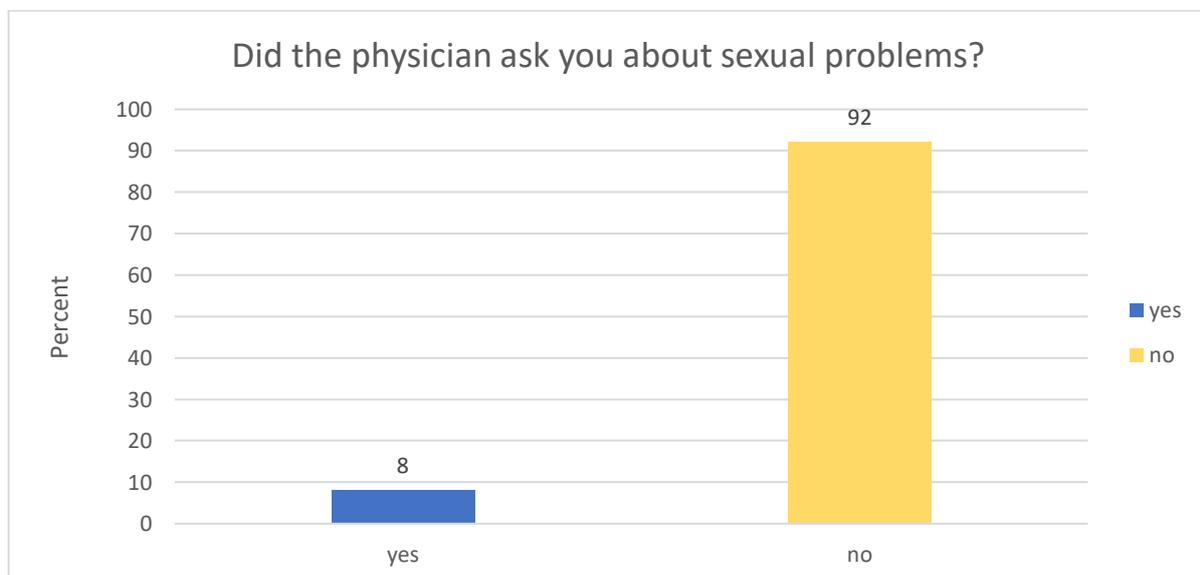


Figure 9. Did the physician ask you about sexual problems?

Source: author's own analysis

Sexual behaviour of participants of the senior clubs in Gdańsk

Table 4. Spearman's rank correlation matrix. Statistically significant correlations are marked in red ($p < 0.05$)

Variable Y	Spearman's rank correlation matrix. Statistically significant correlations are marked in red ($p < 0.05$)															
	Age	Place of residence	Education	Relationship status	Holding hands	Hugging	Kissing	Fondling	Masturbation	Sexual intercourse	Classical sexual intercourse	Cunnilingus/Fellatio	Anal intercourse	The number of sexual intercourses in the last month	The knowledge level of sexual problems	Willingness to get specialized knowledge
Age	1,000000															
Place of residence	0,160727	1,000000														
Education	-0,167599	0,355023	1,000000													
Relationship status	-0,249625	0,011125	-0,026767	1,000000												
Holding hands	0,027698	-0,189219	-0,246403	0,205192	1,000000											
Hugging	-0,047309	-0,166038	-0,141672	0,117899	0,488724	1,000000										
Kissing	-0,094249	0,089024	-0,040481	0,243347	0,448379	0,475059	1,000000									
Fondling	-0,177651	0,125201	0,088897	0,290585	0,172885	0,251892	0,553693	1,000000								
Masturbation	-0,171029	-0,074292	-0,003250	0,189627	-0,009770	0,183044	0,125916	0,163046	1,000000							
Sexual intercourse	-0,351412	0,038651	0,024423	0,391675	0,179600	0,216507	0,582445	0,618351	0,210964	1,000000						
Classical sexual intercourse	-0,322602	0,017802	0,001142	0,280500	0,142498	0,196289	0,545929	0,569262	0,103864	0,939139	1,000000					
Cunnilingus/Fellatio	-0,144749	0,137241	-0,020238	0,133576	0,117680	0,016152	0,268452	0,220762	0,097987	0,484004	0,337246	1,000000				
Anal intercourse	-0,108154	-0,067207	-0,010560	-0,075869	0,041667	0,005719	0,051648	0,078165	-0,025123	0,171370	0,182476	0,354068	1,000000			
The number of sexual intercourses in the last month	-0,376143	-0,005479	-0,037196	0,368994	0,163912	0,188821	0,542346	0,543018	0,153739	0,962597	0,909658	0,490719	0,230665	1,000000		
The knowledge level of sexual problems	-0,198708	0,196425	0,246815	-0,037991	-0,049623	0,206029	0,130710	0,125090	-0,001247	0,066158	0,067474	-0,023290	-0,051648	0,055358	1,000000	
Willingness to get specialized knowledge	-0,002442	-0,124413	-0,140100	0,192585	0,071623	-0,084694	0,020153	0,073354	0,145984	0,273776	0,180704	0,250239	0,066114	0,285102	-0,348230	1,000000

Source: author's own analysis

When asked about the willingness to obtain specialist knowledge from a physician, 29% of the respondents answered affirmatively, while 71% of the respondents did not express such a need (Fig. 10).

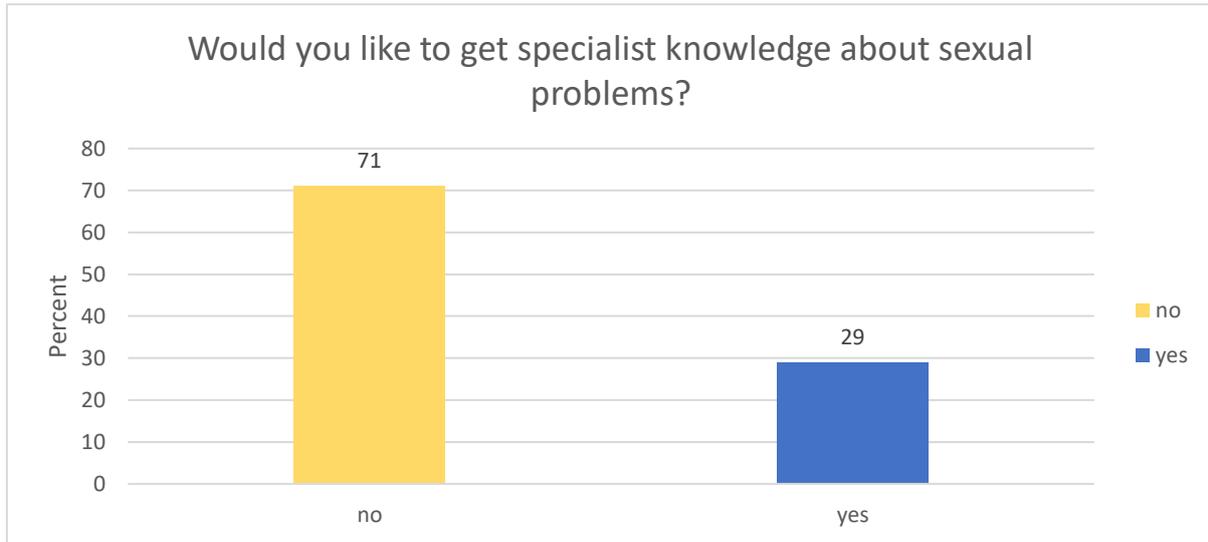


Figure 10. Would you like to get specialist knowledge about sexual problems?
Source: author's own analysis

When asked about the knowledge level of sexual problems, 67% of respondents think it is appropriate, while 33% of respondents answered negatively to this question (Fig. 11).

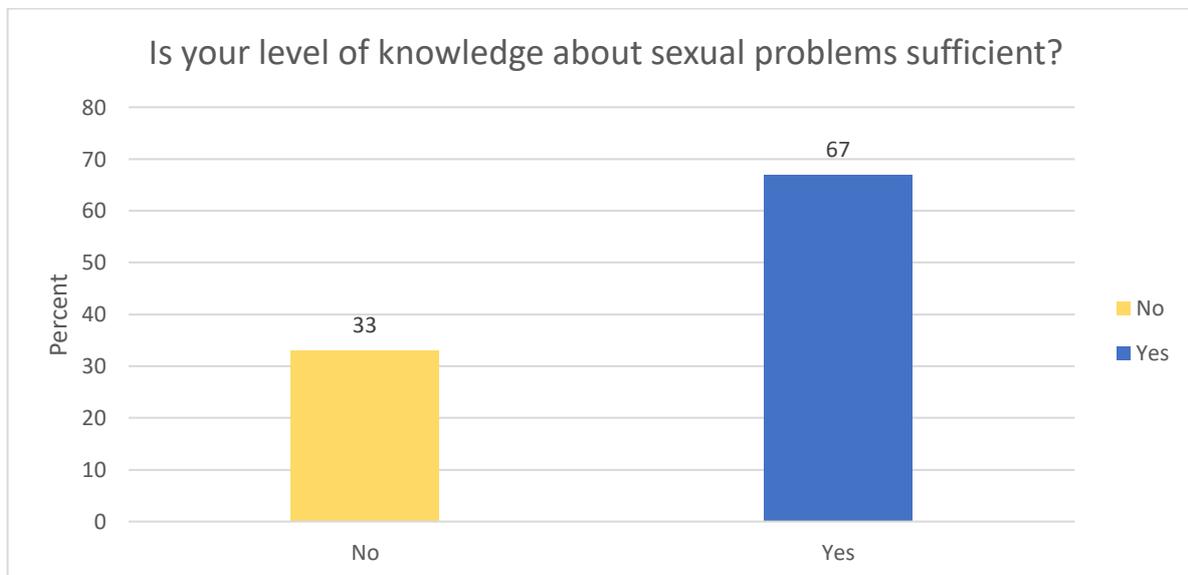


Figure 11. Is your level of knowledge about sexual problems sufficient?
Source: author's own analysis

DISCUSSION

The authors would like to emphasize that they are aware of the fact that the sample was characteristic. Participants of seniors' clubs are socially active and more educated people, and the results of research conducted in a different environment, on a similar age group could be quite different.

The authors have attempted to establish cooperation with the residents of hospitals or nursing homes. However, due to the significant burden of co-existing diseases (e.g. dementia) and negative reception by the group, attempts to conduct the study were abandoned. In addition, a survey was published on the website dojrzaliwspaniali.pl. On the generated 600 page views, only 30 correctly filled in forms were collected. Due to their small number, they were not used in the above research. The highest percentage of returns of correctly filled in questionnaires was achieved by direct meetings with seniors during classes in the senior clubs.

Does sexual behaviour concern older women?

The results of the *English Longitudinal Study of Ageing* (ELSA) conducted among 3432 seniors indicate that 53% of respondents over 50 years of age declared at least one of their sexual behaviour [10]. Whereas in Spain, according to the results published by the *Spanish National Sexual Health Survey* (SNSHS) carried out on a sample of 1018 women, 37.4% of them are sexually active [11]. The research carried out by the author of this study confirms the papers of foreign researchers – 67% of the respondents lead a sexual life through various types of intercourses.

What kind of sexual behaviors concern the seniors in Gdańsk? Do they differ from their female peers from Europe?

In Spain, kissing and hugging are the basic sexual activities involving 25.67% of women over 60 years old [11]. In the United Kingdom, kissing and fondling, i.e. hugging with caresses of erogenous spheres, is one of the sexual behaviors chosen by more than half of mature British women – 65.6% [10]. Our own research confirmed the presented data – among the respondents leading sex life (67%) the most popular are hugging (21.33%), holding hands (16%), kissing (14.67%). Mutual fondling is the least popular (11.56%). Interestingly, according to Nancy R. Hooyman and Asuman H. Kyak, it is kissing, hugging and lying next to each other that significantly affects the assessment of intimacy and sexual satisfaction by seniors [12].

Another example of sexual behaviour is masturbation. According to the report by Professor Izdebski, 83% of Polish men and 23% of Polish women under 45 years of age masturbate [E]. Is it as popular among seniors as in the younger generation?

In Spain, 10.07% of seniors declared masturbation as one of the methods of sexual satisfaction [C]. In England, this percentage was 15.9% [11]. According to the Natsal-3 report (*The National Survey of Sexual Attitudes and Lifestyles*), 26% of 55-64 year old and 19% of 65-64 year old in Great Britain masturbate [F]. Our own research showed that masturbation is chosen in 18% of cases.

What about sex?

The participants of senior clubs in Gdańsk are not much different from their foreign colleagues – 70% of them prefer traditional intercourse, 26% prefer petting and 4% prefer anal technique. Spanish mature women in 55.64% have classical sex, while in 4.8% have oral sex [11]. English women in 50.1% choose the vaginal technique of sexual intercourse. The percentage of women declaring active oral sex is 16% (55-64 years of age), 7% (65-77 years of age) and 15% (55-64 years of age) and 7% (65-77 years of age) of passive sex [10].

On the basis of the above mentioned studies and own results, it can be concluded that ladies aged sixty and over have a rich sexual life. Kisses and hugging are of great interest. The elderly declaring sexual intercourse in most cases enjoy classical sex. Very rarely do they declare oral and anal techniques as a kind of intercourse. It is likely that this has to do with traditional upbringing. In a more sexually liberal England, it is possible to observe the advantage of petting over classical intercourse. It should be noted that in the years of the present youth of seniors, the education of women was significantly different from that of men. The girl, and later the woman, did not agree to talk about sexuality without being accused of promiscuity. Among men, there was greater social consent to discuss sexual issues, as can be seen today. Most men are not ashamed of their own body and sexuality, unlike their female colleagues of the same age.

What else affects the choice of sexual behaviour in late adulthood?

In Gothenburg, Sweden, Nils Beckman has identified positive and negative factors influencing the sexual behaviour of older people based on research. The first group consists of seniors who have a positive attitude towards their changing sexuality, their sexual debut before the age of 20, their psycho-physical well-being, and those who belong to a later born group. Negative factors are long-term smoking and female gender. According to Beckman N., the

correlation between the nature of relationships and physical activity is particularly visible in older women [13]. Among seniors in West Africa, researchers from the *Human Sciences Research Council* have described a strong correlation between body well-being and/or the nature of relationship and sexual activity. The authors of this report concluded that as the age of the subjects increases, their activity decreases and that single seniors have very low chances for sexual behaviour. Other factors listed in the *South National HIV Incidence, Prevalence, Behaviour and Communication Survey* (SABSSM) are chronic diseases, HIV exposure and exercise [14].

Due to slightly different cognitive character of this paper, not all variables were listed in the above paper. The main aim was to show the sexual behaviour of seniors from Gdansk. In the authors' opinion, the correlation between the increase in the number of classical sex and the increase in choice of oral technique during intercourse deserves attention. Ladies who choose oral sex usually also try anal sex. This may indicate that seniors who have sex are looking out for and trying new sensations.

According to the authors, the biggest problem in studies on the sexuality of sixty-year-olds and older women, even the study group (apart from coexisting diseases, of course) are men, namely their statistically shorter lifespan, which makes it very difficult for a woman to find a partner – of the same age – when she becomes a widow after 60 years of age.

In Poland and in the world women live longer than their partners, thus it can be assumed that an older man is more likely to have a partner than a peer in his age. This causes many problems in research, because it is necessary to note that it is difficult to characterize the sexual behavior of a senior females, who usually are already lonely. According to the research results conducted by Osmo Kontuli, the author of the FINSEX research project (research sample of 1019 women), in Finland seniors over 60 years of age are more often lonely than men. Less than half of 70 year old Finns have a husband or a permanent partner [15]. In England the situation seems to be similar, with 40.6% of women over fifty faced with loneliness. The observed difference deepens with the age of respondents – 72.2% after eighty years of age have no partner [10]. In Spain 48.22% of women over 65 years of age are lonely [11]. In South Africa based on the SABSSM analysis, a similar trend was observed where 51% of women over 50 years of age are without a partner [16]. According to the *Global Study of Sexual Attitudes and Behaviors* (GSSAB) in South Korea, 23.7% of women have no partner [16]. In accordance with the report of the Central Statistical Office in Poland, only 34% of women in partnerships or marriages live in the whole group of over sixty-five-year-olds [G]. The research conducted

by the author confirmed this trend – only 22% of the respondents over 60 years of age are in a relationship.

Another factor presented by the respondents during an interview after the survey is a negative public perception, i.e. a single woman, and even worse, a widow over 60 years of age is not suitable for dating. She should sit at home and help to take care of her grandchildren.

Another factor is the respondents' own thinking about intercourse and sex with a non-permanent partner. Most of the respondents did not tolerate such behavior, explaining themselves by ideological reasons, conscience and faith. The ladies stated that they do not want to get close to many men, but they long for intercourse with a permanent partner. The ladies confirmed Izdebski's opinion – Polish men up to the age of 49 have more traditional views on sex in marriage and women's sexual needs, but they are liberal in terms of having more partners. Do physicians ask about sexual problems?

It is well known that the relationship between medical personnel and patients is not easy. If a sexual problem arises, there are even greater barriers in communication resulting from shame, lack of willingness and knowledge of both sides. In 2004, a survey was conducted under the patronage of the Polish Sexological Association on problems with erection, on the basis of which it was concluded that almost 75% of family physicians, psychiatrists or diabetologists refused to participate in the survey, indicating that they are unwilling to ask such questions to patients. The majority of family physicians, as much as 86.5% do not want to discuss this issue, whereas urologists (84.2%) and sexologists (89.5%) discussed such issues with their patients [17]. In South Korea, only 5.2% of adults were asked by their physician about likely disorders. In East Asia, only 4.8% of patients, while in the West – 14.9% [16]. Our own research shows that 92% of seniors did not address intimate issues with their physician. More than half of the respondents (67%) are of the opinion that their knowledge about sexual disorders is sufficient and almost 3/4 (71%) does not want to talk to their physician about it.

What is the deadlock in conversations?

It is possible due to the effect of a cohort (peer group) – i.e. when a neighbor of the same age feels educated, I should also feel educated. He doesn't want to talk to his physician? I won't either.

The next barrier may be the physician's gender, it is commonly known that older women prefer the same gender of medical personnel.

Other factors may be society and educational culture, because in some circles sexual behaviour is simply not talked about.

In conclusion, physicians are not able to talk about sexual issues with their patients. Moreover, women are not interested in such a conversation either.

CONCLUSIONS

1. Over half of the women over 60 years of age who participate in the meetings of the Gdańsk senior clubs lead a sexual life.
2. The respondents presented a wide range of sexual behaviors from holding hands to sexual contacts.
3. Sexual contacts are also varied. The surveyed seniors most often have classical sex followed by oral sex, and anal sex.
4. There are clear, relevant, and almost practical dependencies between the selected parameters.
5. Most of the respondents were not asked by their physician about the sexual sphere, with almost 1/3 of the respondents expressing their desire to talk.
6. Over half of the surveyed seniors claim that their knowledge on sexual problems is sufficient.

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Can Alzheimer's disease be prevented? Selected examples of prevention

Urszula Porycka, Katarzyna Szmuc, Dominika Cuprian, Piotr Modelski, Martyna Zimoch

Medical University Piastów Śląskich of Wrocław, Wrocław

INTRODUCTION

Alzheimer's disease (AD) is a neurodegenerative disorder that clinically manifests itself as a gradual loss of memory and cognitive function, which ultimately leads to a complete picture of dementia [1-4]. AD is the most common neurodegenerative disease. Its degenerative character makes its symptoms appear about 20 years after the initiation of the disease process [1]. The primary cause of the disease is unknown, which makes it impossible to apply a therapeutic effect [2]. It is assumed that the neurodegenerative process is multifactorial and the essence of observed changes is the aggregation of pathological proteins in the brain. Intracellular deposits of beta-amyloid are deposited and in the extracellular space neurofibrillar plexuses with hyperphosphated tau protein are accumulated [5-8]. The course of the disease is relatively fast and leads to psychophysical disability. Ultimately, the disease is fatal [1,4].

EPIDEMIOLOGY

There is an increase in the incidence of dementia diseases in highly developed countries with ageing populations [6]. The age-standardized incidence of dementia ranges from 5 to 7 percent in most developed countries [5]. Alzheimer's disease is assumed to account for 60-70% of all dementia [4]. Considering the overall US population, one in 10 people aged 65 and over suffer from Alzheimer's disease [1]. It affects 5-10% of people over 65 years of age and up to 50% of people over 85 years of age [4]. In the world about 30 million people are affected by Alzheimer's disease, in Poland it is 200 thousand. In 2050 the number of patients is estimated to triple [2]. Gender differences in the prevalence of the disease show that more women than men suffer from Alzheimer's disease [1]. After 80 years of age, the disease is diagnosed more often in men [6]. Studies have shown that African-Americans and Hispanics as ethnic groups are more predisposed to the disease than others [1]. Currently, Alzheimer's disease is officially

recognized as the sixth leading cause of death in the United States. It is mentioned as the fifth most common cause of death of people aged 65 years and older. The disease is becoming an increasingly common cause of death, which is closely related to the process of population ageing. The mortality rate from Alzheimer's disease is 37.3 deaths per 100,000 people [1].

RISK FACTORS

One of the main risk factors for AD is age, but it should be remembered that AD is not a normal part of ageing [1]. Apart from age, the most certain risk factors of Alzheimer's disease are family history towards dementia, hereditary mutations in genes affecting amyloid in the brain and apolipoprotein e4 allele (APOE 4) [5].

Early form of Alzheimer's disease is highly hereditary. Late onset Alzheimer's disease has a more complex genetic basis and is also influenced by the environment, so it has a multifactorial etiology [1,5]. Since biochemical changes in the brain occur long before the clinical signs of Alzheimer's disease, the environmental impact on neurodegeneration requires long-term studies [5].

FAMILY HISTORY

Patients have a 10-30% increased risk of developing the disease if a first-degree relative is affected by dementia. Those whose siblings suffer from late onset Alzheimer's disease have a three times higher AD risk than the general population. Studies show that the risk is similar to the control group if the disease develops in a relative later in life [5]. However, a family history of Alzheimer's disease is not necessary to develop the disease [1].

GENETIC PREDISPOSITION

Mutations that occur in the early form of AD are very penetrating, which means that vectors have an almost 100% chance of developing the disease during their lifetime [5]. They include mutations of 3 specific genes - the amyloid precursor protein gene APP and the preseniline 1 and 2 gene, which results in an increase in the level of toxic beta- amyloid forms [1,3].

The APP gene is encoded on chromosome 21. The presence of an additional copy of chromosome 21 which takes place in Down's syndrome may increase the production of beta-

amyloid produced in the brain. Trisomy in Down's syndrome is therefore associated with an increased risk of disease development. It is estimated that approximately 30% of people suffering from Down's syndrome at the age of 50 have AD [1]. The genotype with the apolipoprotein allele APOE e4 is associated with increased risk of cardiovascular diseases and AD.

APOE e4

Individuals with apolipoprotein epsilon 4 form are more likely to develop AD and cardiovascular disease than the control group. It should be noted that this means an increase in potential risk rather than certainty that the disease will develop. Recent studies show that Afro-Americans have at least one copy of the APOE e4 allele. This ethnic group can therefore be defined as a group of higher risk of the disease [1].

HYPERTENSION

Cerebral vascular diseases and AD often coexist. Cerebral small vessel disease is common in the elderly, and co-occurrence of vascular disease and AD is referred to as mixed dementia. Peripheral vascular atherosclerosis contributes to the risk of AD and dementia. In its assessment, measurements of the thickness of the internal jugular membrane and calcification of the coronary artery can be used [5]. There are many similar risk factors for both cardiovascular diseases and dementia [1]. Smoking, hypertension, obesity and diabetes are all factors that may increase this risk. Studies have confirmed a reduction in the incidence of MCI in intensive pressure-lowering treatments [1].

DYSLIPIDEMIA

An important predictor of AD may be the cholesterol mechanism. Epidemiological studies have shown a link between total or low LDL-C lipoprotein cholesterol levels and the risk of AD especially in middle age, but the interpretation of these studies is complex. The blood-brain barrier does not allow the penetration of LDL-C of peripheral blood unless it is damaged by vascular diseases, among others. The cholesterol that occurs in the brain is synthesized by astrocytes and neurons. High-density lipoprotein complexes (HDL) allow the delivery of cholesterol to the brain, so there is little or no LDL-C or LDL-C at all. In addition,

elevated total cholesterol and LDL-C have been shown to correlate with a decrease in cognitive function in patients with AD. The effect of cholesterol on the deposition of beta amyloid and other factors such as local inflammation or tau protein metabolism is also suggested [5].

DIABETES MELLITUS

Studies have shown that 1.5 higher AD risk is found in people with type 2 diabetes or obesity. This is related to hyperinsulinemia and insulin resistance in the brain and the potential interaction between insulin and amyloid.

TBI

TBI- traumatic brain injury has been found to increase the risk of dementia, including dementia in Alzheimer's disease [1].

ENVIRONMENTAL RISK FACTORS

Identified AD environmental risk factors are passive smoking, air pollution and pesticides [8].

PREVENTIVE FACTORS

The positive effects of physical activity, which reduces the incidence of cognitive impairment and dementia including AD, are supported by research. A meta-analysis of 16 prospective studies showed a 28% reduction in general dementia and a 45% reduction in AD among physically active individuals compared to those who are less active. Social, mental and physical activity reduces the risk of AD and other dementias and can therefore be considered as a form of prevention [1,5].

It has been supported by strong evidence of AD risk reduction through a healthy diet and lifelong learning [1,5]. Scientists believe that the number of years of education allows to build up a 'cognitive reserve' meaning to adapt in some way to changes such as beta- amyloid and tau protein accumulation. The building of such a reserve may also be facilitated by having mentally stimulating work [1].

PATOMECHANISM

The disease process taking place in AD is a gradual and irreversible disorder of homeostasis. A disturbed control system of the inflammatory process in the brain affects the degeneration of its structure and changes in the functioning of the whole organism [8]. The theory of the amyloid cascade determines the sequence of events initiated by the accumulation of insoluble forms of beta peptide, eventually leading to the death of nerve cells which manifests itself in clinical symptoms of AD [6]. The accumulation of beta- amyloid platelets outside the neurons and pathological form of tau proteins inside the neurons are only one of many changes taking place in the brain during the course of the disease. Moreover, calcium ion homeostasis is disturbed, activation of inflammatory factors such as cytokines, reactive oxygen radicals or nitric oxide. Hyperactivation of microglia and astrocytes leads to neuronal apoptosis and damage to the blood-brain barrier. The immunocompetent cells start to accumulate in the brain, which leads to local inflammation [8].

Amyloid beta is a product of enzymatic degradation of an integral membrane protein - a amyloid precursor protein APP. The neurotoxicity of beta amyloid consists, among others, in calcium homeostasis disorder, interaction with cell membrane lipids and activation of specific receptors. PS1 and PS2 proteins are involved in APP proteolysis, occurring mainly in nerve cells, where they function as membrane receptors or calcium channels. Mutations within them lead to the formation of beta amyloid peptides.

Hyperphosphorylated protein tau- is the main component of neurofibrillary tangles NFT observed in the most frequently damaged brain regions in AD [2]. The tau protein is bound to microtubules, helps in their formation and stabilization. In AD, it undergoes hyperphosphorylation and aggregates becoming the main component of neurofibrillar plexuses [5].

Beta amyloid deposits can cause cell death and disrupt neuronal communication in synapses. Pathological forms of tau protein block the transport of nutrients and essential molecules into the neurons. In addition to changes associated with beta amyloid and hyperphosphated tau protein, changes in the brain include inflammatory reaction and atrophy of nerve cells. The immune system is activated - microglycer cells try to remove toxic proteins and dead cells. When the performance limit of the microgleules cells is exceeded and they cannot keep up with their function effectively, an inflammatory process develops. Due to the loss of cells, the brain gradually disappears. Parallel to these changes, the cerebral metabolism of glucose, its main fuel, is impaired [1].

APOE 4

The presence of APOE e4 allele is responsible for intensification of amyloidogenesis processes. The presence of 2 copies of the APOE e4 allele 16 times increases the risk of chA. Elevated cholesterol concentration in the membranes of nerve cells results in increased aggregation of the amyloid produced and induces hyperphosphorylation of tau protein [2].

THE ROLE OF THE IMMUNE SYSTEM IN ALZHEIMER'S DISEASE

Recent studies have highlighted the role of the immune system in the etiopathogenesis of AD. It is believed that the markers of inflammatory reactions or disturbances of immune response processes can be used to monitor the course of disease [2].

At present, the involvement of neuroimmune mechanisms and inflammatory process in the pathogenesis of AD is certain. One of them is "Inflammaging", which means chronic inflammation present during ageing. It is characterized by elevation of markers such as C-reactive protein or interleukin 6 IL- 6. There are many factors that are the source of this condition, which include: accumulation of residues from damaged cells, aging cells and factors produced by them (pro-inflammatory cytokines), overactivity of the clotting system or a decrease in immunity. Microgel is the first line of defence; it has phagocytic properties, releases cytotoxic factors and can act as an antigen-presenting cell. Its proper functioning plays a very important role in maintaining CNS homeostasis. There is a hypothesis about the causes of AD development depending on disturbances in the regulation of the mutual metabolism of microglycer cells. Pathological forms of astrocytes - the most numerous cells in the nervous system performing various functions are also considered pathology in neurodegenerative diseases. Elevated levels of proinflammatory cytokines such as: IL1, IL-6, INF- gamma and TNF- alpha correlate with AD progression. Numerous studies indicate that infections may be responsible for the initiation of neurodegenerative processes in the brain. An example is the reactivation of HHV-1 herpes virus infection. In the early stages of AD, the same areas of the brain were observed to be occupied during the infection characterized by demyelination [10].

At present, we do not know the source of the disease and questions about its cause are constantly asked [9].

It is suggested that amyloid deposits activate the soil cells, which through the complement system causes synapses to be cut. The literature claims that the knowledge of the

biological basis of mechanisms responsible for the loss of synaptic connections in AD is crucial for effective therapy [11].

Recent studies have pointed to the effects of intestinal microflora on brain function. The aging organisms have more and more poor flora and disturbances in the composition of the microflora are associated with the neurological condition. The changes in the composition of the microflora, which coexist with an increase in intestinal permeability observed with age, result in the translocation of microorganisms or their components, followed by the CNS inflammation. The study of the intestinal-brain axis of the microflora in AD still requires a lot of research, however, the data so far suggests modulation of the intestinal microflora through dietary components or probiotics, which can be used to counteract the progress of neurodegeneration. So far, there is no effective cure for AD [12].

Despite the use of many research models, it is still difficult to present precisely how the neurodegeneration process is progressing and thus to answer the question about possible ways to slow down the process. The discovery of mechanisms remains a challenge and encourages interdisciplinary research [8].

CLINICAL PICTURE

Early changes in AD are compensated for and the patient functions normally. However, when the compensatory capacity is exceeded, patients reveal a decrease in cognitive function. A characteristic feature of the disease is progressive memory loss combined with cognitive process disturbances [1]. Early changes in AD are often confused with age-related changes. In the last stages of the disease, however, the patient loses contact with the environment and requires constant care due to failure to cope with basic activities [8].

There are 3 stages of Alzheimer's disease:

1. preclinical Alzheimer's disease : present biomarkers, no development of symptoms or subjective memory disorders, not all people will develop a fully symptomatic disease,
2. mild cognitive impairment MCI due to Alzheimer's disease :
present biomarkers, greater than expected for age decline in cognitive function, changes at this stage may be noticeable for family, friends
3. dementia due to Alzheimer's disease: disturbances impairing the ability to function normally in everyday life [1,6].

Signs of Alzheimer's or other dementias

Memory loss that disrupts daily life
Challenges in planning or solving problems
Difficulty completing familiar tasks at home, at work or at leisure
Confusion with time or place
Trouble understanding visual images and spatial relationships
New problems with words in speaking or writing
Misplacing things and losing the ability to retrace steps
Decreased or poor judgment
Withdrawal from work or social activities
Changes in mood and personality

A long asymptomatic period is characteristic of AD. Initially, declarative memory will be disturbed, which is responsible for learning new facts, then late declarative and episodic memory. Planning, visual and spatial orientation and speech as well as praxis and gnosis begin to cause difficulties. Psychopathological symptoms may develop in advanced stages of the disease. These include psychotic symptoms, emotional disorders, agitation and aggression, abnormal motor behaviour, sleep disorders, personality changes. There are speech disorders of the amnesia aphasia type with paraphasias and semantic impoverishment of speech. Apathy in AD is often confused with depression. Aggression, which is a common problem, makes it difficult to take care of the patient. Mood and emotion disorders usually manifest themselves as depressive syndrome and anxiety disorders. We may encounter the symptom of wandering quite difficult for those taking care of the patient [6]. In some patients with MCI, the cognitive disorders may remain at a similar level for many years, whereas in about 50% of them they are a sign of later dementia, mainly Alzheimer's disease and vascular dementia. Frontal deficits can lead to impairment of skills such as planning, solving complex problems, using current information or understanding and orientation. They are also related to the impairment of the so-called plasticity of mental processes, which prevents AD sufferers from coping with everyday situations and adapting to changing environmental conditions. Contemporary research clearly indicates that depression in the elderly may be a prediction of the dementia process and draws attention to the possibility that depression may be associated with cerebral changes in the same structures that may be the biological cause of both depression and

dementia, i.e. in the frontal lobe and limbic system. The analysis of clinical trials shows that the risk of depression episode in the course of ChA is 20-25%. Other sources estimate the risk of the disease from 0 to 86%, usually in the range 30-50%, and are currently working on determining whether a specific genotype could be a marker of depression in the course of AD [4]. Complications of severe dementia can be stillness, swallowing disorders and malnutrition. They increase the risk of severe acute death threats in patients. An example is pneumonia, which is a leading cause of death among elderly people with dementia.

LIFESTYLE

A wide range of lifestyles affect our physical and mental health, and they are also important for our well-being. There is growing evidence that some lifestyle-related factors are linked to the development of Alzheimer's disease. Many of these are potentially modifiable and include smoking, alcohol consumption, physical activity, education, social engagement, cognitive stimulation and diet. Modification of most of these factors has many health benefits. By increasing the potential benefits of modifying a person's lifestyle, we can reduce the risk of contracting AD [14]. The way and quality of nutrition are important environmental factors influencing the human body regardless of gender, age or coexisting diseases. Proper nutrition is an important element of lifestyle, playing an important role in maintaining the homeostasis of the organism. The quantity and quality of food consumed is an important element in the prevention of non-communicable, chronic civilization diseases, whose frequency increases with age. These diseases include neurodegenerative diseases, among them AD [15,16]. Great importance is attributed to the Mediterranean diet as well as the MIND diet. The authors also specify single nutrients or products that may affect the prevention of neurodegenerative process development.

THE ROLE OF THE MEDITERRANEAN DIET AND THE MIND DIET

The Mediterranean diet is widely recognized as a healthy diet. This diet is rich in vegetables and fruit, legumes, olive oil, whole meal bread, fish and seafood. The Mediterranean diet takes into account reduced consumption of red meat, dairy products, and sweets. Thus, this diet is a source of all necessary nutrients [17,19]. In the Mediterranean diet, its overall nutrition model is important, not only individual products [19]. The improvement in the intake of products included in the diet may directly affect the occurrence of AD, through the

consumption of foods rich in antioxidants and low in cholesterol, and indirectly through the prevention of other AD risk factors, such as high blood pressure, high cholesterol, and diabetes [20]. In the traditional Mediterranean diet, the share of energy derived from fats was about 30% but only 7-8% of energy came from saturated fatty acids. The main source of fats was oil, which has a high content of monounsaturated fatty acids (MUFA), which has a beneficial effect on the blood lipid profile, reduce glucose concentration and insulin demand. Equally significant are polyunsaturated fatty acids, which show anticoagulant, anti-inflammatory and hypotensive effects [17]. The pro-healthy effects of diet are attributed to the consumption of oil, which causes the regression of atherosclerotic plaques. This regression may contribute to slowing the progress of Alzheimer's disease. The essence of slowing down the neurodegenerative process is also attributed to the presence of large amounts of antioxidants, fiber, and anti-inflammatory omega-3 fatty acids, which are present in the Mediterranean diet [17,19]. The influence of the Mediterranean diet on neurodegenerative diseases is discussed; however, most epidemiological studies emphasize its role in reducing the risk of AD development [16,18]. The literature contains many papers confirming the influence of the Mediterranean diet on cognitive function preservation [19]. The observations confirm that among people following the principles of the Mediterranean diet, the risk of development of cognitive disorders is lower by 28% and the risk of Alzheimer's disease is lower by 48%, compared to people following other diets [18]. It has also been noted that the diet used to lower blood systolic blood pressure (DASH) prevents cognitive dysfunction in patients with hypertension and obesity. For this reason, a modification of the Mediterranean diet and the DASH diet, which was called the MIND diet, were created. This diet places great emphasis on natural food of plant origin, especially promoting increased intake of berries and green leafy vegetables, with limited intake of animal food and strongly saturated fats [20]. The diet distinguishes 10 products that are desirable and positively affect the brain: green leafy vegetables, other vegetables, nuts, berries, beans, whole-grain products, fish, poultry, olive oil, wine [21]. Especially from the group of green vegetables, kale, spinach, lettuce, cabbage and berries are distinguished as a separate product due to high content of antioxidants compared to other vegetables [19]. The group of harmful products included: red meat, butter and margarine, cheese, cakes and sweets, fried products and fast foods. Additional general guidelines for the MIND diet include consumption of at least three portions of whole grains, lettuce and one other vegetable as well as a glass of wine every day. Nuts should be eaten as a snack most days and beans every other day. Poultry and berries are recommended at least twice a week, and fish at least once a week [21]. The main studies on the effect of the MIND diet on cognitive

dysfunction were conducted by Morris M. as part of the *Rush Memory and Aging Project*. Slower decrease of cognitive functions was observed in the highest MIND dietary indexes. MIND diet index results correlated with all domains of cognitive function. However, a particular effect of this diet based on the results can be distinguished in episodic memory, semantic memory and rate of perception. A high MIND index reduced the risk of progression to dementia by about 10% compared to the whole group [19,22]. Another study by Morris M. showed that the subjects with the highest MIND index (tercel 8.5-12.5) had a 58% lower risk of Alzheimer's disease compared to those with the lowest index (tercel 2.5-6.5). From the presented studies it can be concluded that high adherence to the Mediterranean diet reduces the risk of AD, but adherence to the MIND diet already significantly reduces this risk [20]. The answer to the question of whether the diet will be an effective method in the prevention of dementia diseases still requires many studies; however, the presented results are promising [19].

THE ROLE OF B VITAMINS AND HYPERCYSTEINEMIA

B vitamins play an important role in the correct metabolism of homocysteine amino acids. This amino acid is an indirect link between exogenous methionine consumed only with proteins in food and endogenous cysteine. The second metabolic pathway of homocysteine is methylation into non-toxic methionine. The donor of the methyl group is folic acid and the cofactor of methionine synthase (MS) is vitamin B12 (cobalamin). Elevated concentration of homocysteine is associated, among others, with decreased plasma concentration of B group vitamins (B12 and folic acid) [23]. In the initial, randomized controlled study involving elderly people with increased risk of dementia, it was demonstrated that treatment with high doses of vitamin B (folic acid 0.8 mg, vitamin B6 20 mg, vitamin B12 0.5 mg) slowed down the contraction of the entire brain within 2 years. Another study revealed that the treatment with vitamin B reduces the brain atrophy as much as seven times in these grey matter regions particularly sensitive to the AD process, including the central temporal lobe [24].

CHOLESTEROL

The brain is the richest organ in cholesterol; with only 2% of body weight it contains about 20% of cholesterol in the body. About 70% is found in myelin, and about 30% is metabolically active and is found in the membranes of glial cells and neurons, where it is

recycled mainly to repair and rebuild neurons. Cholesterol in the brain is synthesized from acetate in situ. Basically, there is no cholesterol to get into the brain from the peripheral circulation [26]. However, epidemiological studies have demonstrated an increased risk of dementia in individuals consuming large amounts of foods containing saturated fatty acids, trans-fatty acid isomers and cholesterol [25].

Research shows that cholesterol is consistently associated with AD. Despite a reduction in cholesterol levels in healthy aging brains, lipid analysis of AD patients' brains reveals increased sterol levels in sensitive regions. Moreover, retrospective epidemiological studies show that middle-aged hypercholesterolemia is a risk factor for AD. There are suggestions that cholesterol may also regulate the removal of A β . The faster delivery of A β to lysosomes and increased degradation have been described in the microglia after lowering the cell cholesterol level; however, molecular mechanisms are still not fully explained [27].

Recent evidence strongly suggests altered sterol homeostasis in AD etiopathogenesis. The disease is associated with aging, characterized by progressive degeneration of neurons, gliosis and accumulation of intracellular inclusions. There was no clear correlation between blood cholesterol levels and AD, although a relationship between systemic cholesterol metabolism and susceptibility to AD was suggested [26,28]. Plasma and cerebrospinal fluid 24-hydroxycholesterol concentrations are significantly higher in patients with early AD and vascular dementia compared to age-matched and dementia-free controls. This is also suggested by statins, which in fact reduce the risk of AD [28]. Several possible mechanisms could explain the effects of high cholesterol levels on AD development. Cholesterol may increase the activity of β or γ secretion enzymes, which produce A β from APP, decrease the flow of APP (amyloid precursor proteins) through the non-amyloid pathway of α -secretase or affect various non-amyloid factors, such as local inflammation or tau metabolism [29].

Nowadays, there are only two types of licensed drugs that are used for symptomatic treatment of Alzheimer's disease. Both, acetylcholinesterase inhibitors and NMDA receptor antagonist (memantine), were developed in the late 1980s'. Since then there are a lot of trials for regulatory approval of new drugs. Unfortunately all of them fail.

US Food and Drug Administration (FDA) and the European Medicines Agency (EMA) require proof of efficacy on two endpoints for every newly tested medicine. The first of them is global cognition for both institutions and global clinical change for the FDA or activities of daily living for EMA [30].

All currently licensed drugs for AD were tested in at least two 6-months independent clinical trials. Results must prove an advantage over placebo on both of these endpoints [30].

Most of them are currently targeting the cholinergic system, $\alpha 7$ nicotinic acetylcholine receptors (nAChRs), γ -secretase modulators, β -secretase, tau, *N*-methyl-D-aspartate receptor (NMDAR), inflammatory mediators and glucagon-like peptide-1 (*GLP-1*). Nevertheless the high failure rate is the major problem of this studies. Researchers need new methods of drug efficacy testing, technics, strategies and targets [31].

I want to present recent observations related to AD's prevention and treatment hereunder. Scientists were looking for connections between lipid-lowering drugs and AD risk according to vascular factors in its pathogenesis. They developed theory saying that antidiabetic medication may slow the formation of the amyloid plaque and divided drugs into four groups: statins, PCSK9 inhibitors, ezetimibe and mipomersen. No relation between lipid-lowering drugs and AD risk was proved. Results showed neither clinical effect nor damage. Moreover PCSK9 inhibitors increased this risk. In conclusion, lipid-lowering drugs protect from coronary artery disease but there is no preventive activity with AD [32]. Other, prospective trials also gave only inconclusive results [33].

Other studies were focused on the main cause of AD which is amyloid- β aggregation. Passive immunization with monoclonal antibodies was analyzed. Immunotherapy seems to be the best possible strategy among approaches pointed at A β . First as well as every alternate tested monoclonal antibody were generally well-tolerated and save. However all the trials were terminated because of no clinical effect. Maybe higher doses of drugs are required or the wrong A β species were targeted. Testing these theories are the most important aim of ongoing trials. A similar story relates to tau aggregation which is another important treatment target. Unfortunately early clinical phases also failed [34].

There are some promising conception concerning prevention and treatment AD. Raloxifene, a selective estrogen receptor modulator, impact on AD pathogenesis in postmenopausal women. This mechanism isn't totally clear this day. The last research showed that raloxifene slows A β aggregation and destabilizes already formed A β plaques due to interaction with certain domains of A β peptides. Moreover, the drug helps to reduce the expression of TNF α and reduces the indirect toxicity of A β to neuronal cells. This is the possible explanation for the neuroprotective function of raloxifene in AD pathogenesis [35].

In respect of the plausible relationship between AD pathology (inflammation, glial cell activation and A β deposition) and change in redox status it developed galantamine research. Galantamine, an acetylcholinesterase inhibitor, probably has oxidative stress inhibitory function. During the trial the drug was administered orally to mice before showing up A β plaques (preplaques phase). In results galantamine reduced production of proinflammatory

cytokines and equilibrated redox state. This test suggests that galantamine could be useful as one of a few drugs for the prevention of AD [36].

One of the latest research allowed us to identify four (A01-A04) novel scaffolds AChE inhibitors thanks to virtual screening. They assisted in collecting brand new information for future modification. Moreover pathophysiology mechanism of AD such an A β aggregation and neuroprotective activity against nerve cell injury caused by A β were well investigated. Two of these scaffolds A01 and A03, turned out to be able to protect cells against A β -induced injury. This work has a chance to groundbreaking. Four new skeletons of selective AChEIs could be the key to the invention of a new anti-AD drug [37].

In another trial, there were used biomimetic nanochaperon inspired by natural HSPs. They are built from the biodegradable mixed-shell polymeric micelle (MSPM). They are expected to suppress the progression of AD. Research revealed that MSPM manifests neuroprotective activity through decreasing inflammation and A β accumulation. It could give remarkable effects in treatment early stage of AD or even be a prophylactic treatment strategy [38].

Beyond all drug researches mentioned above there are a lot of studies relating to dietary habits. Most of them regarded multiple supplements such as multi-ingredient supplements, soy, B vitamins, vitamin C or β -carotene, ginkgo biloba, vitamin E, bile acids, folic acid, *etc.* They were used independently or in addition to the various clinical trials. Unfortunately, none of them didn't bring forward as they were expected. All in all there isn't adequate evidence to advise any of these supplements for cognitive protection [31].

Other groups of these studies focused on ω -3 fatty acids, docosahexaenoic acid and dehydroepiandrosterone (DHEA) [39,40]. The participants were divided into four groups and took these supplements in different ways. Unfortunately, this experiment showed that this kind of supplementation doesn't bring any benefit. Furthermore, groups that were administered ω -3 fatty acids and DHEA get results significantly worse than the placebo group in a cognitive test [39]. The crucial role of ω -3 fatty acids and docosahexaenoic acid in the pathogenesis of AD and the aging brain isn't still known enough [41]. All biological mechanisms and risk factors are still unknown and not well understood. Probably this is the explanation of these failures. Changing endpoints in most studies also could help to achieve more satisfying and pertinent outcomes [42].

There was also a trial examined collection between moderate alcohol intake during the lifetime and AD pathophysiology mechanism. Researchers came to a surprising conclusion. Cerebral A β accumulation, neurodegeneration of brain cells and cerebral white matter

hyperintensities (WMHs) were measured. The study revealed that lifetime alcohol intake significantly decreased the level of amyloid deposition compared to no drinking. Nevertheless, alcohol didn't give neuroprotective activity - cerebral white matter was injured. That conclusion is arguable. There isn't enough evidence to settle if lower A β accumulation arises from lifetime alcohol intake or just protective mechanisms in the human brain [43].

There are some likely reasons for the unclear relationship between diet and development of AD. Firstly there is no consistency of methods, criteria and outcomes used for evaluation of results. Additionally, some factors which may increase or decrease the risk of AD (such as APOE 4 status, cardiovascular risk or physical activity) aren't taken into consideration. Finally, there is broad diversity among study designs (cross-sectional, case-control, prospective and randomized controlled trials) and length of follow-up.

Appropriate diet habits and way of life seem to be an easy, useful and inexpensive technique to slow down the advancement of AD. There is why relevant and complete therapeutic approaches are pressed for [44].

INFLUENCE OF PHYSICAL EXERCISE ON THE OCCURRENCE AND DEVELOPMENT OF AD

It is commonly known that regular physical activity and doing a properly selected physical training helps to maintain physical function for a longer period of time and also has a beneficial effect on the body, affecting many organ systems. Currently, it is known that physical exercises are one of the forms of prevention of different diseases, especially those affecting the cardiovascular system. However, numerous scientific researches indicate that physical exercises can also protect from developing dementia – for example in Alzheimer's Disease – and can slow this process in patients affected by the disease.

Caspersen defined physical activity as any movement of the body produced by skeletal muscles which leads to energy expenditure. Exercise was defined as a subset of physical activity that is planned, structured, repetitive and its final or intermediate goal is to improve or maintain physical fitness [45].

In the document entitled 'Global recommendations on physical activity for health' the World Health Organization recommends adults aged 65 years old and above 'at least 150 minutes of moderate-intensity aerobic physical activity throughout the week or at least 75 minutes of vigorous-intensity aerobic physical activity throughout the week' [46].

Everything that causes poor blood circulation (high cholesterol levels, arterial stiffness, atherosclerotic plaques in arteries, long periods of inactivity) leads to worsening of intellectual ability, especially the activity of the middle part of the frontal lobe responsible for short-term memory. In contrast, factors enhancing blood circulation (for example aerobic exercises) improve the health of the brain and the whole body [47].

The positive impact of physical activity on mental functioning is proved, inter alia, by the results of long-term research which has been conducted since 1948 including 3 generations of patients. It showed that a daily fast walk is correlated with a 40% decrease in the risk of developing Alzheimer's Disease and other types of dementia in old age [48]. Besides, the researchers from Pittsburgh proved that the elderly people, who are used to walking, have greater hippocampal volume and better cognition test scores [49].

The effects of aerobic training on the brain were proved by scientists from Wake Forest University in 2016. They compared the results of intensive exercises and stretching exercises among the people with mild cognitive impairment. The research participants completed the exercise for 45 minutes, 4 times per week for 6 months. In the group consisting of the participants with high-intensity training, increased blood supply to the frontal lobe (responsible for organization and planning) was detected, as well as greater brain volume, better-developed executive functions and stronger protection from cognitive impairment despite a considerable genetic predisposition to Alzheimer's disease. In the group of the participants doing stretching exercises, the findings connected with the progression of dementia were detected: the brain atrophy and executive functions impairment. On the basis of this study the conclusions were drawn that aerobic and intensive exercises are the most effective ones [50].

As a result of the aging process, neurons and significant connections between them are being lost. However, there is evidence proving that aerobic exercises can increase synaptic connections in the brain. The greater number of them makes the brain functioning more efficient and protects from dementia, especially from Alzheimer's disease.

Regular physical activity may be implemented both as prevention of dementia and a way to slow down the development of the disease. Meta-analysis studies revealed that regular physical activities are correlated with a 30-40% decrease in the risk of development of Alzheimer's disease in comparison with physical inactivity. Physical exercises result in the release of growth factor BDNF (brain-derived neurotrophic factor) in the brain. BDNF is essential for the proper functioning of the hippocampus, nerve cells survival, neurogenesis and synaptic plasticity. Moreover, BDNF supports the process of learning by means of modulation of synaptic changes which are necessary for long-term potentiation [51]. Physical activity

induces also other systemic neurotrophic factors. These molecules may cross the blood-brain barrier and may protect against neurodegenerative disorders like for example Alzheimer's disease. Identifying exercise-induced systemic neurotrophic factors with beneficial effects on the brain could lead to new molecular targets in order to maintain cognitive function and prevent the neurodegeneration process [52].

The effect of aerobic training has been studied the most accurately so far but resistance training (focusing mainly on muscle development with the use of weights) also has a beneficial effect on the functioning of the brain. Particularly strong legs are linked to intellectual ability because leg muscles support blood circulation, which then reaches the brain.

The importance of resistance training was proved in the study of scientists at the British Columbia University in Canada, which was carried out on the group of elderly women. It showed that doing resistance exercises reduces the number of damage to the white matter and improves the ability to concentrate [53]. Another study showed that resistance training enhances mental performance better than stretching and toning exercises. In addition, resistance exercises improve the ability of concentration and logical thinking (the frontal lobe is responsible for these functions) more effectively than they improve long-term memory (for which the temporal lobe and the hippocampus are responsible) [54].

The research at the Florida University revealed that the growth factor BDNF blood level demonstrated a 98% increase after each training session in adults doing resistance training [55]. Resistance exercises also contribute to the health improvement of the vascular system and condition of the arteries, which results in an increased supply of nutrients to the brain. Additionally, it relieves inflammations because, after a few months of such training, there is a decrease in the level of homocysteine. Homocysteine is a substance that leads to inflammation and blood vessels damage associated with it.

According to the studies, implementing the combination of aerobic and resistance exercises increases brain efficiency and slows down the deterioration of cognitive functions or dementia development more efficiently than doing only aerobic exercises [56].

Using a desk with the purpose of working in a standing position to increase activity in the office also appears to be a good idea in order to implement dementia prevention. In a standing position more calories are burnt than while sitting down. Moreover, the legs muscles can be strengthened, which is essential for maintaining healthy vascular system [47].

As for chronic diseases that reduce activity and make the heavy lifting impossible, it is worth to choose less intensive and lighter exercises such as training on the bicycle, in the swimming pool, yoga or ballroom dance.

As part of the aerobic exercises, it is worth trying out: quick walk, stationary cycling, stepper exercises, dancing, walking on stairs or martial arts. Examples of resistance training include: squats, bending exercises, sit-ups, exercises with dumbbells and push-ups.

In conclusion, physical exercises appear to improve neurogenesis, brain blood flow, increase hippocampal volume and allow for the process called adult hippocampal neurogenesis (AHN) [57,58]. Numerous studies indicate physical inactivity as one of the most important preventable risk factors for developing Alzheimer's disease. Physical activity as a form of treatment of Alzheimer's disease decreases neuropsychiatric symptoms, contributes to a slower decline in activities of daily living and improvement of cognitive functions [57]. Of course, it also has fewer side effects than medications.

LEISURE TIME AND MENTAL EXERCISES

The way of spending free time, different kinds of entertainment and doing mental exercises also have an impact on avoiding or delaying the development of brain changes associated with dementia [59]. The research carried out by Fabrigoule and based on questionnaires conducted 1 year and 3 years after inclusion of patients over 65 years old in the surveys, proved that regular participation in social life and active leisure activities, especially travelling, garden work or additional employment, involves a reduced risk of dementia [60]. One of the retrospective studies conducted on persons over 70 years old - both healthy and suffering from Alzheimer's disease - showed that patients with Alzheimer's disease were less active in most areas: physical exercise, mental entertainment (reading books, doing crossword puzzles and rebuses, crocheting, playing musical instruments) and Do-It-Yourself, the only exception was watching TV. The most protective influence was shown by mental entertainment, which among its supporters reduced the risk of getting ill 2.5 times in comparison to the general population, regardless of acquired education and the type of work performed. People who used the intellect intensively in their youth are less likely to develop Alzheimer's disease in old age. Not only the education or profession acquired, but also a hobby is important for later intellectual performance [61]. In a one study, 135 patients with Alzheimer's disease and 331 healthy people aged 40 to 59 years old were examined. They were asked about the number of hours spent daily on 26 different forms of leisure activities. It was shown that every extra hour spent in front of the television increases the risk of AD 1.3 times. This activity is considered to be non-intellectually stimulating, while intellectual and social activity reduces the risk of the disease [62]. Spending a lot of time watching television already

in childhood and early adulthood is associated with worse executive function and has a big effect on cognitive function in mid-life [63]. In another study, the researchers observed 469 people over 75 years old without initial dementia. After 5 years, dementia developed in 124 people and, among the ways of spending free time, some of them were associated with a reduced risk of dementia: reading, board games, playing musical instruments and dancing [64].

CONCLUSIONS

Alzheimer's disease is an increasingly common problem for our aging population. Although we are becoming better acquainted with the pathomechanism of the disease, we still do not know any effective way to slow down its course, let alone treat it casually. However, it has been proven that appropriate lifestyle modification can significantly reduce the risk of developing the disease in old age. Such factors are diet, physical activity, social life, and a variety of life experiences. Lifestyle modification to prevent AD development also prevents many other diseases, such as CVDs, which are one of the main causes of death of all people in the world. Shaping appropriate patient habits is a key procedure in preventing the development of the disease and it is worth stressing that this is largely the role of doctors. It is important to work out precise guidelines for doctors and their patients concerning lifestyle modifications to prevent AD. Further research is needed to help discover the early biochemical markers of the disease and the grip points of possible causative drugs. Screening tests, such as MMSE should be more easily accessible to patients in primary care settings.

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Diagnosis and rapid prototyping of the insoles using CNC machine for treatment patient with diabetic foot syndrome or significant degree of foot distortion - a case study

Aleksandra Bitenc-Jasiejko¹, Arkadiusz Parus², Katarzyna B. Głodowska³

1. Podologia.pl sp. z o.o, Szczecin
2. West Pomeranian University of Technology, Faculty of Mechanical Engineering and Mechatronics, Szczecin
3. Posnan University Medical Sciences, Department of Social Sciences and Humanities, Poznan

INTRODUCTION

Diabetic foot syndrome is a significant range of medical activities related to diabetes complications. The definition according to the World Health Organization (WHO) defines "diabetic foot syndrome" (DFS) as a degenerative state of tissues, manifested as infection, an injury coexisting with neuropathy. Almost 8% of patients can be diagnosed with neuropathy and after 25 years of diabetes it affects more than 50% of patients [1].

Diabetic foot syndrome affects approximately 20% of patients, including 12-18% in type II diabetes and 1-2% in type I diabetes [2].

In 15-20% cases of people with DFS there is a necessity of the lower limb amputation and in the following few years 50% of them also have the second limb amputated [2].

In countries with a high standard of living, diabetic foot syndrome is the most common cause of non-traumatic limb amputation [3].

Diabetic foot syndrome is also the most common complication of diabetic neuropathy in the case of significant ischemia of the lower extremities [4] which manifests as sensory disturbances, both superficial and deep.

The consequence of this is the patient's lack of sensitivity to the first symptoms of problems e.g. pain, temperature, shock, etc. and therefore risk of damage to soft structures. This is a significant problem in the activities aimed at preventing the establishment of the DFS.

In view of these facts' effective methods of diagnosis and treatment DFS patients are still sought. In this paper the modern, comprehensive methodology of diabetic foot syndrome treatment is shown. The whole process consists of three steps:

- diagnosis based on active foot sensors (dynamic parameters) and scanners (static parameters),
- choosing the right treatment procedure and design personalized insoles,
- manufacturing of personalized insoles.

Each step is equally important and cannot be omitted. In the paper all steps are presented in the following chapters. Changes in the structure and functionality of the feet affected by the diabetic foot syndrome and with the wound or a significant degree of deformation required very precisely designed and manufactured insoles. Therefore, handmade insoles cannot be further used because of lack of manufacturing accuracy.

The article presents the use of a CNC machine with a developed CAD/CAM module for design and manufacturing process. As a result, a significant reduction in the time needed to make the insoles was obtained. More important is that precisely made insoles are better suited to the requirements of the patient's foot.

Therefore, higher protection level against further progress of the disease is achieved. Finally, greater comfort life of the patients is provided.

DIAGNOSIS OF THE PATIENTS WITH DIABETIC FOOT SYNDROME OR SIGNIFICANT DEGREE OF THE FOOT DEFORMATION

An important aspect of the prevention of diabetic foot syndrome is preventive diagnostics, in particular periodic screening and diabetic education of patients at risk.

In this regard, according to the *Clinical recommendations for the management of patients with diabetes 2016* [16], the diagnostic methods for the purposes of preventing overload changes are defined. In the study of the Polish Diabetes Association and others [16] the following diagnostic methods in sensation were indicated:

- pressure sensation test - the use of monofilament - with a pressure of 10 g (Semmes-Weinstein 5.07),
- vibration sensing test - using a neurotensimeter - or a 128 Hz calibrated reed (aero phone);
- sensation of pain (sterile needle),

- temperature sensation assessment (research indicator with two types of ends - metal and plastic),
- electroneurophysiological tests [16].

One of the most important risk factors causing damage of foot are: presence of calluses. One of the most important risk factors causing damage of foot are: presence of calluses. Corns and calluses developing within the plantar part of the foot are mainly associated with incorrect distribution of pressure within the foot [1,2,3]. They are one of the main causes of pain in the plantar part of the foot [4,5], which also affects the functionality of the foot and the ergonomics of gait [6-10]. Cutaneous overload changes are also an important determinant of ulcer formation in patients with diabetes, neuropathy and circulatory disorders [11]. Another determinant of the formation of changes as a result of faulty distribution of pressure within the feet is distortion of the foot and increased pressure on the plantar side of the foot [12] Recommendations in prevention action are:

- systematic foot examination using a physical examination (sensory disturbance) and heart rate, ankle-brachial index,
- regular use of podiatric procedures (removal of calluses and hyperkeratosis) as well as footwear and insoles adapted to the shape of the foot [16].

Calluses, corns and hyperkeratosis are changes resulting from punctual, increased, in most cases prolonged pressure within the plantar part of the foot. An example of a foot with calluses is shown in Fig. 1. Among diabetic patients, the problem of overloading changes in the plantar part of the foot may lead to diabetic foot syndrome, especially in the case of a group of patients in whom we observe sensory disorders resulting from sensory neuropathy [6,7].

Foot deformities (posture defects) such as longitudinal or transverse flat feet, in the initial stage manifesting themselves as the aforementioned changes from the hyperkeratosis group (keratinization and hypertrophy of the epidermis in the form of calluses, corns). As a result, they may cause an overload change in the form of a wound in the foot of a diabetic person (Fig. 1a-d). Therefore, an extremely important aspect of the procedure is periodic examination for abnormalities in pressure in the foot area, together with the use of individual solutions in the field of relief supply and rehabilitation.

The patient exhibits overload wounds in the left foot within the 2nd head of the metatarsal bone together with a callus involving the 2nd and 3rd metatarsophalangeal joint (Fig. 1b and 1c), which indicates deformation of the foot in this area (transverse flatfoot). A wound in the toe of the left 2 foot, which was the result of hammering fingers also could be observed

(*digitus malleus*). In prophylactic activities of patients at risk of wound formation, prevention is an important issue, i.e. observation of disorders of the entire foot and non-diseased foot. It is also important to prepare individual orthopedic equipment for the foot without wounds (Fig. 1a).

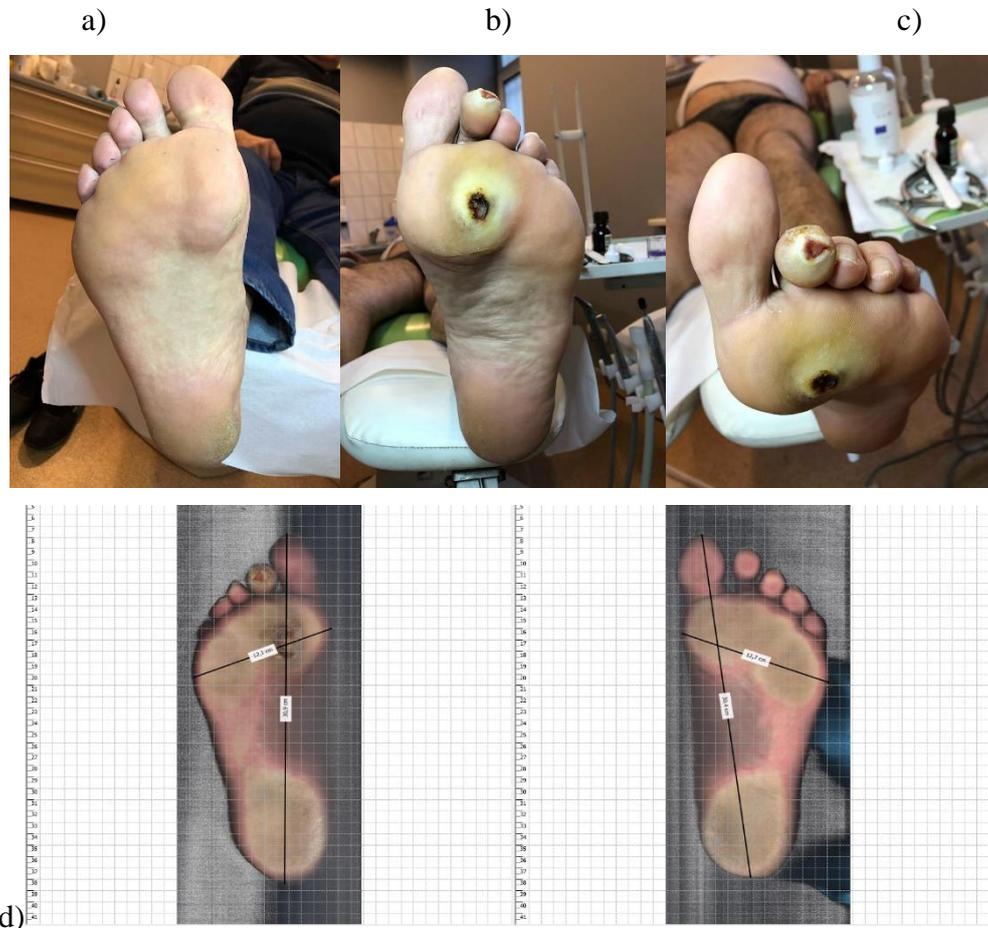


Figure 1. a) image of the left foot - calluses in the heads of the metatarsal bones and in the medial part could be observed, b), c) image of the right foot with wound, d) foot images acquired by podoscanner 2D

Analysis of diabetic guidelines indicates the clear importance of preventing disorders resulting from increased foot pressure on the ground. Therefore, there is a need for an appropriate selection of the diagnostic method that will allow testing the distribution of foot loads to the ground and detects problems at an early stage. This possibility is provided by the method of computer diagnostics called pedobarographic, which allows you to measure pressure on the plantar part of the foot, both while standing (static test) and while walking (dynamic test) [8,9].

In the static analysis presented in Fig. 2 numerous disorders of the pressure distribution could be observed:

- the highest levels of pressure are observed in second toe (with wound),
- increased forefoot pressure resulting in transverse flatfoot,
- lack of participation of the midfoot in support function,
- increased pressure in the media part of the heel (valgus heel).



Figure 2. Static pressure distribution for left and right foot of patient. Results obtained with E.P.S. R1

Furthermore, the most important issue in assessing pressure distribution disorders in patients at risk and with an overload wound is the assessment of propulsion (progression) of the foot. Determining the places where increased pressure is maintained for an extended time, combined with the possibility of dimensioning, allows for the preparation of extremely precise correction and relief elements through the use of CNC technology. Pedobarography (Fig. 3) allows observation of biomechanical parameters of the patient's foot by imaging the plantar part in each phase of gait (propulsion) and determination of time parameters, maximum values and average surface pressure [10,11,13].

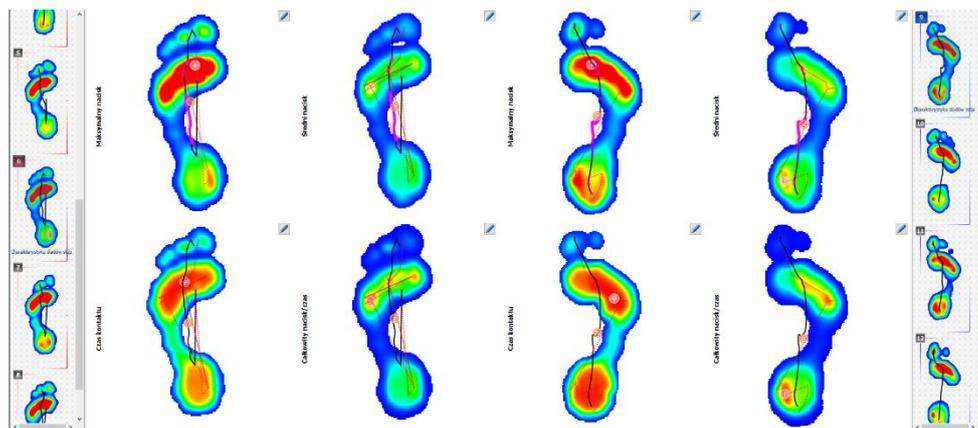


Figure 3. Analysis of foot progression - maximum and medium pressure area

Following issues could be observed from Fig. 3.

- in the left foot significant disturbances in the progression of the foot (dashed line - maximum pressure) - specific returns when rolling. In the design and implementation of individual orthopedic insoles, stabilization of both the longitudinal arch and the heel should be taken into account. Pedobarographic and foot scan measurements should be complemented with measurements of the height of individual areas of the foot (anthropometric measurements of height, width). The results can be supplemented with 3D scanning. Increased pressure of prolonged duration is observed within II metatarsophalangeal joint (exactly at the wound site).
- in the right foot (currently without wounds) calluses within the metatarsophalangeal joints and within the medial part of the heel could be observed. Analysis of subsequent traces using pedobarography indicates prolonged contact time in II and III metatarsophalangeal joints.

When standing (Fig. 4), the patient exhibits compensatory mechanisms, i.e. relieves the left foot (the proportion of pressure in repetitive tests is about 40% on the left foot and 60% on the right foot).

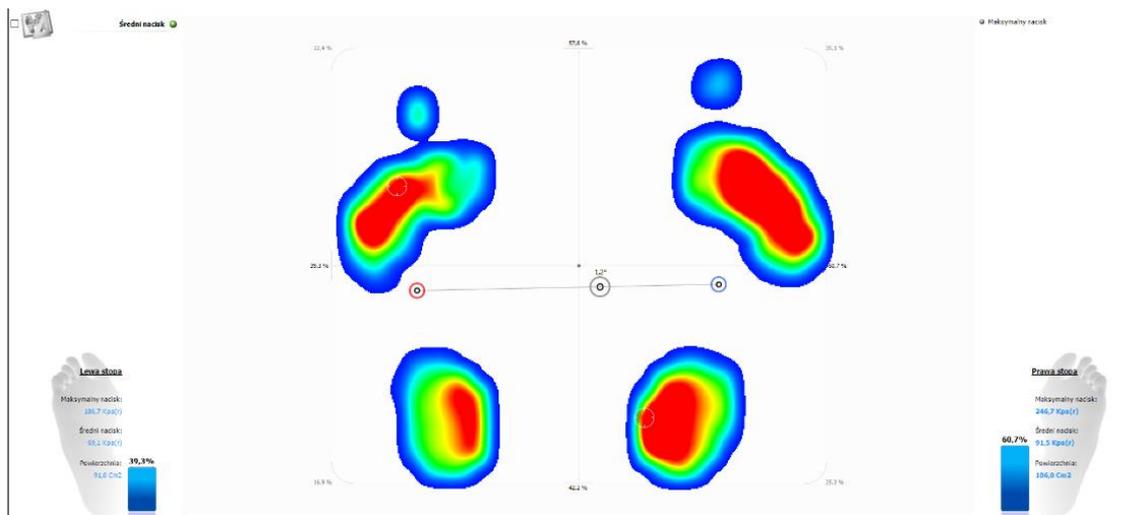


Figure 4. Static pressure distribution for left and right foot of patient with 40/60% disproportion

The described case indicates the coexistence of increased point pressure with the wound resulting from the overload change. However, the result of pedobarographic examination indicates other places of increased pressure within the plantar part, which should be subject to relief (e.g. right foot). This could be done by individually selected orthopedic insoles, designed

based on anthropometric measurements of the feet, functional assessment and pedobarographic statics and dynamics analysis. In addition to the relieving and cushioning element, an important aspect in preventing threats in patients with diabetes is rehabilitation, focused not only on improving and maintaining the patient in good condition, but also on correcting defects [14].

The coexistence of foot defects and deformations with sensory neuropathy causes a loss of soft tissue function. The result of this condition can be both wounds [15] and motor neuropathy, which leads to atrophy of muscle structures within the lower limbs and feet. As a result, weakening ligament structures causes deformations in the osteoarticular structure, which in turn leads to disturbance of the normal gait pattern and biomechanical changes. The risk of permanent, secondary distortions within the foot [16,17] is increased. An example of an advanced change in muscle and bone tissue is neuroosteoartropathy Charkot. This is a complication of diabetic foot syndrome [18]. Compensation mechanisms do not occur so effectively during gait, especially in patients with neurological and diabetic problems (Fig. 5).

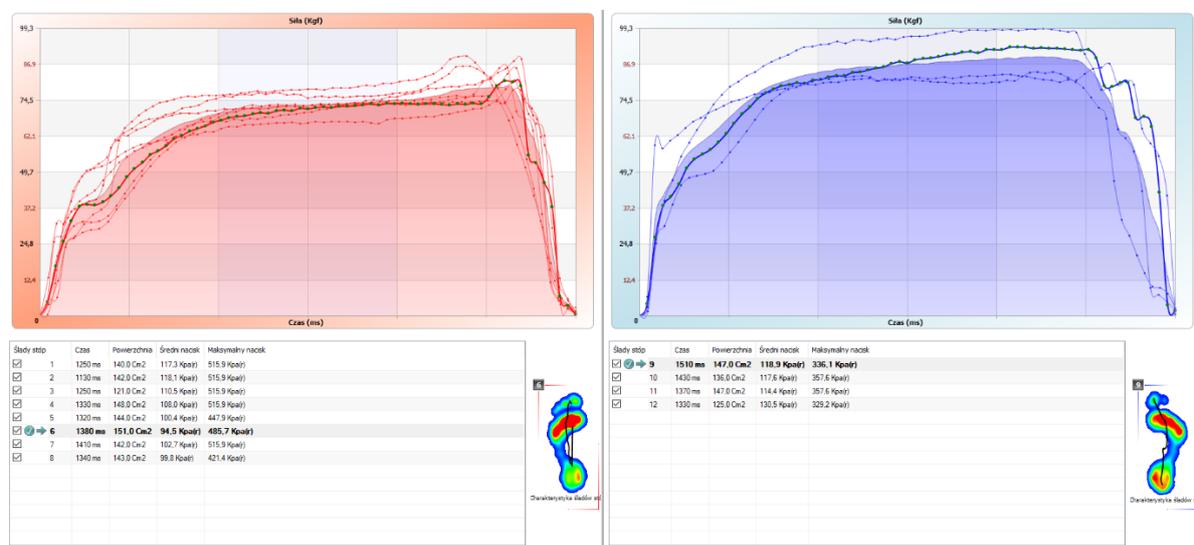


Figure 5. Pressure force of left and right foot in gait analysis. Similar level of left and right force, but in each trace increased pressure in the forefoot could be observed

PROTOTYPING OF THE INSOLES WITH CNC MACHINE AND CAD/CAM SYSTEM

Research results indicate the need for the patient to use an individual orthopedic insole. Following issues should be considered during designing process:

- biomechanical parameters determining during dynamic analysis,
- defects and foot deformities revealed during pedobarographic examination,

- foot anthropometry measurements.

In order to obtain the most beneficial therapeutic results, it is necessary to make very accurate orthopedic insoles. The CNC machine developed during the tests ensures the accuracy of the insert at the level of 0.05 mm (with 0.01 mm resolution). The basic problem that had to be solved was cutting of highly susceptible materials. Typical hardness of materials used to make orthopedic insoles is within 20 – 60 Shore. Moreover, the authors have developed an algorithm to generate tool paths, which allows cutting-prone materials such as foam without deterioration the quality of the machined surface. Many tests were performed in order to select the shape and type of the cutting tool. Finally, the special HSS mills were designed with 6- and 8-mm diameter. The following criteria were considered when choosing the tool:

- final quality of the machined surface,
- the ability to perform as many operations as possible without changing the tool during machining,
- possibility of realizing specific operation in insoles manufacturing,
- time of machining.

The developed CNC machine is presented in Fig 6.



Figure 6. Developed CNC machine

Vacuum table in the machine allows quick fastening of the. Size of the vacuum table is adopted to the maximum insoles size 50. The choice of the kinematic structure of the machine was preceded by an analysis of the pros and cons of the available concepts. The following factors was analyzed:

- way of mounting workpiece,
- efficiency of machining,
- accuracy, for long kinematic chain the errors will cumulate,
- possibility of using dust extraction,
- compact size of machining,
- motors and guide rails service.

In view of analyzed factors, the structure XOZY was chosen, according to the Vragov concept [14].

The workpiece mass is about 300g with the spindle speed about 18000 rpm causes small value of cutting force. This allows to use vacuum table as a main fixing system for the workpiece.

The XOZY kinematics allows to machine with feed up to 125 mm/s. Therefore, the whole manufacturing time of the insoles size 40 is less than 6 min.

The CNC machine is enclosed with the screen and cover and equipped with dust remover. On this basis, machine could be installed in typical pedology office without any additional shields. CNC machine is controlled by PicoCNC and DeskProto software.

CNC machine is one part of the insoles manufacturing system. The second one, equally important, is CAD/CAM software. A modular system for designing orthopedic insoles has been developed in parallel with the CNC machine tool.

This system is dedicated for personalized insoles manufacturing. The diagnostician can made individual changes in the insoles according to results of e.g. pedobarographic tests. The system is very versatile, and operator can make all necessary manipulation.

This is the main advantage of the developed system in comparison to commercially available.

For the patient case study presented in Fig. 1 - 5 medical recommendations include:

- stabilization of the walk and metatarsus to the first and fifth metatarsal phalangeal joint 2 cm high for both feet (Fig. 7),

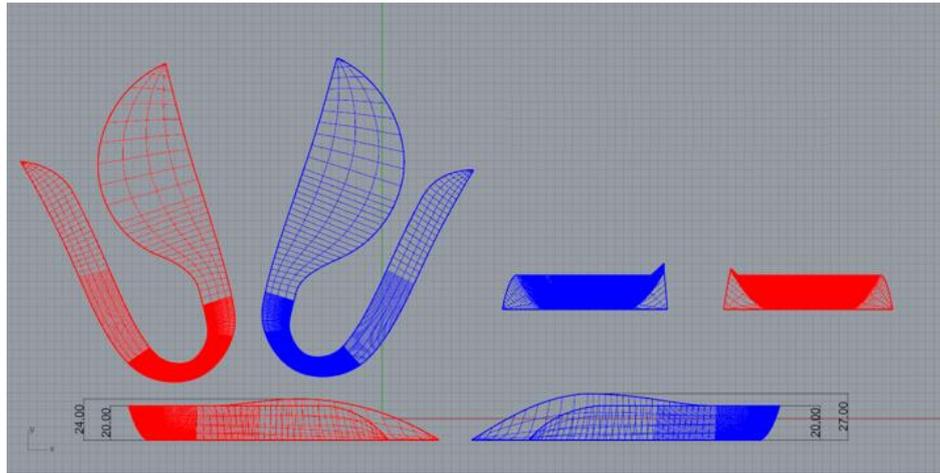


Figure 7. Stabilisation and arch element for right and left foot designed in RhinoCeros (Insoles 3D) – LutraCad

- stabilization of the foot arch through the use of a combined element of the transverse and longitudinal arch, taking into account the varied arch of the right and left foot and the need to secure the lateral arch (dynamic hollow (*pes excavatus*) at 1.5-2mm; no support function of the lateral arch increases pressure on the patient's forefoot). The height of the longitudinal vault is respectively KDL - 27mm / KDP - 24 mm, (Fig.8a., 8b., 9)

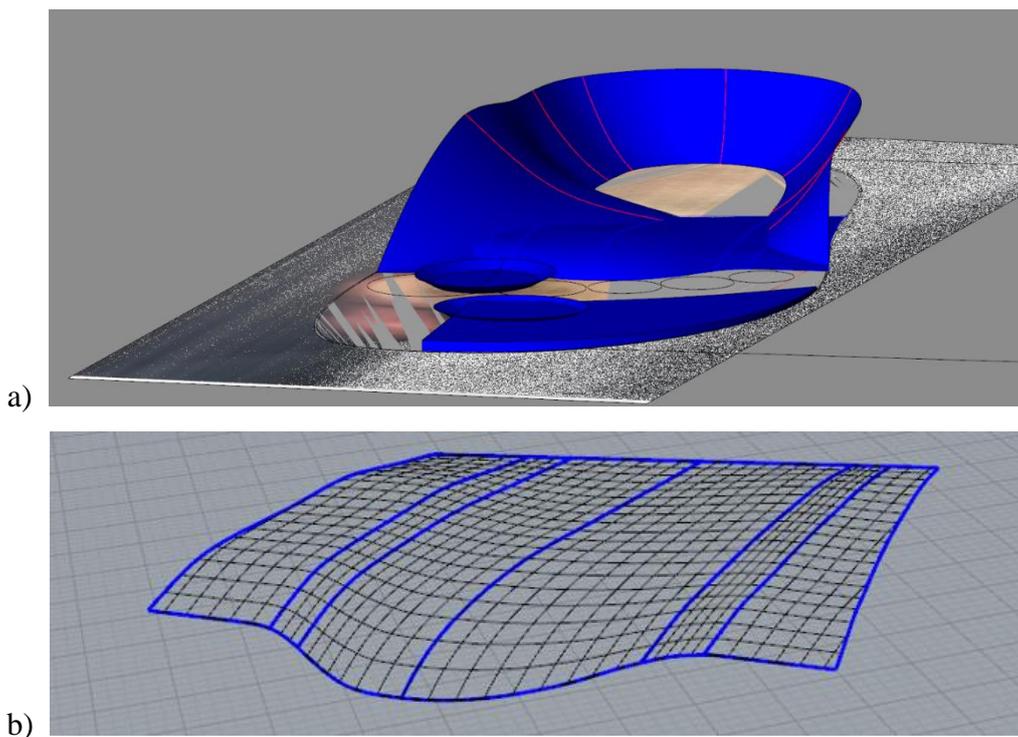


Figure 8. Stabilisation element combined of transverse a) and longitudinal b) arch designed in RhinoCeros (Insoles 3D) - LutraCad

- the use of a correction element within the hammer-toe fingers (*digitus malleus*) 2-5 with varying heights over the entire width of the correction element from 2.5 mm within finger two (covered by the wound) to 1 mm within finger fifth (Fig. 9)

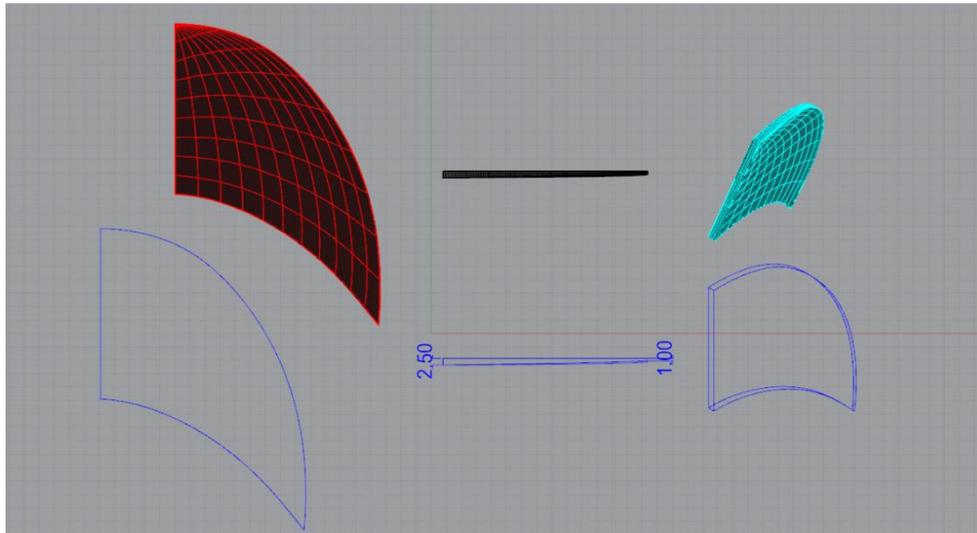


Figure 9. Correction element for right and left foot designed in RhinoCeros (Insoles 3D) - LutraCad

- the use of a correction element within the heel valgus, respectively: KDL - 7 mm, KDP 4mm,
- making relief holes in the wound (i.e. II metatarsal head and fingertip) (Fig.10)

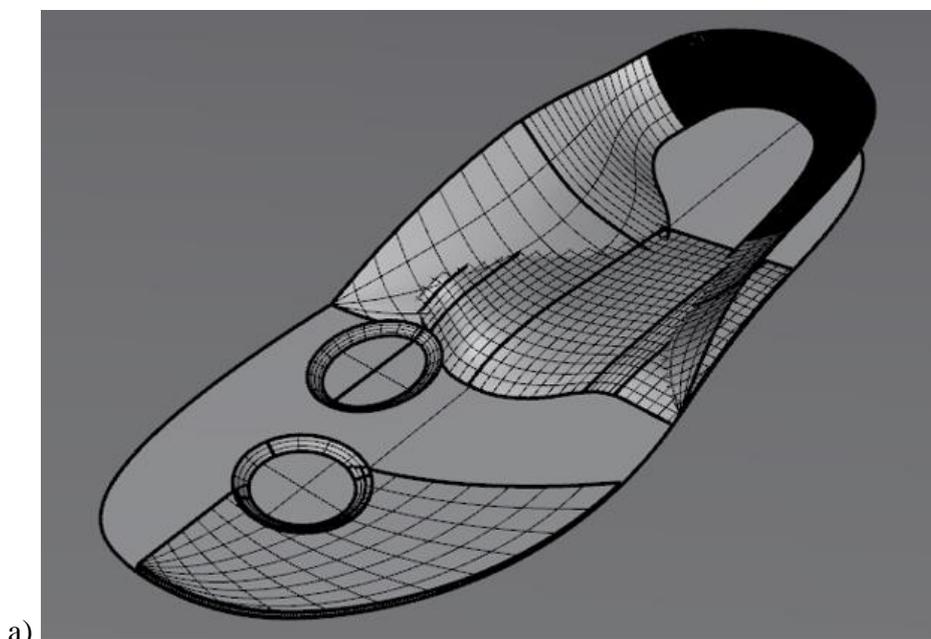




Figure 10. Final view of a) insole designed in RhinoCeros (Insoles 3D) – LutraCad, b) manufactured insole

In the Fig. 7-10 there are presented stages of design and implementation of the orthopedic insoles according to medical recommendations.

CONCLUSIONS

In the paper the complete diagnostic process that allows to design an individual orthopedic insole was described. The standard diagnostic procedures could be used for manufacturing insoles for patients with diabetic foot syndrome or significant degree of foot distortion. All medical recommendations could be incorporated to the insoles design process. The proposed insoles designing system is based on commercially available CAD/CAM software which is specially tuned to that purpose. Therefore, there is no limitations about form and function implemented in the insoles. This is an advantage over the other software designed specially to insoles manufacturing. Developed CNC machine tool is intended to use in typical pedology office or workshop without any additional requirements. There is even possibility of remote manufacturing process based on the files with 3D insoles project. The achieved accuracy and machining speed fulfill requirements of standard pedology office.

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Availability of new glucagon-like peptide receptors agonists in type 2 diabetes mellitus treatment in reference to associated noncommunicable diseases

Aleksandra Krawczuk, Magdalena Paruzel, Barbara Ponikowska, Piotr Potyrała, Michał Puła, Wojtek Tomczak, Katarzyna Zatońska

Medical University Piastów Śląskich of Wrocław, Wrocław

Glucose distribution disorder, what sounds so simple is indeed the curse of 21st-century medicine. Diabetes mellitus that started to occur in described cases as far as 1550 B.C. is now an everyday problem for over 422 billion people and results in the death of over 1,6 billion patients each year. With estimated growth to over 630 billion people till 2045 we can call it the pandemic of the 21st century [1]. The number of patients affected with many slow to cure both angiopathic and neuropathic complications which are costly to treat make type 2 diabetes mellitus an important socio-economic problem. Due to the fact that most of the patients developed their illness for a long time before the first symptoms occur it most likely that only 50-60% of patients have diagnosed T2DM (Type 2 diabetes mellitus) while the 40-50% live without knowing about their health condition.

What is T2DM really about? It's a complicated process with many factors affecting it, but the main mechanism is being either the insufficient insulin release or insufficient insulin uptake by tissues. In both cases not enough insulin affects the tissues and it is a hormone responsible for the regulation of glucose administration and metabolism as well as fats distribution and protein synthesis in the human body.

Being synthesized in beta cells of pancreatic islets, insulin is a polypeptide build from two protein chains (A and B) linked by disulfide bonds. Insulin release is being controlled by lots of factors, but the primary factor is the blood level of glucose, but there are also other substances stimulating this process such as GLP-1 (glucagon like peptide-1) and GIP (glucose-dependent insulinotropic peptide) synthesized by K cells and L cells of duodenum and jejunum. Those hormones called incretins are being released upon meal uptake and help maintain the right glucose levels during the digestion process. Currently to define T2DM we use the criteria specified by the WHO (World Health Organization) [A].

Table 1. Blood glucose levels in diabetes mellitus diagnostics [3]

	Fasting glucose	Venous plasma glucose 2-h after ingestion of 75 g glucose oral load
Healthy	4,0-5,5 mmol/l [72-99 mg/dl]	<7,8 mmol/l [<140mg/dl]
Impaired glucose tolerance (IGT)	5,6-6,9 mmol/l [100-125 mg/dl]	>7,8 and <11,1 mmol/l [>140mg/dl and <200mg/dl]
Impaired fasting glucose (IFG)	6,1-6,9 mmol/l [110-125 mg/dl]	<7,8 mmol/l [<140mg/dl]
Diabetes mellitus	>7,0 mmol/l [>126 mg/dl]	>11,1 mmol/l [>200mg/dl]

Table 2. Types of diabetes mellitus [1]

Type of diabetes mellitus	Summary
Type 1 diabetes mellitus (T1DM)	Genetically determined type of DM, in the process of multi-gen inheritance (at least 24 <i>loci</i> have been located that affect beta cells of pancreatic islets destruction. The strongest relation seems to be the <i>locus</i> IDDM1 that is associated with HLA. The clue of this type of diabetes is autoimmune process that usually has its onset after triggering factor occurs. Main antibodies taking part in this process are: ICA; IAA; anty-GAD; IA-2;
Type 2 diabetes mellitus (T2DM)	Type of diabetes induced both by the genetic conditioning and environmental factors. In this process malfunction of beta cells of pancreatic islets occurs as well as insulin resistance in tissues.
Hybrid forms of diabetes	1. LADA (latent autoimmune diabetes in adults) - autoimmune type of diabetes usually associated with anty-GAD antibodies, however developing in subsequent period of life then T1DM. 2. Ketosis prone type 2 diabetes
Monogenic diabetes	MODY (maturity onset diabetes of the young) - Type of diabetes characterized by early onset (which could point towards T1DM) but with the specification of T2DM.
Endocrine disorders diabetes	Occurs in: 1.Cushing disease 2.Acromegaly 3.Phaeochromocytoma 4.Hyperthyroidism 5.Other
Drugs induced diabetes	Drugs that can induce diabetes: 1.Glucocorticoids 2.Thyroid hormone 3.Thiazides 4.Other
Infection related diabetes	1.Congenital rubella 2.Cytomegalovirus 3.Other

Diabetes mellitus, however cannot be classified as a single disease, this term describes a group of metabolic disorders identified as hyperglycemia in the absence of treatment.[B] Classification of each entity in this group is a challenge because of similar symptoms and the possibility of occurrence of different types of diabetes in one patient. However it's necessary to classify diabetes in order to improve treatment and carry on research to expand our knowledge of diabetes. The most suitable classification in evidence-based medicine so far seems to be the „WHO classification of diabetes 2019”[B].

Symptoms of diabetes are nonspecific and can be ignored by the patient until diabetes is fully developed. In many cases process can be asymptomatic which makes it even harder to diagnose as our only chance to diagnose it is performing blood tests. Symptoms which may occur in diabetes are: Polyuria; Polydipsia; Weight loss; Weakness and somnolence I Frequent urinary tract and skin infections

The vast majority of nowadays diagnosed cases of diabetes are T2DM, this trend is caused by fast economic, cultural and social development of modern society. Aging of society and increased consumption of highly processed foods with a high content of sugar, and lack of physical activity leads to an increase of obesity in society [B]. While obesity is one of the most important environmental risk factors of developing T2DM.

The main goal of T2DM treatment is control of glucose blood level. Glycemia should not exceed 70-99 mg/dl before the meal. According to PTD (*Polskie Towarzystwo Diabetologiczne*) recommendations, achieving an HbA1c level $\leq 7,0\%$ results in a decrease in the frequency of cardiovascular complications and mortality. Patient education focused on lifestyle management, weight loss, blood pressure control and treatment of lipid disorders is also very important [C,2,3,4].

There is a lot of medications used for lowering the plasma glucose level. The oldest one is a biguanide drug called metformin. Nevertheless, it is still the most commonly used anti-diabetic medicament. Metformin works via multiple mechanisms. It interferes with hepatic gluconeogenesis by stopping mitochondrial glycerophosphate dehydrogenase, affects peripheral insulin receptors and increases insulin sensitivity (mainly in the muscle tissue). It also decreases glucose intestinal absorption, causes incretin sensitivity enhancement in β -cells of pancreatic islets and helps with weight loss. It is important that metformin doesn't affect insulin secretion so it does not lead to hypoglycemia [2,3,4].

Sulfonylureas are oral medications that bind with ATP-dependent potassium channels in pancreatic β -cells, causing insulin release and lowering of glycemia. Furthermore, they inhibit glucagon secretion by α -cells of Langerhans islets. Sulfonylureas only work with people

who have pancreatic secretory activity preserved. The main issue with sulfonylureas is that they may lead to dangerous hypoglycemia [2,3,4].

Another class of anti-diabetic drugs is thiazolidinediones (TZDs) represented by pioglitazone. They are PPAR-gamma (peroxisome proliferator-activated receptors) agonists. Because of PPAR-gamma receptor activation TZDs increase peripheral insulin sensitivity, (mainly in muscle and lipid tissue). What is more, it's a beneficial effect on blood pressure and lipid disorders have been proven [2,3,4].

Acarbose is an α -glucosidase inhibitor. It works in enterocytes, causing polysaccharide hydrolysis inhibition. It results in lowering glycemia by decreasing glucose absorption in the small intestine after a meal. Acarbose is a safe medication - there is no risk of hypoglycemia while monotherapy, but combined with some other drugs like sulfonylureas or insulin - such danger exists [2,3].

The latest drugs used to treat type 2 diabetes are incretin mimetic. They include DPP-4 inhibitors (gliptins) and agonists of the GLP-1 receptor. The mechanism of gliptins is to increase GLP-1 level and duration, which affects insulin secretion and inhibition of glucagon secretion depending on glycemia. GLP-1 analogs cause insulin secretion while glycemic stimulus. They also stop α -cells of Langerhans islets from glucagon secretion, inhibit gastric emptying and affect hypothalamus resulting in hunger suppression. Because of that GLP-1 lower plasma glucose level and induce weight loss. One should emphasize the fact that all incretin mimetic lead to insulin release only in response to elevated glucose level [2,3,4].

Another new group of medicaments is SGLT-2 inhibitors, also called gliflozins. They are oral drugs that inhibit sodium/glucose cotransporter 2 (SGLT-2) in a proximal convoluted tube of the nephron. SGLT-2 transporter is responsible for glucose resorption from primary urine, so while blocking it, gliflozins increase glucose excretion with urine. SGLT-2 inhibitors treat hyperglycemia without any correlation with insulin. The fact that they help with weight loss, systolic blood pressure, triglycerides and cholesterol blood level is also worth mentioning. [2,3,4].

The first choice of treatment is metformin. While deciding about combination therapy one must include cardiovascular disorders and chronic kidney disease, risk of hypoglycemia, patient's weight, but also his financial situation [C,3,4]. Basic treatment should always be lifestyle management. Pharmacological therapy can be divided into 4 stages. First is monotherapy using metformin. Alternatively, sulfonylureas, gliptins, gliflozins or pioglitazone can also be prescribed [C,3,4].

When monotherapy fails another drug is added. Usually metformin is combined with sulfonylureas, incretin mimetic, gliflozin or TZDs. At this moment including cardiovascular risk or chronic kidney disease is very important [C]. According to ADA (*American Diabetes Association*), EASD (*European Association for the Study of Diabetes*) and PTD (*Polskie Towarzystwo Diabetologiczne*) recommendation in patients suffering from cardiovascular disease (CVD), metformin should be combined with the drug that has proven to be cardioprotective. Such an effect can be found in some GLP-1 receptor agonists and some SGLT-2 inhibitors. Those drugs have also proven nephroprotective effect so can be used with patients with chronic kidney disease (CKD). During many trials it was proven that the use of those medicaments is linked with evidential reduction of mortality. They have also beneficial impact on obesity treatment [C,4]. Unfortunately, because of significant therapy costs, most available drugs in Poland still are sulfonylureas [C].

When diabetes is refractory to treatment with a progressive defect of pancreatic β -cells it is possible to add insulin in the next step of the therapy [C]. It is strongly recommended when previous treatment didn't bring the desired effect (HbA1c over 8% despite intensive treatment, infections, failure to manage lifestyle, lack of patient cooperation, maximum dosage of anti-diabetic drugs or contraindication to using them) [C,4].

At the beginning of insulin therapy, long-lasting insulin is preferred - it may be isophane insulin (also known as NPH) or long-lasting basal insulin analogs. Insulin analogs are much safer than NPH because of smaller hypoglycemia risk, but the costs of such treatment are also higher [C,4]. Initially, insulin therapy is combined with oral drugs that have been used so far. High efficiency of the combination of basal insulin and GLP-1 analogs in glycemetic control has been proven [C].

If basal insulin is still not successful enough, treatment intensification should be considered. It can be achieved by additional injections of short-lasting insulin (or short-lasting insulin analog), using insulin mix or biphasic insulin [C,4].

Incretins are a group of intestinal hormones having a multidirectional effect on the homeostasis of the body, produced by the K cells and L cells scattered throughout the digestive tract, which are part of the enteroendocrine system. Each type of cell has variable density depending on the gastrointestinal tract segment and produces different hormones. The highest density of K cells falls on the duodenum, they produce glucose-dependent insulinotropic polypeptide (GIP), in the case of L cells it is the ileum and colon, synthesizes glucagon-like peptide-1 (GLP-1) [D, 6]. The secretion of incretins to portal circulation is influenced by digested nutrients, bile acids, other endocrine cells and tissues[6]. In addition to carbohydrates,

proteins and free fatty acids have been shown to influence release [7,8,9,10]. The amount of incretin released in all groups studied by Meier depends on the route of administration of nutrients [11,12]. Oral administration of glucose will result in three times higher concentrations of GIP and GLP-1 than in the case of its intravenous administration [11]. The most important effect of incretins is their effect on the release of insulin from the pancreas. Intestinal hormones released in response to the absorption of digested food affect the Langerhans islets, enhancing insulin secretion (the so-called insulinotropic effect) when plasma glucose exceeds the threshold, estimated at about 66 mm dL-1 [11]. This process is under the influence of three components: incretin hormones, plasma glucose concentration and ANS (Autonomic Nervous System) impulsion [11]. Among listed, incretin hormones play a principal role in physiological circumstances and it ranges between 25% and 75% depending on the amount of glucose consumed[12,13]. As a result of numerous studies, it can be said that disorders leading to loss of insulin secretion are a consequence, not a cause, of the pathology underlying T2DM [14,15]. One model assumes the impaired production of incretin hormones as the cause of T2DM [14,15].

In addition to the insulinotropic effect, intestinal hormones modulate glucagon secretion. A stimulating effect of GIP on glucagon secretion at low plasma glucose level has been found, compared to GLP-1 which has the opposite effect on glucagon release, mainly during hyperglycemia. Moreover GLP-1 inhibits gluconeogenesis regardless of its effect on glucagon levels. Because the liver lacks receptors for GLP-1, this is evidence that incretins exert their effect directly, through their own receptors, which is obvious and indirectly, through neuronal and endocrine pathways. GLP-1 also slows gastric emptying, extending the passage of food through the gastrointestinal tract and having the same effect on glucose absorption resulting in a reduction in maximum glucose serum level after a meal.

Searching for the perfect medicine for T2DM should focus on diabetes as a disease of the whole body and consider not only insufficient work of the pancreas and high serum glucose levels but also a whole list of complications that these changes cause to other organs. Analyzing the mechanism of action of incretins, we should focus on their multidirectional action in various organs (Table 1). Since GIP increases glucagon secretion and does not have an insulinotropic effect in patients with T2DM, it is not used in common therapy. In the opposite of GIP, GLP-1 has been shown to have a positive effect on glycemic control and the prevention of diabetes complications. Unfortunately it has a very low half-life so in order to use it, it needs to be continuously infused. Therefore it was necessary to search for synthetic substances that would work as GLP-1 receptors agonist with a longer half-life. Currently available GLP-1 receptor

agonists offer us a therapeutic effect similar to the physiological action of incretin hormones, which makes them an important element of the treatment of obesity and type 2 diabetes [11,16,17]. The crucial advantage of GLP-1 receptor agonists is demonstrated by the results of large randomized clinical trials, indicating a significant reduction in overall mortality as well as that caused by cardiovascular events [18].

Table 3. Physiological and pharmacological effects of GIP, GIP RA (GIP receptor agonist), GLP-1 and GLP-1 RA (GLP-1 receptor agonist) in various organs, tissues and cells [11,19]

Organ/tissue/cell type	GIP and GIP RA	GLP-1 and GLP-1 RA
Brain	Unknown effect	-Decreased appetite -Reduced food intake
Heart	Unknown effect	-Higher heart rate -Increased glucose uptake -Higher ischaemic tolerance
Pancreas	Mice lacking GIP receptors with higher susceptibility to pancreatic β cell apoptosis	-Higher insulin synthesis -Reduction of pancreatic β cells apoptosis
Digestive tract	Reduced production of stomach acid	-Slower emptying of stomach -Reduced production of stomach acid
Fat tissue	Increased glucose uptake	Unknown effect
Kidneys	Unknown effect	-Lowering albuminuria -Maintenance of GF (<i>glomerular filtration</i>)
Muscles	Increased glucose uptake	Increased glucose uptake
Blood vessels	Unknown effect	-Vasodilatation improving -Lowering systolic pressure

REGISTERED GLP-1 RECEPTOR AGONISTS AND THEIR REIMBURSEMENT (+ REQUIREMENTS THAT PATIENT NEEDS TO FULFIL IN ORDER TO RECEIVE REIMBURSED MEDICATION)

Currently, there are many medications available globally that act agonistically on GLP-1 receptors. Among all GLP-1 receptor agonists, medicines containing exenatide, dulaglutide, semaglutide, liraglutide, teduglutide, lixisenatide and a combination of lixisenatide and insulin glargine are available in Poland [G]. Each of the medicaments containing these substances has different pharmacokinetic and pharmacodynamic parameters as well as the cost of monthly treatment. Based on these parameters, the doctor selects the appropriate drug for the patient's needs and writes down a prescription for him so that the patient can buy it. In case of need of

medicine that is not authorized for sale in Poland physicians may issue an application for target import. It needs to be noted that the registration of a certain medicine is not synonymous with its reimbursement and due to the high cost of the purchase, it is not always possible to carry out effective therapy. Polish list of reimbursed drugs includes two GLP-1 receptor agonists which are dulaglutide and semaglutide. In order to receive a reimbursed drug patient must execute two conditions: patient is eligible for reimbursement and patient meets certain criteria to grant reimbursement for a specific medicine.

Access to a refund is granted to a person who is insured or who is entitled to reimbursement by their additional authorization i.e people with Polish citizenship who are under 18 years of age or who do not have insurance but are entitled to it under an international agreement [E, F].

All medicines with GLP-1 receptor agonistic activity which are stated on the national reimbursement list undergo the same share of the costs. A patient who has received reimbursed medicine from this list pays 30% of the value, the remainder is paid from public funds. The conditions that the patient must meet to qualify for reimbursement of the GLP-1 receptor agonist are identical for all medicines in this group and look as follows: Type 2 diabetes mellitus, in patients prior to insulin initiation, treated with at least two oral hypoglycemic drugs for at least 6 months, with $HbA1c \geq 8\%$, with obesity defined as $BMI \geq 35 \text{ kg/m}^2$ and very high cardiovascular risk understood as confirmed cardiovascular disease or other organ damage manifested by: proteinuria or left ventricular hypertrophy or retinopathy, presence of 2 or more major risk factors among the following: age ≥ 55 years for men and ≥ 60 for women; dyslipidemia, hypertension, smoking [H].

Understanding the mechanism of action and effects of a given drug on the body is very important, especially in T2DM during which pathophysiological processes affect many organs. The analysis of the action of incretin agonists summarized in the previous chapter, compared with the conditions of their reimbursement, shows the lack of understanding of their importance in the developing disease process by legislation.

INCRETINE DRUGS IN DIABETES MELLITUS TREATMENT USED IN GREAT BRITAIN

Our research is so far proven advantages of using incretin drugs in T2DM in specific health conditions. Unfortunately not all drugs available so far on the market are approved to use in Poland. And even those that are available are getting low prescription refunds which

make treatment out of economic range for most of the patients. Knowledge of other countries incretin drug policy so far might guide us to the improvement of the Polish patient's life. Comparison to other European countries will give us a trend to follow in the future.

National Health Service (NHS) in cooperation with the National Institute for Health and Care Excellence (NICE) developed a T2DM treatment algorithm for the United Kingdom. These guidelines as well as in Poland set metformin as a primary drug choice in type 2 diabetes mellitus (T2DM) unless the patient does not tolerate it. In case of maintaining high HbA1c levels (over 58 mmol/mol [$>7,5\%$]) despite the usage of the maximum tolerated doses of metformin National Health Service (NHS) suggests using dual therapy consisting of metformin and one of the following: Dipeptidyl peptidase-4 inhibitor (DPP-4i); Pioglitazone; Sodium-glucose cotransporter 2 inhibitors (SGLT-2i) and Sulfonylurea (SU).

In a scenario where dual treatment did not lower the HbA1c level we consider triple therapy with: Metformin, a DPP-4i and SU; Metformin, pioglitazone and SU; Metformin, pioglitazone or an SU and SGLT-2i

Failing to lower the HbA1c with one of the mentioned triple-drug therapies we need to consider a triple-drug therapy consisting of metformin, an SU and GLP-1 mimetic. However this therapy is reserved for patients with BMI >35 kg/m² and other health implying problems associated with obesity or patients with BMI <35 kg/m² to whom insulin therapy would cause a significant decrease in the quality of life. Moreover GLP-1 mimetic therapy will only be continued with patients that not only reduce their HbA1c by at least 11 mmol/mol (1%) but also reduce their body weight by 3% in 6 months.[I]

Currently in Great Britain drugs prescribed by the National Health Service (NHS) are free of charge in NHS hospitals. Outpatients in England need to co-pay for the drugs 8,40GBP for each prescribed item. Unless they are under 16 years old, over 60 years old or recently gave birth to a child, then prescriptions are free.[K] A multiplicity of available drugs and pharmaceutical companies' competition of making it to the National Institute for Health and Care Excellence reimbursed drugs list, makes GLP-1 mimetic's more affordable. The lower price also means bigger accessibility for patients and higher prescription to buy-out rates.[K]

Based on the information provided we can clearly see that Great Britain guidelines only mention usage of GLP-1 mimetic based therapy as a therapy of the third choice in patients with type 2 diabetes mellitus (T2DM). Obesity-related comorbidities such as cardiovascular diseases (CVD) as a factor in choosing GLP-1 agonist therapy are included however patients also need to meet other factors to qualify for the therapy. Moreover, conditions for GLP-1 mimetic qualification like BMI >35 kg/m² factor exclude many patients with BMI <35 kg/m² to whom

weight loss provided by those drugs could make a significant improvement in the quality of life. Condition expecting patients to lose both weight and lower HbA1c level is also disadvantageous for patients due to the fact that achieving even one of the effects would be beneficial for their health. [I,20].

Table 4. Currently available GLP-1 mimetic’s in type 2 diabetes mellitus (T2DM) treatment [J]

Generic drug name	Substance
Bydureon	exenatide
Byetta	exenatide
Victoza	liraglutide
Saxenda	liraglutide
Rybelsus	semaglutide
Ozempic	semaglutide
Trulicity	dulaglutide
Tanzeum	albiglutide
Adlyxin	lixisenatide

GLP-1 MIMETICS’ POSITIVE EFFECT ON COMORBIDITIES IN TYPE 2 DIABETES MELLITUS

The leading cause of death in patients with type 2 diabetes mellitus is atherosclerotic cardiovascular disease (ASCVD). Despite hyperglycemia, the main risk factors of CVD are hypertension, dyslipidemia, obesity, chronic kidney disease (CKD), lack of physical activity and smoking [21,22,23].

Hypertension defined as lasting blood pressure over 140/90 mmHg is common when it comes to patients suffering from type 2 diabetes. High blood pressure is one of the major risk factors of cardiovascular disease [21]. It was reported that chronic use of GLP-1 agonists has a small, but significant influence on lowering systolic blood pressure (SBP) in patients with type 2 diabetes. Both exenatide and liraglutide were proven to have an impact on the decrease of systolic blood pressure. Analysis of 1-6 LEAD (Liraglutide Effect and Action in Diabetes) trials showed that with liraglutide administration SBP reduction (range from 2.1mmHg to 6.7mmHg) occurred [24,25]. In 6 clinical trials with 2171 subjects it was also observed that exenatide causes a significant decrease of SBP [25]. The beneficial hemodynamic effect of GLP-1 agonists was demonstrated in studies by Katout et al, proving that compared with active comparator therapy they lead to greater SBP decrease. Also meta-analysis by Robinson et al

showed bigger SBP-lowering impact by GLP-1 analogs than placebo or active comparator therapy [24].

Postprandial dyslipidemia is connected with cardiovascular risk in people with T2DM. Insulin resistance in the adipocytes and the liver results in a disturbance in the metabolism of fatty acids, compensatory hyperinsulinemia, hyperglycemia leading to low-density lipoproteins (VLDL) and chylomicrons overproduction. These are believed to be the main reasons of postprandial dyslipidemia. GLP-1 analogs by decreasing intestinal lipids absorption and increasing hepatic fatty acids oxidation may lead to reduce the production of postprandial chylomicrons [26,27]. It was reported that both exenatide and liraglutide lead to reducing postprandial dyslipidemia immediately after administration [26,28].

Obesity increases the risk of T2DM, but also may complicate its course increasing CVD risk. Physiologically glucagon-like peptide 1 regulates appetite and food intake. Central GLP-1 Rs affect appetite reduction leading to weight loss. GLP-1 also inhibits gastric emptying. During clinical studies it weight loss was observed during GLP-1 analogs treatment. A meta-analysis of 21 studies showed that GLP-1 analogs intake leads to clinically relevant weight loss in diabetic patients. It was also proven that liraglutide is the most effective when it comes to weight loss [29]

In addition to their beneficial effects on hypertension, obesity and postprandial dyslipidemia, GLP-1 analogs may also affect the atherosclerotic process. It is suggested that they interact with monocytes and macrophages inhibiting macrophage inflammatory response and resulting in decreasing monocyte/macrophage accumulation in the walls of arteries, reduction in a number of T-cell and macrophages in atherosclerotic plaques and inhibition of MMP-9 and TNF- α expression, which is proven in a study by Balestrieri et al [25,30]. In a meta-analysis made by Song et al. it was observed that GLP-1 analogs therapy results in a significant lowering of atherosclerosis markers in plasma [30].

Large randomized double-blinded trial LEADER („The Liraglutide Effect and Action in Diabetes: Evaluation of Cardiovascular Outcome Results—A Long Term Evaluation“) reported a significant reduction of cardiovascular events while liraglutide treatment. Combined with placebo (14,9%), patients using liraglutide developed fewer (13%) cardiovascular outcomes, defined as myocardial infarct, stroke or death. The total cardiovascular death range was also smaller in the liraglutide group (4,7%) than placebo (6,0%), which lead us to the conclusion that liraglutide has a beneficial influence on cardiovascular outcomes in adults with T2DM. Liraglutide was approved by FDA to minimize major cardiovascular risk in patients with Type 2 Diabetes Mellitus [21,31].

EXSCEL (Exenatide Study of Cardiovascular Event Lowering) observed that the total death number was lower while using exenatide when compared to placebo, but the difference was not statistically important [21].

Another trial was carried for different GLP-1 analog - semaglutide. Compared with placebo, semaglutide occurred to lower the primary outcome range (placebo group - 8,9% and semaglutide - 6,6%). Also, a significant difference in the number of non-lethal myocardial infarction was observed (3,9% versus 2,9% of treated patients) [21,32].

Altogether, in cardiovascular patients GLP-1R agonists seem to be beneficial. Evidence of reducing CVD events is the strongest for liraglutide, then semaglutide and exenatide [21,4].

CONCLUSION

A dramatically increasing number of people suffering from T2DM puts us in indisputable need to constantly look for better solutions. GLP-1R analogs are new promising anti-diabetic drugs that, in addition to strong anti-hyperglycemic activity, have undeniably beneficial effects with high cardiovascular risk patients. In our opinion they ought to be included in as early stage of therapy as possible when CVD risk occurs. Unfortunately, the monthly cost of such a treatment is extremely high which cause an insurmountable problem for an average patient. What is more, not only in Poland access to the reimbursement is complicated. One must meet a lot of conditions before being qualified which postpone the start of the GLP-1R treatment even more with a significant disadvantage for patients. Mindful of patients' welfare, we suggest that the extension of GLP-1R analogs reimbursement should be at least considered.

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Hymenoptera venom allergy – diagnosis and treatment as a new challenge in light of present-day medicine

Anna Żabicka¹, Krzysztof Gomulka²

1. Students' Scientific Society of Adult Allergology, Wrocław Medical University
2. Department and Clinic of Internal Medicine, Pneumology and Allergology, Wrocław Medical University

INTRODUCTION

Hymenoptera venom allergy is a potentially life-threatening allergic reaction following a honeybee, vespid or ant sting. Systemic-allergic sting reactions have been observed in up to 7.5% of adults and up to 3.4% of children. The clinical symptoms can be mild and restricted to the skin or moderate to severe with a risk of life-threatening anaphylaxis [1]. The risk of anaphylaxis to hymenoptera stings is greater in adults compared to children due to increased sting exposure, comorbidities and concomitant medication use.

A diagnosis of venom allergy is made by taking an accurate medical, family and social history, as well as specific allergy testing. Treatment for local reactions involves the use of cold packs, antihistamines, analgesia and topical corticosteroids to reduce swelling, pain and pruritus. Venom immunotherapy is „the treatment of choice” for reducing the incidence of future severe anaphylactic reactions in individuals who have signs of respiratory obstruction or hypotension. Venom immunotherapy is the most effective treatment in reduction of life-threatening reactions to venom and can improve quality of life for individuals. Treatment should only be provided by experienced medical staff who is able to provide emergency care for anaphylaxis and life-threatening episodes. A risk assessment to deliver treatment should be undertaken before treatment is commenced [2].

Hymenoptera venom allergy is an IgE-mediated hypersensitivity to the venom of insects from the Hymenoptera order and is a common cause of anaphylaxis.

There are 24 described orders of insects out of which hymenoptera are the main inducers of severe allergies in humans. Hymenoptera belonging to the genus bee (*Apis*), bumblebee (*Bombus*), wasp (Family: *Vespidae*, Genus: *Dolichovespula*, *Polistes*, *Vespula*), hornet (*Vespa*) and stinging ant (Family: *Formicidae*, Genus: *Myrmecia*, *Solenopsis*), sting humans with high

frequency and thus are the most studied in terms of allergy and allergen-specific immunotherapy [3]. Honeybees (*Apis mellifera*) and yellow jackets (*Vespula vulgaris*, in Europe also called wasp) are observed as the most frequent inducers of allergic reactions to hymenoptera venoms in humans [3].

METHODOLOGY

The systematic literature review was conducted by using three different electronic databases: PubMed, Researchgate and Clinical Key. Electronic databases were searched without any language restriction. All relevant papers issued after 2014 were examined.

The most recent scientific literature addressing problems related to the diagnosis of hymenoptera allergy and to management of venom immunotherapy was analyzed. The potential forms of diagnosis and immunotherapy are the key areas addressed.

RESULTS

Allergic reactions caused by sting of honeybees or wasps are very frequent and responsible for very severe anaphylactic reaction, up to 3,5% of general population. Mostly sting reactions are dependent to IgE immunoglobulin induced reactions type I.

DIAGNOSIS

The diagnosis of Hymenoptera venom allergy (HVA) is based on the clinical history of a systemic/anaphylactic sting reaction and the detection of sensitization to relevant insect venoms by skin testing and/or detection of specific IgE antibodies in patients serum.

Skin tests - SPT, IT

The gold standard for HVA diagnosis is skin testing with venom extracts which should be performed not less than two weeks after the last sting to prevent false negative tests.

Skin prick test (SPT) – as first assessment for HVA, it can be used 100 µg/mL concentration.

Cut-off for positive result is the appearance of a wheal of ≥ 3 mm diameter compared to the negative control in the pricked area after 15-20 minutes observation.

Intradermal testing (IT) - also recommended to perform. Venom extracts are firstly serially diluted to reach end concentrations ranging from 0.001 µg/mL to 1 µg/mL, then

administered at increasing concentrations with intradermal needle injection. The test is stopped at the concentration causing the formation of a wheal (threshold concentration) after 15-20 minutes, or when reaching 1 µg/mL concentration (higher concentrations of venom extracts might exert an irritant effect).

Multiple venoms can be assessed at once, given that the same concentration is used. The outline of the positive wheal reaction should be marked with a drawing pen, transferred to paper using transparent tape and stored in clinical records for both diagnostic and VIT monitoring purposes. The sensitivity of SPT alone is estimated around 64%. Combination of SPT and IT reaches sensitivity to 94%, hence it is recommended to perform both tests sequentially, when available. In case of negative skin tests but presence of a suggestive previous medical history of SR, cutaneous tests should be repeated after 1-2 months, along with serologic testing [4].

Molecular allergology - Serologic testing for IgE antibodies or component-resolved diagnosis (CRD)

Molecular techniques play a key role in the diagnosis of hymenoptera allergy by determining the insect responsible and, thus, the composition of immunotherapy. In recent years, significant advances have been made in the identification of new allergens and the development of techniques for recombinant production of allergens. The best-characterized venom is that of the bee, *Apis mellifera* [5].

The best known and the most potent allergen in venom is phospholipase A2 (Api m1). While phospholipase A2 is a predominant component of bee venoms, phospholipase A1 is highly abundant in wasps and ants. Among other biological roles, during the envenoming process these enzymes cause the disruption of cellular membranes and induce hypersensitive reactions, including life threatening anaphylaxis [6].

Together with melittin - Api m4, they make up most of the dry weight of the venom. According to the recent researches, the relevance of Api m4 has recently been demonstrated and it has been proposed as a biomarker of poor tolerance in patients at the initial stages of immunotherapy. Its advantage is also making it possible to distinguish between two forms of allergy to bee venom. Other relatively scarce proteins reported to be potentially allergenic are acid phosphatase (Api m3), dipeptidylpeptidase IV (Api m5) and icarapin (Api m10), a complex protein with various isoforms [5].

The detection of specific IgE antibodies is an important step for HVA diagnosis to improve the diagnostic accuracy, therefore current guidelines recommend performing both skin and serologic tests. IgEs are antibodies produced after the very first sensitizing event and can

be detected immediately in the serum after the first allergic reaction, although it is recommended to determine their levels 1-4 weeks after the last sting [4].

In the last years it has become more and more evident that testing for specific IgE reactivity against several single allergens (molecular allergology or component-resolved diagnosis, CRD) is evolving as a superior tool to support classical allergy testing [3].

Undoubtedly, CRD plays an important role in the diagnostic assessment of negative skin test results with a positive history of systemic reaction [4].

By using recombinant or natural allergenic epitops, it allows the identification of molecule-specific IgEs, which provides important results for diagnosis and therapeutic management. The main aim is to analyse IgE-mediated sensitizations at a molecular level. Detection of specific immunoglobulin E to marker and cross-reactive venom allergens has been reported to facilitate the discrimination between primary sensitization and cross reactivity [7].

It is particularly important for patients sensitized to both honeybee and vespid venom. CRD enables a better rationale for prescribing venom immunotherapy. Characterization of IgE reactivity to a broad panel of venom allergens has allowed the identification of different sensitization profiles that in honeybee venom allergy were associated with increased risks for side effects of treatment failure of VIT [7].

ImmunoCAP/Phadia

Allergen assays can be performed as singleplex (ImmunoCAP) and as multiplex (ISAC). The singleplex assay determines IgE levels against a single chosen allergen analyte, whereas the multiplex assay uses a fixed array of 112 recombinant or purified native allergen components derived from more than 50 allergen sources [8].

Singleplex ImmunoCAP assays are based on binding of allergen-specific IgE antibodies in serum or other body fluids to allergen coupled in excess to a solid phase, then detected with enzyme-labeled anti-human IgE, where the fluorescence intensity is proportional to the amount of allergen-specific IgE. The major benefit of the singleplex ImmunoCAP is the obtained quantified allergen-specific IgE antibody level (kilounits of antibody per liter [kU_A/L]) based on the total IgE calibration system traceable to the World Health Organization human reference preparation [8].

In addition, the excess of allergen immobilized on the large cellulose derivative surface of a singleplex ImmunoCAP allows complete binding of IgE antibodies, resulting in high sensitivity and a wide linear assay range [8].

ISAC - ImmunoCAP Immuno Solid-phase Allergen Chip

The multiplex ImmunoCAP ISAC is the most complete platform currently available for simultaneously measuring IgE antibody levels against more than 100 allergen molecules. The assay principle is similar to singleplex ImmunoCAP but here the various allergen molecules are spotted in small quantities in triplets on a polymer-coated glass slide instead of one allergen that is immobilized in a large cellulose matrix [8]. Allergen-bound IgE antibodies are detected by using labeled anti-human IgE and fluorescence is measured with a laser scanner [8].

Protein microarray

Protein microarray is a miniaturized multi-analyte, solid-phased immunoassay where thousands of immobilized individual protein spots on a microscopic slide bind are bound to specific antibodies from serum samples, which are then detected by fluorescent labeling, then the image processing and pattern recognition are quantitatively analyzed using advanced algorithms [9].

The flexibility of the number and types of proteins that can be printed on the microarray allows different set of specific IgE-immunoassay analysis to be carried out [9].

Basal Serum Tryptase

The diagnosis of anaphylaxis relies on clinical history after exposure to a potential triggering factor. Serum tryptase concentrations increase on degranulation of mast cells, that is why its levels are measured to diagnose anaphylaxis, however there is no standardized method for assessing total serum mast cell tryptase (MCT). The Working Conference in 2010 proposed a consensus equation (peak MCT should be $> 1.2x$ baseline tryptase + 2 ng/L) to diagnose acute mast cell activation (aMCA) [10].

During the diagnostic workup of HVA, basal serum tryptase levels should be assessed in each patient with SR, to properly identify subjects at a higher risk of developing severe reactions to stings, due to unrecognized clonal mast cell disorders. However, high tryptase levels can also be found in other conditions (e.g. hematologic malignancies, parasitic infections, end-stage chronic renal disease, aneurysms of the abdominal aorta) [4].

Basophile Activation Test – BAT

In the BAT basophils from patients are incubated with allergen *ex vivo* and surface expression of activation markers is measured by using flow cytometry. BAT can be used not only in cases with a clear history but also with negative or unclear results of skin or in-vitro IgE

tests. The measurement of basophil-activation markers may be useful in detecting IgE-mediated sensitization but the relevance for application of the BAT in prediction of clinical reactivity in Hymenoptera allergy is very limited. For this reason, this test currently has no established significant role in the diagnosis and management of patients with insect sting allergy [5].

Treatment – Venom Immunotherapy

Systemic or generalized reactions (SR) or anaphylaxis might include cutaneous urticaria, angioedema, pruritus, flush, unusual nephropathy, central and peripheral neurologic syndromes, idiopathic thrombocytopenic purpura, rhabdomyolysis, vascular or respiratory symptoms, bradycardia, arrhythmia, angina, myocardial infarction, abdominal cramps, gastrointestinal tract and/or uterine smooth muscle contraction.

SRs usually begin 10 to 30 minutes after the sting, but can also arise faster (i.e. in patients with mast cell disorders) or slower (1–4 h) although being less life threatening in the latter case. Anaphylactic reactions due to stings of hymenoptera can cause a rapid death, since cardiorespiratory arrests can be observed in a median time of 15 min after the sting, a fact that leaves people at risk of severe allergic reaction in great anxiety.

The only therapeutic options for venom allergy are the prescription of emergency medication (adrenaline/epinephrine auto-injector, anti-histamines, corticosteroids) or, as the only curative treatment, venom-specific immunotherapy (VIT, also named allergen-specific immunotherapy).

AIT - Allergen immunotherapy

Repeated allergen exposure at regular intervals to modulate immune response to reduce symptoms and need for medication for clinical allergies and to prevent the development of new allergies and asthma (adapted from European Medicines Agency, EMA).

This is also sometimes known as allergen specific immunotherapy, desensitization, hyposensitization and allergy vaccination Allergen immunotherapy (AIT) is a disease-modifying treatment for IgE-mediated allergic disease with effects beyond cessation of AIT that may include important preventive effects [1].

- Subcutaneous immunotherapy (SCIT): Form of AIT where the allergen is administered as subcutaneous injections
- Sublingual immunotherapy (SLIT): Form of AIT where the allergen is administered under the tongue with formulation as drops or tablets

Venom immunotherapy - VIT

AIT where insect venom preparations are administered as a series of subcutaneous injections to eliminate systemic allergic reactions after insect stings.

Venom-specific immunotherapy aims to induce a shift from pro-allergic and pro-inflammatory Th2 conditions, present in an allergic individual, toward a tolerating state of the immune system. The induction of this tolerogenic reaction to venom allergens during VIT is characterized by several changes within cellular and humoral parameters [3].

VIT is indicated in children and adults following a systemic allergic reaction exceeding generalized skin symptoms with a documented sensitization to the venom of the culprit insect with either skin prick tests and/or specific serum IgE tests and/or the basophil activation test (BAT). VIT should also be considered for adults with skin symptoms only but at high risk of re-exposure and/or impairment in quality of life. VIT is not indicated if no sensitization to insect venom can be verified.

Available venoms of *Apis mellifera* and *Vespula* species are available throughout Europe. The use of bumblebee venom would be preferable if the primary sensitization was induced by bumblebee stings. Bumblebee venom for VIT is currently only available in some countries, e.g. in Italy. Worldwide, also ant venoms are available, such as venom of *Myrmecia pilosula* [1].

Throughout Europe, non-purified aqueous, purified aqueous preparations and purified aluminium hydroxide adsorbed preparations (so-called “depot” preparations) are used to perform subcutaneous VIT [1]. The aqueous preparations can be used for build-up protocols including ultra-rush, rush, clustered and conventional, as well as for maintenance phase. Purified aluminium hydroxide adsorbed preparations are used for the conventional or clustered build-up and maintenance schedule. Treatment can be switched from aqueous to depot preparations following the rapid up-dosing phase. Purified aqueous preparations cause smaller local reactions compared with non-purified aqueous preparations. According to the study which was carried out by EAACI a comparative study in honeybee venom allergic patients indicates the superiority of the purified aqueous preparations over the corresponding non-purified aqueous preparation under the same rush protocol in terms of systemic reactions during the build-up phase [1].

Taking into account treatment with more than one venom, selection of the correct venom preparation(s) is important to ensure optimal efficacy of VIT. Sensitization to venom of more than one Hymenoptera species is common in insect venom allergic patients. However, in most of these cases treatment with only one venom seems to be sufficient. A major diagnostic

problem is that currently available tests are not able to distinguish between asymptomatic sensitization and clinically relevant allergy. However, if the initial sting reaction was severe and all allergy tests are almost equally positive to vespid and to honeybee venom, VIT with both venoms should be considered. As there is only limited cross-reactivity between honeybee and vespid venom and *Vespula* and *Polistes* venom, simultaneous injections with both venoms should be safe [1].

Looking for better tolerability of VIT, the preventive pre-treatment was considered. In several double-blind, placebo-controlled trials, it has been shown that pretreatment with H1-antihistamines improves the tolerability of VIT. In detail, it was reported that levocetirizine decreased the rate of SSR (...) [1]. Importantly, effectiveness of VIT was not negatively influenced [1]. Antihistamines were usually administered 1-2 hours before the injections or sometimes twice daily. In case of repeated adverse events during up-dosing, pre-treatment with omalizumab may be recommended [1].

Up-dosing

The recommended starting dose in up-dosing protocols lies between 0.001 µg and 0.1 µg, but it has also been shown that a starting dose of 1 µg is usually safe and not associated with a higher rate of side effects in adults or in children. A maximum dose of 100 µg venom allergen dose usually offers adequate protection against systemic allergic sting reactions in the majority of venom allergic individuals [1].

Maintenance dosing

The standard maintenance dose to be administered is 100 µg of venom. This dose is equivalent to the dry weight of approximately two honeybee stings or five wasp stings [1]. If patients still react to field stings or sting challenge, a dose increase to 200 µg of venom can be recommended adverse reactions [2]. It is recommended in patients who develop systemic allergic reactions following a field sting or sting challenge while on 100 µg maintenance VIT. An increased maintenance dose should also be considered in allergic populations at high risk of multiple stings, such as beekeepers and in exceptional cases where patients have accumulated risk factors for treatment failure [1]. Conventional protocols, where the maintenance dose is reached in several weeks to months, can be administered in outpatient clinics. In an effort to reach the maintenance dose faster, rush and ultra-rush protocols with several injections per day on consecutive days are performed in hospitals. Maintenance dose is reached either within a few hours or within a few days, respectively. Cluster protocols, with several injections per day

usually 1-2 weeks apart, are also a quick alternative to conventional protocols. Importantly, the risk of adverse events is not associated with the severity of initial reactions, high venom-specific IgE levels, or skin test reactivity at low venom concentrations. Conventional regimes appear to be best tolerated while rush and ultra-rush protocols are more frequently associated with adverse events [1,4].

Dosing interval

VIT injections should be administered every 4 weeks in the first year of treatment, every 6 weeks in the second year and in case of a 5-year treatment, every 8 weeks from year 3-5. In the case of lifelong therapy, 12-week intervals may be still safe and effective [4]. The interval for maintenance VIT with 100 µg venom recommended by the manufacturers has been 4-6 weeks for aqueous preparations and 6-8 weeks for purified aluminium hydroxide adsorbed preparations (depot preparations) [1]. Extending the maintenance interval to three months does not seem to reduce effectiveness or increase adverse events, which could be relevant in terms of convenience and economic savings if life-long treatment is necessary. As there is no specific study available for mastocytosis patients with severe initial SSR, caution should be used in extending the intervals to three months in those patients. A dose interval of six months did not provide suitable protection in honeybee venom allergic patients and is therefore not recommended for standard practice [1].

Duration of VIT

VIT should be performed for at least 3 years and some studies have concluded that such period of time may be sufficient, particularly in patients with only mild to moderate initial sting reactions. Nevertheless, most of the studies concluded that a minimum of a five-year treatment is superior for long-term effectiveness [1]. In patients with severe initial sting reactions, at least a 5-year treatment is recommended. Lifelong VIT may be recommended in highly exposed patients with bee venom allergy, honeybee venom allergic patients with high risk of future honeybee stings, patients with very severe initial sting reactions, patients with systemic side-effects during VIT and patients with mast cell disease [4].

When VIT is recommended?

- adults and children with HVA and systemic sting reactions, not limited to skin symptoms

- adults with systemic reactions limited to skin symptoms, if high risk factors or impaired quality of life
- patients with clonal mast cell disorders

When VIT is NOT recommended?

- subjects sensitized to insect venom with no clinical symptoms upon stinging
- unusual / toxic reactions, not immediate type systemic reactions
- patients with active, systemic autoimmune disorders
- patients with severe immunodeficiency
- pregnancy (initiation of VIT)
- special populations

The case of special populations

- patients with cardiovascular disease may undergo VIT, but disease should be stabilized before initiation
- high-risk HVA subjects with malignancy may undergo VIT, only if stable or in remission
- patients with organ-specific autoimmune diseases should undergo VIT, only if stable or in remission
- children below 5 years of age should undergo VIT, only if positive history of severe sting reactions and if cooperative
- ongoing VIT can be continued during pregnancy, if tolerated
- beta blocker and ACE inhibitor therapy may be continued during VIT, but the patient should be informed about possible risks

Adherence

Adherence to VIT is high, possibly because of patients perception of an unpredictable risk of life threatening sting reactions. Regarding to the effectiveness of particular treatments according to recent studies, treatment with ant venom is very effective as 97% to 98% are protected after VIT, the effectiveness of honeybee and vespid VIT is different and ranges from 77% to 84% for honeybee venom compared to 91% to 96% for vespid venom. The underlying reasons are still unclear. It has been speculated that the amount of venom delivered by a

honeybee sting is much larger and more consistent [4]. This may also explain the difference in the reaction rate to sting challenges, which has also been observed in untreated patients.

CONCLUSION

Hymenoptera venom allergy is a potentially life-threatening allergic reaction following a bee, wasp (i.e. paper wasp, yellow jacket or hornet) or ant (i.e. fire ants) sting.

Symptoms range from large local reactions at the sting site to mild, moderate and severe systemic reactions. Mild systemic reactions usually manifest as generalized skin symptoms including flush, urticaria and angioedema. Typically, dizziness, dyspnoea and nausea are examples of moderate reactions, while shock and loss of consciousness, or even cardiac or respiratory arrest, all define a severe sting reaction. Seemingly mild reactions can progress into more severe reactions with little warning. The fear of future severe systemic reactions usually greatly impairs quality of life. Around a quarter of fatalities from anaphylaxis are caused by venom allergy.

Patients are advised to carry an emergency kit comprising adrenaline (epinephrine), H₁-antihistamines and corticosteroids depending on the severity of their previous sting reaction(s). The appropriate diagnosis, treatment and management of HVA is important to modify the natural course of the disease and increase dramatically the quality of life of affected patients. Recognizing specific risk factors for severity and treatment failure and knowing the strengths and weaknesses of diagnostics and currently available treatments should make dealing with HVA a less daunting task.

The only treatment that can potentially prevent further systemic sting reactions is venom immunotherapy (VIT). This may result in long-term clinical benefits and improved quality of life.

Venom immunotherapy (VIT) has been shown to be more than 80% effective in patients treated and is considered the ideal model for the study of the mechanisms of induction of immune tolerance produced by specific immunotherapy with allergens [7]. Furthermore, it has been shown to improve the quality of life of venom-allergic patients when compared with patients who do not receive immunotherapy but carry adrenaline [11]. The management of adverse reactions caused by VIT, the monitoring of the efficacy of VIT and the procedure to be followed in cases of therapeutic failure are common problems in everyday clinical practice [5]. Despite these possible advantages, VIT is still not commonly used by physicians across all European countries. This is likely to reflect uncertainty about the clinical benefits and risks

associated with the use of VIT, uncertainties about the ethics of mounting further formal experimental studies when VIT is established practice in some countries, as well as the practical and economic implications associated with this treatment [12].

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Assessment of functional condition and pain in the treatment of knee cartilage defects with the help of bone marrow concentrate

Krzysztof Koryszewski¹, Jolanta Lewko², Bianka Misiak³, Mirosława Dziekońska¹

1. Department of Orthopedics and Traumatology Medical University of Białystok
2. Department of Primary Health Care, Medical University of Białystok
3. Medical College in Białystok

INTRODUCTION

Active lifestyle and widely propagated increase of sport activity and participation of more people of different ages in it cause that the articular system is subjected to excessive loads. The basic function of the joint, including the knee joint, depends on the presence of a smooth, low-friction surface - articular cartilage. A few millimeters thick articular cartilage covering the articular ends of the femur and tibia bone has a unique structure that allows it to perform an important biomechanical function in the joint. It is made of mesenchymal cells, chondrocytes and a matrix with proteoglycan stroma and collagen fibers. This structure allows articular cartilage to protect the joint from individual overloads that exceed its physiological strength. The load-carrying capability of the tissue is determined by a unique combination of properties such as viscoelasticity, hardness and elasticity at the same time. These features depend mainly on the mutual balance of proteoglycans and collagen fibers, but also from the metabolism of the chondrocytes themselves. It should be emphasized that there is no vascularization or innervation of articular cartilage, which is reflected in its limited healing potential. As long as the proper functioning conditions of joint are maintained, articular cartilage appropriately adapts to greater loads.

Due to the high susceptibility of cartilage covering the joints to various types of damage, osteoarthritis often develops and, as a result, the function of the joint is reduced. This leads to a limitation in human physical activity, including doing sports. This applies particularly to large joints that are heavily loaded such as knee joint. Damage to the cartilage of the knee joint may be the cause of significant motor impairment [1]. The fact that the repair mechanisms of

articular cartilage are insufficient to restore its correct structure was known as early as in the 18th century. However, only the introduction of arthroscopy into knee surgery allowed us to truly understand the pathology of articular cartilage and initiated the rapid development of diagnostic and therapeutic methods in both traumatic injuries and cartilage diseases [2,3,4].

Striving to establish a uniform classification of knee articular cartilage pathologies has been observed in knee surgery for over 60 years. The oldest, but also the most popular is the Outerbridge classification from 1961, which was originally intended for determining the degree of patellar chondromalacia. Over time, this scale began to be used by many "arthroscopists" to assess all types of knee cartilage defects at each location [5]. Classification of articular cartilage damage according to Outerbridge in grades I-IV means: I - clearly softened surface but with cartilage continuity, II – discontinuation of surface with fracture no more than half of thickness of the cartilage, III - deeper damage, but not reaching the subchondral layer, IV - exposure of the subchondral layer [5].

The latest and most accurate classification has been introduced into knee surgery in 2000 by the International Cartilage Repair Society (ICRS) (Fig. 1). This classification is based on a four-degree scale of cartilage damage, taking into account the pathomorphological analysis of cartilage response to injury specifying 0 as no damage, 1 - slight damage, 2 - moderate damage, 3 - significant damage, 4 - significant damage penetrating subchondral layers of bone [6].

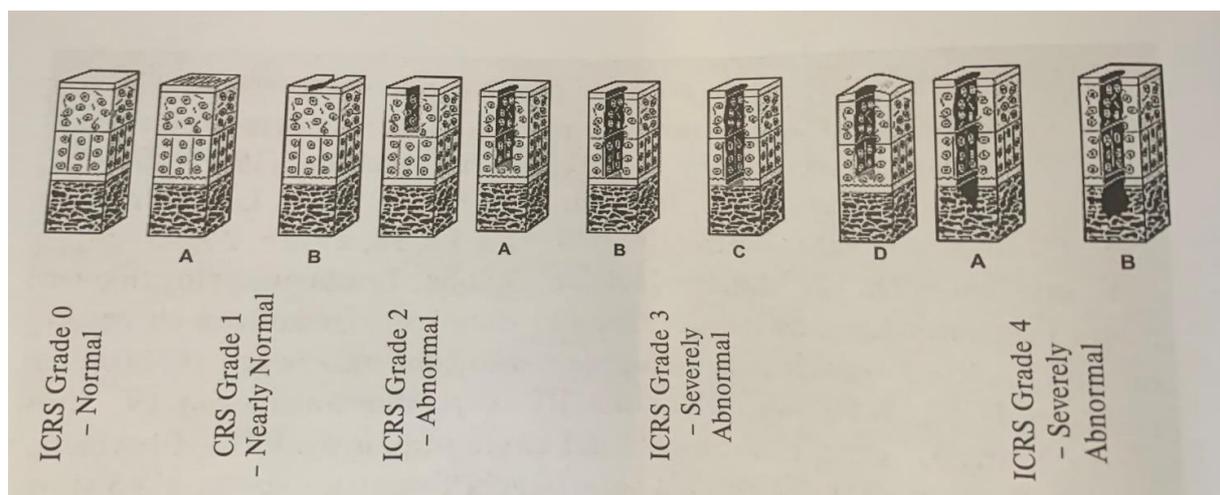


Figure 1. Four-grade ICRS cartilage damage classification [7]

Many mature tissues contain a population of stem cells that have the ability to regenerate or repair after injury, illness or just aging. Stem cells can be found in specific tissues, e.g.

muscles or tendons, but these are characterized by limited possibility of multidirectional differentiation (mono- or bipotent cells) [7,8]. Others cells like mesenchymal stem cells (MSCs) are located in the so-called tissue reservoirs, i.e. in bone marrow, adipose tissue, and these stem cells have multidirectional differentiation possibilities. MSCs can differentiate into mesenchymal tissues such as cartilage, bone, tendon, ligament, muscles, fat or bone marrow [9-13]. It should be emphasized that the transformation of mesenchymal stem cells (MSC) into cartilage cell lines (chondrogenesis) is a complicated process conditioned by many factors. Understanding all of the differentiation and multiplication processes of MSC during chondrogenesis is one of the key challenges facing modern orthopedics. As of today, there is no doubt that this process is stimulated by growth factors (transforming growth factor TGF- β 1, TGF β 2 TGF- β 3 and fibroblast growth factor FGF-2) [14-17].

Currently, researchers focus on better understanding of the physiology of MSC-like cells, and new concepts name them as pericytes, namely perivascular cells. Caplan even postulates changing the name of MSCs to Medicinal Signaling Cells in order to reflect more accurately to the fact that these cells are activated in response to injury or local inflammation and serve to repair the damage by secreting bioactive factors [18] that act immunomodulatory and regeneratively [19]. The bioactive agents secreted by MSC suppress the local immune system, but also inhibit fibrosis (scar formation) and apoptosis, while they increase the angiogenesis and stimulate mitosis together with differentiation of repair cells within tissues or stem cells [20]. In the case of focal injury, pericyte is released from its position on the vascular duct and acts as immunomodulatory and trophic MSC. In the injured area, MSC-induced immunomodulation disables T-cell surveillance and blocks autoimmune reactions. On the other hand, the trophic effect limits the damage area in such a way that no scarring is present and tissue-specific precursor cells replace dead cells [21].

The initiated process of chondrogenesis lasts about 4-6 weeks, but the regenerate reaches its full maturity and metabolic turnover after a few months, which has some clinical implications in the form of specific recommendations in the rehabilitation process.

The goal of treatment for any joint cartilage defect should be filling it in with a hyaline connective tissue that has the characteristics of articular cartilage. The criterion of such tissue is met by tissue that morphologically possesses the characteristics of hyaline cartilage tissue i.e. layering, appropriate tissue cells to the matrix ratio, but also the right proportions of fibers with a predominance of type II collagen fibers. After healing, this tissue should also "enter" the metabolic circulation with the surrounding articular cartilage [22]. In this case, we are talking about cartilage regeneration and the tissue filling the cavity is referred to as a regenerate. It is

worth explaining the terminological ambiguities that are crucial in understanding the processes of chondrogenesis. Regeneration is a replacement of damaged cartilage with a new one - identical in origin and function resulting in formation of hyaline cartilage. All the authors nowadays agree that, as of today, only implantation of autologous chondrocyte cultures can meet the criterion of tissue identity [23,24]. Repair is the process by which damaged cartilage is replaced with a new cartilage tissue, but not identical - most often it is a mix of hyaline and fibrous cartilage.

Usage of methods of treating cartilage defects such as minimally invasive drilling procedure (forage), abrasion or microfractures are nothing more than an attempt to putting mesenchymal pluripotent bone marrow cells into the center of a damage, and as a result to induce a repair tissue reaction. The main problem of these methods is the limited amount of stem cells contained in the hematoma surrounding the cartilage damage. Therefore, these methods are dedicated to supply only cartilage defects below 2 cm² [25,26]. From a clinical point of view, the main criterion for choosing the method of treatment of a defect is its size and its depth, where full thickness defects are those reaching the bone, i.e. to the subchondral layer.

It should be emphasized that in the case of large cartilage defects, an established and commonly accepted procedure is autologous chondrocyte transplantation [1-3]. However, this procedure is a 2-stage surgical procedure, during which, after collecting chondrocytes, it is necessary to multiply them in the laboratory environment and finally implant them in the second stage. Mainly because of cell culture and two operation sessions, this process is time and cost consuming.

AIM OF THE STUDY

Aim of the study was assessment of functional condition and pain in the treatment of knee cartilage defects with the help of bone marrow concentrate.

MATERIALS AND METHODS

Design and Participants

The study was conducted among 54 patients (19 women and 35 men) aged 18-55, average 38.4, hospitalized in the Orthopedics and Traumatology Clinic of the University Clinical Hospital in Bialystok. The size of the reconstructed cartilage defects in the knee joint ranged from 2 to 8 cm² classified as III or IV degree defects according to ICRS scale. The

etiology of defects are 31 traumatic cases, defective patellar track - 6 cases and idiopathic - 17 cases.

Exclusion criteria are: systemic disease, rheumatoid arthritis, bicompartamental or tricompartmental arthrosis, diseases of the hip joint with gait disorders, gout, chondrocalcinosis, angio- and neuropathies, limb deformities and overweight.

Before surgery, each patient had a digital radiograph done on the hip and ankle joint in a standing position, and an axial radiograph of the patella (45 ° flexion). The mechanical and anatomical axis was determined on the radiograph of the limb. MRI examination was performed before surgery, one year after and 5 years after surgery (3T TOSHIBA apparatus). The MRI was performed in the sagittal, frontal and horizontal projection before surgery, as well as during follow-up examinations one and 5 years after surgery (Fig. 2).



Figure 2. Defect on the patella bone, MRI image, before surgery

Isolation of stem cells can be accomplished using a variety of separation techniques with usage of specific growth factors or cytokines. Most often mesenchymal stem cells (MSC) are obtained from bone marrow, adipose tissue or trabecular bone. In our center, MSC are obtained from the bone marrow of the iliac region using specially dedicated Bone Marrow Stimulation kits (Biomet company). This procedure does not require any laboratory testing (it is the usual 15-minute centrifugation) [28,29].

Surgical access in the form of arthrotomy was performed in defects above 2 cm². The scope of the operation (from miniarthrotomy to wide opening of the joint with patellar dislocation) depended on the location and number of defects and thus the possibility of their reconstruction. The next step was the "debridement" of the defect and areas of cartilage damage together with careful treatment of the bottom surface of the cartilage defect (Fig. 3).

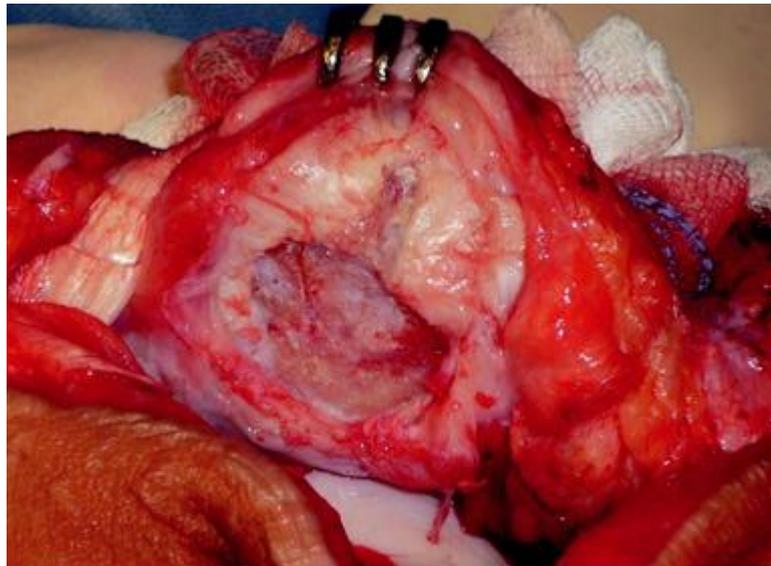


Figure 3. Image of the defect after processing

The key element at this stage of the procedure is accurate vertical excision of the edges of the defect within the limits of healthy cartilage. This action allows to fix the bioabsorbable collagen flap correctly. The collagen membrane is cut according to the template, which was cut out earlier, accurately reproducing the defect. The membrane itself is fixed with 6-0 sutures in intervals of 4-5 mm, and the edges are sealed with fibrin glue. After sealing the membrane, marrow cell concentrate is injected to the bone with a thin needle syringe, while observing if the concentrate does not leak from the membrane (Fig. 4). A slightly different technique should be used for osteochondral defects. In these cases, an additional and necessary element of the management scheme was the reconstruction of the bone defect by filling it with trabecular grafts to the subchondral layer with reconstruction of its shape, e.g. femoral condyle. After filling the defect, the knee extension was performed, observing whether this movement had broken or damaged the prior sutured membrane.

In patients with instability, disturbance of the axis of the limb or defective patellar track, repair or corrective surgery was performed at the same surgical session.

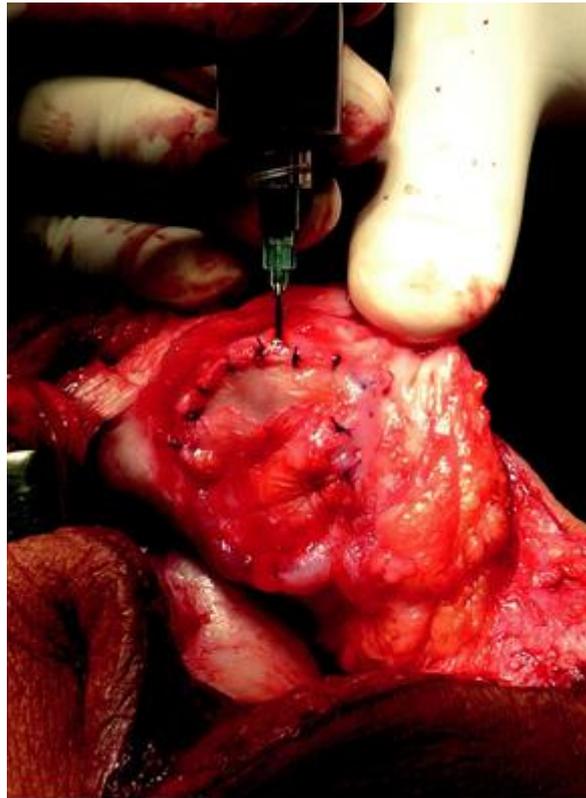


Figure 4. Image of the defect after stem cell implantation

Before surgery, a functional knee joint assessment was performed using the Knee and Osteoarthritis Outcome Score Scale and Lysholm Knee Scoring Scale (Lysholma). Furthermore a pain assessment was performed in the Visual Analog Scale (VAS). An identical study was carried out during check-ups 1 year and 5 years after surgery.

The Knee and Osteoarthritis Outcome Score Scale (KOOS) consists of 6 parts, which assess the severity of symptoms such as swelling, crunching/rubbing, knee clicking during movement, blockage during bending or straightening the knee, limitation in full extension and flexion, stiffness, pain when performing various daily activities. The patient determines the severity of symptoms and restrictions associated with knee dysfunction on a 0 to 4 scale according to Likert scale. The result is calculated separately for each part, and obtaining 100 points indicates the absence of ailments and proper knee joint function [30].

Lysholm Knee Scoring Scale (Lysholm) evaluates outcomes of knee ligament surgery, particularly symptoms of instability. Scale included 8 items:

- limping;
- support;
- stair climbing;
- squatting;

- walking, running, and jumping;
- thigh atrophy.

The revised scale also includes 8 items:

- limping,
- support,
- locking,
- instability,
- pain,
- swelling,
- stair climbing
- squatting.

The total score is the sum of each response to the 8 items of a possible score of 100, where 100 score means no symptoms or disability [31].

Visual Analog Scale (VAS) estimates the severity of pain on a scale of 0 to 10, where 0 means no pain and 10 suggests pain of extreme severity [32].

POSTOPERATIVE PROCEDURES, IMPROVEMENT AND REHABILITATION

On the first postoperative day, patients implemented a rehabilitation programme consisting primarily of passive knee movement in the 0-45° range using the CPM splint.

On the second postoperative day, active quadriceps tense in the rehabilitation protocol was included. Partial loading of the operated limb began two weeks after surgery and the loading value was gradually increased. Full load usually started after 6 weeks.

During postoperative follow-up visits, intraarticular injections were performed containing centrifuged autologous platelet-rich plasma at postoperative weeks 2, 4 and 6, respectively.

The range of movement in rehabilitation exercises was individualized, depending on the accompanying procedures (ACL reconstruction, corrective osteotomy or tuberos transposition).

At week 6 after surgery, patients typically achieved a good range of knee mobility (flexion >120°). The return to sports activity or competitive training followed after a control MRI examination occurred within 24-48 weeks.

Procedure and ethical considerations

The study was performed from January 2014 until December 2019. The research conformed to good clinical practice guidelines, and the followed procedures were in accordance with the Helsinki declaration. All patients signed a consent form to participate in the study. The research was approved by the Bioethics committee of the Medical University of Bialystok (Resolution no. R-I-002/487/2013 from date of 26.09.2013).

Statistical analysis

Statistical analysis was conducted using a statistical package with a rang Kruskal-Wallis test, sequence pairs Wilcoxon test and χ^2 test. Statistical significance was accepted at $p \leq 0.05$.

RESULTS

All patients, except two of them, noted an improvement in the graded scales. There was no postoperative infection or hypertrophy either delamination of regenerate. Most patients – 37 (68,5%) - had cartilage defects of medial condyles of the femur, and the exact anatomical location of the defects is shown in Table 1.

Table 1. Distribution of 54 cartilage defects in patients undergoing cartilage reconstruction using bone marrow cell concentrate

Distribution of 54 cartilage defects			
Localization	Patella	Medial condyle of the femur	Lateral condyle of the femur
Number of defects	10 (18.5%)	37 (68.5%)	7 (13%)

The clinical results after one year and after five years since procedure are shown in Table 2. The average results in all the scales examined in the periods mentioned above showed significant differences.

Results of patients one year after surgery were better on all scales tested and showed statistical significance ($p \leq 0.03$).

The results of patients 5 years after the procedure, compared to the results from year after the procedure, were comparable, but slightly worse and did not show statistical significance ($p \geq 0.2$).

Table 2. General results on the KOOS and Lysholm scales and assessment of pain in the VAS and KOOS pain scales

Scales	Before surgery Mean± SD CI95%	12 months after surgery Mean± SD CI95%	5 years after surgery Mean± SD CI95%
KOOS	64.0 (±2.9) (63.2-64.7)	92.1 (±4.1) (91.0-93.1)	91.6 (±1.9) (91.0-92.1)
Lysholm	56.2 (±4.6) (54.9-57.4)	92.2 (±4.2) (91.0-93.3)	91.3 (±2.8) (90.5-92.0)
VAS	5.8 (±0.06) (5.6-5.9)	1.1 (±0.1) (1.0-1.1)	0.8 (±0.2) (0.70-0.81)
KOOS pain	66.9 (±5.6) (65.4-68.3)	92.1 (±4.1) (91.0-93.1)	90.1 (±3.1) (89.2-90.9)
p≤0.03			p≥0.2

KOOS-Knee and Osteoarthritis Outcome Score Scale, Lysholm - Lysholm Knee Scoring Scale, VAS- Visual Analog Scale

In three cases, a bad result was initially recorded. In 2 cases, the coverage of the defect with regenerate was only partial, which was confirmed by MRI. The „second look arthroscopy” procedures performed in these patients revealed an uncovered bottom surface of the defect, which showed signs of sclerotization of the subchondral bone layer. In the indicated 2 patients, the defect was deepened by resection of the sclerotic layer of bone, then it was supplemented with spongy bone grafts and the bone marrow cell concentrate was reimplanted. Both patients were classified as unsatisfactory results (56 and 59 points on the Lysholm scale and 62 points in KOOS), despite the defects being focal (patella and medial condyle of femur). In the third case, the arthroscopic "second look" only confirmed intraarticular adhesions. Resection of these adhesions allowed to restore a satisfactory knee movement, and the patient was classified as a satisfactory result (83 points on the Lysholm scale and 81 on the KOOS), although the defect in the patella was bifocal.

DISCUSSION

The results of bone marrow cell concentrate treatment in a one-step procedure should be considered satisfactory and comparable with other centers [33,34,35]. It is true that there was a slight decrease in results after 5 years, and the worsening of the final results in 3 patients seemed to be the cause of the gradual degradation of the regenerate and this patients will find oneself in a unsatisfactory results over time.

The whole group of patients should, without a doubt, be monitored further in the event of deterioration of the final result. It is worth noting, however, that the critical limit of 4-5 years time, in which there is a drastic deterioration of the results of the microfractures and mosaicplasty, was exceeded [36,37]. The advantage of repairing cartilage defects with bone marrow concentrate compared to microfractures has also been experimentally proven in an animal model [38]. An important positive aspect of the presented treatment method compared to autologous chondrocyte transplantation is one-step process and significantly lower costs of the procedure. According to Peterson et al., 7-8 years of treatment with autologous chondrocyte transplants are persistent [39]. There is no doubt that further observation of the presented group of patients is needed. It is worth noting that in none of the cases transplantation delamination occurred, and only in two cases the regenerate did not fill the entire cartilage defect. However, on MRI images the integration of the regenerate with the substrate was good, which is very important in the further prognosis [40,41]. On the other hand, other authors also drew attention to the fact that the regenerate, derived from the bone marrow cell concentrate, had definitely better integration with the surrounding cartilage than was the case with microfractures [42,43]. This fact was confirmed by MRI images and histological tests.

Analyzing the development directions of the described method of reconstruction of articular cartilage, it is worth mentioning that the bone marrow concentrate, in addition to stem cells (MSC), contains other cells (monocytes, leukocytes, platelets), which are extremely important for the process of chondrogenesis.

After centrifugation, there are about 7 times more platelets in the bone marrow concentrate than in the bone marrow itself, and these morphotic elements contain many growth factors, which in turn is beneficial for chondrogenesis. In the postoperative period, the positive effect of PRP was additionally used in an attempt to maintain the intensity of chondrogenesis [15-18].

CONCLUSIONS

One-step reconstruction of large cartilage defects with bone marrow cell concentrate is an effective method of treatment. Due to lower costs, this method is a valuable alternative to autologous chondrocyte transplantation.

The presented group of patients requires further observation in case of clinical verification of the presented treatment method.

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Antibiotic resistance – a review of sources, development mechanisms and strategies of control

Aleksandra Krawczuk, Magdalena Paruzel, Barbara Ponikowska, Piotr Potyrała, Michał Puła, Wojciech Tomczak, Paweł Krzyżek

Medical University Piastów Śląskich of Wrocław, Wrocław

INTRODUCTION

The resistance to antimicrobial agents among bacteria is recognized as a major problem of modern medicine in the 21st century. Healthcare systems are facing increasing difficulties in the treatment of infections caused by multidrug-resistant (MDR) pathogens, as they become more severe and impossible to treat with already discovered and commonly used antibiotics [1].

Taking care of patients affected by infections produced by MDR microorganisms also requires longer hospital stays and is responsible for increased healthcare costs [2,3].

Organizations and institutions around the world emphasize the significance of the issue. In 2017, the WHO (World Health Organization) has published “The Priority Pathogens List” to guide research, discovery, and development of new antibiotics. International experts have analyzed many data taking into consideration how serious infections caused by selected microorganisms are and what mortality is associated with it; whether their treatment requires long hospital stays; how frequently they are resistant to existing antibiotics when people in communities are infected; how easily they spread between animals, from animals to humans, and from person to person; whether they can be prevented (for instance through good hygiene and vaccination); how many treatment options remain; and whether new antibiotics to treat them are already in research and development pipeline [A].

Pathogens were divided into three categories according to the urgency of need for a new antibiotic with the first group (critical) containing the most dangerous pathogens [B].

Priority 1: CRITICAL

Acinetobacter baumannii, carbapenem-resistant

Pseudomonas aeruginosa, carbapenem-resistant

Enterobacteriaceae, carbapenem-resistant, 3rd generation cephalosporin-resistant

Priority 2: HIGH

Enterococcus faecium, vancomycin-resistant

Staphylococcus aureus, methicillin-resistant, vancomycin intermediate and resistant

Helicobacter pylori, clarithromycin-resistant

Campylobacter, fluoroquinolone-resistant

Salmonella spp., fluoroquinolone-resistant

Neisseria gonorrhoeae, 3rd generation cephalosporin-resistant, fluoroquinolone-resistant

Priority 3: MEDIUM

Streptococcus pneumoniae, penicillin-non-susceptible

Haemophilus influenzae, ampicillin-resistant

Shigella spp., fluoroquinolone-resistant.

Moreover, WHO points out that not only discovering new antimicrobial agents is important, but also a proper use of existing ones, better prevention of infections and reasonable use of potential new antibiotics is vital to fight MDR bacteria. CDC (Centre for Disease Control and Prevention) reveals alarming data in their “*Antibiotic Resistance Threats in the United States, 2019*”. The report shows that more than 2.8 million of antibiotic-resistant infections occur in the U.S. each year, and they cause more than 35,000 deaths/year [C].

ECDC (European Centre for Disease Prevention and Control) acknowledges the problem and suggests specific solutions in their guidelines. The organization also tries to educate society about the issue by establishing European Antibiotic Awareness Day [D]. Dr. Arjun Srinivasan, an associate director at the Centers for Disease Control and Prevention, stated in 2013 that we are now in the post-antibiotic era [E]. The challenge presented by the spread of pan-resistant bacteria permeates many disciplines from medicine and microbiology through epidemiology, to agriculture and economics. In this paper, we review some of the possible ways in which this problem can be resolved none of them however should be looked at in isolation.

MECHANISMS OF ANTIBIOTIC RESISTANCE

In order to understand antibiotic resistance it is vital to mention the two basic types of this phenomenon, i.e., “intrinsic” and “acquired” resistance [4]. The intrinsic resistance is an inherited characteristic feature of all representative of the selected bacterial specie, developed naturally during their evolution process in order to survive in a hostile environment. Particular attributes of certain types of bacteria provide them resistance to selected groups of antibiotics. For example, Gram-negative bacteria have outer membranes preventing glycopeptides from

reaching their target and killing these microorganisms. Although intrinsic resistance limits therapeutic options and requires administration of suitable antibiotics for certain bacteria, it does not pose a great threat to the patients and healthcare systems. The main concern is the acquired resistance, which occurs in populations of microorganisms that were originally susceptible to the antibiotic. It is especially dangerous, because it can be transferred between different strains (even among phylogenetically distant species/genera). This allows a particular bacterium to obtain more than one resistance mechanism in a short period of time, sometimes limited to minutes/hours [5]. As a result, we observe the appearance of MDR pathogens including ones that cause infections impossible to treat, because all of the existing antimicrobial compounds are no longer effective. Development of MDR pathogens is possible, because bacteria have great ability to adapt to the changing environment due to their genetic plasticity. They use two main genetic strategies to become non-susceptible to an antibiotic action, which are mutations of their own genetic material and acquisition of foreign DNA by transformation (uptake of extracellular DNA), transduction (bacteriophage mediated relocation of genetic material) and conjunction (cell-to-cell exchange of DNA using chromosome fragments or mobile genetic elements). Another very efficient mechanism of gaining and accumulating resistance genes is represented by integrons, which provide the process of addition of new genetic material to bacterial chromosomes [6]. Expression of resistance genes leads to revealing a variety of biochemical mechanisms enabling bacteria to overcome action of antimicrobial agents. Basic mechanisms, categorized according to biochemical route involved in resistance, include:

1. Synthesis of an enzyme which destroys or modifies antimicrobial compounds
2. Disturbing permeability barriers
3. Modifying the target site of antibiotics
4. Synthesis of a new protein that does not bind the antibiotic, but remains its original function in building bacterial cells
5. Bypassing metabolic pathways inhibited by the antibiotic
6. Removing the drug by efflux pumps that extrude the drug from the cell [7]

If a strain of bacteria obtains one of the mechanisms, it dominates the ecological niche, when the population of bacteria is exposed to an antimicrobial agent. This phenomenon is called “strain selection” and contributes to increasing resistance. Spreading of resistant strain is significantly easier in a hospital environment due to extended usage of broad spectrum antibiotics, however external sources are consequently undervalued [8]. Besides to mechanisms conditioning resistance, bacteria have developed processes responsible for tolerance to

antimicrobial substances. From them, the ability to produce multicellular aggregates, known as biofilms, is considered to be the most important. These structures provide protection against environmental factors, such as altered pH, changing temperature, oxygen availability, osmolarity, nutrients insufficiency, and what seems to be the most important, action of antimicrobial agents [9]. It is a complex structure of microorganisms embedded in self-produced extracellular substance composed of free DNA, proteins and polysaccharides. High concentration of cells promotes exchange of genetic material and enables communication between bacteria, a process known as “quorum sensing” [10]. Development of biofilm provides the ability to grow on different surfaces including medical devices (e.g., artificial heart valves, catheters and prostheses), but it can be also formed on tissues, thus promoting development of periodontitis, osteomyelitis and other chronic infections [11]. Tolerance to antibiotics and host's immune cells makes biofilm-associated infections extremely difficult to overcome [12]. Despite they are major global health concern, there are no guidelines to help doctors in treating them [13].

MAIN CAUSES OF GROWING RESISTANCE

The reasons why the medical world is facing crises in the area of antibiotic therapy are mainly inappropriate use and overuse of these drugs, both in human and veterinary medicine [14]. Misuse is often associated with pushing the physician by his patient to prescribe antimicrobial agents, even when there is no certainty if an infection is caused by bacteria or viruses. This example also shows insufficient education of society in matters of antibiotic administration [15].

To provide higher production, antibiotics are extensively used in livestock feed which also contributes to spreading antimicrobial resistance [16,17].

Another issue is availability of antibiotics. In some areas they are sold over-the-counter or *via* the Internet, which allows unlimited and unrestricted access [18].

It can also be assumed that the pharmaceutical industry releases huge amounts of antimicrobial compounds into the biosphere during production of drugs. The extreme example of irresponsible disposal is dumping enormous quantities of ciprofloxacin into rivers by pharmaceutical manufacturers in Hyderabad central India [19], but it can be presumed that similar large-scale disposal is performed worldwide [20].

USE OF ANTIBIOTICS IN AGRICULTURE

Having looked at the problem of antibiotic resistance, it is important to mention that the majority of antibiotics is used in agriculture. For example, in 2009, 80% of all antibiotics sold in the United States were bought by farm owners [21,22,23]. Genes responsible for antibiotic resistance have been discovered in dairy and meat, as well as in fruit and vegetable produce, and it is possible for them to be transferred into other niches. Many MDRs have been identified in cattle, poultry and swine, some of them being zoonotic bacteria (meaning one that can be commonly transmitted between animals and humans [24]), such as *Salmonella*, *Escherichia coli* or *Enterococcus faecalis* among others [23, F].

In meat farming, antibiotics are commonly used to promote growth, as well as anti-infection prophylaxis. This is problematic because the particular substances used for those purposes in agriculture are also used in medical settings. About 90% of the antibiotics administered to the livestock are then returned to the environment through animal excrements, which contaminate ground waters and crops and contribute to an increase in MDRs [21,23, F].

An American study carried out in 2001 showed that 20% of ground meat tested contained traces of *Salmonella*, of which more than 80% was resistant to at least one antibiotic [25]. One instance of antibiotic resistance stemming from agricultural practices is colistin resistance. Colistin (also known as polymyxin E) is a last-line option treatment in the case of severely antibiotic-resistant bacteria, such as XDR (extensively drug-resistant) *Acinetobacter baumannii* and *Pseudomonas aeruginosa*; it is also used in veterinary medicine for the purpose of prevention and treatment of *Enterobacteriaceae* infections in cattle and swine [26].

Increasingly, plasmid-mediated colistin resistance is identified in samples from patients, who were not treated with colistin, leading researchers to believe it is caused by ingestion of meat contaminated with resistant strains [27]. Due to this, measures are being taken to counter the rise of plasmid-mediated colistin-resistance in animals by decreasing its agricultural use. For example, in 2016 the Ministry of Agriculture in China banned the use of colistin as a growth promoter in live-stock, and the European Medicines Agency recommended that it should only be used to treat infections for which there are no effective alternative drugs [28].

Another example of agricultural use impacting MDRs is the case of Denmark, where tylosin used to be a popular growth promoter in cattle. Unfortunately, tylosin is a representative of macrolides, a group of antibiotics frequently used to treat bacterial infection in humans [29].

Due to the extensive agricultural use, in 1996 an estimated 93% of *Enterococcus faecium* was resistant to tylosin, creating a danger of resistance to other macrolides. Legislation

was introduced banning the use of tylosin and as early as 2001 the percentage of *E. faecium* resistance dropped to 26%, and continued to decrease in 2004 [F].

To limit the rise of antibiotic resistance in agriculture appropriate legislation banning the use of antibiotics as growth stimulants must be put in place; for example, the European Union banned it completely in 2007, allowing antibiotic use only for the purpose of veterinary treatment [30]. Education schemes for farmers and agriculture workers would also help increase awareness and offer alternatives to the use of antibiotics.

NEW ANTIBIOTICS

With MDR pathogens making treatment options extremely limited including antibiotics associated with higher toxicity and more severe side effects [31], a need for a new substance arises. Introducing a new, innovative antibiotic which will prove effective in treating MDR pathogens is a complicated process. The course of developing a drug and introducing it onto the market takes between 10 and 20 years and consists of many different steps [G]. From basic research, considering many different substances, searching for one with the highest therapeutic potential. Those selected in this process are then tested in pre-clinical studies in *in vitro* (using eukaryotic cells) or *in vivo* (using animals) conditions. If a drug passes the pre-clinical trials and fulfills strict standards and guidelines, it can then be introduced for clinical testing on patients, and eventually become a licensed drug [H,I]. Additionally, an important factor affecting research of new antibiotics is the economic point of view. Antibiotic treatment is short-term, usually lasting around a week [J]. In addition, the newly introduced antibiotics which target MDR pathogens would be used as a last resort (not first- or second-line treatment), making them unprofitable for producers. Big pharmaceutical companies are mainly focused on drugs in chronic, non-bacterial diseases (e.g., cardiovascular and oncological diseases), huge quantities of which are sold worldwide, ensuring financial gain [32].

PRESCRIPTION AND DIAGNOSIS

In Europe, around 80-90% of prescriptions for antibiotics are written by general practitioners. A big part of that is written for patients with symptoms of upper respiratory infections, which can be caused by both bacteria and viruses - without proper diagnostic tools it can be hard to differentiate between these two, especially at early stages of an infection [21,33].

If a patient gets prescribed an antibiotic treatment as a precaution without confirming that the pathogen responsible is in fact a bacterium, taking it does not only fail to cure the infection but contributes to the rise of antibiotic resistance in human microflora as well. To limit this phenomenon we can look at the practices promoted in Scandinavian countries, where it is very common to employ different diagnostic practices to reduce the number of inaccurate treatment prescriptions. Examples of such practices are rapid antigen-detection tests (e.g. Strep A Test) as well as measuring levels of procalcitonin or C-reactive protein (CRP); research has shown that doctors determining levels of CRP, while diagnosing lower respiratory infections, decreased the number of antibiotics prescribed from 53% to 31% [34].

In a situation where a physician decides to opt for empirical antibiotic therapy, for example due to rapidly progressing conditions (such as meningococcal meningitis), it is crucial to obtain appropriate samples from the patient before implementing the treatment. This allows for microbiological testing and determining the pathogen responsible for the infection, simultaneous to treatment, enabling modification of the chosen antibiotic when results are available. It is important in cases of empirical therapy to follow the most recent available guidelines, if possible one that is suited for a particular hospital or ward. Taking those steps can minimize the spread of MDRs.

Research shows that between 40 and 75% of patients with viral upper respiratory infections are treated with antibacterial agents. To decrease that percentage General Practitioners (GP) can apply a method of “delayed prescription”. It involves prescribing antibiotics to a patient that they will not start immediately after the visit, but upon waiting 2 to 3 days in order for the symptoms to intensify. If they decline instead, the infection might have been of a viral origin and the antibiotic treatment will be obsolete [K].

One research concluded that by employing delayed prescription the number of antibiotics used decreased, without negative impact on the patient as compared with those starting therapy immediately after consulting a GP [35].

The use of antibiotics in Intensive Care Units (ICU) is often unavoidable and it is the ICU patients who frequently require second- and third-line antibiotics are employed. Guidelines often point to prolonged antibiotic treatment, but its usefulness has been questioned. According to some research, reduced treatment duration can lower rates of antibiotic resistance without diminishing therapeutic potential. For example, one study looked at 401 patients with Ventilator Associated Pneumonia (VAP) who were treated with antibiotics empirically with one group receiving the therapy for 8 days and the other for 15 days. Outcomes of the treatment were similar in all cases [34,36]. Data like this points to the conclusion that it may be possible

for some antibiotic use guidelines to be reviewed, keeping in mind that decreased quantity of antibiotics used is not only positively impacting the fight against MDRs, but also lowers the cost of therapy.

STEWARDSHIP PROGRAMMES AND EDUCATION

A crucial element of responsible medical practice is communication with the patient. The task of a physician is not only arriving at a diagnosis and prescription of an appropriate treatment, but also, equally as importantly, informing the patient of their disease and the process of its management. On the other hand, for antibiotic treatment to be successful there is a need for cooperation on the patient's side. This especially concerns situations in which the patient takes the commonly prescribed, first-line antibiotics to treat respiratory and urinary tract infections when staying at home. If the patient fails to follow the appropriate regimen, for example skips doses or takes them at large time intervals, the antibiotic may be ineffective despite being prescribed correctly, causing the patient to require another antibiotic treatment. A study conducted in the Netherlands looked at the correlation between undergoing training on the topic of patient-doctor communication by physicians and the quantity of prescribed antibiotics, arriving at a conclusion that those who received the training prescribed antibiotics less frequently in the case of upper respiratory infections than those who have not [32, 37].

An important element of rational and safe antibiotic use are antibiotic stewardship programmes [34].

They help physicians to adjust the dosage, type, and time of therapy. In order for the programmes to fulfill its purpose hospitals and other medical facilities should be able to provide epidemiologists and microbiologists with feedback. This enables obtaining appropriate data, which can be gathered and the programmes altered to fit the needs of specific facilities. A review of 24 reports from years 1996 and 2010 concludes that stewardship programs helped decrease mean therapy duration and reduction in antibiotic use [21, 34, L].

In 1994, in Sweden, STRAMA (Swedish Strategic Programme for Rational Use of Antimicrobial Agents and Surveillance of Resistance) was introduced, which purpose was to monitor the emergence of MDR pathogens, antibiotic use and work on implementing rational usage practices.

By 2004 the number of antibiotic prescriptions fell from 536 to 410 per 1000 people per year [38], showcasing usefulness of such initiatives.

ALTERNATIVES TO ANTIBIOTICS

One of the alternatives explored in the post-antibiotic era is bacteriophage therapy. Bacteriophages are viruses able to kill bacterial cell without damaging the eukaryotic cells [39,40]. During the treatment a “cocktail” of different types of bacteriophages is usually used to avoid bacteria to develop resistance [41]. It is considered as an experimental therapy and is administered in only a few institutions around the world. One of the advantages of bacteriophage therapy is that it is specific to one specie or even strain of bacteria, allowing it to destroy the cause of an infection without affecting the patient’s physiological microbiome. This situation stands in contrast to antibiotics, which are known to cause alteration to the natural microflora, sometimes weakening the balance between physiological and pathological microorganisms [39,42,43]. Another interesting advantage of bacteriophages is their ability to cross the blood-brain barrier, possibly making them useful in treating infections of the central nervous system [39]. So far, with its limited use, bacteriophage therapy has an 80% success rate at treating enterococcal infections, and 90% at treating patients with *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Escherichia coli* [41]. There are, however, many questions to be answered before bacteriophage therapy can be approved for widespread use. Many factors, such as optimal dosage, route of administration, frequency and duration of treatment have to be determined, as well as addressing concerns about bacteriophages being potentially immunogenic (able to trigger reaction for the immune system) [43]. Another limitation of bacteriophage therapy is simultaneously its advantage – the specificity of the treatment. Such targeted cure comes at a cost; before therapy can even begin, physicians would have to accurately identify the pathogen responsible for an infection, since bacteriophages will only infect their specific bacterial hosts. That could generate further costs of microbiological testing and could prove difficult in a hospital setting [39]. Nonetheless, bacteriophage therapy is an interesting solution to the MDR pathogen problem, with more and more research being conducted around it.

CONCLUSIONS

In summary, the issue of antibiotic resistance is a complex problem, requiring measures to be taken in many different fields. It is important to note that some solutions discussed in this paper would have to be confronted with reality, which varies significantly from one country to another. Funds, hospital equipment, availability of alternatives are all factors that influence

effectiveness of the fight with antibiotic resistance. Examples of studies from all over the world analysed in this paper reinforce that antibiotic resistance is a world-wide phenomenon, making it difficult to find a one-fit-all solution. Nonetheless, it is important to continue the efforts to mitigate this crisis, regardless of nationality or specialty.

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Bacteriophages – allies or enemies of humans?

Hubert Kasprzak¹, Filip Borkowski¹, Adam Jasiura²

1. Faculty of Biological Sciences, University of Wrocław
2. Wrocław Medical University

INTRODUCTION

Recent advances in viral metagenomics allowed scientists to recognize the most abundant organisms in the biosphere. They live in entirely different environments – from the deep ocean to the human intestine. Today, phages are believed to have a huge impact on the environment by affecting the biology of microorganisms. Phages are important for researchers since they are useful in better understanding of horizontal gene transfer, novel therapeutic agents, as well as they can be applied as diagnostic tools in genetics. Phages' genomes are composed of either DNA or RNA, which may be double or single-stranded. Most of them have a dsDNA genome packed into a capsid, which is connected to a tail [1].

Although phages inhabit different environments and show diversity at the nucleotide sequence level, even if no sequence homology exists between them; there are strong similarities between their structural proteins [2].

This phenomenon could be explained by convergent or divergent evolution. It is suggested that viruses form polyphyletic lineages, which sets them apart from cellular life forms, which had a monophyletic origin. BTV (Bluetongue virus), a eukaryotic virus belonging to the *Reoviridae* family, and *Cystoviridae* phages *phi 6* share a similar inner coat protein. Their genome is segmented and packed in a double-shelled capsid [3].

Another example is MCP fold (major capsid protein) from *Tectiviridae* phage PRD1, which is highly similar to mammalian adenoviruses and archaeal virus STIV [4,5].

All of these organisms originated differently, and they can variously impact host populations.

In this paper, we review the possible interactions between the bacteria and bacteriophages as well as their consequences to the phage therapy appliance in the current medicine.

LIVE CYCLES OF PHAGES

Bacteriophages have several possible life cycles. One of them is lytic cycle, in which they infect and kill the host cell. It is a predator-prey interaction, where predator is shaping bacterial population dynamics and exerting a selective pressure on the victim. Thereby, phages influence the pool of genes of the victim's population. Experimental evidences have also shown that both bacteriophages and hosts populations fluctuate over time [6], which is typical for predator-prey dependencies. Bacteriophages can also produce a stable relationship in the lysogenic cycle. Viral genome can be integrated with the host's, allowing the organisms to interact as commensals. However, viral genetic material can exist as plasmids within the host cell as well. Plasmids may carry genes that are not expressed by the host and thus increase its adaptation. This type of interaction is called proto-cooperation. It is observed in CTX ϕ bacteriophage, which carries the genes required by *Vibrio cholerae* to produce the cholera toxin [7]. However, it is possible for bacteriophage to stop its development in the host cell without either killing the host in lytic cycle or stable replication and maintenance in the cell line in lysogenic cycle [8]. In this situation, bacteriophage awaits until appropriate condition occurs to choose either lysogenic or lytic cycle.

BACTERIOPHAGE IN ANIMAL-ASSOCIATED MICROBIOME

The human body is densely colonized by microorganisms, as they occupy up to 54% of the total weight of human feces [9]. The phage interactions in the animal-associated microbiological system are even more complex since there occurs a correlation not only between the phage and the bacterium but also the environment of the animal organism. Phages can be digested by enzymes or macrophages, transported through the blood, or between organisms. In the human colon, Coliphages can be found associated with anaerobic microbiota – especially *Escherichia coli* [10]. Under normal conditions, they appear minorly. Although, in patients with internal diseases or immunodeficiency, their number was considerably higher [11].

Moreover, *Campylobacter jejuni* bacteria can be found in the intestine of the domestic chicken. They are subjected to selective pressure by the phage related to them. The natural selection leads towards increased phage resistance, however simultaneously bacteria have reduced the ability to colonize the intestine [12].

CONTRIBUTION OF PHAGES TO BACTERIAL FITNESS

Earlier studies focused on the destructive function of virulent phages, neglecting the ecology of temperate phages [13,14]. However, modern discoveries show that the role of temperate phages can be beneficial from a bacterial point of view, and defense against them can reduce bacterial adaptation. CRISPR-Cas is a well-known method of protecting bacteria against phages, which has found wide application in genetic engineering. It involves introducing short segments of phage DNA into the genetic material of bacteria, called spacers [15]. Therefore, during the next infection, it becomes possible to recognize foreign DNA by bacteria and to neutralize the bacteriophage.

Phage transmission can occur in two ways - through a lytic and lysogenic cycle. In lytic cycle the bacterial cell breaks down and the virus particles are released, therefore it can be called a horizontal transmission. Whereas, in the lysogenic cycle the virus integrates with bacterial DNA, becoming a prophage, thereby it can be named a vertical transmission [16].

Acquiring new spacers is much more efficient if the pre-existing spacer is partially complementary to the phage. This process, known as "priming," is widespread in CRISPR-Cas type I systems and provides protection against phage mutants that overcome host resistance by point mutation at their target sites [17,18]. However, this incomplete alignment can trigger immunopathological reactions when the virus is in the lysogenic cycle. The reason is that in this process not only the phage DNA is damaged, but also the host DNA, which leads to the release of the SOS response [19]. To study the evolutionary consequences of such events, Rollie et al [20] conducted an experiment in which *Pseudomonas aeruginosa* (strain PA14) were exposed to the temperate phage DMS3 and its virulent version - DMS3vir. It turned out that wild-type bacteria showed higher adaptation in the case of lytic infection, but in the case of lysogenic infection $\Delta cas7$ mutants with the impaired CRISPR-Cas system gained advantage, unless the virus carried the *acr* genes that suppress the immune system of the host. Therefore, in this context, *acr* genes not only provide phage benefit both during horizontal [14,21,22] and vertical transmission, but they are also beneficial to the host by preventing autoimmunity [20]. The confirmation of the Rollie et al. hypothesis that such results are caused by incomplete matching of spacers to the phage was an experiment with the $\Delta cas1$ PA14 mutant, which is not able to acquire new spacers, but is fluent in detecting and destroying complementary DNA – which is called an interference. Thereby, in case of wild-type bacteria, the percentage of mutant bacteria carrying prophage decreased in the initial period of infection.

Note that the CRISPR - Cas type I system in *P. aeruginosa* cannot distinguish between phages that enter lytic or lysogenic cycles even despite the opposite immune system reactions in both horizontal and vertical DMS3 transmission. Therefore, it can be concluded that moderate phage infection can lead to a rapid loss of CRISPR-Cas immune systems from bacterial genomes due to immunopathological effects [20]. Further, bioinformatics analysis suggests that in nature probably non-adaptive action of CRISPR-Cas against moderate phages is likely to occur. In addition, it has been shown that about 60% of all bacterial genotypes do not have CRISPR-Cas systems, and differences in possession of this system are noted even among closely related strains. This indicates that these systems are often obtained and lost from bacterial genomes [23]. In addition, high frequencies of autoimmune events suggest that this is probably a major factor in the evolutionary loss of CRISPR systems in nature [20].

Another example of a beneficial interaction between a bacterium and its temperate phages is the interaction in general of *Roseobacter*, in particular *Sulfitobacter sp.* (Strain CB2047) with two genetically similar phages – ϕ -A and ϕ -D examined by Basso et al. [24]. These phages have different characteristics – CB-A (bacterium carrying prophage of ϕ -A virus) has a higher frequency of spontaneous induction of prophages, as the doubling times of CB-D (bacterium carrying prophage of ϕ -D virus) and CB-A in liquid culture are respectively 75 and 100 minutes. Consistent with earlier evidence that cell lysis increases biofilm formation, CB-A produces twice as much biofilm biomass than CB-D. In the case of planktonic growth, CB-A prevails over CB-D in a 3:1 ratio. However, during biofilm growth the ratio was opposite. These results suggest that genetically similar phages may have a different effect on the competitiveness of their shared hosts in different environmental niches, probably due to the complex form of phage-mediated allelopathy [24].

Temperate phages offer bacteria an increase in resistance to homologous phages [25-28], as well as an increase in host virulence due to toxins encoded by prophage [25,29,30]. In earlier studies, phages were treated as "time bombs" whose ignition could be due external factors damaging to the host's DNA. Such events would lead to the expression of lytic genes encoded by prophages [29,31]. However, it turns out that internal factors can also favor the transition to the lytic cycle. This process is referred to as spontaneous prophage induction (SPI) [30]. Recently, the positive effect of SPI on bacterial adaptation, such as the release of extracellular DNA, which may be important for biofilm formation [30] and phage production as a competitor with susceptible hosts has been noticed [32]. Lysogens are thought to be common in marine environments [33,34], where about half of the sequenced bacterial genomes contain prophages [27,35].

Each of the two lysogens exhibits resistance to superinfection by phage particles of identical genotype: CB-D is resistant to ϕ -D infection, and CB-A is resistant to ϕ -A infection. In general, each strain is susceptible to lytic infection with a different phage genotype, which is accompanied by the induction of prophage already in the cell, although sometimes phage substitution may occur. The mechanism of such substitution has not been known yet [24].

Viral lysis of microbial cells in marine ecosystems leads to quantitatively significant effects on food webs and biogeochemical cycles [36]. Superinfection with other phages is a biotic factor affecting the induction of prophages, probably by inducing an SOS response [37,38]. It is not known, however, whether lytic infection and prophage induction occur simultaneously in a single cell or in different subpopulations of cells, one of which is lytic infected by a new, exogenous phage, while the remaining cells induction of previously resting prophage occurs [24]. It is possible that a sub-population superinfected with one phage could communicate with uninfected counterparts, thus initiating lytic induction, as described for Bacillus phage [39].

Therefore, maintaining both types of phages in a population can be beneficial for a given host for many generations in marine ecosystems. The challenge remains to develop tools to track individual cells and viral particles to better explain the dynamics of each player in this complex interaction [24].

THE SPATIAL COMPLEXITY OF THE ENVIRONMENT HELPS BACTERIA AVOID ATTACK BY VIRULENT PHAGES

In the previous paragraphs, we discussed the latest discoveries regarding the ecology of temperate phages. However, research on virulent phages also reveals new mechanisms, although in this case there can be no longer any positive effects on hosts. The role of the complexity of spatial structures on the millimeter and submillimeter scale has long been overlooked in research on virulent phages [40]. Eriksen et al. conducted experiments with bacteria and their phages in environments of varying spatial complexity. Their results revealed that the additional heterogeneity of space, even in a seemingly homogeneous environment, delays bacterial extinction caused by phage activity, because different regions of space experience different phage densities. Spatially diverse environments increase bacterial survival and make the bacteria survive 100-fold higher phage density, and the spatial structure at the microcolony level allows survival with phage density up to 1000-fold higher. In addition, space has been shown to significantly increase bacterial survival at low initial bacterial density, but

survival at high initial bacterial density depends on the delay time between phage infection and cell lysis. Gene expression diversity may be an additional obstacle in the fight against bacteria [41-43]. The ability of phages to penetrate bacterial colonies is likely to depend on several factors, such as cell geometry, colony packing density, and whether the cells form a biofilm [40]. Therefore, learning and effectively modeling ways of bacterial avoidance of phages will improve phage therapy parameters as well as better use of bacteria in industry and in human gut reimplantation [40].

BACTERIOPHAGES USAGE IN FIGHTING BACTERIA

The therapeutic potential of bacteriophages has been already noticed in the early 1920s, when they were used to treat dysentery. Developed mainly in France, Georgia, Russia and Poland, until the end of 1940s phage therapy had burgeoned, however later it was displaced by commonly used antibiotics [44]. Though, bacteria are constantly subjected to the selective pressure and through years of antibiotics administration they were acquiring the resistance. Therefore, today scientists come back to the phage therapy, as a potentially effective method to fight nuisance antibiotic-resistant bacteria. Here we review several innovative human trials, in which bacteriophages were used to treat the bacterial infections.

Phage therapy is successfully used in the treatment of chronic otitis caused by antibiotic-resistant *Pseudomonas aeruginosa*. In 2009 a controlled clinical trial was performed using *Biophage-PA* on 24 patients, whose disease lasted at least two years. In the trial, 6 phages were selected – BC-BP-01 to 06, based on the bacteria-sensitivity, which were mixed and administered to 12 patients (treatment group) through 2 weeks. As a result, there was a significant clinical improvement observed in the phage-treated patients. It has been confirmed in VAS (Visual Analogue Scale) scores measured by patients as well as clinicians. Mean total VAS scores were statistically significantly lesser, with the highest improvement about day 23 – which is the mean phage replication duration. Neither severe side effects nor significant adverse changes in the audiometry were noticed. Despite the necessity for the treatment repetition every 3 weeks in patients with chronic otitis media, phage therapy has improved their condition. Phages have reduced the number of *P. aeruginosa* organized in a biofilm, which was previously undefeatable by antibiotics and the immune system [45].

In 2019 a study in Australian hospitals was performed, where patients with severe infections, caused by *Staphylococcus aureus*, were instituted a therapy of three bacteriophages from the *Myoviridae* family – AB-SA01. It was the first study to administer an intravenous

phage therapy in severe sepsis. There were selected 13 patients aged 21-87 with ongoing for at least 2 consecutive days *S. aureus* bacteremia, whose predicted six-month mortality ranged 10-87% (average 44%). All of them has been provided with antibiotic therapy of flucloxacillin, cefazolin or vancomycin with prospective addition of ciprofloxacin and/or rifampicin. In the result, 8 of 13 patients (62%) showed clinical improvement after 14 days of therapy. However, 5 patients (38%) died in the first 28 days. In 11 of 13 patients (85%) a reduction of inflammation markers was observed, during or soon after the treatment. This study showed phage therapy is a safe and well-tolerated therapy in patients with severe sepsis. None of the new adverse effects were observed, including fever, hypotension, and rash, after the phages appliance. It has been noticed that after intravenous administration, phage distribution in the organism reached an effective level after just one dose. Phages were detectable up to 12 hours after the administration, through the whole therapy duration, with the highest escalation in days 7-10. Bacterial DNA has been eradicated within days to a week from the treatment beginning in the majority of patients (62%). However, based on this study, it is impossible to evaluate the benefits of phage therapy in addition to antibiotic therapy. Further studies, with selective groups of patients comparing exclusively phage therapy and antibiotic therapy, are required [46].

Moreover, phage therapy can be instituted to treat unhealing ulcers in patients with diabetic foot and osteomyelitis, as a result of antibiotic-resistant bacteria infection – mainly *Staphylococcus*. A series of studies have been conducted with Eliava BioPreparations' *Sb-1* bacteriophage, known from the anti-staphylococcal properties. Six patients, previously treated unsuccessfully with antibiotics, were selected, whose ulcers were mild to moderate severity and localized within toes; as well as the amputation was indicated if the improvement was not achieved. Phages were administered into the ulcer cavity and kept undried for 48h. Therapy was applied 2 times with a weekly interval. As a result, in all patients, an improvement was observed. The administration of phage therapy led to the decrease of the bacterial biofilm, and therefore to the reduction of inflammation and improvement of the microcirculation. Due to the phage therapy, ulcers healed sooner, not progressing to the severe infection, which would require an amputation – thereby conserving patients' extremities [47].

DISCUSSION

In the era of constantly increasing the number of antibiotic-resistant bacteria, phage therapy can be a self-reliant, safe therapy. Further, in case of infections with antibiotic-sensitive microorganisms, it can be not only a mere but also a synergizing method of treatment in co-

therapy with antibiotics. Recent studies of the bacteriophage ecology show however that phages not always are allies in humans' fight with bacteria. Temperate phages can positively influence their bacterial hosts and therefore impede the treatment. On the other hand, recognition of molecular mechanisms of viral virulence may allow the modification of temperate phages with the genetic engineering to make them virulent to bacteria. Moreover, complex interactions are important not only between the phages and bacteria but also animals within the bacteria live in. It is crucial to shaping bacteria-killing effectivity by virulent phages in complex spatial heterogeneity, since due to it, there may be a need to use even a thousand time higher phage dose for the therapy to result successfully. Furthermore, phage therapy requires deep personalization – it is pivotal to select the phages to which bacteria are susceptible, and simultaneously characterized by adequate life cycle and non-toxicity to humans. These restrictions limit the phage therapy usage, as searching for proper phage-bacteria models emerge to be difficult. Therefore, further studies are important to obtain the greatest and widest potential of phage therapy in fighting bacterial infections. However, other areas of phage appliance are worth forward exploration as well – like biotechnology and microbiota restoration therapy. In this case, temperate phages may turn out profitable, increasing the fitness of bacteria, which are beneficial for humans.

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Ecological aspects of aging

Borkowski Filip¹, Kasprzak Hubert¹, Jasiura Adam²

1. Faculty of Biological Sciences, University of Wrocław, Wrocław
2. Medical University Piastów Śląskich of Wrocław, Wrocław

INTRODUCTION

Aging is an irreversible process that occurs at all structural levels of life. Therefore, since it disrupts the body's balance, it reduces the ability to respond to the environmental stress [1].

In the consequence functioning of the body is impaired and the progressive accumulation of harmful cell changes leads to their death. This subject, however, is not completely obvious. Despite the fact, bacteria also age, their death does not appear as a natural end to their existence. Interestingly, simple animals such as jellyfish, sponges, and corals, as well as more complex ones, like hydrozoa representative – *Turritopsis nutricula*, often show longevity and potential immortality [2]. In plants, this phenomenon is also widespread as the oldest plant genet of *Lomatia tasmanica* is 43,600 years old [3]. Although the aging is inevitable, its effects can be successfully delayed and reduced. A good example of this is the 30-year extension of the average age of people in the 20th century in developed countries.

THEORIES OF AGING

Over the years, scientists have come to many theories explaining the aging process. These include: poisoning theory, wear-and-tear theory, cross-linking theory, Hayflick's limited cell divisions theory, Orgel's error catastrophes theory, shortening of telomeres, somatic mutations theory, mitochondrial theory and free radical theory [4].

The most popular one is the shortening telomeres theory; however, it is burdened with many problematic issues. Telomeres are repetitive nucleotide sequences found at the end of the chromosomes, which are not fully copied during the replication. Studies show that yeast cells of *S. Cerevisiae* have DNA telomerases - nuclear enzymes which add the telomere sequences,

hence the telomeres are not shortened in subsequent divisions. Nevertheless, these organisms have a limited number of cell divisions. Therefore, it is assumed that there are more indicators informing the cell when it should stop dividing. The main determinant of the body's resistance to external stress is its ability to repair DNA. Unfixed mutations not only lead to an accelerated aging, but also underlie many inherited diseases and cancerous transformations [5].

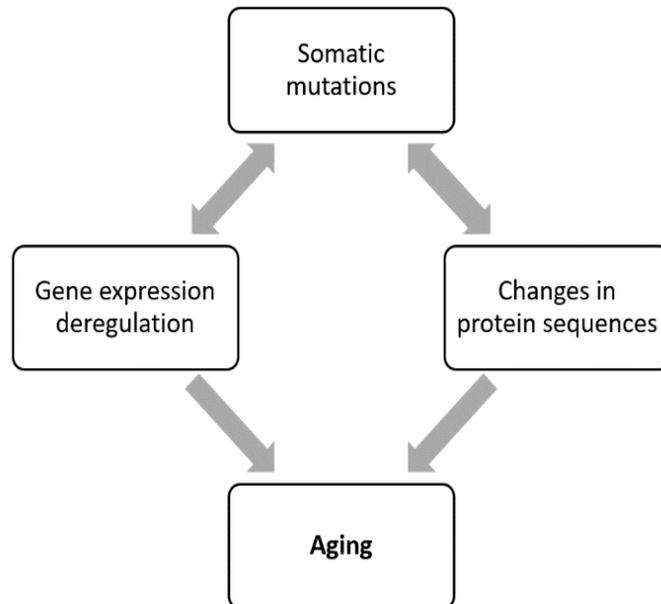


Figure 1. Model of the catastrophic error theory.

Although there are various studies conducted to prove different theories, many of them have been already confirmed, as the whole process is caused by many factors internally related to each other. In the mid-twentieth century, two theories were proposed to explain the aging process. One of them was the model of two-hit, developed by a physicist Leo Szilard, which is still popular today. It shows that somatic mutations accumulate in the genome, disrupting the proper expression of the genes, but both the genetic and cellular redundancies of the organism prevent impaired gene expression leading to aging, until they are completely spent. About the same time Leslie Orgel proposed a different theory of aging. In contrast to Szilard's suggestion of a constant rate of DNA mutations occurrence, Orgel attributed aging to the exponential increase in proteins' structure errors. Since proteins are responsible for the translation, decreasing their architectural fidelity can result in the positive feedback loop of the incorrect synthesis. Thus, the exponential escalation of this process leads to the increased aging. However, this theory has not been proved yet, but it may be caused by the technical limitations [6]. It may be worth combining some elements of both Szilard and Orgel's models, since the

catastrophic error theory explains why the number of somatic mutations increases exponentially with age and what role they play in the aging promotion (Fig. 1).

To fully understand the mechanisms of aging, physiological, molecular, as well as evolutionary approach should be applied. In this part of the work, we will present general patterns of the evolution of aging and consider the influence of the reproductive strategy on the survival of the Y chromosome and men's viability.

HOW DOES THE EVOLUTION SHAPE THE LIFETIME?

Recent findings show that basic assumptions about the functional basis of aging, such as the molecular damage caused by reactive oxygen species generated in metabolic and immune processes, must be reconsidered. New hypotheses, such as the hyperfunction hypothesis i.e. carry-over of activity of early life programs for growth and reproduction into later life, are gaining in importance [7].

Since the beginning, the evolution of aging has been attributed to compromises in response to the patterns of external mortality in adults. It emerges as a by-product of selection and leads to maximizing reproduction. This compromise occurs because mortality originating from the external sources, such as predation or random causes (i.e., external mortality), weakens natural selection in later life. With high external mortality, few individuals survive to reproduce at a later age, therefore, significantly reducing the role of selection which promotes longevity. This allows the accumulation of harmful mutations, whose effect manifests at a later age of individuals. It also enables favoring the fertility at an early age at the sacrifice of the longevity, which is called antagonistic pleiotropy [7]. Comparison of the infant mortality rate in the US in the early twentieth century to today's shows that there is a relevant decrease in external mortality in human populations when good access to medical care is provided [8]. This creates new conditions for natural selection, which can nowadays favor "longevity genes", not in the context of increasing the number of offspring, but care for grandchildren and even great-grandchildren, especially in the case of women's inheritance in the feminine line [9].

More recent publications draw attention to linking the condition of individuals with their survival. This may apply not only to factors with little impact on the human, such as the ability to escape from the predator, but also the significant ones faced by people, such as the resistance to diseases and stress. Studies on *Caenorhabditis elegans* have shown that when increased mortality was induced by heat shock (i.e. condition-dependent), life expectancy increased in subsequent generations, while shorter life span evolved when mortality was

random. In addition, Shokhirev and Johnson's research suggests that high external mortality promotes longevity when the cost of reproduction is high and the access to resources is unlimited. Contrarily, high external mortality would relevantly shorten the life span when resources were limited and reproduction was inexpensive. Moreover, the epigenetic factor is also greatly important, because such changes may affect the features of several generations [7]. It is also worth paying special attention to the recently discovered impact of the Y chromosome on longevity.

WHY IS IT BENEFICIAL FOR MEN TO BE FAITHFUL?

The human X and Y chromosomes were formed about 200-300 million years ago in eutherian mammals. Sex chromosomes transferring sex-determining genes are subject to unique evolutionary forces and play a significant role in many evolutionary processes, such as speciation, adaptation and genomic conflict [10]. There have been many evolutionary models proposed to explain Y degeneration. Their common feature is that the net effectiveness of natural selection is strongly reduced on the non-recombinant chromosome. Oppositely, in a recombinant chromosome, selection can work independently of each mutation. However, in case of recombination absence, selection works on whole chromosomes. The whole chromosome will be fixed in the population if a favorable mutation appears on it, or the whole chromosome will be deleted if it carries a harmful mutation. There are several following factors, which relate to this issue.

Muller's ratchet mechanism refers to the irreversible accumulation of harmful mutations in a finite non-recombinant population. In finite populations, chromosomes can accumulate mutations due to random effects. Recombination at X chromosome allows the reconstruction of mutation-free chromosomes, whereas this genome loss is irreversible to the non-recombinant Y chromosome and preserves a specific harmful mutation on Y.

In genetic hitchhiking the newly created beneficial mutations can occur on the chromosome which also contains harmful mutations. By recombination, preferred alleles can be fixed on the X chromosome without dragging along the associated deleterious mutations. However, on the non-recombinant Y chromosome, the preservation of the preferred mutation simultaneously preserves the linked deleterious mutation. In genetic hitchhiking it is required for the selective advantage of the preferred mutation to outweigh the effect of the combined harmful allele so that the Y chromosome containing the preferred mutation has a net selection advantage. If it is not, rube in the rubbish occurs.

In [ruby in the rubbish](#), Y chromosomes may undergo a smaller adaptive evolution in relation to the X chromosome, due to the connection of beneficial alleles with harmful mutations. Preferred low-performance mutations can be detached from linked, highly deleterious mutations by recombination on the X-chromosome and consolidation in the population. However, in the case of non-recombinant Y chromosomes, they will be eliminated by purifying selection. Genes linked to the X chromosome will continue to adapt and introduce beneficial mutations, while the Y chromosome will lag behind. Ultimately, it may be beneficial for a male not to express already incompatible Y-conjugated genes and silence or deactivate them [10].

Transmission limited to males implies that Y is an ideal part of the genome to transfer genes increasing the efficiency of males, because mutations beneficial to males are always transmitted by the sex in which they are beneficial and protected against counter-selection in females if they are sexually antagonistic (i.e. good for males but harmful to females). Thus, predictions about the content of the Y chromosome gene are clear – gene degradation predominates, and the surviving genes associated with Y are enriched with features beneficial to men.

However, while the general properties of these models are well understood theoretically, their relative contribution to the observed degeneracy of Y chromosomes in natural populations is less pronounced. Significant mechanisms are described below. The process of Y chromosome depletion may suggest that further gene loss will lead to the final disappearance of the human Y chromosome. These predictions are based on the model of the rate of gene loss from the Y chromosome. However, recent theoretical and experimental studies have clearly showed that Y degeneration does not occur in this simple, linear way and disproves sensational claims about the complete disappearance of the human Y chromosome [10].

Lobo and Onody showed in computer modeling that the Y chromosome degenerates, even if both XX and XY pairs do not recombine. They tried to explain these phenomena by specific differences in the inheritance of the sex chromosomes – the Y chromosome never experiences selective pressure in the female body, whereas the X chromosomes spend one third of the time of evolution in the male body and two thirds in the female body. Exclusion of recombination between sex chromosomes is insufficient to induce degeneration of the Y chromosome. It is rather a specific reproduction strategy that is responsible for this process [11].

Biecek and Cebrat, using a computer simulation, showed that in panmictic populations, when females (XX) are free to choose a male partner (XY) for reproduction from the entire

population, the Y chromosome accumulates defects and ultimately the only information it carries is the male's determination. As a result of the shrinking of the Y chromosome, male genomes lose one copy of the X chromosome information, resulting in males burdened with higher mortality, also observed in human populations.

If the model assumes that the presence of a man is indispensable at least during the pregnancy of his partner and he cannot be seduced by another woman at least during one reproduction cycle – the Y chromosome retains its content, does not shrink, and the life expectancy of women and men is equal. Thus, the Y chromosome shrinks not because of being in one copy, without the possibility of recombination, but because it is under weaker selection pressure. In panmictic populations without the need to be faithful, a significant proportion of males are unnecessary and can be eliminated from the population without reducing its reproductive potential [11].

SIRTUINS – A KEY TO AGING INHIBITION?

Recently there are more and more studies conducted focusing on the sirtuins functions, especially their influence on cell aging inhibition and, due to this fact, their possible usage in therapy of age-related diseases. The family of SIR2 proteins (Sirtuins), described as Silent Information Regulator 2, are enzymes classified as NAD⁺-dependent deacetylases. Initially discovered in *Saccharomyces cerevisiae* yeast, are currently found to be present in nearly every organism [12].

Research has proved that decreased sirtuin level in *Saccharomyces cerevisiae* can lead to many metabolic disorders, related with DNA repair, recombinational silencing and cell senescence [13]. It has been shown that prolonged caloric restriction stunts aging processes by inducing the sirtuins genes expression. However, this effect was not observed in case of SIR2 genes damage, whereas it was increased in overexpression [14].

Sirtuins have been divided into 5 classes: I-IV and U (undifferentiated). Homologues present in prokaryotes were assigned to class II and III, the eukaryotic ones to class I-IV, while the U class includes enzymes produced by gram-positive bacteria and some archaea. In *Saccharomyces cerevisiae* beside Sir2p there can be distinguished 4 more sirtuin genes sequences: Hst1-4. Whereas in mammals there were 7 homologues found so far named SIRT1-7. SIRT1-3 form class 1, SIRT4 compose class 2, SIRT5 – class III, as SIRT6 and 7 constitute class IV. Yet, no clear correlation has been observed between sirtuins function and this

classification. All SIRT1s comprise similar catalytic domain though and require NAD⁺ as a cosubstrate to their deacetylating enzymatic activity [15].

SIRTUINS STUNT AGING IN VARIOUS WAYS

SIRT1 is the best known of sirtuins and the most similar to the yeast Sir2p as well. Localized in the nucleus, it is responsible for DNA damage repair, gene silencing and genome stability. SIRT2 present in cytoplasm is functionally associated with tubulin and therefore with chromatin condensation during mitosis [16]. SIRT3-5 are located mostly in mitochondria, where they affect the mitochondrial enzymes regulating apoptosis, oxidative stress and cell energy metabolism [17]. SIRT3 and 4 improve the efficacy of the electron transport chain, whereas SIRT5 is able to activate mitochondrial superoxide dismutase. SIRT6 and 7 are mainly responsible for genome repair and maintaining its stability [18].

The most studied and known function of sirtuins is epigenetic modifications, the majority of which have positive impact on aging stunting. As the NAD⁺-dependent deacetylases, SIRT1s are responsible for deacetylation of lysine residues of N-terminal tails of mainly H3 and H4 histones, leading to more compact structure of chromatin and hence extending the lifespan of cell as a result of transcriptional repression. This functionality is related to SIRT1, 2 and 6 activity [19]. Furthermore, scientists observed that inhibition of SIRT1 in mammalian cells through hyperacetylation of H3K56 histone increases genome instability. It is inferred that elevated level of H3K56Ac (an acetylated form of H3K56) induces SIRT1 genes, what is also related to DNA damage repair due to its key role in binding newly synthesized DNA to chromatin [20, 21].

The influence of SIRT1 on the cell lifespan can be a result of histone methylation as well as DNA itself. Not only is it responsible for direct deacetylation of histones, essential for further methylation, but it also induces and activates histone methyltransferases. Moreover, by enhancing methylation of damaged DNA regions, it leads to epigenetic silencing, which can be a potential target for tumor research and the future therapy [22].

Through the regulation of transcription factors SIRT1 is affecting metabolism control, in particular, promotion of hepatic gluconeogenesis and β -oxidation of fatty acids. It is possible because of enzymatic modification of transcription factors by deacetylation. It results in decreased inhibiting activity of CRTC2, which through ubiquitination leads to its degradation, and STAT3, which turns less active without acetyl group, as well as FOXO1 and its cofactor PGC-1 α , responsible for increased stimulation after deacetylation. Therefore, the

maximalization of gluconeogenesis effectivity is achieved during fasting [23]. Activation of SIRT1 also enhances lipolysis and free fatty acids mobilization. By downregulating expression of nuclear receptor PPAR- γ , which stimulates adipogenesis and is present mainly in fatty tissue, the fat loss is favored. SIRT1 likewise impacts the carbohydrate metabolism, increasing insulin secretion by beta cells in pancreatic islets, as a result of reduction in UCP2 level due to direct suppression of genes encoding them. UCP2 are mitochondrial inner membrane proteins allowing proton leakage from the cytoplasm to mitochondria. Lowered UCP2 level affects intracellular ATP level in islets, elevating it, therefore increasing the insulin release [24,25].

According to the studies, sirtuins influence the inflammation. NF- κ B - a nuclear factor kappa-light-chain-enhancer, related to B-cells is a protein complex, essential in initiating immunological response to infection. It is also able to induce proliferation and angiogenesis, as well as inhibition of apoptosis, which may promote tumorigenesis. SIRT1 stunting the signalization of NF- κ B by deacetylation of its subunits leads to the decrease in tissue inflammation. However, NF- κ B downregulates SIRT1 by expression of factors such as miR-34a (a regulatory microRNA) or interferon gamma, thus enhancing inflammation [26].

In nuclear SIRT6 beside the deacetylation there has also been discovered ADP-ribosyltransferase activity. Researches show that in many tumor cells overexpression of SIRT6 generates massive apoptosis, requiring this enzymatic functionality. Nevertheless, such phenomenon has not been observed in nontumorous cells, which may turn to be crucial in studies on carcinogenesis [27].

SIRT1 impacts the cell aging also by regulating the activity of p53 – the guardian of the genome. The p53 is induced by many stress factors the cell undergoes and damage cumulating through the lifespan. It regulates DNA repair and, if it cannot be avoided, leads to apoptosis in order to protect the organism from tumorigenesis. It has been proved that SIRT1 through deacetylation of p53 inhibits its ability to promote apoptosis and, due to it, precocious cell death. Hence, it is suggested that SIRT1 acts as a balance regulator between death and repair of cells [28]. Although SIRT1 was initially classified because of those findings as a potential carcinogenesis promotor, it can stunt it as well. Antineoplastic activity is achieved through favoring maintaining the genome integrity and DNA repair by Ku70 protein, responsible for erasing DNA strand breaks produced by radiation. It has been shown, that Ku70 is not active enough in case of deficit of SIRT1 activation. Furthermore, restriction of tumorigenesis can be conducted by SIRT1 through inhibition of inflammation, as described above, as well as by influencing the localization of some regulators inside the cell, for example by deacetylation of

β -catenin and thus promotion of cytoplasmic localization of the nuclear-localized oncogenic form of β -catenin [29,30].

PHARMACOLOGICAL REGULATION OF SIRTUINS ACTIVITY

Sirtuins activity can be regulated through various endogenic and exogenic factors, which can be used to beneficially and selectively influence the cells. Since NAD⁺ is a necessary cosubstrate for SIRTs to fulfil their deacetylation, studies have been conducted to find, whether its level relevantly impacts sirtuins activity. It has been shown that the NAD⁺/NADH ratio is crucial, because excess of NAD⁺ increases SIRT activity, whereas NADH is its competitive inhibitor. One of the main factors impacting the NAD⁺/NADH ratio is energy supply status of the organism. Caloric restriction leads to displacement of this balance towards NAD⁺ and therefore allosteric activation of the majority of SIRTs, except SIRT4, which is inhibited by low energy supply [31]. Exogenic elevation of NAD⁺ level by its supplementation can result in beneficial influence on the organism functioning. It pertains to the therapy of cardiovascular diseases in particular, as SIRTs can protectively enhance the endothelial condition. SIRTs influence is also noticeable in neurodegenerative disorders, especially in Alzheimer's disease – an age-dependent disease as well, where SIRT1 acts neuroprotectively, decreasing β -amyloid production. However, in SIRT2 such an observation has not been made. On the contrary, it increases the risk of neurodegenerative diseases in case of overexpression. Having in mind the information introduced above about SIRT1 activity on energy metabolism, studies indicate a beneficial impact of caloric restriction in preventing aging, in context of sirtuins functions [18,32].

According to the studies, low energy effect can be elicited pharmacologically as well. Hence, such compounds act as indirect SIRTs activators. One of them is resveratrol, present in grapes peel and therefore in red wine. Resveratrol does not directly activate SIRTs, however directly inhibits some phosphodiesterases (PDE). Through enhancing the level of intracellular second messenger cAMP, and thus by Ca²⁺-dependent signaling pathway, it leads to the activation of AMPK, an AMP-activated protein kinase. This results in increased NAD⁺ level and elevated SIRTs activity. Furthermore, SIRTs can be activated concurrently through their phosphorylation by cAMP-dependent PKA (protein kinase A), due to the increased cAMP level, as mentioned above [33].

Beside activators, there have also been found exogenic SIRTs inhibitors. One of the products of NAD⁺-dependent deacetylation conducted by SIRTs is nicotinamide, a form of

vitamin B3, found to be a strong uncompetitive SIRT inhibitor. Therefore, to proper activity, SIRTs require expression of PNC1 genes, responsible for nicotinamidase synthesis, which hydrolyses nicotinamide to nicotinate acid, preventing from its excessive accumulation, and thus SIRTs inhibition [34,35]. Other SIRTs inhibitors are β -naphthols, with representatives such as sirtinol, splitomicin and much more stable and effective cambinol. These compounds are unselective inhibitors of SIRT1 and 2, although their impact on other SIRTs has not been observed. Their binding site is different from NAD⁺'s, which is suggested to decrease their potential toxicity. There have also been found inhibitors sharing their binding site with NAD⁺, such as EX-527, effective in low nanomolar range. This indole derivative, competitively prevents NAD⁺ binding to SIRTs, disallowing their activity, since they require NAD⁺ as a cosubstrate. SIRTs inhibitors, alike activators, can find a use in the therapy of age-related diseases. Through blocking SIRTs, resulting in increased antitumor-acting p53 activity, they enhance cells apoptosis. Therefore, by promoting cell death leading to stunting tumor growth, they find a potential use in treatment of neoplastic diseases, such as lymphoma or melanoma [36].

CONCLUSIONS

Aging is not only a physiological phenomenon, but also has significant ecological fundaments. Therefore, the evolutionary approach should be applied in medicine. Natural selection does not always promote longevity; hence knowledge about the evolutionary factors these processes are dependent on is a key to control them competently. It is possible that lower lifespan of men is presented by evolutionary paths of Y chromosome and prolonging men's life may be reached by modification of this chromosome. The future of aging counteracting lies in the recognition of epigenetics mechanisms and epigenome editing. Proteins like sirtuins, regulating many metabolic paths can turn out crucial in achieving increased longevity. Thus, further studies over substances inducing or inhibiting their particular activity are desirable.

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SELECTED PROBLEMS PSYCHIATRY/PSYCHOLOGY



Depression in personal and professional life of artists... An analysis of narration of the music business representatives

Tomira Chmielewska-Ignatowicz

Medical Faculty, Collegium Medicum, Cardinal Stefan Wyszyński University in Warsaw

INTRODUCTION

Depression is one of the most common civilisational diseases. It's ranked number 4 on the list of the most common diseases of the 21st century. According to the WHO data, until 2030, depression might become the most frequent cause of medical problems of the modern man, especially given that today over 350 million people are diagnosed with it [A].

Depression is an affective syndrome, characterised by mood decline. As prof. dr. hab. med. Jacek Wciórka points out, a typical clinical example of depression also consists of: activity inhibition, suicidal thoughts, intentions or attempts, anxiety, somatic symptoms, especially connected to the central drive system (sleep shortening, decreased appetite and libido), biological rhythm disorders and autonomous system activities disorders. There are many different kinds of depression, depending on the clinical picture, intensity and causes, that are not entirely clear [1].

Nevertheless, in medical terminology, there is a distinguished division between endogenous depression and reactive depression. The first one is associated with genetics, while the other one is a result of an occurrence of a painful memory, for instance a traumatic event, loss of a loved one or losing a job. There are many scientific and medical concepts characterising depression, that include it's psychosociological conditions. A. Beck's model of depression assumes, that certain mood disorders occur to a depressive person even before they become actually depressed. The disorders include: low self-esteem, lack of confidence in one's abilities and possibilities, negative estimation of one's experiences and actions, as well as a negative vision of the future. Among depressed people, traits such as introversion and schizothymia dominate and their premorbid personality is melancholic [1]. Some people's vulnerability to depression is also connected to certain personality traits. It has been proven that people of extraordinary creativity are more vulnerable to depression. An American psychiatrist – Arnold Ludwig – has conducted a survey on a group of 59 writers. The research has shown

that depressive episodes occurred to over half of the group [2]. When comparing other research groups of different professions, he proved that occurrence of depression is more frequent among poets (66%) than among military workers (5%). Another American psychiatrist – Joseph Schildkraut – has got similar results. He showed that 50 % of expressionist painters he surveyed have had symptoms of depression. What's more, studies conducted by the Help Musicians foundation from Great Britain, demonstrated that musicians are three times more vulnerable to depression than other professions. Lack of appreciation of their own art, difficult working conditions, financial instability and impossibility of planning their future are being given as the main causes of such state of affairs [B].

It's common knowledge that depression may lead to suicidal actions. As statistics show, only in Poland in 2016 there were more people killed by suicides than in car accidents. In the same year, the National Police Headquarters recorded 9861 suicide attempts, 5405 (nearly 55%) of which resulted in death (85,5% of which are suicides committed by men) [C].

Emil Durkheim emphasised that self-destruction should be considered as a social fact. The sociologist searched for the causes of such state of affairs in a tremendous disintegration of society, that is unable to prevent such incidents. He introduced a famous division of suicides into a few categories: egoistic (committed by people who don't feel any connection to the society), anomic (related to a disorder of structures that make up the essence of society), altruistic (committed by people who feel too much connection to a certain group that leads to lack of individuality) and fatalistic (in result of a tragic situation). Durkheim's theory assumes that the number of suicides has an inverse relationship with a degree of unity of a certain social group, to which an individual belongs [3].

Statistic data show that for each successful suicide there are 10-20 earlier attempts. The same studies demonstrate that women tend to have more suicide attempt than men, while at the same time, men's attempts result in death more often than women's.

People struggling with recurring or bipolar depression are the group that is the most vulnerable to suicidal tendencies. Among the general population, nearly 80% of suicides are results of depression [D].

AIM OF THE STUDIES

An analysis of many famous artists that had suffered from depression and committed suicides (among others: Robin Williams, Lee Thompson Young, Curt Cobain, Chris Cornell, Chester Bennington), as well as interviews of Polish artists about their experiences of

depression, published in a journalist Anna Morawska's book "Twarze depresji" ("Faces of depression"), were an inspiration for conducting studies on a group of 15 young musicians, that have just begun their careers in Polish and European music industry. Media actions, taken by many artists in order to attract attention due to the fact of publicly admitting suffering from depression, makes you think, whether a disease, that is depression, can be used by the artistic community for PR purposes? It's commonly thought, although perhaps stereotypically, that artists characterize by extra sensitivity and fragility. For that reason, it seems understandable to find out what are the opinions of young musicians on depression. Whether it is a serious obstacle in an artist's career (more serious and more frequent than among other professions) or is it a specific tool that could be used to manage one's PR in the world of attention-seeking and mass media.

Is depression actually an occupational disease of the artists community? May a situation of – even temporary – loss of popularity, lead artists to lean towards using the fact of vulnerability to depression as a mean of self-promotion and a marketing tool?

Another impetus to perform a qualitative research on a group of young musicians were Aleksandra Tomczak's pilot studies, conducted in 2017 under scientific supervision of author of this article [4].

It's worth to mention that statements of Anna Morawska's book's characters characters (painters, poets, writers, musicians or actor that either suffer or are close to people who suffer from depression), make a clear expression that the artistic community should speak about experiences of depression openly and publicly. Tomasz Jastrun – Polish poet and prose writer – points out that depression should be treated as a part of life, as it becomes more common due to the civilizational reasons [5]. Kamil Sipowicz (poet, writer and sculptor) however, claims that as far as breaking taboo on the topic of depression goes, there is a certain trend of being depressed right now.

"Our first time publicly talking about our depression was several years ago on the pages of "Wprost". Depression used to be taboo back then. It isn't now. The truth is that many people are depressed and, on the other hand, many decided that it's appropriate to be depressed. There's a belief that if you are not depressed, you can't be a good artist. I think that plenty of people say they're depressed to make themselves look good, that it's a good PR because nowadays it behooves them to be depressed" [5].

The writer's words may bring us to conclusion that admitting to being depressed by a public person, without any doubts, brings attention of the media. The judgement of validity of that person and their statement, however, depends on many different factors, such as the

situation they were in, their personal attitude towards depression and the way they talk about it. The key factor may be the medium the person used in order to publicly admit to be suffering from depression. All characters of the previously mentioned book, notice a need for educating others on the topic of depression and accustoming the society to it. Tomasz Jastrun and Kamil Sipowicz treat the memory of their experiences with depression as an impetus for their creative activities.

Having regard to the stories of people known from the media, as well as of characters of Anna Morawska's book, it seemed reasonable to find out young artists' opinions on whether artistic community's representatives are actually more vulnerable to depression, or is it a way of creating their media image. It also seemed to be important to ask the respondents' opinions on the following topics:

- Are artists more vulnerable to depression? If so, how? Why?
- Should the famous people speak about depression publicly? Why?
- Can an experience of suffering from depression be an impetus for creative activities?

MATERIALS AND METHODS

Qualitative method (in-depth interviews) was used in conducting this study. In-depth interviews, conducted with each young musician individually, allowed the interviewed to have more freedom of expression. As David Silverman noticed: *“the way we shape the research problem, inevitably reflects a connection (shown or hidden) to a certain model describing functioning of the world”* [6].

RESPONDENTS' CHARACTERISTIC

The number of respondents taking part in the research is 15 people – 8 men and 7 women, in the age varying from 21 to 36 years old (specific summary shown in the table nr 1). All respondents live in large Polish cities: Warsaw, Gdańsk, Białystok.

They're all connected to the musical community. 9 of them have musical education. Each respondent plays at least one musical instrument. Every one of the respondents performs publicly in solo or band concerts.

None of them have suffered from depression themselves but 9 have had direct contact with a depressed person.

Table 1. Respondents' characteristics, own study

	Name	Age	Musical Education	Musical instrument
1	Weronika	21	yes	Violin
2	Anna	28	yes	Voice
3	Piotr	35	yes	Saxophone
4	Adam	23	no	Drum kit
5	Grzegorz	29	yes	Saxophone
6	Piotr	30	no	Drum kit
7	Sebastian	38	no	Voice
8	Wioletta	26	yes	Piano
9	Monika	26	yes	Clarinet
10	Beata	31	yes	Piano
11	Maria	26	yes	Voice
12	Anna	28	no	Voice
13	Bogdan	33	no	Guitar
14	Zdzisław	36	no	Drum kit
15	Aldona	31	yes	Violin

The studies were conducted in December 2019 in places chosen by the respondents. Each conversations lasted between 30 and 90 minutes and was recorded using a tape recorder and then transcribed. The respondents were informed of the aim of the study. All respondents agreed to take part in the study. Every respondent was asked a question on their general thoughts on depression. A question about knowing a depressed person turned out to be a key question for the course of the conversation, as this specific fact differentiated respondent's attitude towards depressed people. The next step was connected to the musical environment – a question about respondent's view on the characteristics of the problem in the environment they live in.

RESULTS AND DISCUSSION

Without any doubt, the research was a peculiar experiment due to the fact that it was conducted on a small group. What especially caught our attention during the studies, was the fact of direct contact between each respondent and depression (their own or a loved one's). That factor generated the most extra questions, emotional involvement in answering questions and

created a need for going deeper into the topic. Those respondents who had contact with a depressed person, emphasised seriousness and social meaning of this disease. In their statements, they accentuated that depression is a disease that “pacifies” one’s life.

“It’s an event in that leaves a mark for the rest of your life... It never goes away (...) Such person cuts themselves off from the world, they don’t want anything.” /Adam, drum kit/

The respondent clearly highlights that depression is a chronic problem and is impossible to cure ultimately. It’s important that the respondent is close to a person who was suffering from depression.

“On the one hand it is a terrible, scary thing that temporarily excludes one from the society. Depressed people are being excluded because they don’t leave their houses, don’t work often... On the other hand, when you leave it behind, in some aspects, it makes you – perhaps not stronger – but lets you see many things from different perspectives.” /Adam, drum kit/

The respondent notices a positive side of taking up treatment of depression. He emphasises the essence of psychological, personal and social change of the way a person views the world around them after struggling with depression.

“To me, depression is based on the fact that the depressed person is misunderstood (...) A depressed person doesn’t feel anything. On the outside it may seem that everything is all right, while – in fact – on the inside, the person is broken.” /Weronika, violin/

“It’s a dangerous disease... because it’s silent, inconspicuous and because of that often unnoticed. People with depression are smiling and talkative on the surface... But when no one’s looking they can’t stand other people or themselves... it’s a prison.” /Agata, voice/

As can be seen from the statements above, each of the respondents had direct contact with depression in the artistic community, and each one of them knows personally a depressed person, but none of them was depressed themselves. The respondents’ descriptions of the disease were all very similar. Each of the interviewed showed awareness of the dangers and consequences of the disease, that is depression. The respondents, who didn’t have any contact

with depression (knowing it only from the media), interpreted the disease as a kind of specific weakness and not a disease:

“Everybody has their moments of weakness, kind of melancholy (...) Depression is a lack of ability of dealing with temporary failures. It’s a matter of personality, not a disease.”
/Bogdan, guitar/

“I’ve heard that people suffering from this disease feel that they’re in an impossible, no-win situation... Meanwhile, there’s always a way you can solve a problem, and when you’re struggling, you should try a different approach. (...) There’s always some medical excuse when you fail for some longer time... It’s a kind of explanation to themselves and others.” /Grzegorz, saxophone/

Respondents who didn’t know any depressed person directly, describe depression as a kind of sadness, while those who had contact with the disease, know that a person who hasn’t experienced depression is unable to understand how helpless and weak a diseased person becomes. They realise that it is a disease that one cannot face alone and that an intervention of their loved ones and prompt start of treatment is necessary.

It’s worth to notice that one of the respondents needed to emphasise that in her musical school students are under the constant care of a psychologist. Therefore, an intervention of psychologists and psychiatrists in the musical community is a common thing. In response to a question on whether – in her opinion – this type of specialist care is needed for this specific occupational group, the respondent referred to the scientific studies quoted before in this work, claiming that musicians are 3 times more likely to get depressed than other professions. She simultaneously emphasised her understanding for the necessity of constant specialistic psychological support dedicated to the creative workers that are constantly being judged by others.

The other respondents, who were asked a question on whether they think that artists are more vulnerable to depression, noticed artists’ specialness and sensitiveness.

“They’re being constantly judged. They’re more vulnerable to various emotional states, they experience and feel everything more intensely.” *“An artist may set themselves a standard so*

high that it all might become a loop and when there are also some difficulties in concerts... You need some support.” /Piotr, drum kit/

The respondent thinks that the depression is strongly connected to emotions that are artist’s “work tools” and that it might cause vulnerability to the disease.

“A musician is always being judged, it’s a restless rat race, envy, criticism, falsehood of the community... And that pressure that you’ve got to stay the best or you’re out of the game”. “A musician is always oriented towards judgement of everything they do... Not only the music they create but also what they wear and who they meet.” /Aldona, violin/

“Musicians must be tough rocks... Otherwise, they won’t be able to do their job publicly.” /Beata, clarinet/

The respondents emphasise the value of the constant pressure in the artistic community that is a factor predisposing them to depression. They all noticed that musicians set their standards too high. The respondents’ answers correlate with scientific studies that showed that the causes of depression among musicians include lack of psychical support in the community and lack of recognition for their own work [E].

“When it comes to musicians, it’s important they have distance to the world and to themselves. It’s very much needed in that profession... Otherwise you get depressed and damage yourself and the community” /Bogdan, guitar/

“When entering the stage we are conscious that some will like our work and some won’t and the comments may be unmeritorious, vicious... And that may lead to depression” /Zdzisław, drum kit/

The respondents noticed that musicians show a tendency to set themselves many additional psychical and personal demands, which often goes beyond their possibilities and may lead to depression (which is – according to one of the respondents – “a blockade, wall that people can’t see through to see how much potential is inside them”. /Aldona, violin/)

In the search of the causes of depression amongst artists, each respondent noticed existence of a common factor that increased artists' vulnerability to this disease, being never-ending competition between musicians.

“As a musician you have to be – first of all – mentally resilient, because on the stage even your colleagues-musicians judge everything, not always favourably. You’ve got to be able to distil the comments, as there is constructive criticism and criticism that brings nothing. It’s easy to say... but it often hurts...” /Aldona, violin/

“Artists are more vulnerable to depression because if something gets judged differently than we had assumed, it’s difficult to just shake it off... On the other hand, success may also be a reason for your friends to push you away due to envy.” /Maria, voice/

Additionally, the respondents brought attention to how important it is to a depressed person (especially an artist) to get support from another human. This confirms that the respondents are aware of the necessity of treatment and know that people that are alone with their disease are unable to handle it.

In the next part of the study, an issue of the need of speaking about depression in the media by famous people was brought up. The respondents noticed the emphasis of the function of authority of the famous people speaking up on the topics of diseases that are taboo.

“(...) When a person that’s famous in the media speaks openly about depression, a regular man dealing with the same thing feels that he’s not alone... And when people hear that some famous persons won the struggle with depression they probably have more faith in themselves, that they too may handle their disease.” /Aldona, violin/

“An artist speaking about their depression in the media may – with their attitude – show a way out of this situation.” /Bogdan, guitar/

The respondents noticed a positive impact of publicly speaking about depression by artists but mainly for the sake of raising awareness of this disease and calling on to seek help and treatment. This implies that the respondents not only see a professional success in one's fame but also the influence they have on motivating others.

It's worth to mention that only one of the responders saw depression as a way of creating one's public image:

"It's a popular disease, that is talked about in the media in order to attract attention."
/Grzegorz, saxophone/

The others admitted their lack of knowledge on whether anyone from the artistic community would promote their work through public declaration of diagnosis of depression.

"I see it this way, that people who speak up publicly in the media are the ones that have already won with depression (...) Perhaps only at the moment of defeating the disease one realise they were depressed?" /Piotr, drum kit/

The respondent finds public behaviour of famous people as authentic, as long as their statement is preceded by treatment or defeating the disease. He emphasises that only when one has distance to their depression, they may speak about it.

Additionally, the respondents who had direct contact with depression said stated that people actually struggling with depression don't want to talk about it at all, especially publicly. What's more, this disease turns out to be a creative blockade and not an artistic inspiration or an impetus for creative activities:

"It's hard to talk about depression when you're struggling with it... So when someone admits to it with ease, it's unbelievable to me and is used to attract attention." /Aldona, violin/

"If someone said that they wrote a piece during depression, it'd mean that it was over quickly."
/Grzegorz, saxophone/

The common thing between all the respondents is a belief that credibility of a diseased person can be judged to some extent, by the fact how easily a person talks about depression. A person who has trouble speaking about their experiences of depression publicly is recognised as credible. While someone who talks about it with much ease is recognised as a rather inauthentic person, who tries to use depression as a way to attract more attention.

The last issue brought up in this study is a question on whether an artist may find inspiration in depression. According to the respondents' answers, nobody noticed a cause and effect relationship between deep depression and inspiration. However, an additional common reflection came up on the topic of the meaning of emotions (as an artist's tool) and the disease (as an impetus for work).

“Assertion that one can get inspiration from the disease is rather for show. A depressed person is a wreck. Depressed people don't want to do anything but a lighter phase may result in reaching for an instrument. You can express what you feel, an autotherapy, forgetting for a moment, longing for the listeners.” /Aldona, violin/

“When struggling with depression, I don't think it helps in creative work. Only in the moment of defeating the disease. Artists tend to get inspiration from extremes.” /Piotr, drum kit/

“Experiences and attempts to express them through music are important. There's no way to play something fakely, the other person will always hear it. Various experiences make playing instruments more interesting, even if you have some technical lacks you can train it.” /Beata, clarinet/

The respondents statements prove that the inspiration usually is in the emotional experiences. They claim that that is the base of artistic activity, as due to outstanding sensitivity, an artist has predispositions to use emotions in creating art. It's worth to notice however, that the respondents see the source of inspiration in an interpretation of emotional experiences and not the disease itself. What's more, too much ease in sharing the information of struggling with depression by the famous is seen as a sign of lack of authenticity of their words.

CONCLUSIONS

The respondents noticed an existence of specific personal traits that predispose artistic community representatives to higher vulnerability to depression. The results obtained from this study show a correlation with scientific research showing much bigger frequency of getting depressed among musicians [B]. The fact of directly knowing a depressed person, turned out to be the most essential factor differentiating attitudes of the respondents to the judgement of a

disease that is depression. Thus, emotionality that accompanies the experience of contact with a depressed person greatly affects the view on the scale of the problem.

Inconclusive opinions of the respondents on the topic of credibility of the artists sharing the information of their depression publicly showed clear difficulties in unambiguous assessment of celebrities' behaviour (behaviour often generated by the media).

The results of the study showed compliance of opinions of the respondents on the topic of experience of depression as an inspiration for creative work, which is a proof for an awareness of the specificities of the disease and the difficulty of its diagnosis and treatment by the respondents. The recognition (by the respondents) of the influence the famous people have on others and their ability to spread information about depression shows necessity of correct use of images of the famous for the sake of raising effectivity of social campaigns for support for the depressed persons.

As it was mentioned in the beginning of this article, the study – on such a small group of responders – was merely a research sample and continuation of Aleksandra Tomczyk's 2017 research under supervision of this article's author. It was however, a deliberate intent, as statements of these people showed new research areas, supplementary to the original assumptions of this work. The results obtained from the study may serve as a great research potential for the scientific community of psychologists, psychiatrists working with artists, music pedagogues. It would seem to be very interesting to see an extension of this study to opinions of other artistic community representatives, such as painters, actors, dancers that have not experienced depression themselves and who have had depression or had direct contact with a depressed person.

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Animals in the therapy of psychosomatic diseases

**Paula Bigos¹, Konrad Dendys¹, Monika Hejnowicz¹, Weronika Mazurek¹,
Jadwiga Kuciel-Lewandowska¹, Aneta Hauzer¹, Jan Gnus^{1,2}**

1. Wrocław Medical University
2. Regional Specialist Hospital in Wrocław, Research and Development Center, Wrocław

INTRODUCTION

Animal-assisted therapy (AAT) is a newly evolving field that holds great promise and potential for many populations. It involves using close and natural contact with animals to assist in the healing of psychological, emotional and physical problems, rather than using them as sources of antibiotics which are better obtained from pharmacological sources [1]. This type of therapy is directed at people with disorders and deficits in the psychomotor sphere, for example children with Down syndrome, autism, cerebral paralysis, ADHD, muscular atrophy and adults with depression, schizophrenia, cardiovascular diseases, dementia, Alzheimer's disease, rheumatoid arthritis, high level of fear and anxiety and other diseases [2,A].

The creator of animal therapy is considered to be William Tuke, who was a director of an English psychiatric hospital. He showed in the seventeenth century that patients in psychiatric hospitals are more effective at controlling their emotions through contact with animals [A]. This view is still valid and used in therapeutic work. The most intensive development of AAT took place in the 20th century. In 1903 W. Flower Bucke published a collection of 1,200 children's essays that described dogs. The analysis of these texts contributed to the thesis that the animal may play a special role in the life of a lonely and suffering child [2].

The term "animal therapy" was first used in the 1960s by Levison who published his observations on friendship between animals and autistic children [A].

Dr. Boris Levinson accidentally used animals in therapy with children when he left his dog alone with a difficult child, and upon returning, he found the child talking to the dog [3]. Since then, animal therapy has been recognized as a method of supporting psychological therapy.

There is an increasing appreciation of the therapeutic function pets can enter in relation to mental, emotional and physical health. The first reports about the beneficial effects of pets on health was an early study of 92 heart-attack victims in which 28% of pet owners survived for at least a year as compared to only 6% of non-pet owners [4]. These reports led to increased interest in AAT and generated plentiful research on the positive impact of interacting with pets. Examples are many, among them: stroking dogs and cats, watching tropical fish in an aquarium, and even caressing a pet boa snake have been reported to lower stress levels and blood pressure [4]. The most valuable of these studies was a clinical trial in which hypertensive stockbrokers were randomly assigned to the group with pet or no-pet [4]. Six months later, studies showed a smaller increase in blood pressure during stressful situations in the group with animals than in the group without animal conditions [4]. Pet owners have more ambition, greater life satisfaction, higher self-esteem and lower level of loneliness and these are psychological benefits accrued from living with animals, that was confirmed in studies; animal ownership is also often combined better health and well-being, for example, among 11,000 German and Australian adults, pet owners were in better physical condition than non-pet owners, and they paid 15% fewer visits to the doctor, a potential savings of billions of dollars in national health expenditures [4]. Another epidemiological study of Chinese women found that pet owners slept better, exercised more, felt more physically attractive, and missed fewer days from work than women without pets. Further, these effects were particularly strong for individuals who reported that they were very closely attached to their pets [4].

It is very important to remember that animals can cause humans infection and trauma. There are over 200 different zoonotic infections, which can be spread by animals used in AAT; however their exact incidence has not been documented and remains unknown [B]. Similarly, there may be traumatic injury from animal bites or scratches, but how frequently this takes place as well as the impact of the events is uncertain [B].

Different types of animals are used in therapy: dogs, cats, birds, horses, llamas, alpacas, dolphins, rabbits, lizards, and other small animals can be utilized in AAT. Dogs are the most frequently used animals because of their training and sociability skills [2]. The most commonly used are so-called gentle breeds, e.g. Retrievers, Newfoundland, Collies, Bernardines, Huskies and German Shepherds. The decisive factor in their selection is the already mentioned gentleness and the ability to learn quickly. The type and selection of exercises involving dogs are adapted to the needs and diseases that the person participating in the therapy is struggling with [2]. Cats are often used for therapeutic purposes, contact with them helps to cope with loneliness, makes time more pleasant and soothes the mental state of people participating in

therapy. The therapeutic effect has especially stroking the cat and listening to its purr [3]. Horse-assisted therapy, which is called hippotherapy, is next often used in therapy. The rider experiences a specific interaction with the animal with which he shares a relationship and space during horse riding [2]. A connection is thereby established, resulting in satisfaction and motivation, which in turn mitigate pathologies. In horse therapy observations are made on the effects on the patient's neuromuscular system caused by the mechanical influence of the horse's motions [2]. Specific to the horse therapy is that the patient continuously receives impulses from the horse's walk, which leads to a relaxed perception of the body, balance, and coordination of movement [2]. Dolphins are gentle mammals, contact with them stimulates the release of endorphins, therefore they are also used in AAT [A].

The therapeutic effect have particularly ultrasounds issued by dolphins. These sounds stimulate the brain and improve the overall condition of the patient [A]. In addition, contact with the animal in the aquatic environment encourages movement in the water. Alpacas and llamas are these days also often used in therapy because of their ability to train them in a way appropriate for future contact with people and their impact on endorphin release [C].

THE ELDERLY

The health benefits of pet ownership are truly undeniable, especially among the elderly. Having a pet may provide benefits both for the mental and physical health of older persons. It nullifies the feelings of loneliness, boredom and social isolation. Several studies have reported that dog walkers show more physical activity than themselves before having a dog. What is more, dog walking can lower blood pressure and prevent other cardiovascular complications [B]. In fact, a lot of hospitals and elderly facilities have successfully instituted Animal Assisted Therapy in palliative care, Alzheimer's disease, and other dementias treatment.

At the Interdisciplinary Centre for Palliative Medicine of the University Hospital in Duesseldorf the ATT program began in 2014 [5]. The results after the first year of the program were reported. The median age of the participants was 65 years. The effects of the AAT sessions were mainly patients becoming more relaxed and willing to play with the dog. Furthermore, the animals helped them to slowly overcome their anxiety and simultaneously initiate social interactions. According to these results, AAT in palliative care appears as a valuable method, this should be further developed [5].

There are several studies that show a significant improvement in behavioral and psychological symptoms in demented patients treated with AAT. Most of these therapies

included dogs, but birds, fish and horses were also practiced [B]. A reduced amount of behavioral disturbance, improved ability to socialize, fewer signs of agitation, a positive influence on depressive symptoms are only a few of the described benefits in demented patients after some AAT sessions [6-9].

A particular form of AAT is a dog-assisted therapy (DAT). There are different studies that demonstrate the positive influence of a therapy dog on well-being and quality of life in persons with Alzheimer's disease. According to these, patients expressed feelings of joy, tenderness and affection while spending time with the dog. Moreover, they were able to behave like healthy, empowered, confident people by deciding what is good for the dog. Probably taking care of the dog let them recall some memories and experiences, on which these decisions were based. The treated persons appeared to be more outgoing and secure. It may be possible that they were able to find some pieces of their inner personality and were ready to express themselves. Spending time with the dog and caring for it gives patients the feeling of being important and valuable. While they are focused on and interested only in the dog's needs, their mind could be present [10,11]. However, most of these studies were not controlled and the certainty of the evidence was unclear. That is why the effectiveness of AAT in symptoms of dementia has been questioned. Some systematic reviews that include only controlled and randomized studies show no significant impact of AAT in self-care ability, agitation, Quality of Life, disorientation, social behavior or daily activities in patients with dementia. A slight reduction of depressive symptoms was found. Only one study found a slightly positive effect on apathy [12,13]. Undoubtedly there is a need to increase the certainty of the evidence, create standardized survey instruments and describe the adverse effects so a clear conclusion about the benefits of AAT in patients suffering from dementia can be made.

AAT can also be used in combination with other non-drug therapeutic methods. The integrated elderly play therapy (IEPT) aims to regain lost abilities and memories by participating in various plays. In many studies a combined intervention of AAT and IEPT was conducted. Most of the participants suffered from geriatric diseases. The results showed reduced symptoms of depression and increased self-esteem in elderly people living in nursing homes. These patients also expressed a higher amount of positive feelings and emotions, described by them as enjoyment, happiness, pleasure and excitement. Moreover, the elderly living alone demonstrated increased cognitive functions and also a lower level of depression. Thereby, the combined intervention of AAT and IEPT may have a significant impact on a healthy mental state. It gives more benefits than a single-intervention. As it appears as an effective, appropriate and interesting approach, it needs to be continuously expanded [14,15].

THE CHILDREN

Many experimental studies with children show the physiological, psychological, and emotional benefits of AAT (Rossrtti & King, 2010). Before therapy starts, goals are necessary to set. Treatment is often individual. During a session progress should be recorded in special charts. Visits should be scheduled with the patient's needs in mind (Pet Partners, 2012a). During the therapy a child can be invited to play with buckles, leashes or collars. Its aim is to improve the patient's motor skills. AAT can be used also to improve children's ability to sequence events by giving patients a container of treats and feeding small pieces of food to the cat. The patients benefiting from those activities are for example children with early-onset schizophrenia with estimated to be 1:30 000 children [16]. Furthermore animal-assisted activities like volunteering can also be beneficial.

AAT can be beneficial in many fields of pediatry, from mental to physical. The most common physical goals of AAT are improving motor skills, wheelchair skills, or balancing while standing. To mental goals belong attention skills, recreation skills, increasing self-esteem, reducing anxiety and loneliness [17]. It is extremely important, while many children suffer from some form of mental illness and this type of therapy can help a lot of them.

The most common animals in AAT are dogs. To be allowed to work with children they have to be specially trained. Experts say that allergy or fear of dogs are not contradictions to AAT with dogs [18].

AAT is mostly directed to children with Down Syndrome, Myelomeningocele, Children's paralysis, autism, delayed intellectual and motor Grover, blind or with ADHA [19]. Furthermore, it can be used with healthy children who must learn how to live in society. They learn through cleaning and feeding animals skills necessary in their adult life. The activities like petting and touching dogs can increase the secretion of endorphins, reduce stress, and improve patients' immune systems [20].

AAT could have an especially beneficial effect on children with Autism Spectrum Disorder (ASD). It is a developmental disorder characterized by difficulties with social interactions and communication and is a cause of behavioral challenges [D].

Animals have features that are not available to humans and at the same time can target the symptoms of autism. Animals provide a powerful multi-sensory stimuli: repetitive sounds, vibrant visuals, smell and pleasant touch [21]. Due to this stimulation and also due to the secretion of hormones as a response to interaction with the animal, the symptoms of autism are diminished. The most important benefits of AAT include improving the child's social

communication as well as limiting and controlling restricted and repetitive behavior. Furthermore, an animal companion not only motivates the child to further social interaction and communication, but also can be a point of concentration for the child in case of over-stimulating sensory situations at the same time [22].

The newest therapy with dogs is reading education assistance dog - READ [23]. It was used for the first time in Utah in 1999. It is directed mostly to children who have problems with reading and its aim is to improve reading ability, communication or interest in books. An important advantage of this method is the fact that a child is not feeling pressure to read to a dog as opposed to a teacher or a class. Meetings with dogs should last longer than 30 minutes and include a greeting, a warm-up, reading and at the end time to play with an animal. The study suggests it has a beneficial impact on reading education [E].

In conclusion, animal-assisted therapy has many benefits in the fields of pediatry. It improves not only the physical and mental well-being of young patients, but it can also help them obtain skills important in adult life, like reading.

CONCLUSIONS

Although the term "animal therapy" in medical literature was first used in the 1960s, the presence of animals has had a positive effect on human lives for much longer.

Animal-assisted therapy leads to several important benefits for patients suffering from various psychological, emotional and physical disorders, regardless of age. These benefits draw from many various disciplines, for example biology, behaviorism and psychology. Implementation of this kind of therapy can provide emotional support and stress relief, opportunities and motivation for physical activity and training as well as stimuli to learn cognitive self-care skills.

But most importantly it can improve social interactions and cultivate social skills. Interactions with animals can be both a therapy itself as well as a part of a traditional one. There is no limitation regarding which animal can be used in the therapy and thanks to that patients have a choice which type of therapy is tailored to their potential and which brings the most benefits.

There is no doubt that the benefits of AAT are far greater than the disadvantages. Therefore, research on this subject should be broadened and deepened, and the use of animals in this kind of therapy regulated by law, in order to be able to use it more often in medical practice.

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Suicide in the elderly - risk assessment and prevention

Marta Madej¹, Magdalena Pogorzelska¹, Krzysztof Wróblewski¹, Maria Gaj¹, Agnieszka Wichary¹, Jan Gnus^{1,2}, Krzysztof Dorna³

1. Wrocław Medical University
2. Regional Specialist Hospital in Wrocław, Research and Development Center, Wrocław
3. Pontifical Faculty of Theology in Wrocław

INTRODUCTION

In recent years we can observe huge progress in all fields of medicine. Lots of diseases incurable in the past now can be easily treated or at least their development can be inhibited. New medicines with improved performance and fewer side effects are produced. As a result, people live longer and life expectancy is increasing. According to the World Health Organization (WHO) between 2000 and 2016 global average life expectancy increased by 5,5 years, which is the fastest increase since the 1960s. In 2016 global life expectancy at birth was 72,0 years (74,2 for females and 69.8 for males), for Europe it was 77,5 years (80,8 for females and 74,2 for males) [A].

For the first time in history, most people can expect to live longer than 60 years [B]. However, aging has its limitations. Longer life doesn't always come with an extended period of good health. Most older people suffer from chronic diseases and have to face some limitations in physical or mental capacity. Some longitudinal research conducted in high-income countries has suggested that there may be a slight decline in the prevalence of severe disability, but in the past 30 years, no significant change in less severe disability has been observed [1].

A supportive environment could help to maintain a good quality of life, even for people with many health issues. Unfortunately, health systems are not ready for demographic transitions - levels of health professionals trained in geriatrics or gerontology are low and there is not enough emphasis on the education of caregivers [C].

Elderly people in many countries lack contribution to the life of society and they may feel excluded. Older people often have insufficient income security, limited access to social and

health care and at least 10% of them are victims of some form of elder abuse [D]. An increase in life expectancy leads to an increase in the number of generations in the family living at the same time. However, due to the cultural changes and migrations of younger members of the family, different generations are these days more likely to live separately than in the past. The proportion of older people living alone is rising dramatically, for instance in some European countries more than 40% of women aged 65 and over live alone [E]. Living alone has been suggested to be a risk factor of isolation and suicide, especially for men [2].

Suicidal behavior among older adults should be taken seriously. Elderly people are reluctant to share suicidal and depressive thoughts with their doctor, therefore it is difficult for clinicians to recognize the problem. Despite reduced symptom-reporting, rates of suicide increase across the life course [3].

According to WHO suicide rates are highest in persons aged 70 years or over for both sex in almost all regions of the world [F].

Many deaths by suicide could be prevented, but it requires a raise in awareness of this issue in society and improvement in the recognition and treatment of the vulnerable persons by primary care physicians. In this paper we analyzed the epidemiology of suicide in the elderly, ways to identify risk factors and prevention strategies.

MATERIALS AND METHODS

A literature search using databases such as PubMed and UpToDate was performed to find recent studies focusing on suicide in later life written in English or Polish language. Moreover, we analyzed statistical data from the World Health Organization (WHO) and the Polish Police regarding suicide attempts and suicidal deaths.

REVIEW, ANALYSIS

There is no single definition of suicide. Commonly suicide is defined as intentional self-destructive behavior leading to biological death. From a psychological standpoint suicide is a series of interrelated thoughts and deeds that can last for many years [4].

Erwin Ringel, an Austrian psychiatrist and neurologist who is considered one of the pioneers of suicidology, distinguished a set of features that are characteristic for a person who is about to commit suicide (presuicidal syndrome) [5].

The features are: experiencing anxiety, threats and feelings such as lower value and insufficiency, resignation, autoaggression, escape from real problems into fantasies about death, making suicide plans [5].

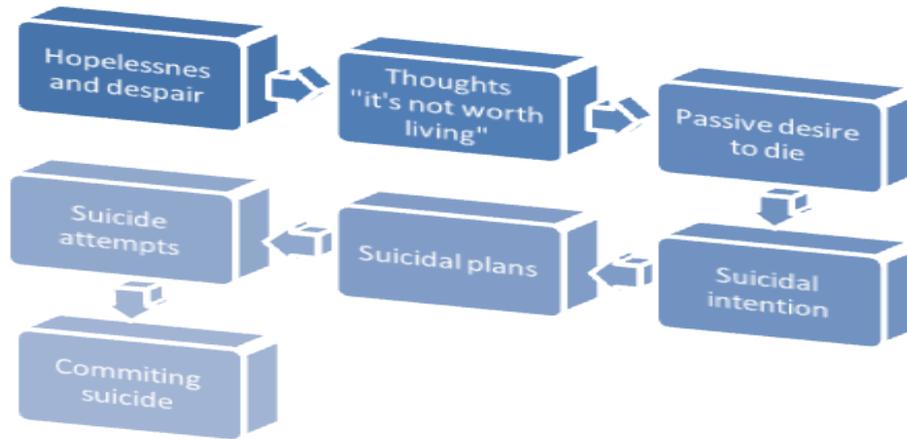


Figure 1. Model of suicidality

In the years 2017-2019, 15713 suicidal deaths were recorded in Poland. 4735 of them were suicides of people over 60 years of age (Fig.2). Men committed suicide about 6 times more often than women [G].

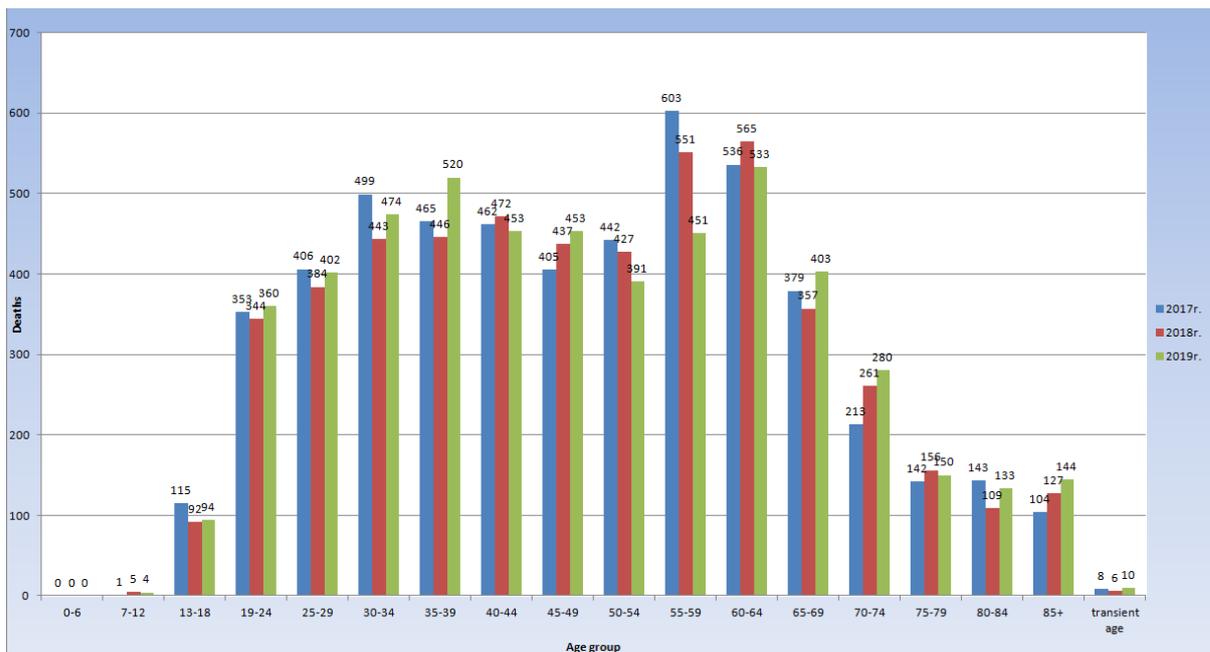


Figure 2. Total suicides, by age group: Poland, 2017-2019

Source: Authors' own work based on statistics of Polish Police [G]

We have analyzed data on the number of suicides from 1999-2019, we have necessarily divided this period into two parts: the first of them covers the years 1999-2012 (Fig.3a) and the second concerns the years 2013-2019 (Fig.3b). In 2013, the method of collecting and generating statistical data on suicide bombings changed, which may result in the difference between the reported number of suicides in these two periods of time.

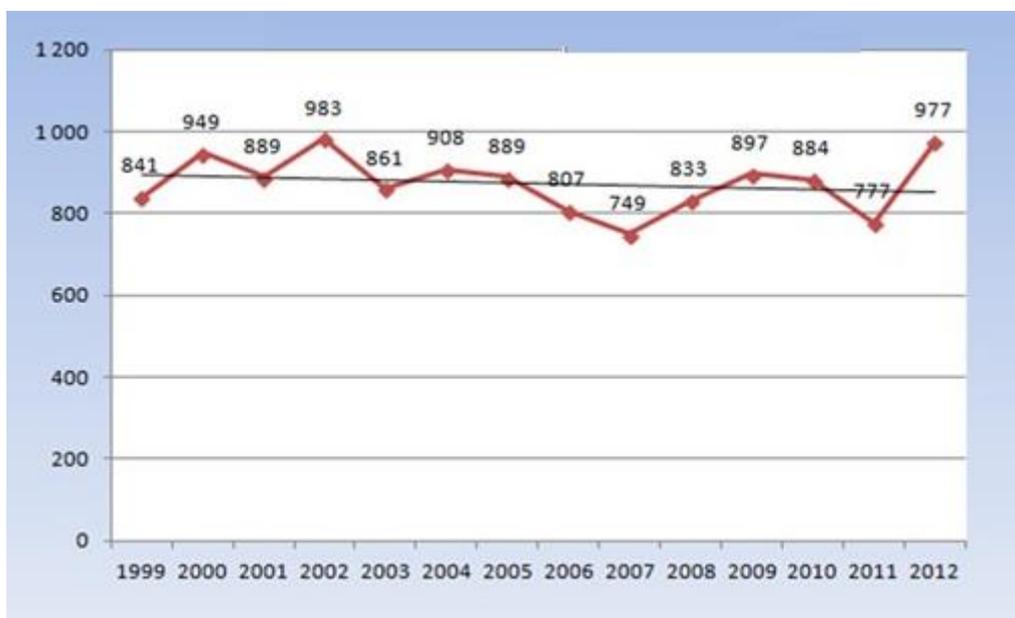


Figure 3a. Suicides in the elderly:Poland, 1999-2012
Source: Authors’ own work based on statistics of Polish Police [G]

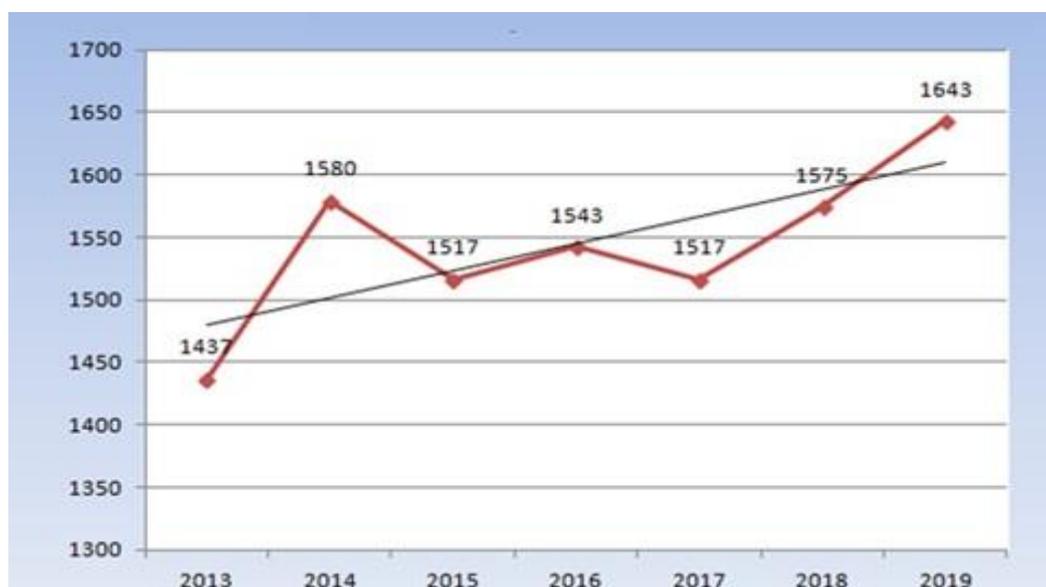


Figure 3b. Total number of suicides in the elderly: Poland, 2013-2019
Source: Authors’ own work based on statistics of Polish Police [G]

On the faith of available data we can observe an upward trend in the number of reported suicidal deaths in people over 60 years of age in the years 2013-2019 [G]. Furthermore, an increasing proportion of all suicides are elderly suicides (in 1999 they constituted 17.91% of all suicides, in 2019 they constituted 31.33%) (Fig.4). Data quality is low due to under-, mis- and non-recognition. Even less is known about suicide attempts, it was estimated that in the general population there are 10 to 20 attempts per one suicide, while among older adults it is only 1 to 4 attempts for each suicide [6]. The suggested reasons, why older people are more likely to die from their suicide attempts, are as follows: their choice of method (relatively more frequent use of firearms than in other age groups), worse body condition due to age, which makes injuries more likely to be fatal and social isolation, which makes quick medical services reaction less likely to happen [7- 10].

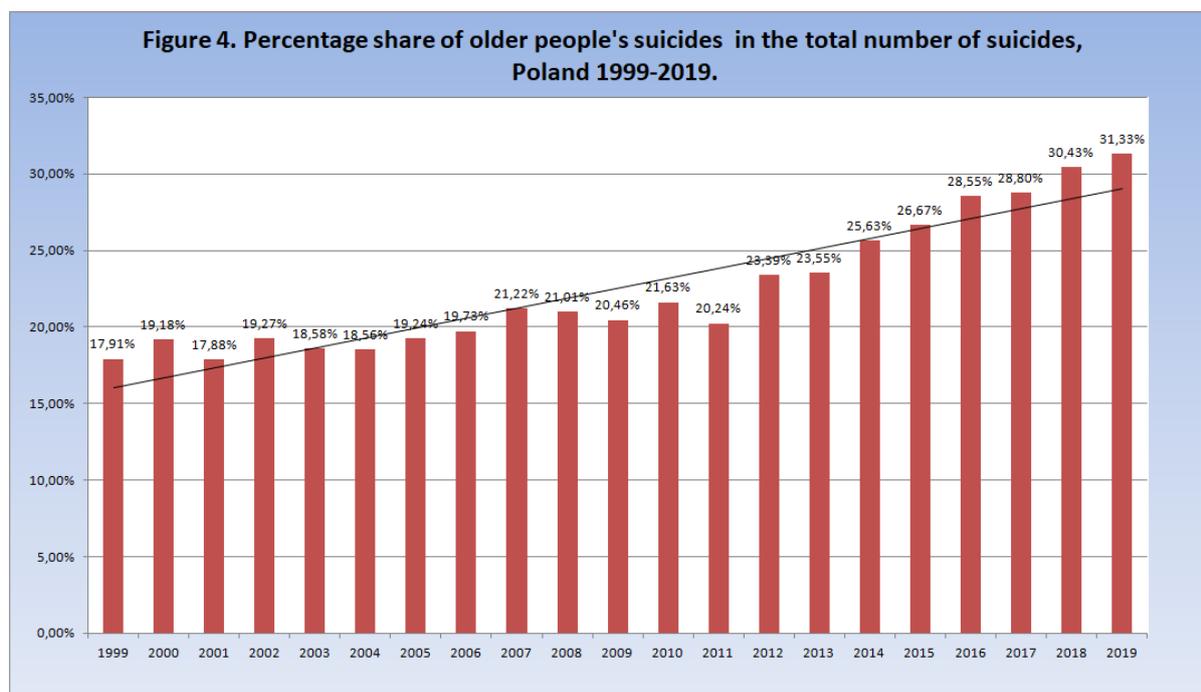


Figure 4. Percentage share of older people’s suicides in the total number of suicides, Poland 1999-2019; Source: Authors’ own work based on statistics of Polish Police [G]

RISK FACTORS, RISK GROUPS

Decades of research have enabled to define characteristics of people at high risk of suicide. However, suicide risk factor identification has not significantly diminished the number of suicide cases. The main factor that influenced suicide is difficult to determine, we think we know less than half of the reasons people took their lives.

Mental disorders are a major cause of suicide in the 21st century according to the statistics of WHO and Polish Police. Not only depression, but also schizophrenia, affective illnesses, anxiety disorders, posttraumatic stress disorder, etc. can be the cause of self-destructive activities [7,8]. In previous years, many scientific papers focused on the correlation between sleep problems and the risk of suicide. Bernert et al. [11] in their review article on this topic confirm sleep disturbances as an evidence-based risk factor for suicidal ideation, suicide attempts, and death by suicide. According to standard methodological considerations suggests that sleep disturbances, such as insomnia and nightmare symptoms may serve as modifiable, empirical risk factors for suicidal behaviors, presenting an unusual opportunity for suicide prevention.

Functional impairment, physical pain and burdensome treatment procedures are often a component that accompanies elderly people who suffer from chronic diseases or lethal disorders. Every chronic disease may be associated with an increased risk of suicide. Among others, the literature lists kidney failure (with the need for hemodialysis), multiple sclerosis, epilepsy, systemic lupus erythematosus, asthma [7]. Tseng et al. [12] provide that the mere fact of being hospitalized seems to increase the risk of suicide. Taiwanese study indicates that suicides in hospitals were eight times more common than in the general population.

Table 1. Indicators of risk of suicidal behavior in old age.

- 1. Depressive illness, mental disorder (schizophrenia, affective disorders, etc.), insomnia;**
- 2. Somatic diseases (functional impairment);**
- 3. Substance abuse (alcohol, drugs, smoking);**
- 4. Reaching for methods with a high risk of death, e.g. firearms;**
- 5. Previous suicide attempts;**
- 6. Pessimism, feeling hopeless, stiff thinking, suicide fantasies, hostility, aggression, irritability**
- 7. Alienation, loneliness, limited social support, inadequate housing, homelessness, overcrowding;**
- 8. Stress, difficult life experiences, economic difficulties;**
- 9. Suicidal behavior in the family;**
- 10. Problems and/or pathologies in the family;**
- 11. Convergence of the above factors, escalation of suicide crises**

According to WHO [A], substance-use related disorders are the second most common cause of death from unnatural causes. There is little research on the correlation between the abuse of alcohol and the risk of suicide, they are often fragmentary and referring to a specific population. Research also differs in methods, hence discrepancies in outcomes may result. Among other things, in a study conducted on the Swedish population, Waern [13] proved a strong association between alcohol use disorder and suicide in persons 65 years of age and above.

PREVENTION OF SUICIDE

There is a widespread view that suicide prevention includes the consideration of risk and protective factors and related interventions. The World Health Organization (WHO) has recommended the classification of preventative interventions. This allows to distinguish three strategies (Institute of Medicine, 1994):

- 1) **“universal interventions”** targeting whole populations. Examples include legislation (for instance restricting access to potentially lethal means) or public health messaging campaigns, e.g. on the importance of social engagement for all older people.

The simplest measure towards suicide reduction is the blocking of access to respective means: poison, potentially poisonous medication such as paracetamol, bridges, firearms, and railway [7]. The media remains to play a major role in older adults' lives. The mentioned medium should educate about depression, fight against stigmatization of mentally ill people, fight taboo, inform people at risk where they can seek help. Interventions should focus on taking advantage of the media considering its strong message. In the future the Internet and social media will be the main area of communication among older adults therefore there may be a need to create discussion forums targeted at the elderly. Community awareness programs can improve the prevention system. Helplines and public education at workplaces and schools to increase knowledge and reduce stigma may help the elderly. Every year there are organized depression campaigns to familiarize yourself with the difficult mental health subject. February 23 is an official depression day in Poland. Not only European countries but the vast majority of them have a similar campaign which seems to be auspicious.

- 2) **“selective interventions”** interventions that target higher-risk groups with more distal risk factors. These types of interventions could be provided by general practice doctors. They mainly care for the elderly, because the older adults rarely go to psychiatrists [8]. The

elderly with chronic and cancerous diseases should be in a higher- risk group. Statistics of WHO says that of those dying by suicide, approximately 45% will have seen their primary care provider within the month before their death, while only 20% will have seen a mental health professional in that period. A selective preventive intervention that provided supportive phone calls to older adults (mostly women), called the Tele Help—TeleCheck program, was found (in a quasi-experimental design) to significantly reduce the number of suicide deaths [14].

- 3) **“indicated interventions”** addressed to individuals at risk. These types of interventions emphasize the importance of psychiatric, psychological and social workers care. Greater consideration should be given in the future to the collaboration between primary care physicians with psychiatrists. To make the investigations more effective the primary care physician should participate in the treatment of patients with depression. Creating a support group for people after attempted suicide could be the right idea. Taking into consideration involving a family of a patient with depression disorder in the treatment process could be a step forward in suicide prevention [15].

The vast majority of researchers emphasize that the evaluation of suicidal patients should focus on contemporaneous factors and the needs of the patient, rather than probabilistic notions of suicide risk. What is more, it is important to have a clear understanding of the risks of suicide according to methods of prediction and what commensurate preventative measures might be.

MEDICAL INTERVIEW

A thorough medical interview is a key issue in detecting suicidal thoughts. A general practitioner should be able to devote sufficient time to the consultation, to not only take care of the main purpose of the visit but also talk to a patient, observe and try to assess the risk. Studies showed that doctors, whose average consultation length was less than seven minutes, were less likely to recognize and deal with long-term problems and psychosocial problems than slower doctors, with an average consultation length of nine minutes or more [16]. Unfortunately, in public healthcare physicians usually are forced to limit the length of the visit. However, it is very important to give a patient as much time as he needs to open up and talk about his problem. As the patients in old age are reluctant to signalize their sad mood and depressive thoughts to the health workers, it is important to build trust and good doctor-patient relationships. Questions should be asked subtly, the structure of the question should not suggest an answer and should give the possibility of free expression. In a study on how health professionals communicate to

access suicide risk 75% of questions were negatively phrased, e.g. “No thoughts of harming yourself?”. This way of formulating questions biased patients’ responses towards reporting no suicidal ideation [17]. For most physicians asking about suicidal thoughts is hard and uncomfortable. WHO’s resource for general practitioners suggests that it could be helpful to start a conversation with a sequence of these questions:

1. Do you feel unhappy and helpless?
2. Do you feel desperate?
3. Do you feel unable to face each day?
4. Do you feel life is a burden?
5. Do you feel life is not worth living?
6. Do you feel like committing suicide [H]?

Asking straight whether someone wants to commit suicide may feel too direct. Instead of that, it may be: *„Did you ever think that life doesn’t make sense anymore, that you’d like to end it?“*. When the patient confirms the presence of suicidal ideas, the physician should continue with further questions about the frequency and severity of the idea. It is important to know whether the patient has made any plans, does he own a gun or has access to pills.

If the patient doesn’t verbalize suicidal thoughts, it doesn’t mean there is no risk of suicide. The physician should pay attention to the patient's behavior. Such indicators as social isolation, providing brief answers to questions about mood, avoiding answers about personal or family situations, denial of food or indifference may be warning signs [18].

SCREENING TOOLS

Screening tools could theoretically help physicians identify any patients at risk of suicide and depression. In practical terms, all the invented scales have limitations. Nowadays, many health professionals regard them as erroneous instruments which you cannot fully trust. Some psychiatrists reckon that the scales are inaccurate, and therefore useless to support specific suicide therapies [18]. Even the authors of the scales write that no instrument is suitable for risk assessment. Moreover, the use of scales can lead to wrong conclusions which may be dangerous. The most important intervention is a holistic assessment of psychosocial factors related to the patient [19].

It should be noted that one of the oldest and essential tools for suicide risk assessment is SSI Beck's suicide scale. The main hypothesis tested by the authors of the scale was the assumption that the feeling of hopelessness is more associated with the severity of suicidal

thoughts than the assessment of depression. The example of the scale for studying a particular population is, the InterSePT scale [20], used to assess the risk of suicide in schizophrenia. A systematic review of scales carried out by Kodaka et al. [21] to assess attitudes towards suicide suggests that all rated scales can be used for research purposes, but three scales deserve special attention. These scales are: Suicide Opinion Questionnaire (SSQ) [22], Suicide Attitude Questionnaire (SUIATT) and Attitudes Toward Suicide (ATTS) [23].

When estimating suicide risk WHO recommends taking into account factors such as emotional state (distress) assessment, early detection of mental disorders, alcohol abuse, access to the tools most commonly used to commit suicide [H].

National Guideline Clearinghouse, an American institution providing guidance based on evidence-based clinical practices for assessing the risk of suicide recommends clinical history, Beck scales: hopelessness, suicidal thoughts, suicidal tendencies, Beck depression scale, Hamilton depression scale [I].

Polish authors undertook to construct a scale to assess the risk of suicide in people hospitalized in a psychiatric ward. T. Koweszko [24] emphasizes that such tools are more used to assess attitudes towards suicide than to predict it.

A useful tool for risk assessment suicide in patients discharged from the psychiatric ward turned out to be the scale for the assessment and intensity of suicidal crisis (Suicidal Crisis Syndrome - SCI) - especially shortly after discharge from the hospital [25]. So the scale is different from other scales that assess risk in the long run. The authors considered SCI to be a tool to predict the risk of suicide, and the most important predictor proved to be entrapment subscale.

Noteworthy is also The Columbia-Suicide Severity Rating Scale - C-SSRS). It consists of two parts defined as 'suicidal thoughts' and 'suicidal behaviors'.

AN APPROACH TO CARE MANAGEMENT FOR HIGH-RISK SUICIDE PATIENTS

Summarizing WHO recommendations for general practitioners (Geneva, 2003) it is important to remember about a few crucial steps while dealing with patients prone to suicide. Initially the physician ought to devote as much time as the patient needs despite a long queue in front of the doctor's office. First and foremost attention must be paid to showing comprehension and building a good relationship. At the same time the general practitioner should evaluate available support systems and ask a particular person close to the patient for help. Furthermore, the physician should make an assessment which enables him to decide

whether or not to refer the patient to a psychiatrist or immediate hospitalization. WHO emphasizes that it is essential to be able to behave properly when hospitalizing a patient [J].

CONCLUSIONS

Suicides of elderly people are already a major problem and we observe a rising trend in the number of suicides in this group. The general practitioners are often the only health professionals old people visit, that is why they should pay particular attention to the possible suicide risk factors and should know how to react properly. The elderly are significantly burdened with chronic underlying diseases that affect their psyche and increase the risk of suicide. Hence the physicians should conduct a more conscientious interview without losing sight of the elderly's mental health. Patients prone to suicidal thoughts should be referred to a psychiatrist. All the risk assessment tools should be seen as having a subsidiary character. The intuition of an experienced physician is a natural tool, worth more appreciation in medical practice, especially in a delicate area, which is a psychological condition.

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The Future of Psychedelics — old substances with potential novel uses

Sebastian Fedorowicz¹, Karolina Radzikowska¹, Michał Turniak¹, Michalina Grzelka¹, Jan Gnus^{1,2}, Paweł Barnaś¹, Anna Kolcz¹

1. Wrocław Medical University
2. Regional Specialist Hospital in Wrocław, Research and Development Center, Wrocław

INTRODUCTION TO PSYCHEDELICS

Hallucinogens are substances whose one of the primary effects is a shift of sensory perception, auditory and visual changes, altered mental state, change of mood and thought schemes. They have been used for millennia for enhancing religious rituals, trying to achieve different states of consciousness or for entertainment [1]. The first chemically synthesized hallucinogen was lysergic acid diethylamide (LSD), developed in 1938 by Albert Hofmann who accidentally found out about its psychedelic properties himself in 1943 due to being exposed to the drug during his laboratory work time [1]. At first, it was meant to be introduced as an anesthetic, then an idea shifted towards an adjuvant drug accompanying the process of psychotherapy and then found its way to masses as a recreational drug in the 1960s. Soon after reaching popularity it was banned by the government of the United States through federal law in 1966 and since then and due to the subsequent “War on drugs” couple years later the access to hallucinogenic substances, even in scientific field and clinical trials, had been severely restricted. Nonetheless, there is a rising trend of investigating therapeutic uses of LSD, especially in the treatment of severe depression, anxiety in palliative care patients, management of tobacco and alcohol dependence and therapy of treatment-resistant depression [2]. Moreover, some experiments were designed to test psychedelic substances as a potential truth serum, adding it to war arsenal or a tool enhancing interrogation for military purposes [3]. American government through Central Intelligence Agency continued its top secret trials during two decades and were in search for “*substances which will promote illogical thinking and impulsiveness, substances which will cause temporary or permanent brain damage and loss of memory, substances which will*

promote weakness or distortion of eyesight or hearing faculties, preferably without permanent effects or substances which alter personality structure in such a way the tendency of the recipient to become dependent upon another person is enhanced” and therefore psychedelics, especially LSD, were thoroughly tested during that underground operation [4].

MECHANISM OF ACTION

Traditionally psychoactive substances divide either into a category of stimulants or depressants of the nervous system. Their action accordingly invigorates our mental state, boosting energy levels or slowing down thinking processes, emotions resulting in a relaxed, often euphoric emotional state. Psychedelics work beside that mechanism. Their mechanism of action is explained molecularly mainly by effecting as agonists of 5-HT_{2A} serotonin receptor, and they induce mental states unsimilar to that experienced on a daily basis. These unrealistic states of consciousness are described as similar to dreaming, mystic trance, religious encounter or ecstasy. Moreover, psychedelics are relatively safe due to not provoking dependence [5].

CLASSIFICATION OF PSYCHEDELICS

Due to the fact that numerous psychedelic substances act on multiple receptors it is unfeasible and impractical to classify them exactly to one group.

The criteria based on mechanism of action is only contractual and serves for listing psychedelics for further description.

As a consequence we will depict the origin, use, dosage and potential future use of lysergic acid diethylamide, psilocybin, N, N-dimethyltryptamine, mescaline, MDMA, PCP, ketamine, dextromethorphan and active substances of *Amanita muscaria* and *Salvia divinorum*.

A. (5-HT_{2A}) SEROTONERGIC RECEPTOR AGONISTS

Pharmacological mechanisms underlying effects of psychedelics are not completely understood, although they are linked to the ability of drugs to bind to 5HT_{2A} receptors in pyramidal cells of neocortex which is the cause of their cognitive effects. Moreover, some of them provoke

sympathomimetic effects such as tachycardia, hypertension, increased preload, glycogenolysis, thermogenesis and mydriasis. The substances that belong to that group are: LSD, psilocybin, DMT and mescaline [6].

Lysergic acid diethylamide (LSD)

As aforementioned in the introduction, lysergic acid diethylamide (LSD) was the first chemically synthesized hallucinogen, which took place in 1938 by Albert Hoffmann. Since then it is perceived as the prototype of all psychedelics and provides a number of scientific studies in this topic. It shifts sensory perception, provokes abnormal thought scheme and mood alteration, although the exact pharmacologic mechanisms are unknown [1].

The most popular forms of LSD are tablets, capsules and liquid dissolved in sugar cubes or pieces of colorful blott papers soaked in the solution due to the fact that the color of pure substance is clear and it has no odor. Usually the route of administration is oral. The single dose consists of 40 up to 500 micrograms of substance however even 20 micrograms may induce the action [A].

In terms of dependence it is known that the lysergic acid diethylamide is believed to not cause physical addiction, nonetheless there are proofs suggesting that repeated doses may lead to developing of tolerance and thus increased dose must be consumed to achieve the same effect [5]. The prevalence of LSD had risen according to results from 49 757 psychoactive substance reports from all poison centers in the United States that shows that the LSD exposure had increased the most from all psychoactive significantly over 10 years since 1st of January 2007 to 31st of December 2017. Nevertheless, this prevalence is believed to be low due to multiplicity of other hallucinogen substances and other psychoactive drugs [7].

LSD had been tested in terms of being an adjuvant to psychotherapy for anxiety connected with terminal, life-threatening diseases in a double-blind, randomized pilot study and the results showed that it may be useful in reducing anxiety under condition that the substance is administered with safety precautions in a scientific, medically supervised psychotherapeutic setting. Although this clinical trial was just a pilot study performed on a group of only 12 participants, it demonstrated the possibilities: the state of anxiety had been significantly reduced without chronic adverse effects linked with treatment [8].

Moreover, there is evidence of a positive outcome of using LSD in the treatment of alcoholism and binge drinking behavior, based on a meta-analysis of randomized control trials which take into account 536 people with alcohol addiction. The results show an improvement understood as abstinence or reduction in binge drinking episodes. The improvement was statistically significant even after 6 months after the therapy. In addition, the number needed to treat using LSD was 6, for instance naltrexone's number needed to treat is 9.

Furthermore, the potential future use of LSD in the treatment of post-traumatic stress disorder (PTSD) is currently being investigated but there is a rationale that its distinctive qualities may provide to be helpful, for example facilitating psychotherapy process. Likewise, other psychedelics, this issue needs to be investigated thoroughly [9].

Psilocybin

Psilocybin is a prodrug of psilocin which is an agonist of serotonin 2A receptor. It can be found naturally in mushrooms and thus has been used by humans for millennia [10]. New findings describe use of psilocybin together with psychotherapy in a variety of psychiatric diseases such as terminal illness anxiety, obsessive compulsive disorder, tobacco and alcohol dependence [10]. Moreover, recent studies consider its use also in treatment-resistant depression which suggests, based on the results of clinical trials with volunteers, that it can induce long-lasting improvement of mental health, even just after a single dose [11].

How does it affect the brain? The exact mechanism are unknown but whole-brain analyses by quality pre and post treatment fMRI indicates that decreased cerebral blood flow in the temporal cortex, including amygdala, correlated with reduced depressive symptoms which were observed in 100% of patients one week after treatment and in 47% of patients after 5 weeks. Post-treatment changes in resting-state functional connectivity are related to clinical outcomes. Authors of this clinical trial propose a mechanism of “therapeutic reset” of brain activity [12].

In 2019 United States Food and Drug Administration granted a status of the “breakthrough therapy” to use of psilocybin in the treatment of major depressive disorder — a condition resistant to traditional therapy regimen in patients whose state had not improved after undergoing two subsequent, different antidepressant treatments [B]. The status of breakthrough therapy surely quicken the unrushed and deliberate procedure of drug development. Notwithstanding it does not necessarily imply that a drug is a “breakthrough”, neither that there are high-quality evidence of

treatment efficacy, it rather allows the Food and Drug Administration (FDA) to grant a priority review if preliminary clinical trials indicate that the new treatment regimen may offer significant advantage over existing options for patients with serious or life-threatening disease, enabling a fast-track designation, approval and review [13]. On the other hand, results from the clinical trial conducted by nonprofit Usona Institute are exceptionally promising — it is focused on the efficacy of treating patients with major depressive disorder with a single dose of psilocybin [C].

N, N-Dimethyltryptamine

N, N-Dimethyltryptamine or DMT can be found in the body of many mammals, including humans, and moreover in different types of plants, for instance in the ayahuasca vine, *Psychotria viridis* or *Mimosa hostilis* which had been serving as components of traditional hallucinogenic ceremony in South America. This custom of Amazonian indigenes now plays a role of touristic draw for travelers seeking psychedelic experience from all over the world due to properties of DMT such as: a sense of enlightenment, experiencing a mystical or religious event, similar to dream or even described by participants as something like death, close to phenomenon called Near Death Experience (NDE) [14].

The drug was discovered by the Hungarian scientist Stephen Szára who also investigated the effects, metabolism and potential use in therapy of DMT, as well as its derivatives. His research subjects were volunteers and people with chronic mental illnesses such as schizophrenia [15].

Typical route of drug administration is oral, through drinking a beverage that is a mixture which consists of leaves and plant stems although there is evidence of insufflation, inhalation and injection. When it comes to drug effects it turns out that it depends on the dosage. Thus lower doses (up to 0,05 mg/kg) can induce emotional responses but without psychedelic effect and hallucinations and doses higher than 0,4 mg/kg induce vivid, intensively colorful visual hallucinations with abstract and rapidly moving forms or images with additional experience of euphoria, calm, fear or anxiety [16,17,18].

Some investigators had observed a similarity of psychedelic experience after DMT with near death experience that is with complex experiential episodes that occur in association with death or the perception that it is impending, for instance including patients with heart attack. This mental state is typically characterized by a sensation of calm, traveling through a dark region, tunnel or 'void', encompassed by an enlightenment of a bright light, an imagination of crossing

the borders of realms or reality and even communicating with “other beings”. These experiences are parallel to sensations after intoxication with dimethyltryptamine. A striking similarity worth underlying is the effect of dissolved ego or so called ego-death, which resembles mystical out-of-body experience [19].

The question is whether there is a grain of truth in shamanic wisdom of ayahuasca ceremony and its alleged therapeutic use? Could it potentially serve as an alternative treatment of psychiatric diseases? It is all about the problem of clinical trials with substances of rigorous legal status, which applies to all psychedelics — further investigation needs to be done but it's being impeded by severe law limitations. Nevertheless, some scientific paper suggests that ayahuasca may be potentially useful in the treatment of various psychiatric disorders and addiction [20].

When it comes to adverse effects they appear to be relatively mild but further investigation needs to be done. Moreover, a side effect such as psychosis is found to be rare during traditional amazonian ayahuasca ritual sessions as well as trials in experimental settings. However, there is evidence of few case reports with such symptoms and therefore the psychiatric screening before drug administration is advised due to the fact that a risk of side effects rises when a patient presents a positive family history of psychosis, for instance psychotic mania, psychotic depression or schizophrenia [21].

In terms of future use, there is evidence suggesting that ayahuasca may present neuroprotective potential in terms of Parkinson's disease [22].

Mescaline

Mescaline is an active substance of *Lophophora williamsii* — a cactus that can be found near the border of Mexico and the United States. Its buttons may be dried or dried and powdered and then brewed as an infusion, with a standard dose of 8-10 cactus buttons. The mescaline evokes distorted perception, especially including visual perception and sensation of dazzle, although the drug is found to be milder than LSD but very similar in action due to the fact that they are both 5-HT_{2A} agonists. Side effects include possible anxiety, fear, diarrhea, headache, nausea, vomiting, psychosis, tachycardia, amnesia or even hallucinogen persisting perception disorder which is a state of returning persisting flashbacks of images or “visual snow” after psychedelic experience, quality of life [23].

B. SEROTONIN RELEASERS (EMPATHOGEN-ENTACTOGENS)

Empatogens or entactogens are names of groups of psychoactive drugs that cause emotional and prosocial effects. Experiences after taking empathogens are characterized by a feeling of emotional openness, euphoria, empathy, love, connectedness, communion with others and mild audio and visual distortions.

Major substances of this class include 3,4-methylenedioxymethamphetamine (MDMA), 3,4-methylenedioxy-N-ethylamphetamine (MDEA), 3,4-methylenedioxyamphetamine (MDA) [24].

MDMA

MDMA is a synthetic substance commonly known as ecstasy or molly. It was first synthesized by the German pharmaceutical company Merck in 1912 as a parent compound for medications that control bleeding [D].

In the 1970s Alexander Shulgin re-synthesized MDMA and tried it himself. After his experiences he called the drug “window” in order to its effects that allows users to strip away habits and perceive the world clearly [25].

In 1977, A. Shulgin gave MDMA to Leo Zeff PhD, who used it as an adjunct in psychotherapy [25].

Despite the fact that MDMA was not pharmacologically tested for the use in humans, a group of psychiatrists included it in their therapy in order to enhance communication during sessions and allow patients to achieve insight about their problems [D].

In the 1980s ecstasy also became popular in the nightclub and at all-night dance parties called raves, as well as it started to be more available on the streets. In 1985, due to extensive non-medical use of the substance DEA declared an emergency ban on MDMA [D].

Recently, MDMA has been re-examined for possible medical use. It may become beneficial for use in psychotherapy of post-traumatic stress disorder (PTSD). Reducing fear and increasing interpersonal trust caused by MDMA helps patients to open up in front of the therapist [9].

Ongoing studies, which are in the third clinical phase, show that MDMA in combination with psychotherapy is effective and well tolerated in reducing PTSD symptoms [26, E].

C. DISSOCIATIVES

Dissociative drugs are psychoactive substances that cause visual and auditory distortion and produce feelings of dissociation from the environment and self. They work by disrupting the action of the brain chemical glutamate at N-methyl-D-aspartate receptors [F].

Effects depend on the amount of drug taken and mostly are unpredictable. Low and moderate doses usually cause numbness, disorientation, confusion, loss of coordination, nausea, vomiting, changes in sensory perception, hallucinations, feelings of dissociation, increase in blood pressure, heart rate, respiration and body temperature [F].

While high doses cause hallucinations, memory loss, physical distress like dangerous changes in blood pressure, heart rate, respiration and body temperature, additionally marked by psychological distress, for instance feeling of extreme panic, fear, anxiety, paranoia, invulnerability, exaggerated strength and aggression [F].

Combination of high doses of dissociative drugs and alcohol or other central nervous system depressant can lead to respiratory distress or arrest, resulting in death. The group includes phencyclidine (PCP), ketamine, dextromethorphan (DXM) [F].

PCP

PCP, which is also known on the streets as “angel dust”, is a hallucinogenic drug. It was first developed in the 1950s by the name of Sernyl for general anesthetic, but later in 1965 became abandoned for this use in humans due to its side effects, such as postoperative dysphoria and hallucinations.

Until 1978 PCP was used in veterinary.

PCP re-emerged as a drug of abuse during the 1960s and 70s. It is available as tablets, capsules, colored powders and can be smoked, taken orally or nasally [G].

Recreational use of PCP in doses 1 to 10 mg causes rapid onset of euphoria and feelings of omnipotence, superhuman strength and social and sexual prowess. While higher doses can lead to progressive confusion, disorientation, coma, seizures, malignant hyperthermia, shock, rhabdomyolysis, renal failure and even sudden death. Effects of chronically drug use are associated with severe violent and aggressive behavior and episodes of acute psychosis [G].

Ketamine

In the 1960s during laboratory investigation of PCP derivatives, ketamine was developed. The US military used it as an anesthetic during the Vietnam War, and then ketamine gained popularity as a substance for starting and maintaining anesthesia, which is used up to nowadays [27]. Ketamine causes pain relief, sedation, memory loss, and at the same time remains heart function, breathing and air reflexes. Intravenous injection starts work after 1-5 minutes and lasts for approximately 10-20 minutes [28].

Ketamine also became a drug of abuse for its hallucinogenic and dissociative effects. Many young people used it and other drugs as part of the “make love, not war” protest. Recreational ketamine is known as Special K, Kit Kat, CatValium, Vitamin K, Monkey Mix [27]. In moderate doses can lead to amnesia, sedation and immobility. Effect called K-hole is possible to achieve with high doses of the drug. It is described by the ketamine users as a terrifying feeling of almost complete sensory detachment compared to near-death experience [F].

Ketamine is considered as a drug for pain relief. It is proven that ketamine reduces pain, opioid requirements and decreases postoperative nausea and vomiting in a perioperative setting. Refractory cancer pain or pain in palliative care also may be treated by low-doses of ketamine combined with opioid. There is not enough data about safety in long-term or repeated treatment by ketamine of non-cancer, chronic pain [29]. In addition, ketamine was tested in chronic refractory migraine. The study showed short-term improvement in pain severity in 6 out of 6 patients. This may show new ways of treatment, but still requires additional research [30].

In the last decades, ketamine was found to be effective in treatment of depression. As it was hypothesized that glutamate plays a role in mood modulation, the noncompetitive antagonist of N-methyl-D-aspartate (NMDA) receptors was considered to help in depression therapy. Further studies demonstrated that ketamine has antidepressant effects in treatment-resistant depression [31]. What is more, due to its rapid action it may be useful in treatment of suicidal behaviors [32]. PTSD psychotherapy assisted by ketamine also may be effective. Due to its ability to enhance fear extinction and block memory reconsolidation ketamine may increase receptivity to psychotherapy [9].

There are also studies that show use of ketamine in refractory status epilepticus (RSE). The disease is caused by prolonged seizures which led to decrease of the numbers and activities of GABA receptors, thus, the first-line and second-line antiepileptic drugs fail. At the same time, the

numbers and activities of glutamatergic NMDA receptors increase. Ketamine, a noncompetitive antagonist of glutamatergic NMDA receptors, appears to be effective and relatively safe for the control of RSE [33].

Dextromethorphan

Dextromethorphan (DXM) is a commonly known medication for cough. It was found as a nonaddictive substitute of codeine. In 1958, DXM was approved and introduced as an over-the-counter drug. The substance is D-isomer of the codeine analog, levorphanol, and contains a chemical structure that is seen in other dissociatives agents. DXM works on medulla oblongata and inhibits the cough reflex. It is absorbed quickly from the digestive tract, starts working after 10-30 minutes and lasts 6h [H].

Syrups typically contains 15 milligrams per teaspoon, tablets 15 milligrams per tablet. When the drug is taken in higher doses may cause euphoria, hallucinations or even depressant effects, that is why it is often misused [I].

Dextromethorphan is the only one approved drug for emotional disturbance characterized by uncontrollable episodes of crying or laughing, or other emotional displays, called pseudobulbar affect. The drug has diverse pharmacological actions in the central nervous system so it may become effective for many neurological and psychiatric diseases [34].

D. OTHERS

Amanita muscaria

A basidiomycota fungi called *Amanita muscaria* contains psychoactive substances and therefore is known to be toxic in spite of the fact that death intoxications are rare nowadays due effective treatment and usually good prognosis [35].

The biologically active agents are muscimol, ibotenic acid, muscarine and muscazone. They produce a state of sedation, function as the depressants with additional feeling of detachment and most of all, can cause a psychedelic effect. However, in some individuals sometimes paradoxical reactions may be revealed through state of simultaneous agitation [36].

The poisoning treatment normally includes administration of active charcoal or gastric lavage, depending on the time after ingestion.

Salvia divinorum

Salvia divinorum is a plant species that has psychoactive properties, grows 1 meter high and has ovate to acuminate leaves 12 to 15 centimeters long. It comes from Sierra Mazateca in Oaxaca, Mexico. Aztec people have used it for many healing and divinatory rituals. The plant leaves may be chewed or dried and smoked [37].

The effects after taking include elevated mood, revisiting childhood memories, contact with entities, cartoon-like imaginary, synesthesia. *Salvia divinorum* is considered as a hallucinogen [38]. The plant has become popular among young people because of its easy availability on the internet.

The active substance isolated from *salvia divinorum* is Salvinorin A, which is a selective high efficiency kappa-opioid receptor agonist. Acute activation of these receptors cause anti-addictive effects in the brain by regulation of dopamine levels. According to that Salvinorin A and its analogues may be promising substances in treatment of drug abuse [39].

CONCLUSIONS

Many psychedelics are considered as substances of abuse by the government because of their misuse by people. However, the researchers have shown that some of them have potential to be used in therapy of specific diseases.

Psychotherapy of PTSD associated with MDMA appears to be effective and currently is in the 3rd phase of clinical trial. LSD and ketamine are also considered in this field. While in treatment-resistant depression may help psilocybin after just one dose or ketamine which works rapidly and may be also useful in suicidal behavior.

DMT from ayahuasca vine may be neuroprotective in Parkinson's disease. Ketamine is also considered in pain management especially perioperative pain, refractory cancer pain and pain in palliative care. Use of ketamine in refractory chronic migraine is promising but needs more research. What is more, ketamine shows to be an effective and possible new way of treatment in RSE while working on different receptors.

DXM appears to be useful in pseudobulbar affect. In addition, psychedelics may be used in treatment of various addictions, for instance *Salvia divinorum* and its analogues are promising in therapy of drug abuse, while LSD may be effective in tobacco and alcohol dependence.

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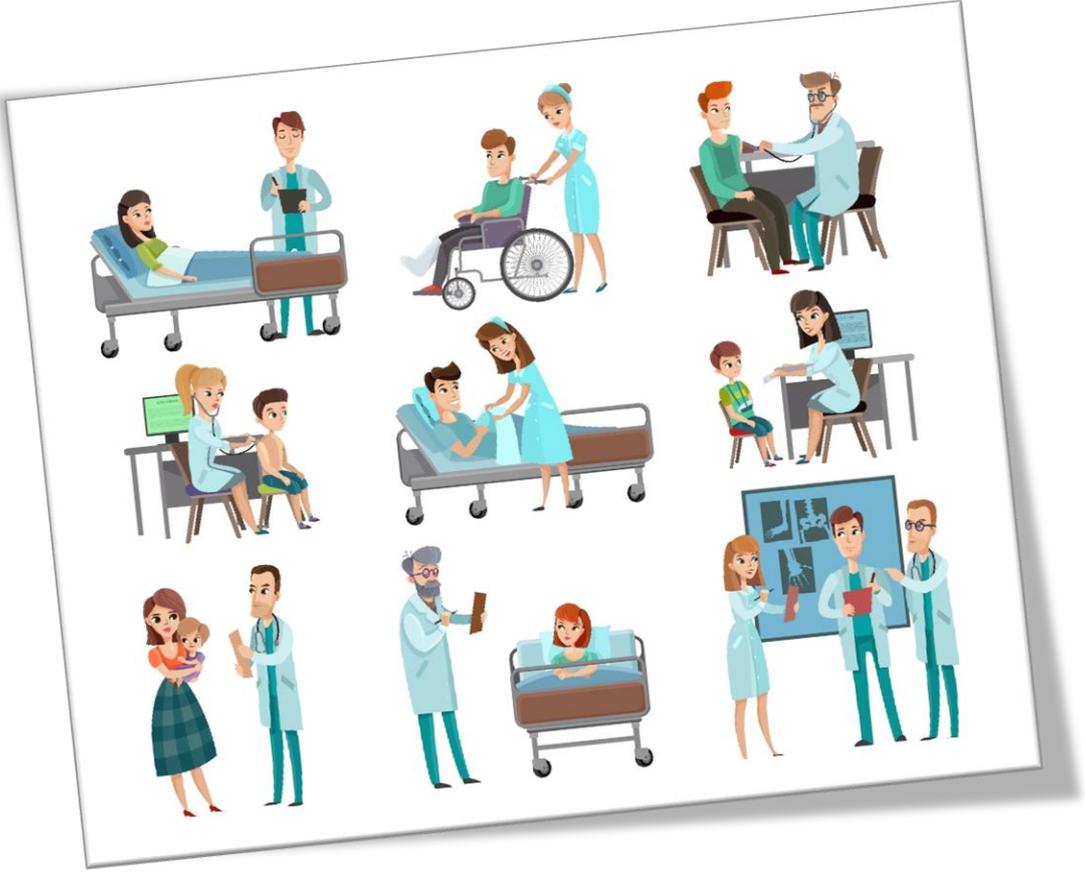
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PATIENT AS A PARTNER



Patients' perception of trust in doctors and nurses

Elżbieta Krajewska-Kulak¹, Cecylia Łukaszuk¹, Mateusz Cybulski¹, Andrzej Guzowski¹, Paulina Aniśko², Wojciech Kulak³

1. Department of Integrated Medical Care, Medical University of Białystok
2. Doctoral School, Medical University of Białystok
3. Department of Pediatric Rehabilitation, Medical University of Białystok

INTRODUCTION

Patient participation in the treatment process is a relatively new phenomenon that has recently been of interest to researchers, but it is the main determinant of proper medical care for the patient [1] and a motivating factor for changes in the entire treatment process [2].

Castroa and colleagues [3] distinguished patient participation at the micro level (individual healthcare), mezzo level (services development, healthcare evaluation) and macro level (policy), suggesting that each level is associated with other types of activity.

The authors [3], based on a thorough review of the literature, proposed their own definition, according to which patient participation is based on the patient's rights and ability to influence decisions regarding medical care through communication and dialogue adapted to the patient's preferences and potential and resulting from a combination of doctor and patient contact.

The literature on the subject emphasizes the importance of the relationship between patient participation in the treatment process and trust in the doctor. The patient's trust in doctors results from the expectation that doctors act in the patient's interest [4] and use the latest knowledge and achievements in the field of medicine. These expectations become more important in healthcare systems that provide patients and physicians with greater freedom of action [5,6].

Savage [6] notes that analysing the level of patient trust in physicians gives insight into patient attitudes regarding the extent of participation in the treatment process. The increase in trust means patients prefer to leave medical decisions in the hands of a doctor and rely solely on their medical knowledge, i.e. passively participate in the healthcare process. Then the

likelihood of conflict between doctors and patients is reduced, and accordingly, the scale of complaints and lawsuits [4]. However, in a situation where trust in the doctor is eroded, patients try to minimize their dependence on the doctor and take more control over their health. Therefore, it seems the level of trust in doctors may be a factor influencing the extent of patient participation [5].

Previous studies clearly show the relationship between trust in the doctor and the patient's willingness to participate in the treatment process [7,8].

The quality of nursing care by a patient depends on many factors [9] such as: organisation of healthcare, expectations of care and previous experience of patients external environment - cleanliness, meals, comfort; communication, good cooperation between doctors and nurses; interpersonal relations between a nurse and a patient; patient participation and commitment to care decisions; rested staff; and sociodemographic factors: age (older patients are more satisfied with care than younger), gender (men present a higher level of satisfaction than women), education (patients with higher education have lower levels of satisfaction)

Trust is a defining element in any interpersonal relationship, but is particularly central to the patient-physician relationship [10,11].

Although evidence shows that the majority of patients continue to trust physicians to act in their best interest, concern is growing that the rapid and far-reaching changes in the healthcare system have placed great pressure on that trust and may be undermining it.

One of the measures used to assess interpersonal trust in a patient-physician relationship is a scale developed by Anderson and Dedrick entitled the Trust in Physician Scale (The authors defined trust as a process which occurs when relations between a physician and a patient take place [5,7,12,13].

Most research on participation either diagnoses the essence of this issue or focuses on its consequences, i.e. the quality of medical care, patient satisfaction or loyalty. However, few studies are devoted to trust in a doctor, and even fewer to trust in nurses [14].

The purpose of this study was to assess patients' trust in their doctors and their nurses.

METHODS

We compared patients' trust in their doctors to their trust in nurses. We analysed data from a cross-sectional survey.

We summarise the survey methodology below.

Design

We completed analysis of the data from a cross-sectional survey conducted from March 2018 to February 2019. In that survey, patients completed two questionnaires: the Trust in Physician Scale and the Trust in Nurse Scale (Table 1). Approval of the study was obtained from the Bioethics Committee of the Medical University of Bialystok (R-I-002/426/2017).

Study setting and sample

The study was carried out in collaboration with family doctors' offices in Bialystok, Poland. Fourteen family doctors and 1.200 adult patients participated in the study.

Variables

We used two questionnaires: the Trust in Physician Scale and the Trust in Nurse Scale. The *Polish* language versions of these scales were each validated.

Table 1. Changes in item wording on the scale

	Trust Scale	
	in Doctors	in Nurse
1	I doubt that my doctor really cares about me as a person.	I doubt that my nurse really cares about me as a person.
2	My doctor is usually considerate of my needs and puts them first.	My nurse is usually considerate of my needs and puts them first.
3	I trust my doctor so much I always try to follow his/her advice.	I trust my nurse so much I always try to follow his/her advice.
4	If my doctor tells me something is so, then it must be true.	If my nurse tells me something is so, then it must be true.
5	I sometimes distrust my doctor.	I sometimes distrust my nurse.
6	I trust my doctor's judgments and opinions.	I trust my nurse's judgments and opinions.
7	I feel my doctor does not do everything he/she should for my medical care.	I feel my nurse does not do everything he/she should for my medical care.
8	I trust my doctor as to the method of treating my medical problems.	I trust my nurse as to the method of nursing.
9	My doctor is a real expert in treating medical problems.	My nurse is a real expert in nursing.
10	I can tell my doctor if he/she made a mistake.	I can tell my nurse if he/she made a mistake.
11	I sometimes worry that my doctor may not keep the information we discuss totally private.	I sometimes worry that my nurse may not keep the information we discuss totally private.
	The reliability of the Cronbach coefficient of this scale	
	0.981	0.980

The Trust in Physician Scale validation was completed with the agreement of Robert F. Dedrick and published in *Prog Health Sci* 2018, 8, 1, 27-35. Cronbach's alpha coefficient was 0.85. The Trust in Nurse Scale was similarly validated.

Procedure

The study used the Anderson and Dedrick Trust in Physician Scale (TIPS) covering 11 issues (Anderson), Polish validation by Krajewska-Kułak'2018 [15], as well as the Trust in Nurse Scale, based on the standardized Trust in Physician Scale, validation by Krajewska-Kułak'2019 [16]. The data were derived from the two questionnaires. Patients completed the questionnaires before visits to their family doctors. Researchers provided patients with the questionnaires and explained how to complete them. 1,350 questionnaires were distributed to patients. Of those, 1,264 (93.6%) were returned and 1,200 (88%) were fully completed and analysed. Data extraction procedures were applied to the questionnaires, and the data were anonymised.

Analysis

The thematic scope of the scales is illustrated in Table 1. Patient responded using a five-point scale: 1- I definitely disagree, 2- I disagree, 3- neither yes nor no, 4- I agree and 5- I definitely agree. In the result analysis, we calculated the percentage and average values of points obtained from the responses to individual scale questions. All statistical analysis was performed with Statistica 13.0. Wilcoxon test signed rank test was applied to compare difference between groups. The critical level for all tests of significance was $p < 0.05$.

RESULTS

Generally, no differences were found in the level of patient–doctor trust and patient–nurse trust. In addition, differences were found in favor of doctors regarding the belief that the doctor or nurse really cares about the patient; that whenever they make a mistake, they can be told; that if they say something, it must always be true; that patients always adhere to a doctor's or a nurse's advice; and that, sometimes, patients worry that their doctor or nurse will not keep their private information confidential ($p < 0.0001$). There were also differences between the lack of trust in the doctor and the nurse; these were sometimes greater in the case of doctors than nurses ($p = 0.01$); trust in the decisions and opinions of the doctor or nurse was found to be greater in the case of doctors than nurses ($p = 0.003$) and in the belief that the doctor is an expert

in the treatment of diseases and the nurse is an expert in nursing care; this was greater in the case of doctors than nurses ($p= 0.0004$).

Table 2. Respondents' responses to scale issues

Item	N=1200	Categories					Mean/ SD	P value
		1 pts	2 pts	3 pts	4 pts	5 pts		
I	Nurse	22.5%	39.8%	27.0%	9.3%	1.3%	2.27±0.96	<0.0001
	Doctor	18.5%	32.8%	60.6%	13.8%	4.25%	2.53±1.07	
II	Nurse	1.2%	7.5%	48.0%	35.2%	8.2%	3.42±0.79	NS
	Doctor	4%	8%	39.4%	37.1%	11.5%	3.44±0.93	
III	Nurse	2.7%	6.8%	30.3%	53.5%	6.7%	3.55±0.82	<0.0001
	Doctor	2.1%	5.2%	24.8%	50%	17.9%	3.77±0.87	
IV	Nurse	4.5%	9.7%	34.0%	43.0%	8.8%	3.42±0.94	<0.0001
	Doctor	3.1%	8%	28.6%	47.4%	12.9%	3.59±0.92	
V	Nurse	15.3%	34.7%	29.5%	15.8%	4.7%	2.60±1.07	0.01
	Doctor	9.75%	34.8%	29.8%	20.3%	5.3%	2.77±1.05	
VI	Nurse	2.8%	11.3%	28.9%	49.3%	7.6%	3.48±0.89	0.003
	Doctor	3.1%	9.1%	29.2%	43.6%	15.3%	3.59±0.95	
VII	Nurse	14.0%	25.8%	36.0%	17.5%	6.7%	2.77±1.10	NS
	Doctor	11.6%	33.3%	27.2%	22.3%	5.3%	2.77±1.09	
VIII	Nurse	1.0%	6.2%	29.7%	54.7%	8.5%	3.64±0.77	NS
	Doctor	2.3%	7.3%	26.8%	47.9%	15.3%	3.66±0.92	
IX	Nurse	3.7%	6.7%	31.2%	48.3%	10.2%	3.55±0.90	0.0004
	Doctor	1.6%	7.6%	28.2%	47.3%	15.3%	3.68±0.88	
X	Nurse	8.8%	13.3%	44.2%	28.8%	4.8%	3.08±0.98	<0.0001
	Doctor	4.7%	15.5%	24.8%	47.3%	15.3%	3.38±0.99	
XI	Nurse	38.0%	17.1%	27.0%	9.3%	8.6%	2.33±1.30	<0.0001
	Doctor	26%	25.7%	21.1%	22.8%	4.4%	2.54±1.22	
TOTAL	Nurse	10.4%	16.2%	33.4%	33.1%	6.8%	3.16±1.1	NS
	Doctor	7.9%	17%	28.2%	36.4%	10.5%	3.24±1.1	

NS- not significant

No statistically significant differences were found in regard to the belief that the doctor or nurse usually considers the patient's needs and puts them first; the perception that the doctor or nurse does not do everything he or she should in terms of medical care; and trust in the doctor regarding the method of treatment and in the nurse regarding the care method. Details are shown in Table 2.

No significant differences in the level of trust placed in male doctors and nurses were found.

Statistically, patients were more often convinced that a doctor rather than a nurse could say that they had made a mistake ($p < 0.0001$), that a doctor considers the patient's needs more than a nurse, and that patients trust and follow their doctor's advice more than they do with Nurses ($p = 0.0002$).

Patients had greater concerns about nurses than doctors about secrecy ($p < 0.0001$). Details are shown in Table 3.

Significant ($p < 0.0001$) differences (in favour of doctors) in the level of trust placed in women by doctors and nurses were demonstrated.

Significant differences in favour of doctors compared to nurses were found in matters of patient care ($p < 0.0001$), always following his advice ($p < 0.0001$), belief that he tells the truth more often ($p < 0.0001$), trust in his decisions and opinions ($p < 0.0001$), and belief that the doctor is an expert in the treatment of diseases.

Patients were more in favour of nurses when it comes to considering patient needs and putting them first ($p = 0.0121$) and sometimes trusting them more than doctors ($p < 0.0001$).

Women were also more convinced that nurses did not do everything they should for their medical care ($p < 0.0001$) and that the doctor may not keep a secret more often than nurses do ($p < 0.0001$). Details are shown in Table 4.

No significant ($p < 0.0001$) differences in the level of trust held by city residents towards doctors and nurses were demonstrated.

Significant differences in favour of doctors compared to nurses were found in matters of advice ($p < 0.0001$), belief that the doctor tells the truth more often ($p < 0.0001$), trust in his decisions and opinions ($p < 0.0001$) and greater faith in his judgements and opinions ($p < 0.0001$).

Significant differences compared to nurses were found in the belief that the doctor does not do everything he should for proper care ($p < 0.0001$), in occasional distrust of the doctor ($p < 0.0001$) and in the more common belief that a nurse is more of an expert in nursing than a doctor is in treatment ($p = 0.0022$). Details are shown in Table 5.

Table 3. Statistics of the scale patient-doctor trust depending on men

Item	No= 600	Categories					Mean/SD	P value
		1 pts	2 pts	3 pts	4 pts	5 pts		
I	Nurse	28.3%	36.8%	20.7%	11.5%	2.6%	2.23±1.07	NS
	Doctor	25.8%	34.7%	25.3%	10.2%	4%	2.32±1,08	
II	Nurse	1.7%	8.3%	62.7%	20.3%	7%	3.23±0.78	0.0009
	Doctor	4.7%	9.3%	40.8%	32.2%	13%	3.4±0.98	
III	Nurse	2%	8.8%	27.2%	57%	5%	3.54±0.80	0.0002
	Doctor	1.7%	6.2%	27.2%	48.8%	16.2%	3.72±0.87	
IV	Nurse	4%	12.7%	34.7%	36%	12.7%	3.41±0.99	NS
	Doctor	3.2%	11.3%	33.2%	40.5%	11.8%	3.47±0.95	
V	Nurse	18%	40%	23%	14%	5%	2.48±1.09	NS
	Doctor	15.7%	47%	21.3%	12%	4%	2.42±1.02	
VI	Nurse	2.3%	14.3%	22.8%	52%	8.5%	3.5±0.92	NS
	Doctor	3.5%	13.7%	28%	40.3%	14.5%	3.49±1.01	
VII	Nurse	19.7%	35%	22.5%	16.7%	6.2%	2.55±1.56	0.0005
	Doctor	14.5%	22%	33.8%	26.8%	2.8%	2.82±1.08	
VIII	Nurse	0.8%	7.3%	25.5%	54.3%	12%	3.69±0,81	NS
	Doctor	2.5%	9.3%	24.8%	48%	15.3%	3.64±0.93	
IX	Nurse	4%	6.7%	24.2%	51.7%	13.5%	3.64±0.92	NS
	Doctor	2%	6.3%	31.2%	37%	23.5%	3.74±0.95	
X	Nurse	6.2%	8.2%	43.7%	36%	6%	3.28±0.92	<0.0001
	Doctor	6%	12.7%	37.5%	35.3%	8.5%	3.78±0.99	
XI	Nurse	27.5%	17.2%	27%	15.3%	12.2%	2.68±1,34	<0.0001
	Doctor	44.5%	27.3%	17%	6.5%	4.7%	1.995±1.14	
TOTAL	Nurse	10.4%	17.8%	30.4%	33.2%	8.2%	3.11±1.11	NS
	Doctor	11.3%	18.2%	29.1%	30.7%	10.8%	3.11±1.16	

NS- not significant

Table 4. Statistics of the scale patient-doctor trust and scale reliability depending on women

Item	After 600	Categories					Mean/SD	P value
		1 pts	2 pts	3 pts	4 pts	5 pts		
I	Nurse	16.7%	42.8%	33.3%	7.2%	0	2.31±0.83	<0.0001
	Doctor	11.2%	31%	35.8%	17.5%	4.5%	2.73±1.01	
II	Nurse	0.7%	6.7%	33.3%	50%	9.3%	3.61±0.77	0.0121
	Doctor	3.3%	6.6%	38%	42%	10%	3.49±0.88	
III	Nurse	3.3%	5%	33.3%	50%	8.3%	3.55±0.85	<0.0001
	Doctor	2.5%	4.2%	22.5%	52.2%	19.7%	3.81±0.88	
IV	Nurse	5%	6.7%	33.3%	50%	5%	3.43±0.88	<0.0001
	Doctor	3%	4.67%	24%	54.3%	14%	3.72±0.87	
V	Nurse	12.7%	29.3%	36%	17.7%	4.3%	2.72±1.03	<0.0001
	Doctor	3.8%	22.7%	38.3%	28.5%	6.7%	3.12±0.96	
VI	Nurse	3.3%	8.3%	35%	46.7%	6.7%	3.45±0.86	<0.0001
	Doctor	2.7%	4.2%	30.3%	46.8%	16%	3.69±0.88	
VII	Nurse	8.3%	16.7%	49.5%	18.3%	7.2%	2.99±0.98	<0.0001
	Doctor	8.7%	44.5%	20.5%	18.7%	7.7%	2.72±1.1	
VIII	Nurse	1.2%	5%	33.8%	55%	5%	3.58±0.72	NS
	Doctor	3%	5.3%	28.7%	47.8%	15.2%	3.67±0.9	
IX	Nurse	3.3%	6.7%	38.2%	45%	6.8%	3.45±0.85	0.0006
	Doctor	1.2%	8.8%	25.2%	57.7%	7.2%	3.61±0.79	
X	Nurse	11.5%	18.5%	44.7%	21.6%	3.7%	2.86±0.996	<0.0001
	Doctor	3.3%	18.3%	12.2%	59.2%	7%	3.48±0.98	
XI	Nurse	48.5%	17%	26.2%	3.3%	5%	1.99±1.15	<0.0001
	Doctor	7.5%	24%	25.2%	39%	4.3%	3.09±1.05	
TOTAL	Nurse	7.98%	11.3%	51.1%	25.4%	4.2%	3.09±1.05	<0.0001
	Doctor	4.6%	15.8%	27.3%	42.1%	10.2%	3.35±1.01	

NS- not significant

Table 5. Statistics of the scale patient-doctor trust and scale reliability depending on city

Item	No=665	Categories					Mean/SD	P value
		1 pts	2 pts	3 pts	4 pts	5 pts		
I	Nurse	15.8%	46%	23.8%	13.8%	0.6%	2.40±0.9	NS
	Doctor	19.7%	31.4%	31.7%	12.2%	4.96%	2.51±1.09	
II	Nurse	1.8%	12.8%	36.5%	44.1%	5.4%	3.40±0.83	NS
	Doctor	3.5%	7.2%	39.7%	35.3%	14.3%	3.50±0.94	
III	Nurse	3.6%	9.5%	32.5%	43%	11.4%	3.50±0.90	<0.0001
	Doctor	1.2%	4.7%	20.6%	53.4%	20.1%	3.91±0.83	
IV	Nurse	3.3%	13.2%	57.6%	24.4%	1.5%	3.11±0.75	<0.0001
	Doctor	2.3%	8.4%	26%	48.6%	14.7%	3.65±0.91	
V	Nurse	7.7%	16.4%	46.6%	25.9%	3.5%	3.00±0.93	<0.0001
	Doctor	11.9%	42.1%	26%	14.7%	5.3%	2.60±1.04	
VI	Nurse	4.2%	17.6%	35.8%	32.8%	9.6%	3.30±1.00	<0.0001
	Doctor	2.6%	10.5%	21.4%	47.5%	18%	3.80±0.97	
VII	Nurse	7.5%	25.9%	36.99%	23.9%	5.7%	2.90±1.01	0.0001
	Doctor	12.3%	36.8%	25.1%	20.2%	5.6%	2.70±1.09	
VIII	Nurse	0%	9.9%	17.6%	59.8%	5.7%	3.80±0.798	NS
	Doctor	1.8%	7.2%	18.9%	52.8%	19.2%	3.80±0.89	
IX	Nurse	0%	6.3%	17.6%	59.8%	12.6%	3.90±0.76	0.0022
	Doctor	1.5%	4.5%	23.9%	47.7%	19.4%	3.76±0.9	
X	Nurse	0%	15.3%	32.6%	42.4%	6.6%	3.40±0.82	NS
	Doctor	4.5%	20.9%	31.1%	43.9%	10.7%	3.51±0.96	
XI	Nurse	20.8%	29.3%	31.3%	12.6%	6%	2.30±1.1	NS
	Doctor	34.1%	31.3%	20.9%	8%	2.7%	2.21±1.16	
TOTAL	Nurse	5.9%	18.4%	33.8%	34.8%	7.14%	3.21±1.01	NS
	Doctor	8.7%	17.9%	25.9%	34.9%	12.5%	3.32±1.15	

NS- not significant

Table 6. Statistics of the scale patient-doctor trust and scale reliability depending on country

Item	After 535	Categories					Mean/ SD	P value
		1 pts	2 pts	3 pts	4 pts	5 pts		
I	Nurse	30.8%	32.1%	31%	3.7%	2,2%	2.14±0.97	<0.0001
	Doctor	17%	34.6%	29.2%	15.9%	3.4%	1±0	
II	Nurse	0.7%	0.9%	62.2%	24.1%	11.96%	3.46±0.74	<0.0001
	Doctor	4.7%	8.97%	39.1%	39.3%	8%	1.92±0.68	
III	Nurse	1.5%	3.6%	27.7%	66.5%	0.7%	3.61±0.64	<0.0001
	Doctor	3.2%	5.8%	30.1%	45.8%	15.1%	2.29±0.76	
IV	Nurse	5.98%	5.2%	4.7%	66.2%	17.9%	3.85±0.98	<0.0001
	Doctor	4.1%	7.5%	31.8%	45.98%	10.7%	2.08±0.74	
V	Nurse	23.7%	56.3%	12.7%	2,3%	5.05%	2.09±0.95	<0.0001
	Doctor	7.1%	25.8%	34.6%	27.1%	5.4%	1.58±0.49	
VI	Nurse	1.1%	3.6%	20.4%	69.9%	5.05%	3.05±0.53	<0.0001
	Doctor	3.7%	6.9%	38.9%	38.7%	11.8%	2.15±0.75	
VII	Nurse	22.1%	25.8%	34.8%	9.5%	7.9%	2.55±1.16	<0.0001
	Doctor	10.7%	28.8%	29.7%	25.98%	4.9%	1.37±0.48	
VIII	Nurse	2.2%	1.5%	44.7%	48.2%	3.4%	3.49±0.69	<0.0001
	Doctor	3.9%	7.5%	36.4%	41.9%	10.3%	2.098±0.74	
IX	Nurse	8.2%	7.1%	48%	34%	2.6%	3.16±0.91	<0.0001
	Doctor	1.7%	7.7%	33.5%	46.9%	10.3%	2.35±0.65	
X	Nurse	19.8%	10.8%	54.8%	11.96%	2.6%	2.67±1.01	<0.0001
	Doctor	4.9%	22.6%	17%	51.4%	4.1%	1.71±0.45	
XI	Nurse	59.4%	1.9%	21.7%	5.2%	11.8%	2.08±1.44	<0.0001
	Doctor	15.9%	18.7%	21.3%	41.1%	2.99%	1.07±0.25	
TOTAL	Nurse	15.9%	13.5%	32.9%	31%	6.6%	2.92±1.13	<0.0001
	Doctor	6.98%	15.9%	31%	38.2%	7.9%	3.25±1.04	

NS- not significant

In general, and in all aspects of trust examined, differences in the level of trust given to the doctor and the nurse by the villagers were demonstrated in the doctor's favour ($p < 0.0001$). Details are shown in Table 6.

DISCUSSION

One objective of the study was to determine whether patients have higher trust in doctors or nurses. Our data support the conclusion that, in general, levels of patient–doctor trust and patient–nurse trust do not differ. Nevertheless, patients in the sample mostly believed that doctors cared about them more than nurses did and could be more easily informed of their mistakes. In particular, women and rural residents in the sample had far more trust in doctors than in nurses.

Blenkiron [17] and Mahoń [13] have suggested that to produce good outcomes, treatments require both the professionalism of healthcare professionals and patients' satisfaction with treatment in their subjective assessments of the quality of medical services. Along with participating in making decisions about their treatment, today's patients also expect medical services from healthcare providers that reflect up-to-date medical knowledge. At the same time, today's healthcare systems no longer cast patients as passive recipients of medical services but as clients who assess and judge the facilities where they receive healthcare services, including ones provided by nurses. According to Lawthers [18] patients' most important expectations of healthcare professionals are easy accessibility, good communication and information provided in understandable ways, respect for patients' rights and preferences when choosing treatment methods, the continued coordination of therapeutic processes, psychological comfort and care matched to individuals' expectations and needs.

Amongst the most important types of healthcare services, nursing care is provided in direct contact with patients and forms part of a subsystem that determines the overall quality of services [14,19].

In 200 described five thematic groups of features of so-called “good care”: meeting patients' needs, treating patients in caring ways, caring about patients, being competent and providing prompt care [20].

Pałyska et al., [21] have suggested that viewing care from the patient's perspective prioritises his or her satisfaction, which reflects the extent to which the quality of care meets the patient's requirements and affords him or her expected benefits. In subjective assessments of the quality of medical services, patients take into account not only the quality of services

provided but also the entire environment of service provision, the attitudes of medical staff and even interpersonal relationships amongst staff members.

According to Mahoń [13] the results of scientific research indicate that satisfaction with nursing care is the most important indicator of patients' satisfaction with the overall care provided in healthcare facilities.

Amongst other findings, the more attention that nurses devoted to patients, the greater the patients' satisfaction, and nurses' emotional commitment to patients' care increased their satisfaction as well [9]. Patients have added that the emotional involvement of nurses in providing care was even more important to them than their manual or technical skills.

Lemke [22] has shown that amongst patients who rated their hospital stays as being very satisfactory, 95% also rated their nursing care as being very satisfactory. By contrast, amongst patients who rated their overall stays as being less satisfactorily, only 34% rated their nursing care as being very satisfactory.

According to of factors affecting patients' overall satisfaction with their hospital stays, nursing care was the most important, followed by the hospital admission procedure, the care environment, the staff's approach to family and relatives, medical care and discharge from the hospital. They added that to improve satisfaction with overall hospital stays, it is first necessary to improve patients' satisfaction with nursing care [23].

In Jóźwicka et al. [24] study, doctors showed subjectivity for patients under their care. Respect for the patient's personal dignity was described by 62% of patients as being very good, 30% as being good and 6% as being average. More than half of the patients rated the provision of information as being very good or good, and only 3.6% of respondents were dissatisfied with the information that they had received over the course of their care [24].

Analysing patients' sense of trust in doctors, Derczyński [25] demonstrated that most respondents in his sample trusted the doctors who were treating them. Although only every sixth respondent described that trust as being very high, nearly two-thirds of the sample reported that it was high.

Krajewska-Kułak et al. [7] compared patients' trust in doctors in Poland versus Belarus. Their results revealed that, amongst Polish and Belarusian respondents respectively, 53.3% and 56.0% did not doubt that they have received appropriate care; 83.3% and 56.0% claimed that their doctors usually considered their needs and prioritised their prerogatives; 91.7% and 56.0% trusted their doctors and always followed their doctors' advice; 76.6% but only 8.0% were convinced that their doctors always told the truth; and 76.7% and 84.0% believed that their doctors were truly experts in treating their diseases.

According to Świdyrowicz, [26] literature addressing patients treatment contains several studies showing that patients perceive the need to receive detailed information about their health, progress in treatment and the risks of therapy. People seeking information about their health should receive comprehensive answers, which reduces their sense of anxiety and encourages them to actively participate in their treatment. On the contrary, poor communication is one of the most common causes of malaise in patients.

In a representative sample of 989 randomly selected adult residents in Poland, the Public Opinion Research Center (CBOS) [27] observed that 70% of respondents appreciated the competence of doctors, and most reported that primary care physicians tend to correctly recognise patients' problems.

CONCLUSION

In conclusions, in general, levels of patient–doctor trust and patient–nurse trust do not differ. Nevertheless, patients in the sample mostly believed that doctors cared about them more than nurses did and could be more easily informed of their mistakes. In particular, women and rural residents in the sample had far more trust in doctors than in nurses.

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Globalisation in the aspect of doctor's work

Michał Borkowski¹, Natalia Markiewicz¹, Magdalena Noparlik¹, Paweł Borkowski¹, Jan Gnus^{1,2}, Łukasz B. Lewandowski¹, Jadwiga Kuciel-Lewandowska¹

1. Wrocław Medical University
2. Regional Specialist Hospital in Wrocław, Research and Development Center, Wrocław

INTRODUCTION

Globalization is a continuous process that began in the era of great geographical discoveries, and its greatest intensity falls at the turn of the 20th and 21st centuries. Depending on the source, it is defined as tendencies or processes leading to the unification of the image of the world, which consists of phenomena related to economics, politics, social life and culture [1]. None of us can avoid the effects of this phenomenon and it has an obvious impact on our lives.

The spontaneous development of humanity leads to unprecedented diffusion of societies around the world. This is the result of influencing us with various new technologies that make it easier for us to travel and communicate. Globalization has a obvious positive impact on human life, e.g. by giving him new opportunities to learn, work and develop, which has never been seen before. However, it also carries many risks that directly and indirectly affect the work of a doctor.

Medics from various backgrounds have to deal with diseases that were previously unknown to them. Societies around the world must be constantly prepared for epidemiological threats arising from unstoppable global travel and population migration. Never before has the transformation of a local epidemic into a global threat in the form of a pandemic been so easy. The progressing unification of the world and the creation of a global village irreversibly change the reality of everyday work of doctors [A]. They pose unique problems and challenges that will change their business forever. This makes the analysis of the impact of globalization on the work of medical professionals multidimensional and complex, and this issue should be one of the most important in today's discussion on health care.

PANDEMICS AND EPIDEMICS – IMPLICATIONS

An epidemic is defined as a situation in which people are diagnosed with the occurrence of a specific disease in a specific time and area, in a number of cases higher than average [B]. In contrast, a pandemic is called an epidemic of particularly large size, covering countries and even continents [C]. Both phenomena pose a great challenge to health care. In recent days, the SARS-CoV-2 coronavirus-induced pandemic is impossible to overlook, in which globalization has played a significant role. On March 11, 2020 The World Health Organization (WHO) has announced that the disease caused by this virus, COVID-19, has been characterized as a pandemic [D]. Coronaviruses are a large family of viruses that cause diseases - from the common cold to such as severe acute respiratory syndrome (SARS) [E]. Novel coronavirus has gained considerable public and government attention in recent weeks, shortly after its first detection in the Chinese province of Wuhan in December 2019, followed by an epidemic in China, the Republic of Korea, Iran and Italy. From March 12, 2020 125,000 cases from 118 countries and territories were reported globally, with forecasts indicating that the number of subsequent cases will increase rapidly [F]. This led the government to support a range of public health activities recommended by the WHO, including four key aspects of the action:

1. Preparation and readiness - countries should quickly and firmly strengthen preparedness and response based on their national risk assessment and the four transmission scenarios provided by WHO for countries without identified cases, with identified first cases, case groups and identified transmission communities.
2. Detection, protection and treatment - all countries should consider combining response measures: finding a disease case and finding a patient's contact network; restraining or other measures to delay the appearance of soaring numbers patient increases where possible; the use of measures such as public awareness, promotion of personal hygiene measures, preparation of health care systems for the rapid growth of seriously ill patients, stronger infection prevention and control in healthcare facilities, social care homes and long-term care facilities, as well as postponing or canceling large-scale public meetings.
3. Reduction of transmission - countries where there are no or only several cases of COVID-19 infection should consider: active supervision of these people, isolation, promotion of practicing social distance, preparation of their healthcare system and population for the spread of infection.

4. Introducing innovation and developing science - low- and middle-income countries that require WHO support should be fully technically and financially supported. Financial support should be sought by these countries and WHO.

STAG-IH (Strategic and Technical Advisory Group for Infectious Hazards) emphasizes the importance of further rapid sharing of public health data in medical journals that provide quick reviews and online publications - without a subscription fee. Sharing information in this way, as well as collaboration between clinicians, epidemiologists and virologists, provides the world with an understanding of COVID-19 [2].

Cooperation of professions related to health promotion, both clinical and non-clinical - epidemiologists, scientists dealing with environmental health, public health doctors, infectious disease specialists, general practitioners, nurses, related health professions, health decision makers, health planners, health geographers and many more - is important in reducing the risk associated with pandemics and raising public awareness of the real threat and the possibilities of preventing it or reducing the rate of its spread. The profession of health promoter plays a significant role in pandemics, and this is clearly seen in countries associated with COVID-19. Health and hygiene news, in particular handwashing, are one example of the role that health promotion has played using the expertise in health education and the implementation of health-related mass media and social marketing. Over the past two decades, information technology and social media have changed the way we can reach people during an epidemic. Indeed, globalization and social media have made it easier to reach large populations and even entire countries, giving the opportunity to spread information, especially for those at risk. On the other hand, social media can have its negative effects. Misinformation and fake news are common today. Potentially, this can stifle efforts to promote health. Therefore, it is important to know the source of the information being disseminated and ignore those that are not reliable. In health promotion, it is important to look for new communication strategies. Health messages should be formulated in a concise manner, which significantly facilitates the understanding, navigation and action of citizens. It is also worth considering how the content is transmitted via electronic communication channels and find the right frequency level of such communication to achieve optimal impact. Without this, there is the possibility of strengthening social ambivalence at one end of the spectrum and causing panic at the other [G]. It is worth considering who is the most sensitive during a pandemic. Although COVID-19 has the potential to affect every individual in society, these effects will be felt depending on how you prepare, protect, treat, limit transmission, and introduce innovative solutions. It is important to recognize that pandemics are influenced by social, economic and political health

determinants. The WHO Director-General stated that: "All countries must strike with an exact balance between health protection, prevention of economic problems and social disturbances and at the same time respect for human rights" [F].

Despite the fact that we still know little about the nature of SARS-CoV-2, we have information on how pandemics can affect sensitive populations. We know that developing countries do not have properly prepared health systems, health resources and health infrastructure ready to slow down the harmful effects of COVID-19. It is known that there are vulnerable populations such as the elderly, people with disabilities, people deprived of liberty, island communities, people with chronic diseases and people from culturally and linguistically diverse environments. COVID-19 will have a disproportionate impact on their lives, especially when health promotion is absent [3]. It is also known that it is highly probable that people from low socioeconomic strata, many racial and ethnic minorities, will not have sufficient financial resources to provide themselves with adequate conditions to isolate and reduce the spread of disease at home [4].

Despite the fact that we may not be able to accurately predict the time of occurrence of natural disasters and infectious pandemics, we can be certain they will reappear in the future. The COVID-19 coronavirus experience is not the first and will not be the last. As recent events have shown, telemedicine plays a large role in the fight against similar phenomena. Its advantages include the ability to quickly implement a large number of health services, facilitate medical segregation to improve the operation of medical services, provide clinical services in the event of damage to local clinics or hospitals, and reduce the risk of infectious diseases transmitted through personal contact. There are also restrictions on the use of this technology. Some consultations require physical examinations that may be difficult to perform remotely, e.g. auscultation, and specialized diagnostics impossible to perform at the patient's home, e.g. imaging using computed tomography or grow of the bacterial culture. It is important not to forget about restrictions on the use of telemedicine during the clinician's training today. The importance of providing remote healthcare also applies to people who are not infected during a pandemic. It is important that the development of telehealth strategies to deal with global and national crises be based on the assumption that telemedicine can become a major element of the health care system. This assumption can be realized through [G]:

- Providing all healthcare professionals with adequate education and training in telemedicine
- Introduction of telemedicine accreditation for healthcare professionals
- Provide financing that adequately covers the costs of telemedicine

- Redesign of the clinical care model
- Support all stakeholders with effective communication and change management strategy
- Establish systems to manage telemedicine services in routine cases

Considering all the above-mentioned requirements, their complex nature and all aspects related to the occurrence and spread of a pandemic, it is easy to say that health care has found itself in a sensitive and demanding moment in history. It will irrevocably change our image of medicine and allow us to draw conclusions that will help humanity in the future.

MEDICAL TOURISM AND ITS GLOBAL IMPACT

The global increase in the flow of patients and medical staff, as well as the development of medical technologies have led to the emergence of new patterns of consumption and production of medical services in recent decades. Free movement of goods and services contributed to the liberalization of trade in health services.

Medical tourism is one of the forms of health tourism, which is rapidly growing on the global market. In medical tourism, patients travel to neighboring or distant countries to receive medical help, often in combination with rest and treatment. Its development is also observed in Poland. Citizens of Germany, Great Britain and Scandinavian countries are increasingly using health services offered in Polish.

Development of medical tourism has its beginning in ancient times, when journeys took place not only for religious, commercial or military purposes, but also to improve physical and mental health. However, in the first decade of the 21st century, the development of medical tourism became more dynamic, mainly due to its promotion in mass media such as television, internet and magazines [5]. In the era of easily accessible and efficient communication, trips to other countries are more and more common. People who want to look for savings and the best standard of living successfully use the opportunities of medical tourism to take care of health.

That is why the development of this tourism sector will accelerate even more in the coming years.

Health is paramount for most people. WHO defines it as a state of complete physical, mental and social well-being, and not just no illness or impotence [H]. Achieving or at least approaching this state gives a person more confidence, the ability to achieve higher earnings or better contact with other people.

Medical tourism is part of the phenomenon of health tourism, which includes [6]:

1. Spa tourism - present in recreational areas, which includes treatment of chronic diseases, rehabilitation, preventive education and health promotion.
2. Spa and tourist well-being - containing cosmetic treatments, massages, gymnastics as well as typical products stimulating physical and mental well-being, such as stress management methods and motivational workshops.
3. Medical tourism - traveling to other countries to obtain health services combined with relaxation, physical and mental regeneration, sightseeing and entertainment in the broad sense.

This term is usually associated as much with traveling to another country as with traveling along one's own country to obtain health services at the highest level [7].

The correct identification of a person involved in medical tourism is an important issue. There are terms in the literature such as: customer, consumer, patient, tourist, patient-tourist and tourist-patient [5]. The term 'medical tourist' is considered to be the most accurate, mainly because the patient does not travel alone and largely combines travel to improve health with a regular tourist trip.

The phenomenon of medical tourism can also be considered from the point of view of economics, which distinguishes two types of medical tourism, resulting from [5]:

1. Quality orientation - including medical tourists that come from economically underdeveloped countries who are looking for medical services in other countries with the highest quality level or better treatment offer, or want to emphasize their own socio-professional status.
2. Expenditure orientation - including medical tourists that come from developed and economically prosperous countries, looking for services of similar quality at lower costs.

The WHO distinguishes several factors influencing the development of the phenomenon of medical tourism: more advanced technologies, better quality of care, faster access to treatment and lower costs associated with it. Along with its continuous development, the need to add attractions and amenities also increases. Countries and institutions that are tourist destinations are using more and more marketing tools to attract the largest number of customers and create attractive offers for them. In order to better adapt to market needs, new professions have been created to meet customer requirements. That's how the Mediator of Medical Tourism was created, which mediates in organizing and supporting the management of medical tourists [8]. This mediator is also responsible for contact and assistance given to tourists at various stages of their trip, ranging from location decisions to the organization and

control of the medical facility where the services will be performed. In Poland, this profession is known as a specialist in the field of medical tourism. Due to the high demand for this profession, many universities began to open faculties in this field, including the Warsaw School of Economics and the Lazarski University.

In recent decades, the global increase in the flow of patients, health professionals and medical technologies has led to the emergence of new models of consumption and production of medical services. The free flow of goods and services has contributed to the liberalization of the medical services market. One of the first countries that noticed the potential of health tourism and began to use it, attracting thousands of patients, was India. In 2009-2011, the number of medical tourists in India increased by 30% [I]. The next leading countries are Singapore, Thailand and Mexico. In Europe, they are Turkey, Hungary and the Czech Republic.

The main reason for this situation is the high level of services rendered in the abovementioned countries.

In connection with the approval by the member states of the European Union Directive on the proposal on the rights of patients in foreign healthcare (Directive 2011/24 / EU of the European Parliament and the European Council of March 9, 2011), patients have the freedom to choose healthcare throughout the European Union. As a result, public and private hospitals in the EU can treat foreigners, and the cost of treatment is covered by the health care of the country of origin of the patient in the same value of the service that applies in the patient's country of origin. Outside the European Union, Turkey is a country with a great development in medical tourism. In 2010-2014, there was a 32% increase in the medical tourism market, and the goal of the Turkish government is to reach USD 10 billion in 2016, with the support of 1 million patients [J]. The development of medical tourism is also present in Poland. It is estimated that in 2012 the market reached PLN 800 million thanks to 300,000 patients from other countries. Dental and plastic surgery services as well as stays in health departments are most often chosen by foreigners.

The strong increase in medical tourism is mainly due to the increased awareness of citizens and lifestyle changes. It is expected that this growth will continue in the near future, which is influenced by several factors [5]:

1. Reform of health systems providing subsidies for services provided outside the country.
2. Lowering the prices of medical services in other countries.
3. Poorly developed health care in other countries.
4. Long queues for surgery.

These are the main factors contributing positively to the development of medical tourism in the world. However, along with its development, there is also the development of competing and serving unique trend services [9]:

1. Development of natural medicine.
2. The increase in the popularity of organic products and an ecological approach to the medical industry.
3. Development of medical tourism based on a holistic health model.
4. Development of gastro-tourism, education and promotion of healthy eating.
5. Possibility of providing medical services in a more flexible way, for example in different locations, indicated by the patient - home, work place or hotel.

MIGRATION OF DOCTORS AROUND THE WORLD

The migration of medical workers is part of the phenomenon of internationalization of work and the consequence of introducing, to an increasing extent, liberal rules of moving between countries and living in specific countries. By narrowing the category of health care workers to doctors, nurses and midwives, WHO estimated a deficit of health care workers at around 4.3 million people. A large contrast was observed between highly developed and developing countries [10].

The issue of migration of doctors from developing countries to developed countries is the subject of many discussions around the world. This topic is often raised because of the general fear of further weakening and the progressively deteriorating health system, especially in developing countries with a huge population. WHO has repeatedly highlighted the need to strengthen systems, and one of the most important steps to achieve this was to discourage doctors from migrating. According to WHO, there are a shortage of over 4 million doctors and other healthcare professionals worldwide, and the region most affected by this deficit is Sub-Saharan Africa. Migration of doctors began in the 1950s and 1970s from India, Sri Lanka, Pakistan, Nepal and Bangladesh, and the wave of migration continues. India is the largest exporter of doctors to the United Kingdom, Canada, Australia and the Gulf countries. So far, about 6.5 thousand doctors from Pakistan left the country [11].

Migrations are social phenomena that respond relatively quickly to changes in political and economic conditions. That is why flows of people are fundamental to the functioning of labor markets. The relationship between migration and the labor market is two-sided. This means that on the one hand, population movements can have a modifying effect on the labor

market situation, on the other hand, a specific situation on the labor market can stimulate individual migration flows. The free movement of persons in the European Union is one of the fundamental freedoms guaranteed by Community law. On the other hand, employees are one of the categories of persons entitled under Community law to the freedom of migration [K].

Many discussions have been held to find the most effective way to solve the problems of medical migration in Europe. There have been skeptical opinions in recent debates that so-called 'replacement migration' is not a solution to Europe's demographic problems in the long run. This is explained by the fact that immigrants are getting older, and moreover, when they are rooted in a new society, they quickly adopt their own cultural customs and value system, which among other things means a decrease in fertility among immigrant groups. Recent discussions, however, are dominated by voices suggesting that this is the best short-term solution and the well-known selection of a foreign workforce may have a long-term positive impact on labor markets. It is primarily about selection due to qualifications as well as due to their lack [K]. A very important conclusion can be drawn from these discussions - skillful management of labor force migration seems to be the best short-term solution for an aging Europe at present, which is characterized by very low birth rates.

Migration of healthcare workers from low- and middle-income countries to rich countries is a long-term phenomenon that affects entire healthcare systems in the countries where these workers leave. In 2000, it was estimated that about 1.5 million specialists from developing countries work in industrialized countries [11]. Some sets of skills and competences are so specialized and have so little resources that they are acquired worldwide at all costs. Migration covers primarily healthcare workers, mainly doctors and nurses. As a result of this global movement, some health resources are lost to other countries. This significantly limits the ability of the healthcare system to provide services equally and access diagnostic and therapeutic procedures. This problem mainly affects developing countries. The migration of healthcare workers is therefore of great importance. Human resources are the foundation for the provision of health services, and policy plays a key role in implementing healthcare sector reforms in low- and middle-income countries and for achieving specific development goals [L].

Low and middle-income governments have expressed serious concern about the impact of migration on their healthcare systems. Clear arguments have been put forward for strengthening human resources for health in poor countries to limit the influx of medical workers from these countries. Factors affecting migration were investigated, these include push factors, i.e. factors pushing an employee towards work in another country, and pull factors, i.e.

attracting an employee to a given place of employment [M]. The first of them cover all the circumstances prompting a potential health care worker to leave the homeland and move abroad. These include low salaries, poor job security, inadequate infrastructure, insufficient drug supply, and no post-graduate education or career prospects. It is also worth mentioning such aspects as mediocre economic background, political instability or security problems in a given country [L,12]. What's more, the students' contact with the doctors who conduct the classes often discouraging graduates from staying in a low-income country is one of the most serious aspects of the migration problem. Attracting factors include all recruitment in highly developed countries, vacancies in specific jobs with high salaries and better conditions, facilities as well as access to training and opportunities for self-development [12].

A comprehensive approach to concluding international agreements to reduce the negative side effects of oversupply of healthcare workers in low and middle-income countries is a key to solving this problem. Cooperation between these countries makes it possible to strengthen general and specialized health services due to greater capacity to monitor and plan the movement of the workforce [L]. Migration of doctors will probably continue to be a problem until international aid develops a program to strengthen human capital and healthcare systems - that does not affect the right to health of the general population.

Although the migration of doctors to more developed countries is unfavorable for "exporting" countries, it is worth noting some interesting phenomenon. The "exporting" countries are making efforts to launch international medical training programs at their universities, which are then accredited by major organizations and associations, primarily those from the United States of America [13]. It can be speculated that the assumed goal here is to attract foreign students who are to pay for their studies in a selected country. International competition, however, forces universities to obtain international accreditation for most of their fields and specialties, including those where domestic students are educated. The result is a situation in which Indian universities, having the appropriate accreditation of American associations, train doctors ready to work in the USA, which only strengthens the brain drain from India. Therefore, one can risk the thesis that globalization in the field of medical staff education will strengthen the migration of this professional group to more developed countries [14].

Although human health resources in low- and middle-income countries should have increased significantly over the past 50 years, in practice (despite the considerable efforts of universities and colleges) there have been losses due to brain drain to richer countries that draw a significant proportion of their service workers health from poorer countries [L].

BRAIN DRAIN

Brain drain is a phenomenon of attracting high-class specialists to work in another, more industrialized country, by providing them with better economic conditions, modern work organization and better wages [N]. It is a flight of human capital that is a phenomenon in which qualified employees or young potentials migrate and leave their country. Although brain drain is nothing new, its effects are much greater in a globalized world where qualified employees can travel the world freely. Many countries have limited their migration policies, but highly qualified and well-trained employees are often welcome and even encouraged to come to the Western world [O].

There are many reasons for this qualified migration, which may vary from region to region. Skilled workers living in Eastern Europe are migrating to the United States (US) or European Union (EU) because they have greater career opportunities there, higher wages and better social security. African migrants sometimes run away from violence, poverty, political instability or corruption [O].

The modern education system has more and more market relations covering new countries. Globalization is primarily manifested in the possibility of learning mobility. The increase in international mobility of research staff is an integral feature of scientific globalization, and in this context the problem of brain drain in developing countries is particularly worrying. It has been estimated that between 15% and 20% of university graduates in Kazakhstan leave the country in search of self-fulfillment possibilities. The problem of brain drain is considered a threat to national security. Justifying brain drain is not enough to eliminate this phenomenon. Countries make serious political, social and legal efforts. However, the course of this phenomenon is practically impossible to reverse. Brain drainage should be officially recognized as a problem and, above all, comprehensive measures should be taken to reform education and science systems [15].

The process of political unification of countries should be used to create international research centers, higher education institutions and international educational funds. Undoubtedly, they would improve the quality of education and create more opportunities to find decent work in the country and minimize the need to look for offers outside the country. In addition, particular attention should be given to the creation of a national secondary and higher education system based on international practice as well as to achievements in the field of science, technology and humanitarian aid [15].

Globalization processes should also be considered in the opposite direction. World universities could help donor countries by creating scientific consultations, helping to prepare curricula based on global trends, and by updating certain fields of science or training doctors. It would be necessary to involve scientists from developing countries in research related to the problems of their homeland, the results of which could be used to solve more global problems on a larger scale. This would create conditions for exchanging experiences, receiving additional financial assistance, as well as using the scientific achievements of scientists without the phenomenon of brain drain outside the country. Globalization is the result of cross-border information exchange, while researchers and young people leaving the country are the result of social, political, financial and residual problems that solve the problems of science and education [15]. Thus, the solution to this problem is mainly based on solving the root causes and preventing their irreversible consequences.

The latest report shows that some regions of the European Union have been the victims of a brain drain. Poland is the largest of them. Almost 580 thousand Poles after studying in 2017 lived in another EU country. This is the highest number out of 28 countries belonging to the Union. In 2017, the EU population was around 511 million citizens. All people of working age who moved (mainly in search of better employment conditions) to another EU country, there were almost 17 million, or about 3%. Two EU countries attracted more than half of these people: Germany (33%) and the United Kingdom (20%). Of the 17 million Europeans who changed their country of residence, one in four (around 4.2 million) had a university degree. The share of highly qualified employees among migrants increased steadily in the years 2014-2017. The report shows that the highest number of the best educated people in 2017 came from Poland (around 577,000), Germany (around 473,000) and Romania (around 468 thousand). These people most often chose cities in the northern part of the Union: Sweden, Ireland, Estonia, Denmark, as well as in several regions of Great Britain. Italy turned out to be the least attractive to these employees. The authors of the report emphasize that in some regions of the Union, the free movement of employees has led to such a significant migration of well-educated people that, as a result, it has led to the phenomenon of brain drain. These movements were mainly due to growing competition and the fact that the regions where people leave are unable to create attractive conditions for these employees [P].

The social and economic situation of host regions is one of the basic foundations and reasons for attracting highly qualified migrants. In this context, the following were mentioned: economic growth, higher wages, solid social security, high Gross Domestic Product per capita,

language and cultural similarity, easier access to the labor market, as well as for example, a well-established knowledge-based economy.

In regions where there is drainage, negative conditions in the labor market motivate to leave - high unemployment and low wages, as well as administrative barriers, recession and bad political environment [P].

Brain drain may result in staff shortages, limited ability to innovate and the adoption of more advanced technologies by the state. Brain drain also has fiscal consequences - reducing tax revenues and market - reducing consumption. What's more, it limits economic growth and also reduces productivity. In turn, the positive effects for the regions to which qualified employees arrive are the result of an increase in innovation potential, economic growth, a growing competitive advantage as well as an increase in consumption and demand for services [Q].

Noted is also the so-called "brain waste", which happens when highly qualified employees migrate to another region (in this case in another EU country), not having sufficient knowledge about the demand for labor in a given country, city or workplace. This often results in unemployment or work in a profession that does not require his or her skills. Notwithstanding the regime for the free movement of workers, there are still some obstacles to labor mobility within the European Union. These obstacles are common to all employees, regardless of their skill level. Among them, the language barrier is key. Evidence of this is to be observed in the phenomenon of higher mobility in neighboring regions of different countries that have the same language common to each other. Other obstacles include cultural differences, a lack of information on local labor demand and a lack of full recognition of qualifications [Q].

MEDICAL MISSIONS

At the end of the colonial era, in addition to evangelizing activities for Christian missions, medical, charitable and educational assistance to indigent, peripheral communities of newly emerging independent countries became extremely important. It was a period of increased expansion and development of medical mission centers, also on the Polish side, despite many difficulties created by the then authorities. However, the role of medical assistance began to change as international public health programs developed into so-called global health. This very intensive development caused changes in health care and disease prevention in mission medical centers. Numerous non-religious governmental and non-governmental organizations began to allocating large sums to strictly controlled health

programs. The focus was primarily on combating acquired immune deficiency syndrome (AIDS), malaria, tuberculosis and caring for mother and child [16].

Successful cooperation with national health institutions requires many compromises, in particular the implementation of international standards for modern diagnostics, treatment and prevention of tropical diseases. Rational world indications cannot always be applied due to the local situation and culturally different communities in developing regions of the world whose beliefs conflict with the implemented health protection methods. In these cases, national medical associations and local episcopates play a very important role. The advancement of medical knowledge about medical missions requires the existence of well-stocked and well-run mission medical centers that need external help until developing countries are able to address the health problems that affect them [16]. The cooperation on which the missionary initiative is based consists of implementing actions aimed at changing human behavior, health education, and environmental protection. It is largely based on volunteering leading to significant strengthening of local social structures, the so-called community health [16].

The establishment and implementation of the development goals initiated a number of activities under the general name of global health. The emergence of dangerous epidemics has contributed to their emergence, regardless of "economic" globalization. As an example, we can use dengue fever on the South American continent, SARS, influenza, Ebola virus, and recently also the SARS-CoV-2 coronavirus epidemic, all crossing national borders thanks to the rapid and international communication. The threat to wealthy countries with massive cases of little-known viral infections forced them not only to seal their borders, but also to try to eliminate outbreaks on distant continents. Regardless of the institutions dealing with international health, incl. WHO and Centers for Disease Control and Prevention (CDC), a number of organizations have been created to deal with or support health care globally. However, they lack global coordination. The reason for this is the complexity of many factors determining the level of health in societies: social, economic, environmental and political. It is unlikely that the impact of these factors on global health will be effectively corrected at international level in the near future. Therefore, health protection at the basic level remain priority. Here, health-promoting activities of religious missions are of great importance, especially those with medical centers [17].

Effective eradication of smallpox in 1980 showed a real opportunity to control one of the most dangerous infectious diseases [O]. We owe it to our joint international effort. This situation has significantly put an emphasis on the global dimension of health care, which depends on the action of the world's population as a community, not on separate entities.

Together, significant progress has been made in looking after children under five and in combating leprosy. Significant progress has also been made in controlling AIDS, tuberculosis and malaria, but research and trials are still underway for their complete eradication [17].

The WHO has taken the initiative to combat previously underestimated tropical diseases. The role of missions is usually underestimated in strengthening social ties, which are extremely useful not only during epidemics and other natural disasters, but in the general concept of society as a united group. However, during some epidemics, the mission's involvement in the spiritual aspect, consisting of gathering the faithful at services or funerals, is inadvisable, such as during the recent Ebola epidemic in Africa. Mutual cooperation in the field of health protection requires not only mutual compromises, but also the adaptation of missionary medical centers to international standards, both in terms of professional and managerial [17].

With globalization, there has been a significant increase in the number of short-term business trips (MST) from high-income countries (HIC) to low-income and low-income countries (LMIC). Although MSTs provide significant care, medical literature and public health devote relatively little attention to the impact of these interventions on served populations [18].

Most of the health professionals involved in short-term medical missions came from the US. The United States, Canada, Australia and the United Kingdom are four countries which articles from the medical mission have been published in the last 25 years, indicating highest percentage of doctors sent on missions came from these countries. Regarding destination countries, the two main ones mentioned in the mission publications were Honduras and Papua New Guinea. The next to which medical missions were most often sent were: Afghanistan, Bolivia, China, Ethiopia, Haiti, India, Liberia, Mexico, Peru, Russia, Somalia, Sudan and Uganda. The US most often sends short-term medical missions to Honduras, Canada to Somalia, Australia to Papua New Guinea, and Great Britain equally to Sri Lanka, Peru, Ghana, Tanzania and Uganda. Most of the articles described short-term medical missions that were designed to meet the needs of patients arriving at a particular clinic or hospital. Missionary doctors provided patients with care in many different ways, depending on the diseases they were reporting on - from primary health care, injury response, mother and child care, to vaccine distribution and infectious disease management. Of these missions, which defined the health of patients and the problems they reported, the most common were: cleft lip and palate (23%), oral and dental health (6%) and vaginal fistula (5%) [17].

States send missionary doctors to countries with which they have existing political relations. For example, the United States sends most of its missions to Honduras, as well as to Nicaragua - two countries with which it has significant socio-political ties. This formula has been well documented in previous studies. For example, in their review of the distribution of foreign assistance, Alesina and Dollar analyzed the flow of bilateral assistance notified by the Organization for Economic Cooperation and Development and found a strong correlation between colonial status and the amount of foreign assistance received [19].

Every day about 22,000 doctors, nurses, midwives, clinical psychologists and representatives of many other professions work in various parts of the world, helping people who were deprived of medical care for various reasons. They are all members of one organization: Médecins Sans Frontières (Doctors Without Borders). The conditions in which they help are often very difficult, dangerous and require a lot of sacrifices. However, without this help, many people would not receive any support, and some would probably die of disease, hunger or wounds sustained in the fighting [R]. According to the WHO, the highest proportion of global disease burden falls on regions that also suffer significantly from shortages of doctors [20].

Health professionals have found that many missions have brought them many benefits, especially in spiritual and ideological matters. They defined missions as an opportunity to remember the reasons and the real reasons why they decided to become doctors [21].

The global ebbs of migration, both among doctors and patients, reveal many emotional conflicts that are relevant to us all. Feelings caused by uncertainty, war, torture, hatred and despair are balanced by feelings generated by heroism, beauty, kindness, compassion, humanity and empathy - values that should be an integral part of clinical practice. These conflicts can be hard to deal with. "*Hardened by a professional fight against tragedy and confusion, missionary doctors create a seemingly convenient border to stop the feeling of care and compassion*" [21]. To work as missionary doctors, including MSF in the new century, the need for a new shared set of professional values around global social responsibility arose [21].

Some diseases are rare in high-income countries, which means that doctors decide to volunteer for medical missions to improve their skills and test themselves in conditions where they would never be able to work otherwise. Medical missions are often unable to provide the full spectrum of care required for complex diseases. An example are patients with cleft lip and palate who need maxillofacial surgeons for the initial surgical repair of the abovementioned defects, with greater postoperative care often being required. Patients may require follow-up visits to general practitioners or plastic surgeons, future visits to orthodontists to repair

damaged teeth and jaws, and speech therapists to improve speech challenges - care that local conditions and staff cannot provide upon return of the missionary team to their homeland. In these cases, a long-term support is required that not every missionary physician can provide [22].

Members of the local community who received missionary assistance often stated that they felt the presence of medical missions. They were grateful that the outside world had recognized their difficult situation and was helping them in this way. They mentioned a sense of solidarity when foreigners come to new communities for them at the distant ends of the map, to provide medical assistance. Everyone replied that the arrival of the doctor, even for short periods of time, was extremely helpful to the community, because he was able to point out their problems and gave them hope that continuous help would be implemented and they would not be left alone [23].

However, many times attention is drawn to the less positive issue of medical missions to developing countries. It is said that the quality and effectiveness of medical care provided by foreign doctors is worrying. Presumably, they may not be familiar with local health needs, local culture, and the benefits and limitations of the healthcare system in which they must leave their patients for further care [24]. Doctors who are not qualified enough to carry out a certain type of surgery in their country are often put in situations during medical missions in which they must provide care for which they are not properly prepared or confident, and in their home country, they would never be allowed to carry out certain actions. Practicing physicians and surgeons may not be covered by the typical higher-level supervision they may have at home when performing procedures that they are not familiar with. This can cause serious medical complications for patients, and local doctors feel aversion to medical missions. However, it can be argued that this may be the best potential patient care at a specific place and time [17].

The Lancet report emphasizes that the need of the population is the basic link between vocational training on the one hand and health conditions on the other. The Lancet report also states explicitly that "20th century educational strategies are not suitable to meet the challenges of the 21st century" [25]. Medical education programs have remained unchanged with the development of the world [21].

IN CONCLUSION

The modern world is an extremely complicated place, full of divisions, inequalities and injustice. At the same time, it is becoming more and more uniform as a result of increasing

globalization. There are common difficulties that we all have to face. More and more people are struggling with similar problems that are becoming universal between social strata around the world. Similar phenomena affect national health systems and ideas that are above state divisions. Wealthier community members around the world have the opportunity to seek better healthcare in other countries on different continents. Increasingly, they combine leisure with medical consultation or minor surgery. At the other end of the spectrum are the poor, whose health depends only on what the country in which they live can offer. It is on them that all problems resulting from globalization leave their mark.

The consequences of this process for doctors are mostly positive. They open up the possibility of additional income in the medical tourism sector, allow for scientific exchanges and internships abroad. Emigration in search of a job offering better pay and conditions has never been easier. There are unique opportunities to gain professional experience, e.g. due to participation in medical missions in Third World Countries. Globalization, unfortunately, also has negative effects. Migrations are not just about healthcare professionals. Immigrants from poorer countries are coming in significant numbers to many countries, who often bring with them rare, exotic diseases that challenge local medics. Doctors are forced to treat complications of medical tourism procedures, often in the absence of adequate medical records of the patient. The national and global effects of globalization are also partly negative. There is a brain drain that negatively affects local healthcare systems. It is an additional challenge that the poor, with little to offer, state have to fight in order to maintain minimal health care for their citizens. It calls into question the stability of entire systems and leads to a deepening of inequalities between individual countries. In response, organizations are created to bring medical help to the poorest regions of the world.

There is no doubt that globalization is a process that will continue. Probably, as time passes, its development will accelerate, which will bring about unknown effects. It is necessary to create and adapt appropriate legal regulations that will limit the negative consequences of this process and will protect the most sensitive parts of our communities. At the same time, the promotion and development of all benefits of globalization is key. As humanity, we must make every effort to ensure that the positive consequences of this process can outweigh the negative ones.

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Patient's rights in Poland and in the world - between the East and the West

**Patryk Kubiak¹, Natalia Markiewicz¹, Jan Gnus^{1,2}, Bartosz Kurzelewski¹,
Karolina Pawlak², Aneta Hauzer¹, Anna Kolcz¹**

1. Wrocław Medical University
2. Regional Specialist Hospital in Wrocław, Research and Development Center, Wrocław

INTRODUCTION

Human rights are a universal idea aimed at protecting human dignity. The first documents about them were created in the 13th century. Since then, many organizations have been created whose main task is to safeguard compliance with the rules, including the United Nations Human Rights Council and the European Court of Human Rights.

Some of them include the right to life and health protection. Caring for these rights required formulating the concept of patient rights. The implementation of this initiative usually takes place separately in each country, in each of them to a different extent and with multiple effects.

Access to medical services is different in each country, which is largely determined by the economic and geopolitical situation of the region.

The availability of doctors and the approach to broadly understood prevention of many diseases is different. The multitude of factors that make up final health care can be overbearing. Seemingly unrelated events, which seem to be only its background, can have a colossal impact on health care.

Combining all the circumstances into a whole can be difficult, especially in the first attempts to reflect on this topic.

These issues are often the subject of ongoing social discussions. The legal status and its practical implementation are compared between countries around the world.

Debates on similar issues are the driving force of change that is necessary to improve the situation of patients in our country.

Before considering this topic, it is worth getting acquainted with the realities prevailing elsewhere in the world, both those less and more developed than Poland.

HEALTH CARE SYSTEMS

To date, no ideal healthcare system has been developed. Both those based on the private sector - e.g. the United States (US) - and those built on public funding do not provide comprehensive, unlimited care to all citizens.

Health care in the US is seen through the prism of the latest therapies and an extensive program for the development of new drugs as a health system close to ideal. In reality, however, it is a system of great extremes, in which all possible therapies and medicines are available, but a small part of society can actually use them. The privatization of healthcare has resulted in insurance companies and healthcare providers over valuing all medical procedures. As a result, insurance is expensive and does not cover all treatments and therapies. In addition, this has limited access for people who do not have health insurance. It is not uncommon for loans to be repaid for many years and taken to pay for the costs of treating illnesses or accidents that are trivial from a European point of view [1].

In Poland, the private sector is responsible for around 30% of healthcare financing [A]. The vast majority of costly services are carried out as part of public funding from the National Health Fund. Health insurance is easy to obtain - it is available to everyone working under a contract of employment or a mandate contract, registered unemployed, members of the insured's family, disabled persons, children and adolescents under 18 years of age, students and doctoral students up to a certain age, pregnant women, perinatal and many other social groups, e.g. drug addicts, patients with psychiatric diseases [A,B].

PROVIDING INFORMATION AND CONSENT FOR TREATMENT IN POLAND

The Act on the Medical Profession of 1996 gives the physician the obligation to provide full information about the state of health, diagnosis, possible therapy and prognosis to the patient to whom it relates in a clear and understandable way. However, the patient has the right to decide whether and to what extent he wants to receive such information, and who has the right to receive it in addition to him. In the absence of such authorization, information cannot be given to even the immediate family. The exception to this situation are cases of persons under 16 years of age, whose information is obtained by parents or legal guardians. Information may be transferred to previously unauthorized persons, but only those who can document a close relationship with patients - in case of patients with limited ability to understand the transmitted information and these in unconscious state. Because of the difficulty in establishing

the identity of the person calling or communicating through other media, the provision of information in this way should not be practiced.

VACCINATIONS

In recent years, the number of people avoiding vaccination in Poland has been increasing [C]. In connection with this trend, an increase in the incidence of diseases is observed, which previously has been very rare and was associated mainly with the migration of people from across the eastern border, where the vaccinated percentage was insufficient to provide collective immunity [D].

An example of such a disease is measles caused by a virus from the paramyxovirus family, which in 2019 affected nearly 1,500 people - up to 4 times more than previous year and almost 24 times more than in 2017 [E]. The trend continues for another year in a row and due to the fact that in Europe few countries have a vaccination rate at the level recommended by the WHO of 95% of the population, we are dealing with an increasing number of outbreaks of this disease - in countries where it was previously considered practically absent. To prevent this, other countries introduce the obligation to vaccinate children, for example Germany [F]. The chance - unfortunately paid for thousands of victims - for a change in trend and reduction of anti-vaccination movements may be the SARS-CoV-2 pandemic currently prevailing in Europe, causing COVID-19 disease. Due to restrictions on the movement of people and a significant reduction of people-to-people contacts in most European countries, statistics of diseases such as measles or rubella will probably fall, however, one should not ignore the threat that results from them, because with a high percentage of complications that they carry. Also these have the potential to paralyze European healthcare when vaccine numbers fall below a critical limit.

When in Western European countries the incidence of infectious diseases fell, an increasing proportion of the population, also under the influence of anti-vaccination movements, did not vaccinate children - in countries where vaccination was not mandatory [G]. Such behavior leads to an increase in the incidence again. Vaccinations are compulsory in Poland or Lithuania, therefore, despite the large migration of people from Eastern Europe, especially from Ukraine with several dozen thousand cases a year, we still do not have massive number of cases [H]. The situation of Ukraine is also difficult because of the ongoing armed conflict and unstable domestic policy - despite the vaccination obligation, the vaccinated percentage is constantly falling.

Fortunately, there is a tendency to legal changes that adjust the recommendations into vaccination obligations, which is undoubtedly beneficial to the entire society - only in this way can one reduce the incidence and even eradication of pathogens. Thanks to universal polio vaccination, the illness is well on its way to completely disappearing from the environment. It was announced in 2019 that polio type 3 virus was eradicated. This means that only one type of wild polio virus occurs in nature - outbreaks are found in Afghanistan and Pakistan, and due to the numerous travels of residents around the world, as long as this virus is not considered removed from the environment, it will be necessary to maintain vaccination further for polio virus that causes poliomyelitis. After the eradication of extremely deadly smallpox virus, it is the eradication of polio virus, first in America and then in Europe in 2001, which served as an example of the importance of international cooperation and social solidarity in the fight against infectious diseases [I].

DENTAL CARE

The teeth condition of Poles, unfortunately, differs significantly from the European average - a higher DMFT (Decayed, Missing, Filled Teeth) among EU (European Union) countries is observed only in Croatia. In western countries it is several times lower. Despite the growing public financial support for dental care, the vast majority of patients use private practices, which translates into smaller and smaller outlays for this sector. Despite the high costs of treatment, only a third of cases are conducted using public dental care [J]. It results from queues and little social trust for free dental visits. Patients value time and quality highly, appearance is more and more important - Poles are increasingly using orthodontists and are increasingly less likely to use tooth extraction as a treatment method. They are not interested in filling cavities with an amalgam seal, and prefer composite filling, which NFZ does not refund. Pains located in the mouth strongly affect everyday life, hamper eating, speaking or sleeping, hence the patient wants to receive help immediately and to the fullest extent. Frequent fear of visiting a dentist can be reduced by believing that when you pay enough for the service, they will be less painful [2]. The United States, as a country based on private financing of health care and high costs of treatment, is struggling with the plague of dental diseases and periodontal disease of its citizens [K]. Those who, due to their financial aspect, are deprived of access to dental care benefit from free treatment campaigns financed by various foundations. The equipment is transported by trucks and temporary offices are grouped e.g. at stadiums. The interest in this type of service is huge because as many as 130 million Americans do not have

dental insurance. The quality of teeth in the US is closely related to the amount of household income [L]. In the most affluent, almost half of the people have a very good condition of teeth and mouth, while in the poorest it is only 15%. As many as 60% of Americans would like to go to the dentist regularly, but they do not because of the associated cost. Every fourth US citizen with the lowest income level does not have a single tooth. The high cost of treatment and therefore the very low access of the less well-off is another example of the extreme in American healthcare, note the dentist is one of the best-paid professions, with an average annual income of 200-250 thousand dollars [M].

EUTHANASIA

The exact definition of the term "euthanasia" is not specified. It depends on the country and legal situation, religious context and social approach to the topic of death. This term etymologically means "good death", which means that depending on the religion, material situation and many other factors it can have completely different meaning for different people. Increasingly, for the society of Western Europe, this term means not only the right to choose the date and method of death, but also the right to receive help from third parties in this procedure. In Central Europe, euthanasia is usually illegal and seen as a negative phenomenon, which may be caused by a higher percentage of practicing Christians [3].

Over the years, the attitude of the world's largest religions has remained unchanged relative to euthanasia, and to this day only Hinduism justifies a kind of acceleration of human death, but it clearly separates it from suicide both in the spiritual context and the actual conduct of the procedure. Other religions, such as Islam, Judaism or Christianity, clearly prohibit the deliberate shortening of human life, regardless of the reasons and means of carrying out such a procedure [4]. In recent years, all these religions have formulated an opinion that allows the abandonment of futile therapy and do not treat this type of decision as a sin. However, it is forbidden to give agents accelerating death or to use other ways to shorten human life, even in terminally ill and suffering persons. Equally condemned is providing such type of resources to patients or giving advice enabling suicide, then called assisted suicide.

In the legal context, euthanasia in Poland is illegal, and the doctor is not allowed to give funds or apply procedures accelerating the death of the patient, even at his explicit and documented request. Despite this, in the Criminal Code such a crime is not treated as homicide and is punishable by imprisonment in the amount of three months to 5 years imprisonment, and in special cases, the so-called compassionate killing. In such cases, the court may remarkably

reduce the penalty or completely withdraw it. Although the Act lacks specific requirements regarding the degree of illness that a patient must meet, in practice, for such an act to be classified as euthanasia murder, the patient must be terminally ill [N].

Society is divided about attitudes towards euthanasia, and due to the high percentage of people of Catholic religion, no changes in legislation are currently planned, explaining this by the risk of abuse and the lack of explicit social acceptance for legalizing euthanasia.

Uncertainty in society can be also explained by the lack of a homogeneous definition of euthanasia - public debates do not always emphasize that one of the requirements is the patient's conscious will to shorten his life.

While in the case of people suffering from diseases that inevitably lead to death or permanent disability with a significant limitation of the possibility of independent functioning, social moods are distributed similarly, where the issue strongly dividing society is the requests for the death of partially handicapped, mentally ill or completely healthy people, as well as matters regarding children.

For these reasons, if it is physically possible, there are trips to countries where the law is less restrictive, e.g. to Switzerland where, despite the fact that euthanasia is illegal, there are legal institutions which, through the assisted suicide procedure, help patients to die in the right for them moment and conditions [O]. Patients can receive a vial of the fatal medication, usually barbituric acid derivatives, after appropriate procedures. Despite the procedures to prevent abuse, there are situations in which healthy people are committing assisted suicide, utilizing forged medical documentation which proves a serious illness.

ABORTION

The 7 January 1993 act on family planning, protection of the human fetus and conditions for the admissibility of termination of pregnancy grants every human being the inherent right to life. It imposes on a number of institutions the obligation to provide care for both fetus and pregnant woman. Material, legal and medical assistance is considered as care throughout the entire pregnancy and perinatal period. People expecting children are also provided with information about the possibility of obtaining further benefits for the poorest, the possibility of renouncing parental rights and access to psychological help in difficult times. Schools are also obliged to help pregnant women in learning to complete their education, for example by setting new exam dates if the reason for not being able to take the exam at the original date was pregnancy, childbirth or the puerperium [5].

From the moment of conception, the Act, giving legal status to the unborn child, a number of rights, including the most important right to life, allows only medical examinations and procedures that are to protect the life and health of it and its mother. In the case of a significant family genetic load or reasonable suspicion, prenatal tests that do not significantly increase the risk of miscarriage are allowed, provided that it is possible to cure or improve the child's health before and after delivery. Such tests can also be used to verify suspected severe fetal damage.

The June 6, 1997 Act of the Penal Code imposes penalties on persons who make abortions, incline to abortion, or lead to abortion [6].

Premature termination of pregnancy as a result of external intervention is treated as a crime and punishable by imprisonment of up to three years, but the pregnant woman is not punished.

Premature termination of pregnancy using pharmacological or surgical methods can be made by a doctor of a public health care institution only in exceptional situations that have been clearly defined by the Legislator. Such situations include severe, irreversible damage to the fetus; pregnancy as a result of a criminal act or when the pregnancy presents a risk of loss of life or serious loss of health to the mother. In all cases, unless there is a need for immediate medical intervention to save a woman's health or life, it is necessary to document the situation by the decisions of two other doctors or by a prosecutor's certificate. This procedure can be performed up to 12 weeks of pregnancy, but this limit does not apply in the event of a woman's life being in danger [5].

The Act also contains a provision on the obligation to provide citizens with free access to resources and information enabling an informed decision on procreation, which further emphasizes the fact that the Legislator does not see premature termination of pregnancy as a form of contraception. Medical practice aimed at saving the health or life of an unborn child or pregnant mother which may result in bodily injury or loss of health of the conceived child was also excluded from criminal liability. Under Polish law, it is unacceptable to terminate a pregnancy for other reasons or at the mother's request [5].

According to official data, in 2018, 1076 abortions were performed in Poland (an increase of 19 compared to the previous year). The vast majority (1050) was caused by severe fetal damage. The number of legal abortions has been at a similar level for several years, but there is a lack of reliable data on abortion performed illegally, often using primitive tools, and so-called abortion tourism, i.e. trips to neighboring countries where there are clinics dealing with abortion for a fee [P].

Poland is the only country in this part of Europe where abortion based on wish of the mother is not available. Similar regulations apply in Brazil, in the Faroe Islands (husband's

consent is also required there) and in some states of Mexico. In most African countries, the rules are stricter or similar to Polish.

Views on the right to abortion are the subject of disputes and each person has the right to make their own decisions. It is not decidable which countries have a more appropriate abortion policy, however, the list of countries with similar regulations regarding these matters shows that the same country can be comparable in one aspect to the best-developed health care systems, and in others to systems that do not enjoy such status. Regardless of the discussion about which approach is the right one, one can see in which direction most countries are heading, including those on which the Polish health system is modeled. It appears that the Polish system is its own creation, not based on another, but adapting to social trends prevailing for years and constantly trying to adapt. This is confirmed by the situation from recent years, when it was not possible to introduce in the country a law changing the right to abortion as a result of mass protests [R].

CONCLUSIONS

Comparing the legal status and the way it is enforced, the first conclusion is that the Polish health care system, although it does not enjoy a good reputation inside the country, is definitely not the worst one. In the face of a relatively short history of a free Polish state and significantly limited financial resources, access to well-managed treatment should be assessed high. The healthcare system in Poland is unique in many respects. Some of the health care activities must be paid for by the patient. As mentioned, many of the regulations strongly reflect cultural influences in our region, which do not necessarily coincide with global trends in legislation. Unfortunately, many of the applicable legal regulations may limit the right to make decisions consistent with one's conscience and belief, the position in the currently ongoing social discussion on topics such as euthanasia or the right to abortion.

Health protection regulations established in Poland are not modeled on neighboring or richer countries, but adapted to the current political and economic situation of the country and the social requirement. Patients have a legal right to information about their health and treatment. Certainly, discussions on the right to abortion or euthanasia and how to distribute funds will continue in the coming years – this is in order to reduce unnecessary spend on extensive bureaucracy. However, none of the healthcare systems, as shown in the richest countries in the world, provide the perfect solution yet. However, efforts should undoubtedly be made to adapt the law to the economic and social situation and also patient education as

soon as possible, which will undoubtedly improve the quality of health services. The health care system in Poland differs in many sectors from neighboring countries, but without a broad social discussion it cannot be assessed whether it is better for a Polish citizen or worse. No other country healthcare system should be used as a model, however use of known, confirmed solutions should be encouraged.

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MEDICAL PROBLEMS OF PALLIATIVE CARE



Treatment of patients with breast cancer – a present-day perspective

Adam Jasiura¹, Izabela Dera¹, Karolina Dupla¹, Mateusz Gorzel¹, Paulina Sztucka², Joanna Zmonarska¹

1. Wroclaw Medical University
2. Faculty of Biological Sciences, University of Wroclaw

INTRODUCTION

Breast cancer (BC) is one of the most common malignant tumors among the inhabitants of highly civilized countries. It accounts for 23% of all cases of malignant neoplasms in women, resulting in 14% mortality. Every year 1.5 million women are diagnosed with breast cancer worldwide, and about 400 000 die from it. Therefore, it is a subject which modern medicine has to challenge today in order to treat patients more effectively [1].

In Poland, breast cancer is responsible for 22% of all cancers in women. According to the National Cancer Registry, over 18,500 cases were recorded in 2017. It is estimated that in the next 3 years this number will exceed 21 000, and the incidence will approach a comparable level observed in Europe [2].

With the molecular diagnostics, through evaluating the expression of markers such as ER, PR, HER-2 and Ki-67, breast cancer can be divided into 5 biological subtypes, including: luminal A, luminal B, basal-like, HER2+ and seminormal (normal breast like). In addition, 12-17% of patients with breast cancer are diagnosed with triple-negative breast cancer (TNBC). Histologically, TNBC is a cancer with poorly differentiated cells, lacking the expression of estrogen and progesterone receptors as well as HER2, therefore the prognosis is usually worse than in the other subtypes [3].

The diagnosis and further treatment of breast cancer are proceeded by multi-specialist teams. They adjust the therapy to the patient's individual predispositions and contraindications. The choice of the treatment method is based on both clinical and pathomorphological assessment including histological subtype of the neoplasm, the stage of the primary tumor as well as the presence of metastases [4]. In this paper, we delineate the possible treatment directions for patients with breast cancer, focusing on those currently used, but also presenting

some promising techniques which can be used in the future. These include surgical methods, radiation therapy, and systemic treatment. Bearing in mind that the treatment of breast cancer is a complex therapy, we present the indications for combining different therapies in order to maximize therapeutic effects, according to the respected guidelines.

SURGICAL TREATMENT

Surgical operation is one of the main methods of breast cancer treatment. It can be separated into two types: breast conserving treatment (BCT) and radical resection [5].

BCT is focused on saving as much of a gland tissue as possible and on limiting intervention in its structure. This allows for a future reconstructed organ to be similar to the breast before treatment. Due to the pathological state constraint, this procedure is indicated only for a certain group of patients. Breast conserving treatment is dedicated to patients with tumor whose diameter does not exceed 3 cm, local metastases that reach only the I-II level of ipsilateral axillary lymph nodes and when any distant metastases have not been found [6]. Additional restrictions include multifocal cancer, mutation in BRCA gene, numerous microcalcifications in mammography, relapse after previous treatment and lack of a technical possibility to dissect the tumor with further reconstruction of the breast [5]. Radiotherapy is an integral part of BCT procedure, therefore if radiation therapy cannot be performed, another type of procedure must be recommended to the patient. Pregnancy is a relative contraindication - undergoing therapy is possible in the second and third trimester, since the planned irradiation takes place after the labor [7,8].

Conserving treatment consists of tylectomy, in which tumor dissection is performed with a 2 cm margin of tissue, lumpectomy - 1 cm margin, and tumorectomy - when only macroscopically changed parts are dissected. If pectoralis muscles cross the border of incision, their fascia must also be removed during surgery [9]. However, one of the newest reports prove that a wider margin in this procedure does not ensure higher effectiveness of the treatment. Present studies concentrate on creating guidelines that will enable precise margin estimation, thus saving greater volume of a gland and increasing treatment effectiveness [10].

Small, impalpable tumors are marked during biopsy before surgery. Metal markers or clips with additional material that fills the tumor bed are typically used for this purpose. Bovine collagen, polylactic acid, or hydrogel are among the most commonly used filling materials because they expand and stabilize the tissue. Marking is recommended especially when biopsy must be taken from several places. Clips are seen in ultrasonography, mammography, and MRI.

Observation of the tumor after a biopsy is conducted to monitor its changes during neoadjuvant treatment. A successful reduction in the size of neoplasm allows a patient to undergo BCT instead of mastectomy. Additionally, clips indicate to the surgeon a place of the incision [11,12].

It is recommended to use four clips on the margins of the sample and one in the center to mark the boundaries of the dissected tissue. The assessment of the excision is made by using the same imaging techniques as used before the operation. Standard methods include USG and mammography, due to their accessibility, ease of use and low costs of the scans. In case of a positive result residual tissue is removed and examined similarly to previously dissected tissue. Intraoperative histopathological examination on frozen samples is not a part of a standard practice - it is characterized by lower sensitivity than testing on paraffin blocks and it extends the time of surgery [5,13,14].

Radical resection is chosen, when BCT cannot be performed. The conventional method is a simple amputation, which is based on the complete removal of the entire breast tissue with pectoralis major muscle fascia and excessive skin. Therapy is dedicated to patients with ductal carcinoma in situ (DCIS) and invasive breast carcinoma without metastasis in axillary lymph nodes. The procedure is also a part of the palliative treatment of locally invasive neoplasm. One of its modifications is a subcutaneous mastectomy that allows sparing of most of the skin around the gland. Saved tissue will be used to cover the implant during further reconstruction. This method is mainly proposed to patients diagnosed with DCIS without the possibility of BCT and as a prophylactic measure to patients with a family history of breast cancer or BRCA gene mutation [13].

Extended amputation, instead of a gland only, additionally includes axillary lymphatic nodes levels I-III. When a tumor infiltrates major pectoralis muscle a total or partial resection of muscle is a necessity. Nowadays resections are mostly based on a modified radical mastectomy by Madden method, which involves resection of major pectoralis muscle fascia and is recommended to patients who do not qualify for any of the above-mentioned surgeries. Another method is modified radical mastectomy by Patey. It differs from Madden's idea of the dissection of minor pectoralis muscle, resulting in better access to lymphatic vessels. Today it is performed only in case of a neoplasm infiltration of the drainage pathway between minor and major pectoralis muscle named Rotter's lymph nodes [13].

For women diagnosed with breast cancer in the first trimester of pregnancy surgical treatment is the only method that can be proposed at this point. Metastasis in lymphatic vessels suggests choosing the modified radical mastectomy by Madden. Unaffected lymph tract allows

for a simple mastectomy with biopsy of sentinel lymph node (SLNB) using isotope technique. During the examination, small doses of radiation are generated, but it presents no risk to the fetus. In the second and third trimester of pregnancy, patients can undergo any surgical operation like women before the gestation period [8].

Successful treatment depends on accurate examination of axillary lymphatic nodes. It is performed as an inter-operational biopsy of sentinel lymph node marked by the isotope. Tests undergo patients with clinically negative node at the time of diagnosis confirmed in USG or fine needle biopsy. At least four hours before the surgery technetium isotope Tc99m on a support of albumin is injected subcutaneously near the nipple. The substance is accumulated along the draining lymph nodes and is detected by a gamma probe. The found sentinel lymph node is cut out and scanned. Absence of metastasis or presence of micrometastases in histopathology allows withdrawal from further axillary surgery. Lymphadenectomy is not a necessity among patients with tumor stadium T1-T2, 1-2 positive sentinel lymph nodes, planned BCT with whole breast radiotherapy and without applied chemotherapy before the procedure. When all of the above-mentioned conditions cannot be met or sentinel lymph node localization fails, lymphadenectomy levels I-II is performed. Clinically negative nodes and positive biopsy results can be resolved by postoperative radiotherapy instead of the node dissection.

Minimum three positive nodes in clinical exam or imaging must be tested by fine or core needle biopsy. It also involves neoplasm stage $\geq T2$ or $\geq N1$ with planned preoperative systemic therapy and tumors T2-T4, N1-N3, M0. If the result of biopsy is negative, the sentinel lymph node is examined during operation and follows the procedure described in the paragraph above. On the other hand, detection of metastases requires lymphadenectomy levels I-II or SLNB in certain cases [13,15-17].

The most common complication of breast cancer resection with lymphadenectomy is lymph flow disturbance. At an early stage in the operating area pseudocyst filled with lymphatic fluid is formed. It must be drained or regularly incised to evacuate excessive fluid, otherwise it could significantly disturb radiation treatment. Later complications include lymphatic edema of the ipsilateral upper limb. In its worst exacerbation swelling causes severe pain and constricts the movement of the joints, also increasing the risk of the overlapping infection or additional stasis in venous circulation. The persistence of this condition leads to rise of angiosarcoma defined as Stewart-Treves syndrome. The disease is rare and affects less than 0,5% of women after breast amputation [18]. Surgery also has an impact on the nervous system. Impairment of sensation in axillary area and inner side of the shoulder, resulting from transection of cutaneous nerves, is an expected side effect. Defect improves gradually, but sense never fully regenerates.

Sensation impairments also include hyperesthesia, paresthesia, neuropathy or phantom pain [19]. The dissection of the gland runs the risk of the surrounding vascular or nerve bundle transection, which results in limited upper limb mobility in pectoral girdle and muscle atrophy, which can lead to postural defects in later life. Damage of long thoracic nerve inactivates serratus anterior muscle which is manifested as winged scapula. Other complications like fibrosis with contracture and hypertrophy of the scar can also occur [5,19-21].

Changes to how their body looks after the surgery are hard to accept for women. Mastectomy significantly affects psychological domain - commonly leading to the despondence and lack of self-acceptance. Psychological aspect is one of the main indications for reconstruction surgery and patients should be informed about such possibilities before the surgery [5,19,22].

Nowadays, methods of breast cancer surgical treatment give satisfying final therapeutic effect also ensuring a reduced interference in organ structure. After surgery, patients must be consulted by a multidisciplinary team in order to establish a form of adjuvant treatment [13].

RADIOTHERAPY

Radiotherapy (RT) is a constantly developing method of breast cancer treatment. It uses the energy of ionizing radiation to destroy cancerous cells. Currently, radiotherapy is one of the basic elements of multidisciplinary breast cancer treatment. According to the 2004-2010 data it is instituted in 60% of patients affected by this disease. All stages of cancer can be cured with RT. In the case of early-stage tumors Tis, T1-T2, N0, or N1, it is used as a part of breast-conserving treatment (BCT). Patients with the advanced neoplastic process (tumors IIIa, IIIb) benefit from adjuvant radiotherapy after mastectomy, because it significantly reduces the number of local recurrences. What is more, RT, due to its analgesic properties, contributes to the reduction of symptoms and improvement of life quality in the case of patients with metastases to bones or central nervous system [24,25].

Radiotherapy became an indispensable part of breast cancer sparing therapy, which is now being chosen by more and more patients. This kind of treatment results in a high esthetic, as well as the therapeutic effect. An increased appliance is noticed due to the possibility of preserving mammary gland. This option is available for women in the early stage of disease - if the diameter of primary lesion is less than 3 cm and nodes are negative [26]. In the case of bigger tumors - up to 5 cm - it is possible to combine a few methods of treatment and start with inductive chemotherapy. If a patient's response to the therapy is satisfying and a diminishment

of lesion is observed, further sparing treatment is possible - tumorectomy followed by radiotherapy [27].

The indications for adjuvant radiotherapy after mastectomy include: T4 tumors (infiltrating chest wall or skin), tumors in patients after non-radical surgical treatment, the occurrence of metastases in 4 and more axillary lymphatic nodes and T3 tumors – with >5 cm in diameter. Postoperative RT can also be performed if the occurrence of metastases was observed in 1 to 3 axillary lymphatic nodes and there are additional locoregional recurrence risk factors such as: cancerous invasion of lymphatic vessels, third degree of neoplasm's histological malignancy (G3) or embolism of cancerous cells in peritumor blood vessels [4,28]. Moreover, performing RT of a nodal area in all patients with the N+ clinical trait results in decreased risk of tumor recurrence and reduced number of deaths caused by neoplasm. However, this procedure should not be indicated in case of patients who went through lymphadenectomy, since the risk of recurrence after lymphatic node resection is low. Furthermore, nodal area irradiation after lymphadenectomy is more likely to cause a sequela in a form of upper limb edema. The exceptions to this rule are patients with high risk of cancer recurrence, e.g. in case of a massive cancerous infiltration through the capsule of a lymph node or when the occurrence of metastases is observed in more than a half of removed lymphatic nodes [4].

The technique of choice in breast cancer treatment is 3D conformal radiotherapy (3D CRT), which enables to strictly adjust the amount, size, the shape of radiation strips, and their angle of entry to the area of irradiation - that is to an individual patient's anatomy. The homogenous distribution of dose is remotely calculated by a computer system on the basis of a patient's computed tomography (CT). CT is shot in a therapeutic position with a coordinated system marked on a patient's skin, which helps to repeat the same position on the therapeutic machine during every exposition. The area of irradiation enfolds the whole treated breast or, in case of mastectomy, the chest wall with a scar after the surgery. If the occurrence of metastases in lymphatic nodes was stated, RT is also performed to the ipsilateral axillary, supraclavicular, and retrosternal lymphatic nodes [24,29].

In adjuvant radiotherapy the effective dose of radiation is 50 Gy in 25 fractions - 2 Gy during every exposition in 5 expositions a week, which gives in total 5 weeks of treatment. In some cases, there is a possibility to shorten the time of RT to 3 weeks by using hypofractionated radiotherapy. It consists in heightening the dose for a singular fraction to 2,66 Gy which involves decreasing the total dose to 40,2 Gy (15 fractions during 3 weeks' time) [24].

Radiotherapy has many advantages, nevertheless it is also related with some negative side effects. Heart and lungs are the organs mainly exposed to ionizing radiation due to their proximity to mammary gland. Disease entities following RT in them include inflammation and fibrosis of pericardium, diffuse fibrosis of cardiac muscle, coronary vessels disease, fibrosis and inflammation of lung [30,31]. In 2000 Early Breast Cancer Trialists' Collaborative Group (EBCTCG) presented the results of their research of the radiotherapy role in reduction of locoregional recurrences and mortality caused by breast cancer. It showed a 4,8%-drop of cancer-induced mortality with concurrent 4,1%-growth of mortality caused by other reasons - mostly by cardiovascular complications [32].

Mentioned research became a prime mover in searching and developing modern techniques of radiotherapy in order to minimize its toxicity. These methods include intensity-modulated radiation therapy (IMRT), deep inspiration breath hold (DIBH) irradiation method and stereotactic radiotherapy used in the treatment of metastases in the brain and liver [24,33].

IMTR is a dynamic technique which enables to modulate the radiation strip intensity, thereby in the same time different doses of radiation are applied to the certain areas. Introducing this method permits administration of a homogeneous dose in the irregular area of treatment or otherwise - to provide a higher dosage of radiation in the region that is more threatened with cancerous recurrence – which is called simultaneous BOOST (SIB). Moreover, those techniques significantly reduce doses of radiation that penetrate to critical structures (heart and lung) and therefore decrease the toxicity of the treatment [24,34].

Other techniques of radiotherapy which contributed to the lower risk of cardiovascular complications are accelerated partial breast irradiation (APBI) and irradiation of larger breasts in the prone position [24]. Proton Beam Therapy (BPT) and tomotherapy (TOMO) are considered to be an alternative for 3D CRT that also decrease the amount of radiation reaching critical organs [35].

Another field where radiotherapy can be used is palliative treatment. Its main aim is to inhibit growth of invasive neoplasm and therefore to improve patient's life quality. The radiation dose is usually lower than those used in postoperative or radical radiotherapy. In this case, alleviation of symptoms is achieved due to anti-inflammatory, anti-swelling, vascular and cytotoxic effects of radiation. Palliative radiotherapy is desirable for patients with non-operative tumor of mammary gland and with a locoregional recurrence of neoplastic process [36].

Radiotherapy is a method of treatment that will be constantly developing. New techniques of irradiation, more precise dosage and determining optimal treatment duration can lead to decrease in the number of complications in the future, especially those concerning

cardiovascular system, and to more and more effective eradication of cancer. Undoubtedly, progressions in development of other breast cancer treatment methods, often combined with RT, will contribute to enhancement of positive effects arising from radiotherapy [24].

SYSTEMIC TREATMENT

Besides surgical methods and radiotherapy, in breast cancer treatment systemic therapy is crucial as well. It consists of diverse methods, which can be divided into chemotherapy, hormonotherapy (HT), and immunotherapy. Such a treatment can be applied either before the surgery as induction therapy or neoadjuvant regimen, or post-surgically as adjuvant therapy. The administration of the treatment requires histopathological and molecular diagnostics to evaluate the expression of estrogen receptor (ER), progesterone receptor (PR), human epidermal growth factor receptor 2 (HER2), and Ki-67 antigen; from the core-needle or open surgical biopsy. The qualification for the treatment should be conducted by a multidisciplinary team, which considers various approaches to the therapy and chooses the most favorable and beneficial regimen for the patient. Many factors are taken into account, such as the risk of the recurrence based on the prognostic assays, cancer sensitivity to the treatment, advantages and contraindications as well as potential side effects of the proposed therapy eventuated from the patient's both mental and physical condition [4].

CHEMOTHERAPY

Chemotherapy is one of the systemic treatment methods, in which cytostatic drugs are administered, often mixed in various regimens in order to reduce the growth of rapidly dividing cells – including cancer cells. Chemotherapy essentially can be divided into adjuvant and neoadjuvant chemotherapy [37].

Adjuvant therapy

Adjuvant therapy is a complementary treatment provided after radical surgery. It is a part of the combined therapy, which is presently standard cancer care. Studies show that an institution of adjuvant therapy decreased the case fatality rate (CFR) by 30% in comparison to the nonuse of such a method. However, its effectiveness declines with the time elapsed from the surgery [38].

According to the breast cancer treatment guidelines, choosing a therapy regimen should be based on the biological subtype of the cancer. Adjuvant chemotherapy is primarily

recommended in triple-negative (TNBC) and non-luminal HER-2 breast cancer, which additionally can be treated with targeted therapy. In case of luminal B cancer chemotherapy employment should be based on the many predictive factors. Whereas, in luminal A cancer chemotherapy is rarely applied, only for patients with multiple positive lymph nodes since the hormonotherapy is the treatment of choice [39].

Drugs used in chemotherapy regimens

In chemotherapy multi-drug regimens are used, which consist of various cytostatics (table 1). They are administered in the maximal tolerated dose (MTD), therefore they act rapidly however simultaneously not leading to the unacceptable side effects. Drugs are provided in 4-8 cycles, every one usually lasts 3 weeks and the whole therapy takes ordinarily 3-6 months. During the breaks between the consecutive cycles, the patient's organism has time to recover from the side effects induced by the treatment, however cancer tissue regenerates too resulting in tumor neovascularization [40].

Table 1. Regimens used in adjuvant chemotherapy, sorted by generations [42,53]

I generation	CMF	Cyclophosphamide, methotrexate, 5-fluorouracyl
	AC	Doxorubicin, cyclophosphamide
	FEC50	5-fluorouracyl, epirubicin (50 mg/m ²), cyclophosphamide
II generation	FAC	5-fluorouracyl, doxorubicin, cyclophosphamide
	FEC100	5-fluorouracyl, epirubicin (100 mg/m ²), cyclophosphamide
	TC	Docetaxel, cyclophosphamide
	AC → Pq3w	Doxorubicin, cyclophosphamide → paclitaxel every 3 weeks
III generation	TAC	Docetaxel, doxorubicin, cyclophosphamide
	AC → Pq1w	Doxorubicin, cyclophosphamide → paclitaxel every week
	AC → T	Doxorubicin, cyclophosphamide → docetaxel
	FEC100 → T	5-fluorouracyl, epirubicin (100 mg/m ²), cyclophosphamide → docetaxel

In the 70s, CMF regimen was the first one to be used in the therapy of breast cancer after the radical surgery. It is based on three drugs: cyclophosphamide, methotrexate and 5-fluorouracil. Cyclophosphamide is an alkylator, which forms chemical bonds with functional groups of the DNA-building compounds, therefore resulting in DNA damage and further cell death. Whereas methotrexate (an analog of folic acid) and 5-fluorouracil (a pyrimidine analog) are antimetabolites, which derange DNA synthesis through interfering pathways of proper metabolites by competitive inhibition.

Currently, a standard chemotherapy comprises anthracyclines, including doxorubicin, epirubicin, daunorubicin, idarubicin and mitoxantrone. They act by insertion into DNA where they interfere with its synthesis and repair, disrupt topoisomerase I and II, as well as generate reactive oxygen species; thereby through these mechanisms they lead to the programmed cell death. However, these drugs have many cardiotoxic side effects, which can limit their usefulness, therefore it is important to monitor ejection fraction (EF) of heart. Especially for the older patients a taxanes-based regimen is worth considering as a less toxic therapy. Nevertheless, most adjuvant therapies are based on the anthracyclines regimens. They can be provided in double-agent regimens with cyclophosphamide – AC, or triple-agent with an extra addition of 5-fluorouracil: FAC or FEC [41,42].

Studies show no significant difference in therapeutic effectiveness between CMF and AC or FAC regimens. However, they differ in adverse effects as in CMF therapy patients reported mostly myelosuppression, hair loss, gastrointestinal distress and weight gain, whereas anthracyclines-based regimens led mainly to vomiting and alopecia [43,44]. Though, FEC regimen manifested increased effectiveness compared to CMF – with overall survival (OS) 77% vs 70% [45].

Taxanes are another group of cytostatics, on which the therapy regimens are mainly based on. The most common clinically used taxanes are paclitaxel and docetaxel. These drugs disrupt the microtubule function by stabilizing them, hence they inhibit the mitosis. As the most frequently reported side effect is an anaphylactic reaction, a premedication with corticosteroids and antihistamine drugs is required. Taxanes can be administered in the double-agent regimens with cyclophosphamide (TC) or trastuzumab (TH) as well as with both of them in the triple-agent regimen (TCH). Studies indicate increased therapeutic effectiveness of TC regimen in comparison to AC (OS after 7 years: 87% vs 75%) [46]. Whereas THC compared to the sequential AC → TH regimen has fewer side effects and is better tolerated, despite comparable efficacy [47].

The most aggressive adjuvant therapies consist of regimens based on anthracyclines and taxanes. Studies show that for patients provided with AC regimen (4 cycles) it turned beneficial to add extra 4 cycles of paclitaxel (AC → P) [48]. Moreover, further studies indicate increased 5-year survival rate when paclitaxel was administered weekly in comparison to application every 3 weeks. However, such a regimen resulted in enhanced neurotoxicity in tested patients [49]. Similar benefits were discovered when to the 3-cycle FEC regimen additional 3 cycles of docetaxel were provided (FEC → T). Such a treatment relevantly increased disease-free survival (DFS) and OS as well as it decreased the risk of cariological adverse effects [50].

Furthermore, studies show relevantly increased survivability of regimens combining anthracyclines, taxanes and cyclophosphamide (TAC) in comparison to FAC, especially in patients with HER2+ cancer, before the menopause or with metastases in 1-3 regional lymph nodes. However, such a therapy resulted in increased toxicity and significant hematologic side effects [51]. Further studies were conducted to compare TAC with the sequential regimen of 4 AC cycles followed by 4 cycles of docetaxel (AC → T). Results show, these therapies were equally effective, they differed in toxicity profile though [52].

Currently, guidelines recommend regimens based on anthracyclines and taxanes in multi-drug sequences. However, they should not be combined simultaneously, like in the TAC regimen, due to their high toxicity and many adverse effects. Further, it is inadvisable to institute FAC or FEC regimens if surgery is considered. Moreover, guidelines recommend not to use platinum-based drugs in the post-surgical adjuvant treatment of patients with BRCA mutation [4].

Dose dense therapy

Presently, dose-dense therapy is becoming more and more popular. It was developed by shortening the periods between cycles of cytostatics, whereas the dosing remains the same. Therefore, increased destruction of cancer cells is possible, resulting in a higher effectiveness of high-risk breast cancer (HRBC) treatment. However, such a therapy simultaneously impacts cytostatically bone marrow cells, thereby it could lead to iatrogenic neutropenia. To decrease the myelosuppression-related side effects, an additional G-CSF protective treatment was introduced, using filgrastim or pegfilgrastim, which reduced the time of neutropenia and associated infections and mortality. Metanalyses confirm dose dense therapy increased both OS and DFS in patients diagnosed with HRBC in comparison to conventional adjuvant therapy. It decreased the risk of death by 40% as well in comparison to nonuse of adjuvant therapy at all, whereas a conventional therapy decreased that risk by 30%. Furthermore, studies show dose

dense therapy prolongs the time to recurrence in comparison to conventional therapy. According to researches the risk of side effects was not relevantly increased compared to conventional therapy. However, adverse effects of necessary G-CSF therapy should be regarded, such as pulmonary toxicity. Nevertheless, adjuvant chemotherapy with dose dense regimens is a preferred treatment of patients with HRBC [4,38,54,55].

Metronomic therapy

An alternative method of chemotherapy dosage are metronomic regimens. This therapy, still under clinical trials, provides cytostatics with shortened breaks between the cycles but also in reduced dosages. Drugs are administered every day or in short intervals with small doses, opposite to conventional therapy, where they are close to the maximal tolerated dose (MTD). Metronomic therapy acts multilevel – it stimulates the immune system, inducing the maturation of its cells; enhances antigen presentation; increases Tc lymphocytes cytotoxicity as well as decreases suppressor activity of Treg cells. Furthermore, it inhibits angiogenesis, preventing the regeneration of blood vessels necessary to tumor growth, which is allowed in breaks between the conventional treatment cycles. According to studies, metronomic therapy is non-toxic, well tolerated and cheap. It should be considered for older patients, if receiving a quick response to treatment is not crucial as well as in palliative therapy. It can be also a promising later line treatment in luminal subtypes of breast cancer, which acquired hormone resistance. Currently, studies over metronomic therapy of breast cancer are conducted with cyclophosphamide + capecitabine regimen as well as with vinorelbine [40,56].

Neoadjuvant therapy

Neoadjuvant treatment differs from adjuvant therapy as it is instituted before the surgical treatment. It can be provided to patients, who primarily do not qualify for a surgery – it is called inductive treatment then. Another group of patients are the ones with early stage of breast cancer, who potentially can undergo the surgical treatment, but neoadjuvant therapy relevantly improve the prognosis and increase total effectiveness of the therapy through DFS, OS and pathologic complete response (pCR) [37]. A primary recommendation for neoadjuvant treatment is stage 2 and 3 triple-negative (TNBC) and HER2+ breast cancer [39].

There are many benefits of neoadjuvant treatment. Due to the pre-surgical chemotherapy, breast conserving therapy may become possible instead of radical mastectomy. Moreover, neoadjuvant therapy allows monitoring the changes of neoplastic tissue and its surroundings with the evaluation through biological markers after the institution of the therapy;

which cannot be done in adjuvant treatment as the tumor is removed. However, neoadjuvant therapy has some possible disadvantages as well, such as increased risk of local recurrence, complications in later radiotherapy and surgical treatment, or tumor acquisition of the resistance to cytostatics. Furthermore, prognostic microscopic evaluation of primary tumor and its metastases in lymph nodes cannot be provided. Therefore, a proper qualification for a treatment is important, as well as the care of the patient by a multidisciplinary team to possibly minimize the adverse effects of the therapy. Nevertheless, advantages are greater and neoadjuvant therapy relevantly decreased the risk of death in locally advanced breast cancer [57]. However, recent metanalysis show that both OS and DFS are equal in neoadjuvant and post-operative adjuvant therapy [58].

Regimens used in neoadjuvant therapy

According to National Comprehensive Cancer Network (NCCN) recommendations, there are many adjuvant chemotherapy regimens possible – all should be considered, and the therapy needs to be individually selected for each patient. Guidelines propose in case of HER2-subtype to initiate a dose dense therapy based on AC regimen followed by paclitaxel (PCL) or TC regimen. Treatment of HER2+ subtype should be provided with AC regimen followed by trastuzumab with prospective addition of pertuzumab or with TCH regimen [17]. European Society for Medical Oncology (ESMO) guidelines are less strict and allow teams to choose the treatment more liberally – they recommend basing the therapy on anthracyclines and taxanes as well as TC or CMF regimens [59]. Moreover, guidelines recommend in patients with BRCA1/2 mutation to consider platinum-based neoadjuvant chemotherapy regimens [4].

HORMONOTHERAPY

Cancer hormonotherapy (HT) is a therapeutic method depending on suppressing the effect of hormones on cancer cells or inhibiting the process of their endogenous synthesis. Breast cancer is one of the most common types of cancer which is sensitive to this type of therapy. The development and proliferation of hormone-dependent cancer cells are directly dependent on the action of certain hormones - in the case of breast cancer, these include estrogens and gestagens. By suppressing their influence on cancer cells, these hormones are assumed to inhibit the tumor growth and, consequently, slow down the progression of the disease.

Before hormone therapy is administered, the potential susceptibility of cancer to this type of treatment needs to be investigated. The standard diagnostic procedure is an immunohistochemical examination of biopsy material to detect the presence of ER and/or PR on the cell surface using labeled antibodies [60]. Tumor cells expressing estrogen and/or progesterone receptors (referred to as ER-positive) are potentially susceptible to therapy and account for approximately 80% of all diagnosed breast cancer cases yet in women <45 years old only about 50% of cases are ER-positive tumors [61]. Nonetheless, it should be noted that the presence of receptors does not determine a positive response to the treatment. It has been shown that with the accumulation of mutations within the ER gene in cancer cells during disease progression, the receptor loses its function and ceases to exhibit metabolic activity despite proper estrogen fusion [62]. This leads to the independence of the tumor development from hormones despite the presence of an estrogen receptor. The exact mechanisms of resistance and prediction of the response to hormonotherapy remain unknown.

Therefore, hormonal treatment is recommended as first-line therapy for the treatment of disseminated forms of hormone-sensitive cancer. Medications used in hormonal therapy can be classified into several groups with different mechanisms of action and therapeutic effectiveness. The main groups of drugs and selected representatives are listed in the table 2.

Table 2. Selected pharmacologic agents used in mammary gland cancer hormonotherapy

SERMs	<ul style="list-style-type: none"> • Tamoxifen • Toremifen
Aromatase inhibitors (AI)	III generation non-steroid AI: <ul style="list-style-type: none"> • Anastrozole • Letrozole • Atemestane Steroidal AI: <ul style="list-style-type: none"> • Exemestane • Fulvestrant
Gonadoliberin analogs (GnRH)	<ul style="list-style-type: none"> • Goserelin
Progestogens	<ul style="list-style-type: none"> • Megestrol acetate • Medroxyprogesterone acetate
Obsolete forms of therapy	<ul style="list-style-type: none"> • High doses of estrogens • Androgens

Hormonal treatment is used in cases of local cancer - as an adjuvant therapy after tumor removal surgery. However, it is instituted much more often in the treatment of generalized disease (node-positive BC), to slow down the progression and improve the effectiveness of other treatment methods - mainly chemotherapy or palliative therapy. The great advantage of hormonal therapy is its low toxicity and the associated low severity of side effects compared to conventional chemotherapy. Moreover, hormone therapy provides longer survival for patients with disseminated disease compared to monotherapy with cytostatics [63]. The disadvantages of this type of therapy include the relatively long treatment response time and the need for its long-term use – for 5-10 years, according to ESMO guidelines [59].

Selective estrogen receptor modulators

Selective estrogen receptor modulators - SERMs (so-called "anti-estrogens"), whose tamoxifen is the most commonly used representative, are pharmacological agents that have different effects on the estrogen receptor (ER) depending on the tissue. Tamoxifen is a non-steroidal compound whose main mechanism of action is blocking estrogen binding to receptors in cancer cells, thereby inhibiting their proliferation. At the same time, tamoxifen has agonist properties in relation to the estrogen receptor of bone tissue and other visceral organs - thereby it has a slight estrogenic effect: it inhibits osteoporosis and lowers plasma cholesterol. Furthermore, the drug increases NK (natural killers) cell activity and the production of transforming growth factor beta (TGF- β), which inhibits cell proliferation [64]. The most commonly reported adverse reactions are facial flushing, vaginal discharge, rarely thromboembolism and hypercalcemia. The most dangerous complication of tamoxifen therapy is the increased risk of developing endometrial cancer. Therefore, every patient treated with tamoxifen is advised to have a gynecological examination at least once a year. Obtaining a positive response to tamoxifen therapy depends on the presence of ER and/or PR markers in cancer cells. Positive tumors reach a 50-60% response rate, nonetheless, the absence of these receptors lowers the chance to about 10%. The average duration of remission is 9-12 months [65]. Other SERMs, such as toremifene or droloxifene, do not show an advantage in safety or efficacy over tamoxifen.

Aromatase inhibitors (AI)

Drugs from this group are the treatment of choice for women with hormone-dependent breast cancer in postmenopausal age. Aromatase is an enzyme that converts androstenedione to estrone. In the postmenopausal period, this process is inhibited in the ovaries and occurs only

in peripheral tissues - adipose tissue, skin, muscles, liver, breast stromal cells, and in the tumor tissue itself. The drug blockage of the enzyme inhibits the endogenous estrogen synthesis.

Aromatase inhibitors in terms of chemical structure and mechanism of action can be divided into two main groups - steroid, which are derivatives of androstenedione, including formestane, exemestane and atemestane, and non-steroidal imidazole derivatives - letrozole or anastrozole. Steroid compounds irreversibly connect to the active center of aromatase. Nonsteroidal agents cause selective aromatase inhibition by blocking cytochrome P-450 reductase. Third-generation nonsteroidal AIs are characterized by high selectivity, therefore the synthesis of other steroids in the adrenal glands remains unaffected, resulting in the significant improvement in their safety profile compared to older generation drugs [66]. Compared with tamoxifen, AIs are less likely to cause menopausal symptoms and thromboembolic complications, while osteoporosis is observed more often. On that account, calcium and vitamin D3 preparations should be administered during the treatment with AIs. Metanalyses of numerous clinical studies have shown that compared to tamoxifen AI are more effective in slowing down the course of advanced disease in 18-30% of cases. However, they do not significantly increase overall survival [67].

Gonadoliberin analogs

Gonadoliberin (GnRH), also called luteinizing hormone-releasing hormone (LH-RH) is a peptide hormone produced in the hypothalamus. Its pulsatile secretion stimulates the pituitary gland cells to synthesize and release luteinizing hormone (LH) and follicle-stimulating hormone (FSH), the main regulators of ovarian functions. The only synthetic gonadoliberin analog registered in breast cancer therapy is goserelin. Its long-term stable supply interferes with the natural, pulsating nature of stimulating the pituitary gland by endogenous GnRH. Therefore, it desensitizes and stops the secretion of LH and FSH, resulting in the inhibition of estrogen and progesterone synthesis in the ovaries. This phenomenon is called "pharmacological castration". This method replaced other, historical methods, such as surgical removal of the ovaries or their thermal ablation. In addition to the significantly lower invasiveness of the pharmacological technique, its greatest advantage is the potential reversibility of ovarian suppression. Due to the principle of action of gonadoliberin analogs, this treatment can only be used for premenopausal women with preserved ovarian function. The main side effects include menopausal symptoms - hot flushes, emotional lability, weight gain, decrease in bone density. The effectiveness of gonadoliberin analogs in monotherapy is lower than other types of hormonal therapy - remission is observed in 20–40% of patients for an average period of 10–14 months. Therefore,

it is usually used in combination with tamoxifen, which increases the chance of a positive response to treatment [68].

Progestogens

Progestogens, which are progesterone receptor (PR) agonists, are rarely used in the treatment of breast cancer and are reserved as last chance therapy in the absence of a sufficient response to other hormone therapy methods. Their mechanism of action has not been yet fully understood.

Progestogens have been shown to inhibit several enzymes, mainly sulfatase and 17 β -hydroxysteroid dehydrogenase, which transform endogenous estrogens into their high-activity forms within the tumor tissue. Furthermore, progestogens activate the enzyme sulfotransferase, which is responsible for the inactivation of estrogens into their inactive sulfates. The effectiveness of progestogens is controversial with significant discrepancies in clinical trial results. Lack of evidence for their effectiveness and burdening with dangerous side effects, including thromboembolic complications (with the risk of life-threatening pulmonary embolism), edema, intermenstrual bleeding and weight gain, assure the absence of progestin therapy in most of current therapeutic recommendations [69].

HER2-TARGETING THERAPY

Apart from chemotherapy and hormone therapy, systemic treatment also comprises immunotherapy with adjuvant HER-2 therapy. To this group belongs trastuzumab (H), which is a humanized monoclonal antibody directed against the human epidermal growth factor receptor 2 (HER2). For 15-20% of breast cancer patients whose tumors overexpress HER2, trastuzumab treatment is important in both early and advanced stages of disease [70]. Studies show that supplementing standard chemotherapy with trastuzumab extends metastatic disease survival and reduces the risk of recurrence in the early stages by approximately 50% [71,72]. Its use implies however low or moderate risk of cardiotoxicity, which is usually manifested by asymptomatic reduction of left ventricular ejection fraction and less frequently by clinical heart failure [73].

Following the introduction of trastuzumab, three other HER2-targeted agents have been developed: pertuzumab, T-DM1 and lapatinib.

Pertuzumab (P) is a monoclonal antibody that binds subdomain II of the HER2 extracellular domain and prevents the formation of HER-2/HER-3 heterodimers [74]. Studies

have shown that the addition of pertuzumab to trastuzumab and docetaxel (THP regimen) in first-line therapy for metastatic disease prolongs survival by 7.7 months, compared with the TH regimen [75]. According to the Phase III CLEOPATRA trials on HER2+ node-positive patients, compared to the TH regimen, THP resulted in improved overall response rate (ORR, 80% vs 69%), improved progression-free survival (median 19 vs 12 months), and improved overall survival (OS; median, 56.5 vs. 40.8 months) [76]. In contrast, neoadjuvant treatment with HP added to the docetaxel and carboplatin (TCHP) gives the highest pCR (66.2%) reported to date [77].

Another drug, ado-trastuzumab emtansine (T-DM1), consists of trastuzumab conjugated to a chemotherapeutic emtansine moiety. It improves progression-free survival (PFS) as well as OS compared to lapatinib and capecitabine in HER2+ breast cancer patients who have progressed on a taxanes and trastuzumab [78].

Although data on pertuzumab and T-DM1 are limited, available evidence support the view that these drugs may be less cardiotoxic than trastuzumab. Trastuzumab itself has intrinsic immunity-modulating activity capable of both mediating ADCC (antibody dependent cell cytotoxicity) [79] and supporting HER-2 specific T-cell response [80].

T-DM1 may further enhance immune priming by modulating dendritic cell activity [81]. The data show the benefits of T-DM1 usage in combination with trastuzumab in women with HER2+ type who have progressed after previous trastuzumab-based therapy, as well as in first-line treatment in combination with trastuzumab and docetaxel.

Another developed substance is lapatinib. It is a small molecule dual tyrosine kinase inhibitor of EGFR (ErbB1, epidermal growth factor receptor) and HER2 (human epidermal growth factor 2 receptor) [82].

Early clinical studies suggest its activity in women with advanced breast cancer. In addition, studies show that this drug, unlike trastuzumab, can penetrate the central nervous system. What is more, it may be effective in the treatment of brain metastases in combination with capecitabine - an objective response in the CNS was obtained in 20% of patients, maintaining a stable state or response to therapy in extracranial locations [83].

HER2 therapy in combination with hormone therapy is an alternative option for patients whose disease does not progress rapidly or symptomatically, or does not have significant visceral involvement (i.e. multiorgan metastasis). For these patients, hormonal and HER2 treatment may offer a less toxic first-line approach compared to typical HER2 treatment in combination with chemotherapy [84,85].

SYSTEMIC THERAPY REGIMENS

In systemic therapy, all of the available methods of treatment should be taken into consideration, including chemotherapy, hormonotherapy (HT), and HER2-targeting immunotherapy depending on the cancer subtype, which is determined with the molecular and histological markers. The result of the biopsy is crucial when choosing the appropriate treatment as every subtype requires a different therapy regimen to match individually to the patient depending on their predispositions and contraindications.

In case of luminal A breast cancer (ER+, HER2-) the treatment of choice is hormonotherapy. European Society for Medical Oncology (ESMO) guidelines advise the usage of aromatase inhibitors (AI), tamoxifen or fulvestrant, with potential addition of CDK 4/6 inhibitors. Moreover, National Comprehensive Cancer Network (NCCN) recommendations suggest in case of a tumor size >0.5 cm and negative lymph nodes to consider a 21-gene RT-PCR assay in order to evaluate the risk of the recurrence. If the risk is increased, chemotherapy should be added to hormonotherapy. Further, if more than 4 lymph nodes are positive, there is an indication for chemotherapy followed by hormonotherapy. For the patients resistant to HT or in the visceral crisis a sequential single-agent chemotherapy is advised, using anthracyclines, taxanes or – if the patient has been previously treated with the anthracyclines/taxanes regimens – capecitabine, eribulin or vinorelbine [4,17,59].

The systemic treatment of luminal B HER2- (ER+) cancer is similar to the luminal A subtype. Administration of chemotherapy and its regimen depends on the number and the response of hormonal receptors as well as the risk of recurrence and individual indications [4].

According to ESMO guidelines, luminal B HER2+ (ER+) subtype should be treated using chemotherapy with HER-2 dual-blockage (trastuzumab + pertuzumab). If chemotherapy is contraindicated due to the patient's condition, a HT with dual-blockage (trastuzumab + pertuzumab or trastuzumab + lapatinib) is suggested instead of chemotherapy. If the treatment achieves the intended results and the progression is not noticeable, it should be continued as a maintenance therapy using HT + HER-2 dual-blockage. However, if the progression is detectable further treatment is required. If possible, T-DM1 should be administered. In case of no response to the therapy, HT + trastuzumab + lapatinib can be used if such a regimen has not been previously applied, or trastuzumab in combination with previously unused chemotherapy and hormonotherapy drugs as a later line treatment. Furthermore, NCCN recommendations allow the treatment to begin with HT in case of tumors >1 cm and node-negative or with micrometastases pN1mi, with further addition of chemotherapy [4,17,59].

In non-luminal HER2+ (ER-) subtype ESMO guidelines suggest regimens with chemotherapy and HER-2 dual-blockage (trastuzumab + pertuzumab). If chemotherapy is not possible to administer, there is indicated a treatment with trastuzumab or HER-2 dual-blockage, where pertuzumab or lapatinib is added to trastuzumab. Further therapy depends on the remission achievement. In case of no progression, an anti-HER-2 maintenance treatment should be applied. However, if the progression is noticeable T-DM1 is advised if available. Then, later lines therapy consists of trastuzumab with addition of a previously unused chemotherapy drug or trastuzumab + lapatinib (dual-blockage) [4,17,59].

Triple-negative breast cancer (TNBC) is associated with a worse prognosis. In compliance with ESMO guidelines, sequential single-agent chemotherapy should be applied. If the patient with stage 2 or 3 cancer has not been treated with anthracyclines/taxanes regimens before – such treatment is indicated. Moreover, recommendations after St. Gallen Consensus Conference 2019 suggest in case of stage 1 TNBC to use alkylators and taxanes omitting anthracyclines. However, if anthracyclines/taxanes regimens have been previously administered without achieving the intended outcome, the decision about further treatment should be based on the BRCA genes analysis. In BRCA mutation poly-ADP-ribose polymerase inhibitors (PARPi) or carboplatin are advised. Whereas, in BRCA wild-type beside carboplatin, there can be used capecitabine, eribulin, and vinorelbine as well [4,17,39,59].

IMMUNOTHERAPY

Cancer immunotherapy is a treatment method that involves interfering with the dormant immune system to increase or modify defense mechanisms against developing cancer. Immunotherapy can be divided into passive and active, each can be specific or non-specific [86].

Vaccines used in the treatment of breast cancer are intended to increase the population of lymphocytes specific for tumor's antigens. Initially, they contained antigens common with healthy tissues and tumor in which their expression was significantly increased. To avoid self-aggression, the selection of lymphocytes in the thymus resulted in the removal of high avidity clones, leaving the lymphocyte population with lower affinity. Unfortunately, this response is weaker and has no clinical benefit. Vaccines containing antigens specific for mutations present in the tumor are an alternative - more commonly in tumors with a higher degree of mutation, since they are seen as foreign). Produced antibodies have greater avidity and more effectively destroy tumor cells [87].

Many clinical studies have tested breast cancer vaccines in combination with chemotherapy to reduce the suppressive effects of Treg lymphocytes [88,89]. Others have tested peptide and whole-cell vaccines that provide HER-2 along with trastuzumab – which can increase immune priming, enhance effector CD8⁺ T cells activity and promote immune memory [90,91]; or with trastuzumab and a low dose of cyclophosphamide combination – to suppress the Treg lymphocytes, obtaining increased immunity [92]. Another combination of vaccines against breast cancer with CTLA-4 (cytotoxic T cell antigen 4) or PD-1 (programmed death receptor-1)/PD-L1 (programmed death ligand-1 antagonists) stays in the area of high interest, as it abrogates signaling that shuts vaccine-induced T cells down in the tumor site.

Immune checkpoint antagonists, specific for CTLA-4, PD-1 and PD-L1, have revolutionized cancer therapy, inducing a durable response, which translates into increased overall survival (OS) in many types of cancer [93,94].

CTLA-4 (cytotoxic T cell antigen 4) is a protein that appears on the surface of T lymphocytes activated in contact with an antigen and inhibits the further lymphocyte response. CTLA-4 antagonists, humanized monoclonal antibodies - tremelimumab and ipilimumab, have been tested in several breast cancer studies in which evidence of their immunomodulatory activity was obtained. Increasing doses of tremelimumab with exemestane have been studied in 26 patients with ER⁺ HER2⁺ breast cancer [95]. A significant increase in ICOS⁺/FoxP3⁺CD4⁺ T cell ratio was observed in most patients. In another breast cancer study, a single dose of neoadjuvant ipilimumab was given as monotherapy or with cryoablation to 12 patients with early breast cancer before mastectomy [96]. Combined immunotherapy induced circulating Th1 lymphocyte cytokines, ICOS⁺Ki67⁺CD4⁺/CD8⁺ T cells, and an increased ratio of CD8⁺/FoxP3⁺ T cells in the tumor.

Collected data also suggest that humanized monoclonal antibodies that target PD-L1 (avelumab and atezolizumab) or PD-1 receptor (pembrolizumab) may induce sustained clinical response in some patients with metastatic TNBC and probably also have significant clinical activity in ER⁺ HER2⁺ types of breast cancer. The PD-1 receptor binds ligands, PD-L1 and PD-L2, which, by interacting with it on cancer and immune cells, induce signaling counteracting T-cell activation during the effector phase of the immune response [96]. For avelumab, the response was highest for TNBC [97]. It is noteworthy that the ORR for 52 patients with metastatic PD-L1+TNBC, for whom pembrolizumab was the first-line treatment, was higher than for patients overall and amounted to 23.1%. This is also in line with atezolizumab data, where the responses in metastatic TNBC appear to be highest when given in the first line and/or to selected patients with PD-L1 [98]. Early data from the I-SPY study

revealed that the addition of pembrolizumab to neoadjuvant paclitaxel gives an estimated pCR at 46% vs 16% in HER-2 patients, 60% vs 20% in patients with TNBC and 34% vs 13% in patients with ER+ HER2+ cancer [99].

CONCLUSION

Years of experience in breast cancer treatment have shown the greatest results are achieved in combining several therapies, instead of using monotherapy. Therefore, combined therapy is considered to be the most efficient way to fight breast cancer. Future studies should focus on finding new therapy combinations, which would show even greater synergy, thereby improving survivability scores, simultaneously decreasing adverse effects rates in patients.

The development of the BC therapies not only resulted in an increase of the treatment effectivity but also in the improvement of patients' life quality – both psychically and socially. For many women, especially younger, breast-conserving therapy is an important part when choosing the treatment. Some of the described in this paper methods can be used in palliative therapy as well.

All of the systemic treatment methods – chemotherapy, hormonotherapy, and immunotherapy – have been developed in the XX century. If such a progress rate of breast cancer therapy is going to continue, it is possible for new methods to eventuate in the near future, which would even better suit patients' needs. Constantly improving diagnostic and therapeutic methods as well as multi-specialists teams care are going to further decrease patients' mortality and significantly improve their life, possibly even curing cancer eventually.

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Role of Epstein-Barr virus in the pathogenesis of lymphoma

Magdalena Paziewska¹, Patrycja Gierszon², Agata Stachura³

1. Department of Experimental Hematooncology, Medical University of Lublin
2. Department of Applied Psychology, Department of Psychology, Medical University of Lublin
3. Chair and Department of Epidemiology and Clinical Research Methodology, Medical University of Lublin

INTRODUCTION

Infectious agents are associated with approximately two million new cases of cancer per year [1].

Three mechanisms of infection-related malignant transformation have been demonstrated [2]:

- Direct malignant transformation through infection with the oncogenic virus: Epstein-Barr virus (EBV) - Hodgkin lymphoma, Burkitt lymphoma, diffuse large B cell lymphoma, post-transplant lymphoproliferative disorders, T/NK cell lymphoma, and HIV-related non-Hodgkin lymphoma, Human herpesvirus 8 (HHV8) - primary effusion lymphoma, plasmablastic lymphoma, Human T-lymphotropic virus 1 (HTLV-1) - adult T leukemia/lymphoma, Kaposi sarcoma-associated herpesvirus (KSHV) - primary effusion lymphoma, plasmablastic lymphoma
- Indirect mechanism of malignant transformation as a consequence of chronic antigenic stimulation leading to selection and clonal expansion of B cells. This model include infection with *Helicobacter pylori* - gastric MALT (mucosa-associated lymphoid tissue) lymphoma, *Chlamydia psittaci* - ocular adnexal lymphoma, *Campylobacter jejuni* - immunoproliferative small intestinal disease, *Borrelia burgdorferi* - primary cutaneous B-cell lymphoma, Hepatitis C virus (HCV) – marginal zone lymphoma (in particular splenic type), diffuse large B cell lymphoma
- Mechanism of malignant transformation associated with immunosuppression through depletion of T helper lymphocytes as a result of infection with Human

immunodeficiency virus (HIV) or as a consequence of an immunosuppressive treatment - Hodgkin lymphoma (with EBV), Burkitt lymphoma, diffuse large B cell lymphoma, extranodal marginal zone lymphoma, T-cell lymphoma

Oncogenic viruses play a causative role in 12-15% of human cancers. The first discovered human oncogenic virus is EBV that causes approximately 1% of human cancers [3].

EBV is double-stranded DNA, an enveloped virus belonging to the human herpesvirus family. EBV has a tropism for B cells and epithelial cells. Almost everyone is exposed to EBV infection at some point in their lives. Infection with EBV is most often asymptomatic. However, it cannot be eradicated and a person once infected for the whole life has B lymphocytes containing EBV latent form. Therefore, EBV is widespread in all populations and chronic latent infection occurs in more than 90% of the world's population [4].

The life cycle of EBV is subdivided into two phases: lytic and latent. An important component in EBV infection is epithelial M cells that transport molecules and antigens to the lymphoid tissue. CD21 on B lymphocytes is the receptor for EBV glycoproteins involved in the fusion of the virus to the host cell: gp350/220, gp85, gp42, gp25 and gp110 [5].

In the lytic phase, EBV assumes a linear form. After viral replication progeny virions are released and infect other B lymphocytes [6].

In the latent cycle, the virus exists in B cells and replicates with the genetic material of the host without expressing any viral protein to avoid immune response [6].

Three models of gene expression were identified. EBV nuclear antigen 1 (EBNA-1) and EBV-encoded small RNAs (EBER) expression is associated with latency type I [7].

Latent membrane protein 1 (LMP-1) and 2 (LMP-2), EBNA-1 and EBER are characteristic of latency type II. Latency type III is associated with LMP-1, LMP-2 and EBNA: EBNA-1, EBNA-2, EBNA-3A, EBNA-3B, EBNA-3C, EBNA-LP [7].

The role of EBV latent protein has been shown in Table 1. In addition to latent proteins, an important role is played by microRNAs transcribed from *BART* and *BHRF1* which target both viral and cellular mRNAs. EBV microRNAs interfere with processes related to oncogenesis including cell proliferation, apoptosis and escape from immunosurveillance. Moreover, EBV microRNAs may regulate microenvironment through exosomal transport between cells [8].

The latent gene expression type (0, I, II, III) is characteristic for types of infected cells and types of disease (Table 2).

Table 1. Role of latent proteins [9-11]

Latent protein	Role
EBNA-1	immortalization of B cells; transcription activator; decrease of p53 activation and apoptosis response to DNA damage;
EBNA-2	immortalization of B cells; <i>c-Myc</i> activation; induces cyclin D2 expression in resting B cells leading to G1/S transition promotion; transcription activator – mimic activated Notch receptor downregulation of BIK expression (proapoptotic Bcl-2 family protein) and upregulation of BFL-1/A1 (prosurvival Bcl-2 family protein) expression
EBNA-3A	regulation of cell survival; interaction with RBPJ proteins; decrease of Bim (proapoptotic Bcl-2 family protein) expression; inhibition of CDKN2 transcription; downregulation of <i>c-Myc</i> transcription
EBNA-3B	interaction with RBPJ proteins; upregulation of CXCL10
EBNA-3C	interaction with RBPJ proteins; modulation of Rb, p53 and p27 activity; decrease of Bim (proapoptotic Bcl-2 family protein) expression; induce CDKN2, aurora kinase B and chemokines; downregulation of <i>c-Myc</i> transcription; promotion of G1/S transition;
EBNA-LP	transformation of B cells; <i>c-Myc</i> activation; induce cyclin D2 expression in resting B cells leading to G1/S transition promotion (in co-operation with EBNA-2)
LMP-1	promotion of B cell immortalization; mimic CD40 signalling; activation of NF- κ B, JAK/STAT, PI3K/Akt, JNK, p38 MAPK, ERK signalling pathways; promotion of cell cycle progression by regulation CDK2 and Rb; regulation of telomerase activity
LMP-2A	mimic BCR signalling; interaction with PI-3K/Akt and ITAM/Syk signalling pathways; Myc upregulation (promotion of B cells G1/S transition); p27 degradation (promotion of B cells proliferation)
LMP-2B	regulation of LMP-2A activity
EBERs	transcription regulator; promotion G1/S transition by activation of the Pi-3K/Akt signalling pathway and inhibition of the tumour suppressor PKR and the cell cycle inhibitors p21 and p27

Abbreviations:

Akt - protein kinase B, BCR – B cell receptor, BIK - Bcl-2-interacting killer, Bcl-2 - apoptosis regulator family, CDK2 - cyclin-dependent kinase 2, CDKN2 - cyclin-dependent kinase inhibitor 2, CXCL10 - CXC chemokine, IFN- γ inducible protein-10, EBER – EBV-encoded small RNAs, EBNA - EBV nuclear antigen, Erk – extracellular signal-regulated kinase, ITAM - immunoreceptor tyrosine-based activation motif, JAK - Janus kinase, JNK - c-Jun N-terminal kinase, LMP - latent membrane protein, LP- leader protein, MAPK - mitogen-activated protein kinase, NF- κ B - nuclear factor κ B, PI3K – phosphatidylinositol 3-kinase, PKR - protein kinase RNA-dependent, Rb - retinoblastoma protein, RBPJ – recombination signal-binding immunoglobulin kJ region, STAT - signal transducers and activators of transcription, Syk - spleen tyrosine kinase

Table 2. EBV latency type

EBV latency type	Latent EBV protein expression	Types of cells infected/disease
0	EBERs	Memory B cells
I	EBNA-1, EBERs	Burkitt lymphoma, Plasmablastic lymphoma, Primary effusion lymphoma
II	EBNA-1, LMP-1, LMP-2A, LMP-2B, EBERs	Hodgkin lymphoma, Diffuse large B-cell lymphoma, NK/T-cell lymphoma
III	EBNA-1, EBNA-2, EBNA-3A, EBNA-3B, EBNA-3C, EBNA-LP, LMP-1, LMP-2A, LMP-2B, EBERs	Post-transplant lymphoproliferative disorders, Diffuse large B-cell lymphoma

Abbreviations:

EBER - EBV-encoded small RNAs, EBNA - EBV nuclear antigen, LMP - latent membrane protein

EBV is the aetiological agent of infectious mononucleosis. Furthermore, EBV has been associated with a wide range of malignancies including Hodgkin lymphoma, Burkitt lymphoma, diffuse large B cell lymphoma, post-transplant lymphoproliferative disorders, T/NK cell lymphoma and HIV-related non-Hodgkin lymphoma [11].

HODGKIN LYMPHOMA

According to the World Health Organization (WHO) classification, Hodgkin lymphoma (HL) can be divided into classical HL (cHL) and nodular lymphocyte-predominant HL (NLPHL). The association of EBV infection with lymphomagenesis has been shown only for cHL. The histological subtypes of the cHL based on the morphologic characteristics of the Hodgkin Reed-Sternberg (HRS) tumour cells (CD30+, CD40+, CD15+, IRF4/MUM1+) that represent only 1-2% of the tumour mass and the composition of the tumour microenvironment are nodular sclerosis (NS) cHL, mixed cellularity (MC) cHL, lymphocyte-rich (LR) cHL and lymphocyte depletion (LD) cHL. EBV is mainly associated with MC HL and LD HL [12]. The proportion of EBV-positive cHL varies also with age (a high incidence in children under 10 years and adults over 80 years), gender (a male predominance), socioeconomic status, and geographic region (a high incidence in underdeveloped countries) [13].

HRS cells are characterized by constitutive activation of canonical and noncanonical nuclear factor kappa B (NF-κB) signalling as a result of expression of different TNF receptors, including CD40 and RANK as well as genomic alterations such as mutations/deletions in *TNFAIP3/A20* and overexpression of *BCL3*. NF-κB pathway contributes to the survival of HL cells. Moreover, enhanced activation of JAK/STAT and phosphatidylinositide-3-kinase (PI3-K) signaling pathway been observed. BV-positive HRS cells express the II latency viral type.

LMP1 can activate the NF- κ B, JAK/STAT and PI3K/AKT signaling pathways and modify cellular transcription [14]. LMP-1 is also an important agent for immunosuppressive microenvironment due to the influence on the production of the immunosuppressive cytokine, such as IL-6, IL-8, and IL-10 [12]. Tumour microenvironment may support HRS cell growth and survival as well as help the tumour cells escape from immune surveillance. Moreover, LMP1 may induce expression of the collagen receptor – DDR1 (discoidin domain receptor 1) in B cells protecting them from apoptosis. LMP2 may provide a survival signal by activating RAS/PI3K/AKT pathway. Moreover, suppression of B cell lineage gene expression by LMP2 has been described. LMP-2 has also probably BCR-independent functions which are important in the pathogenesis of HL. EBNA1 might contribute to the growth and survival of B cells by inhibition of p53-mediated apoptosis and immune evasion of EBV-infected HRS cells due to upregulation of chemokine CCL20 expression of the in HRS cells that promote the migration of regulatory T cells [14].

Interestingly, EBV-positive HL is associated with human leukocyte antigen (HLA) polymorphisms. An overrepresentation of HLA-A*01 and an underrepresentation of HLA-A*02 was observed in patients with EBV-positive HL. Therefore, antigen presentation and the immune system function play a significant role in cHL pathogenesis [15].

BURKITT LYMPHOMA

Burkitt lymphoma (BL) is a highly aggressive non-Hodgkin B cell lymphoma characterised by monomorphic medium-sized cells expressed CD19, CD20, CD79a, CD10 and Bcl-6. The tumour cells are negative for CD5 and CD23. Macrophages containing apoptotic tumour cells are described as a “starry sky” pattern. BL is the fastest growing human tumour. Cell doubling time is 24-48 hours [16].

Endemic Burkitt lymphoma (eBL), sporadic Burkitt lymphoma (sBL) and HIV-associated Burkitt lymphoma are three forms of BL based on geographical distribution. All three BL are characterized by chromosomal translocation juxtaposing the *Myc* gene on chromosome 8 and one of the immunoglobulin (Ig) loci, heavy chain locus on chromosome 14, kappa light chain locus on chromosome 2 or lambda light chain locus on chromosome 22. Bringing the *Myc* gene under the control of an Ig locus leads to high Myc protein expression and uncontrolled proliferation of BL cells. Despite the high level of Myc protein causes a tp53-mediated apoptotic stress response, approximately 30% of BL carry tp53 mutations to counteract this apoptosis. Moreover, BL with tp53 wild type carries other mutations or

epigenetic changes that deregulate the expression of tp53 regulatory proteins. Also, recurrent mutations in genes related to cell cycle have been described. CCND3 mutation is associated with a deregulation of cyclin D3 protein which is one from three D-type cyclins (CCND1, CCND2 and CCND3). Cyclins are cofactors that regulate cell cycle progression by binding to cyclin-dependent kinases (CDKs). CCND3 promotes the G1/S phase transition [17].

Additionally, increased activity of the transcription factor TCF-3 as a consequence of mutation in *TCF3* gene or its negative regulator ID3 (inhibitor of differentiation) in almost 70% of patients with BL has been observed. TCF-3 amplified antigen-independent (tonic) B-cell-receptor (BCR) signaling and thus engage the PI3K/AKT pathway what lead to sustaining BL cells survival. A consequence of cooperation TCF-3 with Myc is high aggressiveness of BL [18].

Almost 100% of eBL, 10-15% of sBL and 40% of HIV-associated BL are EBV-positive. Therefore, EBV is considered as an important agent in BL pathogenesis [19]. It is worth noting that BL is the first human tumour whose pathogenesis has been associated with the virus [12]. BL cells express II EBV latency type. The aetiological role of EBV in BL pathogenesis might be related to increased survival of BL tumour cells carrying *Myc* translocation as a result of inhibition of apoptosis induction by EBNA-1 protein and inhibition of the proapoptotic BIM protein upregulation by the latent transcript LMP-1. Furthermore, the role of BART-microRNAs in B cell growth transformation and maintaining the BL phenotype has been shown [17]. EBV might also contribute to genomic instability and induce DNA damage [16].

A mechanism in which EBV, inducing the activity of the AID (activation-induced cytidine deaminase) enzyme that regulates somatic Ig gene mutation and following Ig class switch recombination, promotes the generation of *c-Myc* translocations has been also proposed [17].

DIFFUSE LARGE B-CELL LYMPHOMA

Diffuse large B-cell lymphoma (DLBCL) is the most common non-Hodgkin lymphoma (NHL) worldwide. Gene expression profiling (GEP) allowed for better understanding of DLBCL heterogeneity and classification of DLBCL cases based on cell origin into two major subtypes: germinal center B cell (GCB) and activated B cell (ABC). Morphologically, the centroblastic, immunoblastic and anaplastic are the most common variants of DLBCL. DLBCL cells express CD19, CD20, CD22 and transcription factors characteristic for B cells (PAX5,

BOB.1, OCT2). DLBCL cells are negative for CD5. Expression of CD10, BCL6, MUM1 is used to determine origin of cells (GCB vs non-GCB) [20,21].

The most common chromosomal abnormality $t(3;q27)(q27;v)$ which involves *Bcl-6* leading to its inactivation. Translocations $t(14;18)(q21;q32)(IGH/BCL2)$ and $t(8;14)(q24;q32)(IGH/MYC)$ also commonly occur in patients with DLBCL [22].

Gene mutations in DLBCL are involved in B-cell receptor signalling and NF- κ B signalling pathway (*MYD88*, *CD79A/B*, *CARD11*), PI3K and JAK-STAT pathway signalling (*PTEN*, *FOXO*, *GNA13*, *SGK1*), apoptosis (*Bcl-2*, *tp53*), histone methylation or acetylation (*EZH2*, *EP300*, *CREBBP*, *KMT2D*), immune-regulation (*TNFRSF14*) [21].

EBV has been detected in approximately 10% of DLBCL patients. However, the prevalence of EBV in patients with DLBCL varies among countries, in Asian and Latin American countries accounts 10-15% whereas in the United States and European countries it is less than 5% [1]. Most EBV-positive DLBCL cases are ABC type [19]. EBV-positive DLBCL was originally observed in patients greater than 50 years old. EBV is characterized by a lifelong latency. As a consequence of age-related immunosenescence, the diversity of B cells decreases. Similarly, functional dysregulations of T cells including decreased cytotoxic activity of NK/T cells, decreased the number of T effector lymphocytes (as a result of development impairment) and decreased T cell receptor (TCR) diversity are observed. Therefore, there are more EBV-specific cells. EBV-positive DLBCL arising in older patients show a latency III gene expression profile. However, EBV-positive DLBCL may occur also in younger immunocompetent patients. Then it is characterized by a latency II gene expression profile [23]. Evaluation of GEP profile signatures shows enhanced activation of the NF- κ B and JAK/STAT signalling pathway in EBV-positive DLBCL compared to EBV-negative DLBCL. The NF- κ B and JAK/STAT signalling pathways play a critical role in many cell process including proliferation, differentiation and survival [24].

PYOTHORAX-ASSOCIATED LYMPHOMA

Pyothorax-associated lymphoma (PAL) is a subset of DLBCL associated with longstanding (more than 20-year) chronic pyothorax or chronic pleuritis as a result of the artificial pneumothorax in the treatment of pulmonary tuberculosis or tuberculous pleuritis [25]. EBV-positive PAL cells express III viral latency type. EBV infection and *tp53* mutations, which were found in almost 70% of PAL, immortalize B cells. These cells are exposed to proinflammatory cytokines such as IL-6 which support their proliferation as well as reactive

oxygen species which induce alterations of DNA damage–response genes (*ATM*, *ATR*) contributing to gene instability [26].

EBV-ASSOCIATED T/NK LYMPHOPROLIFERATIVE DISEASES AND LYMPHOMAS

Although the EBV has mainly tropism to B cells, it may uncommonly infect T or natural killer (NK) cells. According to World Health Organisation (WHO) classification, there are 6 categories of EBV-associated T and NK-cell lymphoproliferative diseases (LPDs): chronic active EBV infection (CAEBV) of T/ NK-cell type, EBV-associated hemophagocytic lymphohistiocytosis (HLH), systemic EBV-positive T-cell lymphoma of childhood, extranodal NK/T-cell lymphoma (nasal type), primary EBV-positive nodal T/ NK-cell lymphoma and aggressive NK-cell leukemia. CAEBV of T/NK-cell, EBV-associated HLH, and systemic EBV-positive T-cell lymphoma of childhood are prevalent in both pediatric and adult patients, while the others EBV-associated T/NK lymphoproliferative diseases occurs usually in adult patients [27].

CAEBV of T/NK-cell type a reactive process of EBV-associated T and NK-cell LPDs that may evolve into T/NK lymphoma [27]. EBV-positive T/NK lymphoma cells express the Latency II pattern of viral genes [28].

LMP-1 plays an important role for lymphoma microenvironment by promoting secretion of proinflammatory cytokines such as TNF- α and IFN- γ and increasing expression of programmed death protein ligand 1 (PD-L1) as a result of regulation of a wide range of signaling pathways including the MAPK/ERK1/2, JAK/STAT, NF- κ B, and PI3K/Akt pathways [28]. Moreover, LMP-1 protects T cells from apoptosis by suppression of TNF-receptor expression and interaction with TRADD (tumor necrosis factor receptor type 1-associated death domain protein) that activate caspase cascade [17]. Expression of EBV proteins – EBNA1 and LMP1 increase genetic instability. The most frequent chromosomal abnormality is 6q21 deletion which is associated with loss of suppressor genes such as *PRDM1*, *FOXO3*, *AIM1*, *ATG5*, and *HACE1* [29].

POST-TRANSPLANT LYMPHOPROLIFERATIVE DISORDER (PTLD)

T lymphocytes through the mechanism of cellular immunity maintain immune surveillance over EBV infection in immunocompetent patients. After hematopoietic stem cells

or solid organ transplantation patients are heavily immunosuppressed. The absence of EBV-specific immune response may lead to escape from immune surveillance and uncontrolled expansion of transformed B cells. 60-80% of PTLD are EBV-positive. EBV-positive cells in PTLD express a latency III pattern [30].

SUMMARY

EBV causing approximately 1% of human cancers is an important aetiological agent of malignancies including haematological disease (Hodgkin lymphoma, Burkitt lymphoma, diffuse large B cell lymphoma, HIV-related non-Hodgkin lymphoma, post-transplant lymphoproliferative disorders, and T/NK lymphoproliferative diseases). Currently available antiviral drugs are effective only against the virus in the lytic state and do not affect the latent form of the virus. Therefore, new targets for virus-associated malignancies are investigated including drugs that reactivate the virus.

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Tanatopedagogy as a holistic approach to the needs of patients

**Mateusz Nowak, Jakub Kasalik, Gabriela Gut, Aleksandra Kurczak,
Katarzyna Kałuża, Magdalena Towarek**

Medical University Piastów Śląskich of Wrocław, Wrocław

INTRODUCTION

Death has always aroused extreme emotions in people. People naturally feel fear of death, regardless of the value system on which human communities were built. Although the process of aging and dying has been an inseparable part of human existence, societies (especially in the current 21st century) would like to delay old age and death for as long as possible. Therefore, disease which may accelerate this death, has become something that scares people a lot. From the dawn of time, diseases have driven people anxious. The evidence of that is the middle-ages personification of death in European art in the form of a grim reaper.

The epidemiology of terminal diseases and the mortality of diseases has varied over time. Simultaneously, the development of science and public awareness have been arising. The following generations faced new, previously unknown diseases and learned how to deal with them. People during their illness often searched for support in religion or culture. Disease has been not only a source of suffering for the patient, but has also caused a change in the worldview and perception of reality. Therefore, development of civilization and progress in science have determined the changes in the view of terminal diseases.

Can we skillfully deal with life with the awareness of death in the 21st century, being aware of all the past events and with the whole knowledge of history? Can we also use the achievements of modern psychology to help people who have heard diagnosis of terminal disease? At first glance, this seems even more demanding than in the past. Contemporary Western societies are characterized by the disappearance of mechanisms that help assimilate death and perceive it as a constructive element of life. In the current societies, behavioral models that cultivate the perception of death as something unacceptable, something that should be hidden and denied are more often observed. Therefore, human death and life itself

are gradually dehumanized. Consequently, it was necessary to implement the educational model that would be able to compensate for this in our culture. Tanatological education is suitable in this field to build human awareness about the responsible experience of death itself - both one's and someone else's.

The science which describes how to deal with death is tanatopedagogy. It supplements modern medicine with the psychological aspect of the patient. Therefore, implementation of its techniques is an opportunity to optimize therapies and meet the needs of patients. Thinking about all needs of patients fits in with the current holistic approach that Western medicine is currently seeking. Tanatopedagogy is both part of tanatology (meaning science about death) and a subdiscipline of pedagogy (which means studying about educational processes) [1]. Due to its multidimensionality, this field of science has various definitions. According to J. Binnebesel, it is "*a science of upbringing with mortality awareness, inscribed in the nature of human being, based on the fundamental principle of respecting the dignity of every human being and the inviolability and a priori value of human life*" [2]. According to P. Grzybowski, Tanatopedagogy is "*a pedagogical sub-discipline dealing with education in the aspect of suffering, illness and dying*" [3]. Nevertheless, tanatopedagogy can be divided into two basic branches: theoretical and practical. Theoretical considerations include concepts about human life and attempts to understand death. Practical tanatopedagogics, on the other hand, aim at education about death, activities such as hospice volunteering and also support for the dying person and their family [2,4].

THEORETICAL TANATOPEDAGOGY. SCIENTIFIC CONCEPT OF DEATH

Over the years, the definition of death has changed many times. Classically, it was considered to be a permanent and irreversible cessation of heart rate and/or breathing [5]. The current definition was developed at an international expert meeting that took place in Montreal in 2012 and it reads as follows: "*Death is permanent loss of consciousness and permanent loss of all brainstem functions. It can be caused by permanent cessation of circulation or critical brain damage. In the context of declaring death, the term "permanent" means a loss of function that cannot return spontaneously and will not be restored by intervention*" [6]. Death can last longer or shorter, but regardless of the time it is a process, which has its chronology and is divided into several stages: clinical, individual and biological death.

Usually, just before death, the phenomenon of agony occurs, in which the body's vital functions weaken. This is a transitional stage, when there is still a chance for complete recovery. At first, the functions of major systems, such as the respiratory, circulatory and central nervous systems are impaired. It is defined as the period of reduced life (*vita reducta*). In the second stage, respiratory and circulatory functions decrease so that they can be very difficult to observe on a routine examination. This is called minimal life (*vita minima*). When this period extends, it resembles real death, which is commonly called lethargy (apparent death).

When the restoration of normal system functions is impossible, the body goes into the interletal period. It is characterized by varying degrees of viability of individual cells and tissues of the body. It's length depends on many factors, including temperature, humidity, causes of death and prior health. Different tissues may still properly respond to stimuli that cause so-called interletal or supravital reactions. They are only a local tissue reaction, not a reflex observed during life, in which the reflex arc of the nervous system is involved.

The interletal period precedes clinical death. Clinical death can be a gradual process (which can be observed, for example, in incurable diseases) or it can progress quickly. In the latter, it is possible to restore life by cardiopulmonary resuscitation. The final phase of clinical death is inhibition of breathing and circulation with loss of consciousness. Just 3-6 minutes after the cessation of circulation and breathing, the activity of the cerebral cortex is turned off. Only the next few minutes separate an organism from total brain death (individual death).

Individual death is equivalent to death according to the law, even when circulation and breathing (spontaneous or artificial) are still maintained. As a result of irreversible damage, mental, regulatory and coordinating functions stop in the brain. After this time, when oxygen supply is inhibited, the body's cells die at different times. When all body functions have ceased, the last stage occurs. The total death of the body, called biological death, is characterized by progressive autolysis. The first process is a tissue breakdown by its enzymes, while the second is by the enzymes of putrefactive bacteria, which were previously symbiotic with the body [7,8].

Tanatopedagogue is responsible for making people aware of the inevitability of ongoing processes, as well as for helping them to get used to the terror and fear. We should be equally aware of the inevitability of processes occurring during dying and that death itself is not a single event, but a process stretched in time that can be divided into individual stages. Nevertheless, not everyone perceives their vision of death in such a matter-of-fact and literal

way. In most religions, death is not the end, but a way to a new stage of life. In monotheistic religions, the physiological process of dying is not synonymous with human death. Both Christians and Jews, as well as Muslims, believe in the existence of a spiritual form of a man, who re-assembles and resurrects after death. On the other hand, dharmic religions, such as Buddhism or Hinduism, assume the transformation of consciousness into another form of life on earth. They call the process a reincarnation. According to those beliefs, death gives a chance for another human life. A thorough understanding of the concept of the dying process in a medical context and the fusion of scientific facts with the patient's beliefs allows better understanding of this phenomenon in a social context. This should translate the tanatopedagogue into a mentor leading the patient throughout the process, reducing his anxiety and accustoming to upcoming events.

WHY DID THE EUROPEAN CULTURE OF THE 21ST CENTURY DEVELOP THE TANATOLOGICAL CURRENT?

In the face of civilisational development, the European culture has started treating the issue of death and illness as a taboo. Instead of being a part of everyone's life, the subject of death is often concealed from children and youngsters in the apprehension of their reaction. Thus, the present generation does not learn how to truly understand those phenomena. The history of Europe has various events, by which demise was perceived as a natural part of the human existence. Currently, the part of tanatopedagogical reflections is an attempt to break the stereotype regarding tanatology recognised as a branch of science only needed by those who are sick, suffering and dying, and their relatives. The subject of tanatopedagogy's interests is not only the biological and medical aspect of death (described in the previous chapter) but also the humanistic viewpoint [10]. Its purpose is not only education related to the process of dying but also teaching how to live with the awareness of the end of life.

Tanatological education was not always needed in human history. In the past, infectious diseases have had the greatest impact on the formation of mortality and functioning of society back in the day as contracting an infectious disease was once inevitable. From the beginning of Western civilization, chroniclers paid the most of their attention to descriptions of wars and epidemics. The very first statements of outbreaks related to antiquity, the Peloponnesian war in Athens, when an unknown disease decimating the city's population appeared on the island's area, its victim was also Pericles [11]. Similarly, Republican Rome

and later the Roman Empire have repeatedly fallen victim to all kinds of epidemics that directly affected both prevailing laws and customs, leaving the remains for future generations. One of the most important ones was a plague during the reign of Marcus Aurelius, described by Galen. According to the preserved chronicles, up to 2,000 people died as a result of this disease. The strict regulations regarding the creation of cemeteries near household facilities were introduced back then, which are still in force today, almost unchanged [11]. However, the most famous epidemic from antiquity was the Plague of Justinian. Historians estimate that 25-33% of the Byzantium's population died because of it, and in Constantinople alone, up to 16,000 people a day have lost their lives [11,12]. Society has turned strongly to religion, believed that God sent the disease as a punishment for sins [12]. After 750, there were no major outbreaks in Europe until the fourteenth century, when the plague known as Black Death has come. The pandemic consumed 30-60% [13] of the current European population. The topos of the "plague doctor," a person wearing a black coat and a mask with a bird's beak, has become established in the collective conscience. Whereas in Art topos of Danse Macabre gained popularity - a pageant of people from all social strata led by a skeleton. It symbolized the equality of all states in the face of imminent death. They depicted it in their paintings, among others, Michael Wolgemut and Guy Marchant [14]. However, the most deadly pandemic in recent human history turned out to be the A/H1N1 pandemic, also known as Spanish flu. The number of cases is estimated to have been circa 500 million, while victims are estimated to have been between 50 and 100 million people. The authorities were unable to restore control of the situation. Restaurants, churches, theatres were closed, and war operations were suspended for some time [14]. The abovementioned unforeseen epidemics and ecological disasters such as volcanic eruptions, earthquakes or tsunamis have always undermined the sense of security and reminded us of the fragility of our existence. They influenced the functioning and development of communities and cultures.

Since the time of the great pandemics and world wars, civilization progress began to increase rapidly. There has been enormous economic growth. Thanks to the discoveries of the 19th and 20th centuries such as antiseptics and antibiotics, life expectancy has increased significantly, and infectious diseases have become less lethal than before. On the other hand, the cases of mass disasters, because of the media hype created in the 21st century, ceased to be events that reminded society for a long time about the vision of the fragility of human life. Suspending such events in collective memory earlier allowed individual reflection on one's life and getting used to the vision of death. The actual flow of information gives people the

opportunity to choose what news they want to listen to. Many people are displacing adverse information because of this, living in a bubble of only positive beliefs. So how is humanity dealing with similar events today? In most situations, society does not process these life lessons as a knowledge, but as a source of unresolved anxiety and fear.

Until the mid-twentieth century, the most common causes of death were acute diseases, i.e. infectious diseases of childhood, tuberculosis, pneumonia and others. With the start of the trend to improve the quality of life for Europeans, death has stopped coming rapidly. The improvement of sanitation, the introduction of vaccinations, antibiotics have led to a significant reduction in mortality from those diseases [15]. Chronic diseases, which weaken the body for years, have become the main problem of the 21st century. They are usually caused by an improper lifestyle, lack of physical activity and progressive pollution of the environment [A,16]. Along with the development of the industry, the incidence of the so-called civilization diseases is rising. Diabetes, hypertension or cancer, in terms of their course, slowly begin to lead to destabilization of the body's homeostasis. In neoplastic diseases that were previously incurable, nowadays, thanks to the development of medicine, the patient does not die overnight, has to adapt to the new situation, learn how to live with the disease. The prospect of growing infirmity, depending on caregivers or death can lead to a mental breakdown in a sick person, which worsens the quality of life and can shorten it significantly. Tanatopedagogy provides tools that allow preventing such situations by educating the patient and their loved ones. The more we talk openly about death, the less unknown it becomes, making it less feared.

LIFE WITH THE AWARENESS OF INEVITABLE DEATH. STAGES OF ACCEPTANCE OF DISEASE AND DEATH

Due to the technological progress and discoveries of new medications, the approach to terminal diseases has changed. Death is more often preceded by periods of treatment that improve the state of functioning, but do not always guarantee the recovery. Therefore, the patient has to deal with the awareness of impending death. According to Eurostat reports, in 2016 the second most common cause of death in European countries was cancer [A,17].

The word "cancer" brings to mind the patient's feelings such as death, helplessness and a sense of hopelessness. In situations of patients finding out that they are given a certain

amount of life, living with the awareness of lethal disease undoubtedly generates above-average stress. This situation requires dealing with the stress in various aspects of life from the patient and his close environment. Such awareness is a chronic stressor. As a result of stress, the organism becomes weaker and makes the patient feel helpless and scared of death, such feeling may accompany him for the rest of his life [16, 18].

The feeling of losing oneself makes people divide their life into parts of before and after diagnosis. It's a very difficult period emotionally. There are five psychological stages that the patient usually passes through after finding out about terminal disease. This psychological concept of internal coping with the diagnosis was first proposed by the American doctor Elisabeth Kübler-Rose in 1969 in the book "On Death and Dying". In this work, the author assumes that the diagnosis of terminal disease is a huge challenge for the patient. Therefore, awareness of impending death does not appear immediately, but gradually evolves. This psychological theory divides time from diagnosis to acceptance into five stages: denial, anger, negotiation, depression and acceptance. At each of the following levels, the patient requires appropriate care from a psychologist or tanathopedist. Basic knowledge in the field of psychology and the ability to differentiate the patient's current mental state allows to introduce adequate responses of the tanatopedagogue, especially in critical situations, when immediate action is required [10].

These stages are assigned not only to the patient but also to his family and people in the closest environment. The stages are as follows:

1. **NEGATION**. Initial reaction is refusal, followed by shock and speechlessness. The patient believes that the diagnosis has an error and displaces reality.
2. **ANGER**. The next stage is anger, which is mainly passed onto family and friends. The patient cannot accept that this situation has affected him.
3. **NEGOTIATIONS**. The third stage pertains to religious people in particular. The patient promises to a higher power, which he believes in, that when he recovers, he will change. Often at this stage there is an internal discourse between the sick and the object in which he sees the center of his religiosity. In exchange for the recovery, he promises God that he will improve as a person e.g. he will stop smoking.
4. **DEPRESSION**. When the patient in the fourth stage realizes that he will not save himself from death, depression and the accompanying sadness come, as well as dislike of society and even self-mutilation.

5. **ACCEPTANCE.** The final stage is acceptance. The patient then agrees with what awaits him, he is overwhelmed with peace, with the desire to recall beautiful moments from his life. He wants to stabilize all matters before death, both financial and emotional.

PATIENT PERSONALITY AS A PARTICULAR DETERMINANT ATTITUDE TOWARDS DEATH

The above mentioned pattern of five accepting stages differs depending on patients' personality, beliefs and life experience. Patients may stop at particular phase, undergo it for longer period of time and reinforce the reaction. For example, they may affirm in anger or depression, while suppressing, delaying accepting attitude towards disease. Acute stress and adjustment disorders developed at the negation stage on the example of patients after diagnosis of acute leukemia [20,21]. Incorrect and inadequate long anxiety response had a significant impact on the course of the somatic disease. It has been proven that adjustment disorders expanded at early phases without correct diagnosis and therapy increased the risk of cardiac death and suicide number. In turn, symptoms like depression or anger were estimated around 10-18% in population of patients with acute leukemia disease [22]. The previous study performed Hosaka et al. [23] on another group of patients have demonstrated, that stopping at stage four (of five accepting stages), concerned even 40% patients without mental illness diagnosed before. This situation was connected with developing symptomatic depression [23]. Treatment duration has impact on patients' symptoms and mental health condition as well. Zittoun et al. reported that depression frequency increases with longevity, from 23% in first day of chemotherapy to 35% in 21st day among patients with acute leukemia [24]. In these cases, implementation by tanatopedagogists self-therapy methods causes better decision-making about treatment and overcoming the fear related with disease. Analysis contained in peer-review articles described higher susceptible among young people (below 33y.o.) on negative psychological effects caused by disease. Younger patients feel helplessness and stress more often what may be associated with depression development. This is probably due to less life experience. This indicates the greater need for further psychological support among young patients during treatment [25].

The study considers other factors which may have positive impact on emotional, mental state like: gender, civil status, social relations, educational level, profession. However,

they don't have significant impact. Outcome of studies shows that types of personalities have impact on mental health condition of adults during acute leukemia remission. The best results were observed in vigorous, persistent, independent patients with "fighting spirit" [26]. According to this, Nelson described that in mixed patient group with various cancers, there was a significant correlation between "fighting spirit" characteristic and mental health well-being [27]. Self-denial, valor and tenacity are very important in acceleration of disease remission. Specialized tanatopedagogue should be aware of these factors to be able to identify which aspects in patient's life may help with the acceptance of diagnosis.

PATIENT AND HIS FAMILY IN PERCENTING THE DEATH MESSAGE; METHODS OF COMBATING WITH INTERNAL FEAR

Cancer is a disease that causes, among many extreme emotions, most often intense anxiety. It accompanies patients at each of the five-step acceptance stages. The expression can be regressed by activating defense mechanisms. The first defense mechanism is always repression resulting in the denial of illness. On the other hand, defensive mechanisms such as blocking or passive-aggressive activities transform fear on the subconscious level into anger, giving vent to their emotions in aggressive behavior. In addition to examining personality strengths and the current defense mechanisms used by the patient, it is also very important to know the source of his fears. In the comparative analysis of patients from the Czech Republic, Poland, Ukraine and Italy, the most frequently indicated components of the so-called tanatic anxiety, fear of death, included fear of pain, abandonment by family and friends, loneliness and a negative vision of the future [28]. The basic way to support a patient is always conversation. In the case of a conversation with a suffering person, it is crucial to develop an atmosphere of mutual trust [4]. The feeling of aura of security and the lack of assessment of the patient's views gives the opportunity to look into his personality and learn about his fears. Expressing problems by the patient and identifying the source of anxiety itself helps to reduce stress and brings relief, and the interlocutor gives the opportunity to provide the best mental support possible [29].

It is worth mentioning that consequences of diagnosis have influence not only on patients but also on the closest family and friends. The close surrounding is not indifferent to patient's feelings. Family is a witness of anger, pain, tears and share these feelings with patients [30]. De Walden- Gołuszko explains that care-givers' reactions are a reflection of

emotional state and frequently, they are more severe [31]. Consequently, tanatopedagogists should not focus on patients only but on their surroundings as well. Community of suffering person is a crucial element in understanding patient's perception of the disease. Tanatopedagogist should constantly pay attention to changing emotions in each person and define ties that bind the closest [4]. The role of family and friends is essential in patient's life. He needs mental support. The presence of the closest helps to fight with fear, relieve tension and develop healthy defense mechanism in response to information about inevitable death. Last stage- acceptance, gives the opportunity to experience this difficult time consciously but without as much sorrow [10].

The tanatopedagogist's support in this situation provides long-term care during dying and also after the death. It's important to remember that as a result of loss, mourning period is challenging, when pathological defense mechanisms may occur in response to sorrow. It can manifest through isolation, hipochondria, obsessive-compulsive disorder development, different addictions. The tanatopedagogist's support is intended to help mourning person with developing a sense of life again and return to previous hobbies, duties [32,B].

COMMON THREADS OF RELIGIOUSNESS AND TANATOPEDAGOGY

From the dawn of time, religious systems have been providing mental support for the terminal patients. For most of humanity's existence, it was not the tanatopedagogists who had the function of reducing the tension associated with death, but it was spirituality. Religion was undoubtedly the factor that significantly facilitated and assimilated patients with the thought of death. Therefore, tanatopedagogy should draw, as much as possible, inspiration from the benefits and advantages of religions. Well-educated tanatopedagogists should be able to identify and use analogous and effective methods in everyday practice with patients. One of the key functions of religion in the face of danger is to give hope for another life after death. This thought reduces tension and fear of death which improves mental well-being and affects on a better perception of one's health [33].

Another benefit of religion is also the feeling of belonging to a larger group, and the associated social acceptance. People living in religious minorities report, as it turns out, more often than others, feelings of depression. This is due to a higher percentage of discrimination in the local community. This phenomenon is independent of the challenge [34].

Tanatopedopaedists therefore use group therapies, involving patients in active participation in the life of the group, to arouse a positive sense of belonging in patients.

PATIENT'S RELIGIOSITY AS A USEFUL TOOL FOR TANATHOPEDAGOGISTS

The true indicator of the religion advantages and potential benefits is how the patient interprets his faith. The most important thing is not the type of religion, but personalized approach to it. Noticing the diversity of attitudes towards religion, David Wulff in his book "Psychology of Religion" (1991, 1999) proposed a new viewpoint, based on the analysis of cognitive approaches to religion. According to Wulff, people's religiosity is characterized by two fundamental features. These features change their intensity depending on the patients. The first is the scale of belief in transcendence (belief in naturalism versus belief in mysticism). The second is the interpretation of the events we encounter (literal versus symbolic interpretation). The scale of transcendence assesses to what extent the patient accepts the metaphysical or mystical reality. The scale of interpretation assesses to whether the religious rituals and symbols are interpreted literally or symbolically.

As a result of combining those two dimensions, four fields (quarters) are created. Each quarter reflects the potential style of thinking about religion. Respectively, it is Orthodoxy, External Critique, Second Naiveté and Relativism (Wulff, 1991, 1999) [35]. Using the concept of Wulff, a Belgian psychologist, prof. Dirk Hutsebaut, created The Post Critical Belief Scale (PCBS). Thanks to this scale, it is possible to objectively assess the cognitive attitude towards religion [36].

Based on the described above Wulff's religiosity model and the PCBS scale, the relationship between religious style and attitude towards death was examined. The results of these studies have shown that religious people are more likely to show an accepting approach to death. For second, people who interpret religion literally report greater fear of death. Furthermore, literal and dogmatic interpretation of religion was associated with a greater fear of death and a stronger tendency to avoid and suppress thoughts about death. "Orthodoxy" type of people (convinced of the reality of mystic world, with a literal interpretation of religion) presented an attitude of acceptance or avoidance. "Second Naiveté" - type of people (who accept the fact that transcendent reality is real, but ideas are symbolic) presented an accepting attitude. "Relativism" - type of people (who reject the mystic reality and recognize religion as symbolic) presented a neutral or avoidance attitude. "External Critique" - type of

people (who understand religious claims literally, but reject their truthfulness) presented anxiety or avoidance.

Taking into consideration the above-mentioned facts, it can be stated that it is impossible to answer the question: whether exists one religion which provides human well-being and give better techniques for dealing with information about death. The mental state of the patient depends not so much on the type of religion, but on personal attitude to its assumptions. Therefore, it is extremely important to adopt the attitude of an individualized approach to each patient. Current patient's mental state should be taken into consideration. Well-educated tanatopedagist should be able to assess whether the patient's approach to his religion will facilitate or hinder the process of education about death. Tanatopedagist should be able to recognize the patient's mental state. The ability to qualify the patient for one of the religious styles allows a deeper understanding of the nature of their fears. These skills help in the selection of content and forms of conversation with suffering people. Depending on the situation, the goal of a tanatopedagists may be different. Some patients might fear a lot and would require hopeful words. Meanwhile, the others want only passive attendance without any conversations [10].

Therefore, tanathopedagogy consists of building a relationship based on empathy and an attitude of openness to experience another person. It is crucial to understand sense of the patient's existence, and thus also death. The task of tanatopedagists in this area is to stimulate reflection about life and the meaning of death. Tanatopedagogy also forces looking critically at culturally common patterns and stereotypes, including those related to the issues of death and illness.

SUMMARY. THE ROLE OF TANATOPEDAGOGIST AT THE CURRENT TIME

The tasks of tanatopedagogics, as the article above shows, are implemented on many levels. They require continuous improvement of the educational process of pedagogues, teachers and people accompanying patients in their suffering. Inspirations found in culture and art that contribute to creating the appropriate attitudes towards suffering, dying and death may be useful in developing teaching resources for tanatological education. Its main goal is to provide specialists with the necessary knowledge, determine their consciousness and instil appropriate competence in leading the discussion concerning the topics of suffering and death. The task of tanatopedagogues is also to overcome the stereotypical approach to death.

Education about suffering and death should be addressed to people of all ages, which is why various programs are being created using appropriate educational methods, adjusted to the target audience. They require reflection on the values that are important in the experience of the suffering of oneself or a loved one, formulating the principles of self-education in illness and preparation for the co-suffering and co-dying [4].

Tanatopedagogue's work also involves preparing programmes and projects for cooperation with mature people who are in the late stages of their lives. It should focus on issues related to building and strengthening relationships with loved ones, as well as on conscious being engraved in the memory of other people. The main goal of tanatopedagogical education is to develop people's ability to accept their and someone else's death in a conscious and mature way.

The key issue that tanatopedagogics explores is upbringing to death, as well as upbringing to life. It raises universal issues, common for all people. These include categories of illness, death, suffering, loss, but also the meaning of life. These categories refer not only to people who are affected by the disease, but also to their relatives, friends or strangers who display a high level of empathy. In this context, the tanatopedagogue's task is to educate people to co-suffer, and thus to make them aware of the meaning of life. Promoting hospice volunteering and active participation in similar activities contributes to the individual development of the volunteer and his awareness, and at the same time provides patients with the significant support. Such actions significantly modify society's attitude towards the sick, neutralize fear and anxiety caused by the vision of death and the process of dying, and lead to more conscious living [4,29]. Thus, internal attitudes of volunteers based on understanding themselves and the surrounding world are built. In this sense, tanatopedagogical education enables to gain new ways of understanding yourself and other people through open and honest dialogue.

The methods of helping the sick include not only the above-mentioned psychological support for the patient and his loved ones, but also assistance in organizing the optimal place for the patient's stay [4]. 75% of those who did not suffer from any serious illness say they would rather die at home [29]. However, it should be born in mind that in many cases, patients in hospices stay in better conditions than the ones that would be possible to provide at their place of residence. Hospice staff use all available means to alleviate the physical discomfort accompanying the patient in his last days. The aim of hospice care is also to

prevent from the patient's feeling of being lonely and unnecessary, help him and his family to overcome anxiety and inform relatives about the best possible ways of care for the sick [29].

Death and suffering are natural and inherent phenomena occurring in all societies. The need of gradual assimilation with the vision of death and acceptance of the inevitability of human fate seems obvious and essential. Tanatopedagogue should not impose his beliefs or views on anyone, judge or discriminate, but rather seek to broaden his knowledge. This makes him a good teacher and mentor for people experiencing difficult incidents and for himself is a source of expanding his mind and horizons.

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The impact of physical exertion in the therapeutic process and modification of the course of cancer disease

Gabriela Gut, Mateusz Nowak, Aleksandra Kurczak, Aleksandra Grzegorzczak, Maria Greniuk, Natalia Nowak

Medical University Piastów Śląskich of Wrocław, Wrocław

INTRODUCTION

Carcinoma is a heterogeneous group of diseases that, in most cases, significantly reduce the physical functionality of patients. The implementation of aggressive therapies to the infirm has many negative effects, including weakening of cardiovascular system function, decrease in physical fitness, nagging pain, and fatigue. Decrease in muscle mass often leads to a gradual impairment of the patients' activity [1,2].

Weakened muscles and general fatigue cause worse prognoses by prolonging oxidative stress and inflammation. Physical activity plays a positive role here and has a beneficial influence on the negative effects of cancer in the body. It allows maintaining physical condition and muscle mass, and encourages lowering body fat that allows maintaining normal physiological function of the body and reduce inflammation parameters by regulating cytokines. In some cases, it stanches the progress of cancer and the undesirable effects of cancer therapy, which is described in detail in each of the respective sections.

The effectiveness of the exercises depends on their form and intensity. Aerobic and resistance exercises have been found to be the most effective [3].

Greater muscle mass gives patients the strength to perform further training and daily activities. The introduction of oxygen training to the patient's daily activities significantly improves their quality of life. Furthermore, by maintaining constant physical activity, the risk of developing depression and fatigue is reduced. It also improves bone density, level of hemoglobin in the blood and improves lung oxygen capacity [4].

Cancer patients are therefore, recommended 150 minutes of moderate aerobic exercises per week. However, the expected gain in muscle mass will be achieved through resistance exercise.

PHYSICAL EXERTION AMONG SPECIFIC ONCOLOGICAL PATIENTS

Physical exertion is activity aimed at exerting muscles in various ways to keep fit and its benefits to the body are manifold. It stimulates physical, motor, and psychosocial development. It leads to decrease in blood pressure, improves cardiac function, enhances oxygen transport, develops the network of vessels surrounding the heart, enforces full ventilation of the lung, and a lot of more. Physical activity is well known to prevent diseases, but in this article we want to focus on its impact on the course of treatment and to suggest that it should, therefore, be implemented for preventive purposes as well as during therapy. A preliminary review of the current literature suggests that certain types of cancer respond well to the implementation of physical activity in the treatment process. The following paper will characterize the impact of exercise sequentially on breast cancer among women, colorectal cancer, and hematological diseases. Existing literature indicates that the implementation of physical exertion in these particular diseases has the best results in improving patients' prognosis, reduces side effects of cancer treatment, and ameliorates patients' mental condition. In addition, it appears to be safe for patients in the most cases. They do not have to be advanced sets of exercises, it seems beneficial to undertake any activity as evidenced by the scientific reports cited below.

PHYSICAL ACTIVITY AMONG FEMALE PATIENTS WITH BREAST CANCER

Breast cancer is still considered to be a major oncological problem in developed countries, despite enhanced diagnostic and therapeutic medical techniques. It is the second most common cancer in the world (after lung cancer). Morbidity in Poland is constantly increasing, with around 1,33 million women living with the diagnosis of breast cancer as of 2013 year [A]. Breast cancer is malignant and often requires introducing intensive and long-term treatment (chemotherapy, radiotherapy, and surgical treatment), which is connected with undesirable effects. It is important to reduce the side effects of treatment and improve quality of life [5].

In the mid 20th century, physical activity didn't constitute an important element of breast cancer therapy. Nevertheless, there is a statistically significant correlation between physical activity, psychological well-being, and subjective opinion about improved quality of life [6].

One of the earliest research studies conducted in 1989 on 45 women (with II stage of breast cancer during chemotherapy), found that 10-weeks of interval aerobic exercises enhanced body composition and also reduced vomiting associated with chemotherapy [7]. Exercising during breast cancer treatment is currently recommended because it improves quality of life by reducing the side effects of treatment and improves the quality of sleep. It likely increases the survival time as well [8]. A study on 1327 women with breast cancer among the Norwegian population, observed a particular connection between physical activity among women after cancer diagnosis and reduction in overall mortality [9].

Studies conducted on patients with breast, prostate, and colorectal cancer, show a strong correlation between doing physical activity after being diagnosed and longer survival times. Reducing mortality in breast cancer is already visible after taking a 3-hour-long walk during one week [B]. In research published by Cochrane Library that studied 5761 women with breast cancer, the participants who practiced varied types of sport (aerobic exercise, riding a bike etc.), achieved better results in cognitive function studies and had fewer symptoms of depression, fatigue and instead had high self-esteem [10].

Obesity is a risk factor of breast cancer development and worsens prognosis [11]. It has been proven, that adipocytes, which are located in the closest surrounding of the tumor, react with cancerous cells by sending signals that stimulate growth and increases invasiveness [12]. Furthermore, higher quantities of fat induce chronic inflammation and increase anaerobic metabolism. Chronic inflammation, in turn, contributes to the development of the cancer [13,14]. Hyperlipidemia associated with obesity, can also increase the risk of cancer and intensify the potential of metastasizing. Proliferation of tumors is higher in patients with hyperlipidemia [15]. The molecular basis of this correlation is associated with the fact that the cholesterol metabolite 27-hydroxycholesterol (27-HC) is considered to be an agonist of the estrogen receptor. Because of this, 27-HC may have a big impact on breast cancer growth among women with tumors expressing estrogen receptors (ER+ breast cancer). In addition, activation of phosphoinositide 3-kinase enhances tumor cells multiplying [16]. It was also observed that preoperative patients with higher levels of HDL cholesterol, benefit in overall survival [17,18]. Nevertheless, the role of hyperlipidemia and the impact of individual types of cholesterol on breast cancer growth, requires further research.

Physical activity inhibits the start of pouchitis, fatigue, and cardiotoxicity caused by chemotherapy [19]. Lymphedema caused by surgically removing lymph nodes or damaging by radiotherapy is reduced due to muscle function improvement [20]. Moreover, aromatase inhibitors used in ER+ breast cancer therapy, intensify osteosclerotic changes and result in

burdensome joint pain. Regular exercise reduces the intensity of pain by improving control over it and increasing the bone density [21].

No harmful effect of physical activity on the health of breast cancer patients has yet been proven. Exercising has a positive effect on mental and physical health, and also leads to slowing down cancer's progress in the organism. The slow down of breast cancer development indicated by physical activity is estimated to be between 15% and 80% [22]. It has a positive effect on primary and secondary prophylaxis. Starting physical activity at any age is beneficial. However, maintaining a regular, active, and healthy lifestyle gives the most promising results [23].

PHYSICAL ACTIVITY AMONG PATIENTS WITH COLORECTAL CANCER

Colorectal cancer is also closely connected with physical activity. Carcinomas in the large intestine and anus, are the third most common cancer in polish men, second in polish women. The number of new cases is greater among the populations of developed countries populations, increases with age, and is 1.5-2 times more common in men [C]. Accordingly, it is a real threat in European countries. It has been found that the implementation of walking 6 hours per week causes a reduction in mortality among patients with this cancer [24,B].

Devine research reports that high-intensity workouts are very safe for these patients. Exercises help with the function of respiratory and circulatory systems and optimize percentage body composition [25]. Furthermore, Vallace's analysis presents a positive impact of moderate and more energetic physical activity on colorectal cancer treatment and a noticeable improvement in the mental health condition of patients [26]. In 2006 Meyerhardt published a prospective study that included 832 patients in stage three of colorectal cancer treated by chemotherapy. Analysis indicated that higher levels of physical activity was connected with tumor growth shrink, relapses reduction, and benefit in overall survival [27].

The CHALLENGE (Colon Health and Life-Long Exercise ChaNGE) trial, carried out by the National Cancer Institute of Canada Clinical Trials Group, investigated the impact of exercising on the course of the disease among patients with colorectal cancer in stage II and III after chemotherapy. The study included 932 people. They were randomly divided into two groups. Participants from one group had to start working out daily. Parameters like general quality of life, fatigue, level of fear, sleep quality, depression, were assessed by the Past Year Total Physical Activity Questionnaire every 6-12 months. Blood tests, tumor markers, electrolyte panel, insulin, and IGF levels were also checked. Results showed a positive impact

of beginning the physical activity, especially on side effects caused by chemotherapy. Physical and mental health conditions improved in the group of patients engaged in physical exercise. The authors of the study suggest that in the future, it would be possible to reduce treatment costs because people engaged in physical activity may not require antiemetics, pain killers, and sleeping pills [28].

There are several plausible mechanisms that can explain the correlation between physical activity and colorectal cancer growth. One of them is the metabolic consequences of obesity that include inhibited intestine passage, lower levels of insulin, reduced insulin sensitivity, higher levels of prostaglandins, inhibited production of bile, and changed gut microbiome. Unfortunately, these changes have a major impact on digestive system functioning. They stimulate abnormal proliferation and inhibit normal cell apoptosis, which contributes to cancer development [29].

The lack of balance between proliferation and apoptosis is the result of mitochondrial malfunctioning. During exercise, biochemical mechanisms are initiated. Their target is to generate enough energy. Constitutive stimulation of these processes during regular activity promotes mitochondrial biogenesis, oxygen metabolism, PGC-1 α protein production and activation. Protein PGC-1 α is a mitochondrial regulator. It is involved in thermogenesis, fatty acid oxidation, and glucose metabolism [30]. Furthermore, the elevated level of this protein inhibits inflammatory markers in the organism and reduces inflammation. Chronic inflammation induces tumor growth [31,32]. Lack of physical activity has the opposite effect. It leads to the release of free radicals, changes in glucose metabolism, and finally to apoptosis inhibition [33,34].

PHYSICAL ACTIVITY AMONG PATIENTS WITH HEMATOLOGICAL TUMORS

Hematologic malignancies are caused by abnormal proliferation of hematopoietic stem cells, blood morphotic elements, and cells that colonize lymph nodes. Incorrectly functioning morphotic elements circulating throughout the body negatively influences many systems. Therefore, these types of neoplasms seriously affect the daily physical functioning and quality of life of the patients. Especially in this group of patients, there is deconditioning of the cardiopulmonary and neuromuscular systems. Neoplastic process leads also to nutritional deficiencies and cognitive disturbances such as attention and memory disorders. Furthermore, impaired proprioception, reduced autonomic tension and neuromuscular weakness increases the risk of patient falls. In addition, co-occurring thrombocytopenia and reduced bone mineral

density are also risk factors of pathological fractures and heavy bleeding which could be very difficult to control.

MULTIPLE MYELOMA (MM)

Taking into consideration the data available so far, moderate exercises in hematological malignancies, seems to be safe, feasible and effective. They can also lead to functional improvement. Multiple myeloma (MM) is one of the haematological malignancies in which physical exercise appears to have a significant impact on improving the patient's quality of life. MM is a B cell tumor characterized by clonal proliferation of plasmatic cells. It is the second most common blood cancer (13%) in the United States, after non-Hodgkin lymphoma. It is also the most common primary bone cancer [35].

Patients with multiple myeloma initially suffer from back and bone pain. This characteristic pain is caused by osteolytic changes that develop during the course of the disease. Bone related complications occur in approximately 80% of patients at the time of diagnosis. The most commonly affected locations include long bones, ribs, skull and pelvis.

Excessive osteolysis is the result of more frequent fractures. Population studies show that pathological fractures affect 26% to 34% of patients with multiple myeloma. High bone turnover (which means increased bone resorption) also significantly correlates with higher proliferation rate and longer survival of cancer cells in multiple myeloma. [36-38] Pathological fractures cause severe pain, reduce mobility, and may require surgery and hospitalization. These complications increase the risk of death by 23% - 32% [39]. The most common fracture site is the lumbar spine (55% - 70%) [40]. Moreover, weakness and paraesthesia of the lower extremities may occur as a result of the described compression lumbar fracture among these patients [41,42].

The solution to the problem of reduced bone density in this group of patients may be education and implementation of moderate physical exercises in patients' daily routine. According to the research cited below, exercises have a beneficial influence on bone mass and bone metabolism.

The work of Brahma et al. on exercise-related bone metabolism has shown a statistically significant change in bone reorganization in a group of healthy participants. The study was conducted on participants who underwent a 35-minute effort. Three blood samples were taken. The first sample was taken immediately after the physical exertion, the second - 30 minutes after the end of activity, and the third - 24 hours later. A statistically significant correlation was detected between exercise and decrease in plasma volume at initial stages of regeneration. In

"zero-minute" and in thirtieth minute the plasma volume decreased by 4.3% and 15.1% respectively in comparison to the initial results. Whereas after 24 hours, an increase of plasma volume by 7.5% was observed. The other surprising fact was that serum osteocalcin concentration remained unchanged throughout the study, but total production increased at the final stage when plasma volume increased by 7.5%. The serum PTH concentration also finally increased in proportion to the intensity of the exercise and remained elevated during prolonged regeneration. During post-exercise regeneration, bone metabolic changes took place. PTH secretion was subject to constant stimulation, with a simultaneous increase in BMD (bone mineral density). These observations correlate well with the beneficial effects of exercise and training on bone mass [43].

In the meta-analysis including 13 cohort studies, moderate or intensive physical activity has also been found to correlate significantly with a reduced risk of hip fracture by 45% for men and 38% for women. These results suggest that the risk of falling is reduced in physically active people, with potentially increased risk in the most active and inactive people [44].

MYELOPROLIFERATIVE NEOPLASMS (MPN)

Another group of patients with hematological malignancies among whom physical activity contributes to a better quality of life are patients suffering from myeloproliferative neoplasms. Mildly progressive myeloproliferative neoplasms (MPN) such as polycythemia vera or essential thrombocythaemia treated properly could become chronic diseases. During the course of the above diseases, fatigue is the most commonly reported symptom among this group of patients. Chronic Fatigue Syndrome (CFS) will be described in detail in the next chapter below.

Nevertheless, in the research conducted on 1.179 patients with MPN, 81% of patients reported fatigue [45]. In the another research conducted on 1.788 patients with MPN, 73% of them tried to exercise in order to control their fatigue. Statistical analysis showed that among regularly exercising patients 63% reported an improvement in their quality of life and assessed exercise as an effective strategy to reduce the unpleasant effects of their disease [46]. This is very important considering the fact that patients with MPN report fatigue and CFS as the most troublesome symptom that they would like to resolve most often [47].

Moreover, the patients with MPN are at greater risk of thrombotic episodes. Incidents such as deep vein thrombosis, ischemic stroke, or myocardial ischemia are relatively more common in this group of patients compared to the general population. Exercise has been shown

to increase both process of coagulation and fibrinolysis while the balance between these two processes remains unchanged. Consequently, this reduces the risk of developing thromboembolic incidents in patients at increased risk [48]. Considering all the available research on physical activity in hematological diseases, it seems to have a justified impact on patients' quality of life. However, more research is needed to assess for which patient groups it will be most effective

PHYSICAL ACTIVITY THERAPY IN COMPLICATIONS AFTER AGGRESSIVE CHEMOTHERAPY

With the increase of cancer survival, the severity and range of long term side effects directly linked to chemotherapy gains in importance. From the doctor's and physiotherapist's point of view, not only the process of treating the patient but also their future functioning should be of the utmost importance. It should not be forgotten that patient's quality of life should be as close as possible to their life before the illness. In light of the above physical activity seems to be a good form of therapy also for patients after antineoplastic treatment. It contributes to reducing the most common long term side effects which are chemotherapy-induced peripheral neuropathy and chronic fatigue syndrome.

CHEMOTHERAPY INDUCED PERIPHERAL NEUROPATHY (CIPN)

One of the most severe illnesses linked to chemotherapy is chemotherapy-induced peripheral neuropathy(CIPN). The illness occurs in patients who are taking chemotherapy which induces axonal degeneration such as taxane-, platinum-, or vinca alkaloid-based medication [49,50].

The symptoms of the illness are tickling, numbness, burning and pain in legs and/or hands. Additionally, patients often suffer from imbalance, hyporeflexia, limb weakness, cramps, and less frequently: vertigo, loss of hearing, and constipation. Mentioned symptoms can contribute to decrease in physical activity and lower quality of life, which often translates to high mortality after antineoplastic treatment [49,51]. Statistically, 68% of patients after chemotherapy suffer from CIPN (after a year the rate decreases to 33%) [52].

Options for treating CIPN symptoms are limited due to lack of information about this illness. Some doctors recommend lowering doses of chemotherapy, which is causing these symptoms, but this approach can have quite severe consequences in the case of antineoplastic

treatment, and as such is not considered as optimal[49]. In spite of many efforts to use different substances in treating CIPN consensus on how to treat it is yet to be reached [D]. In light of these facts researchers currently draw more attention to physical activity as a form of treatment of CIPN.

Research shows that prescribing (after proper physiotherapeutic education) nerve gliding exercises to patients with breast cancer can significantly influence perception (measured with NPRS- Numeric Pain Rating Scale) of pain caused by effects of CIPN, power of a grip and levels of pain felt during pressure [53]. Balance-based exercises according to Kneis considerably improve proprioceptive reception of impulses linked to balance in patients with CIPN. Those patients initially, due to the illness, base their balance on information from vestibule rather than from proprioceptors [54]. Other benefit from physical exercises, which is outlined in Kleckner's research is decrease in sensation of hot/cold feet and/or palms. This paper also showed that the impact was greater in elderly patients [50]. Very interesting findings come from research on the impact of yoga and meditation on CIPN. During this study, patients were subjected to 1,5-hour sessions of somatic yoga and meditation twice a week. After 8 weeks a considerable increase in flexibility joined by improvement of overall physical ability and decrease in risk of a fall were observed. Another benefit for the patients, which came from this practice was creation of patient support groups which gave them a sense of community and companionship in their struggle with CIPN [54].

Many more studies should be conducted in order to find a holistic approach to treatment of chemotherapy-induced peripheral neuropathy. Physical exercise should be taken in consideration as a part of the solution as they can deliver substantial improvement in patient's health and if conducted in groups they can provide moral support for the ill which is one of the most important factors in successful treatment.

CHRONIC FATIGUE SYNDROME (CFS)

Another complication, that may develop in patients who were cured of cancer, is chronic fatigue syndrome (CFS). It is a medical condition characterized by long-term fatigue, with other symptoms, such as weariness, lack of energy, tiredness, difficulty with thinking and starting a physical activity. As opposed to weakness occurring physiologically after an exercise, fatigue felt by patients does not subside [55].

CFS may occur during the advanced stage of a tumor, as a result of the treatment or after the therapy [56]. The treatment and its long-lasting effects significantly lower general

condition. The chemotherapy prevents the tumor from growing and spreading by destroying the cancer cells, as well as the healthy tissue. It is an unavoidable side effect. Consequently, CFS is diagnosed in 70-100% of patients who underwent chemotherapy, immunotherapy, and radiotherapy [57]. Studies have shown, that chronic fatigue syndrome in oncological patients causes difficulty in coping with the disease and promotes destructive strategies [58,59].

Physical activity is an effective method of the therapy for chronic fatigue syndrome. It is important to adjust the workout plan individually, considering comorbidities (e.g. cardiovascular diseases). The activities cannot also exacerbate tumor disease symptoms (e.g. pathological bone fracture caused by exercise in bone metastasis). The studies have shown that optimally individualized activity plans lower the symptoms of CFS in up to 50% of the patients [60]. The hematological parameters are also improved, which may be the reason for the improvement [61,62]. The research suggests rhythmical, repeated exercises, such as walking or riding a bike, as the most effective method of dealing with CFS if oncological patients [63].

Furthermore, physical activity helps patients to adapt and increases awareness about energy strategies. GET (Graded Exercise Therapy) is a movement therapy which provides gradual prolongation and intensification of exercises. During this program the patients learn their individual levels of physical capacity. Moreover, because of the gradual elevation of range motion, starting from a few easy moves and then adding more advanced positions, general physical functionality rises [64]. The studies had shown, that GET therapy partly lowers the level of general malaise and after- exercise tiredness and enhances work and social adjustment [E].

RESTRICTIONS ON THE APPLICATION OF PHYSICAL EXERCISE AMONG CANCER PATIENTS

Nowadays, physical exercise is a very important subject of many studies that analyze its impact on the cancer patients course of treatment. Most of the studies proved a lot of beneficial effects of exercising during the therapy and important improvement in general condition of their health. Intensive researches are currently in progress and scientists are trying to set a list of guidelines to improve the effectiveness of cancer treatments. Despite all the positive effects of the implementation of physical effort in the therapeutic process of the patients, one should always keep in mind their general good and the awareness of the potential damage that such an effort can cause. Medical recommendations should be individualized and adapted to the current clinical situation of each patient.

As previously mentioned, breast cancer is the most common malignant cancer diagnosed in women. People who are qualified for medium or high-risk groups are treated with complementary therapy in which a special group of chemotherapeutic agents called the anthracyclines are given. One of their most significant side effects is cardiotoxicity. This group of agents show the highest cardiotoxicity potential among all the chemotherapeutic agents routinely used [F]. Scientists decided to look into their impact on exercise tolerance. For this purpose, two groups of female patients in the same age range were examined. The first group consisted of the healthy women and the second group was the breast cancer patients treated with the anthracycline. The results indicated that the group of patients treated with the anthracyclines showed a reduced maximum oxygen consumption index (VO_{2max}) and a reduced cardiac index during the exercises [65]. Another article published in the magazine “The Oncologist” described a strong correlation between the anthracycline therapy and a structural change in muscle tissue. In comparison with the results at the beginning of the treatment, there was a clear increase in body fat with reduced muscle mass, without significant changes in their bioenergetics. Also in this case, a reduction of the VO_{2max} peak was observed. Generally speaking, this shows a cardiopulmonary limitation to exercise among this group of patients. The measured VO_{2max} parameter is an indicator of the functional capacity and it defines the exercise tolerance threshold [66,67]. Thus, the patients treated with the anthracyclines require special attention considering that physical effort may become a risk factor through the exacerbation of treatment-induced cardiac disease.

In addition to popular therapy, some specific types of cancer also include the stem cell transplant (SCT) procedures as a form of the treatment, which were mentioned while discussing the hematological diseases. This method is associated with many complications that include immunosuppression, which is a condition where even a usually harmless germ can cause a dangerous infection, or graft-versus-host disease which may also cause life-threatening complications. In these cases, exercising should not be recommended, and all daily activities should be performed with extreme caution. The above-mentioned complications significantly translate into a reduction in muscle mass and its function, and negatively affect the functioning of the circulatory and the respiratory system, exacerbating the body's inefficiency, which could significantly decrease level of the physical tolerance [68].

Furthermore, the time elapsed since the end of the treatment is also a factor in the efficacy of physical effort. Performed within 2 hours of radiotherapy and within 24 hours of intravenous chemotherapy, they accelerate circulation, which translates into an increase in the effect of the drug - enhancing, both its positive effects and the side effects. The physical activity

may increase fever and cause hyperthermia which may result in multiple organ failure [G]. Nausea and / or vomiting that might occur within 24 hours of treatment may be aggravated.

Moreover, the occurrence of thrombocytopenia, leukopenia or anemia are also indications for discontinuation of exercise therapy due to the risk of bleeding or tissue damage [69]. Confusion, blurred vision, fainting, pallor, night pain or non-traumatic pain are conditions in which the patient should rest and temporarily stop physical exertion if they have previously used it. It was also noted that the concomitant diseases such as cardiovascular insufficiency, acute infectious diseases, and metabolic diseases endure physical exertion less well, therefore more research is needed to determine the safe tolerance range of physical effort for this group of patients with cancer, noting that the main task of physicians and scientists is to help the patient improve his functioning, not to generate new side effects [70].

CONCLUSIONS

Taking into account all research described above, it can be stated that the implementation of physical activities in patient therapy improves the final effects of treatment. Moreover, it supports the patient during the treatment process by increasing his tolerance to the negative effects of chemo and radio therapy. Rehabilitation of people with cancer can play a key role not only in reducing the unpleasant feeling of therapy related side effects, but also in reducing their frequency incidence. Improving the quality of patients functioning during therapy is associated with patients' greater motivation. Thanks to this, the patients not only physically improve their body's efficiency and better tolerate aggressive treatment, but also improve their psychological well-being. Reasonable implementation of sport and physical activity in the future may become an inseparable element of therapeutic schemes among cancer patients. In fact, there seems to be an emerging trend in modern biomedicine to view disease in a holistic way and individualize treatment.

Therefore, the authors recommend spreading awareness about the impact of physical activity on the course of cancers that will allow for better results in the fight against cancers. This must also be accompanied by extensive and well-planned scientific research that will provide key information on the type and duration of exercises that will be most optimal for specific groups of patients.

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Awareness of tanatology in modern society- do we lack an education?

**Patrycja Bolla¹, Natalia Markiewicz¹, Jan Gnus^{1,2}, Łukasz B. Lewandowski¹,
Jakub Migoń¹, Małgorzata Paprocka-Borowicz^{1,2}, Krzysztof Dorna³**

1. Wrocław Medical University
2. Regional Specialist Hospital in Wrocław, Research and Development Center, Wrocław
3. Pontifical Faculty of Theology in Wrocław

INTRODUCTION

The science of dying and death has both a scientific and philosophical dimension, it can be understood in countless ways. For some, it is limited to a purely practical view and provides answers to specific questions. Forensic specialists, detectives and coroners, whose institution is not yet present in our country, are people who use the empirical dimension of Thanatology.

They deal with the description of tangible phenomena that help them determine the circumstances of human death, such as their time or mechanism. The development of medical thanatology has resulted in the creation of interesting initiatives aimed at expanding knowledge in its field, one of them is Body Farm. These are places examining the impact of various factors on the decay of corpses and all phenomena that occur during this process. Numerous scientific research results in an increase in the number of academic textbooks and publications in this field.

Thanatology is also less empirical. In the field of humanities, it focuses on the way in which a person experiences the process of dying, on reflections, thoughts and emotions that accompany him. It is looking for ways to help a person who learns that his disease is incurable and terminal. It is a multidimensional field addressing topics not only directly related to the death of an individual. At the same time, this science discusses how death affects people in the immediate vicinity of the dying person, including medical staff.

There is no doubt that the process of dying and death is an integral part of the doctor's work. One cannot forget about its spiritual and emotional aspects. They are the most important for people in mourning around the world, regardless of cultural and social conditions. The

reconciliation of the scientific aspect of thanatology with its humanistic dimension is a challenge that medical staff must face in their daily work.

WHAT IS THANATOLOGY

Thanatology according to the commonly accepted definition is the science of human death. This word comes from Greek mythology, from the god Thanatos [1]. In the literature, he is usually depicted as a young man with black wings on his shoulders, which symbolize death, and with an extinguished and inverted torch in his hand. According to beliefs, he would appear unnoticed and cut off a lock of hair of the dying person. After death, people were sent to the underground kingdom of the god Hades, where they were judged. The dead went either to the Elysium Fields - equivalent to paradise, or to Tartrate - equivalent to hell.

This science particularly deals with the qualitative description of the causes of death, regret and other emotions that accompany it. Thanatology is an interdisciplinary field using, among others from knowledge of such issues as: acquired immune deficiency syndrome, pain, euthanasia, philosophy, medicine, funeral, suicide, aging, art and war. It can deal with the meaning of death for both individuals and communities and cultures.

Thanatology also examines medical intervention in the process of death to prevent death or alleviate agony. One of the reasons for the development of this discipline was the progress of medical technology that allows the extension of a human life. Thanatology recognizes that death is ultimately inevitable. It works to develop guidelines to facilitate the dying process. Knowledge in this field is widely used by doctors in the field of forensic medicine, as well as medics of other specialties [2].

IN LITERATURE

Thanatology has been a theme in literature since the dawn of time. According to the Old Testament, death is the end of every existence, because God alone is immortal and will never pass away [3]. Christians treat death as "ianua vitae," or the gate of life, an escape from worldly suffering and scarcity, and at the same time as a punishment for sins, a tragedy that will not bypass anyone. Death takes everything and everyone regardless of their origin, property or social status. In the Middle Ages, death was often depicted through the theme "Danse macabre" - a death dance that was medieval image of the procession of people of

different states and professions led by death. This dance reflected the ubiquity of death, which treats all people the same. This allegory is to make people realize mutual equality at the end of life, to cause fear and sadness.

Thanatology is also a topic often taken up by contemporary authors. In the book Sue Black entitled "All That Remains: A Life in Death," the author particularly points out that the human view of death has changed dramatically over the past centuries [4].

Our ancestors treated death as a friend, something they cannot escape, so the only way out is to reconcile with her and accept her. Contemporary people, on the other hand, consider her an opponent, an uninvited guest from hell, who must be fought with or avoided as long as possible.

However, no matter what humanity believes, life and death are inextricably linked and belong to the same continuum. Neither of them can exist without the other. It is also certain that, unfortunately, regardless of the efforts of modern medicine, death always wins at the end. Sue Black is particularly interested in the way people depict death in literary works [4]. It has been given ominous nicknames, such as the Pale Rider, the Grim Reaper, Kostucha, and the Great Equalizer, and it is most definitely presented as a slender skeleton with a hood on its head, holding a deadly scythe, ready to separate the soul from the body with one sweeping motion. Although in the vast majority of languages the word death is feminine, it is most often portrayed as a man.

Thanatological motifs currently appear even in literary works intended for the youngest readers [5]. Taboo subjects have always existed in children's and youth literature. This, of course, resulted from the educational and educational function of this literature, which, being subordinate to adult literature, is full of numerous restrictions. Today, "prohibited topics", including regarding illness, suffering and death, penetrate into the literature for children [6].

WORLDWIDE

In different cultures, the death of loved ones and commemoration of passing is different. All Saints is a holiday celebrated on November 1 in the tradition of the Catholic Church. This ceremony is mainly derived from the worship of martyrs who gave their lives for faith in Christ, and who were not mentioned either in local martyrologues or in the canon of the Holy Mass. On the Feast of All Saints, Poles visit cemeteries to decorate the graves of their loved ones with flowers and light candles. It is a day full of prayer and reflection.

Mexicans, on the other hand, believe that their loved ones who died, on the first and second of November can return to their homes to enjoy the company of the family [A]. That is why souls should be helped to find their way home and greet them best. Mexicans decorate their houses with arches made of yellow marigolds or marigolds. They raise altars full of flowers, colorful candles and fruit. There is no shortage of food for the ghosts to eat after a long journey. In the Mexican tradition, the Day of the Dead is a joyful occasion, because the dead still exist, and their souls, evoked and guided by velvet buds, sympathize, enjoying in the company of their celebrating families. The traditional festival "La dias de Muertos" is celebrated mainly in Mexico and Central America, but has also taken root in many places in the USA, and wherever Mexicans and immigrants from Central America have settled [B].

Another holiday associated with the dead is Halloween, which is the most celebrated in the United States, Canada, Ireland and Great Britain. Although the day is not an official holiday, it enjoys the great popularity. Halloween in Poland appeared only in the 90s, but it is still a foreign tradition that does not supplant the commonly practiced celebrations of All Saints 'Day and All Souls' Day, but is treated rather as innocent fun. This is associated with a different attitude to the issue of death in our culture and a great respect for cemeteries. Not without significance is the fact that most Christian denominations, and above all the Catholic Church condemns Halloween, identifying American play with the cult of Satan. Despite this, Halloween successfully operates in mass culture [7]. The main symbol of this holiday is a hollow and backlit pumpkin with jagged teeth. Other popular themes include ghosts, demons, zombies, vampires, witches, and skulls.

The traditional Chinese festival of the dead is called the festival of the spirits. According to the Chinese calendar, it falls on the fifteenth day of the seventh month. The Ghost Festival is organized in cemeteries as well as in private homes. It is common to launch small reed boats with lanterns that are to guide lost souls [8].

QUESTIONNAIRE

A survey between medical student was carried out to examine knowledge of thanatology. It was assumed that most of them should be familiar with this concept, which is closely related to the nature of the faculty they study. Study of thanatology, such as medical ethics and forensic medicine, are carried out during various years of training. Consequently,

there should be no significant difference in knowledge of this issue in the group of students surveyed.

The survey was conducted anonymously. The study group consisted of 69 medical students, 45 women and 24 men. Among the respondents, 40 people (57.9%) were familiar with the concept of thanatology, while the remaining 29 people (42.1%) have never heard of it (Fig. 1).

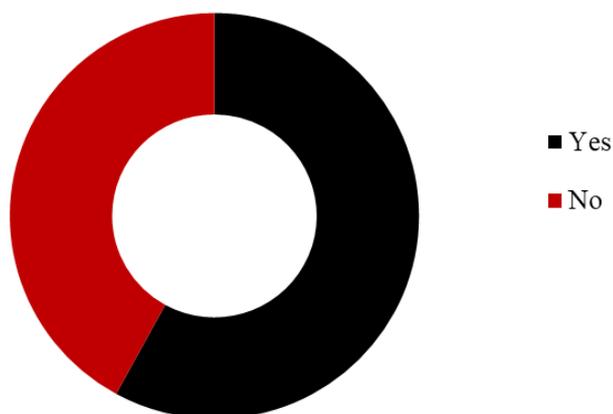


Figure 1. Are you familiar with the term "tanatology"?

Respondents who answered in the negative to the question of knowledge of the term thanatology were then asked if they were going to find out what it means. Among them, 22 people (75.9%) answered this question in the affirmative, 3 people (10.3%) denied, and 4 people (13.8%) remained undecided (Fig. 2).

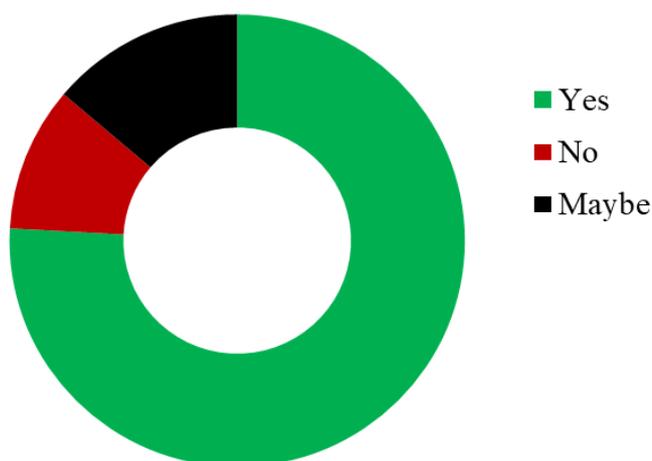


Figure 2. Are you going to make yourself familiar with the term "tanatology"?

The vast majority of respondents seemed interested in the issue presented. When asked about the probable meaning of this term, they associated it with the science of death and the Greek god of death, Thanatos. Thanks to the widespread use of the topic of death in culture, most of us subconsciously associate the notion of thanatology with death, even if we have not encountered it before. Only a few were unable to explain what thanatology could possibly mean. Individuals developed this concept as a science dealing with an unspecified method of treatment or as a field of preparing a body for burial.

A group of people who were familiar with the concept of thanatology were asked to answer multiple choice question in which they were asked to indicate how they were able to get acquainted with it (Fig. 3). Half of them chose academic classes as the only or one of the options. 30% of respondents indicated non-academic literature, and 25% also academic textbooks. 20% of people indicated that they were familiar with the term via the Internet, and 7.5% of respondents heard about it for the first time from friends. It is therefore evident that classes and academic textbooks are the main source of knowledge about thanatology among medical students.

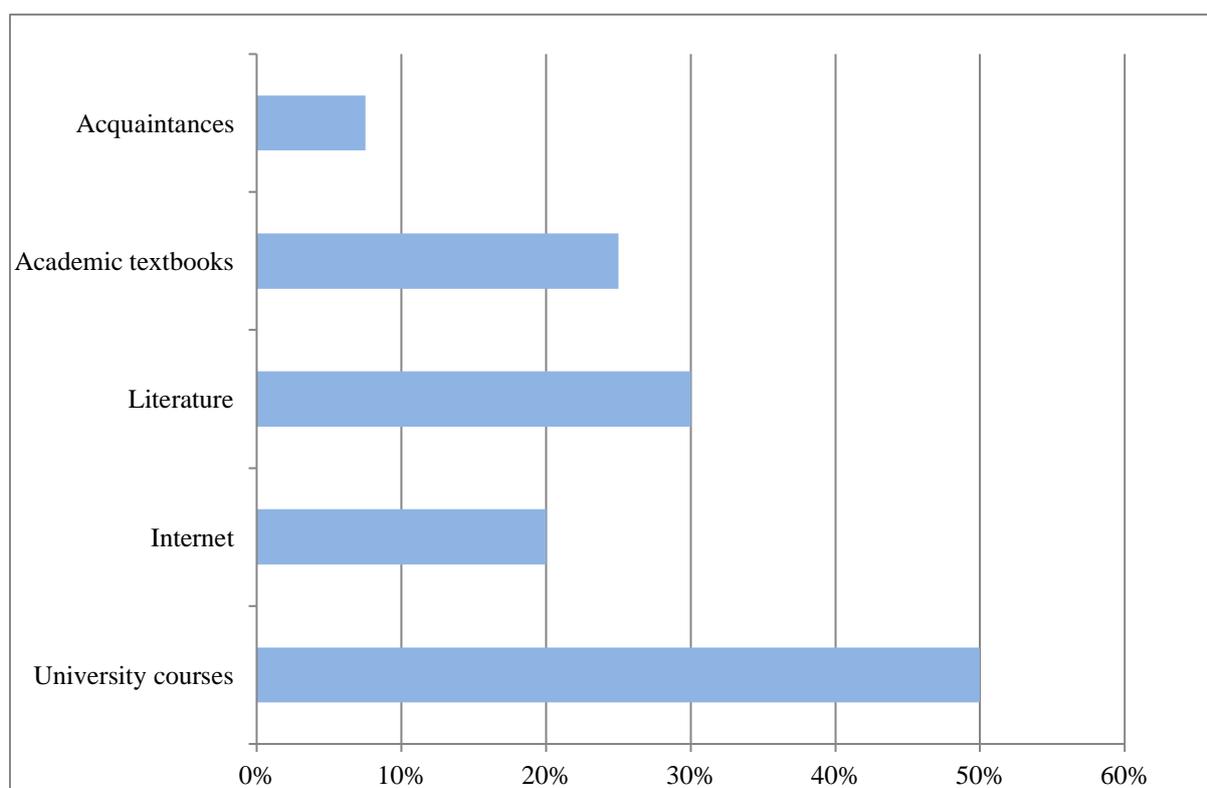


Figure 3. How did you become familiar with the term "tanatology"?

After answering the above, the examined group was asked another two questions, this time of single choice. Questions were aimed at finding whether they consider it thanatology as interdisciplinary science (Fig. 4) and whether they believe that topics related to it should be raised during their medical studies (Fig. 5).

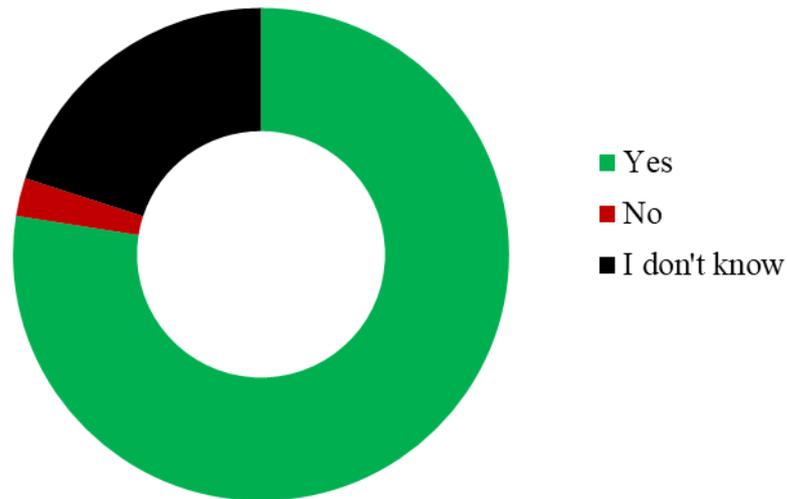


Figure 4. Do you think that tanatology-related issues should be discussed during university classes?

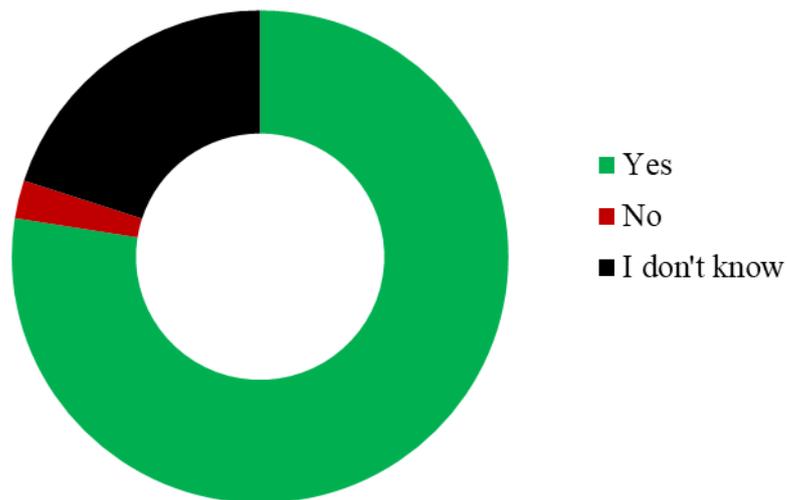


Figure 5. Do you think that tanatology is a cross-disciplinary field?

The vast majority of respondents, as much as 77.5%, considered thanatology as an interdisciplinary branch of science. Only 2.5% of respondents disagreed with this statement, while another 20% were unable to give a definite answer. The second question obtained the same percentage distribution of answers among the study group. 77.5% of respondents believe

that the topics related to thanatology should be raised during their studies in medicine, 2.5% do not agree with this statement, and another 20% could not give a definite answer. However, the answers to these questions showed no correlation. If the respondent answered the first of them in the negative or could not answer, usually the answer to the second question was different from the first one.

IN CONCLUSION

Death is an inseparable part of life; it accompanies man at its every stage. We have to face Death saying goodbye to our loved ones, and then, at some point, it will concern us directly. Usually, death is an unexpected phenomenon, although one cannot be really ready even for the expected event. Closeness of the death resulted in human interest in the subject, which can be observed in literature and art over the centuries, from ancient times and the building of tombs in the form of pyramids, to modern times and the ubiquitous fascination with forensics and criminology.

The survey attempted to find whether the field of science directly related to death and the dying process is close to medical students. People who will inevitably have to face death in their professional lives and be a support for the families of people who are in the process and subsequently die. As shown, unfortunately over 40% of students did not know the concept of "thanatology" before completing this anonymous survey. Importantly, most of these people subconsciously associated it with death. Among those who knew this term before completing the survey, the vast majority believe that this is a topic that should be addressed during the education of future doctors. As shown, unfortunately, knowledge of thanatology probably requires dissemination among students of medicine.

Doctors are a professional group that very often accompanies other people in the process of dying. It is essential that they are well prepared for it. Without extensive knowledge in this field and relevant experience, they will not be able to support patients and their families in this extremely difficult and often overwhelming situation, which is death. Young medical students should be educated in this matter from the moment they choose their future career path. The study program should be full of issues related to thanatology, not only those falling within the scope of forensic medicine. Discussing the process of death should go beyond its physiology, it should also focus on its psychological and spiritual aspect. These are the most important for a dying person and their relatives.

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THE ETHICAL ASPECT OF MEDICINE



Different outlook for the cornea- donation of new possibilities

Aleksandra Jany¹, Urszula Nowak¹, Jan Gnus^{1,2}, Joanna Bogusławska²,
Agnieszka Kowal-Lange², Magdalena Targońska², Anna Kurzelewska¹,
Paweł Barnaś¹, Wojciech Borowicz¹, Michał Karaszewski¹

1. Wrocław Medical University
2. Regional Specialist Hospital in Wrocław, Research and Development Center, Wrocław

INTRODUCTION

“ Every organ transplant has its source in the decision of great ethical value, the decision to selflessly donate a part of one's own body for the health and well-being of another. This is what the nobility of this act is about, which is an authentic act of love ”

- John Paul II

The uniqueness of corneal transplantation (keratoplasty) in ophthalmic surgery relies on restoring the quality of vision. For a patient with corneal blindness, life after keratoplasty can be compared to starting a new life. Therefore, allowing starting a new life can be seen as saving another life. An important part of this procedure is the prior professional collection, preparation, and storage of eye tissue for the recipient. Moreover, it was the first solid tissue that was successfully transplanted.

The first mention of the corneal transplantation appeared in the book "Zoonomy" by Erasmus Darwin in 1796.

The pioneering successful transplantation was carried out in Egypt on gazelles' eyes in 1835 by an Irishman Samuel Bigger.

In 1838, Richard Kissam performed the first transplantation using pig cornea. Eduard Konrad Zirm conducted the first successful corneal graft, a full-thickness graft called penetrating keratoplasty on a 45-year old patient with corneal blindness secondary to burn with lye. According to the majority of sources, this surgery took place in 1906 [1].

In Poland, in 1926, this procedure was initiated by Professor Wincenty Majewski, who was working in the Cracow Ophthalmology Clinic [A].

At the beginning of the history of transplantations, it was necessary to perform the cornea implantation immediately after its collection from the donor due to the lack of tissue storage capacity. The popularization of this microsurgical procedure forced to find the way of tissue storing [B]. One of the first methods of their conservation was developed by Vladimir Filatov in the 30s of the XX century [1].

At the same time Ramon Castroviejo introduced to the world of medicine some new tools designed specifically for corneal transplantation. The first eye tissue bank was established in 1944 on the initiative of R. Towney Paton in order to store more and more donated eye tissues. After about twenty years, several banks this type decided to set up the Eye Bank Association of America (EBAA) to standardize the rules and procedures of transplantation interventions [2].

PRESENTATION OF THE ISSUE

The number of corneal transplantations in Poland is estimated at about 1,000 per year, in whole Europe 20,000 and in the USA around 40,000 [2].

According to the Polish law, it is possible to collect corneas from a donor due to the fact that the alleged consent is valid. This means that a person who does not express his or her objection and does not declare his or her decision to the Central Register of Objections during life can be qualified as a potential donor [3].

The National Waiting List includes data of the number of people looking forward to transplantation in Poland. It has an upward trend from 2014 to the current year: in 2014 the number of awaiting patients was 2638, in 2017 - 3046, and in 2020 – 3147 (last update on February 2nd, 2020). In Poland in years 2010- 2019 the number of deceased potential donors oscillates between 639 (2019) and 786 (2012), while the number of actual donors is between 503 (2019) and 615 (2012) [C].

According to the statistical data prepared by FRK HOMOGRAFT Sp. z o.o., the highest number of donations in years 2015-2019 was recorded in the Opolskie and Śląskie Voivodeships. The number of corneal collections in Poland is the highest next to the southwestern border of the country [D].

Currently, there are 27 corneal transplantation centres in Poland with current permission from the Minister of Health. The cities in which they are based include: Białystok, Bydgoszcz, Bytom, Gdańsk, Katowice, Kędzierzyn-Koźle, Kraków, Lublin, Łódź, Olsztyn, Polanica-Zdrój, Rzeszów, Poznań, Sosnowiec, Szczecin, Warsaw and Wrocław. The patients on the urgent list are the most common in Bydgoszcz, Łódź and Lublin [C].

DETAILED EXPLANATION

Qualification the material for collection

The majority of eye tissue institutions accept corneas from donors between the ages of 2 and 75 years. Corneas from donors under two years of age are primarily very flaccid what is making them difficult to collect and transplant. Currently, the optimal time between the moment of death and the procedure of collection is less than 12 hours, however, in some situations, it is also acceptable to collect tissue within 24 hours from death. An important criterion is also the number of endothelial cells, the requested number of which is 2000 cells per mm².

Special attention is also paid to the occurrence of risk factors such as recently performed tattoos, high-risk behaviours, imprisonment, infectious diseases (syphilis, caused by HBV and HCV, Ebola virus, HIV, rabies, bacterial inflammations, leukaemia, lymphomas, Down's syndrome, retinoblastoma and melanomas) [2].

The elimination of these factors is aimed at avoiding the transmission of infectious diseases and malignant tumors [1].

The confirmed diseases that have been transmitted during corneal transplantation are: rabies, hepatitis B, Creutzfeldt-Jakob prion disease, retinoblastoma, bacterial or fungal keratitis and bacterial or fungal inflammation of interior part of eyeball. Potentially transmitted diseases include HIV infection, herpes virus infection and prion diseases [2].

The morphological analysis of collected tissue is based on stating the absence of abnormal cells, deposits, scars, foreign bodies and changes typical for development-related disorders [1].

After the tissue collection, the aspects of serological examination of donor blood, ophthalmological evaluation in the slit lamp and possible results of section are also checked in the tissue bank [D]. The surgeon is the decisive person and ultimately determines whether a given tissue is suitable for transplantation [2].

Corneal collection procedure

At the beginning, the surgical field is prepared in the operating room or dissecting room [D]. Preparation for corneal collection resembles standard preparation for any eye surgery - the first step is to sterilize the skin of the eyelids and the skin around the donor's eye. Then the conjunctival sac should be rinsed several times for 1 minute using the 5% solution of Betadine and the transplantation from a donor is then started. There are two techniques of the procedure: it is possible to collect the entire eyeball or cut out the corneal-sclera petal in situ, which is the preferred method of collection. To excise the petal the 360 degree conjunctival peritomy should be performed near the limbus (which is the most peripheral part of cornea). Then the proper corneal-sclera petal is performed starting up to 5 mm from the limbus. The operator should make a preliminary scleral incision with a knife cut at a 15 degree angle. The operator opens the eye with primal incision and then cuts out the corneal-sclera petal with Castroviejo scissors or a scalpel [1]. The tissue prepared in this way is placed in a glass container filled with corneal storage fluid: Eusol C, Optisol GS or Licorol medium, described by a unique code [1, E].

All these fluids additionally contain antibiotics, antioxidants, antioedematous factors, growth factors, cell membrane nutrients and stabilizers and [1].

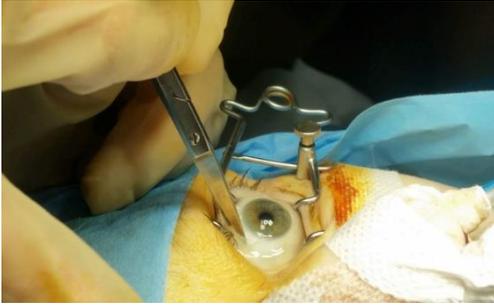
The mentioned media allow to store the tissue for 7 to 14 days at the temperature of +4°C (Eusol C: +2 to +8°C) - the so-called Cold Storage technique [2].

Tissue banks in Poland most willingly use the material for transplantation within 1 to 10-12 days. A situation in which the tissue is used on the 14th day after donation is avoided mainly due to organizational reasons [J].

After the completion of donation, special hoods are put on the donor's eye surface and eyelids are sutured [1]. In this technique, some specialists also practice the removal of the central part of the eyeball and its cleaning by means of sterile compressors from the gas. If a technique is chosen in which the entire eyeball is removed, after peritomy the extraocular muscles are found using a strabismus hook. The surgeon then cuts the visual nerve behind the eyeball and the medial and lateral straight muscles with scissors [1].

The organ is transferred to the humid chamber with added gentamicin and transported to the tissue bank [D].

Next, the operator puts on the prosthesis in the orbit and, as in the first eyelid technique, protects with sutures [1]. An important criterion to qualify the donor tissue or organ for transplantation is a positive serological evaluation of blood serum [F]. The blood is usually obtained from the femoral vein [1].



Peritomy



Corneal-sclera petal incision



Corneal-sclera petal excision



Removal of corneal-sclera petal



Nutrient-conservative liquid



Removal of the central part of the eyeball



Cleaning the central part of the eyeball



Filling with sterile gas compressors



Protecting with sutures



Sutured eyelids

POSTCOLLECTION PROCEDURE IN EYE BANK

Previously, the corneas were stored within the entire eyeball in glass jars at 4 degrees Celsius. This method significantly limited the lifetime of tissue for transplantation. It could be used within 24-48 hours after collection [2].

The procedure of transport and method of cornea transfer to the tissue establishment requires the presence of documentation, which is adequately protected and easy to recover throughout the whole storage period and compliant with data protection regulations. The transport of such tissue requires its placement in the appropriate transport packaging [4].

The centre workers examine the corneal-sclera petal or the whole eyeball in the slit lamp for the presence of scars, foreign bodies, gerontoxon, inappropriate shape, epithelial defects and check the cornea translucency. Macroscopic impurities on the eyeball are removed, then it is sterilized in a 10% solution of Betadine for about 2 minutes and rinsed successively in sodium thiosulphate. A corneal-sclera petal from the full eyeball is obtained by the same method as for *in situ* collection. Corneal flakes prepared and qualified for transplantation in this way are stored on Eusol C medium or the before mentioned media until the moment just before the transplantation operation [1,G].

An important aspect in the final qualification of tissue for grafting is the evaluation of endothelium, which includes such parameters as:

- cell density (standard: 2000/mm² for PK (Penetrating Keratoplasty) and 2500/mm² for DLEK (Deep Lamellar Endothelial Keratoplasty)),
- percentage of hexagonal cells (degree of polymorphism- standard: > 50 %),
- coefficient of variation of cell surface (measure of polymegatism- standard: 20.0-30.0),
- possible presence of abnormal cells, dystrophic changes or sediments.

Images of the endothelium to be evaluated shall be taken at room temperature, under a mirror microscope with its appropriate downward orientation. Other types of microscopes can also be used, such as light, phase-contrast and light field ones. Using hypotonic solutions such as BSS (balanced salt solution) or PBS (phosphate buffered saline) we are able to differentiate normal cells from dead cells. Normal cells due to the osmosis effect will swell, whereas in the abnormal ones this trend will be unnoticeable.

An alternative technique is the use of trypan blue staining (1% dye solution in PBS), which dyes late apoptotic and necrotic cells to blue [H].

Their number is determined as a percentage of the total number of endothelial cells per defined area. Tissue culture extends the time of transplantation storage to 35 days and is therefore very popular in European tissue banks although it requires higher financial resources and is more complicated [2]. In this method, additional microbiological tests are necessary to detect abnormalities, pathogens and confirm its sterility immediately before tissue release. The currently available methods do not allow us to detect the presence of mycosis within less than 10 days and therefore cornea from the tissue culture cannot be released earlier. If undesired results are obtained during corneal endothelial evaluation or after serological tests, the tissue is disqualified and transferred for utilization [1].

SUMMARY

*“ The only way to go forward is to keep raising the standards.
The only measure of success is the effort we have put into achieving it.”*

- Bruce Lee

The procedure of corneal transplantation is therefore fundamental in the world of ophthalmic surgery. It is worth noting that according to the WHO, corneal diseases are one of the main reasons for occurring blindness among people, which is connected with their impairment of functioning in society and everyday life. As reported by the Vision Share Consortium of Eye Banks, over 10 million people are affected by these diseases. Each year, there are between 1.5 and 2 million people whose ulcers or injuries cause blindness [B].

Corneal transplants have the highest percentage of successful surgeries compared to other tissues and organs [K].

With the development of technology and medicine, corneal collection techniques are being improved, which provides an opportunity for more and more people to regain their visual abilities. Corneal transplantation is also the hope for patients with a severe defect in vision refraction; the cornea equals functionally a very strong lens, refracting light rays falling on the eyeball suitably. Thanks to the work of tissue banks, it is possible to significantly extend the usefulness of tissues and organs before handing them over to a person requiring a transplant. However, there are new challenges, which include transplants in children up to the age of 3 years old, as they are currently among the worst prospective [A].

This is a field for the next generations of surgeons and ophthalmologists.

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Mediatization of death

Zofia Hak¹, Klaudia Wijata¹, Jan Gnus^{1,2}, Michał Kasperczak¹, Krzysztof Dorna³, Wojciech Borowicz¹, Jadwiga Kuciel-Lewandowska¹

1. Medical University of Silesian Piasts in Wrocław, Wrocław
2. Research and Development Center, Provincial Specialist Hospital in Wrocław, Wrocław
3. Pontifical Faculty of Theology in Wrocław

INTRODUCTION

Along with the development of technology and cyberspace, all spheres of life have changed [A].

In times of globalization and migration, social bonds and family connections have loosened up. A big part of people have moved to densely populated areas in bigger cities and therefore have gained individuality but also anonymity. Simultaneously they have lost direct support of their loved ones during difficult situations. The mourning process has been moved from private and personal space to the internet. This phenomenon is relatively new and reflects on the need of returning into the collective experience of death [A].

Old rituals are replaced with new ones. Due to widely available internet, people become accustomed to the aspect of dying, identify with mourners or provide support to people who have lost close ones.

Death and funeral can also become a public event and as a result of media accessibility, everyone can turn into a spectator without leaving home.

The purpose of this work is to demonstrate how the customs related to the process of dying and experiencing the death of loved ones have changed as well as how the media enabled the appearance of previously absent behaviour and new commemorative and mourning practises in funeral culture. Mourners stopped experiencing personal tragedies in isolation or in small groups, which in many cases made the process of reconciling the loss and overcoming the pain associated with it harder. Mourning began to appear in the media, thereby taming us with death.

VIRTUAL MOURNING

Losing a loved one is an experience that affects or will affect every human being. Many various cultures handle these situations differently. In Polish tradition, when a person dies, his/her close family hangs an obituary in special cabinets on the street or publishes it in a local newspaper so that all the interested people can take part in funeral rituals.

Currently, publishing the obituary on internet websites and forums as well as Facebook is becoming more and more popular. On the virtual map of activity related to the memory of the dead and experiencing mourning, social media are worth to notice. Most Internet users react to the obituary - "lighting" a virtual candle: [*], (*), * /, [^], and / or writing a few words in order to reminisce or say goodbye to the deceased person [1]. Social media, like Facebook or Instagram, are particularly important in the context of commemorative practices. For example Facebook gives the option of leaving the account 'in memoriam'. As we read in Facebook Help Center, such accounts become places where users can share memories of deceased people. Those profiles do not appear in public spaces e.g. suggestions for People You May Know or birthday reminders. A legacy contact can manage such an account or ask to create one. Any user of Facebook has a possibility to choose the legacy contact. This approved person may, among others: "share a farewell message on your behalf or provide information about the funeral ceremony, respond to new invitations to a group of friends (e.g. from old friends or family members who did not have a Facebook account before)" or change the photo profile and page background image " [B]. A group of friends of such a user becomes at the same time a closed community of mourners, in which everyone has the opportunity to share their thoughts on the In memoriam wall. On the profile wall you can find photos of the current decor of the grave, supplemented with expressions of gratitude, e.g. from parents or siblings of the deceased person, addressed to everyone who has lit candles or gifted fresh flowers in a non virtual world. Another possibility is choosing to delete Facebook accounts after our death - this option is available in account's settings.

In the case of an Instagram profile, a close person must complete the request and provide a death certificate of the deceased person. The other option is to hand over a document that states who is a legal representative of the deceased person or an executor of the will. Both of those actions give the possibility to delete a person's account [C]. Sharing photos from funerals marked with the hashtag #funeral, often with a description commemorating the deceased and with memories related to him, is also a more and more popular phenomenon [2].

Internet users have been joining virtual communities for years to share their experiences and moments of their lives. All this by placing information on more or less important events such as weddings, birthdays, birth of children. Account holders share many aspects of life, both positive and negative, therefore we should not be surprised that more and more obituaries and information about mourning or funerals are also appearing. [3]. Analyzing this kind of posts, it can be stated that in Poland some customs or rituals have not disappeared, but due to the Internet they changed their form and moved into virtual space.

VIRTUAL CEMETERIES

Internet users build virtual equivalents of the surrounding reality, which are a digital representation of the physical world. Virtual cemeteries, more and more often found on the Internet, are such an example. Traditionally, in the cemetery, to commemorate the deceased, his tombstone or monument is visited, the wreath is made and candles are lit.

There are several types of Internet cemeteries - a reflection of a physical cemetery with a cemetery gate, numbered alleys and characteristic memorial iconography. There are also less realistic ones, where their character is determined only by funeral graphics, with no specific virtual location of the burial place.

The user can - most often for a small fee - create an e-tombstone by choosing its pattern from templates, and decorate it with a selected type of flowers, candles, photo albums and inscriptions (the most common ones include birth and death dates, commemorative sentences and the information about real burial sites) [D]. Such an e-gravestone has a similar function as the "in memorium" account on Facebook - it allows sharing memories and photos related to the deceased person. According to Magdalena Gajewska, *"tombstones and cemeteries are a symbolic gate, giving us a chance to meet the dead. Thanks to them, the dead do not disappear completely, and we do not lose them at all"* [4]. Additionally, visitors to a given tombstone can share their private memory about the deceased person or buy an additional candle or flowers. Both virtual necropolises and social networking sites are governed by the principle of equality. Regardless of gender, education or occupation - everyone can have their own profile / gravestone which they will cherish. The rules for maintaining such an account are identical for all users.

Virtual cemeteries for animals are also a topic worth mentioning. The spectrum of possibilities is the same as in the case of a virtual cemetery for people - from designing the grave to choosing candles and flowers. In addition to published photos, pet owners also share

memories related to a dead animal. Several animal cemeteries can be distinguished. Besides the general animal cemetery, users can visit online graveyards for reptiles, rodents, horses, cats, monkeys, insects, amphibians, dogs, birds, fishes, other mammals, other animals, VIPs [5]. Very often people can "bury" their pet next to a relative's grave, which virtually does not cause as much controversy as in the case of a traditional cemetery.

ONLINE PRESENCE AT FUNERALS

With the spread of new media, changes in the sphere of funeral services can also be seen. New solutions are proposed, such as the ability to record funeral ceremonies and their transmission via the Internet or television.

The funerals of famous people are currently the most popular. According to American anthropologist John Caughey, people tend to treat their idols as people who are emotionally close to them. This often results in a very strong reaction to the death of a famous person, as if he/she belonged to one's close family [6]. We often spend more time (by watching subsequent episodes) with people known from TV series stars than with family members.

What can be described as the phenomenon of 'media events' is gathering a large number of people in front of a television set when the event is broadcast live [7]. The whole country or even the world freezes in front of TV. Some people, in order to emphasize the importance of the event, dress up elegant and then sit down to watch the broadcast.

In Poland, one of the biggest media events was the funeral of Pope John Paul II, which was watched by 94% of Poles and about 2 billion people in the world [8]. As many as 47% of Poles surveyed believed that the experience of this event will change people for the better [F]. Professor Godzic points out that television through 'media events; can change our psyche to such an extent that we get the impression that after participating in the ceremony X "the world will no longer be the same as before" or that the event Y changed our thinking about life. We could hear similar comments after the funeral of the Pope [8]. Interestingly, he also points out that those kinds of shows arise in widespread acceptance. Sometimes there even is pressure from society which wants information and discussions about the deceased.

Media events are peculiar monuments in electronic form and their joint experience strengthens interpersonal relationships and a sense of collective identity, including mutual understanding. Family bonds and friendships are renewed. The solemn broadcast integrates society - people call each other, look for someone with whom they can talk about what they saw on TV, arrange to watch the event together at home or in the bar [7].

The funeral of Michael Jackson, the wedding of Prince Charles or the travels of John Paul II are examples of such celebrations. Television broadcasts of those events were in a sublime mood, and viewers were involved in "distance participation": "Such tele-participation in the ceremony is possible only through electronic devices and is an example of a special modern form of ritual"[9]. There is no evidence, however, for funerals of famous people to be viewed by more people than, for example, royal marriage.

Not only great personalities can have funeral broadcasts online. The so-called 'family distance funeral service' was created, which is offered by some funeral homes in Poland. When we are unable to arrive at the ceremonies (e.g. when we live on another continent, or are unable to attend due to healthy reasons), an employee of a funeral company records the ceremonies or connects with us directly. Most often, the recording of such a ceremony expires after 7 days and the cost of 20 minutes of transmission is PLN 60 [5]. In addition, there is also the option of condoling the closest relatives of the deceased and writing a short memoir text or placing the funeral wreath with personalised ribbon.

PLAN YOUR FUNERAL ONLINE

This is a relatively new service on the Polish market because there is still a dominant belief that funeral organization is a family matter. Frequent pre-planning can be understood as purchasing by the elderly or sick person a place in the cemetery and possibly putting up a tombstone. However, this usually results with ambivalence from the family, who treats it as a "bad omen" [E].

In Western countries, the pre-funeral planning service is becoming increasingly popular. Due to it, we can design our perfect burial without leaving it to the family burdened in sadness and despair [5]. The customer can choose every detail of the funeral - from choosing a casket and music to decisions about cremation, burial or donation of the body for scientific purposes [G]. He/She can also be sure that the ceremony will proceed according to his/her preferences. In the United States, DIGNITY PLANNER also has a variety of funeral themed packages; *Fiesta*, *Gardener* or *Golfe*. According to the description of the *Fiesta* package, we read: Spicy dishes, refreshing drinks and vibrant music are the hallmarks of a fiesta. Hang a piñata full of your favorite memories and ask friends and family to each read one — reflecting on memories that made your life colorful [H]. The company informs that each of the plans can be individually adapted to the customer's preferences. We can purchase and personalize them lying in bed with a laptop, we do not have to go to the funeral parlor to determine all the details.

SUMMARY

Currently, to experience mourning, we do not have to leave the house - an access to TV and computer is all what is needed. This is a great convenience for people who are ill and who cannot attend a given event. People can "meet" virtually and support each other in difficult times. In the era of media, nobody is forced to grieve alone. More and more, new, computerized funeral rituals are moving into everyday life, no longer being understood as "extravagance".

A modern manifestation of mourning, as well as a reaction to news of someone's death is very often a picture; on discussion groups - a graphic sign of a candle; in the space of e-cemeteries also a photo or a film from the life of the deceased. In a traditional cemetery there is very little space for stories about people who are gone, and its possible forms are limited. In contrast, the virtual cemetery gives the opportunity to talk about the decedent using the language of contemporary life - not only words but also visuals.

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Funeral rituals and approaches to death over the past hundred years in Poland

Zuzanna Mazurkiewicz¹, Zuzanna Pilszyk¹, Zofia Hak¹

Student Scientific Association of Adult Allergology, Wrocław Medical University

INTRODUCTION

Various funeral rites have been practiced for thousands of years to help to cope with death, which has always been a sorrowful experience. Their aim was to help people accept the death of their beloved ones. These rituals have changed along with civilization development but their function remains the same. Over the last hundred years, the evolution of funeral rites has occurred faster and affected many areas of life. The reason for such rapid changes were historical events, intensive technological development, and significant changes in lifestyle and family structures. The aim of this paper is to analyze and discuss the impact of the above factors on changes in funeral rites in Poland.

APPROACH TO DEATH, TABOOISATION AND THE BEGINNING OF DETABOOISATION

The 20th and the beginning of the 21st century are considered as a period of great taboo when it comes to the subject of death [1]. On the other hand, a new approach to death has appeared, which is manifested as a growing demand on the right to euthanasia [2]. According to the research carried out in 2009 by the Centre for Public Opinion Research (CBOS), 48% of respondents believe that doctors should fulfill the will of those who suffer by delivering lethal agents. However, 39% of the respondents disagreed with this judgment and 13% did not have an opinion on the subject [A]. Nowadays in Western culture death is mainly associated with severe fear and grief [2]. Old age is not identified as a glory moment of one's culmination of life, rather as infirmity, and burden for society [3].

Nowadays, Poland is a country representing the western approach to the subject of death. It should be noted, that in eastern European culture death is regarded differently - it represents the possibility to free ourselves from the suffering, ailments, difficulties, and anxieties [4]. It is worth remembering that many religions, including Christianity, proclaims that death is not the end of human existence, but the transition from biological to spiritual life [5]. Yet, 93,5% of Polish citizens over 16 years identify themselves with religious institutions, of which 91,9% belong to the Roman Catholic Church. About $\frac{3}{5}$ of respondents declare themselves as believers [B]. Catholic Church denies the practice of euthanasia and the use of persistent therapy [2].

So-called persistent therapy is an important issue. It stands for the use of medical procedures to support the life functions of the terminally ill. It involves suffering, prolongs dying, or even violates a patient's dignity. Persistent therapy does not include basic care, relief of pain, feeding, and hydration, as long as they serve the patient [6]. Justification for renouncement of these actions is assurance of natural dignified death. It is not meant to shorten a patient's life but to avoid prolonging dying and delaying imminent death [7]. According to the Code of Medical Ethics, in terminal states a doctor is not obligated to conduct resuscitation, persistent therapy or use of emergency measures.

Another important subject is the taboo of death, which means holding back emotions, and hiding signs of mourning. Term "Taboo" stands for something that should not be talked or written about [C]. Goeffrey Gorer, British sociologist and anthropologist, even described the death as "new pornography" [8]. The causes of taboos of the death includes: secularization of society, changes in structure of family, urbanization and the development of the family [9]. In addition, the cult of carnality was born in the 20th century: for society, a beautiful and healthy body became supreme value, displacing the image of illness and death. Changes in family structures include disappearance of multi-generational families [5].

Along with these changes, a natural way of taking care of elders vanishes, as well as the chance to experience the death of relatives [10]. In the past, youth took care of the elders or ill, so eventually, they were close to their death [5]. The growing distance between the subject of death and younger generations, which took place in the mid-20th century, reduced that phenomenon [1], and families sought help in special institutions [4]. The matter of death has changed from family events to medical cases [11]. The twentieth century brought death free from suffering, but it was transferred to hospital rooms [2]. In the 1970s 62,9% of deaths took place at home when only

35,9% in hospitals. In 2015 the situation reversed and 35,3% of deaths occurred at home, and 51,5% in hospital [D].

According to data from 2012, 66% of citizens of Poland would rather die at home, 8% in hospital, and 4% elsewhere while 22% have no opinion on this subject [12]. Families often lack adequate conditions, equipment, or do not have nursing skills or experience with dying. Taking care of severely ill, disabled, or dying requires time, commitment, and money and might lower the life quality of other family members. In addition, it carries an emotional burden. In such cases, the support offered by professional assistance, i.e. hospitals, hospices, and palliative care departments becomes crucial [11].

The 20th century was the time of significant progress in the field of medicine. It had a huge impact on the way people think about death. At the turn of the 50s and 60s, we started to think more about the health of the elders. The second half of the 20th century was marked by the fight against the main causes of death of old people, such as heart disease, cancer, stroke, and respiratory disease. It was also the time when the number of people over 65 years old greatly increased. What is more, science, medicine, and industry started to concentrate on older people [1]. Diseases that were considered terminal began to be treated [11], demise was no longer considered a necessity, but as the natural order of things [1] and eventually, death became an anomaly [8]. Development of medicine and technology, the possibility of delaying or accelerating death [3], depending on the will of doctors, the wealth of family or country [4], can give the false impression of immortality [2]. Scientific progress has contributed to extending human life and improving its quality but also caused self-alienation [8]. Modern man is a man that dies in solitude [2,4].

Medical technology is focused not on the needs of the dying, but on restoring their health [10]. The development of medicine had an impact on the definition of death as well. The classical definition says that death is an irreversible cessation of breathing, and cardiovascular system functions. “New” definition specifies death as the irreversible cessation of brain function. The latest definition declares that death is found when the brain stem function ceases irreversibly [12].

World War I and II have also influenced the process of death tabooisation. They accelerated it as people of the post-war civilization wanted to erase Wars from their memory [1]. The trauma after World War II involved remembering the deaths of millions and the close experience of illness and dying in the war period. The denial of difficult memories was a defense mechanism [13].

Macabre, bloody, unreal deaths, ubiquitous in pop culture and deaths caused by accidents, wars, terrorist attacks or homicides about which mass media inform everyday are the exception of tabooisation [1,4].

Despite the clear tendency of social withdrawal from the subject of death, statistics show that many Polish people think about passing away every day. According to CBOS research from 2019, 67% of Polish people think about their death [E].

The end of the 20th century heralded upcoming changes of social attitudes towards death. May be acknowledged, that as a society we started a process of opening up to the problem of death and dying [11]. The process of detabooisation has slowly begun and it is still going on. Mass media more often show the topic of dying and it forces people to confront their own mortality. Reports on death which are showing also its hardships and suffering are the proof of social changes taking place. Ill people go online and start blogs in which they write about their struggles with death. Publishing on the Internet a journal of a person consciously approaching the end of life may have therapeutic features [14]. Ill person reports his or her experience and shares thoughts. It is the way of showing that death does not have to be a taboo [11]. This type of report becomes more and more popular. Both sides benefit from them as the sick one and the reader slowly become accustomed to the nature of dying. More often they even make contact with each other. Sick people often suffer from isolation associated with the progression of the disease and thanks to this way of communication they get a sense of closeness with another person. Attempting to establish a relationship results from the willingness to share and support in the struggle related to the disease and the inevitable end of life. It is a completely new attitude compared to the past ones, where death was a secret and patients were not informed by their families or doctors about their health [3].

Previously mentioned, almost unreal pictures of death present in pop culture and those presented by mass media also have their share in detabooisation as they show tangibility of death, provoke reflection and participation in the drama of passing. The reason to reflect on life, its sudden or even unexpected end are also funerals of famous people transmitted by television stations [15].

A big step towards the detabooisation of death [1] and slow re-opening the consciousness of modern society to the problem of death is hospice movement [11]. Hospice is the facility for

incurably ill people [F]. The first such institution was St. Christopher Hospice founded in London by Cicley Saunders in 1967 [11].

Hospice activity included caring about spiritual, emotional and health needs of incurably ill patients, regardless of their religion, origin or social status. Hospice does not deny death, it does not treat it as a scandal nor it promises illusory recovery to people terminally ill. This is a place of full focus on the dying person [2] which creates maximum comfort for dying ones to the last days of their lives [11].

In Poland, the precursor of the hospice movement was Hanna Chrzanowska. In 1981 in Cracow came into being „*Towarzystwo Przyjaciół Chorych – Hospicjum*” (The society of the friends of the ill ones – a hospice) which initiated the construction of the first stationary hospice in Poland. The palliative and hospice care officially became a part of the Polish health care in 1991 [1].

FUNERAL RITUALS IN THE LAST HUNDRED YEARS IN POLAND

Changes in the perception of death over the past hundred years show the transformation in funeral rituals. For centuries, people tried to get used to the experience of the death of their family members and friends by creating a number of rituals, beliefs and superstitions. Representatives of folk culture were more accustomed to death and more prepared for it [5].

Funeral rituals help in surviving the loss, colliding with it and accepting it [4].

Polish funeral rituals are the result of religion, old beliefs and eschatological imaginations [16]. Among Polish people lots of archaic beliefs and practices can be found [17] but archaic ways of grieving the dead ones are actually only in small communities [12].

Funerals have a hygienic-sanitary function, it helps the dead person to pass to the other side/nonentity/other form of existence, it soothes the shock of the close ones caused by death and gives the opportunity of setting the emotions free [9]. Old but still in use death and funeral rituals include: covering the mirrors and television (so the dead one will not look at himself and stay among alive) [17,18], turning chairs upside down (it prevents the soul of the dead one from staying in the house) [17] and stopping the clock in the hour of death [9]. The coffin was placed on the table in the house [16]. Watch by the corpse lasted for three days and was accompanied by prayer [9,18]. Deceased one was washed and dressed at home [9]. Seniors were dressed in black, young

people wore white and unmarried women wore wedding gowns. Shoes were an important element. It was believed that the dead one cannot go to heaven barefoot. Dead person was holding in hand a rosary, a picture of a saint, a cross or a prayer book [9,17]. Money was placed in the coffin [18] and some things of everyday use such as tissue, glasses or toys for children [17]. Equipping the dead one with items is a custom which occurred due to the belief that human life continues after death in another dimension [16]. Coffin with the deceased was taken out of the house in a way that the dead one would have his feet in the front. Another custom was hitting the coffin against the threshold of the house three times [9,16,17]. The coffin was carried to the graveyard by a horse-drawn carriage [9]. It was accompanied by a funeral procession. Procession with the coffin stopped by the small chapel or cross, it was a symbolic way of the dead one saying goodbye to the village [16]. Deceased were buried in the graveyard, the place of prayer and reflection [3]. It was forbidden for relatives to throw soil on the coffin to say goodbye to the dead person. It was thought that if a relative did this, he will soon die too [19]. The period of mourning included prohibition of singing, dancing and going to the parties and weddings [16]. Black clothes worn by relatives indicate the mourning [9]. A catholic tradition practiced till today is receiving the sacrament of the last anointing by the dying person [2]. Priests, as representatives of The Catholic Church, played an important role in the transformation of funeral customs, shortening customary practices as they were not a catholic tradition [9]. An important funeral tradition of the 20th century was funeral photography. It was popularized in the interwar period. Its function was preserving the memory of dead ones, recording their last moments on earth and enabling to show the dead person and the funeral to people who were not present. There was a tradition of a collective coffin portrait which presented relatives, neighbours and friends of the deceased gathered around his body lying in the open coffin. The tradition of photographing the dead ones was still practiced in the 1960s [17].

MODERN POLISH FUNERAL

What we feel, think and believe about death and which values we attribute to it, largely translates into the ways in which societies and families part with their members. CBOS [E] research from 2019, as many as 50% of respondents said that the body of the deceased can be cremated. In 1994, only 37% of respondents said alike. In Poland, cremation as a funeral custom has been around for a relatively short time. The Catholic Church allowed the cremation of the dead

rather recently, only in 1963 [G]. Over time, the approach of the Polish episcopate has changed and nowadays both forms - traditional burial as well as cremation, are accepted. This is the main reason for changing the approach of Polish society in the matter of funeral rites. However, the church recommends the traditional form of interment that has been conducted until the debate but it does not formulate an absolute prohibition on burning the bodies of the deceased. People who actively practice religion (visiting the temple at least three times a week) are in favor of traditional burial. Some of the elderly refuse cremation, among other things, due to experiences during World War II.

The dynamically developing funeral industry offers many different burial forms: from traditional, cremation and burial in the urn, to unusual forms, such as processing of ashes into diamonds, sending the ashes in space in a special metal capsule [20].

In Poland, unusual forms of burial make up a very small percentage, while in Western countries they are becoming more and more popular.

Along with urbanization and the increase in the affluence of society, restaurants specializing in organizing funeral feasts were opened [21].

According to CBOS, in 2019 as much as 82% of such events were organized in restaurants, while only 17% in the home of the deceased. Often, the family not organizing the funeral is associated with socioeconomic status and poor education. Studies show [E] that the cost of the entire funeral and funeral feast, regardless of social status and earnings, is similar.

The funeral ceremony itself currently goes according to the tradition, while the symbolism around the funeral rite has changed. Compared to 20th century rituals, the “funeral hero” has changed - previously it has been a photo of the deceased while nowadays mourners have become one. Few pictures portray a deceased person in a casket, the photographs mainly show family and funeral procession, which once were only the background for the funeral. Emotions experienced by loved ones come to the fore, not death itself. In addition, the majority of society wants to remember the deceased in his “best years” of life and thus do not decide to posthumously consolidate the image of the deceased. Also for this reason, the number of caskets closed during vigil and service is increasing.

It is worth mentioning that a new service has recently appeared in Poland - an online funeral transmission. The transmission is conducted by the employees of the funeral parlor. It contains the streaming of the entire funeral service with a casket being laid inside the grave [22] Later such

transmissions are placed on servers to which access is only available to the loved ones [20]. This is an example of how the funeral service becomes easier to approach in times of common migration. One who cannot attend a funeral service, can easily watch it online or even buy on the Internet a funeral wreath or a snitch from the funeral house which later will be placed on the grave by employees of the funeral parlor.

We have now departed from a large number of symbols and funeral superstitions. Despite this, in some circles new funeral rituals have developed, e.g. placing charged cell phones in a casket, which are supposed to be a rescue in the event of being buried alive. In the case of children's burial, however, there is a tradition of putting toys, rattles, and teddy bears into the casket. For newborns - blankets, dummies, or things that a living child of this age needs [18]. Very often, specific coffin portraits of dead newborns or children are made.

CONCLUSION

Not only funeral rites have changed over the years, but also the approach to death itself. Many factors influence this change. One of them is technological progress, which, translating into the development of medicine, makes it possible to extend life, slowly removing the image of the dying process from social consciousness. Even issues as prosaic as the long distances separating family members turned out to be important, which was the impetus for the creation of a funeral transmission service. The combination of all components gives the effect of tabooing death, which is the main reason for changes in the development of funeral culture. On the other hand, we can observe the reverse tendencies, which aim to re-familiarize society with death. The development of blogs and other forms of online journals reporting the fight against a serious illness, public funerals of famous people, or finally the most important, i.e. the development and popularization of the hospices. This allows us to conclude that, like every sphere of life, also death and saying goodbye to our loved one, must maintain a balance and sacred–profane dichotomy. Partial departure from ancient rites deprives death of its sacral dimension and transfers it to the taboo sphere. At the same time, however, many new habits reintroduce the topic of dying into social consciousness. Such activities create a new balance, and thus a new funeral culture, whose further changes and development will certainly be observed along with the progressing development of civilization.

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Knowledge among medicine students from Poland about euthanasia

Weronika Mazurek¹, Monika Hejnowicz¹, Karolina Radzikowska¹,
Sebastian Fedorowicz¹, Anna Kolcz¹, Jan Gnus^{1,2}, Krzysztof Krzysztof³

1. Wrocław Medical University
2. Regional Specialist Hospital in Wrocław, Research and Development Center, Wrocław
3. Pontifical Faculty of Theology in Wrocław

INTRODUCTION

Euthanasia, also called mercy killing, is an act of painlessly putting to death persons suffering from a painful and incurable disease or incapacitating physical disorder [A].

It is a complex legal challenge but also an ethical one. European Union does not put restrictions on its members in this matter. Therefore laws differ from country to country. According to article 150 of *Penal Code* euthanasia is illegal in Poland with penalty form 3 months up to 5 years of imprisonment [1].

That is the only legal document that regulates euthanasia. According to police statistics, in the years 1999 – 2017, a total of 4 cases of these criminal offenses were found [B].

On the contrary, in the Netherlands, Belgium, and Luxembourg euthanasia is legal under several conditions [2,3,4], with Belgium being the country that allows euthanizing children.

Although the term euthanasia and assisted suicide are often confused with the cessation of persistent therapy and they are often mentioned together in public debate, we would like to focus on euthanasia. There is no doubt that it is both a controversial and interesting topic that will be increasingly discussed. Therefore, a graduate of a medical university should have appropriate knowledge including legal aspects of this subject.

AIM

The study aims to compare the knowledge of students of Polish medical universities about euthanasia.

MATERIALS AND METHODS

The research group consisted of 264 students of medicine from several universities in Poland. 41.7% of the respondents were students of the Wroclaw Medical University, 10.4% of the Medical University of Lodz, 7.6% of the Medical University of Warsaw, 6.1% of the Medical University of Lublin, 4.5% of the Poznan University of Medical Sciences. The remaining 29.7% were students of other medical universities.

The majority of the respondents were students of the second year – 28.8%, the second in terms of amount were the third-year students – 26.1%, then 5th year – 11.4%, 4th year – 15.9% and 1st year – 9.8%, 6th year- 6.8%, the 1st-year-resident – 1.2%. The research was carried out using the original questionnaire created by the authors, containing 5 questions regarding the knowledge of students on the euthanasia in Poland. The survey was conducted online using Google Forms. The criterion for inclusion in the survey was holding the status of a student of faculty of medicine in the medical university of Poland.

RESULTS

This part of the work will present the results of the survey.

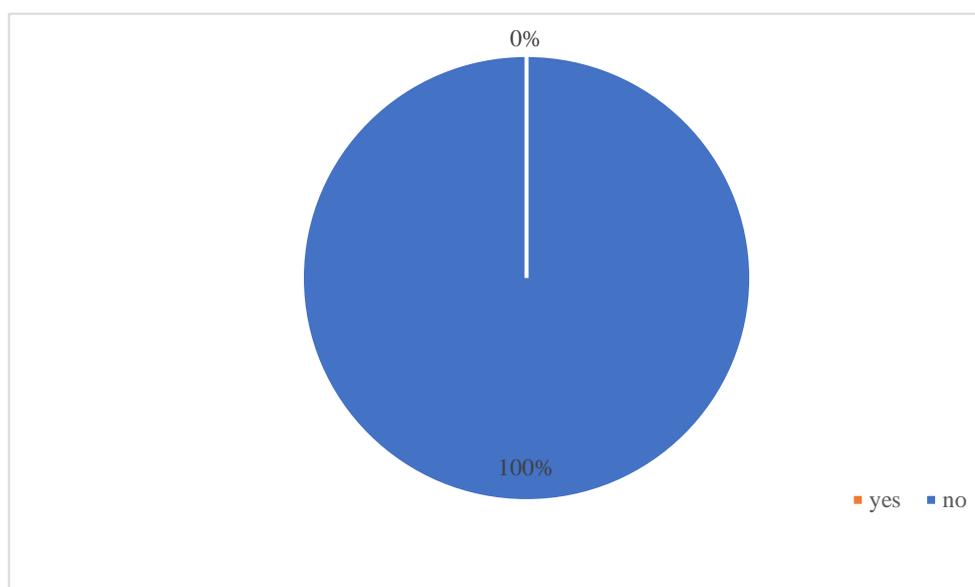


Figure 1. Students' knowledge about the legality of euthanasia in Poland

Figure 1. shows the students' answers to the question of whether euthanasia in Poland is legal. 100% of the respondents answered correctly indicating that euthanasia is not legal in Poland.

No student gave an incorrect answer.

In the survey we asked students what the definition of euthanasia is. Figure 2. Shows that most of the respondents, 54.9%, answered correctly that euthanasia is defined as giving the patient, by a doctor or another person, lethal means on a voluntary and informed request in an intended manner. 4.5% pointed out that discontinuation to carry out any further activities that could extend the life of the terminally ill and suffering person as the definition, which is an incorrect answer. A wrong answer is also both of the above, which pointed out 40.20% of the respondents.

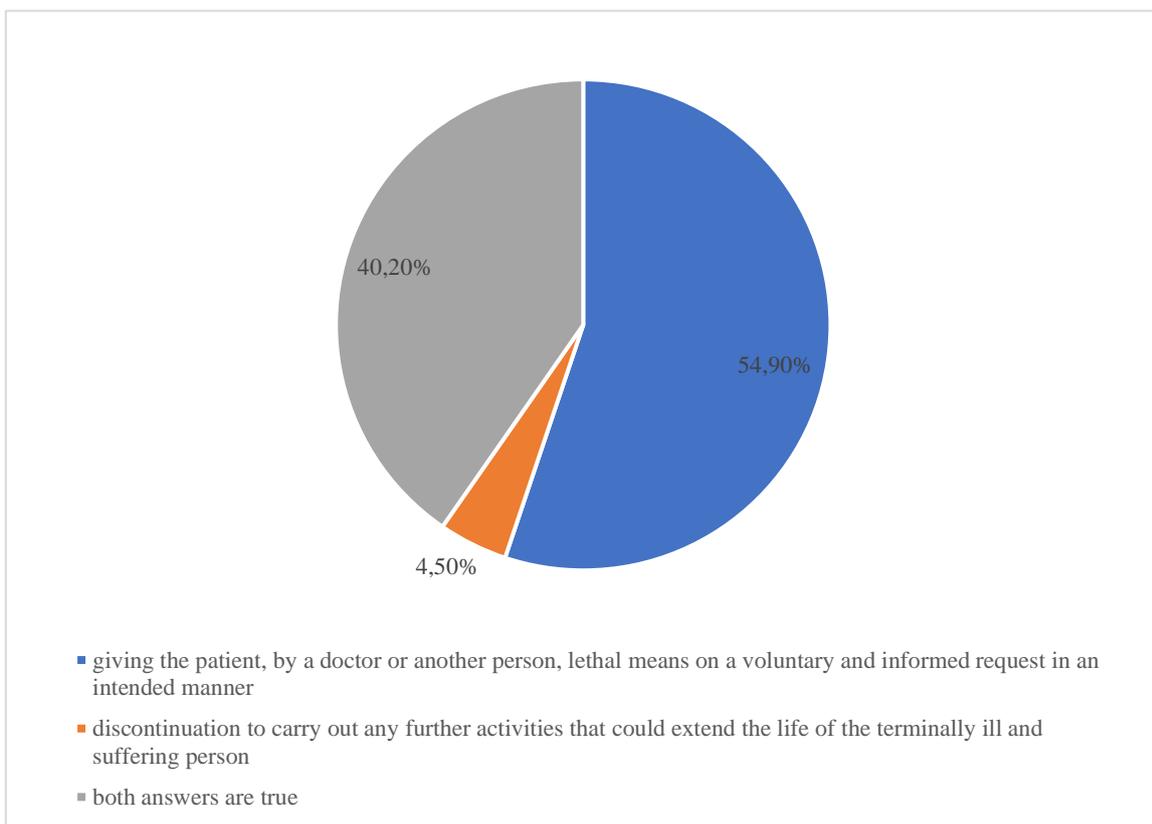


Figure 2. Students' knowledge about the definition of euthanasia

In the further part of the study we asked respondents to point out in which European countries euthanasia is legally allowed. The correct answers are Luxembourg, Netherlands and Belgium which were pointed out by 48.7%, 91.3% and 66.2% of respondents respectively as it is shown in figure 3. The other answers are incorrect and these are: Austria with 23.2% of respondents, Malta – 8.4%, Greece – 1.5% and Poland with 0.8%.

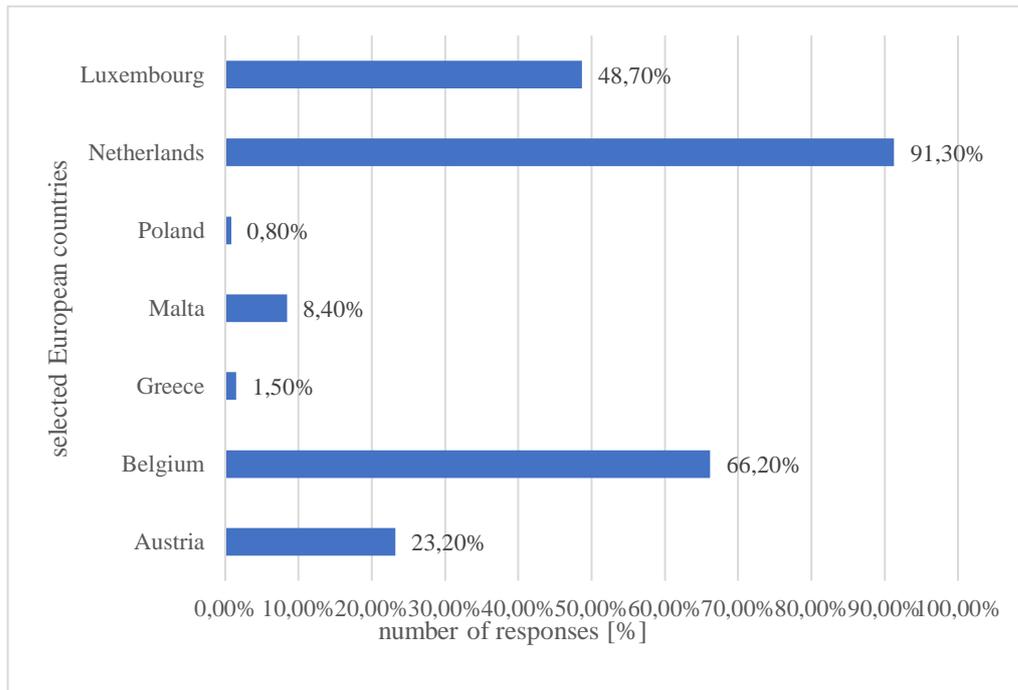


Figure 3. Students' knowledge about the legality of euthanasia in Europe

In the next question we wanted to see if the students know what is the punishment for euthanasia in Poland. As can be seen in figure 4., 33.7% of the respondents pointed out correctly from 3 months to 5 years as the answer. In total 62.9% of respondents gave the wrong answer: 57.6% answered from 3 to 15 years and 5.3% answered from 20 to 50 years. 3.4% admitted that they didn't know.

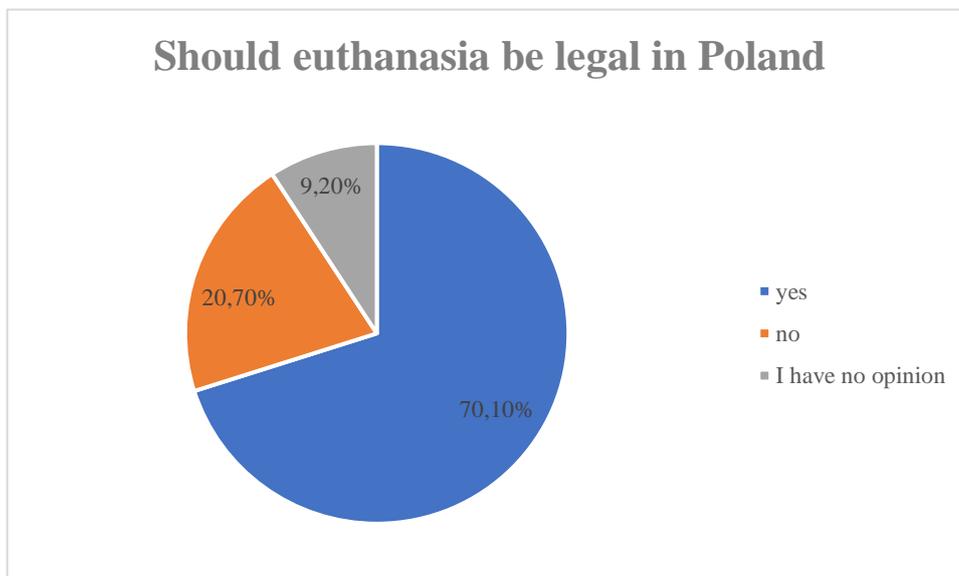
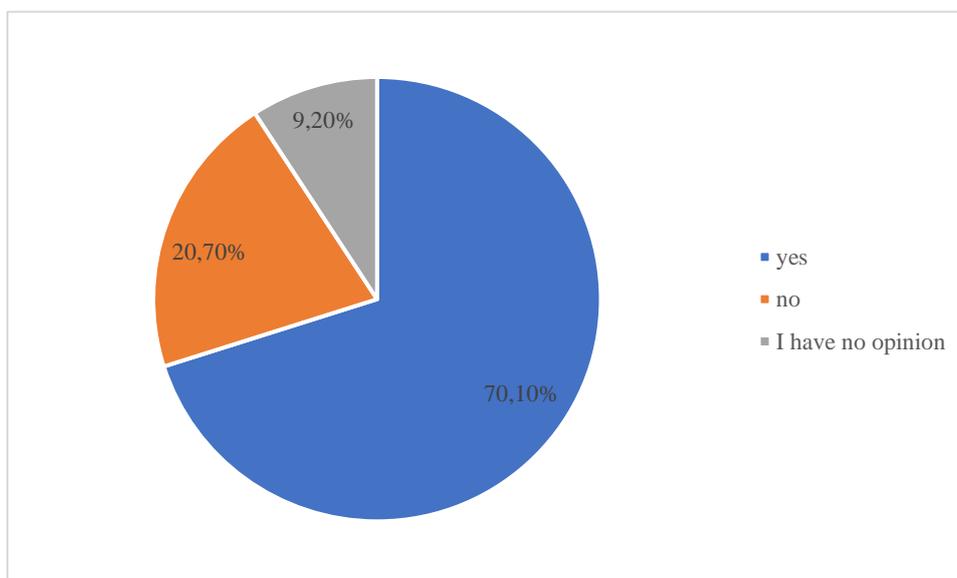


Figure 4. Students' knowledge about the penalty of euthanasia in Poland

In the penultimate question we asked the students if they think that euthanasia in Poland should be legal. 70.1% of the respondents said yes, it should be legal. 20.7% answered no, and 9.2% admitted that they have no opinion.



In the last question we asked students, who answered “no” in the previous question, why they think euthanasia should remain illegal in Poland. 44 students gave a written answer and that is 16.6% of the respondents. We will refer to the answers in the discussion.

Figure 5. Students’ opinion on whether euthanasia in Poland should be legal

DISCUSSION

The subject of euthanasia and medically assisted suicide is a very controversial topic all over the world. Nowadays, thanks to constant medical progress, life expectancy is longer and the quality of life is better. On the other hand, this progress also enables us to artificially shorten life. Furthermore, the amount of palliative illnesses is increasing. Given such reality, the questions regarding the end of life have been and still are the topic of public debate.

Our study aimed to examine the level of knowledge of students of medical universities in Poland regarding the legal aspects of euthanasia. The study covered 264 students from various medical universities in Poland.

The results of the study showed that all of the respondents (100%) answered correctly about euthanasia’s legal status in Poland. That shows a completely satisfactory level of knowledge in this area.

In the next question we wanted to check the students' knowledge on the definition of euthanasia. Over half of the respondents, that is 54.9%, pointed out correctly. The remaining 45.1% gave the wrong answer. On one hand, this shows acceptable awareness of students. On the other hand, as many as 40.2% of the students answered that euthanasia is both the administration of a lethal agent and the cessation of persistent therapy. The difference between these two is very important and essential for doctors, especially for palliative therapy specialists. There is no exact definition of cessation of persistent therapy in Polish law [C]. However indirect reference to persistent therapy can be found in *the Code of Medical Ethics* [5] and *Act of 6 November 2008* [6]. The first one states:

“1. In terminal states, a physician is not required to undertake and conduct resuscitation or persistent therapy and emergency measures.

2. The decision to stop resuscitation belongs to the doctor and is related to the assessment of therapeutic opportunities.”

In the *Act of 6 November 2008* is stated:

“1. The patient has the right to respect intimacy and dignity, in particular when providing health services to him.

2. The right to dignity also includes the right to die in peace and dignity. A patient in the terminal state has the right to health services providing relief of pain and another suffering.” This “right to die in peace and dignity” indicates a ban on taking medical actions against the patient that would violate his or her dignity in the terminal phase. Bp. Dr. hab. Józef Wróbel, the chairman of *Team of Experts of the Polish Episcopal Conference on Bioethics*, says that “Resignation from futile therapy does not mean leaving the patient and cannot be equated with euthanasia” [D]. Moreover, the above-mentioned *Team* issued a document confirming this position [D].

Students as future physicians should know the differences between euthanasia and cessation of persistent therapy, especially if they have different legal consequences. Therefore, we believe that students' knowledge is not satisfactory enough and should be deepened and broadened in this matter.

Another issue that we addressed in the survey is students' knowledge about the legality of euthanasia in Europe. Most respondents correctly identified countries in which euthanasia is legal and that is accordingly: Luxembourg – 48.7%, Netherlands – 91.3% and Belgium – 66.2%. The incorrect answer that got the most votes (23.2%) was Austria. There euthanasia is illegal and is considered a criminal offense according to article 77 of the Austrian Criminal Code [7], with sentences ranging from 6 months to 5 years of imprisonment.

As most of the respondents indicated correctly, in Belgium, Luxembourg and the Netherlands, euthanasia and medical assistance in suicide are legally allowed for several years based on legal acts which are respectively: The Belgian Act on Euthanasia of May, 28th 2002 [2], the Law of March 16, 2009 [3] and Termination of Life on Request and Assisted Suicide Act which went to effect of April 1, 2002 [4]. Those legal acts are a result of public discussion by media about euthanasia in different countries in the last years [E, F, G]. However it is important to remember it is euthanasia under specific conditions [8,9,10] that are clearly stated in these three legal acts [2,3,4]. They are to ensure the transparency and control of medical procedures and to consider the possibility to end life voluntarily. These conditions are the following:

1. The request must be voluntary, without any external pressure, carefully considered and repeated. Therefore the patient must be conscious at the time of making the request. The patient must be in futile condition and unbearable pain, with no prospects of improvement. Moreover the physician along with the patient has concluded that there is no reasonable alternative solution. And this conclusion is based on several conversations between the patient and the doctor spread out over a reasonable period.
2. The physician must consult another, fully independent physician. This consulted physician reviews the patient's medical record, conducts a physical examination and must be sure of the patient's serious and incurable character of the disorder. Based on their findings the consulted physician makes a report of the case.
3. The request must be in writing.
4. All formulated requests along with any actions undertaken by an attending and consulted physician must be regularly noted in the patient's medical record.
5. Each decision to carry out euthanasia or assisted suicide is ultimately taken by a commission especially created for this, which includes doctors of medicine, legal experts/practicing lawyers and experts on ethical or moral issues.

The laws in these three countries differ regarding the patient's age and legal capacity. In Luxembourg only a patient who is of legal age, with full legal capacity, and conscious at the time of application may request euthanasia. "Minors, persons of legal age under guardianship or protection, and legally incapable persons may not legally request euthanasia or assisted suicide, nor may their parents, guardians or trustees make such a request on their behalf" [H]. In the Netherlands, the law is less restrictive in this matter. Underage patients, between 12-18 years old, with full capability of making a reasonable decision, may request euthanasia. The attending physician may comply with the patient's request under these conditions: if the patient

is from 16 to 18 years old – after consultation with his/hers parents or legal guardian; or if the patient is from 12 to 16 years old – after parents/legal guardian’s permission to do so [4]. Belgium has removed all reference to an age restriction [11]. On February 13rd 2014 the lower house of the Belgian legislature voted in favor of an amendment to euthanasia law [I]. Under this new law, a terminally ill child, regardless of age, can request medically assisted suicide. Before fulfilling the child’s request, the child's parents must give their consent and the capacity of the child's understanding and judgment must be verified by a psychologist or child psychiatrist. Additionally, euthanasia is legal to some extent in Switzerland. Assistance in suicide in some cases takes place in other countries like Germany or Finland [J].

Knowledge about the health care system in other countries and the possibility of comparing them with the Polish one is broadening and mind-opening and allows improvement. Therefore, both students and doctors should constantly expand their knowledge about European standards.

The following question was about the punishment by polish law. Most of the responders, 57.6%, have given the wrong answer, saying the expected punishment is between 3 to 15 years of imprisonment. According to article 150 of *Penal Code* punishment for euthanasia is from 3 months to 15 years of imprisonment [1]. Ignorance of the laws is dangerous and knowledge is the best protection. Protection from possible legal trial and imprisonment especially for doctors, whose profession has a lot of responsibility. Therefore, the lack of students' knowledge in this sector is alarming. It suggests inadequate knowledge of polish medical students about the law they have to abide by.

The last questions involved students’ position on euthanasia. Most of the responders, 70.1%, expressed willingness of changing the law to be more accepting of euthanasia. They believe that it could help palliative patients to pass away and remain dignity and comfort in the last moments of life, before the pain will become insufferable. On the other hand, responders’ most common arguments against changing euthanasia law were that it is against their and most of society’s religious beliefs. Christianity has a significant role in the lives of many people in Poland. Law against one of the principles of religion would be met with public backlash and scrutiny. Furthermore, students said that in modern society, with advanced medicine, palliative patients should be offered therapy not euthanasia. They indicate that pain-relieving drugs or different types of pain-relieving therapy should be available so people won’t be forced to consider euthanasia as an option. It is a very idealistic idea that we can only hope will be possible in the nearest future.

Another important issue against the legalization of euthanasia was that this law could be abused or misused. Respondents pointed out that many elderly, mortally ill patients could feel obligated to undergo euthanasia so they won't be a burden on their families. Even darker scenario is that severely ill patients could be forced to proceed with euthanasia to unburden the crumbling medical system. People fear that allowing euthanasia could be a start of a society where we do not take care of elderly people or people with disabilities.

CONCLUSIONS

1. Students of medical universities in Poland have some knowledge about the legal aspects of euthanasia. However, this knowledge is incomplete, especially in the matter of the consequences of euthanasia.
2. Ignorance of the law is dangerous. Knowledge of legal acts protects the doctor from harmful consequences, embarrassment and possible legal process. This knowledge is crucial for medical students, as their future profession has a direct responsibility for the health and life of the patient.
3. Universities should provide an appropriate level of medical law classes so that the graduate starts her/his professional career with sufficient knowledge that is necessary in medical practice.
4. Euthanasia is a complex and difficult subject, which certainly will be discussed in the future. This discussion should involve specialist lawyers, doctors and spiritual guides, as it is a controversial topic concerning many ethical and moral matters, as well as religious ones. But most importantly, it evokes strong emotions on both sides. That is why this discussion should be led by firmly educated people.

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Feminity, partnership, family and work of women with disability - preliminary results

Józefa Dąbek¹, Maja Surma², Halina Kulik², Magdalena Szynal^{1,3}

1. Department of Cardiology, School of Health Sciences in Katowice, Medical University of Silesia in Katowice, Katowice
2. Chair and Department of Nursing Propedeutics, School of Health Sciences in Katowice, Medical University of Silesia in Katowice, Katowice
3. PhD Studies, School of Health Sciences in Katowice, Medical University of Silesia in Katowice, Katowice

INTRODUCTION

When disability appears in the family, the whole world of its members turns upside down. All of the family life areas such as: marriage, parenthood and sexuality are being put into question. Anxiety and loss, which accompany the family, additionally intensify the social reception. Questioned is also femininity that significantly drops compared to being an able-bodied mother, wife and lover. Dreams of marriage, sexual intercourse and having children terrify the disabled woman. Moreover, the often cause doubt and disapproval in the society [1,2].

Case gets a bit different when a woman was already disabled when starting a family. Her able-bodied, or disabled partner was aware of his or hers chosen one disability from the beginning. Accepting, understanding and being used to this fact the partner is ready for marriage and to have children in future. In mentioned case won't be any surprise factor of a new, uneasy life situation, there won't be any: fear, consternation, moment of rejection and disbelief that are present in a sudden disability, caused by accident.

Unfortunately, there is still a belief in the society that we live in that disabled people are incapable of many daily activities. Environment is led by such stereotypes, myths and prejudices as: "all disabled people should be isolated", "disabled woman is infertile", "disabled woman isn't able to raise children", "disabled woman isn't a woman", "why is he even with her?"

Most important thing is to remember, that in mentioned before situations maintain respect and love for the other person in love, family, sexual and parenthood life. Everyday struggle to keep the family life as previously isn't easy, family members have to go through and fight many obstacles tied to disability. Societies task is to not only not disturb them in this fight, but also where it is possible – help.

Due to stereotypical thinking, disabled women aren't seen in the light of their femininity, but through their disability. Because of that, disabled women are treated as worse, unattractive and also asexual by the rest of society. They are perceived in the same way also by men (especially able-bodied), so their potential partners. Unfortunately, it happens that having a partner, husband and family by the disabled woman only remain in her dreams and hope and that it is only reserved for world of able-bodied people being meanwhile unobtainable for disabled women [1,2].

The truth is that able-bodied women that are attractive, slim and looked-after are in a better position to find a partner. What appeals to men in first place is undoubtedly appearance – according to individual preferences. Woman with particular disability finds herself in a more difficult situation because she is perceived as a less attractive person. For the disabled woman, awareness of relation with an able-bodied partner is associated with hope for being taken care of by her partner, sense of security, tenderness and support in everyday problems. From the point of man, relation with a disabled woman may come from emotions that feels to her, readiness to support and help her. Unfortunately, due to society's stereotypical thinking, an able-bodied man is only with the disabled woman “out of pity” [1,2].

Situation where disability appears suddenly due to, for example, an accident or disease, is completely different. In one possible situation, the man remains with his partner to support her in their common misfortune, to comfort her and face the new disabled reality together. In another situation, disability will be the main and only cause of breaking the relation with disabled woman, who is a less attractive wife/partner then. It also happens that it is thanks to disability that man and woman come closer to each other and establish a relationship. There are known cases in which people started to love each other during rehabilitation period – feeling between patient and the therapist. Sometimes woman finds her real love after losing physical ability by meeting a disabled man, for example, in a disabled peoples club.

Most of disabled women choose disabled men for their life partner. The cause of such approach is sense of understanding, common limitations and health issues, as well as activities,

better tolerance towards each other and especially sense of equality in the relationship. Every human, no matter sex, skin color, religion or sexual orientation wants to love and be loved [1,2].

The need of sexual activity applies to every human no matter their disabilities. Sexual intercourse has a procreative and hedonistic role strengthening relations of two people. It is a natural development area assigned by nature, leading to development and self-realization and it is maintaining good level of both mental and physical health. Until recently area of disabled people sexuality was a taboo case. Nowadays more and more scientific publications dig into this issue [2,3,4]. According to prof. Zbigniew Lew-Starowicz “*sex for disabled people is particularly important, because they feel normal thanks to it*” [2,5].

When a disabled woman finds an appropriate man fulfilling his role as a husband, next step of building relations has to be made: starting a family and planning to have children. Widespread stereotypes on wife and mother role bring them to fulfill specified functions. Society has designated an existence area of wife to take care and satisfy her husbands need, and mother's duty to raise children. Disabled wife's and mother's role were brought to the same duties, but additionally limited by the fact of disability.

Contemporary society learn tolerance and empathy for disabled people as well as for relationships of disabled with able-bodied people and having children by them. Unfortunately, sight of a disabled woman with a disabled or able-bodied man and child still rises mixed emotions and controversies [2,4].

Problems of disabled wife and mother consist of: fear of society disapproval and rejection, stigmatization as well as stereotypical picture, awareness of imperfection, sense of control and observation from society point of view and also fear of yourself: “what if I won't make it?”, “what if I won't be able to help my child?”, “will I be a good mother?”, “am I able to give my own child security?”.

Disabled women extremely carefully and responsibly make decisions about marriage. They are aware of opinions and stereotypes put on a disabled mother by the environment. Disabled women are thoroughly aware of their own motive limitations and related to them impediments with caring for a newborn baby [2,4].

Medical care for a disabled pregnant woman should be the same high standard as for an able-bodied woman accounting special needs, necessary conveniences and help due to physical disability of future mother. However, medical personnel in hospitals and maternity wards is still not ready to accept and care for patients with motive disabilities.

Women moving on a wheelchair will perform differently as a mother, women without upper limbs differently and blind mothers yet in another way. All of disabled mothers will find

a convenient solution for themselves simplifying care for the child. During years of their disability's women has learned many ways to manage with their limitations. During motherhood they will use acquired skills just as effectively.

AIM OF STUDY

Purpose of this paper was recognition of disabled women ability to self-realization in family, work and partnership areas.

MATERIALS AND METHODS

Research tool was a customized questionnaire consisting of 32 questions regarding main topic and 6 questions which asked for age, place of residence, education, marital status and number of children as well as work.

Main part of questionnaire asked about men's attitude towards respondents and vice versa, sense of attractiveness of disabled women and their sense of fulfillment in specified categories: woman, mother, wife and lover.

Questions asked as well about fulfillment of disabled women in mothers' role and professional work of disabled women.

RESULTS

General characteristics of examined group of physically disabled women

Research included a group of 100 physically disabled women. Criteria of being included into examined group was physical disability and female sex. Among examined group of disabled women (n=100) youngest was aged 21, however oldest was aged 65. Average age of examined women was at 40.2 years old. Residents of villages were 12 (12%) examined women, however rest 88 (88%) lived in a city.

Analysis of obtained results revealed that just over half of examined women (52%; 52) remained in marriage, next 22 (22%) women were single and 12 (12%) were divorced due to their disability. However for 10 (10%) of respondents reasons of divorce weren't bound to disability and 4 (4%) remained in informal partnerships.

Most of women (52; 52%) graduated from university, however 36 (36%) had secondary or vocational education, and rest 12 (12%) had primary education.

Maternity was declared by almost two thirds (74; 74%) of examined women, including 52 (70%) had one child, 22 (30%) two, three or more offspring, and rest 26 (26%) didn't have children. Most of women were professionally active (68; 68%), including 56 (82%) performing mental work, and 12 (18%) – physical work. Rest 32 (32%) women didn't work professionally.

Femininity, family life and partnership of examined physically disabled women

Among examined group of 100 women there were 74 (74%) mothers, 78 (78%) women had a husband or a partner and 66 (66%) were sexually active. Characteristic of examined physically disabled women including their femininity, family life and partnership was shown in figures 1., 2., and 3.

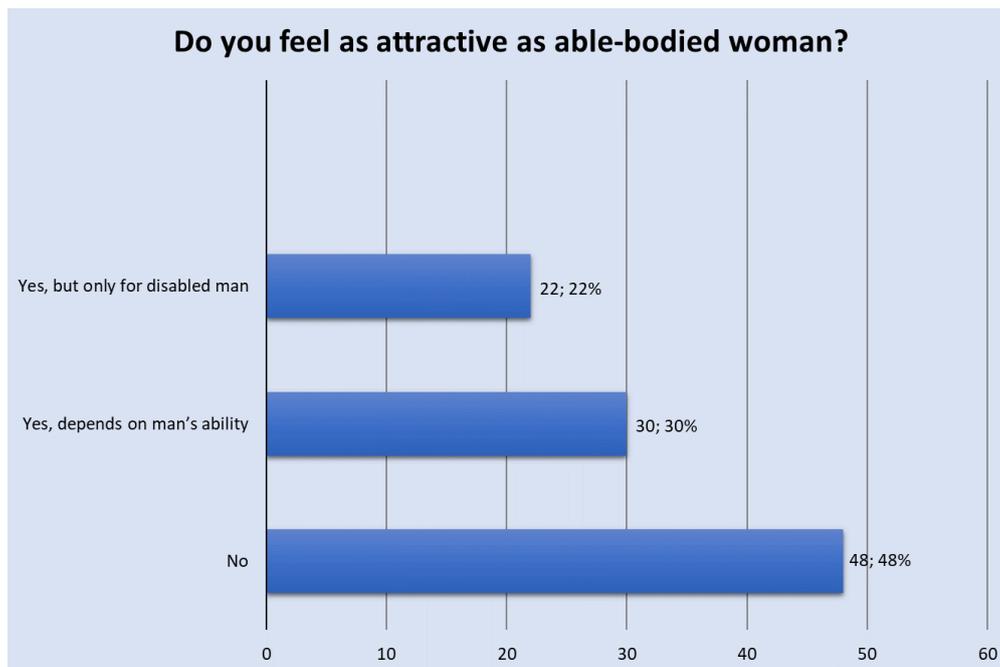


Figure 1. Attractiveness of examined physically disabled women for opposite sex

As much as 48 (48%) of examined women didn't feel as attractive as able-bodied, however 22 (22%) women felt as attractive but only for a disabled man.

Among group of 100 examined women:

- 68 (68%) felt fulfilled women,
- 70 (95%) of women that are mothers – fulfilled mothers,
- 72 (92%) of women having a partner – fulfilled partners
- 42 (64%) of sexually active – fulfilled lovers.

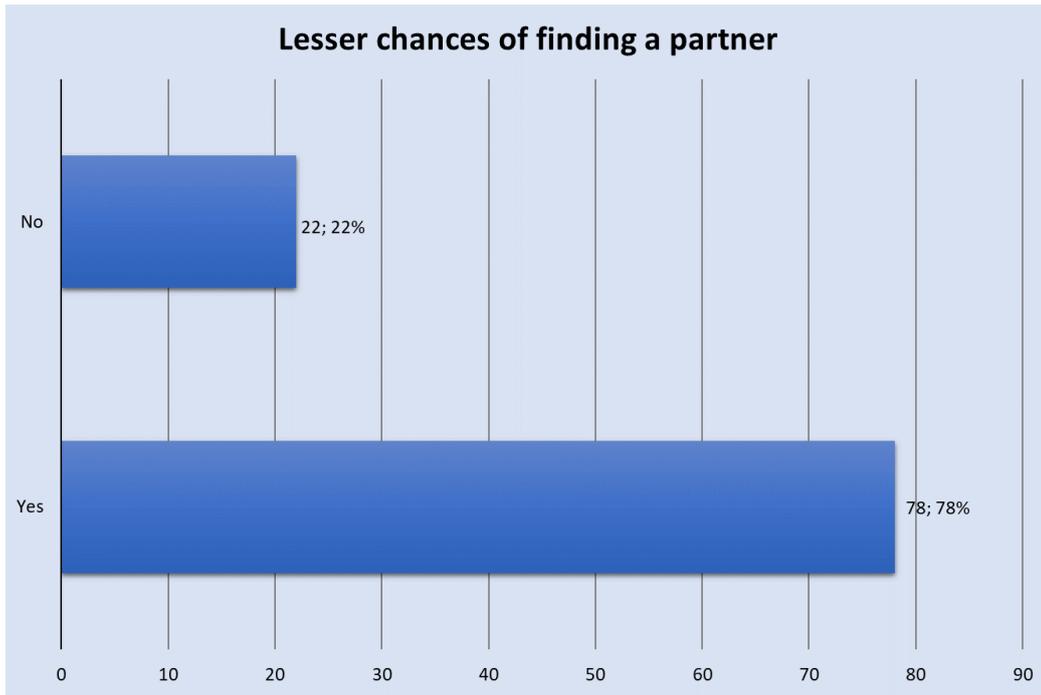


Figure 2. Chance of finding a life partner evaluated by examined physically disabled women

As much as 78 (78%) women marked that they have lesser chances of finding a partner due to their disability.

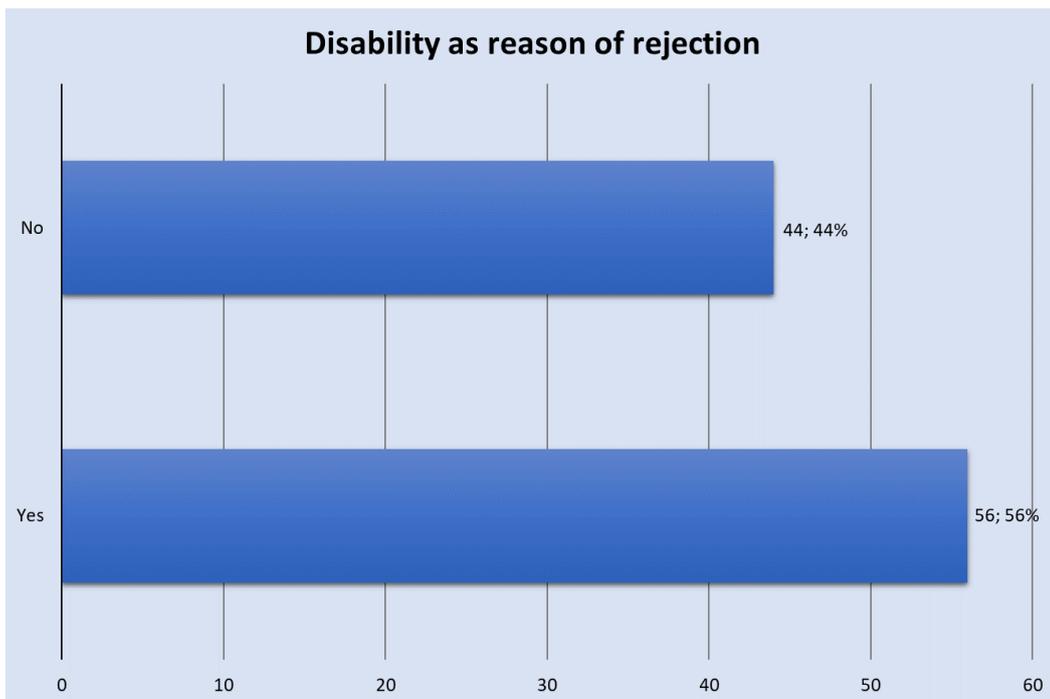


Figure 3. Disability as a reason of rejection in marriage or partnership of examined physically disabled women

More than half of examined women (56; 56%) declared that they encountered rejection from their partner due to their disability. As much as 68 (68%) of them expressed fear of feeling that their partner stays with them out of pity.

Intimacy sphere and sexuality of examined physically disabled women

Out of 100 (100%) questioned women, as much as 66 (66%) were sexually active. Characteristic of examined group of physically disabled women including their sexuality was shown in figure 4.

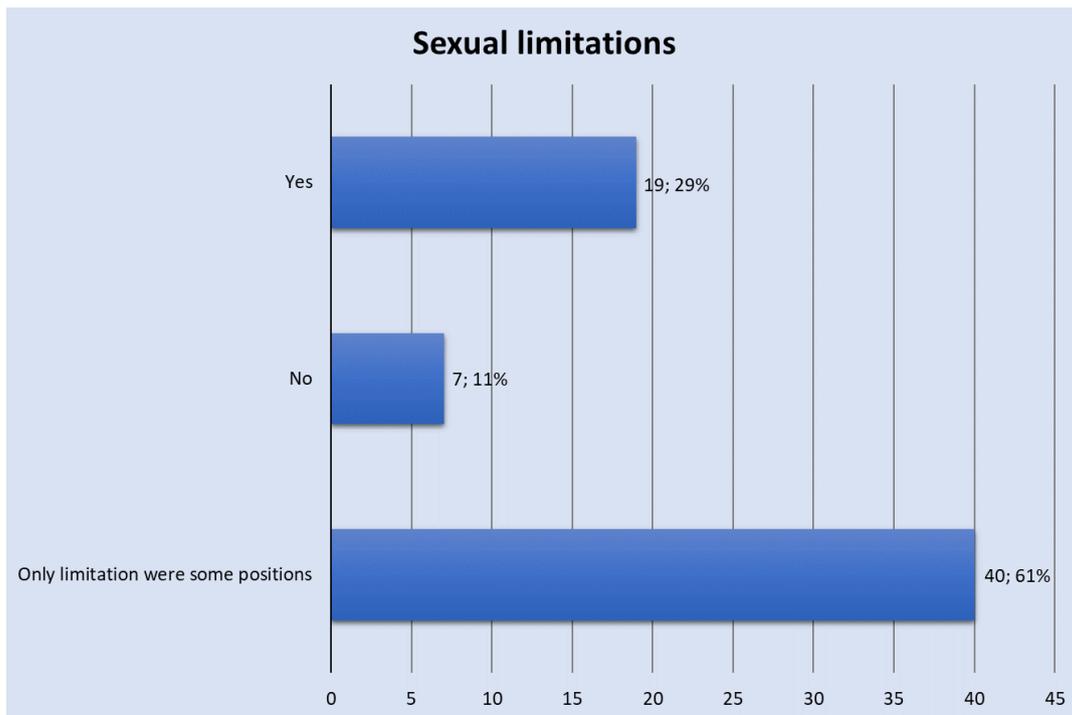


Figure 4. Sexual limitations of examined physically disabled women

The most numerous group of examined women (40; 61%) thought that only limitation in sexual intercourse are some sexual positions, however 19 (29%) thought that they have the same sexual capabilities as an able-bodied woman. Only 7 (11%) of examined thought that mentioned capabilities are smaller.

Maternity in examined group of physically disabled women

Out of 100 (100%) examined women as much as 74 (74%) were mothers. Characteristic of examined physically disabled women including their maternity was shown in figures 5., 6. and 7.

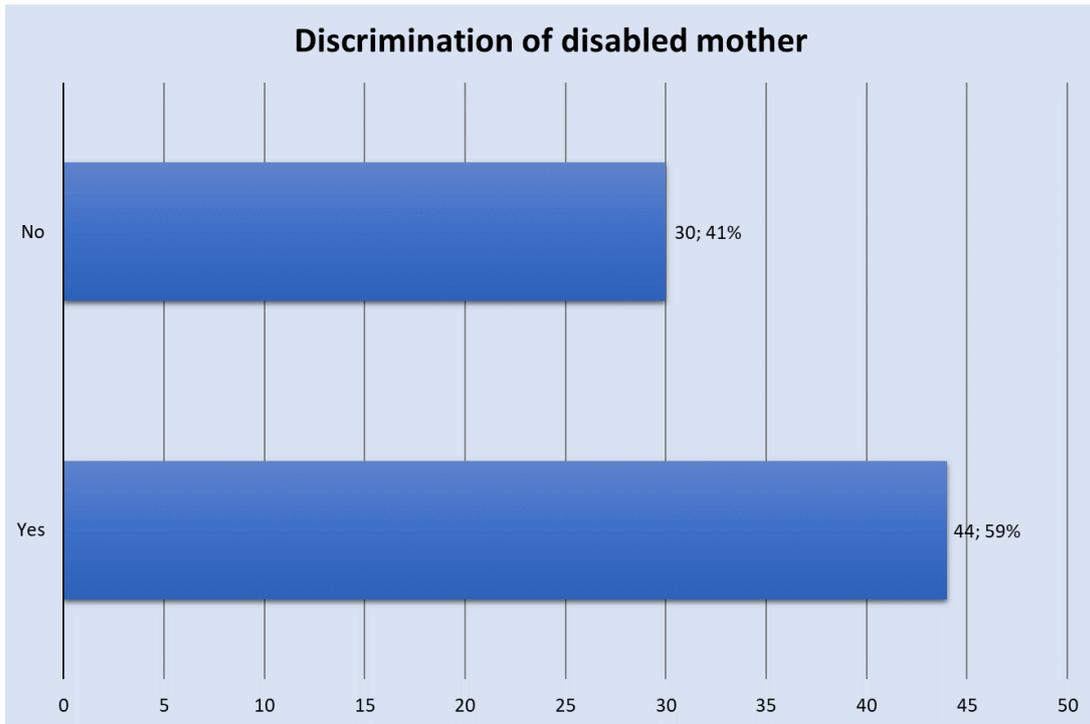


Figure 5. Experience of discrimination of examined physically disabled women as mothers

More than half of examined mothers (44; 59%) felt controlled, stigmatized and discriminated due to fact that they became mothers.

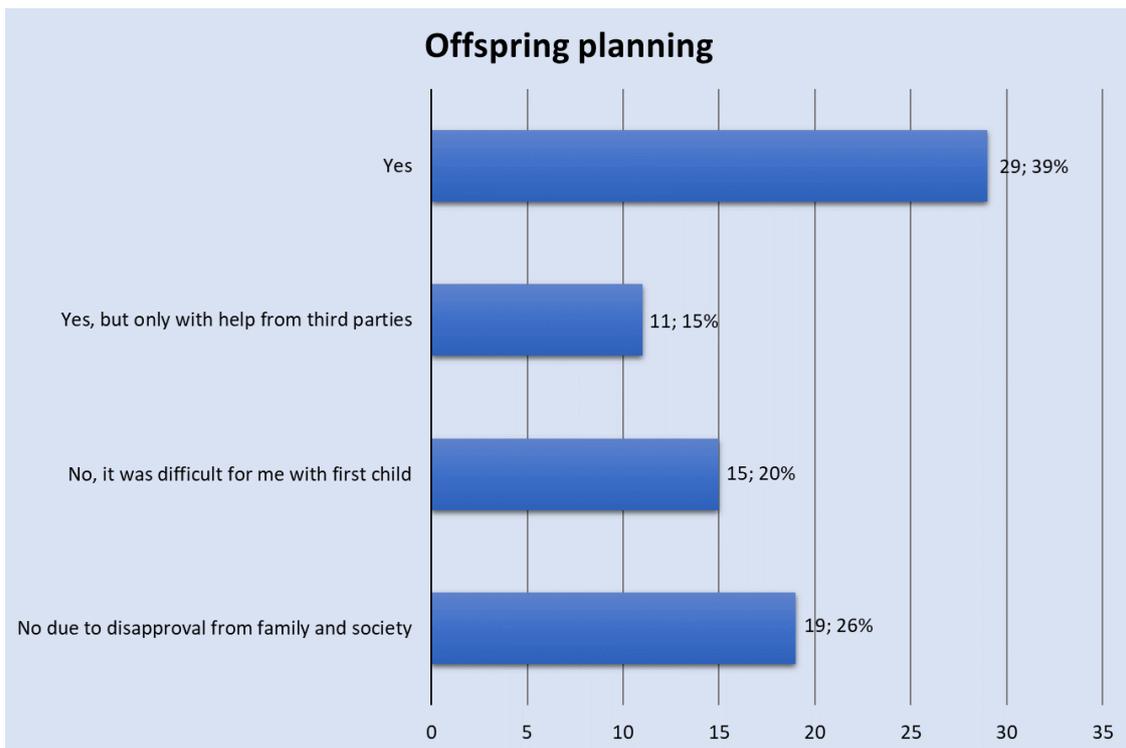


Figure 6. Offspring planning by examined physically disabled women

Out of mothers group: 11 (15%) declared desire to have next child with provided assistance to take care of him, next 29 (39%) – decided on having next child without any assistance provided, 19 (26%) didn't decide to have next child due to disapproval from family and society, and 15 (20%) stating that they poorly managed to take care of their first child.

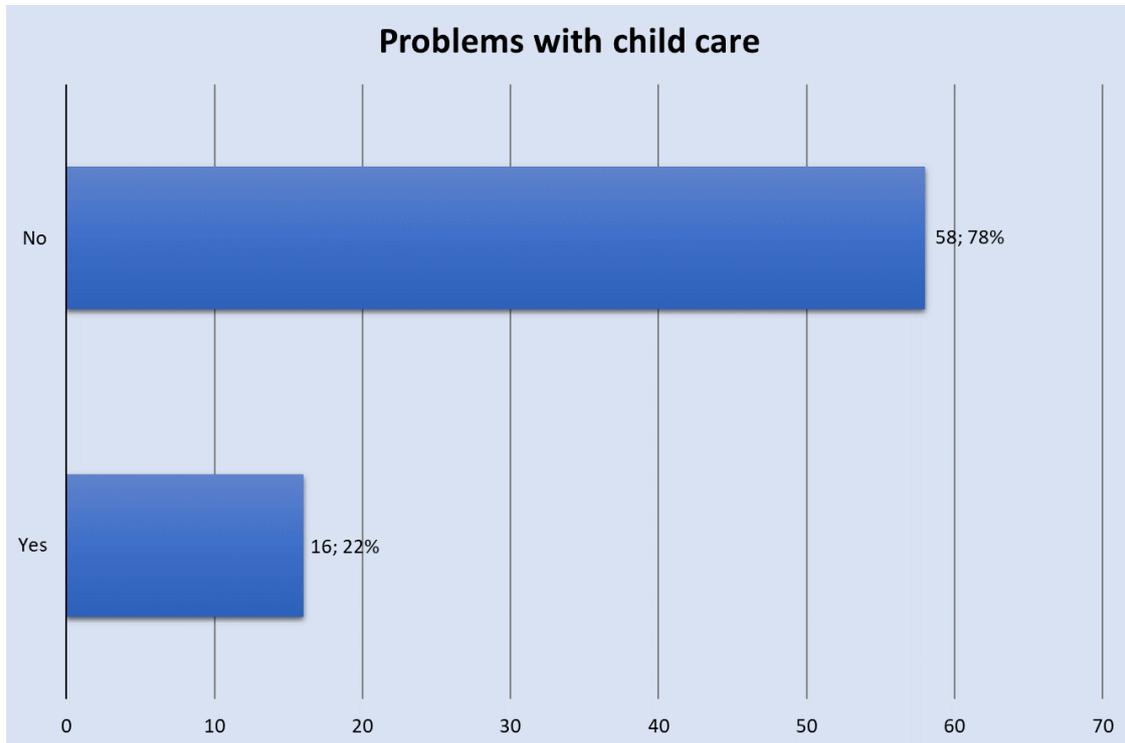


Figure 7. Problems with children care among examined physically disabled women

Most of examined group of disabled mothers (58; 78%) didn't have problems regarding child care, the rest 16 (22%) experienced such problems. Problems stated with children care by disabled women were mentioned such as:

- issue with quickly getting to the child in case of it crying,
- bringing down or up baby carriage,
- going for a walk independently,
- shopping,
- getting through architectural limitations,
- issues with care in case of worse level of own health.

Professional work of examined physically disabled women

Characteristic of examined physically disabled women including professional work-related aspects was shown in figures 8. and 9.

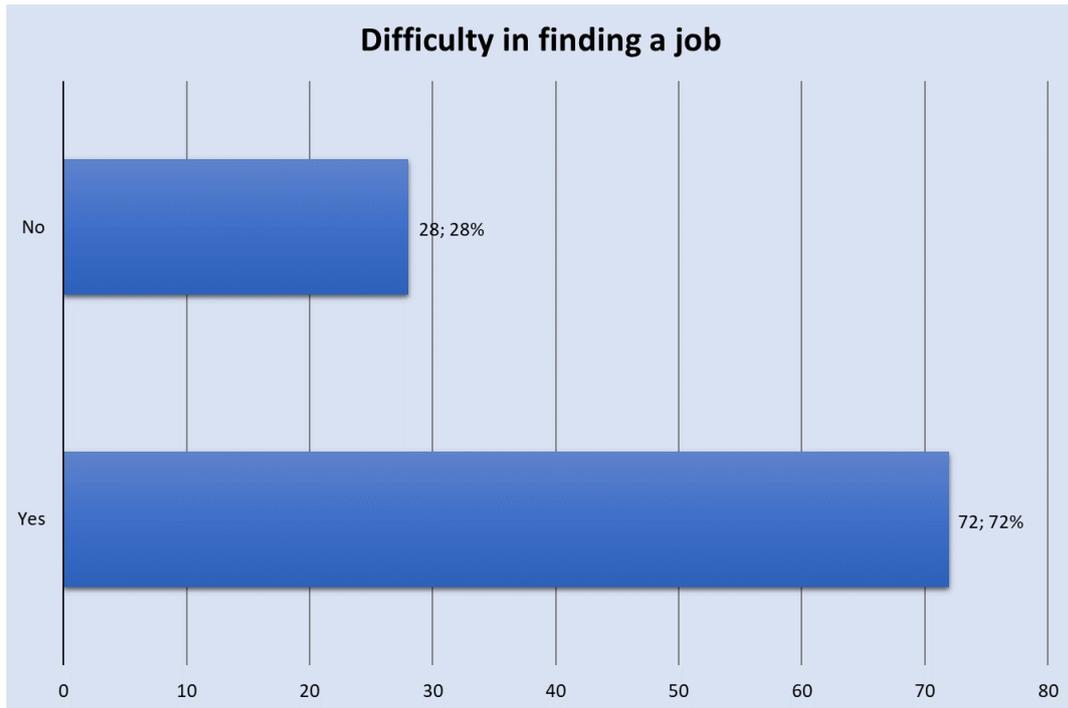


Figure 8. Issues with finding a job position by examined physically disabled women

Out of all 100 examined disabled women, as many as 72 (72%) had problems with finding a job due to their disability.



Figure 9. Unemployment reasons among examined group of physically disabled women

In the examined group 32 (32%) women didn't have a job including 11 of them (34%) that didn't feel the need to work and 10 (31%) who were raising a child, however 5 (16%) women feared of discrimination at work and 4 (13%) didn't believe in their capabilities.

DISCUSSION

In order to conduct the research a customized own authorship questionnaire was used. Research included a group of 100 physically disabled women. Youngest woman of the examined group was 21 years old and oldest one was 65 years old. Collected results pointed out a high level of education of examined disabled women: 52 (52%) graduated with university degree, 36 (36%) had secondary or vocational education, 12 (12%) – primary education. Out of all women participating in the research 78 (78%) remained in partnership or marriage, and 74 (74%) were mothers.

Own research proved that 78 (78%) of examined women confirmed difficulties in finding a partner due to their disability. Out of 100 (100%) women participating in the research, 68 (68%) felt fulfilled as a woman. Disabled women usually are seen as asexual due to stereotypes widespread among society. Disabled women encounter a number of limitations related to dating as well as in physical and mental sphere. Current canons of beauty force certain standard which cause complexes for disabled women, feeling of being worse, incomplete, not inscribed in beauty standards [6].

Results of own research deny the myth of supposed asexuality of disabled women. 78 (78%) of respondents stayed in stable partnerships or marriages, however 72 (92%) of them felt fulfilled as a wife/partner. Moreover, as many as 42 (64%) women from 66 sexually active felt fulfilled in that role and 40 (61%) of them admitted that only limitation in intimate intercourse are some of the sexual positions. Beleza in her research also accounted the stereotype of asexuality of disabled women. She stressed on existence of unfounded stereotypes about disabled women's inability to be in the role of mother and wife and inability to give birth [A].

In own research as many as 74 (74%) of women were mothers. Maternity is a time which most of women want to experience in their lives. It results from satisfying many needs as well as in biological, mental and social spheres. Desire to fulfill as a mother also referred to disabled women [7]. In own research as many as 70 (95%) from the group of disabled women felt fulfilled in their roles. Those results confirm the fact that maternity is an exceptional period for women which is experienced in special way no matter the mobility level.

In own research it was proved that disabled women have problems with finding a job. As many as 72 (72%) of examined disabled women marked that fact. However, only 32 (32%) women from examined group didn't have a job including 11 of them (34%) didn't feel the need to, 10 (31%) were raising a child. However, 5 (16%) women were afraid of discrimination in work and 4 (13%) didn't believe in their abilities. In Gadzinowska research most common reasons of reluctance to take on a job by disabled people was: bad health level, no faith in finding a job, negative stereotypes regarding people with medical certificates of disability functioning in the society, which caused waiting strategy, lack of own initiatives and ideas to improve their fate. It resulted in weakening the position of disabled people in the society and seeing them through the light of only one feature – disability [8].

Disabled people often didn't decide to take on a job due to lack of adjusted working conditions for them. Not all places can provide suitable working environment for disabled people. Propitious working place is one of main reasons of disabled people to work. Supposedly if workplaces could provide better work conditions for people with disabilities, unemployment issue in mentioned group would lower or wouldn't be present at all [9].

In article titled „Double discrimination of disabled women in workplace” of Pawłowska-Cyprysiak accounts for phenomenon of discrimination with twice the force. From questionnaire research titled “Psychosocial diagnosis attempt of disabled women in Poland” it appeared that 53% of women covered by the examination admitted to be discriminated due to their disability and only 15% felt discriminated due to being a woman. In quoted research, 28% of women related difficulties in finding a professional work with their disability and 8% felt that disability is a factor that completely prevents them from having their dream job. Despite the development and higher tolerance of disabled people in recent years, research revealed rise of discrimination of women as employees and showed reluctance of employers to hire women, especially disabled ones [10].

All of the barriers that women are put through by fate, or especially society of able-bodied people, are possible to overcome. Biggest issue remaining is the need to overcome negative myths, stereotypes and judgements, that are still functioning, about disabled women. Important fact for disabled women is social support, but the most important is tolerance and empathy.

Radziewicz-Winnicki had similar assumptions when he wrote about necessity to increase social interest in the area of disability. In order to repeal all social barriers related with disability, it is needed to reduce the barrier between environment of able-bodied and disabled people [11].

CONCLUSIONS

1. Examined disabled women mostly fulfilled themselves in feminine roles such as: mother, wife and partner,
2. Physical disability of examined women was often a limitation for them to find a job.
3. Still a current challenge for society remains integration with disabled people.

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The gift of vision – the use of eye tissues from deceased donors in ophthalmology

Zuzanna Marek¹, Karolina Borowska¹, Alicja Lachowska¹, Kornelia Kręcisz¹, Maria Myślicka¹, Jan Gnus^{1,2}, Joanna Bogusławska², Agnieszka Kowal – Lange², Magdalena Targońska², Michał Karaszewski¹

1. Wrocław Medical University
2. Regional Specialist Hospital in Wrocław, Research and Development Center, Wrocław

INTRODUCTION

The use of cells, tissues, and organs from deceased donors is now a recognised and widely used method of treatment worldwide. Treatments with their use are often the only available therapeutic pathway for normal functioning. In ophthalmology, the most important transplant surgeries are corneal and sclera transplantations. Corneal transplantation is one of the most frequently performed procedures in the world and has the highest success rate among solid tissue transplants [A].

The number of eye tissue grafts has undoubtedly increased in recent years, which is related to the development of microsurgical techniques and modernization of eye tissue preservation/banking methods [B].

In Poland, 1264 corneal transplants were performed in 2019 [C]. Unfortunately, this is not a sufficient number and patients have to wait in long queues for the surgery. In the whole of Europe, about 20,000 such procedures are performed annually, while in the United States of America alone the number is about 40,000 [D].

EYE TISSUE BANKS

Eye tissue banks prepare for transplantation tissues such as cornea and sclera. Corneal transplantation is primarily based on tissues from deceased donors obtained during tissue or multi-organ donations [1]. Tissue collection is possible after confirmation permanent, irreversible cessation of brain function of the donor [E].

In Poland, a potential donor can be anyone who has not objected to the Central Register of Objections during his or her lifetime or has not expressed an objection in the form of a written statement with his or her own signature or an oral statement made in the presence of at least two witnesses and confirmed by them in writing [E]. A doctor who intends to perform tissue removal from a deceased person must first establish his or her personal data, determine the time and cause of death and obtain information whether the person's objection is placed in the Central Register of Objections. The doctor should also talk to the family of the deceased to verify the existence of other forms of objection [1].

PRE-QUALIFICATION OF EYE TISSUES FOR TRANSPLANTATION

Tissue collection must be preceded by completion of the deceased's medical records, medical history and a general examination of the corpse for risk factors. Tissue banks follow sterile procedures and strict donor selection criteria to prevent the transmission of infectious diseases and malignant tumours from donor to recipient. To this end, lists of relative and absolute contraindications to the use of tissues from a deceased donor are created. Unfortunately, it is not possible to eliminate the risk completely, but by following the procedures it is possible to reduce it to an acceptable level. Corneas from donors below 1. year old usually are not useful because they are very flaccid and can cause big post-operative astigmatism. Corneas from donors over 70. year old should also not be used due to the small number of endothelial cells [2]. Eye tissues cannot be admitted to the eye tissue bank if there are no data on a deceased donor [3]. The criteria for the exclusion of a corneal donor are presented in the subsection „Keratoplasty”.

The exclusion criteria for sclera donors are similar to those for cornea. Additionally, people after surgical procedures, such as freezing or eyeball strapping, are disqualified [1].

TISSUE COLLECTION

Tissue collection must be performed up to 16 hours after death for refrigerated corpses and up to 20 hours for uncooled corpses [1]. The shorter the time from death to tissue collection is, the better for preserving corneal endothelial viability [2]. After initial positive qualification of the transplant donor, a macroscopic evaluation of the knobs and protective eye apparatus is performed [1]. The entire procedure should be performed in sterile conditions. For this purpose, a sterile surgical field is prepared before tissue removal, eyelids are washed with an antiseptic

substance, and the eye and conjunctival sac are rinsed with a solution of betadine or an antibiotic [1,3]. Currently, in Poland, cornea is taken mainly in two ways. The first one consists in excision of the corneoscleral graft *in situ* and this is the most commonly used method. After collection, special caps are put on the eye surface and eyelids are secured with stitches. The tissues are transported to the eye tissue bank in a coded glass container with corneal storage fluid. The container is placed in the transport fridge, where the temperature is +4°C [1,4]. The second way is to take the whole eyeball, which is transported to the bank in a humid chamber. The humid chamber is placed in a special thermal container filled with ice. The main purpose of such actions is to keep the temperature inside 2-6°C [1,3]. After collection, eyeball prostheses are placed in the eye sockets and the eyelids are secured with stitches. The final stage is the collection of blood from the femoral or subclavicular vein of the donor for serological tests for hepatitis B, C, HIV and syphilis infection [1,4].

PREPARATION OF TISSUES FOR TRANSPLANTATION

Tissues are transported to the tissue bank at a temperature of 2-6°C. Both corneoscleral grafts and entire eyeballs are examined at the slit lamp. The following parameters are evaluated: corneal translucency, presence and location of scars, foreign bodies, infiltrates, oedema, senile periphery, shape abnormalities, epithelial defects and the degree of Descemet's membrane corrugation [1]. In order to minimize the risk of transmission of infectious agents such as viruses, bacteria or fungi, the donor's eyeballs are decontaminated. After pre-cleaning, the eyeball is sterilised for 2 minutes in 10 % betadine solution and rinsed for one minute in sodium thiosulphate. Next, a corneoscleral graft is excised and put into a special preservative medium. Modern corneal storage media contain gentamicin and streptomycin for antibacterial protection. Both described procedures are performed in a laminar airflow chamber [1,3]. Sclera can also be obtained from the entire eyeball. After the removal of the conjunctiva, rests of the muscles and the excision of the corneoscleral graft, the inside of the eyeball is removed. Before preservation, the tissue is rinsed for 20 minutes in gentamicin solution or quarantined for 14 days in 70% ethanol solution, which is then replaced [1].

A very important stage in the qualification of the cornea for transplantation is also the assessment of endothelial cells. There are methods that enable its examination in both closed and open corneal collection systems. The closed corneal collection system is based on the fact that the cornea is removed from the medium only in the operating room, before the transplantation. In this case, endothelial cell examination is possible using a specular

microscope. The open system uses a light microscope. By direct examination of the corneoscleral graft, additional assessment of endothelial cell viability is possible. First, hypotonic solutions are added, which results in swelling of the endothelial cells. Then trypan blue staining is used to recognize dead cells and detachment of Descemet's membrane. Modern software in microscopes for endothelial assessment allows precise morphometric evaluation. The evaluated parameters include endothelial cell density. Depending on the type of transplantation, certain graft values are required. For a posterior layered graft, a higher endothelial cell density is required than for a hollow graft. Morphological assessment is also important in terms of possible presence of abnormal cells, sediments or changes typical for dystrophy [1].

STORAGE OF TRANSPLANTATION MATERIAL IN EYE TISSUES BANKS

There are several ways of storing eye tissues. None of these techniques are perfect. Every method has its advantages and disadvantages. Differences between them mainly concern on the storage temperature, the composition of the medium, the presence of osmotically active ingredients and the storage time of the transplantation material [4].

The entire eyeball can be stored up to 48 hours after collection in a humid chamber at a temperature of 2-6°C. This is a simple method that doesn't require any special equipment. Unfortunately, it does not take into account such important aspects as microbiological contamination or the risk of transmission from donor to recipient. Nowadays, this method is less frequently used [1,4].

Corneoscleral graft can be stored using hypothermic method at the temperature of 2-6°C or in a tissue culture at the temperature of 31-37°C. Longer storage time permits greater flexibility in the use of the donor tissue and prevents wastage [5].

The use of the hypothermic method allowed to extend the term of validity of the tissue up to 14 days, depending on the type of medium used [1]. Low temperature lowers the corneal metabolism to a minimum [4]. The medium contains antibiotics, substances such as dextran and chondroitin sulfate to prevent corneal swelling *in vitro* and other agents such as energy sources, antioxidants, growth factors and membrane stabilizers. Tissue control with a slit lamp or specular microscope can be performed in a closed system. EUSOL is currently the most commonly used medium in Poland, refrigerator storage with minimal handling. It requires no complex or expensive equipment [1,5].

For the tissue culture method at the temperature of 31-37°C the cornea storage time is about 4 weeks. It is more complicated than the hypothermic method and requires the use of specialized equipment [1,5]. It is based on the assumption of long-term storage of an isolated human cornea under conditions as close as possible to physiological [4]. Examination of the endothelium is performed not only before storage but also after storage using a light microscope. In this method, additional microbiological tests are required during storage and before tissue release from the bank. The shortest time of tissue culture is 10 days - this is the time needed to detect a possible fungal infection. The vulnerability of organ culture to microbial contamination can be turned into an advantage because it allows the detection of residual microorganisms on the cornea before surgery [5]. Preservation medium in this method is enriched with calf serum, antibacterial and antifungal antibiotics. Anti-oedematous agents must not be added to the medium because they accumulate at this temperature in the vacuoles of the corneal cells. Lack of these agents causes significant corneal oedema. For this reason, the tissue is transferred to a transport medium containing dextran, in which the cornea can be stored for 1-7 days [1]. Because organ culture involves extensive testing of the quality of the donor cornea, there are theoretically no preset limits on donor age and postmortem time [5].

Both preservation techniques seem to result in similar graft survival. The method of choice for preservation of the donor cornea is dictated by a number of factors. Currently, the method of choice in Western Europe is tissue culture [4,5].

The method of storing sclera is different. Before conservation, sclera is rinsed for 20 minutes in gentamicin solution or quarantined for 14 days in 70 % ethanol solution. It can be stored at room temperature - in 70 % ethanol, glycerine or formalin, in a refrigerator in Optisol-GS solution or a saline solution with antibiotics or frozen after dehydration. Sclera transplantation requires its prior preparation - rinsing in saline and antibiotics, thawing and hydration, depending on the method of storage [1].

RULES FOR RELEASE OF THE TISSUE FROM THE TISSUE BANK

The final acceptance of tissue for transplantation is determined by the results of serological tests of blood samples from the donor for HBV, HCV, HIV and syphilis infection. If any of these tests are positive, even if the tissue is of very good quality, it is disqualified for transplantation. Each donor has a briefcase in the bank and each cornea has its unique identification number, in accordance with the European tissue codification system for transplantation. Before the tissue is released, it is checked again. The appearance and colour of

the medium is assessed. Turbidity may indicate tissue infection and then the tissue can not be released from the bank. The surgeon receives with the tissue the donor's medical documentation (cause of death, autopsy results, serological results, corneal evaluation). Two forms are attached to the documentation: a post-operative report, which should be sent back to the bank after tissue transplantation, and an adverse reaction report, which should be sent back to the bank in case of adverse reactions related to the transplanted tissue [4].

KERATOPLASTY (CORNEAL TRANSPLANTATION, GRAFTING)

Keratoplasty is a procedure in which the recipient's pathologically altered corneal tissue is replaced by the donor's healthy corneal tissue. The pathologically altered cornea loses its transparency, is not shiny, its structure is not smooth, and the shape disorders do not undergo any method of correction [6,7].

Indications for keratoplasty

The basic indications for corneal transplantation can be divided into 4 groups:

1. Keratoplasty due to optical indications - used to improve vision
 - a. turbidity
 - b. bullous keratopathy after cataract removal
 - c. pathologically altered corneal curvature (corneal cone and dystrophies, degeneration and scars) [7,8]
2. Keratoplasty due to tectonic indications - performed to restore or maintain the integrity of the cornea in patients with severe changes in its structure
 - a. postoperative fistulas after cataract removal or anti-glaucoma surgeries
 - b. after excision *en bloc* of a tumor, epithelial cyst, external fistula with necrotic wound edges
 - c. thinning of the corneal stroma skeleton
 - d. posttraumatic corneal injury [7,8]
3. Therapeutic keratoplasty - performed to remove the cornea damaged by an infectious agent when the applied pharmacological treatment has not brought the desired effect
 - a. deep keratitis: a granulomatous reaction from Descemet's membrane in viral inflammation (HSV)
 - b. primary or secondary endothelial disease
 - c. corneal perforation in the course of ulceration [7,8]

4. Cosmetic keratoplasty - sometimes carried out in the situation of damage of the optic nerve (in a patient with total blindness with no chance of proper vision), when the damaged cornea is a cosmetic discomfort for the patient - leucoma [7]

An additional indication may also be an autologous grafting of corneal tissue at cataract removal in the case of a very small cornea to gain access to the lens [9].

TRANSPLANTATION MATERIAL

The basic principle is to collect the cornea from a deceased donor within 24 hours of death. The best transplantation material are corneas of donors from 1 to 70 years old. This is associated with too much tissue flaccidity in infants, which can result in high postoperative astigmatism and too few endothelial cells in donors over 70 years old - for this reason they are not a useful transplantation material [7].

Preoperative evaluation of the donor's cornea includes examination at the slit lamp and in a specular microscope [10].

Contraindications for cornea collection from the donor side:

- unknown cause of death
- infectious diseases of the central nervous system (Creutzfeld's disease, systemic sclerosing encephalitis, progressive multifocal leukoencephalopathy)
- AIDS, viral hepatitis, syphilis, sepsis
- leukaemia and disseminated lymphoma
- diseases of the donor's eye (cancer, active inflammation) or past ophthalmological surgeries [10]

Contraindications for keratoplasty from the recipient side:

- pathological lesions forward of the Bowman layer (papillary lesions, cancer *in situ*, epithelial dysplasia and psoriasis) - lesions requiring removal of the altered tissues.
- Band keratopathy
- Sclerocornea
- Microcornea in Peter's anomaly

In the case of the above mentioned lesions, it is sufficient to use other therapeutic methods (removal of calcium deposits using sodium wersenate with preservation of the anterior part of the corneal framework in Band keratopathy) or no improvement or even postoperative eye loss in children with Sclerocornea or Microcornea in Peter's anomaly [8].

The course of keratoplasty may be adversely affected by such abnormalities in the recipient, as: eyelid edge inflammation, ectropion, entropion, abnormal eyelash growth; recurrent or progressive conjunctivitis; massive stromal vascularisation, no corneal sensation, massive corneal thinning at the site of the anticipated fusion of donor and recipient tissue and active keratitis, as well as unstabilised glaucoma or uveitis [7].

TYPES OF KERATOPLASTY

In a general way, rafting can be divided into all the layers of the cornea (penetrating) graft and the individual layers of the pathological process (lamellar) graft. Depending on whether the graft concerns the superficial layers of the cornea or reaches Descemet's membrane, a layer graft can be divided into superficial and deep. In order to precisely plan the scope of the treatment, it is helpful to use OCT [6].

I Penetrating Keratoplasty

1. Procedure technique - mechanical

Before the start of keratoplasty, the graft size should be determined by means of an examination in a slit lamp using different widths of the light beam, and during surgery using trepans of different diameters. The optimal transplantation diameter is 7.5 mm [7].

The corneo-scleral segment should be about 0.25 mm larger than the planned diameter of the recipient's corneal excision. Place it on the concave teflon endothelial block upwards and then trim it with a trepan [7].

It should be made after the removal of the donor's cornea, taking care not to damage the recipient's iris or lens (this can be prevented by narrowing the pupil with a pilocarpine) [7].

Four single main sutures should be inserted and the wound closed with a continuous suture. Quite deeply, using monofilament nylon 10-0 thread to ensure the adhesion of the Decemet membrane to the donor and recipient [7].

2. Contraindications

- transient increase in intraocular pressure
- massive stromal vascularisation with no Bowman layer
- eyelid regurgitation
- abnormal tear film
- epithelial ingrowth

- very thin recipient's cornea [9]

3. Factors of risk of failure

- the donor material is of poor quality (low endothelial density and morphology; transmission of viral infections to the recipient)
- Graft rejection (immunological reactions)
- dangers of intraocular surgery
- relapse of the underlying disease after transplantation [9]

4. Postoperative treatment

- locally: steroids (initially 4 times a day for several weeks, then the dose can be reduced depending on the condition of the eye. It is usually practiced to administer small doses of steroids for a long time, e. g. once a day for a year). Mydriatics 2 times a day for 2 weeks or more if there is uveitis
- orally acyclovir (when HSV keratitis was previously present)
- after 9-12 months suture removal (in elderly patients and patients with keratoplasty due to keratoconus, sutures should be left for longer, up to two years) [11]
- hard contact lenses (to obtain satisfactory visual acuity in patients with post-operative astigmatism). Only used after all seams have been removed [7]

5. Non-immune complications of penetrating keratoplasty

Graft opacity resulting from endothelial cell dysfunction visible from the first day after surgery. This is due to poor quality of the donor endothelium, surgical trauma, as well as problems with the eye surface leading to unilateral mucus deficiency syndrome [7,8]:

- displacing haemorrhage and postoperative infections
- impaired wound healing (abnormal adhesion of the wound edges, very thin recipient cornea, preoperative epithelial ingrowth, steroid administration and related side effects)
- secondary open angle glaucoma after massive fibrinous exudate and incomplete restoration of the anterior chamber (in children in hot keratoplasty)
- secondary open angle glaucoma or ocular hypertension as a side effect of steroids, alpha-chymotrypsin injection or sodium hyaluronate administration.
- severe pathological changes on the surface of the cornea and conjunctiva, such as eyelid regurgitation, dry eye syndrome, abnormal growth of eyelashes
- incomplete cure of infection
- primary failure of endothelial cells in the donor
- recurrence of disease in a transplant (e. g. stromal dystrophy)

- scars after burns which may cause all the above mentioned complications
- irregular or severe astigmatism [9]

6. Immunological rejection of the transplant

In the first 6 months, about 50% of all transplant rejections occur, most of them in the first year after surgery. We can distinguish epithelial and endothelial rejections.

- Epithelial rejection

It is characterised by a linear opacity of the epithelium, which may not show subjective symptoms. There are no clinically significant consequences. Over time, numerous small subepithelial infiltrations resembling the Krachmer spots (adenovirus keratitis) appear. It may be accompanied by iritis. Intensive local steroid therapy is necessary.

- Endothelial rejection

It is characterised by a more dangerous course due to the lack of regeneration of damaged endothelial cells. This can result in permanent corneal swelling. The first symptom is visible as an inflammatory process on the connection of the donor disc with the recipient tissue or in the form of iris inflammation. Khodadoust lines on the endothelium and corneal oedema are then observed. It is necessary to apply peribulbar and topical droplet steroid therapy and sometimes general immunosuppression [7].

7. Predisposing factors for immune rejection of the transplant

The first factor to be mentioned is the small distance between the tissue of the transplanted cornea and the vessels of the cornea. In this case, the diameter of the centrally located graft can be very large (with a diameter of >7.5 mm) or the graft can be eccentrically located. Another factor are pathological changes in the host cornea, such as: vascularisation, anterior adhesions, and inflammation of the cornea or intraocular tissues. Abnormal wound healing caused by suture irritation, for example, is also an important factor predisposing to immune rejection of the transplantation [8].

The treatment is topical and general steroid therapy and, if necessary, general immunosuppression. In initial endothelial decompensation beta-blockers and carbonate anhydrase inhibitors turn out to be useful in the treatment [8].

II Deep Anterior Lamellar Keratoplasty

The treatment consists in removing the entire opaque cornea to the level of Descemet membrane. The predisposed method is the big bubble technique, during which air bubbles press

the Descemet membrane. The endothelium, which is the main target of the immune process, is not transplanted, what is connected with a lower risk of rejection [7].

1. Indications

The basic indication for the procedure is corneal damage involving the anterior 95% of tissue thickness in the absence of scars or tears on the Descemet membrane and in the preserved structure of the endothelium. Second, the disease processes that carry an increased risk of rejection of the transplantation should be mentioned. For example, chronic atopic keratoconjunctivitis and cornea [7].

2. Advantages and disadvantages of the procedure

An undoubted advantage is the greater availability of transplantation material, because the quality of the donor endothelium does not matter. In the case of deep lamellar keratoplasty there is no risk of endothelial rejection, although the risk of epithelial rejection cannot be excluded. Compared to penetrating keratoplasty, a stronger eyeball structure and less astigmatism can be observed [7].

The disadvantage is the difficult and time-consuming technique of the procedure, which is associated with the risk of perforation in older patients [7].

Visual acuity obtained after surgery may be affected by haze between the recipient's Descemet membrane and the transplanted donor tissue [7].

3. Postoperative procedure

The principle is similar to the treatment after penetrating keratoplasty, but fewer steroid drops are used and the sutures can be removed after 6 months [7].

III Anterior Lamellar Keratoplasty

The procedure consists in cutting out the incomplete thickness of the corneal stroma, but leaving the endothelium and deep part of the stroma [7].

1. Indications

Corneal diseases with preserved endothelial function, corneal cone, post-inflammatory and post-burnal turbidity, Reis-Bucklers dystrophies (reticulate and granular) and limited thinning or forming *descemetocèle* should be listed here [7,8].

2. Contraindications

Corneal endothelial diseases should be listed here, as well as Descemet's granulomatous reaction, spotted dystrophy, Fuchs'; dystrophy, neurotrophic corneal degeneration, no tears, and corneal thickness in the center below 300µm [8].

3. The procedure technique

Technique is similar to the penetrating keratoplasty but applies only to a part of the corneal thickness [7]:

- preparation of the recipient's bed The first stage of the procedure is to mark the optical centre of the pupil. Next, using the Hesburg-Baron vacuum trepan, a cornea cut should be made to 80% of its thickness. Separation of corneal stroma from Descemet membrane is the most difficult moment and can be done with a spatula, air, viscoelastic, fluid or mirror effect after air injection into the anterior chamber. When completely uncovered, Descemet's membrane looks like a sheet of ice [8]
- preparation of the donor's corneal transplant (corneal graft).

The donor's corneal graft can be obtained in two ways: from the corneo-scleral graft or from the entire eyeball. In the first case, preparation starts with cutting out the full thickness of the cornea, whose diameter is equal to the prepared recipient's lodge. The cornea prepared in this way should be placed on a teflon block with the endothelial side facing upwards and the Descemet membrane should be rolled with a spongosteen stick. When choosing the second technique, the preparation should be started by cutting a scleral tunnel in the seam, through which the stroma from Descemet's membrane is then separated with spatulas. The level of Descemet membrane is determined using the Melles technique. After the stroma is delaminated, the cornea with sclera flange should be cut off and the endothelial side facing up must be placed on the Teflon block. Then, a graft with a diameter equal to that of the recipient's lodge should be excised with a trepan through the entire thickness of the cornea. The final stage is to remove the Descemet membrane from the cornea with tweezers [8].

- suturing a corneo-scleral graft

As recommended, it is best to use 10/0 single or continuous nylon sutures, taking care to control their tension intraoperatively with a manual keratoscope. It should be taken into account that in exceptional cases it is necessary to perform an anterior chamber puncture to close the wound tightly [8].

4. Intraoperative complications

The most common are perforations of Descemet membrane. They can be divided into those with small diameters - there is no complete abolition of the anterior chamber and perforations with large diameters - there is tearing. In the first case, it is sufficient to apply air under the surface of the cornea to allow the Descemet membrane to be applied. In the second case, you should proceed to a penetrating keratoplasty. Therefore, it is important that the

anterior lamellar keratoplasty procedure is protected by a cornea suitable for a penetrating graft [8].

During the procedure, an intra-corneal clot formed from a bleeding, pathological vessel may develop. In such a situation, it is necessary to expose the graft and thoroughly clean the Descemet membrane, as well as resuturing [8].

5. Postoperative complications

In rare cases, a non-adherence of the graft may occur. It is then enough to inject air into the front chamber to apply the Descemet membrane. Sometimes it is necessary to apply topical medication when the patient's intraocular pressure increases after the administration of intravitreal air. Sometimes there is a rejection reaction concerning epithelium and stroma [8].

Persistent mydriasis is extremely rare. It should be noted that in patients with perforated Descemet membrane, cataracts develop more frequently [8].

Chronic mild inflammation may be caused by the ingress of talcum particles, threads or epithelium [8].

6. Postoperative procedure

The patient may be discharged home the day after the surgery because there is no communication with the inside of the eyeball during the procedure.

- topical antibiotics for 10 days
- corticosteroids and moisturizing drops for two months

Suture removal is possible faster than in case of penetrating keratoplasty, because from 4 months after the procedure. Coherent tomography is useful when guiding a patient after surgery [8].

7. Conclusions

Compared to a penetrating keratoplasty, patients require a shorter recovery period and visual acuity is comparable in these patients. It is worth noting the greater safety of lamellar keratoplasty. A disadvantage is the long time of the procedure and the time needed to train the operator [8].

IV Posterior Lamellar Keratoplasty

The procedure involves transplanting the posterior corneal stroma from Descemet's membrane and endothelial cells into a lodge from which similar anatomical structures were cut out in the recipient [8]. When only Descemet's membrane with endothelial cells at the donor is removed, the treatment is called descemetorhexy [8].

1. Indications

The procedure most often affects patients with Fuchsa dystrophy and pseudophakic and aphakic corneal oedema. There is a loss of corneal transparency with swelling as a result of abnormal endothelial cell function [8].

2. Contraindications

They are associated with the presence of scars and vascularization in the corneal stroma [8].

3. Operating technique

- preparation of the recipient's lodge

The first stage is to mark on the cornea the diameter of the excised endothelial petal and incision of the conjunctiva in the limbus (from 1. to 5 o'clock) while coagulating the sclera vessels. It is important that the sclera should be cut in a linear manner 5mm wide at a distance of 3mm from the cornea. In the next stage of the procedure, two types of knives are used: using crescent knife, the mid-corneal tunnel should be hollowed out; a slotted knife is used to enter the front chamber. The administration of air and the transplanted petal occurs after two paracentesis- on both sides of the cut. Then, a viscoelastic is administered into the anterior chamber and with the inverted hook Sinsky we scratch the Descemet's membrane. To completely remove the viscoelastic substance, the front chamber is filled with BSS solution. In patients who cannot lie flat after surgery, Melles' original technique can be used [8].

- preparation of the endothelial flap

The basis is that donor cornea has a minimum of 2800 cells per mm².

Two ways of preparing an endothelial flap at the donor should be distinguished: from the corneo-scleral graft or from the entire eyeball. The technique is at the operator's choice: manual or automatic (requires a special microkeratom) [8].

In a similar way as in the recipient, the eyeball should be cut by layering the cornea with a spatula, but at the depth of the Descemet's membrane using the Technique of Melles (mirror image). Then, a corneo-scleral graft should be placed on the block endothelium upwards. It is important to separate that the Descemet's membrane from the rest of the cornea, and apply a drop of viscoelastic substance on the endothelium. In the artificial ventricle (filled with a viscoelastic), the corneo-scleral disc should be placed, and then the cornea should be delaminated [8].

The advantage of the automatic method is the smoothness of the stroma surface used for the transplant of the flake and its repeatability. The disadvantage is the thickness of the flake, as well as the high cost of the microkeratom [8].

- implantation of the endothelial flap

Marking the stroma side of the petal with a medical mazama. The prepared petal should be spread over 60/40% and inserted through the tunnel into the front chamber with McPherson tweezers. In order for the petal to develop properly, through paracentesis, air is injected into the anterior chamber. The cross seams close the tunnel exit. It is important to squeeze the liquid out of the space between the petal and the corneal stroma. This should be done by smoothing the anterior surface of the cornea with a spatula. In case of increased tension of the eyeball, it is necessary to drop the air. The final stage of the procedure is the administration of atropine 1%, timolol 0.5% and antibiotic and corticosteroid [8].

When Melles technique is used, air should be replaced with BSS in the anterior chamber. Then the use of atropine is not necessary [8].

4. Post-operative procedure

It is necessary that every patient after the procedure lay on the back in order to better press the transplanted graft through the air (this does not apply to Melles's technique, as there is no air left in the anterior chamber). It is important to constantly control intraocular pressure, especially during the first two hours after the procedure. The pressure increase above 35 mmHg is an indication to drop the half of the air from the front chamber. Beta-blockers, antibiotics and corticosteroids are administered topically. In the seventh day, the patient can be discharged, with a recommendation for monthly control. Steroid drops should be used 4 times a day for three months [8].

5. Intraoperative complications

The most common include confusion between the sides of the petal (the sides of the petal should be marked in advance), the iris falls through the tunnel (requires reposition) and bleeding under the petal and the anterior chamber (cleaning the cornea from clots and precised rinsing) [8].

6. Postoperative complications

- dislocation of the petal (it should be controlled by optical tomography, sometimes it is necessary to administer the air into the anterior chamber)
- increase in intraocular pressure after air administration into the front chamber
- rejection of the transplant
- the loss of endothelial cells with time is 12% after a year, while after two years - 27% [8]

7. Conclusions

Compared to penetrating keratoplasty, the advantages of this method are: the ability to replace only the corneal layer, which has been pathologically altered and leaving its healthy parts. Visual acuity without correction is much better after the procedure, and the recovery time is shorter. In the procedure performed with this technique, the problem of loosening of the seams, as well as the wound dehiscence was minimized, because the operation does not affect the curvature of the cornea [8].

Scleral graft

The sclera has been used in ophthalmic surgery for many years, this tissue is very widely used. Sclera is one of three membranes - the outer membrane, building the walls of the eyeball. It is made of collagen fibers, surrounds the eye from the orbital side, passing in the anterior part of the eyeball in the cornea, and in the posterior part into the optic nerve capsule. The tissue consist of three layers, the outermost episcleral plaque, the correct layer of the sclera, the brown plaque sclera that adheres to the uveal membrane. The sclera's function is to protect the inside of the eyeball, stabilize, and maintain its proper shape [1,4].

Use of the tissue

1. Wrapping orbital implants

The sclera is commonly used to wrap orbital implants inserted after enucleation of the eyeball. This procedure involves removing the eyeball along with part of the optic nerve, while leaving other nerves, blood vessels, orbital fat, eyelid and muscles moving the eyeball. Instead of an eyeball, an orbital implant is implanted in order to maintain the proper structure, better cosmetic effect and facilitate the mobility of each subsequent eyeball prosthesis. The presence of an orbital implant also prevents "enucleation syndrome", which is characterized by collapse of the orbit, drooping of the upper eyelid and deepening of the furrow in the skin of the upper eyelid. This syndrome can cause problems when choosing the right prosthesis and cause facial asymmetry, which is why it is so important to implant such implants to children to allow the correct, symmetrical development of the facial skull.

Wrapping the orbital implant with other tissue is used to prevent migration and exposure to infections of the implanted orbital implant. The materials used for this purpose are very diverse, scleral or donor sclerosis, broad thigh fascia, temporal muscle fascia, donor dura mater and even bovine pericardium. The latter is due to the high costs and potential risk of contracting Creutzfeldt-Jakob disease through tissue transmissions. When comparing the use of bovine pericardium, the use of sclera is cheaper and has fewer postoperative complications, so it is

safer because after testing it was shown that cattle pericardial transplantation in 23% of operations has a risk of complications compared to sclera transplant, after which the number of complications was 3, 8%.

Postoperative complications when using the sclera are less common than when using other alternative materials during wrapping. The key aspect is the contractor's surgical experience. Complications have been shown to be more common in operations performed by less experienced surgeons, complicated cases and requiring additional surgery are also associated with additional risk. Infections are a rare risk of complications and usually affect the implant itself, not the surrounding material [F,G,H].

2. Thinning of the sclera

Scleral thinning may occur as a result of scleritis, which is an important etiological factor of autoimmune diseases such as rheumatoid arthritis, systemic lupus erythematosus, systemic sclerosis, Wegener's granulomatosis, psoriatic arthritis, nodular arteritis, Reiter's syndrome or giant cell arteritis. Scleritis can be divided into anterior diffuse necrotic, where the inflammatory process covers the entire sector or anterior part of the sclera, anterior necrotic nodules, accompanied by the formation of a nodule in the sclera, and anterior necrotizing inflammatory process, this is deep irritation which is diffuse or sectoral. Scleral transplants are used for the latter - necrotizing scleritis, which causes thinning. It usually occurs in women with rheumatoid arthritis. The large majority of patients are cured with pharmacotherapy, but in the case of 5 - 10% surgical intervention is needed to eliminate sclera defects, repair perforation in the eyeball, or cover the translucent choroid.

The first who use the term "scleromalacia perforans" to describe degenerative scleral lesions was Van der Hoeve in 1934. He and suggested that the use of mucous membranes may serve as transplant material.

Since then, various materials have been used, including broad fascia, periosteal tissue, aorta, polytetrafluoroethylene, but the most popular and most commonly used material is the sclera [G,I,J].

Surface cancer and eye melanomas can also cause thinning of the cornea. Thinning of the sclera can occur directly as a result of cancer invasion, or secondary through local resection or radiotherapy (up to 12% of radiotherapy treatment for melanomas has caused necrosis). However, studies show that scleral transplantation is not the first choice in this case, because a large amount of tissue transplant is usually necessary [G].

3. Corneal diseases

In the case of extensive corneal perforation, the sclera may be an emergency option in the absence of corneal tissue available. Thanks to this, it is possible to restore the integrity of the eyeball [G].

Further more a scleral patch can also be used in other diseases, e.g. corneal ulceration, perforation in neurotrophic keratopathy, tear deficiency syndromes (Sjogren's syndrome, GVHD), corneal perforation in eyes not eligible for corneal transplantation or in the absence of corneal transplantation [G].

Methods of sclera retrieval

In order to prepare the tissue for transplant, all eyeball muscles and the donor's conjunctiva should be removed. Then all remaining contents of the eyeball are removed - retina, choroid, lens, vitreous humor. This is possible by rolling the entire eyeball and cleaning the residue by scratching the inside of the eyeball with a hockey knife, which is overturning on the outside. The tissues thus prepared can be stored until they are needed for the transplant. There are many different methods of tissue storage, each with its advantages and disadvantages.

Storing dry tissue is the most common way. The sclera is placed for 7 hours in a 70% ethanol solution, then for 24 hours in a 96% ethanol solution. After this time, the transplant should be cleaned with sterile, isotonic NaCl solution, and finally the sclera is dried thanks to the residual heat of the hot air sterilizer. The transplant prepared in this way can be stored for at least a year in a sterile sample in the refrigeration device. Another method is to store the sclera in a 90% methanol solution for a long time at a temperature of 4-8 degrees without any pre-treatment. The use of ethanol is very beneficial because of the low costs, as well as antibacterial, fungicidal and virucidal properties [F]. Another method described is the technique of freeze-drying, i.e. freezing tissue at a temperature of -20 degrees, in antibiotic solution for up to 3 months. The main advantage of this method is the ability to accurately assess the size and shape before the transplant. The donor's sclera can also be stored in glycerin, the use of which seems to be more favorable compared to other materials, because the collagen fibers from which the sclera is built, showed a more regular arrangement than tissue stored on other substrates, thus having greater integrity and are more resistant to stretching compared to other carriers, which is a big advantage, due to the main reasons for using the sclera, i.e. restoring the structural integrity of the eyeball. The tissue can be stored in glycerin for 12 months.

Gamma radiation has also found application as a method of tissue storage. Irradiated tissue is attractive in that it carries a low risk of transmitting any disease to the recipient through sterilization. The tissue prepared in this way can be stored at room temperature for up to 18

months. In some cases, sclera changes have been reported due to radiation exposure, but this risk can be mitigated by modifying the maintenance technique [G].

CONCLUSIONS

Eye tissues obtained from deceased donors are undoubtedly widely used in modern medicine. The development of tissue banking and microsurgery techniques, the use of stem cells, and modern technologies such as femtosecond laser or computer software for microscopes have certainly created many new therapeutic opportunities. With corneal and sclera transplants many people regain their sight. The cornea is a kind of window on the world, which can be closed due to disease. Transplants are coming to help. They allow older people to see their grandchildren for the first time and people with congenital defects to see the world. From the patients' point of view, such surgeries are treatments that give new life. It is a priceless gift that the deceased donors give. Death is not only cruel and ruthless but also life-giving.

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Ecology of viruses

Borkowski Filip¹, Kasprzak Hubert¹, Jasiura Adam², Ludwig Bartłomiej²

1. Faculty of Biological Sciences, University of Wrocław, Wrocław
2. Medical University Piastów Śląskich of Wrocław, Wrocław

INTRODUCTION

Viruses are obligatory parasites that can replicate only in living organisms and cannot really live on their own. Although they are not classified into the three main domains of the life, they infect representatives of each domain and significantly affect the evolution of ecosystems. Since they are remarkably diverse, viruses were divided depending on the host they acquire, into viruses of bacteria (bacteriophages), archaea and eucaryota.

What is more, they can also infect other viruses (satellite viruses) – which is called hyperparasitism.

The viruses emergence is an amazing biomedical challenge which humanity is facing today. Humans have been suffering from virus diseases for a long time.

Nevertheless, more than 10% of human DNA is thought to be viral. Genes encoded by this DNA produce proteins, which affect our health in a variety of still unknown ways.

Viruses consist of nucleic acid enclosed in a protein shell – called capsid. They are not capable of ATP synthesis or other metabolic changes on their own, however they can stimulate hosts' expression of enzymes involved in these pathways [1].

Due to the lack of the cellular structure, they are not recognized as living organisms. Still, they are subject to natural selection – not only as virions but as the DNA molecules too, as Orgel noticed [2].

Due to this fact they can better adapt to the environment which they live in. Through evolution, viruses acquired various infective mechanisms in order to achieve better adaptation. The only way of the inheritance and further passing of these traits is through double-stranded or one-stranded DNA or RNA.

Therefore, viruses have much faster reproduction capacity than all other organisms. Hence, they may be the most evolutionary successful Earth inhabitants.

A COMPREHENSIVE VIEW OF VIRUSES

Some researchers believe a virus can be entirely defined by its encoding capacity. The latest studies prove that genome transplantation from one species to another is possible. It has been proven by transplanting the genome of *M. mycoides* into a cell of another species. As the result the phenotype of the cells was identical to *M. mycoides* [3]. This experiment suggests that organism can be completely defined through genome analysis. What is more, there are many giant viruses with abilities have not been seen before. *Mimivirus* is the largest virus found up to now. It contains a 1.2 mega base pair chromosome that encodes 1 000 putative genes. *Mimivirus* was thought to be *Legionella*-like organism in the past, as in the microbiological examinations it was Gram-positive. Due to such an enormous genome, *Mimivirus* acquired mechanisms have not been previously known in any other viruses. It can encode a part of the translational machinery, which is done entirely by host in other viruses. Further, *Mimivirus* as well as its relatives have their own viruses – called *Sputnik virophages*. Once discovered, it was a completely new virus-virus interaction – called hyperparasitism. Moreover, studies show the *Sputnik*'s genome contains genes from *Mimivirus*, which suggests virophages may serve as horizontal gene transfer carriers [4]. What is more, comparing the genomic sequence of *Mimivirus* and the smallest cellular organisms (*Candidatus Carsonella ruddii* - a bacteria, *Nanoarchaeum equitans* - an archaeon, *Encephalitozoon cuniculi* - an eukaryote) may lead to a conclusion, that there is only a one significant difference between cellular organisms and viruses. It is the number of genes involved in the translation and the energy production. However, it is not a significant difference – viruses have a parasitic mode of life and the reduction of the number of genes involved in such processes could be caused by the convergent evolution, which occurred many times independently [3]. Thereby, it could be possible for viruses to originate from DNA fragments of cellular organisms, which got independent from cellular control and became parasites. It was called the escape hypothesis [5].

THE ENVIRONMENTAL ADAPTATION

During their evolution, viruses have developed various mechanisms facilitating their beneficial adaptation to the environment. The continuing "evolutionary arms race" led to the creation of various strategies that, when they are used together, effectively oppose the host's defence mechanisms. Viruses can adapt depending on environmental conditions. Their populations have to evolve in heterogeneous spatial environments. Further, their hosts are

multicellular, and every cell of them pose a different selective pressure. Due to the populations polymorphism caused by mutations and other factors, natural selection can promote subpopulations specialized in a narrow subset of the cells; or it may select the viruses that are generalists. Recent studies provide a great *in vivo* example of this process. Studies show, *Vesicular stomatitis virus* (VSV) populations which had grown either in homogenous tissue cultures or heterogeneous host cell mixture, evolved in these environments. However, VSV lineages from the spatial heterogeneous colonies could better adjust to the variance of later infected hosts than the lineages from the homogenous environments [6]. Generalist's phenotypes dominated in all viral populations in all kinds of environments. Furthermore, viruses can coevolve with the host using synonymous codons in the translation. Eighteen from twenty proteinogenic amino acids are encoded by more than a one synonymous codon, which are not used in equal frequency. Each organism has its own codon use bias (CUB). Most of viruses do not encode any tRNA since they have a compact genomic structure. Therefore, translation of their DNA is dependent from host's tRNA, resulting due to that strong relationship in the similarity of the viral and host's CUB. Further, excessive expression of viral proteins decreases the translational efficiency of the host, as it causes the repulsion between the CUB of the virus and the host's [7].

HUMAN VIRUSES

A good example of a virus with excellent environmental adaptation is influenza virus, in particular – type A, which belongs, next to types B and C, to the *Orthomyxoviridae* family. Their carrier of genetic information is RNA [8]. Within type A, viral surface proteins are crucial - hemagglutinin (H) and neuraminidase (N), since the division into subtypes is based on them [9]. The most important mechanism, which enables influenza virus to cause annual seasonal epidemics or pandemics, is mutagenesis. The variability of genetic material may occur either in single base pairs, then it is called antigenic drift; or larger RNA fragments, called antigenic shift. An antigenic shift can result in the creation of a completely new virus subtype when the cell is simultaneously infected by two different types of viruses. Because the genome of influenza virus consists of eight different segments, the number of potential combinations is 256. Therefore, if the antigenic shift includes genes encoding neuraminidase or in particular hemagglutinin, pandemic outbreaks occur as people could not have acquired specific immunity against newly emerging pathogens [8,10-12]. Over the past 120 years, the pandemic has been caused by the following viruses: A/H1N1 (so-called "Spanish"), A/H2N2 (so-called "Asian

flu"), A/H3N2 (so-called "Hong Kong flu") and recently A/H1N1v (so-called "swine flu") [13]. Influenza virus is able to infect various species and to transmit the virus between them. Particularly important carriers, which can potentially result in the contagiousness to humans include: birds (H1-H14 and N1-N9), pigs (H1N1 and H3N2), horses (H3N8 and H7N7) and marine mammals (H7N7, H13N2, H13N9 and H1N3) [14]. The mechanisms of crossing the interspecies barrier, especially on the birds-mammals line, have not been thoroughly studied so far, but various differences in the genetic material of viruses infecting these animals have been observed. Studies show, these differences concern mainly the genes encoding the PB2 subunits of the polymerase [12,15].

Another virus perfectly adapting to the host organism is the human immunodeficiency virus (HIV). It belongs to the *Lentivinae* subfamily of the *Retroviridae* family. Its genetic material, similarly to influenza virus, is ribonucleic acid [8]. HIV is thought to have unusual virulence mainly due to three mechanisms: a huge number of mutations; killing cells responsible for fighting infection and viral latency by integration of its own genome with the host cells'. Mutagenicity is important in particular, as it is probably responsible for HIV's "slipping out" of Tc lymphocytes control after their initial management of infection [16]. Human immunodeficiency virus, primarily present in chimpanzees, crossed the interspecies barrier at the beginning of the twentieth century, but it was discovered in the 1980s as a consequence of a sudden increase in the incidence of opportunistic infections [17]. This transition between species occurred three times and each time different phylogenetic lines were created: M, N and O. Then the viruses from individual lines evolved further and more lines developed. Despite such diversity, the world has been virtually dominated by only one form - HIV-1. [18,19].

Interestingly, viral carriers can adapt to the virus as well, thereby they do not show signs of the disease, or if any symptoms occur, they are much less expressed than in other infected animals. Such a relationship has been observed in bats infected with rabies virus, which is an RNA virus belonging to the *Rhabdoviridae* family [20]. The occurrence of disease symptoms in humans, after being bitten by a carrier, is practically synonymous with death [8]. The virus quickly penetrates to the central nervous system using peripheral nerves causing severe encephalitis [11]. However, bats either do not develop symptoms at all or only after many months or even years. This phenomenon is suggested to occur due to the increased temperature of the bat's body, resulted from the animal's flight, as it mimics the inflammatory response in fever. Bats are also a reservoir for other pathogens dangerous for humans, such as coronaviruses, probably due to the same defense mechanisms [20].

VIRUSES OF MICROORGANISMS

As viruses prey on organisms of all three domains of life, they are considered to have major impact on ecosystems. Not only are they the source of the environmental changes but also that altering environment affects them. As the influence on the inter-organismal relations is multileveled, focusing in this paper on the bacteriophages – the most commonly studied viruses of the microorganisms (VoMs), the ecology can be divided into: population, evolutionary, organismal, community and ecosystem ecology [21,22].

Population ecology

Depending on the factors, which influence the VoMs populations and changes in the growth rate, extent and distribution of these populations, the viral ecology can be analyzed as a population ecology. It enables the evaluation of the viral impact on the environment in the local or global scale as well as seasonal and climatic [22]. It is thought as they prey on numerous hosts, VoMs are the most abundant organisms in the world [23]. They can be found in the soil, atmosphere and aquatic habitats – both marine and freshwater. However, their distribution and extent are primarily dependent on the distribution of hosts they infect. Hence the hosts population is in this case crucial. The amount of the viruses is mainly defined by their ability to reproduce. Most viruses, except fungal viruses, infect their hosts by direct cell-to-cell contact, therefore their communicability is defined by host-virus collisions. Supposing the deterministic model, the number of infections increase in the direct proportion to the hosts and the viruses populations, resulting in exponential growth, varying on its rate with the viral life pattern (lytic, lysogenic or chronic). Contagiousness of the viruses can be also affected by their morphology and size since they influence the diffusion rate and the collision chance. Nevertheless, the stochastic factors actually impact the populational dynamics as well – including multiplicity of infection to the host, which leads to decreased growth rate of the viruses due to the reduced resources [24].

Evolutional ecology

In dependent populations, such as hosts and VoMs, theoretical predator-prey model suggests the occurrence of the coevolution, where organisms evolve interrelatedly on the feedback basis. Such coevolution should last long, resulting in cyclic fluctuations of the hosts population. Simultaneously VoMs should maintain hosts population under the maximal amount, defined by the niche capability [25]. Experimental researches of this model seem not

to confirm it effectively. As far as the hosts population resembled the theoretical calculations, the predators population was relevantly lesser. What is more, some studies show even total extinction of the VoMs population due to their purge of the hosts and therefore losing the vital resources; or due to the resistance acquisition by the hosts (without their extinction). Therefore, studies suggest the coevolution in experimental models occurs rarely. The reason can be the difference in the predators and preys evolutionary potential. Hosts mutate quickly and have an abundant population. It leads to the VoMs response, which due to the even greater population, mutate even faster to adapt to the new conditions. However, hosts due to the greater genome have better ability to mutate towards gaining resistance against the VoMs infection. Eventually VoMs are not able to counter the resistance. Therefore, their ability to survive depends on the presence of the VoMs-sensitive hosts and the VoMs capable to survive without the VoMs-susceptible hosts, which can later recover the population [24,26]. Furthermore, some microorganisms developed defense system against the exogenic genetic material – called CRISPR-Cas. It uses the sequences of the phage genetic material, that infected the microorganisms in the past. The functional principle is to detect and destroy the similar to the saved genetic material in the event of another infection. Therefore, this mechanism is considered antiviral [27].

However, the CRISPR-Cas system is not unerring. There has been discovered the viruses of the *Phikzlikevirus* genus, which are *Pseudomonas* phages. They developed the nucleus-like structures in the bacterial cytoplasm, thereby creating a new compartment for the viral DNA, protecting it from the cytoplasmatic CRISPR-Cas effector complexes. Viral genetic material is surrounded by phage proteins and is located in the center of the bacterial cell using the phage-encoded tubulin spindle. Though, the nucleus-like compartment cannot protect the RNA produced by the transcription apparatus, which can be destroyed by the RNA-targeting immunity as it moves to the cytoplasm. Nevertheless, the RNA production is so intense, the immunological mechanisms are not able to keep up. There are suggested promising possibilities of using such viruses in the phage therapy of the infections by bacteria, which are resistant to the known antibiotics. However, as a still incomplete method, it requires further studies, before it can be used clinically [28].

Another important aspect of evolutionary ecology is the competition between the viruses. Natural selection promotes the predator, which better manages to acquire the greater amount of the resources. Viruses compete for the host not only extracellularly but also intracellularly. In case of MOI (multiplicity of infection) greater than 1 – when one host cell is infected by more than a one viral particle – viruses compete with themselves for the resources of the host. For

example, viruses can use capsids with the proteins produced by other concomitant viruses to achieve a greater reproductive success. They can also block the adhesion or internalization of competitive viruses, in case of the infection by viruses of different species [24,29,30].

Organismal ecology

The changes relating to the virus itself are relevant as well. They involve the organismal response to the altering environmental conditions in which virus is currently located, including the change of the inhabited ecosystem or the form of the virus – from the host-infecting to the virion or free-living form. It can be manifested by – for example – restriction of the viral adsorption in the conditions unfavorable for increased reproduction of the virus. Studies prove, bacteriophage T4 is able to retract its whiskers depending on the temperature and the ionic strength of the environment. The whiskers are crucial to proper adsorption of the phage to the host cell. Such a behavioral response is accomplished by conformational changes of the viral protein subunits. It leads to the retraction of the tail fibers and therefore prevention of the adsorption in a location, where number of bacteria is restricted and thereby disallowing the effective reproduction [31].

Another example of the adaptation to the conditions the virus inhabits is pseudolysogeny. Such a change of the life pattern occurs when the resources of the host are restricted and the cell is starving, thereby it cannot provide enough supplies to effective viral replication. Pseudolysogeny consist of withholding the phage development in the host cell without the degradation of the virus. Viral genetic material does not replicate neither in multiplication in the lytic cycle nor with the host's genome in the lysogenic cycle. It remains inactive in the host cell without integration with the host's genome, therefore in case of the division of the host cell it is unequally distributed into progeny cells. If the environmental conditions improve and the host starvation is terminated, the virus enters a true lytic or lysogenic cycle [32,33].

Community ecology

A basic unit described by the community ecology is the relation between the host and the predator in the micro scale. It includes how viruses impact the abundance and the diversity of the host population as well as how hosts influence the VoMs populations. Over time, due to the evolution of the predation mechanisms, viruses developed various ways of interactions with their hosts. These include the most virulent cases, leading to the host's death in the lytic cycle; lysogenic cycle and pseudolysogeny, where genetic material of the virus is inserted into the

host's genome, resulting in the acquisition of the new functions due to the enlargement of the gene pool; as well as non-lethal chronic infection. Therefore, viruses can influence the richness and the diversity of the population, increasing the fitness of their hosts, which result in better adaptation to compete for the niche with other organisms of the ecosystem. Such cases were observed in the gain of the type IV secretion system by *Vibrio cholerae* [34] or phage-encoded diphtheria toxin in *Corynebacterium diphtheriae* [35].

What is more, viruses enable a gene transfer between the organisms, not only theirs but also host's, what is called lateral gene transfer. That include transformation, transduction and conjugation. It is thought to influence the speciation, but additionally viruses act there as a gene reservoir, leading to the organisms specialization, their better adaptation to the environment of the fragmented ecological niche, allowing the increase of the biodiversity of the ecosystem [32].

Concurrently, viruses are considered as ones of the most important predators of the prokaryotic cells. They play a major role in prevention of the ecosystem domination by one – the most adapted by the natural selection – prokaryotic species. Such model has been called 'killing the winner'. It assumes the presence of the highly species-specific lytic viruses, which in the case of the excessive growth of the hosts population lead to the increased lysis of their cells, as a result of the escalated growth of the viral population. It prevents the domination of the ecosystem by only one population decreasing the realized niche by the hosts and expanding it for the other organisms [32].

Such a phenomenon has been observed for ones of the smallest marine bacteria – *SAR11*, which simultaneously are ones of the most abundant organisms on Earth. Despite being thought too small for phage predation or multiplying too slow for an effective viral infection in the past, now these hypotheses has been disproven. Studies have demonstrated in the *SAR11* cells the presence of *HTVC010P* phage, which belongs to the new *Podoviridae* subfamily more abundant than any seen previously. The dynamic equilibrium between the host and predator regulates the domination of each population in the ecosystem, thereby influencing global carbon and nitrogen cycles [36].

Furthermore, it is possible to use above viral mechanisms to fight infections caused by pathogenic microorganisms, which is called the phage therapy. It requires administration of a bacteria-specific virus in order to eliminate or reduce the nuisance bacteria population. Phages destroy the bacterial cells in the obligately lytic cycle solely, therefore bacteria cannot quickly regenerate or acquire the resistance to the therapy, since the lysogenic or chronic cycles are precluded. Due to the high phage-host specificity, this therapy can be applied only to the selected bacteria species. Therefore, it does not disrupt the human microbiota, on the contrary

to the antibiotics, which have a much wider spectrum. What is more, the therapy is auto-dosing, as viruses reproduce to the required abundance on their own after just one appliance. It can be administered in many dosage forms, in the combination with the antibiotics to widen the spectrum as well. It impacts the environment narrowly too, since phages are quickly inactivated after they get out of the host, unlike the antibiotics, excreted mostly in the unchanged form. However, finding a proper phage, which is obligately lytic, has high virulence towards targeted bacteria and simultaneously does not carry any toxin genes, turned out to be highly difficult. Bacteriophages have very narrow therapeutic spectrum as well – even narrower than the narrow-spectrum antibiotics. Nevertheless, as bacteria are gaining more and more resistance to known antibiotics, phage therapy is an extremely promising treatment. It has many advantages and relatively little weaknesses, however it still requires further studies in order to use phage potential at its fullest [37,38].

Ecosystem ecology

By expanding the concept of community ecology with abiotic factors, it is possible to analyze the viral influence on the ecology of the whole ecosystem – how the viruses impact the carbon cycle and the energy flow in the ecosystem. Studies show, viruses play a major role in the mineralization – decomposition of other organisms to the inorganic matter. It consists of the bacteria lysis and results in the release of the nutrients to the extracellular environment. It is especially noticeable in the aquatic habitats, where viruses cause the disintegration of the cyanobacteria.

These bacteria bind CO₂ producing organic forms of carbon, which are available to the higher trophic level consumers. However, the fraction of that carbon despite being consumed is not transferred to the higher trophic level. Viruses, by the decomposition of the cyanobacteria enable the return of the carbon to the primary cycle as DOM – dissolved organic matter, which can be reused by heterotrophic bacteria. Moreover, bacteriophages lead to the liberation of the enzymes, mainly hydrolytic – ectoenzymes, which decompose the organic matter. It can be observed in the soil decomposition, where viruses increase the amount of the inorganic nutrients, which are more soluble and easier to absorb. Further, phages can transfer the enzymes encoding genes – called exoenzymes, which due to the modification of the infected host's surroundings, can secondarily influence abiotic factors of the environment. Besides, phages can transfer carbon and energy as themselves and as they are included in many cycles, these nutrients can be reused [39].

Plant viruses

Since plants produce more than 80% of the biomass and one of the key elements for life - molecular oxygen, the ecology of plant and algae viruses is important as well. Studies show plant viruses have the greatest impact on the functioning of ecosystems compared to viruses of other organisms [40]. Recent discoveries show that plant viruses should not be treated solely as crop destroyers, contrarily – they can be potentially applied for protection of natural ecosystems against invasive species or reduction of pesticides use in agriculture. In this part of the paper, we present a brief overview of ecological mechanisms, which plant and algal viruses are subject to.

Different strategies of algal and plant viruses

The genetic diversity of viruses is greater than in cellular life forms [41-45]. This is due to the complex interactions with many genomes that viruses are subject to. While vectors are not needed for algal viruses, adaptation to a vector may be more important than adaptation to the host itself for some terrestrial plants viruses. Unlike animal viruses, plant viruses are rarely transmitted through the direct contact between infected and uninfected individuals [46-48]. Transmission through natural root grafts or contaminated soil or water is possible [49], but the most effective transmission occurs through vectors, pollen or seeds [40]. The most common vectors of plant viruses are insects, mainly belonging to the *Hemiptera* order. They transmit about 70% of known plant viruses [50]. Hemipterans are particularly well suited to transmitting plant viruses because of their needle-like mouthparts, which they use for sucking sap and/or the contents of plant cells. There are two ways to transmit viruses through insects: non-circular and circular. The non-circular method only involves the single transmission of the virus along with the saliva of the insect. Whereas the circular one is more sophisticated - ingested viral particles traverse the gut epithelia and enter the haemocoel before moving to the salivary glands. Furthermore, there may be interactions with insect endosymbionts. This allows for uninterrupted transmission of the virus in the body until the death of the insect [40]. In addition, viruses can manipulate the behavior of vectors by making infected plants more attractive to insects preying on them and/or by producing substances that promote viral transmission behavior. On the other hand, viruses that are not transmitted by vectors can promote host survival by making infected plants chemically repellent for herbivores. Moreover, exclusively most of, if not all, plant viruses probably evolved from viruses that in the past infected non-plant organisms. This is confirmed, for example, by the fact that most genera including plant viruses have genes and/or genomic structures homologous to arthropods and/or fungal viruses.

It cannot be denied that some of the viruses, which are now considered to be exclusively plant viruses, are in fact capable of infecting also fungi. This may be partly due to the close interaction of plants and fungi - mycorrhiza. However, fungal viruses were only discovered in the 1960s and are not known as well as those infecting plants and animals [51].

An interesting feature of some plant viruses is the occurrence of multipartite genomes, which are packaged as individual genome components into separate viral particles during transmission. This allows a reduction in the size of the virus particles and easier penetration through plasmodesmata [40]. As mentioned before, viruses which do not use vectors can even contribute to fending off herbivore attacks, and thus, to some extent, act in favor of the host. Conversely, higher virulence is expected for viruses transmitted through vectors. Plant viruses usually have a host range that includes several species from one or more different plant families. However, they are rarely transmitted by more than a few very closely related species of insects [52]. It seems that acquiring a new vector by the virus is a much more difficult than acquiring a new host. It was confirmed by the analysis of the capsid proteins of geminiviruses. The phylogenesis of these proteins overlaps significantly better with vector species than with host species. Thereby, many new host species may have been infected by viruses due to the polyphagy of their vectors. Many viruses quickly adapt to the genotypes of new hosts, which in some cases causes epidemics [53].

When using multiple hosts, there may be a problem of competition with other viruses or organisms for the same resources. In this case, the adverse phenomenon is antagonistic pleiotropy, i.e. a situation in which mutations favorable for controlling one host become unfavorable for another. On the other hand, specialization will be promoted when vectors are mono- or oligophagous. In animal viruses 'no-cost generalists' are observed, i.e. viruses that are able to overcome antagonistic pleiotropy and adapt to infect more species [54-56]. Only one experiment has shown such a property of the virus in plants so far [57]. However, the situation is different in algae viruses. They do not need vectors, which means that other virus parameters, such as the ability to infect the host, latent period, burst size and swimming ability are gaining importance. Latent period is the duration of the viral infection in the host and is divided into two types: lytic and lysogenic [58]. While lytic viruses cause host cell death and release a large number of offspring quickly after infection, lysogenic viruses are able to form stable interactions with their hosts by integrating genetic material into the host genome or maintaining it as plasmids – called pseudolizogens [59]. Lysogenic viruses are less effective than lytic viruses [60]. Their direct impact on the ecosystem is not so strong because at the same time they produce fewer offspring than lytic viruses. In addition, it can be expected that in low

productivity areas, infecting hosts of many species will be favored. Algae try to free themselves from the effects of viruses by reduction or total loss of cell wall receptors recognized by viruses [61], producing virus inhibitors [62], biofilm [61] and even colonies [63]. An interesting mechanism occurs in *Emilian huxleyi*, which performs meiosis in the presence of the virus, because haploid cells are resistant to the virus [59].

Impact of plant and algal viruses on ecosystems

Under the conditions of the contact of agricultural ecosystems with more natural ones, there is a constant transmission of viruses from non-cultivated to arable species, with the percentage of infected individuals of the arable species remaining at a higher level. In stable, unmanaged ecosystems, viruses contribute to the preservation of genetic diversity, preventing the dominance of the homogeneous population of one species [64,65]. As for invasive species, on the one hand, the virus may not inhibit invasion if the invasive plant species has less natural pathogens than the native plant species [66] or if the invasive species increases susceptibility to the virus in native plant species [67,68]. On the other hand, a scenario is possible in which the invasive species is not resistant to viruses in the new environment, and failure to adapt the virus to the new host may result in high virulence [69]. Although, as mentioned earlier, viruses that do not use vectors may be somewhat beneficial to the host plant, there is still insufficient data to support this thesis, and the results of experiments tend to be contradictory [40]. It leaves a lot of space for future research on the interaction of viruses with plants. The results of these studies can be used to protect natural ecosystems and reduce pesticide use. Regarding aquatic ecosystems, the structure and diversity of microbial communities can be modified by viruses by selectively removing organisms. It may release some species from predator pressure and/or stimulate other trophic levels. In particular, in phytoplankton, viruses are able to remove 25% of photosynthetically bound carbon [70] and reduce phytoplankton blooms [71]. As recent discoveries show, high CO₂ concentrations and the associated low pH have been linked to higher viral decay in aquatic systems, which in the face of global warming and ocean acidification creates the need for further studies.

CONCLUSIONS

In order to more effectively control the dynamics of viral populations, a deep and multilevel insight of viruses and the environment, which they live in, is required. It is significant not only in the eradication of viral diseases through the reduction of their adaptation, but also

in phage therapy, where viruses are allies of humans. In-depth knowledge of the ecology of algal and plant viruses will allow a better management of both artificial and natural ecosystems. It is possible that some of these studies can be transferred to the ecology of human viruses, which will also benefit medicine. Moreover, the awareness of viral ecology is useful in molding the natural environment – which directly depends on the biogeochemical cycles, modulated through interactions between microorganisms and phages. Therefore, viruses should be furtherly studied, as the understanding of their ecology can be beneficial for humanity.

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