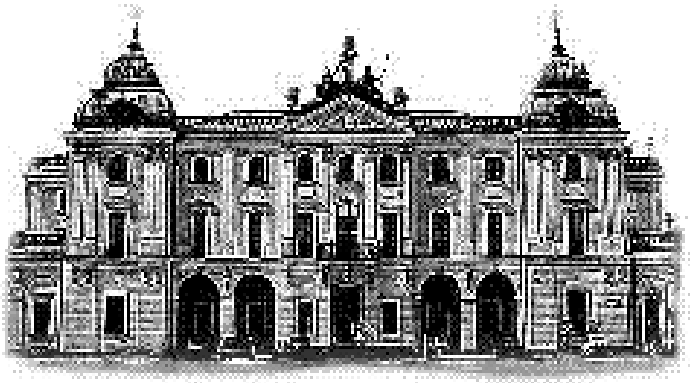
BIOPHYSICS LABORATORY EXERCISES

Edited by
Professor Anna Kostrzewska



Białystok 2005

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Preface

The course book is meant to serve as a didactic aid for students of English Division of Medical University of Białystok in their laboratory class of biophysics. It contains descriptions and guidelines on laboratory activities as well as some basic information concerning physical phenomena underpinning many biological processes, the knowledge of which is essential for a successful completion of a laboratory course in the Institute of Biophysics. The book is used to document the range of work and progress of each student. While not being a substitute for a textbook, the course book aims to facilitate the acquisition of knowledge of physical phenomena fundamental for biological processes and the understanding of the principles and modern methods of both diagnostics and therapy.

Professor Anna Kostrzewska

Safety in the laboratory

The laboratory is a safe place to experiment if you are careful. You must assume responsibility for the safety of yourself and others. Here are some safety rules to help guide you in protecting yourself and others from injury in the lab.

1. Do not perform experiments that are unauthorised. Always obtain your teacher's permission.
2. Study your laboratory assignment before you come to the lab. If you are in doubt about any procedure ask your instructor for help.
3. Always wear protective clothing.
4. Know the location of the phone, fire extinguisher, fire blanket, first aid kit and emergency exit.
5. Report any accident, injury or incorrect procedure to your teacher at once.
6. Handle dangerous materials only under supervision of your teacher. If you spill acid or another corrosive substance, wash it off immediately with water.
7. Keep combustible materials away from open flames.
8. Never eat or drink anything in the lab.
9. Place broken glass and solid substances in designated containers. Keep insoluble waste material out of the sink.
10. When your laboratory work is completed, clean your work area. Return all materials and apparatus to their proper places.

Criteria and number of points assigned for exercises,
the colloquium (test) and the exam

1) **Class (exercise)**
   - theoretical part – max. 5 points
   - practical part – max. 5 points
   - minimum number of points to obtain a pass – 6 points
2) **Section (3 classes/exercises) – max. 30 points**
   - minimum number of points to obtain a pass – 18 points
3) **Colloquium (test) – max. 20 points**
   - class material / topics – max. 15 points
   - lecture material / topics – max. 5 points
   - minimum number of points to obtain a pass – 12 points
4) **Total for classes** – 3 sections and 3 colloquia – max. 150 points
   - minimum number of points to obtain a pass – 90 points
   - Total for classes is calculated as final points by being divided by 10; e.g. 90 points = 9 final points
5) **Exam – max.30 points**
   - lecture topics (20 questions) – max. 20 points
   - class topics (10 questions) – max. 10 points
   - minimum number of points to obtain a pass – 17 points
   - Examination points count as final points
6) **Credit in biophysics – max. 45 final points**
   (15 for classes and 30 for the exam)
   - minimum to obtain a credit – 26 final points + a pass in classes, colloquia and the exam according to the above stated criteria)
7) **Grading based on the number of the total of final points obtained**
   - satisfactory (3): 26 – 29 final points
   - almost good (3.5): 30 - 33 final points
   - good (4): 34 – 37 final points
   - almost very good (4.5) 38 – 41 final points
   - very good (5): 42 – 45 final points

Student's name: .....

Student's results

Exercise		Points			
Date	Number	Theoretical part	Practical part	Total points	Signature
		Test No. 1			
		Test No. 2			
		Test No. 3			
		Correction			
		Total			

Final points

	Points	**Signature**
Classes		
Exam		
Total		

ASSIGNMENTS FOR OPTICS LAB EXERCISES

1.1 Lab Exercise

Determining solution concentrations using a refractometer and a polarimeter

1. Fermat's principle, the law of light refraction, the law of light reflection
2. Phenomenon of the total internal reflection of light
3. Operation principle of the waveguide, endoscopy
4. Operation principle of the refractometer
5. Phenomenon of light polarisation
6. Methods of light polarisation
7. Optical birefringence
8. Optically active substances
9. Optical isomerism
10. Applications of polarimetry in diagnostics
11. Determination of a simple equation by the least square method
12. Types of concentrations: weight to weight, weight to volume, molar and normal

1.2 Lab Exercise

Measurement of focal length and properties of converging lenses

1. Fermat's principle, the law of light refraction, the law of light reflection
2. Thin lenses
3. The lens equation, lens magnification and types of lenses
4. Systems of lenses
5. Focal length and converging properties of the lens and system of lenses
6. Lens aberrations
7. Optical structure of the human eye
8. The lens of the human eye
9. Accommodation of the human eye, accommodation range
10. Resolving abilities of the human eye
11. Energetics of the human vision process
12. Young's model of colour vision

1.2a Lab Exercise

Microscopy. Resolving power and measurement of numerical aperture.

1. Fermat's principle, the law of light refraction, the law of light reflection
2. Phenomenon of light diffraction, phenomenon of light interference
3. Image formation in the optical microscope
4. Image magnification in the optical microscope
5. Resolution power of the microscope
6. Aperture of the microscope
7. Types of microscopes
8. Operation principle of the electron microscope

1.3 Lab Exercise

Determining solution concentrations using an absorption spectrophotometer

1. Types and classification of electromagnetic waves
2. Visible light, ultraviolet radiation
3. Young's model of colour vision
4. Mechanism of absorption spectra formation
5. Bouger-Lambert-Beer law
6. Extinction and transmission
7. Determination of simple equation by the least square method
8. Bohr's model of the hydrogen atom
9. Mechanism of hydrogen spectral series formation
10. Structures of the free atom, the atom in a molecule, the atom in a solid
11. Influence of IR, VIS and UV radiation on human organism
12. Mechanism of emission and absorption spectra formation
13. Line, band and continuous spectra
14. Applications of spectral analysis

LITERATURE:

1. Paul Davidovits – "Physics in Biology and Medicine"
2. Roland Glaser – "Biophysics"

1.1 Lab Exercise

DETERMINING SOLUTION CONCENTRATIONS USING A REFRACTOMETER AND A POLARIMETER

THEORY

Some physical properties of dissolved substances depend linearly on their concentration. By making use of linear relations, it is possible to determine the concentration of solutions. In this exercise to determine the concentrations, we are going to examine and use the linear dependencies of both the refractive index and the rotation angle of light polarisation on the concentration of the solution.

1. *Determining of the refractive index in a solution.*

To determine the refractive index in a solution we measure the critical angle of the total internal light reflection. The instruments used for these measurements are called refractometers. When light hits the surface lying between two optical media then some light rays are partially reflected from the boundary surface while some other pass into the other medium. As a result the light beam is refracted on the boundary of two optical media. This phenomenon is described by the law of light reflection and refraction.

The sine ratio of both the incidence angle and refraction angle is constant for two optical media and is called the refractive index.

$$\frac{\sin\alpha}{\sin\beta} = n_{1,2} = \frac{c_1}{c_2} = \frac{\frac{c}{c_2}}{\frac{c}{c_1}} = \frac{n_2}{n_1} \qquad (1)$$

where:

α - angle of incidence

β - angle of refraction

$n_{1,2}$ - relative index of light refraction of the first medium with respect to the other medium

c – speed of light propagation in vacuum

c_1 - speed of light propagation in the first medium

c_2 - speed of light propagation in the other medium

n_1 – absolute index of light refraction in the first medium

n_2 – absolute index of light refraction in the other medium

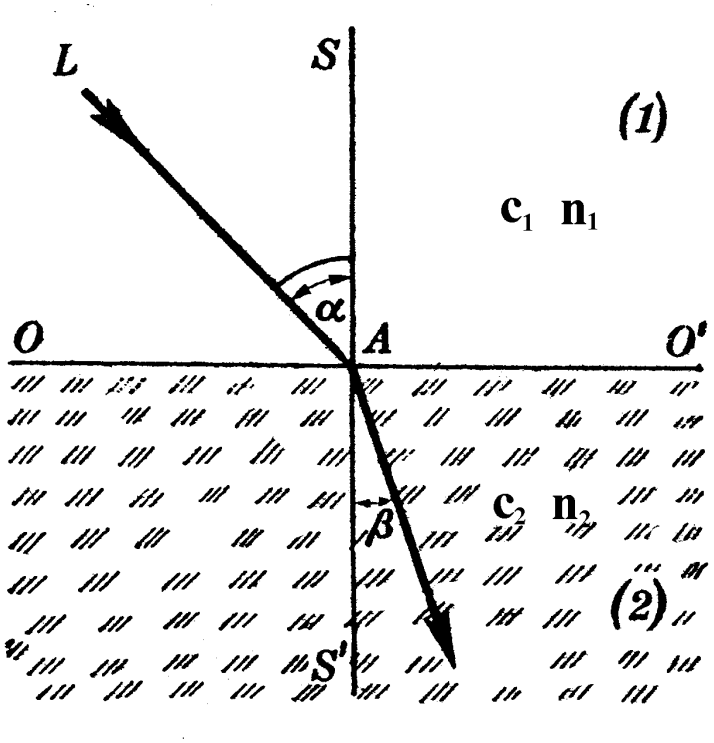


Fig. 1 Light refraction on the boundary of two optical media.

When light passes from a medium in which it propagates at a lower speed to another medium where it travels faster (eg from water into air), then, at some critical incident angle typical of the two optical media, the light ray starts traveling along the boundary of the two optical media. The refraction angle is then equal to 90° and is called the *critical angle*. If the light is incident upon the boundary separating the two optical media at an angle that is greater than the critical angle, then the light ray does not pass into the other medium but becomes reflected. This optical phenomenon is called the *total internal reflection* (Fig. 2).

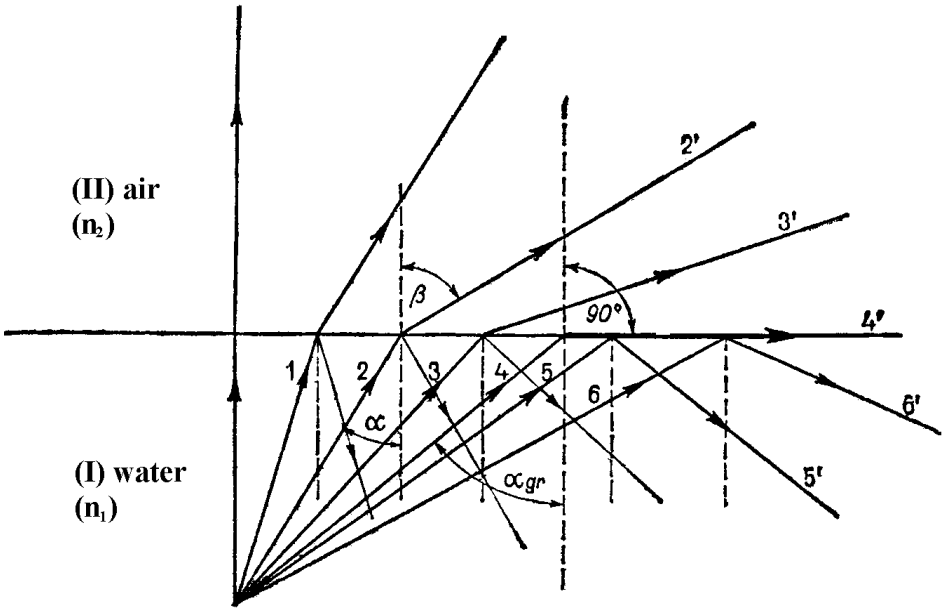


Fig. 2. Total internal reflection of light. Ray 4 hits the surface at the critical angle, rays 5 and 6 undergo the total internal reflection.

For the incident angle that is equal to the critical angle, the law of light refraction takes the following form:

$$\sin\alpha_{critical} = n_{1,2} = \frac{c1}{c2} = \frac{\frac{c}{c_2}}{\frac{c}{c_1}} = \frac{n_2}{n_1} \qquad (2)$$

since $\beta = 90^\circ$ and $\sin 90^\circ = 1$ (cf dependence (1))

If the medium penetrated by the light happens to be vacuum then $n_2 = 1$ and the law of light refraction in such a case, takes the following shape:

$$\sin\alpha_{critical} = \frac{1}{n_1} \qquad (3)$$

As can be seen from equation (3), the absolute index of light refraction in a medium is clearly determined by the critical angle. To measure the critical angle and by the same token determine the index of light refraction in a solution, we make use of instruments called refractometers. In the Abbe' refractometer used in our lab the analysed solution is composed of a layer sandwiched between two prisms made of glass. Here, the light undergoes the total internal reflection on the solution-glass boundary and an appropriate optical system makes it possible to

measure the critical angle. This device is provided with a scale to enable the readout of the light refraction index in the solution.

The value of the refraction index in a solution is directly proportional to the concentration of the solution:

$$n = b \cdot c + a$$

where:

n – index of light refraction in solution

c – concentration of solution

a, b – constant indices related to the type of solution

2. *Polarisation and rotation of polarisation plane*

The visible light consists of electromagnetic waves whose wavelengths range from 380 to 720 nm. In the vacuum the light wave propagates at the speed of 300000 km/s. In space electromagnetic waves propagate by reciprocal induction of both electric and magnetic fields, that is a variable rotational magnetic field induces a variable rotational electric field and in turn a variable rotational electric field induces a variable rotational magnetic field. These fields are described by both electric field vector E and magnetic field vector H. Vectors E and H are always perpendicular to each other and also perpendicular to the direction of the wave propagation (Fig. 3).

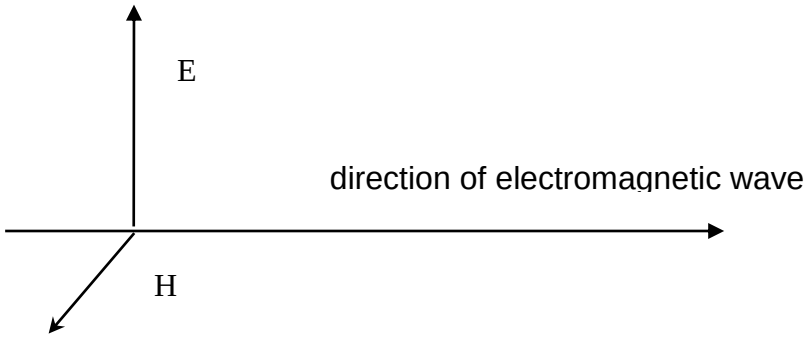
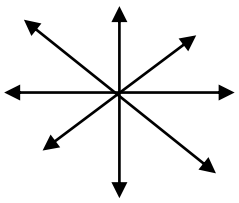


Fig. 3. Distribution of vectors E, H and direction of electromagnetic wave propagation in space.

At a propagation point in space vectors E and H will change their lengths according to the relations $E=E_o\sin(\omega t+\varphi)$ and $H=H_o\sin\omega t$. The ends of both vectors

will behave as weights hanging on a spring. This is described as “field vibrations”. In a light beam there are a great number of light waves, however, each of them may have a different spatial orientation of their electric and magnetic fields. This type of light is called unpolarised light. On the other hand, a polarised light beam will consist exclusively of waves that have the same spatial orientation of both its electric and magnetic fields. They are linearly polarised. If a light ray is directed perpendicularly to the surface of a sheet of paper then the spatial orientation for vectors E for both unpolarised and linearly polarised light rays can be shown as in Fig.4.



unpolarised light wave linearly polarised light wave

Fig. 4. Spatial system of vectors of electric field intensity for both upolarised and linearly polarised light observed along the propagation direction of light wave.

A plane in space determined by the direction of a polarised light beam and the direction of the vibrations of the electric field vector is described as the light polarization plane.

There are several methods of light polarization. The most commonly used is the one performed using anisotropic crystals i.e. such crystals whose physical properties depend directly on the direction. A specific distribution of electric fields inside such crystals will change the incident unpolarised light into linearly polarized light on leaving the crystal. Examples of such crystals include Iceland spat and tourmaline. If we create an optical system of two light polarising crystals we will notice that the intensity of light leaving the system depends on the spatial arrangement of the crystals. The first crystal called the polariser will polarise light linearly creating a polarization plane in space. Next this light hits the second crystal called the analyser. On leaving the analyser the light intensity will depend on the angle created by the polarization planes of both the polariser and analyser. This phenomenon is described by the law of Malus:

$$J = J_o \cos^2 \alpha \qquad (4)$$

where:

J_o – light intensity hitting the analyser

J - light intensity leaving the analyser

α - angle created by the polarisation planes of both the polariser and analyser

As can be seen, in the case when the polarisation planes are perpendicular to each other the light intensity on leaving the optical system is equal to zero.

The human eye is not capable of distinguishing between the polarised and unpolarised light.

There is a group of substances such as sugar solution, tartaric acid, turpentine and a number of other organic compounds which show the property to rotate the polarisation plane of polarised light. The molecules of these substances must have asymmetric structure. The ability of these compounds to rotate the light polarisation plane is called *optical activity*.

Two different forms of the same substance whose molecules are mirror-like reflections of each other are described by the phenomenon of *optical isomerism*. Form L of the substance will rotate the polarisation plane anticlockwise whereas form D will produce a clockwise rotation.

The polarisation plane of light after passing a solution of an optically active substance will be rotated in space by an angle whose value is directly proportional to the concentration of the solution:

$$\alpha = k \cdot c \cdot l$$

where:

α - rotation angle of polarized plane

c – concentration of solution

l – thickness of the layer of optically active substance

k – proportionality coefficient dependent on the type of solution

The instrument used to determine the rotation angle of polarisation is called the polarimeter.

3. ***Method of determining solution concentration***

In order to determine the concentration of a solution using the measurement of the light refractive index or the rotation angle of polarised plane we should make use of the fact that both these parameters of the solution depend linearly on its concentration. This dependence is different for each solution and as a result must be determined experimentally. First, using measuring instruments such as the refractometer or the polarimeter, it is necessary to find out the values of both the light refractive index and the rotation angle of polarisation plane for the solutions whose concentrations are known. Next, using the above data we find the graphical and numerical relation between the physical parameters of the solution and its concentration.

Both of these relations are linear i.e. they can be described by the following equation:

$$y = b \cdot x + a$$

where:

x - the concentration of solution,

y – the value of the light refraction index or the rotation angle of light polarisation plane depending on the value investigated.

Using the least square method we can determine coefficients b and a of the simple regression in equation $y = b \cdot x + a$

$$b = \frac{N\sum xy - \sum x \sum y}{N\sum x^2 - (\sum x)^2} \qquad a = \frac{\sum y - b\sum x}{N}$$

In this way we find the experimental dependence of the investigated parameters on the solution concentration. To determine the simple linear regression equation and the correlation coefficient R^2 we can use a computer application such as “Microsoft Excel”. An illustrative result of such a procedure is shown in Fig. 5.

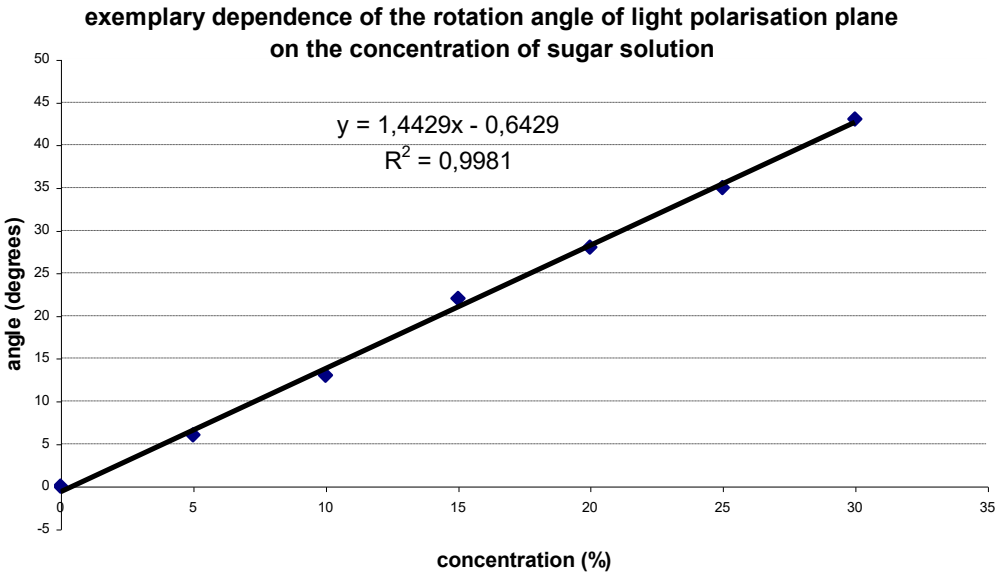


Fig. 5. Example of linear regression.

The dependence is said to be linear and ready to be used to determine concentration only if the correlation coefficient $R^2 > 0.95$.

If we wish to determine an unknown solution concentration of the same substance it is required to measure the value of the light refractive index or the rotation angle of polarization plane using the same measuring instruments. Next, it is necessary to refer the obtained values to the linear regression graph or making use of the known parameters b and a in equation $y = b \cdot x + a$ determine value x knowing measured value y.

For example, if the dependence of the rotation angle of the polarisation plane on the solution concentration is $y = 1.4429 \cdot x - 0.6429$ as shown in Fig.5 and the measured value of the angle for the solution of the unknown concentration is 19 degrees, then the concentration of the solution can be determined by substituting value 19 for “y” and calculating value “x” from the equation. The result obtained is 13.61% (please check the calculations yourselves).

PRACTICAL PART

Discussion questions:

1. Which instrument, the refractometer or the polarimeter, will you use to determine the concentration of table salt (NaCl), and which one to determine the concentration of sucrose? Give reason for your answer.

2. What is “endoscopy” and what physical phenomenon is used in the operation of the endoscope?

EXPERIMENTAL PART

Objective: Determination the concentration of solutions

Materials: a refractometer, a polarimeter

a) Preparation of solutions

- Prepare sugar solutions in water of the following (weight-weight) concentrations 5%,10%,15%,20%,25%,30%, 50 grams of each.
- The students are divided into two subgroups, each of which prepares a 50 gram aqueous sugar solution keeping the concentration value in secret – x_0. Write down the value x_0 of your subgroup here,
- x_0 =

b) Refractometer – measurement of the light refractive index

Spread a thin layer of the solution on the matt surface of the refractometer glass. Next, turn the prism knob to position the refractometer prisms in such a way that the division line between the bright field and the dark one cuts across the intersection point of the crosshairs (Fig.6)

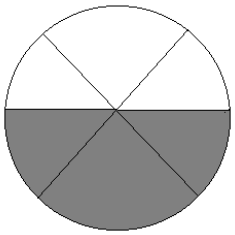


Fig. 6. Image seen in the refractometer eyepiece.

Data and observations

From the scale we read out the value of the light refraction index in the solution for all the prepared solutions and distilled water and enter the results into the table:

Solution concentration (%)	Value of refractive index „n”
0 (distilled water)	
5	
10	
15	
20	
25	
30	

Using a computer program we calculate the linear dependence (simple linear equation and correlation coefficient) for the obtained values of the light refraction index dependent on solution concentration.

Write down here:

- obtained equation: y =

- value of the correlation coefficient R^2 =

Next we carry out the measurement of the light refractive index of the solution prepared by the second subgroup.

Write down here:

- measured value of the light refractive index n =

Making use of the obtained dependence between the value of the light refraction index and the solution concentration we calculate the concentration of solution – x – prepared by the second subgroup.

Write down the calculations here:

Write down here:

- calculated value of concentration x =

We determine the percentage error of the measurement

$$b = \frac{|x - x_0|}{x_0} \cdot 100\% =$$

where x_0 – solution concentration prepared by the second subgroup

c) Polarimeter – measurement of the rotation angle of light polarisation plane

We fill in the polarimeter cell with the solution and place it in the tube. Prior to this, we check the zero point of the polarimeter i.e. on the polarimeter scale we find such a point that corresponds to the image in which all the elements in our field of vision have the same colour (Fig. 7)

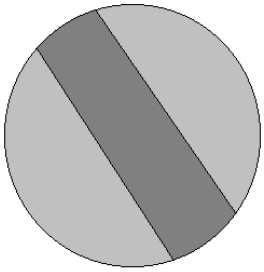


Fig. 7. Image seen in the polarimeter eyepiece with the sugar solution placed inside the tube.

Now we start searching for a new position on the scale to correspond to such an image that would include all the elements of the same colour in our field of vision. We read out the difference between the final and initial position and what we get is exactly the rotation angle of the polarisation plane.

Data and observations

From the scale we read out the value of the rotation angle of light polarisation plane in the solution for all the prepared solutions and enter the results into the table:

Solution concentration (%)	Value of rotation angle „α”
0 (distilled water)	0
5	
10	
15	
20	
25	
30	

Using a computer program we calculate the linear dependence (simple linear equation and correlation coefficient) for the obtained values of the rotation angle of the light polarisation plane dependent on solution concentration.
Write down here:

- obtained equation: y =
- value of the correlation coefficient R^2 =

Now we perform the measurement of the rotation angle of the light polarisation plane in the solution prepared by the second subgroup.
Write down here:

- measured value of the rotation angle of the light polarisation plane α =

Making use of the obtained dependence between the value of the rotation angle of the light polarisation plane and the solution concentration we calculate the concentration of solution – x – prepared by the second subgroup.
Write down the calculations here:

Write down here:

- calculated value of concentration x =

We determine the percentage error of the measurement

$$b = \frac{|x - x_0|}{x_0} \cdot 100\% =$$

where x_0 – solution concentration prepared by the second subgroup

The date	Student's name and surname	Lab assistant signature

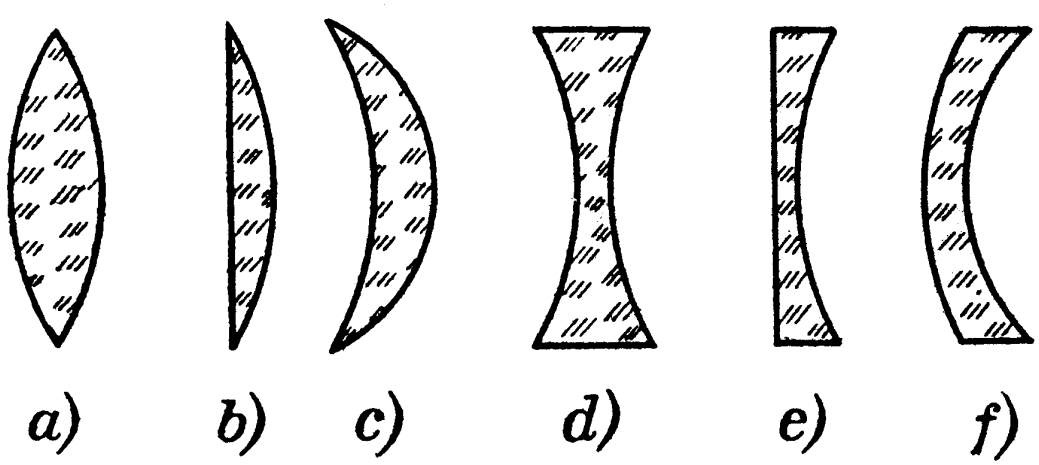
NOTES

1.2 Lab Exercise

MEASUREMENT OF FOCAL LENGTH AND PROPERTIES OF CONVERGING LENSES

THEORY

The lens consists of two spherical surfaces which refract light. Taking their shape into consideration lenses can be divided into the following types:

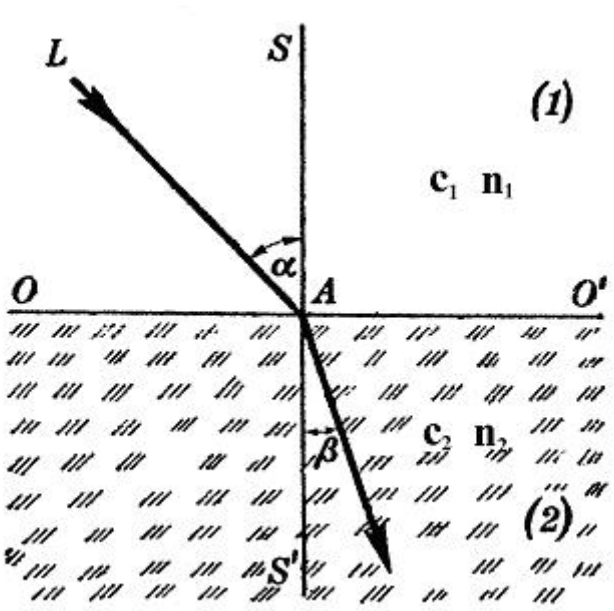


a) double convex, b) plano-convex, c) concave convex, d) ,double concave
e) plano-concave, f) convex concave

Thin lenses – if the linear dimensions of the lens is considerably smaller than the radii of the lens curvature, we make use of the thin lens approximation i.e. we assume a zero thickness of the lens and analyse the processes that occur exclusively on the external surfaces of the lens and the centre of the lens is then the point at which the principal axis crosses the lens.

Light is refracted on the external surfaces of the lens according to the law of light refraction:

$$\frac{\sin\alpha}{\sin\beta} = \frac{c_1}{c_2} = \frac{n_2}{n_1}$$



where: α - angle of incidence, β - angle of refraction, c_1,c_2 – speed of light in medium 1 and 2, n_1,n_2 – absolute indices of light refraction in medium 1 and 2

Light refraction in thin lens

On account of their optical properties lenses are categorised into *converging* and *diverging* ones.

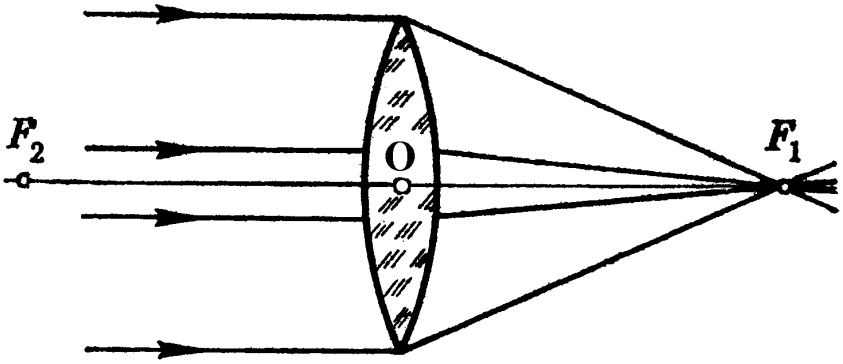


Fig. 1. Schematic diagram of light rays passing through a converging lens.
where: F_1, F_2 – focal points of the converging lens, O – centre of the lens, section $OF_1 = OF_2$ – f – focal length of the converging lens, normal F_2OF_1 – principal axis of the lens.

The focal point - F – of the converging lens is the point lying on the principal lens axis at which incident rays travelling parallel to the axis intersect on leaving the lens.

Focal length – f – of the converging lens is the distance connecting the focal point F of the lens with its geometric centre – O.

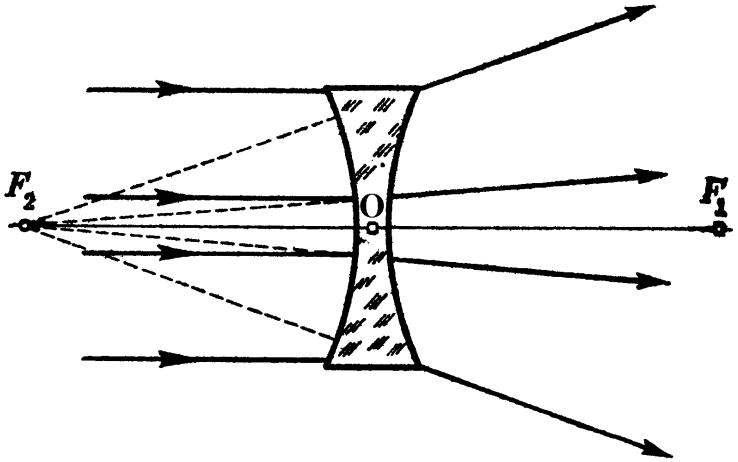


Fig. 2. Schematic diagram of light rays passing through a diverging lens.
where: F_1, F_2 – focal points of the diverging lens, O – centre of the lens, section $OF_1 = OF_2$ – f – focal length of the diverging lens, normal F_2OF_1 – principal axis of the lens.

Virtual focal point - F – of the diverging lens is the point lying on the principal lens axis at which extended rays intersect after refraction, when incident rays travelling parallel to the axis intersect.

Focal length – f – of the diverging lens is the distance connecting the virtual focal point F of the lens with its geometric centre – O.

The properties of the lens (position of focal point – F and focal length – f) depend on the radii of the lens curvature (R_1,R_2), refractive index of the material used to make the lens (n) and refractive index of the medium in which the lens is placed (n_0).

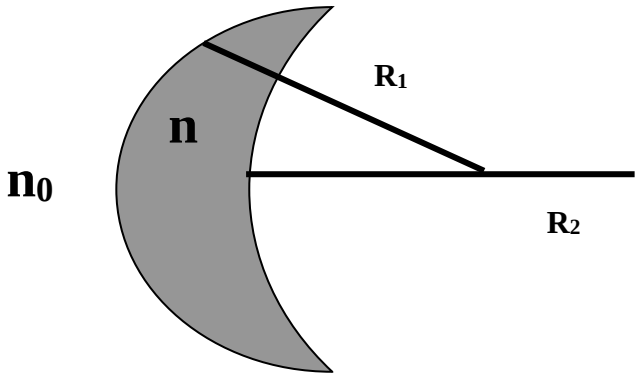


Fig. 3. Parameters of the thin lens.

Focal length – f – is determined by the following relation:

$$\frac{1}{f}=\left(\frac{n}{n_0}-1\right)\cdot\left(\frac{1}{R_1}+\frac{1}{R_2}\right) \qquad (1)$$

The lens curvature is said to be positive if the lens is convex, and negative if the lens is concave. The radius of the curvature plane is equal to infinity.

The lens is said to be converging only if its focal length is positive, and for the diverging lens if its focal length is negative.

The converging property of the lens defined in dioptres is called the reciprocal of the focal length:

$$D=\frac{1}{f} \qquad [1/m] \qquad (2)$$

The converging property of the thin lens system remaining in the close possible contact is equal to the sum of all the properties of individual lenses in the system:

$$D_s=D_1+D_2+D_3+\ldots\ldots \quad (3)$$

Image formation in the thin lens

To find an image of a point in any given optical system (including the lens), it is necessary to determine the path, of at least, two rays in the system. If the image is formed at the intersection point of the rays then a real image is formed and it can be seen on the screen. On the other hand, if the image is formed at the intersection point of the extended rays then a virtual image is formed and it cannot be seen on the screen.

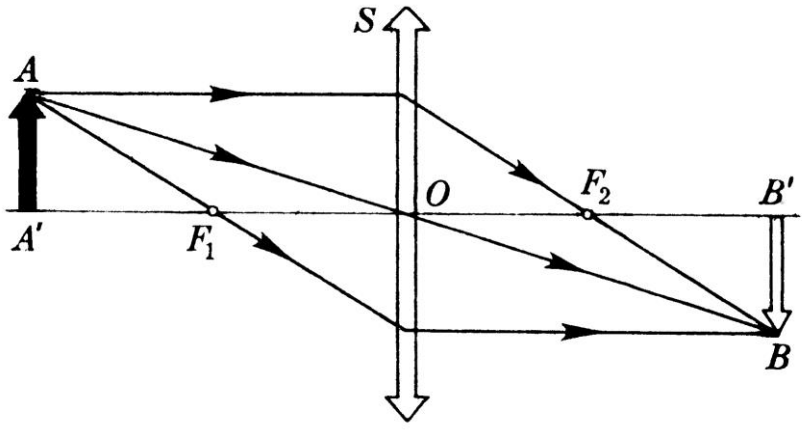


Fig. 4. Schematic diagram of real image formation for object A in a converging lens with the focal length f .

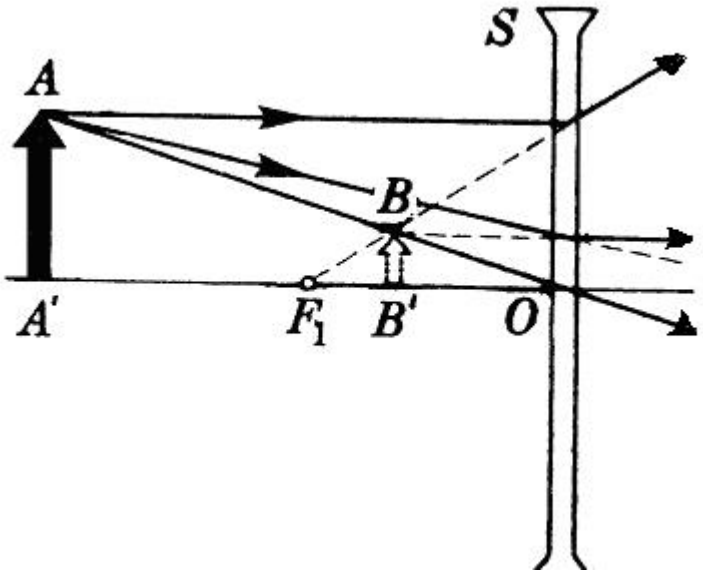


Fig. 5. Schematic diagram of virtual image formation for object A in a diverging lens.

The image formed in a diverging lens is always a virtual image.

The distance of the image from the lens - Y and the distance of the object from the lens – X satisfy the equation known as the lens equation:

$$\frac{1}{X}+\frac{1}{Y}=\frac{1}{f} \qquad (4)$$

After transformation the equation gives the dependence of the focal length f on the lens image (Y) and the distance of the object on the lens (X):

$$f=\frac{Y\cdot X}{X+Y} \qquad (5)$$

By measuring distances X and Y, in the case of real image formation on the screen, it is possible, using equation 5, to determine the focal length of the lens. However, this method cannot be directly applied to measure the focal length of a diverging lens since it does not produce real images. This difficulty can be overcome by making use of a system composed of two lenses: a converging and a diverging ones. From equation 5 we calculate the converging property of system D_s. If converging property D_1 of the converging lens in the system is known, then the needed converging property D_2 of the diverging lens can be calculated from equation 3.

Optical structure of the human eye

Figure 6 shows the cross section drawing of the human eye

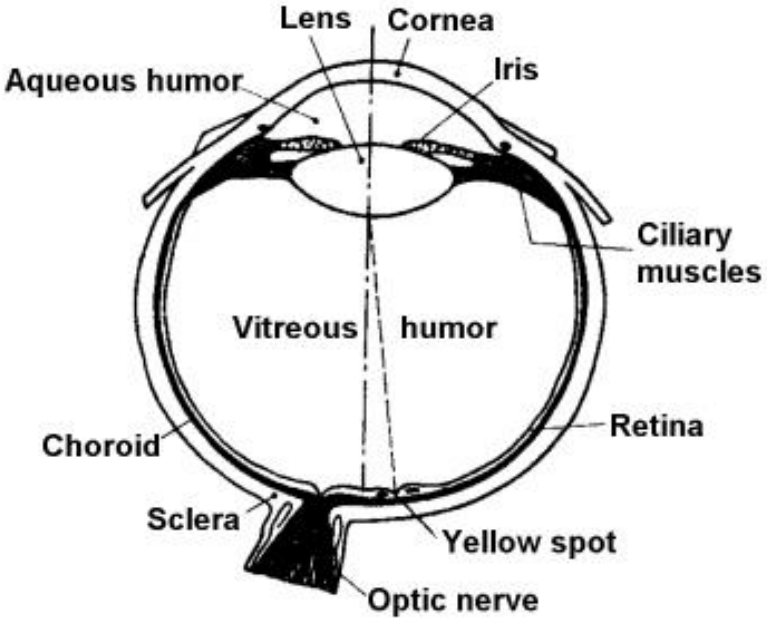


Fig. 6. Cross section drawing of the human eye

The human eyeball has a spherical shape usually 24.4 mm in diameter. However, it is possible to find healthy eyes of distinct vision whose diameter is as low as 21mm or as high as 27.5 mm.

The optical system of the human eye consists of the following parts: cornea, anterior chamber filled with aqueous humour, iris with the pupil, lens, posterior chamber filled with vitreous humour and light - sensitive retina.

It is the external surface of the cornea where light is refracted the most. This is due to the considerable difference of refractive indices when light passes from air into the interior part of the eye. The cornea is usually 11.5 mm in diameter, its thickness ranges from 0.5 mm in the central part to 0.7 mm at the ends. The radius of the external surface curvature is 7.8 mm on average. The converging power of the cornea is 43 dioptres.

Between the cornea and the lens there is the anterior chamber measuring 3.6 mm in depth. It is filled with a water-like liquid. The space between the lens and the retina is filled with jelly-like substance, the so called vitreous humour. Both

media surrounding the lens have the same refractive index whose value is similar to the index of light refraction in water.

The human lens' structure is complex and multi-layered. The kernel of the lens has the highest refractive index. The lens makes it possible for us to see both distant and close objects equally well as its shape and converging properties change adaptively. This ability of the lens is called accommodation. For distant objects' viewing an average lens thickness is 3.6 mm and its converging power is 19 dioptres. For close-up viewing the converging power of the lens increases up to 33 dioptres maximally and becomes a little thicker.

The iris with its pupil's hole is an aperture diaphragm. The iris diameter changes from 8 mm in the darkness up to 1.5 mm at good lighting conditions.

PRACTICAL PART

Discussion questions:

1.What does the resolution of the human eye depend on and how does it work?

2. Enumerate and describe common vision defects of the human eye.

EXPERIMENTAL PART

Objective: Determination the focal length of the lenses

Materials: an optical bench, lenses

To determine the focal length of the lenses under investigation we make use of a system which consists of a light source, a lens and screen placed on an optical bench (Fig.7). All the lenses used in our investigation are considered to be thin ones.

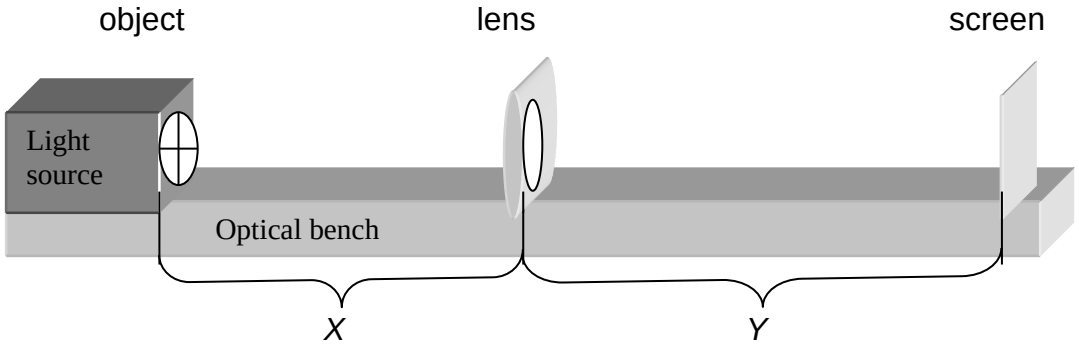


Fig. 7. Drawing of the system used to determine focal lengths.

Having established the distance of the object from the lens, we adjust the distance of the screen from the lens to obtain a clear image of the object. Next we measure the quantities of X an Y on the optical bench and using equation 5 we determine the focal length of the lens. This procedure is repeated at least 5 times changing the distance between the object and the lens by a few centimetres each time. Now keeping the lens under investigation in its position we insert a diverging lens into the frame so that we create a lens system having a converging property. The focal length of the system can be found in the way described for the converging lens above. All the results obtained should be written down in the table prepared according to the arrangement given below:

Data and observations

	X	Y	f	f-average	D-average
Converging lens					
I System of lenses					
II System of lenses					

The date	Student's name and surname	Lab assistant signature

NOTES

1.2a Lab Exercise

MICROSCOPY. RESOLVING POWER AND MEASUREMENT OF NUMERICAL APERTURE.

THEORY

Optical microscopy has widespread applications in various fields of science. The microscope is an instrument that is used to increase the angular vision of objects positioned within a distance of distinct vision. As specimens viewed under the microscope are mostly non-luminous, they must be intensively illuminated either from below (transmission microscopes) or from above (reflection microscopes) if specimens are non-transparent.

The microscope consists of two converging lenses – the objective lens that has a very short focal length and the eyepiece or ocular lens that has a larger focal length (acting as a magnifying glass). Both the objective and eyepiece lenses are situated on the same axis at some distance L from each other. The objective forms a real, magnified and inverted image of the specimen viewed. The specimen is placed in front of the objective at a distance x which is a little greater than the focal length f for of the lens ($x \approx f$). The image formed in this way, when viewed through the eyepiece, is once more magnified. It is also virtual and upright. This image is formed within a distance of distinct vision b = 25 cm.

The magnification of the microscope (p) can be described by the following approximated relation:

$$p = p_{ob} \cdot p_{oc} = \frac{L \cdot b}{f_{ob} \cdot f_{oc}} \qquad (1)$$

The pathway of light rays in a microscope is shown in Fig. 1.

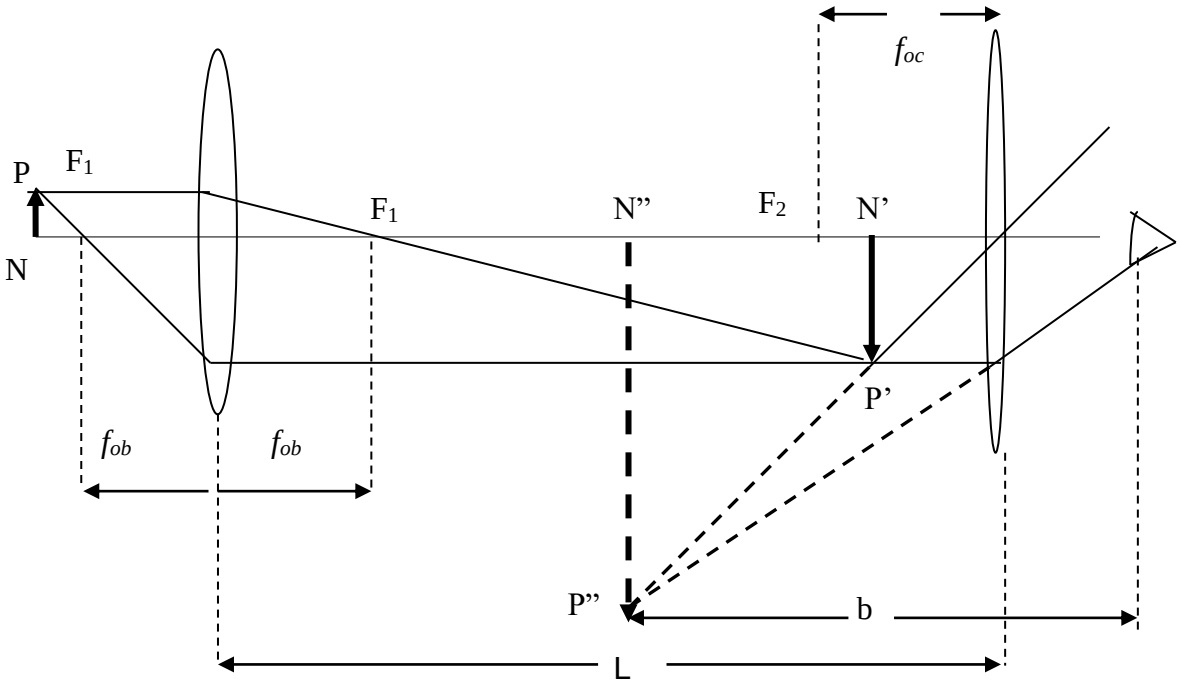


Fig. 1. Course of light in a microscope.

Images formed in a microscope should be not only magnified but they should also expose minute details of viewed specimens. The more detailed image a microscope can produce, the higher resolution it has. This resolving power is conditioned by such wave phenomena as diffraction or interference. It is well known (from practice) that there is a certain definite distance d between two points that can still be distinguished as two separate entities. This smallest distance d (or its reciprocal) that is distinguished (or resolved) by the microscope is a measure of its resolving power. It was Abbe who first observed that very small objects form a kind of diffraction grating that refracts light rays coming from the condenser. The necessary condition of real image formation of an object in the objective is the focusing of, at least, two refracted rays at a single point. The rays leaving the objective lens must be in phase.

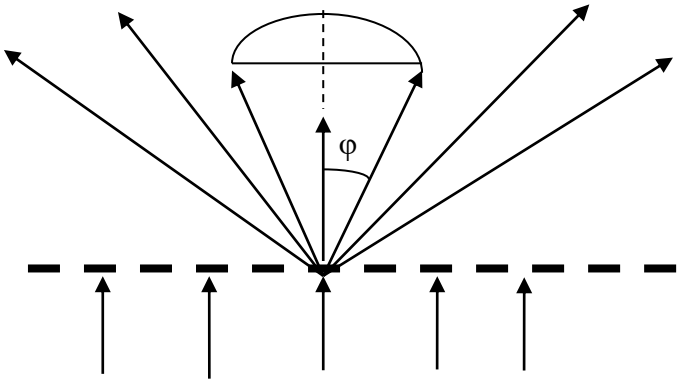


Fig. 2. Diffraction of light rays in the openings of a specimen placed in front of the objective.

Experimental data based on the analysis of the diffraction phenomenon show that the least distance “d” between the points that can be distinguished in the image formed by the microscope fulfils the following condition:

$$d = \frac{\lambda}{n \cdot \sin\varphi} \qquad (2)$$

where: λ – wavelength of the radiation used during the viewing process ,

n – refractive index of the medium from which the rays reach the objective,

φ - angle formed between the boundary radius and the optical axis. It is assumed that the apex of the angle lies within the microscope’s image sharpness field. (Fig. 3).

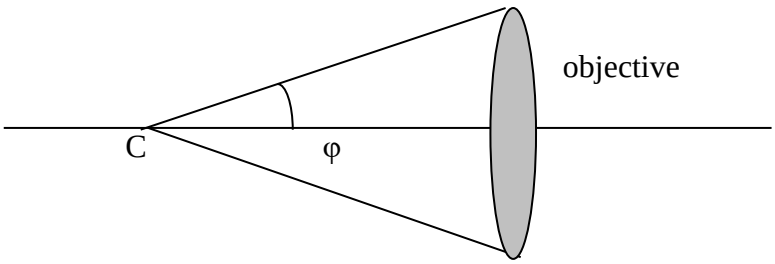


Fig. 3. Aperture angle

The reciprocal of d, i.e.

$$Z = \frac{1}{d} = \frac{n \cdot \sin\varphi}{\lambda} \qquad (3)$$

is a measure of the resolving power of the microscope. The microscope's limited power of resolving is caused by the phenomenon of light diffraction when passing through tiny slits or fibres or other parts of the viewed specimens. For simplicity sake let us assume that the specimen consists of a diffraction grating built of a series of transparent slits spaced at a distance d from each other.

The theory of diffraction grating states that in order to obtain a screen image similar to the viewed object, the objective lens must focus at least one deflected light beam. Condition (2) implies that the deflection angle of a ray when passing through a slit cannot be grater than angle φ. It is only then that we can obtain the first interference image of the slot on the screen.

If the distance between the slits of the diffraction grating is smaller than d, then the deflection angle θ is greater than angle φ, and, as a result, only undeflected rays will reach the objective: there will be no image of the diffraction grating on the screen, only a bright spot.

It follows from condition (2) that the structure of the specimen (e.g. the distance between the slits of the diffraction grating) is the more detailed the smaller the wavelength and the larger angle φ are.

Expression:

$$\mathbf{a = n \cdot \sin \varphi} \qquad (4)$$

from condition (2) is called the *numerical aperture*. The higher the numerical aperture, the better the resolving power of the microscope (formula 3).

To increase the resolving power of the microscope we use an immersion system (i.e. a transparent substance of a refractive index n>1 is placed between the specimen and the objective). In some biological investigations we use cedar oil of n = 1.55 or naphthalene bromide of n = 1.66 as such immersion systems. To improve the resolving power of the microscope we can also shorten the length of light wave λ used to illuminate the specimen.

EXPERIMENTAL PART

Objective: The purpose of the exercise is to get acquainted with the way of determining the numerical aperture of the microscope. According to the definition the numerical aperture is the sine of the angle formed by the critical incident ray still reaching the objective with the principal axis of the objective lens. (Fig.3).

Materials: microscope, optical bench with scale, a small bench with moveable spotlight source.

To perform measurements of the numerical aperture of an objective we make use of Fig. 4.

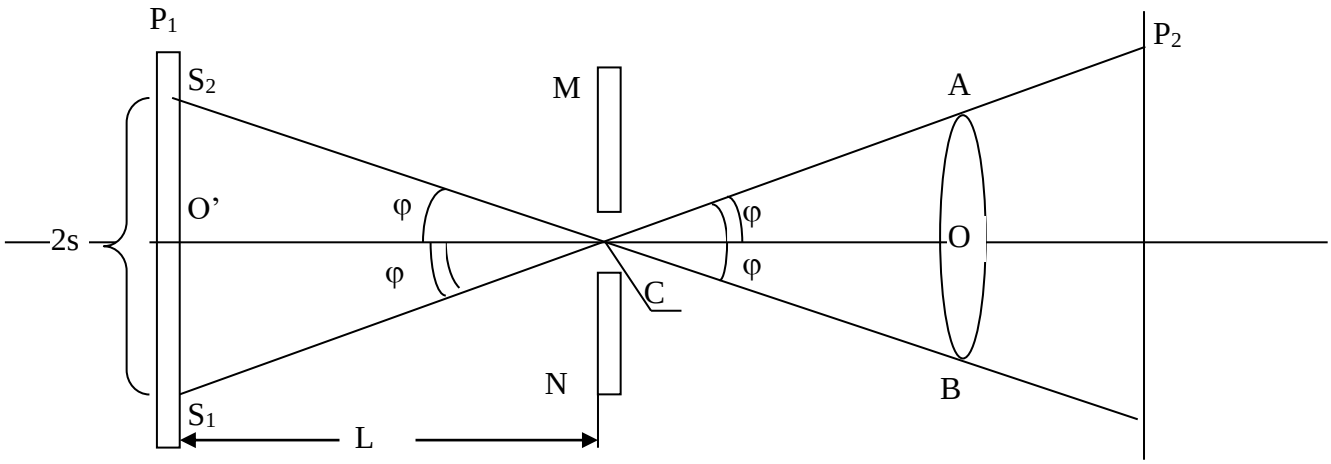


Fig. 4. Diagram useful for numerical aperture measurements.

Let lens AB be the objective lens. MN denotes the plane of sharp image (the surface of the specimen constituting a kind of diffraction grating) in which the point C is situated.

If light is passed through an object placed in the field of sharp image then the rays, having passed through the lens, will create a sharp image on screen P_2. Let's assume that point C of the field of sharp image is situated on the principal axis of the microscope.

In Fig.4 angle φ =< ACO = < BCO forms the aperture. Let's place the small optical bench P_1 perpendicularly to the optical axis of the microscope, along which the spotlight source will be moved. By extending border rays AC and BC towards the bench P_1 we obtain points S_1 and S_2, at which the spotlight source will still be observed. As a result we obtain a ray cone formed by border rays S_1C and S_2C entering the objective of the microscope.

Due to its very small dimensions we can say that the field of the sharp image is point-like. As a result we will be able to notice a ray passing through point C coming from the cone of rays sent by the light source S_1. This ray is the border ray S_1A. Looking into the opening of the microscope's tube after removing the eyepiece we will see a brightening just at the boundary of the objective.

After moving the light source to point S_2 lying at the intersection of the straight line BC (being also a border ray) and the optical bench P_1 we obtain a brightening near the boundary of objective B. The location of S_2 is, of course, symmetrical to the position of S_1.

It follows from Fig.4 that

$$\sin\varphi = \frac{O'S_1}{S_1C} = \frac{O'S_2}{S_2C}$$

Since the value of n air is equal practically to 1, then, according to (2) the numerical aperture of the objective is $a = \sin\varphi$.

Experimental procedures

1. To obtain a sharp image place a specimen on the stage and move the tube. Once you get the sharp image of the specimen, remove it and take out the eyepiece.
2. Place the light source in front of the microscope. It must be on the optical axis of the microscope.
3. Looking into the tube move the spotlight source along the bench P_1 to such a position from which the last visible ray coming from the source reached point A (Fig. 4) – this corresponds to position S_1. Next determine the extreme position for the light source on the opposite site of the bench P_1 so that the border ray leaving point S_2 reached point B.

In the table below write down positions S_1 and S_2 with the accuracy of 1 mm. The measured distance between these two positions denote as 2S, and the distance between the field of sharp image of the microscope specimen and line S_1S_2 denote as L (Fig. 4).

Since the value of n for air is 1, hence the numerical aperture of the objective is

$a = \sin\varphi$.

$$a = \sin\varphi = \frac{s}{\sqrt{s^2 + L^2}}$$

Write down the results of the measurements and calculations into the table according to the arrangement given below.

Data and observations:

No objective	measurement	L	S_1	S_2	s	a	Average value a
	1						
	2						
	3						
	1						
	2						
	3						
	1						
	2						
	3						

The date	Student's name and surname	Lab assistant signature

NOTES

1.3 Lab Exercise

DETERMINING SOLUTION CONCENTRATIONS USING AN ABSORPTION SPECTROPHOTOMETER

THEORY

Investigation of different types of spectra is generally known as spectroscopy or spectrophotometry. Spectrum analysis is commonly used as it makes possible to identify elements or chemical compounds in the examined sample by making use of characteristic lines (bands) in the emission (absorption) spectrum. It should be remembered that the spectrum is defined as distribution of the electromagnetic radiation according to its energy, frequency or wavelength.

In a more general sense spectroscopy involves a complex system of research methods in both atomic and nuclear physics dealing with:

- structural analysis and properties of molecules, atoms and atomic nuclei;
- investigation of elementary constituents of atoms and molecules based on the distribution of electromagnetic or corpuscular radiation emitted, absorbed or dispersed by them.

The basis of spectrum analysis is the fact that atoms of each element have a system of permissible energy states or levels, which makes it possible to distinguish one element from another and also determine the energy state of an atom or molecule. It is advisable to refer to the Bohr Model of the hydrogen atom here. Electrons in both atoms and molecules possess energy connected with their interaction with atomic nuclei. This energy is not unrestricted but it can assume only some definite values (one says that electron energy is “quantised”). It is impossible for an electron to give away less energy than the energy difference between the permissible energy levels for the electron. The spectrum created in this way, the so called line spectrum or band spectrum, is the information carrier of the chemical composition of the sample under investigation.

The electromagnetic spectrum is presented in Fig. 1.

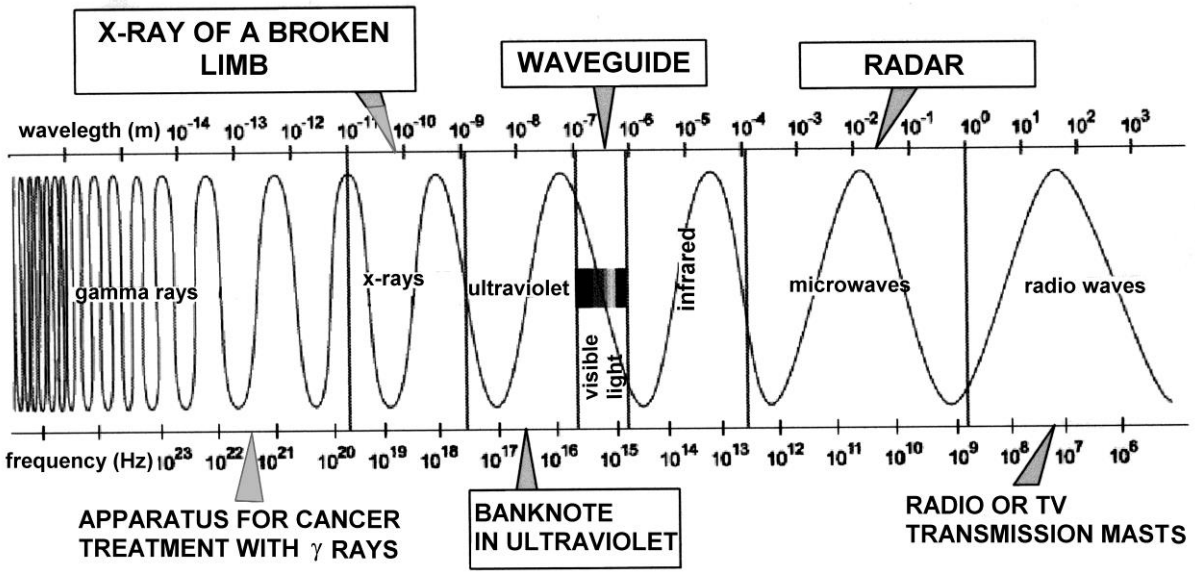


Fig. 1. The electromagnetic spectrum (EM spectrum)

The EM spectrum gives information not only about the source of given radiation (i.e. emission spectrum), but very often about the medium it went throught (i.e. absorption spectrum).

In the case of electromagnetic waves (from microwaves to x and γ rays) emitted by single atoms, molecules or nuclei, the emission spectrum consists of lines characterized be precisely defined energy states.

Absorption spectrum is formed when continuous spectrum passes through matter. In the case of electromagnetic waves, atoms or molecules of a medium will absorb the radiation whose energy corresponds to their energy levels and will then immediately (excitation time - 10^{-12} – 10^{-8} s) and spontaneously emit EM radiation – the emission has an isotropic character. We observe in the absorption spectrum considerable discontinuity of energy, typical of the given substance in direction of the incident electromagnetic wave. This makes it possible to investigate the chemical composition of the absorbent.

There are three types of emission spectra: line spectra (for gases and vapours in atomic states), band spectra (for gases and vapours in molecular states), and continuous spectra (for fluids and solids).

The fundamental instrument in spectroscopy is a spectroscope (or spectrometer, or spectrograph) appropriate to a given type of radiation.

Spectroscopy can be classified according to the type and the range of the radiation investigated, type of sample investigated and the methods of obtaining

the spectra. In spectroscopy the electromagnetic radiation of 10^{-12} to 10^{3} m in wavelength is investigated. According to the wavelength ranges, spectroscopy is divided into :

- gamma ray spectroscopy (investigation of intranuclear processes),
- x-ray spectroscopy (investigation of internal electron shells of atoms),
- UV and visible light spectroscopy (determination of structure and distribution of atomic and molecular energy levels),
- IR spectroscopy (investigation of absorption spectra and using the investigations to analyse the structure of molecules and their oscillation and rotation).

According to the types of the investigated objects whose spectra are analysed, spectroscopy can be categorized into:

15. atomic spectroscopy including structure investigations of atom and ion energy levels. These investigations are carried out on the basis of the analysis of emission and absorption spectra which are created during transition between energy levels of particular atoms. The atomic spectrum has a line characteristic.
16. Molecular spectroscopy that involves the investigation of molecules, their structure and physico-chemical properties. Unlike the atomic spectrum, the molecular spectrum has a band characteristics. This is due to the fact that molecules, apart from motions in their atoms, are characterized by oscillatory motions in their atomic nuclei and also rotational motions of molecules as a whole. The transition process of molecule between the energy levels is accompanied by emission or absorption of a photon whose energy is as follows:

$$\Delta E = \Delta E_{electr} + \Delta E_{oscyl} + \Delta E_{rot}$$

where: ΔE_{electr} – change of molecule's electron energy, ΔE_{oscyl} – change of energy associated with molecule's oscillatory motion, ΔE_{rot} – change of energy associated with molecule's rotational motion.

Due to the fact that $\Delta E_{electr} \gg \Delta E_{oscyl} \gg \Delta E_{rot}$ of each spectrum line which corresponds to the change in electron shell is accompanied by the change of a series of lines connected with oscillatory and rotational levels. In the molecular spectrum we can distinguish:

- Rotational line spectrum ($\Delta E_{elektr} = 0$, $\Delta E_{oscyl} = 0$, $\Delta E_{rot} \neq 0$), spectrum lines lying in infrared and microwave region,
- Oscillatory-rotational band spectrum ($\Delta E_{elektr} = 0$, $\Delta E_{oscyl} \neq 0$, $\Delta E_{rot} \neq 0$) lying in near infrared region,
- Electronic oscillatory-rotational band spectrum ($\Delta E_{elektr} \neq 0$, $\Delta E_{oscyl} \neq 0$, $\Delta E_{rot} \neq 0$) lying in the visible and ultraviolet regions.

Light is an electromagnetic wave. The visible light includes wavelengths ranging from about 380 nm (which corresponds to violet) to about 720 nm (red colour. Between these two extreme colours all the remaining colours of the visible light can be found. Although the range of the visible light is very narrow, it is extremely significant as the light waves of this very range cause light (and colour) sensations in human beings. The human eye can distinguish seven different spectral colours (Table 1).

Table 1. Spectral colours of white light.

colour	wavelength (nm)
violet	380-435
blue	435-485
indigo	485-495
green	495-560
yellow	560-585
orange	585-610
red	610-720

The sensitivity of the human eye varies depending on the particular length of the light wave. Fig. 2 shows a diagrammatic representation of the relative sensitivity of the human eye.

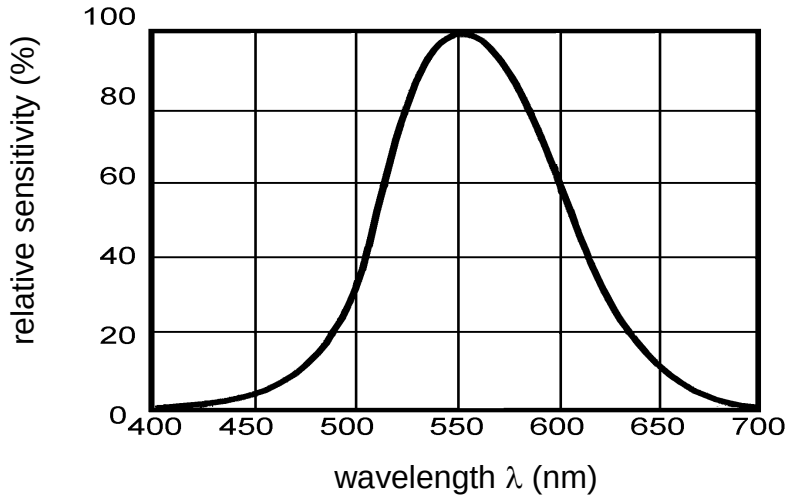


Fig . 2. General characteristics of human eye sensitivity.

As can be seen the central region of the human vision is located at about 555 nm ($5{,}55 \cdot 10^{-7}$ m.). At this wavelength the colour sensation is of yellow and green. It should be noted that the light we deal with on everyday basis is the so called white light (emitted by the sun, ordinary bulbs, etc.). It has no specific wavelength associated with it. The white colour effect is the result of an overall interaction of all wavelengths within the visible spectrum. The white light when passed through a prism is split into specific monochromatic colours. When mixed the individual colours give white light again. However, if we remove only one of the constituent colours from the spectrum, then the remaining light will manifest a complementary colour, e.g. green is the complementary to red and vice versa, the red colour is complementary to the green colour.

If white light hits a body its waves start interacting with the atoms and molecules of this body. Thus, if white light hits an non-transparent body, then the wavelengths that are absorbed are only those whose energy $h\nu = hc/\lambda$ corresponds exactly to the difference $\Delta E = E_2 - E_1$ between the permissible electron energy levels. The remaining wavelengths are not simply absorbed by the body at all, they become reflected and as a result they reach our eyes creating colour effect. If, for example, a specific body absorbs most waves and reflects mainly the ones that correspond to red colour then the body perceived by us will be red in colour. The colour of transparent bodies will depend on which wavelengths are let through by the body since the absorbed or reflected rays do not reach our eyes.

If light of intensity I_0 hits a coloured and transparent solution, then some amount of this light is reflected from its surface (I_{ref}) some is dispersed (I_{dis}), some becomes absorbed by the solution (I_{abs}) and some passes through the solution (I). The whole phenomenon can be expressed by the following equation:

$$\mathbf{I_0 = I_{ref} + I_{dis} + I_{abs} + I} \qquad (1)$$

If we use the same clean cuvettes during our measurements and assume the same dispersion throughout the whole solution (assuming that I_{ref} and I_{dis} are of little importance), then the above equation can be simplified to:

$$\mathbf{I_o = I_{abs} + I} \qquad (2)$$

The quantitative relation between the light intensity I_0 hitting the body and the amount of light leaving the body is described by Bouger-Lambert's law :

$$\mathbf{I = I_0\, e^{-\mu d}} \qquad (3)$$

where: μ – absorption coefficient typical of a specific substance,

d – thickness of the layer penetrated by the light.

Coefficient μ depends on the incident light wavelength, hence equation (3) can refer to both parallel and monochromatic light beam. The curve in Fig. 3 shows the diagram of Bouger-Lambert's law for a specific light wavelength i.e. for μ = const.

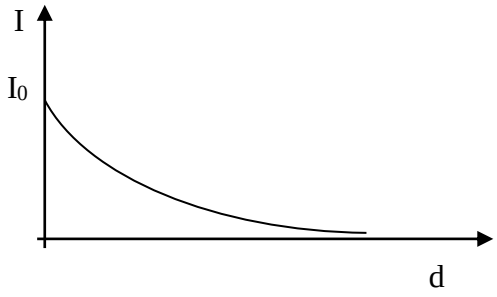


Fig. 3. Diagram of Bouger-Lambert's law I = f(d) for μ = const

For solutions which are composed of two kinds of molecules: a molecule of solvent and a molecule of dissolved substance and also of solutions of low concentration we can apply Beer's law, according to which the absorption coefficient μ is directly proportional to the solution concentration c, namely:

$$\mathbf{\mu = k \cdot c} \qquad (4)$$

where: k is the factor of proportionality independent of concentration and is called molar extinction coefficient (optical density, absorbance coefficient).

Beer's law similarly to Bouger-Lambert's law refers to monochromatic light beam. Inserting equation (4) into (3) we obtain Bouger-Lambert-Beer's law in the form:

$$I = I_0 e^{-kcd} \quad (5)$$

Equation (4) can be used only for solutions whose coefficient μ is linear function of concentration (Fig. 4 sector OA).

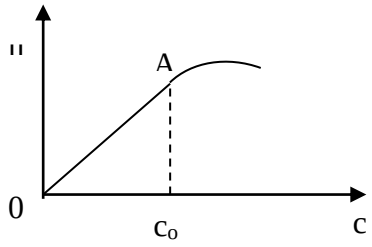


Fig. 4. Dependence curve μ = f(c). Bouger-Lambert-Beer's law refers to concentrations from 0 to c_o.

Dividing both sides of equation (5) by I_0 we obtain a quantity called the transmittance of the solution::

$$T = \frac{I}{I_0} = e^{-k \cdot c \cdot d} \quad (6)$$

Transmittance T provides information about how much of the incident light has been let through a specific solution. Transmittance is expressed in percentages. However, in spectrophotometric measurements extinction is used more frequently to describe this phenomenon. The relation between both these variables is inverse and logarithmic (in character).

Extinction E is the quantity that denotes the weakening of the light beam passing through an absorbent. This quantity can be written as follows:

$$E = \log \frac{I_0}{I} = \log \frac{1}{T} = \log e^{k \cdot c \cdot d} = k \cdot c \cdot d \cdot \log e = 0.43 \cdot k \cdot c \cdot d \quad (7)$$

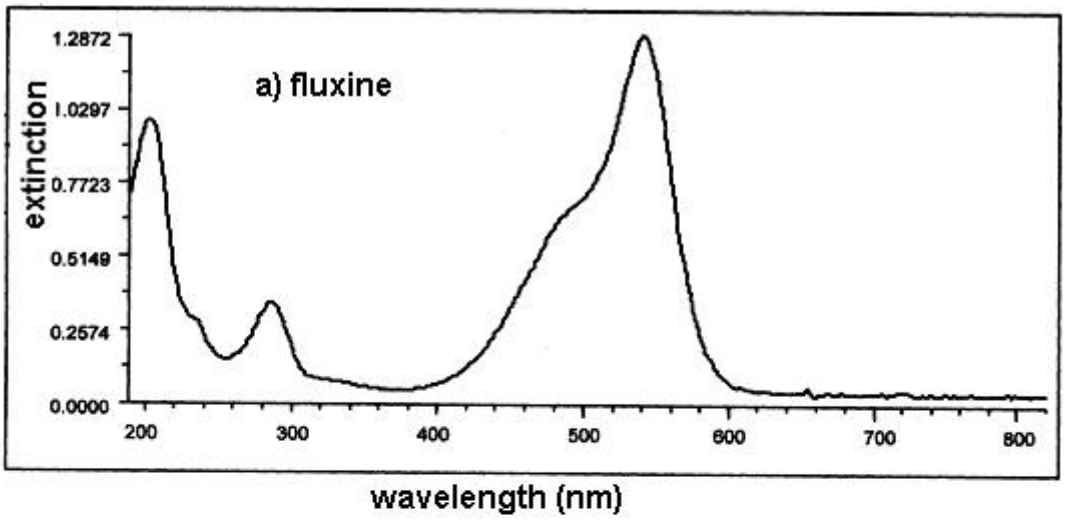
Both (6) and (7) are two different version of the same Bouger-Lambert-Beer's law. Equation (7) states that extinction of a parallel monochromatic light beam

passing through a solution is in directly proportional dependence on both the concentration and thickness of the solution.

Solutions of chemical substances have different extinction values at different wavelengths. This is the reason why it is very important to perform measurements of solutions using the monochromatic light beam. An example of an extinction of water solution of both fluxine and aniline blue in a function of light wavelength is shown in Figs 5a and 5b respectively.

As can be seen the absorption phenomenon is selective i.e. different wavelengths are absorbed in different degrees. For fluxine (Fig. 5a) green colour is absorbed in the highest degree (maximum extinction for $\lambda = 540$ nm). Hence the colour of the solution is red. However, for aniline blue (Fig. 5b) the absorption peak (with in the visible range) is reached at $\lambda = 580$ nm (the solution is blue).

To determine the concentrations of both these solutions in our experiment it is recommended to use exactly the same wavelengths of the monochromatic radiation that corresponds to maximum extinction values. The intensity of the colour according to Bouger-Lambert-Beer's law will be the greater the higher the solution of concentration there is.



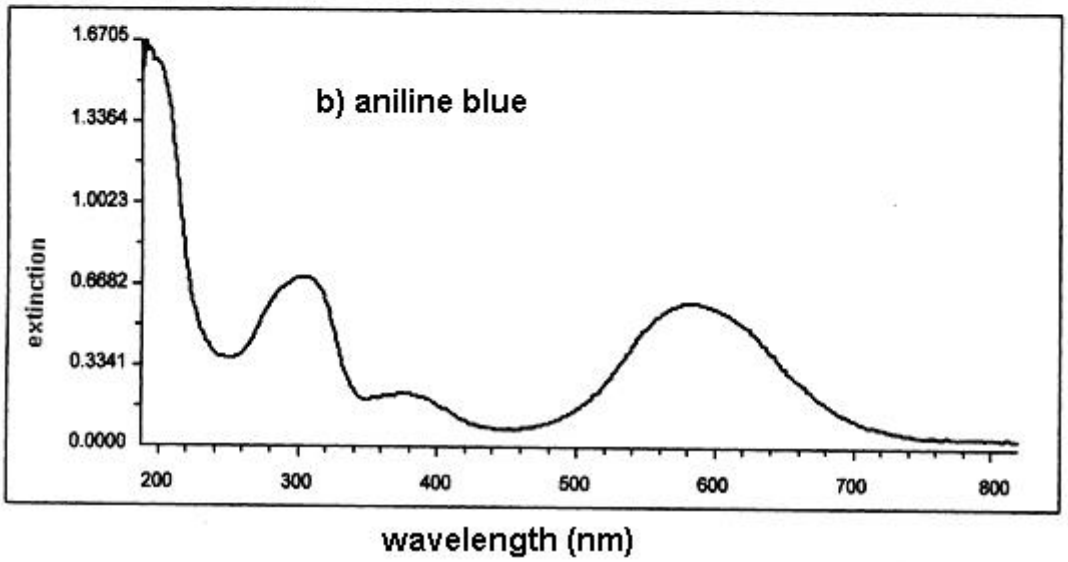


Fig 5. Extinction dependence of water solution fluxine (a) and aniline blue (b) on the light wavelength.

PRACTICAL PART

Discussion questions

1.What is the mechanism of colour vision of the human eye?

2. Describe the Bohr Model of the hydrogen atom.

EXPERIMENTAL PART

Objective: The determination concentration of solution

Materials: spectrophotometer SPECOL, 10% solution of $CuSO_4$, distilled water, cuvettes, pipettes.

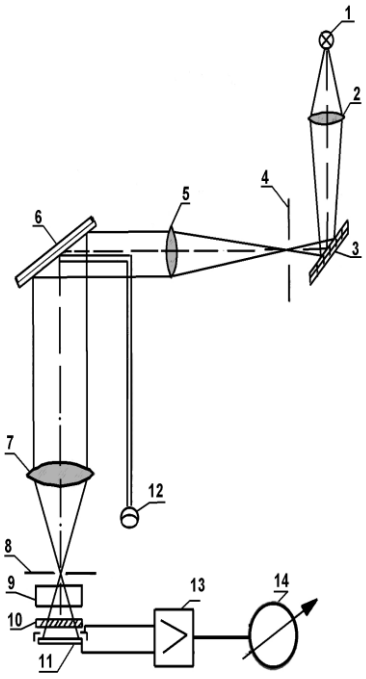


Fig. 6. Schematic diagram of the optical system of SPECOL spectrophotometer

1 – source of light, 2 – condenser, 3 – mirror, 4 – entrance slit, 5 – lens,

6 – reflection diffraction grating, 7 – lens, 8 – exit slit, 9 – investigated solution in the cuvette, 10 – colour filter, 11 – photo detector, 12 – knob for wavelength regulation, 13 – amplifier, 14 – measuring instrument.

Measurement method

The SPECOL Spectrofotometer used in this experiment works as follows:

The dispersed beam of white light coming from bulb (1) after passing through the system of a condenser (2), mirror (3), slit (4) and the second lens becomes a parallel light beam. The diffraction grating (6) splits the light into different wavelengths. By turning the grating with adjusting knob (12) the ray of specific wavelength is directed into the optical system and then onto the body to be investigated (most frequently it is a liquid placed in a special cuvette made of glass or quartz). After passing through the substance investigated the light reaches photodetector (11) which produces a voltage dependent on the intensity of the incident light.

It is not enough to perform a single measurement of the investigated solution to determine its extinction as we do not know the intensity of the light beam hitting the cuvette. The incident beam is absorbed by the cuvette material, the solvent substance and by the molecules of the substance under investigation. As a result SPECOL measurements are performed using a reference standard, which is most commonly distilled water.

To begin the measurements it is necessary to set zero extinction on the instrument for distilled water (i.e. 100% transmission) and next using the same settings we place the investigated substance (instead of the distilled water) on the path of the light beam. Now it is possible to read out either extinction or transmission values directly from the scale of the measurement instrument (14). Using the above method we eliminate light absorption by the solvent itself (i.e. water).

Principles of determining solution concentrations

Measurements of the dependencies of light extinction in solutions on their concentrations make sense only for those light wavelengths whose light bands are absorbed by the solution. If, for example, we examine the absorption spectrum of fluxine concentration (Fig. 5a), we will notice that it has the greatest absorption value at λ_1 = 540 nm, whereas at wavelength λ_2 = 640 nm the absorption is practically equal to zero. If we carried out the measurement of light extinction dependence of fluxine solution on its concentration at wavelength λ_2 = 640 nm,

then we would not be able to find such a dependence as at this wavelength light is practically not absorbed by the solution.

Thus, prior to spectrophotometric determination of solution concentrations it is essential to examine the absorption spectrum of the solution. This analysis will allow us to determine the wavelengths at which light is not absorbed in the solution (it's evident that at such wavelengths we cannot examine the dependence of extinction solution concentration). The analysis will also tell us at which wavelengths light is absorbed in the solution. The influence of the solution concentration on light absorption in the solution will be the greatest at such a wavelength at which the absorption reaches its maximum value. It also means that this wavelength is the one at which we should determine the solution concentration in our investigations.

To determine solution concentration using measurements of light extinction in the solution, we should make use of the fact that this parameter is linearly dependent on its concentration (i.e. it is directly proportional).This dependence is different for each solution and must be determined by experiment. The spectrophotometer is used to determine the extinction values of solution whose concentrations are known.

Making use of these measurement data we can find the relationship between light extinction in a solution and its concentration and express these relations graphically or numerically. We can create relationship charts of light extinction in a solution and its concentration. This dependence has a linear character and thus can be presented by an equation:

$$y = b \cdot x + a$$

where: x is the solution concentration, y is the extinction value.

Using the least square method we determine linear regression coefficients b and a in equation $y = b \cdot x + a$

$$b = \frac{N\sum xy - \sum x \sum y}{N\sum x^2 - (\sum x)^2} \qquad a = \frac{\sum y - b\sum x}{N}$$

In this way we find an experimental dependence of the extinction on solution concentration. To determine the equation and correlation coefficient R^2, a computer program, for example, „Microsoft Excell" can be used. Fig. 8 gives an example of such procedure.

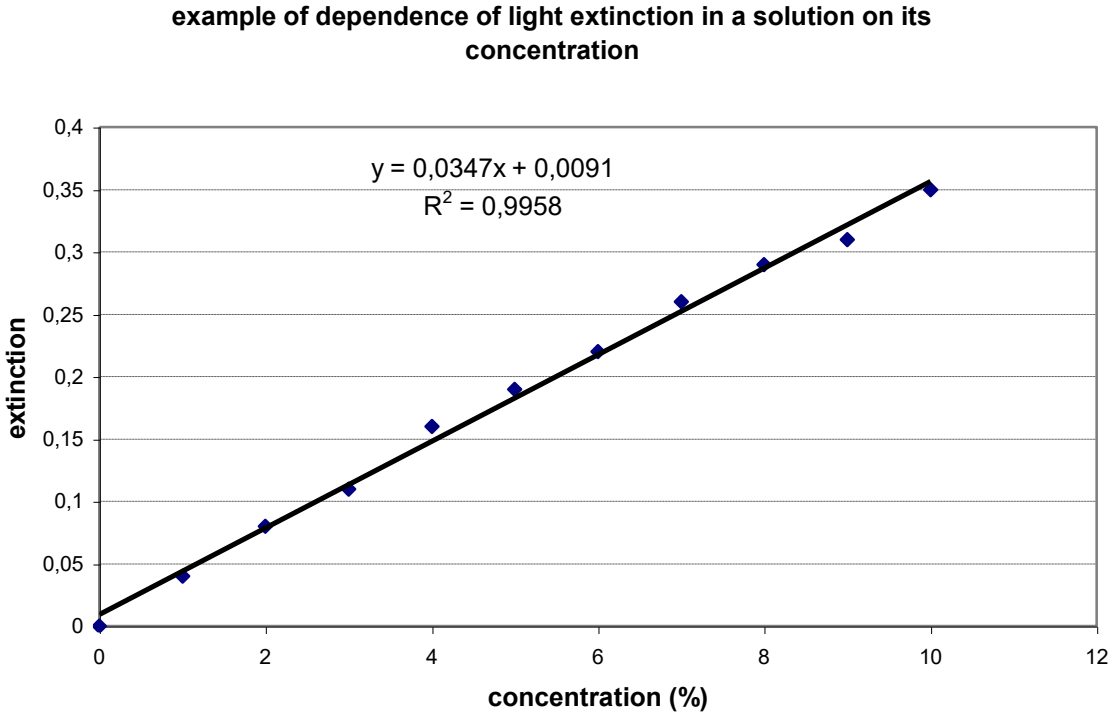


Fig. 8. Example of linear regression.

The dependence is considered to be linear and can be used to determine the concentration only if the correlation coefficient $R^2 > 0,95$.

Now if we wish to determine an unknown solution concentration of the same substance, it is necessary to measure by means of the spectrophotometer the values of light extinction in the solution in the same conditions (i.e. at the same wavelength and in the same cuvette). Next the measured values must be referred to the straight line on the chart and knowing parameters "b" an "a" of equation $y = b \cdot x + a$ we can determine value "x" knowing the measured value "y".

For instance, if the dependence of light extinction in the solution on its concentration is just like the one shown in Fig. 8, i.e. $y = 0,0347 \cdot x + 0,0091$ and the measured extinction value for the solution of an unknown concentration is 0.17, then the solution concentration is determined by inserting value 0.17 instead of "y" into the equation and finding value "x" from the equation. The value obtained is 4,64% (please check the calculations yourselves).

a) Solution preparation

- Prepare aqueous solutions of cupric sulphate ($CuSO_4$) in the following concentrations 1%, 2%, 3%, 4%, 5%, 6%, 7%, 8%, 9% and 10% (10 ml of each).
- Each lab group divided into two subgroups prepares an aqueous solution of cupric sulphate (10 ml) of undisclosed concentration – x_0. Write down x_0 value of your subgroup, x_0 =

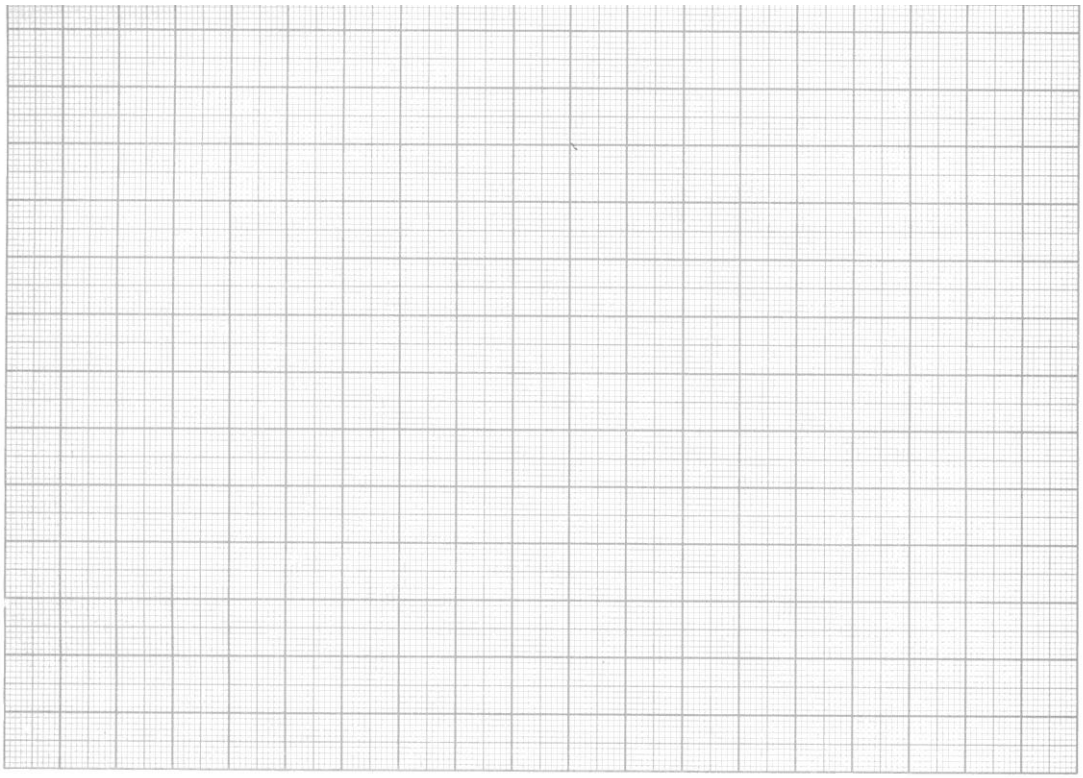
Data and observations:

1. Investigation of $CuSO_4$ absorption spectrum

Find the value of light extinction in 10% solution of $CuSO_4$ changing the wavelength every 10 nm within the visible range of EM spectrum. Write the results into the table below.

λ[nm]														
E														
λ[nm]														
E														

Draw the graphic relation of light extinction in the solution and the wavelength in the graph paper.



- Here write down the wavelength at which the extinction reaches its maximum λ_{max} =

At λ_{max} measure the extinction for all ten solutions of $CuSO_4$ concentrations from 1% to 10%. Present the result in the table below. Make sure to conduct each measurement in the same experimental conditions i.e. use the same dry clean cuvette at the same wavelength.

No	Solution concentration c [%]	E			Average value E
		1	2	3	
1	1				
2	2				
3	3				
4	4				
5	5				
6	6				
7	7				
8	8				
9	9				
10	10				

Using a computer program find the linear dependence between light extinction and solution concentration for the values obtained.

Write down here:

- Equation obtained: y =

- Value of the correlation coefficient R^2 =

Next we measure the light extinction value in the solution prepared by the second subgroup.

Write down here:

- Measured extinction value E =

Making use of the obtained dependence of light extinction value on the solution concentration, calculate the solution concentration „x” prepared by the second subgroup.

Write down the calculations here:

Write down here:

- Calculated value of concentration "x" =

Determine the percentage error of the measurement:

$$b = \frac{|x - x_0|}{x_0} \cdot 100\% =$$

Where x_0 – solution concentration prepared by the second subgroup.

The date	Student's name and surname	Lab assistant signature

NOTES

ASSIGNMENTS FOR ELECTROMEDICINE LAB EXERCISES

2.1 Lab Exercise

The oscilloscope

1. How does an oscilloscope work?
    - thermionic emission
    - acceleration of electrons
    - luminescence
    - horizontal and vertical deflections:
2. Measuring voltage and time period.

2.2 Lab Exercise

Biophysics of phonation

1. Properties of sound: intensity, frequency, sound pattern.
2. Some properties of waves: reflection and refraction, interference and diffraction
3. Resonant standing waves: resonant frequencies
4. Subjective sensation of sound and its correlation with the physical properties of the sound:
    - Frequency and pitch
    - Intensity and loudness
    - Harmonic content (sound pattern) and quality (timbre)
5. The principles of the phonation.

2.3 Lab Exercise

Hearing testing

1. Structures and functions of the outer, middle and inner ear.
2. Types of hearing testing.
3. The principles of the pure tone audiometry.
4. The principles of the tympanometry.
5. Intensity scales.

2.4 Lab Exercise

Electrocardiography

1. Parts of the heart’s electrical conduction system
2. Cardiac action potentials
    - Pacemaker action potentials
    - Non-pacemaker action potentials
3. The "12 lead ECG"
    - The 3 standard limb leads (bipolar)
    - The 3 augmented limb leads (unipolar)
    - The 6 precordial leads (unipolar)
4. Waves, segments and intervals in the ECG
5. Fundamental principles of reading the ECG
    - Measuring time of waves, segments and intervals in the ECG
    - Estimation of the heart rate
6. The mean electrical axis

2.5 Lab Exercise

The measurement of the blood flow velocity

1. Production of ultrasound
    - inverse piezoelectric effect
    - magnetostriction
2. The properties of ultrasound
    - propagation velocity
    - acoustic impedance
3. The Doppler Effect
4. Continuous wave Doppler ultrasound
5. Spectral Doppler waveform measurements
6. Biophysical effects of ultrasound
    - thermal effects
    - non-thermal (mechanical) effects
7. Major uses of ultrasound in diagnostic

2.6 Lab Exercise

The circulatory system – the fundamentals of motion of fluids

1. Blood pressure measurement's methods
2. The auscultatory method
    - viscous force
    - laminar flow
    - turbulent flow
    - Reynolds number
    - critical flow velocity
    - Korotkoff sounds
3. Pressure measurement equipment
4. The role of gravity in the circulation

LITERATURE:

1. Paul Davidovits – "Physics in Biology and Medicine"
2. Roland Glaser – "Biophysics"

2.1 Lab exercise

THE OSCILLOSCOPE

THEORY

Oscilloscopes are used by everyone from television repair technicians to physicists. The usefulness of an oscilloscope is not limited to the world of electronics. With the proper **transducer**, an oscilloscope can measure all kinds of phenomena. A transducer is a device that creates an electrical signal in response to physical stimuli, such as sound, mechanical stress, pressure, light, or heat.

An automotive engineer uses an oscilloscope to measure engine vibrations. A medical researcher uses an oscilloscope to measure brain waves. The possibilities are endless.

How Does an Oscilloscope Work?

An oscilloscope is an electron gun with horizontal and vertical deflection plates. The gun consists of a cathode and an anode, enclosed in a vacuum tube as shown below in Figure 1. The cathode is heated until the temperature is so great that the metal electrons of the filament vibrate strongly enough to knock electrons

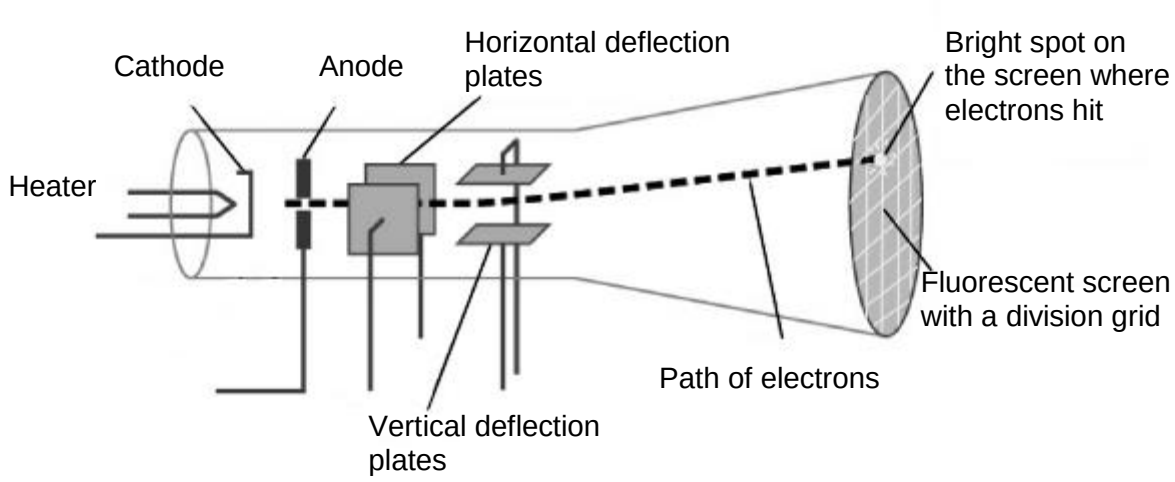
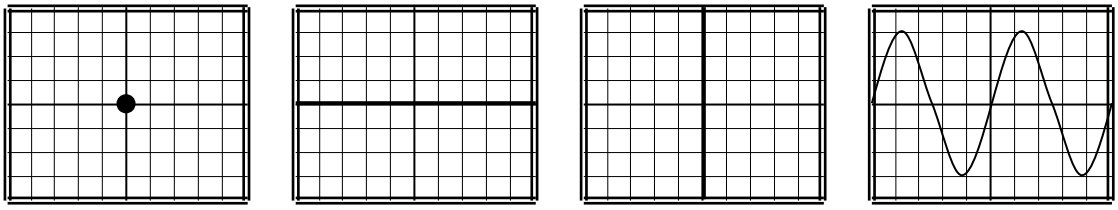


Fig.1 A cathode-ray tube (CRT).

free of the metal (**thermionic emission**). If a potential difference is applied between the anode (positive) and the cathode (negative), the electrons will

accelerate towards the anode. A small hole in the centre of the anode allows a narrow beam of these accelerated electrons to pass through, till they hit the flat screen, which is lined with luminescent material (usually phosphor). When the electrons hit this material, light is emitted (**luminescence**). The phosphor glows at the point of impact of the electron beam, and that glow is persistent enough for the entire trace of the electron beam across the screen to be seen until the next voltage ramp sweeps it across again



(A) (B) (C) (D)

Fig. 2 Different oscilloscope signals on the fluorescent screen.

In the absence of the voltage applied to deviating plates of the capacitor the electron beam will remain in the centre of the screen in the centre creating a spot in the fluorescing layer (Fig. 2A). **Horizontal deflections** can be produced with horizontal electric fields between the parallel plates. These plates have a separation *d* (Fig. 3).

I

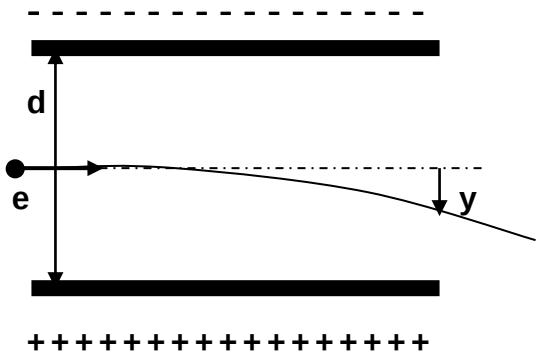


Fig.3 Electrons deflection between horizontal plates.

f they are at a potential difference V_1, there is a uniform horizontal electric field *E* between them.

$$\mathbf{E} = \frac{\mathbf{V_1}}{\mathbf{d}}$$

This field accelerates the electrons, which then strike the screen at a horizontal distance from the centre, which is proportional to V_1. When V_1 is gradually increased the spot gradually moves or sweeps across the screen; when V_1 returns to its original value the spot returns to its starting point. If this sweep is repeated at a high enough frequency, the persistence of the image on the screen and in the eye conceals the motion, and a straight line is seen (Fig. 2B). In the same way, a potential difference V_2 applied to the vertical plates, will caused **vertical deflections** (Fig. 2C). Complex variations of the vertical field with time can be displayed if they repeat at the sweep frequency (Fig. 2D).

Oscilloscope Terminology

Learning a new skill often involves learning a new vocabulary. This section describes some useful measurement and oscilloscope performance terms.

Measurement Terms

The generic term for a pattern that repeats over time is a **wave** - sound waves, brain waves, ocean waves, and voltage waves are all repeating patterns. An oscilloscope measures voltage waves. One **cycle** of a wave is the portion of the wave that repeats. A **waveform** is a graphic representation of a wave. A voltage waveform shows time on the horizontal axis and voltage on the vertical axis.

Waveform shapes tell you a great deal about a signal. Any time you see a change in the height of the waveform, you know the voltage has changed. Any time there is a flat horizontal line, you know that there is no change for that length of time. Straight diagonal lines mean a linear change - rise or fall of voltage at a steady rate. Sharp angles on a waveform mean sudden change. Figure 4 shows common waveforms.

The **sine wave** is the fundamental wave shape for several reasons. It has harmonious mathematical properties - it is the same sine shape you may have

studied in high school trigonometry class. The voltage in your wall socket varies as a sine wave. Test signals produced by the oscillator circuit of a signal generator are often sine waves. Most AC power sources produce sine waves. (*AC* stands for alternating current, although the voltage alternates too. *DC* stands for direct current, which means a steady current and voltage, such as a battery produces.)

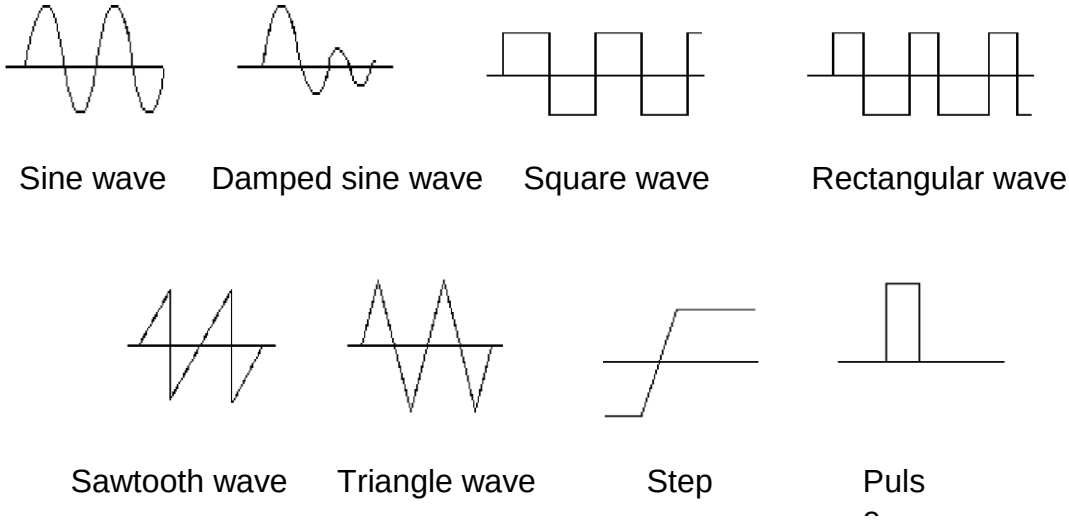


Fig. 4 Common Waveforms

The **damped sine wave** is a special case you may see in a circuit that oscillates but winds down over time.

The **square wave** is another common wave shape. Basically, a square wave is a voltage that turns on and off (or goes high and low) at regular intervals. It is a standard wave for testing amplifiers. Television, radio, and computer circuitry often use square waves for timing signals.

The **rectangular wave** is like the square wave except that the high and low time intervals are not of equal length. It is particularly important when analyzing digital circuitry.

Sawtooth and **triangle waves** result from circuits designed to control voltages linearly, such as the horizontal sweep of an analog oscilloscope or the raster scan of a television. The transitions between voltage levels of these waves change at a constant rate. These transitions are called *ramps*.

Signals such as **steps** and **pulses** that only occur once are called *single-shot* or *transient* signals. The step indicates a sudden change in voltage, like what you would see if you turned on a power switch. The pulse indicates what you would see if you turned a power switch on and then off again.

Measuring voltage and time period

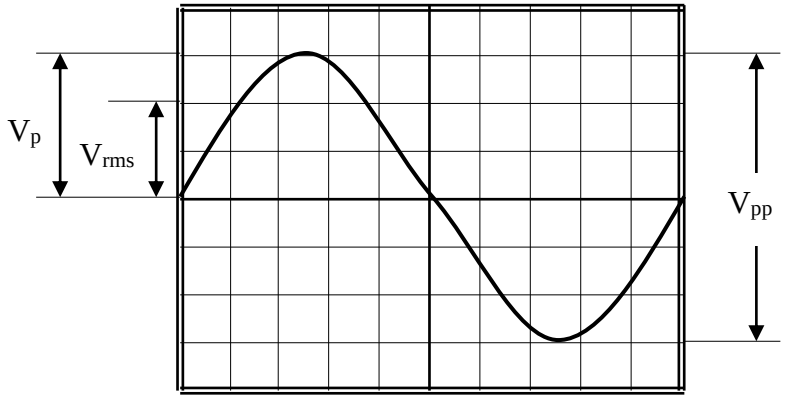
The trace on an oscilloscope screen is a **graph of voltage against time**. The shape of this graph is determined by the nature of the input signal.

In addition to the properties labelled on the graph, there is frequency which is the number of cycles per second.

The diagram shows (Fig. 5) a **sine wave** but these properties apply to any signal with a constant shape. In general electrical engineering, alternating voltage data normally refers to effective values (**V_{rms} -root-mean-square value**). If a sinusoidal waveform, displayed on the oscilloscope screen, is to be converted into an effective value, the resulting peak-to-peak value must be divided by $2\sqrt{2} = 2.83$ (Fig. 5).

Setting the Controls

After plugging in the oscilloscope, take a look at the front panel. It is divided into three main sections labelled *Vertical*, *Horizontal*, and *Trigger*. Your



$$V_{rms} = \frac{V_{pp}}{2\sqrt{2}}$$

$$V_{rms} = \frac{V_p}{\sqrt{2}}$$

Fig. 5 Voltage values of sine curve.
V_{rms} – the effective value; V_p – the voltage of one peak (the crest value);
V_{pp} – the peak-to-peak voltage, which is usually twice V_p.

oscilloscope may have other sections, depending on the model and type (analog or digital).

Notice the input connectors on your oscilloscope. This is where you attach probes. Most oscilloscopes have at least two input channels and each channel can display a waveform on the screen.

The vertical position control lets you move the waveform up or down to exactly where you want it on the screen.

The Y AMPLIFIER control determines the height of the trace. The volt per division (usually written V/div) setting varies the size of the waveform on the screen.

With the designations

H – observed display height in div;

U – required signal voltage in V_{pp} at the vertical input;

D – deflection coefficient in V/div at attenuator switch

the required value can be calculated from the two given quantities:

$$U = D \cdot H$$

An example:
Set deflection coefficient D = 0.05 V/div,
observed display height H = 4.6 div,
required voltage U = 0.05 x 4.6 = 0.23 V

The horizontal position control moves the waveform from left and right to exactly where you want it on the screen.

The **TIMEBASE** control determines the rate at which the dot sweeps across the screen. The seconds per division (usually written as s/div) setting lets you select the rate at which the waveform is drawn across the screen (also known as the time base setting or sweep speed). The time coefficients are stated in ms/div, μs/div or ns/div. The duration of a signal period or a part of it is determined by multiplying the relevant time (horizontal distance in div) by the time coefficient.

With the designations:

L – observed displayed wavelength in div,

T – time in seconds for one period,

f – recurrence frequency in Hz of the signal,

T_c – time coefficients in ms/div, μs/div or ns/div and the relation, $f = 1/T$

the following equations can be stated:

$$T = L \cdot T_c \Rightarrow f = \frac{1}{L \cdot T_c}$$

An example:
Set time coefficient T_c = 100 ns/div,
observed display wavelenght L = 7 div,
required period T = 7 x 100 x 10^{-9} = 0.7 μs,
required rec. frequency f = 1: (0.7 x 10^{-6}) = 1.428 x 10^{6} Hz = 1.428 MHz

PRACTICAL PART

Explain terms:

Thermionic emission:

Acceleration of electrons:

Luminescence:

Horizontal and vertical deflections:

EXPERIMENTAL PART

Precautions

- An oscilloscope should be handled gently to protect its fragile (and expensive) vacuum tube.
- Oscilloscopes use high voltages to create the electron beam and these remain for some time after switching off - for your own safety do not attempt to examine the inside of an oscilloscope!

Part A

Objective: Setting up an oscilloscope

Materials: an oscilloscope, a function generator

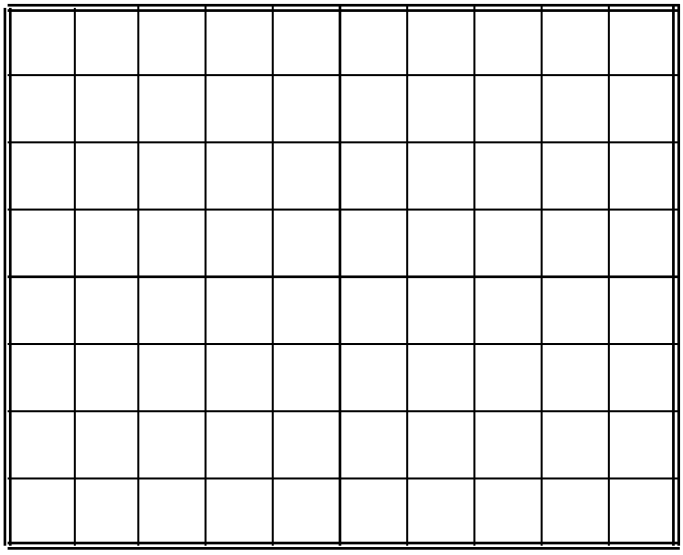
Procedure

Oscilloscopes are complex instruments with many controls and they require some care to set up and use successfully. It is quite easy to 'lose' the trace off the screen if controls are set wrongly!

There is some variation in the arrangement and labeling of the many controls so the following instructions may need to be adapted for your instrument.

1. **Switch on** the oscilloscope to warm up (it takes a minute or two).
2. Set the **Y AMPLIFIER** to **5 V/div** (a moderate value).
3. Set the **TIMEBASE** to **10 ms/div** (a moderate speed).
4. Adjust **Y SHIFT** (up/down) and **X SHIFT** (left/right) to give a **trace across the middle of the screen**, like the picture.
5. Adjust **INTENSITY** (brightness) and **FOCUS** to give a bright, sharp trace.
6. **Switch on** the function generator.
7. **Adjust** the **TIMEBASE** of the oscilloscope or the frequency of the function generator to display one period of **sine wave** and draw this image below

Data and observations



V/div	
time/div	

8. **Calculate** the signal voltage – U; time in seconds for one period (T); the voltage of one peak (the crest value) - V_p; the peak-to-peak voltage - V_{pp} and the effective value - V_{rms};.

U [V]	**T [s]**	**V_p [V]**	**V_{pp} [V]**	**V_{rms} [V]**

Calculations:

Part B

Objective: Measurement of sine wave frequency changes.

Materials: an oscilloscope, a function generator

Procedure

1. **Adjust** the **TIMEBASE** of the oscilloscope or the frequency of the function generator to display **one period** of **sine wave**
2. **Increase the frequency** of the generator to get **two periods** of sine wave
3. Then, **increase** the frequency of the generator to get **three periods** of sine wave and etc.
4. Set your data at the table below

Data and observations

The frequency of a generator [Hz]	The number of periods	The difference of frequency $f_n - f_{n-1}$
f_1 =		
f_2=		
f_3=		
f_4=		
f_5=		

The date	Student's name and surname	Lab assistant signature

NOTES

2.2 Lab exercise

BIOPHYSICS OF PHONATION

THEORY

The voice consists of the anatomical and neurological properties employed in phonation and articulation. The physiological ability to create vocalised speech and the cognitive processes involved in language use appear to be unique traits of human beings. The process of converting the air pressure from the lungs into audible vibrations is called **phonation**, from the Greek **phonē**, meaning “voice” or “sound’.

The **vocal tract** is the instrument of the human voice. The vocal tract is a stopped air column approximately 17 cm long. It consists of the air passages from the vocal cords to the lips, including the nasal cavity (Fig.1). The vocal tract behaves like a resonance chamber, amplifying and attenuating certain

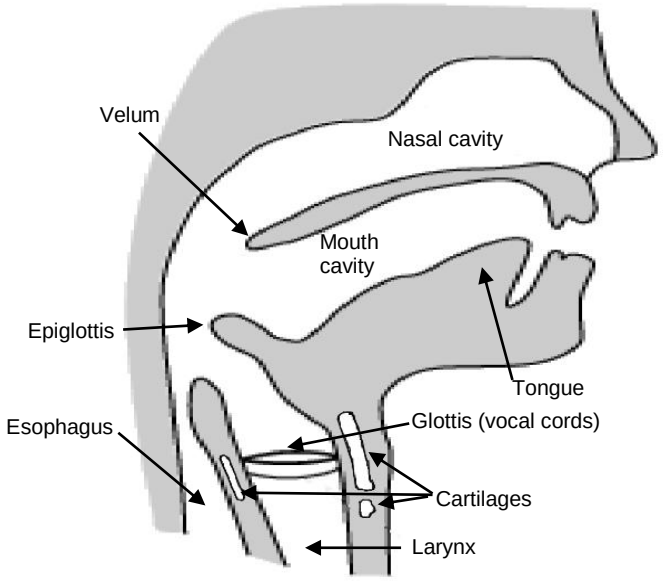


Fig.1 Essential components of human speech mechanism.

frequencies. The shape and size of the vocal tract can be modified by moving the lips, tongue, velum, etc. which results in continually changing resonant frequencies. Consequently a spectrum of frequencies is produced which contains peaks at certain frequencies.

Phonation occurs in the larynx**. The larynx** consists of four basic components: cartilaginous skeleton, intrinsic and extrinsic muscles, and a mucosal lining. The intrinsic muscles of the larynx alter the position, shape, and tension of the vocal folds (vocal cords). The front end of the vocal folds is attached to the thyroid cartilage, the "Adam's apple". The back end is attached to the arytenoid cartilages, which move to separate (Fig.2). The vocal cords are relaxed during normal breathing, when speech is begun the vocal cords tension increases and the larynx is closed.

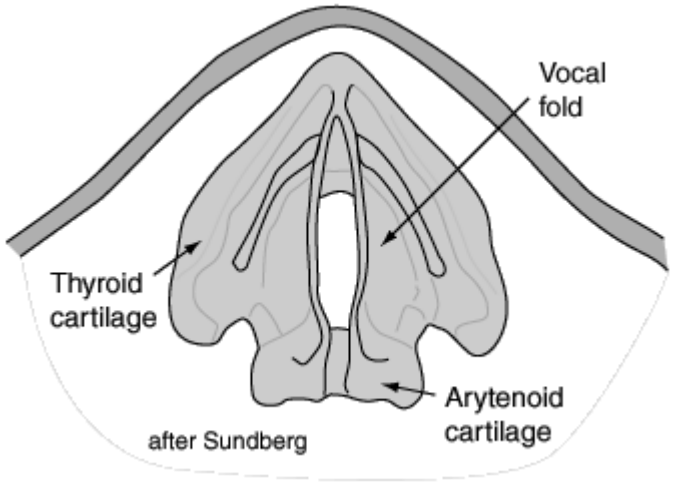


Fig.2 The vocal cords.

The production of voice can be thought of in terms of three components: the production of airflow, the generation and resonance of sound, and the articulation of voice. **The lungs** first supply adequate airflow to overcome the resistance of the closed vocal cords. The vocal cords are finely tuned neuromuscular units that adjust pitch and tone by altering their position and tension. Air pressure develops below vocal cords because of air from lungs during exhalation. Positive air pressure from the lungs forces them open momentarily, but the high velocity air produces a lowered pressure by the Bernoulli Effect which brings them back together. The folds themselves have a resonant frequency which determines voice pitch. The final modification of the voice occurs in the mouth, nose, and throat. This is where the tongue, palate, cheek, and lips are involved in articulating the words we understand.

In an adult male, the vocal folds are usually 17-23 mm long, and12.5 -17 mm in an adult female. They may be stretched 3 or 4 mm by action of the muscles in the larynx.

The vocal cord vibration cycle occurs 200-400 times a second during sound production. The rapid opening and closing of the vocal cords occur in a vibratory pattern and are responsible for sound production.

The male speaking voice averages about 125 Hz, while the female voice averages about 210 Hz. Children's voices average over 300 Hz.

The human oral cavity is a complex structure, but its properties are close to those of simple model of a pipe 0.17 m long with one end open at the mouth and nose and the other end nearly closed at the vocal cords. The resonant frequencies of a pipe open at one end were found to be:

$$f_n = \frac{(2n - l) \cdot c}{4l}$$

where : *n* – an integer (n=1, 2, 3, …)

ℓ – the length of a pipe (note that *2n-1* is always an odd integer)

c – the velocity of sound in the air (344 m/s)

In speech the frequencies of importance are between 300 and 3000 Hz. Using c=344 m/s and ℓ =0.17 m, we find that the fundamental and first two overtones of the pipe, which are near 500, 1500, 2500 Hz, are within this range.

Vocal Formants

If we analyse a typical voice sound, we find that there usually are three resonant peaks that are close to the resonant frequencies of the pipe model. These resonant peaks are referred to as **formants**. So, **the term formant** refers to peaks in the harmonic spectrum of a complex sound which arise from some sort of resonance of the source. Because of their resonant origin, they tend to stay essentially the same when the frequency of the fundamental is changed. Formants in the sound of the human voice are particularly important because they are essential components in the intelligibility of speech. For example, the distinguish ability of the vowel sounds can be attributed to the differences in their first three formant frequencies. Producing different vowel sounds amounts to retuning these formants within a general range of frequencies.

The process of articulation determines the frequencies of the vocal formants. The jaw opening, which constricts the vocal tract toward the glottal end and

expands it toward the lip end, is the deciding factor for the first formant. This formant frequency rises as the jaw is opened wider. The second formant is most sensitive to the shape of the body of the tongue, and the third formant is most sensitive to the tip of the tongue.

Speech Analysis

Phonetics is part of the linguistic sciences. It is concerned with the sounds produced by the human vocal organs, and more specifically, the sounds which are used in human speech. One important aspect of phonetic research is the instrumental analysis of speech.

The instrumental analysis is performed using one or many of the available instruments. These include X-ray photography and film, air-flow tubes, electromyography (EMG), spectrographs, mingographs, laryngographs etc. The aim for most of these methods is to visualise the speech signal in some way, and to try and capture some aspects of the speech signal on paper or on a computer screen. Today the computer is the most readily available and used tool. With the computer the analysis process is much simpler and usually faster than with other tools, however, it does not necessarily produce a result of higher quality.

Oscillogram (waveform) (Fig.3A)

Physically the speech signal (actually all sound) is a series of pressure changes in the medium between the sound source and the listener. The most common representation of the speech signal is the oscillogram, often called the waveform. In this the time axis is the horizontal axis from left to right and the curve shows how the pressure increases and decreases in the signal.

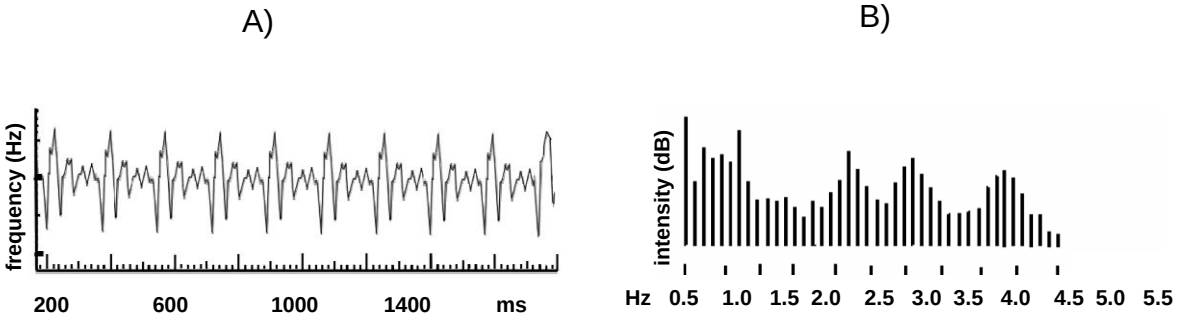


Fig. 3 Speech Analysis – a spectrum of the vowel “e”: A) – An Oscillogram (a waveform); B) - The fundamental frequency (the pich analysis).

Fundamental frequency (pitch, F0) (Fig.3B)

Another representation of the speech signal is the one produced by a pitch analysis. Speech is normally looked upon as a physical process consisting of two parts: a product of a sound source (the vocal chords) and filtering (by the tongue, lips, teeth etc). The pitch analysis tries to capture the fundamental frequency of the sound source by analysing the final speech utterance. The fundamental frequency is the dominating frequency of the sound produced by the vocal chords. This analysis is quite difficult to perform. There are several problems in trying to decide which parts of the speech signal are voiced and which are not. It is also difficult to decipher the speech signal and try to find which oscillations originate from the sound source, and which are introduced by the filtering in the mouth. Several algorithms have been developed, but no algorithm has been found which is efficient and correct for all situations. The fundamental frequency is the strongest correlate to how the listener perceives the speakers' intonation and stress.

PRACTICAL PART

Answer the questions:

1. What are sound waves and what properties of sound do you know? Describe them:

2. Shortly describe some properties of waves: reflection and refraction, interference and diffraction.

3. What are resonant standing waves: resonant frequencies?

4. Shortly describe subjective sensation of sound and its correlation with the physical properties of the sound: frequency and pitch; intensity and loudness; harmonic content (sound pattern) and quality (timbre).

5. What are some functions of the larynx?

6. How do the vocal chords produce sound?

EXPERIMENTAL PART

Objective: Exploration of sounds produced by a sound generator, and sound waves that YOU produce.
Materials: a sound generator, a speaker, an oscilloscope, a tuning fork.

Part A

Procedure and observations

In the first part of this activity a sound generator generates output signals that are played through a speaker and displayed on the oscilloscope screen. Compare subjective sensation of sound to the physical properties of the sound:

YOUR name	The lower limit of heard frequencies [Hz]	The upper limit of heard frequencies [Hz]

1. At **constant intensity** of the sound, **change** slowly it's frequency
2. **Observe** how the sensation of loudness is changing
3. **Write down** YOUR observations

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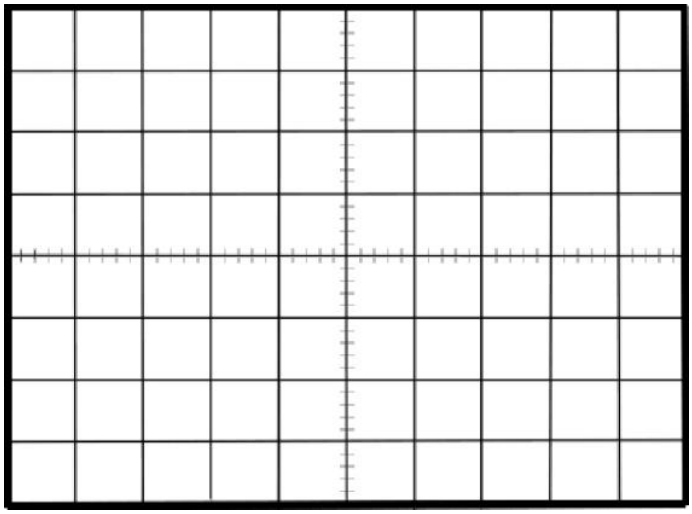
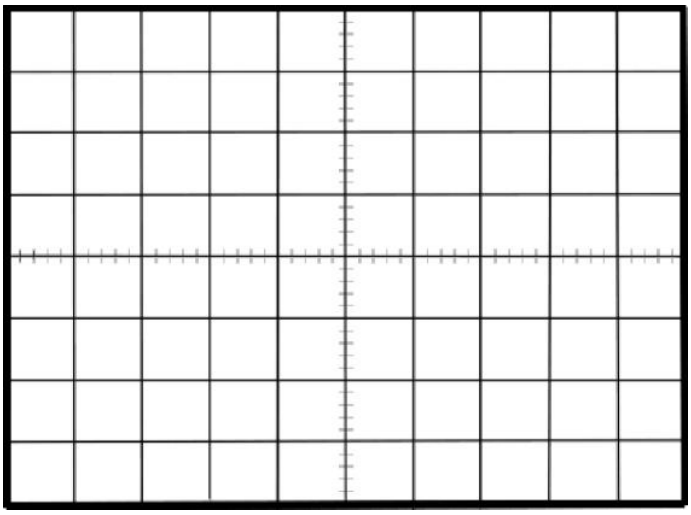
Part B

Procedure and observations

1. **Utter** vowels into the Sound Sensor and observe their waveform on the oscilloscope screen.
2. **Draw** their spectrums below.



spectrum of the vowel.....



spectrum of the vowel.....

Part C

Procedure and observations

1. **Observe** the spectrum of a tuning fork on the oscilloscope screen.
2. **Calculate** the frequency of the tuning fork.
3. **Write** obtained data in the table below.
4. **Fill in** the brackets with the **proper units**.

Timebase [ ]	The number of squares in one period	The period ***T*** [ ]	The frequency ***f*** [ ]

Calculations:

The date	Student's name and surname	Lab assistant signature

NOTES

Lab exercise

HEARING TESTING

THEORY

Hearing testing is a means of evaluating an individual's overall hearing function. Hearing tests help detect hearing loss, identify how severe it is, and determine what is causing it. They measure the ability of sound to reach the brain. Sounds are actually vibrations of different frequencies and intensities in the air around us; air in the ear canals and bones in the ears and skull help these vibrations travel from the ear to the brain. By measuring the ability to hear sounds that reach the inner ear through the ear canal (air-conducted sounds) and sounds transmitted through bones (bone-conducted sounds), hearing tests can also help determine the kind of hearing loss.

Most hearing tests **require a response** to a series of tones or words. These tests include:

- Whispered speech testing, which is a simple screening test that assesses your ability to hear whispered speech across a short distance.
- Pure tone audiometry, which measures your ability to hear sounds that reach the inner ear through the ear canal (air conduction). By using vibrations, this test can also measure hearing through bone (bone conduction).
- Tuning fork testing, which assesses how well sound moves through your ear.
- Speech reception and word recognition testing, which measure your ability to hear and understand speech.

Hearing tests that **do not require a response** (often used to screen newborns for hearing problems) include:

- Otoacoustic emissions (OAE) testing, which measures the inner ear's response to sound
- Brainstem auditory evoked responses (BAER), which evaluates the function of nerve pathways in the brain that are required for hearing.
- Electrocochleography (ECOG) is a variant of BAER where an electrode is placed on or in the ear drum to increase the size of wave 1 of the BAER.

- Tympanometry is a measure of the stiffness of the eardrum and thus evaluates middle ear function. This test can be helpful in detecting fluid in the middle ear, negative middle ear pressure, disruption of the ossicles, tympanic membrane perforation, and otosclerosis.

The pure tone audiometry and tympanometry are briefly described below:

Pure Tone Audiometry

Saying that two sounds have equal intensity is not the same thing as saying that they have equal loudness. Since the human hearing sensitivity varies with frequency, it is useful to plot equal loudness curves which show that variation for the average human ear (Fig. 1). If 1000 Hz is chosen as a standard frequency, then each equal loudness curve can be referenced to the decibel level at 1000 Hz. This is the basis for the measurement of loudness in phons. If a given sound is perceived to be as loud as a 60 dB sound at 1000 Hz, then it is said to have a loudness of 60 phons.

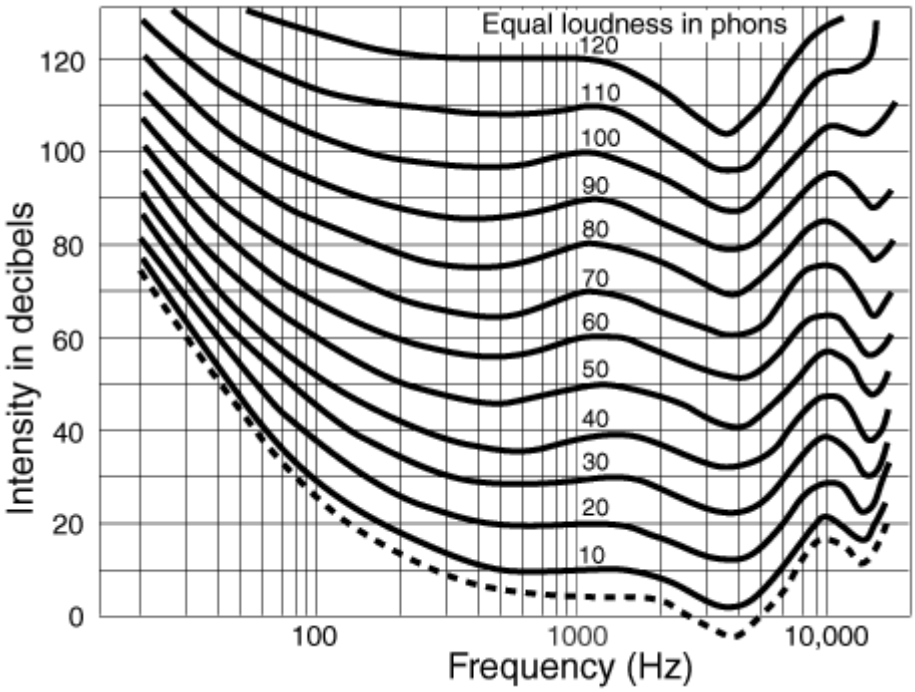


Fig. 1 Equal loudness curves.

Threshold of Hearing (I_0)

Sound level measurements in decibels are generally referenced to a standard threshold of hearing at 1000 Hz for the human ear which can be stated in terms of sound intensity:

$$I_0 = 10^{-12}\,\text{watts}/\text{m}^2$$

Sound intensity is defined as the sound power per unit area. The most common approach to sound intensity measurement is to use the decibel scale. The sound intensity $\mathbf{I}$ may be expressed in decibels above the standard threshold of hearing $\mathbf{I_0}$. The expression is:

$$I(\text{dB}) = 10\log_{10}\left[\frac{I}{I_0}\right]$$

The logarithm involved is just the power of 10 of the sound intensity expressed as a multiple of the threshold of hearing intensity. Example: If $\mathbf{I}$ = 10000 times the threshold:

$$I(\text{dB}) = 10\log_{10}\left[\frac{10000 I_0}{I_0}\right] = 10\cdot 4 = 40\text{dB}$$

The factor of 10 multiplying the logarithm makes it decibels instead of Bels, and is included because about 1 decibel is the just noticeable difference in sound intensity for the normal human ear. This value has wide acceptance as a nominal standard threshold and corresponds to 0 decibels.

The standard threshold of hearing can be stated in terms of pressure and the sound intensity in decibels can be expressed in terms of the sound pressure:

$$P_0 = 2\cdot 10^{-5}\,\text{N}/\text{m}^2$$

$$I(\text{dB}) = 10\log_{10}\left[\frac{I}{I_0}\right] = 10\log_{10}\left[\frac{p^2}{p_0{}^2}\right] = 20\log_{10}\left[\frac{p}{p_0}\right]$$

The pressure P here is to be understood as the amplitude of the pressure wave. The power carried by a travelling wave is proportional to the square of the amplitude. The factor of 20 comes from the fact that the logarithm of the square of a quantity is equal to 2 x the logarithm of the quantity.

The actual average threshold of hearing at 1000 Hz is more like 2.5 x 10^{-12} watts/cm^2 or about 4 decibels, but zero decibels is a convenient reference. The threshold of hearing varies with frequency, as illustrated by the measured hearing curves Fig.1). The nominal dynamic range of human hearing is from the standard threshold of hearing to the threshold of pain. A nominal figure for the threshold of pain is 130 decibels, but that which may be considered painful for one may be welcomed as entertainment by others. Generally, younger persons are more tolerant of loud sounds than older persons because their protective mechanisms are more effective.

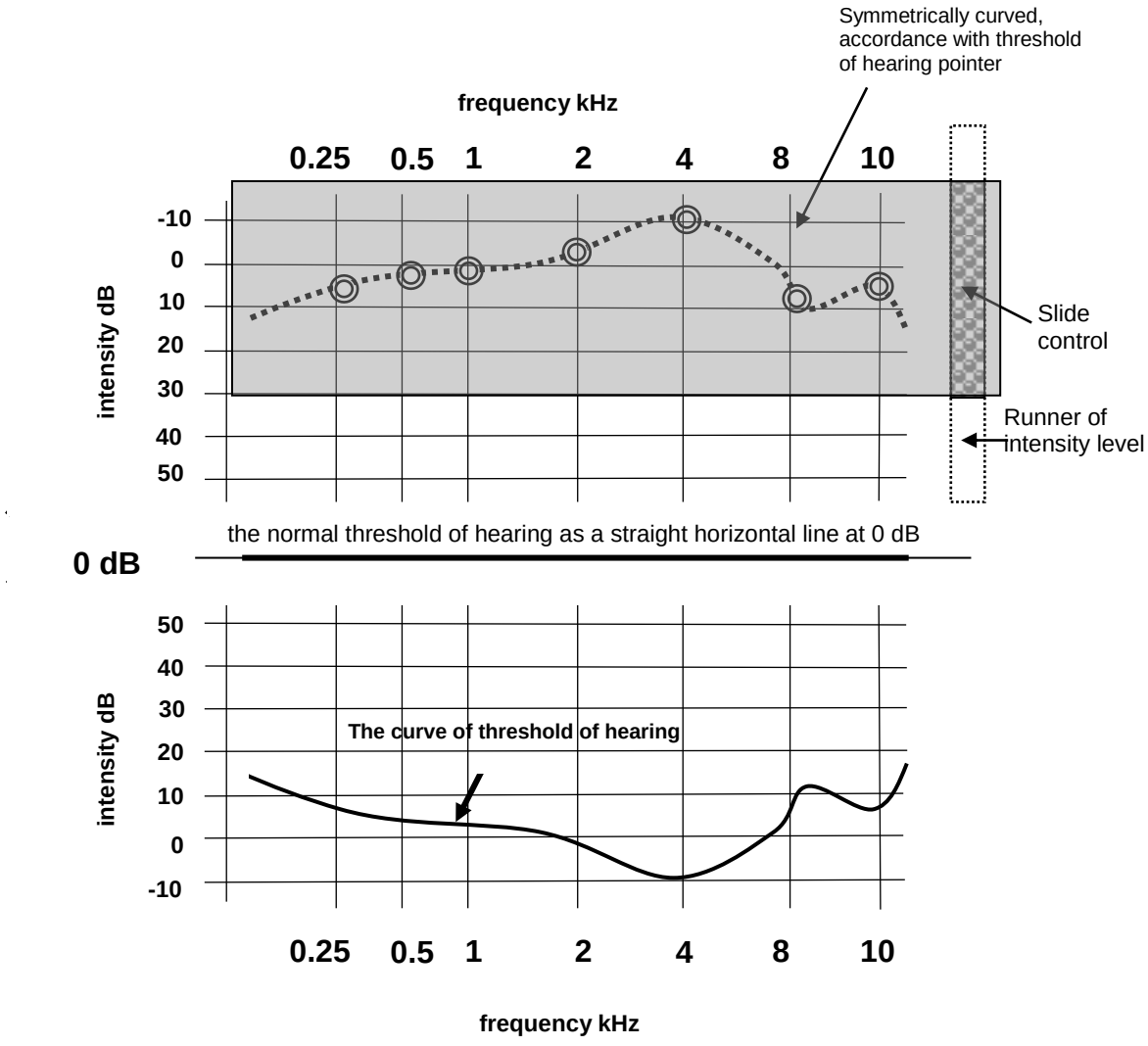


Fig.2 Scheme of the curve of threshold of hearing and its symmetric reflection on an audiometer pointer.

The testing of hearing is most often carried out by establishing the threshold of hearing, the softest sound which can be perceived in a controlled environment. It

is typical to do this testing with pure tones by providing calibrated tones to a person via earphones, allowing that person to increase the level until it can just be heard. Various strategies are used, but pure tone audiometry with tones starting at about 125 Hz and increasing by octaves, half-octaves, or third-octaves to about 8000 Hz is typical. The results of such tests are summarized in audiograms. Audiograms compare hearing to the normal threshold of hearing, which varies with frequency as illustrated by the hearing curves. The audiogram is normalized to the hearing curve so that a straight horizontal line at 0 represents normal hearing (Fig.2). It is possible to have scores less than 0, which indicate better than average hearing.

Tympanometry

The tympanometer measures the compliance (sometimes called "admittance") of the tympanic membrane (eardrum) while different pressures are being applied to the external ear canal. A probe is inserted into the ear canal and emits a sound signal under different pressure conditions (Fig. 3).

The compliance of the tympanic membrane is measured in cubic centimetres,

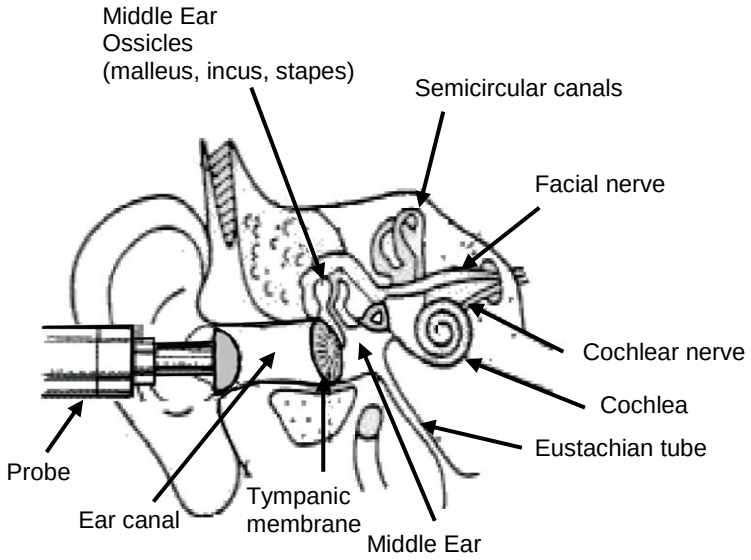


Fig.3 Scheme of tympanometry.

and the pressure in the ear canal is measured in decapascals (daPa). The probe has different sized "plugs" that provide a seal at the entrance to the external ear canal. The tip of the probe has a pressure transducer that changes the pressure in the external ear canal from negative, through atmospheric pressure, to positive

pressure. While the pressure is changing, a sound transmitter sends a sound wave to the tympanic membrane. The wave that is reflected from the tympanic membrane is then picked up by a microphone in the probe. The tympanometer measures the energy of the sound signal reflected by the tympanic membrane at these different pressures.

If the middle ear space is filled with fluid, most of the sound is reflected back to the probe from the stiff tympanic membrane and a flat tympanogram tracing results (low compliance) (Fig.4). If the middle ear space is filled with air, and the

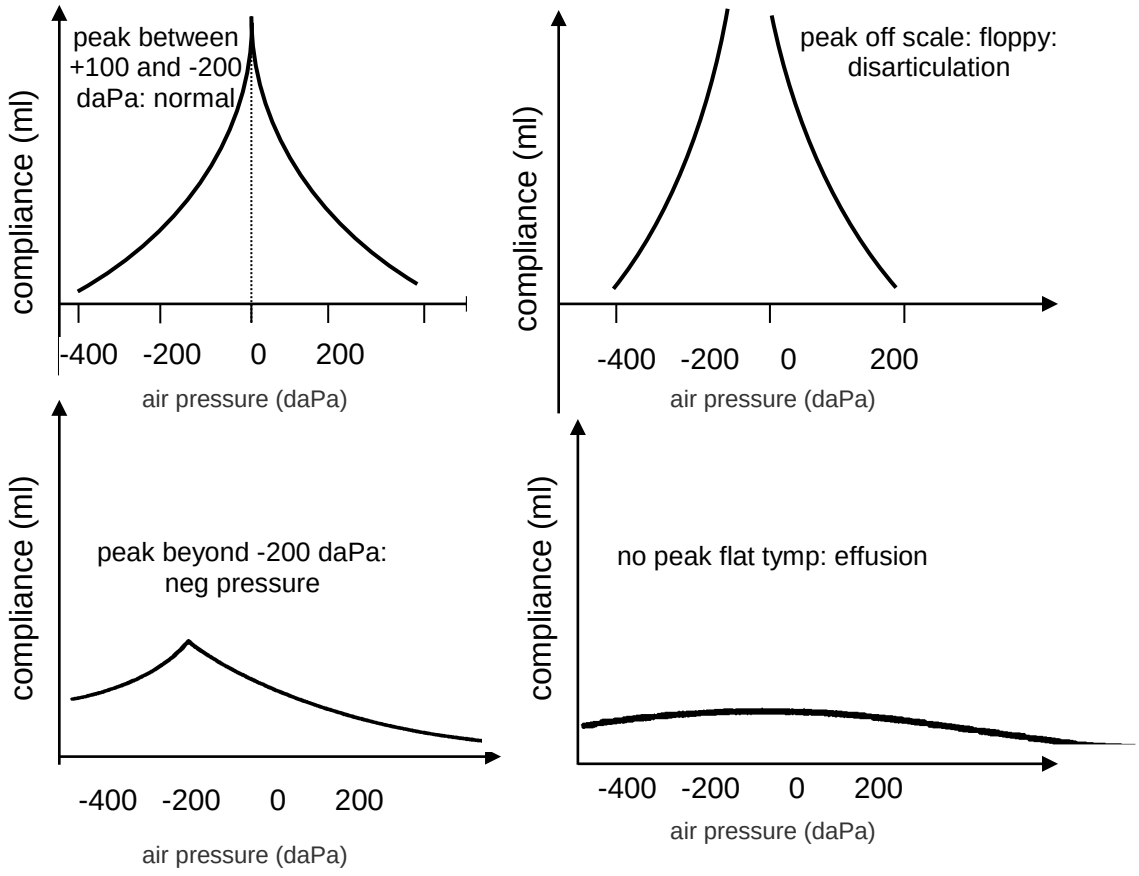


Fig.4 Tympanogram types.

ossicles are intact, energy is absorbed by the tympanic membrane, ossicles, and inner ear structures. The tracing will show a "normal" peak with normal compliance. If there is disruption of the ossicles, or if a portion of the tympanic membrane is flaccid, a large amount of energy will be absorbed into the tympanic

membrane and the tracing will display an abnormal peak (high compliance). Similarly, if there is a perforation of the tympanic membrane, the tympanometer will measure an unusually large canal volume, because the space of the middle ear and mastoid air cells will be included in the volume calculation.

PRACTICAL PART

Answer the questions:

1. What is a decibel?

2. What is a phon?

3. Describe the Weber-Fechner law.

4. What is the function of the outer ear?

5. Functions of the middle ear.

6. What is the function of the inner ear?

EXPERIMENTAL PART

Objective: Audiometry and tympanometry testing.

Materials: an audiometer, headphones

PART A

Procedure: Pure tone audiometry

An audiometer emits sounds or tones, like musical notes, at various frequencies, or pitches, and at differing volumes or levels of loudness. Testing is usually done in a soundproof testing room. The person being tested wears a set of headphones that blocks out other distracting sounds and delivers a test tone to one ear at a time. At the sound of a tone, the patient holds up a hand or finger to indicate that the sound is detected. Each time the person hears a tone, should press a response button. Each ear is tested separately. It is not unusual for levels of sensitivity to sound to differ from one ear to the other.

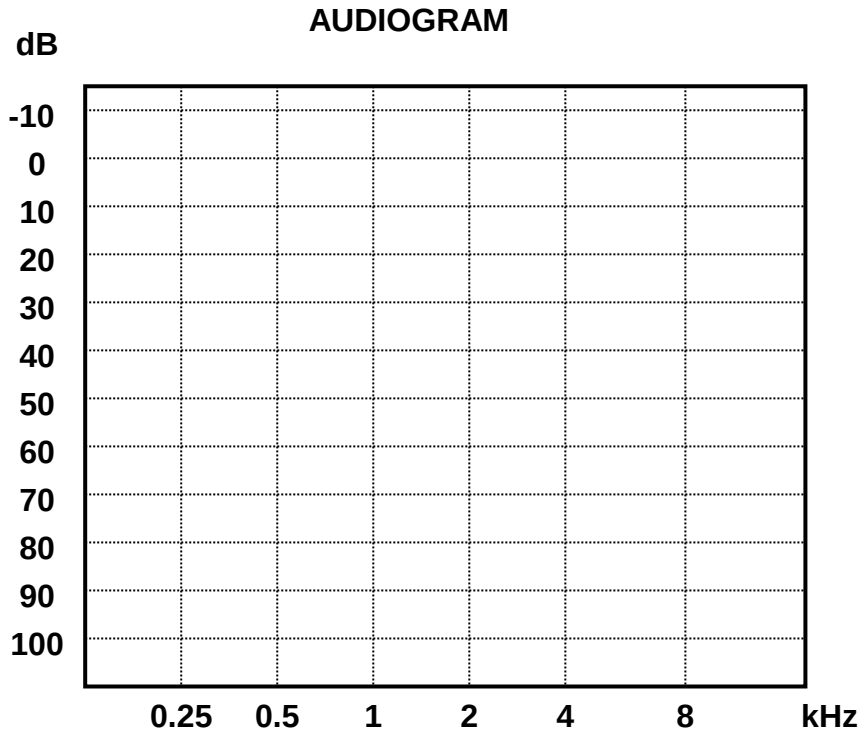
A second type of audiometry testing uses a headband rather than headphones. The headband is worn with small plastic rectangles that fit behind the ears to conduct sound through the bones of the skull. The patient being tested senses the tones that are transmitted as vibrations through the bones to the inner ear. As with the headphones, the tones are repeated at various frequencies and strength.

The results of the audiometry test may be recorded on a grid or graph called an audiogram. Hearing is measured in units called decibels.

Data and observations

The threshold of hearing: a right ear – **x**; a left ear - ●

Obtained points you should connect and receiving the treshold of hearing.



PART B

Procedure: Tympanometry

A device (a probe) is inserted in your ear that will change the pressure in your ear, produce a pure tone, and measure your body's responses to the sound and different pressures.

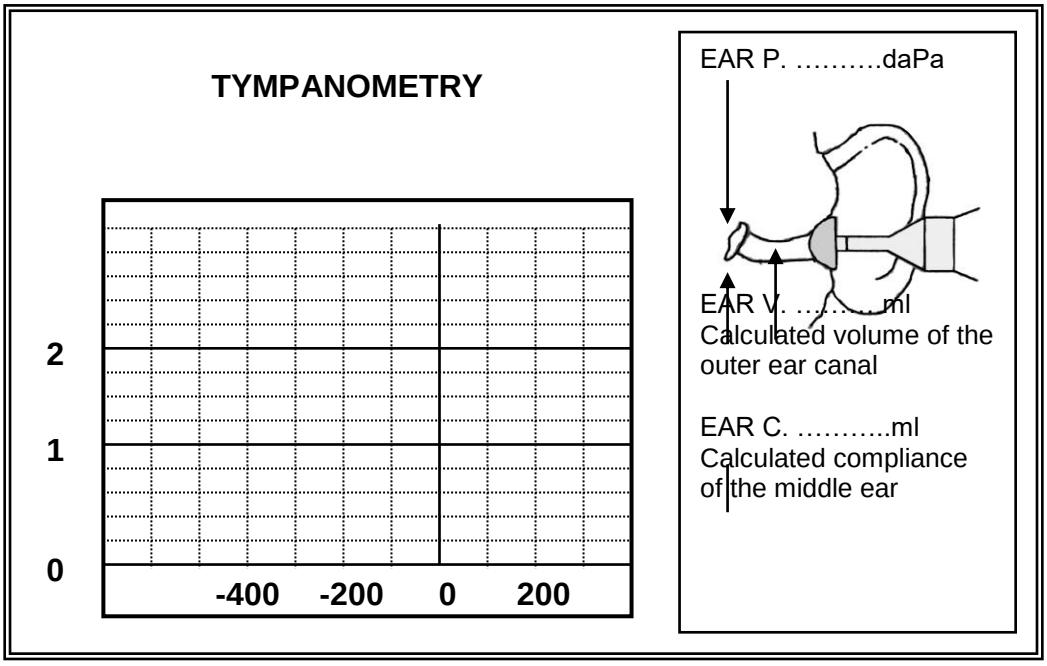
During the test, it is important not to speak, move, swallow, or to be alarmed. All these actions can alter the pressure in the middle ear and invalidate the test results. There may be some discomfort while the probe is in the ear, but no harm will result. You will hear a loud tone as the measurements are taken.

The normal pressure inside the middle ear is 100 daPa (a very small amount). The tympanic membrane should appear smooth and symmetrical.

The results of the tympanometry test may be recorded on a grid or graph called a tympanogram. It is a plot of middle ear compliance as a function of ear canal pressure:

- Pressure is swept from +200 to -200 or -400 daPa
- Should see peak at point where pressures are equal

Data and observations



The date	Student's name and surname	Lab assistant signature

NOTES

2.4 Lab exercise

ELECTROCARDIOGRAPHY

THEORY

The rhythmical action of the heart is controlled by an electrical signal initiated by spontaneous stimulation of special muscle cells located in the right atrium. The natural pacemaker of the heart is called the **Sinoatrial Node** (SA node). It is located in the right atrium. The heart also contains specialized fibers that conduct the electrical impulse from the pacemaker (SA node) to the rest of the heart (Fig. 1). The electrical impulse leaves the SA node and travels to the right and left atria, causing them to contract together. This takes 0.04 seconds. There is now a natural delay to allow the atria to contract and the ventricles to fill up with blood. The electrical impulse has now travelled to the **Atrioventricular Node** (AV node). The electrical impulse now goes to the **Bundle of His**, then it divides into the **Right and Left Bundle Branches** where it rapidly spreads using **Purkinje Fibres** to the muscles of the right and left ventricle, causing them to contract at the same time. All this activity produces electrical waves.

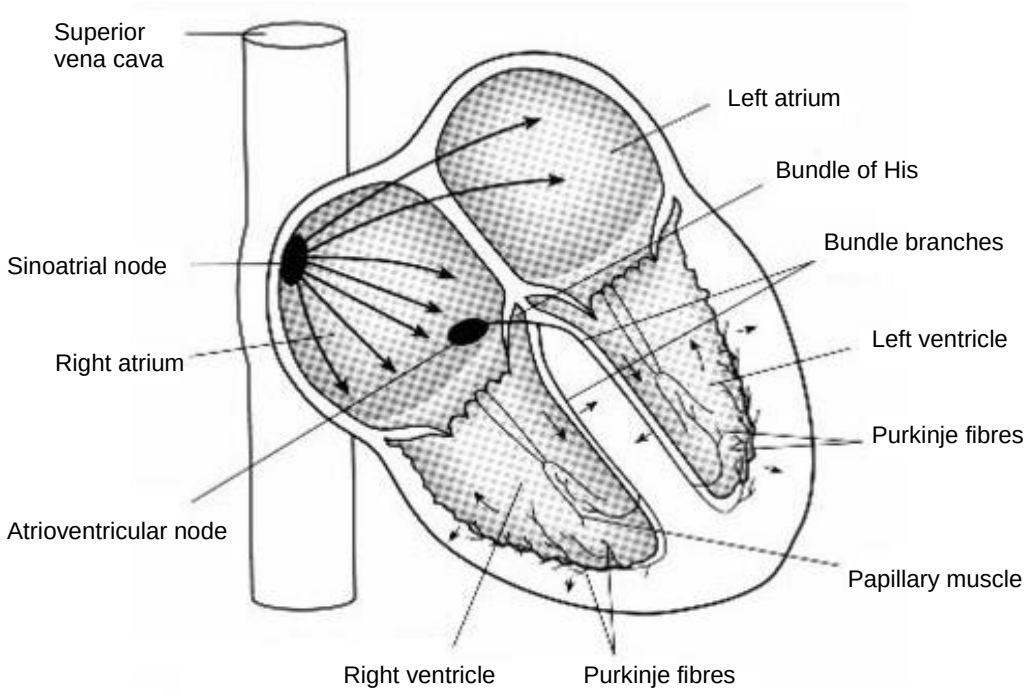


Fig.1 The conduction system of the heart

The electrocardiograph is a device that allows us to record the electrical signals produced by the heart by placing electrodes on the surface of the skin. An **Electrocardiogram** (abbreviated as either **ECG** or **EKG** (from German, Elektrokardiogramm) is a graphic produced by an electrocardiograph, which records the electrical voltage in the heart in the form of a continuous strip graph.

Action Potentials

Certain cells (excitable cells) have ion channels that allow them to quickly change the cell membrane permeability to specific ions. Electrical impulses are the result of the flow of cations (mostly Na^+ and K^+, and some Ca^{2+}) back and forth across the cardiac cell (myocyte) membrane (Fig. 2). A difference in concentration

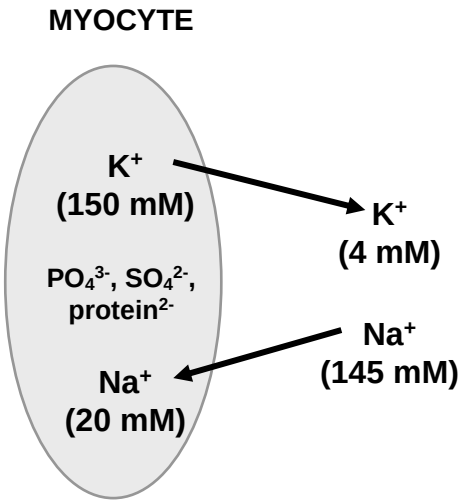


Fig.2 Different concentrations of ions, particularly Na^+, K^+, across the cell membrane in cardiac cells (myocytes).

of these ions across the membrane at any given moment provides an electrical potential (voltage) measured in millivolts. **The resting state** of the cardiac cell consists of: (1) a high concentration of Na^+ outside the cell membrane, (at rest, the cell membrane is impermeable to Na^+); (2) a high concentration of K^+ and anions (PO_4^{3-}, SO_4^{2-}, $protein^{2-}$) is inside the cell, (the cell membrane is always impermeable to these anions); and (3) a negative electrical potential exists across the cell membrane (negative on the inside relative to the outside) and the cell is **polarized**. So, **the resting potential** is a state of disequilibrium where the surrounding extracellular fluid has a positive charge and the myocyte itself has a

negative charge. The resting potential of myocardial cells and conduction cells is –90mV.

In response to the appropriate stimulus, the cell membrane of excitable cells goes through a sequence of depolarisation from its rest state followed by repolarisation to that rest state. Excitation of these cells usually opens Na^+ channels, allowing an influx of Na^+ ions. The cell membrane potential then becomes positive. The cell is **depolarised**. As the Na^+ channels begin to close, K^+ channels also open. K^+ ions then flow out of the cell, and the membrane potential becomes negative again. The cell is **repolarised**. After repolarisation, the membrane potential is usually slightly lower than the resting membrane potential for a short period of time. The cell is said to be **hyperpolarised**. The cell then returns to its normal resting membrane potential. The sequence of events from the depolarisation to the return to the normal membrane potential is the **action potential**.

The cardiac action potential, like all action potentials, is produced through changes in membrane ion permeabilities. Many cells in the body have the ability to undergo a transient depolarisation and repolarisation that is either triggered by external mechanisms (e.g., motor nerve stimulation of skeletal muscle or cell-to-cell depolarisation in the heart) or by intracellular, spontaneous mechanisms (e.g., cardiac pacemaker cells). There are two general types of cardiac action potentials. Non-pacemaker action potentials, also called fast response action potentials because of their rapid depolarisation, are found throughout the heart except for the pacemaker cells. The pacemaker cells generate spontaneous action potentials that are also termed slow response action potentials because of their slower rate of depolarisation. These are found in the SA and AV nodes of the heart.

Non-Pacemaker Action Potentials (Fig.3A)

Ventricular myocytes and Purkinje cells are an example of non-pacemaker cells in the heart. When these cells are rapidly depolarised to a threshold voltage of about -70 mV (e.g., by another conducted action potential), there is a **rapid depolarisation** (**Phase 0**) that is caused by a transient increase in fast Na^+-channel conductance (gNa^+).

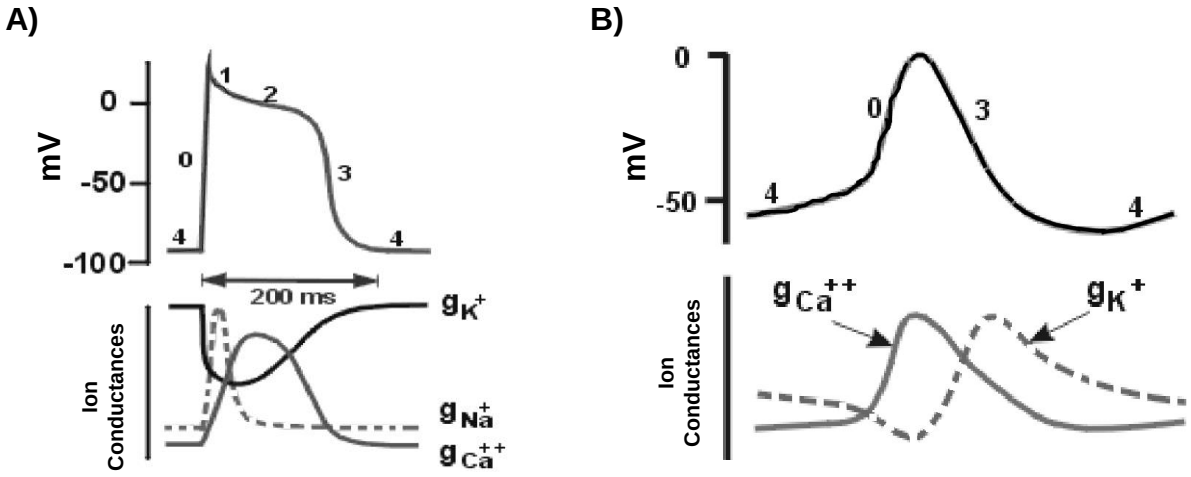


Fig.3 **A)**: Non-pacemaker action potentials (ventricular myocytes and Purkinje cells) and **B)**: sinoatrial nodal action potentials (pacemaker cells);
gCa^{++}, gK^+, gNa^+ - Ca^{2+}, K^+, Na^+ conductance respectively.

At the same time, Na^+-channel conductance (gK^+) falls. These two conductance changes move the membrane potential away from the equilibrium potential for K^+ and closer toward the equilibrium potential for Na^+. **Phase 1** represents **an initial repolarisation** that is caused by the opening of a special type of K^+ channel (not shown in the figure). However, because of the large increase in slow inward gCa^{2+}, the repolarisation is delayed and there is **a plateau phase** in the action potential (**Phase 2**). This inward calcium movement is through long-lasting (L-type) calcium channels that open up when the membrane potential depolarises to about -40 mV. **Repolarisation** (**Phase 3**) occurs when gK^+ increases and gCa^{2+} decreases. Therefore, the action potential in non-pacemaker cells is primarily determined by changes in fast Na^+, slow Ca^{2+} and K^+ conductances. Non-pacemaker cells have a true **resting membrane potential** (**Phase 4**) that remains near the equilibrium potential for K^+.

Sinoatrial Node Action Potentials

Cells within the SA node are the primary pacemaker site within the heart. These cells are characterised as having **no true resting potential, but instead generate regular, spontaneous action potentials**. Unlike most other cells that elicit action potentials, the depolarising current is carried primarily by relatively

slow, inward Ca^{2+} currents instead of by fast Na^{+} currents. There are, in fact, no fast Na^{+} currents operating in SA nodal cells.

SA nodal action potentials are divided into three phases (Fig.2B): **Phase 0 depolarisation** is primarily due to increased gCa^{2+}. Because the movement (or conductance) of Ca^{2+} through their channels is not rapid (hence, the term "slow inward Ca^{2+} channels"), the rate of depolarisation (slope of Phase 0) is much slower than found in other cardiac cells. **Repolarisation** occurs (**Phase 3**) as gK^{+} increases and gCa^{2+} decreases. **Spontaneous depolarisation** (**Phase 4**) is due to a fall in gK^{+} and a small increase in gCa^{2+}. A slow inward Na^{+} current also contributes to **Phase 4**, and is thought to be responsible for what is termed the pacemaker or "funny" current. Once this spontaneous depolarisation reaches a threshold (about -40 mV), a new action potential is triggered.

Leads and wires

Between the patient and the ECG machine is a patient cable, and this is divided into a number of different coloured wires. **A lead** is a view of the electrical activity of the heart from a particular angle across the body, obtained by using different combinations of these wires. To obtain a 12 lead ECG you would have 4 wires attached to each of the limbs, and six wires placed around the chest, 10 wires in total but you get 12 "leads" or pictures.

"Standard limb leads" or the "bipolar limb leads."

Bipolar recordings utilise standard limb lead configurations (Fig.4). By convention,

lead I has the positive electrode on the left arm, and the negative electrode on the right arm. They represent the difference in electrical potential between two specific points in the body. In this and the other two limb leads, an electrode on the right leg serves as a reference electrode for recording purposes. In the **lead II** configuration, the positive electrode is on the left leg and the negative electrode is on the right arm. **Lead III** has the positive electrode on the left leg and the negative electrode on the left arm. These three bipolar limb leads roughly form an equilateral triangle (with the heart at the center) that is called Einthoven's triangle in honour of Willem Einthoven who developed the electrocardiogram in 1901. The

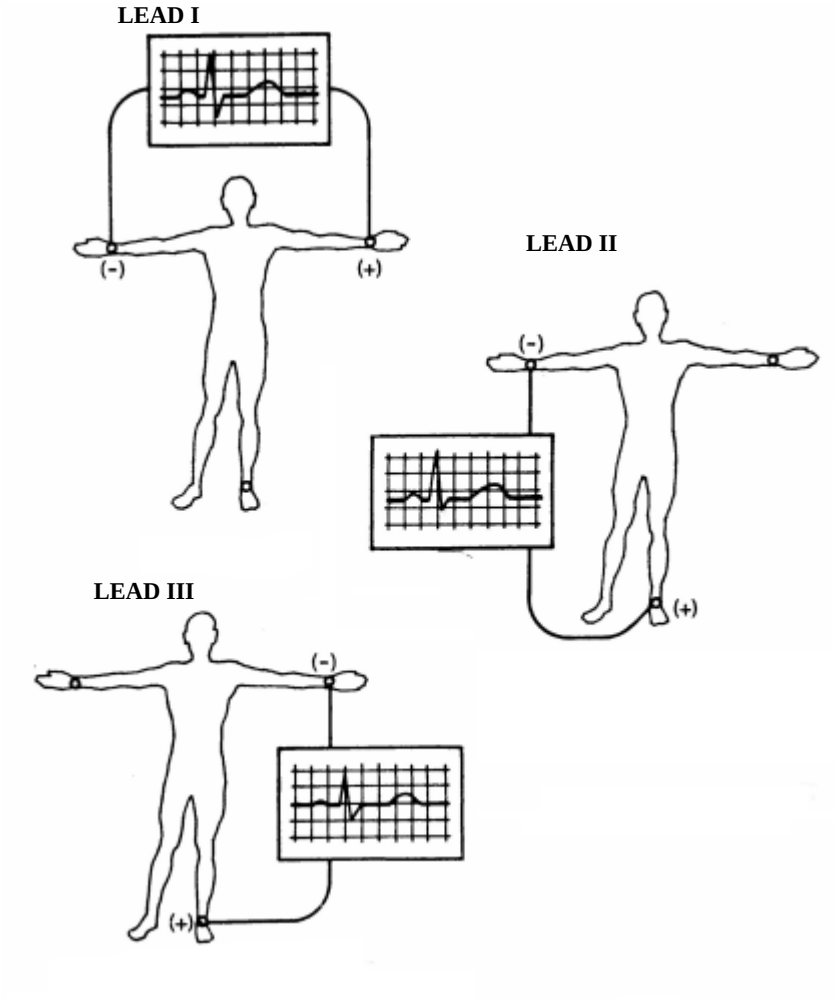


Fig.4 Standard limb leads or the bipolar limb leads measures the potential difference between the two arms.

direction of the lead must be paid attention to. This lead arrangement is called the **standard bipolar limb lead**.

Einthoven's law is stated mathematically as follows:

Lead I + Lead III = Lead II.

Therefore, if any two leads are known at a given time, the third can be determined mathematically.

Whether the limb leads are attached to the end of the limb (wrists and ankles) or at the origin of the limb (shoulder or upper thigh) makes no difference in the recording because the limb can simply be viewed as a long wire conductor originating from a point on the trunk of the body.

Augmented Limb Leads (Unipolar)

In addition to the three bipolar limb leads, there are three augmented unipolar limb leads (Fig. 5). These are termed unipolar leads because there is **a single positive electrode that is referenced against a combination of the other limb electrodes,** which serve as the negative electrode or a zero reference point. The positive electrodes for these augmented leads are located on the left arm (**aV$_L$**), the right arm (**aV$_R$**), and the left leg (**aV$_F$**). In practice, these are the same electrodes used for leads I, II and III. (The ECG machine does the actual switching and rearranging of the electrode designations).

The three augmented unipolar leads, coupled with the three bipolar leads, constitute **the six limb leads** of the ECG. These leads record electrical activity along a single plane, termed the frontal plane relative to the heart.

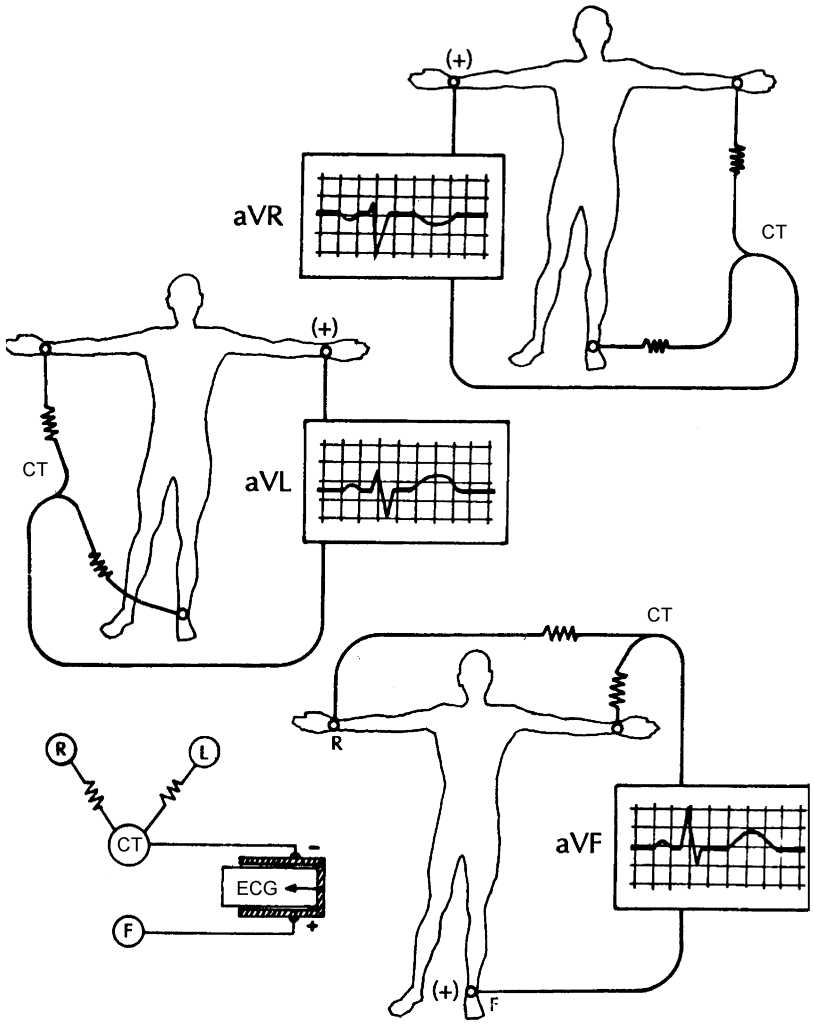


Fig.5 Augmented unipolar limb leads - aV$_R$, aV$_L$, aV$_F$.
CT – the Goldberger central terminal; the zigzag lines – 5 Ω resistances.

Chest Leads (Unipolar)

The last ECG leads to consider are the precordial, unipolar chest leads. These are six positive electrodes placed on the surface of the chest over the heart in order to record electrical activity in a plane perpendicular to the frontal plane (Fig.6). These six leads are named **V_1 - V_6**. The rules of interpretation are the same as for the limb leads. For example, a wave of depolarisation travelling toward a particular electrode on the chest surface will elicit a positive deflection.

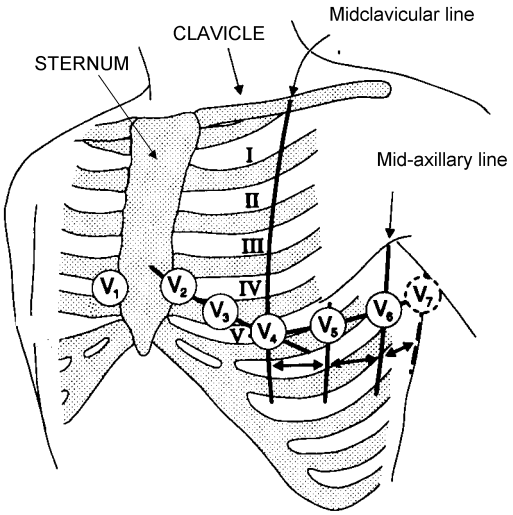


Fig. 6 Placement of "precordial" leads.

In summary, the **"12 lead ECG"** provides different views of the same electrical activity within the heart. It utilises the 3 standard limb leads (bipolar), with the 3 augmented limb leads (unipolar) and the 6 precordial leads (unipolar).

Placement of the limb electrodes

According to the American Heart Association, "The electrodes may be placed on any part of the arms or of the left leg as long as they are below the shoulders in the former and below the inguinal fold anterioly and the gluteal fold posterioly in the latter. Any other placement necessary by deformed or missing extremities must be noted on the record."

Limb electrodes

RA = red - Right arm

LA =yellow - Left arm - wrist

LL = green - Left leg – ankle

RL = black - Right leg (ankle) - as the ground or indifferent lead

Placement of "precordial" or "unipolar" leads (Fig. 6)**:**

- **V_1** = red - 4th intercostal space, just to the right of the sternum
- **V_2** = yellow - 4th intercostal space, just to the left of the sternum
- **V_3** = green - half-way between V_2 and V_4
- **V_4** = brown - left midclavicular line in the 5th intercostal space (ie. about even with the middle of the clavicle and between the 5th and 6th ribs on the left side)
- **V_5** = black - left anterior axillary line in the 5th intercostal space (ie. about even with the front of the armpit and between the 5th and 6th ribs on the left side)
- **V_6** = violet - left mid-axillary line in the 5th intercostal space (ie. about even with the middle of the armpit and between the 5th and 6th ribs on the left side):

The Electrocardiogram (ECG)

Each beat of the heart can be observed as a series of deflections away from the baseline on the ECG. These deflections reflect the evolution of electrical activity in the heart with time which initiates muscle contraction. A typical ECG tracing of a normal heartbeat consists of a P-wave, a QRS complex and a T-wave and sometimes a U-wave (Fig.7).

Waves in the ECG

One part of a typical ECG is a ‘flat line’ or trace indicating no detectable electrical activity. This line is called the **isoelectric line**. The first deviation from the isoelectric line in a typical ECG is an upward pulse following by a return to the isoelectric line. This is called the **P-wave** and it lasts about 0.04 seconds. This wave is caused by the depolarisation of the atria and is associated with the contraction of the atria.

After a return to the isoelectric line there is a short delay while the heart’s AV node depolarises and sends a signal along the AV bundle of conducting fibres

(a bundle of His) to the Purkinje fibres, which bring depolarisation to all parts of the ventricles almost simultaneously.

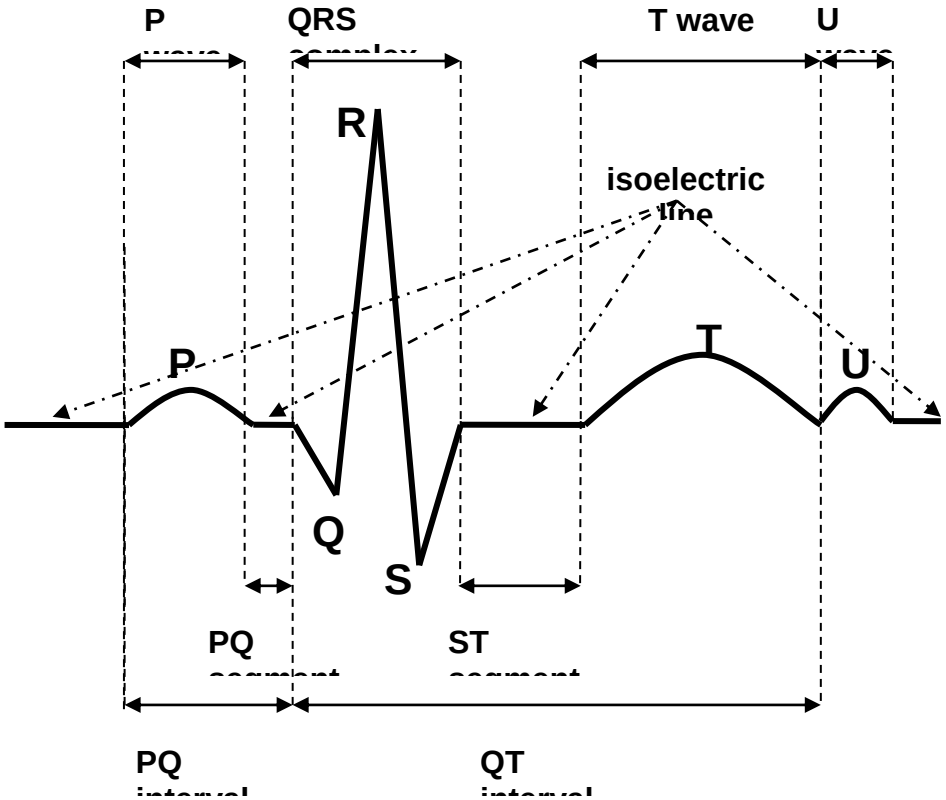


Fig. 7 Normal ECG (waves, segments and intervals in the ECG).

After the AV node depolarises there is a downward pulse called the **Q-wave**. Shortly after the Q-wave there is a rapid upswing of the line called the **R-wave** followed by a strong downswing of the line called the **S-wave** and then a return to the isoelectric line. These three waves together are called the **QRS complex**. This complex is caused by the depolarisation of the ventricles and is associated with the contraction of the ventricles. Every QRS complex may not always have a Q, R, and an S, but it is still labelled as such. Large deflections are denoted by capital letters; smaller deflections (do not exceed 3 mm) by lower case letters. A second positive deflection is given a prime notation (R' or r'). A third positive deflection is designated R" or r". If there is no positive deflection (only a negative deflection), the complex is called QS.

After a short period of time the chemical ions that have been involved in the contraction migrate back to their original locations. The movement of these ions generates an upward wave that then returns to the isoelectric line. This upward pulse is called **the T-wave** and indicates repolarisation of the ventricles. The **U-**

Wave follows the T-wave and represents a small "after-polarisation." Think of the U-wave as a small aftershock (little depolarisations) following an earthquake; the heart tissue, notably the ventricles, is settling down.

The sequence from P-wave to T-wave (or U-wave) represents one heart cycle. The number of such cycles in a minute is called **the heart rate** and is typically 70-80 cycles (beats) per minute (bpm) at rest.

Segments and intervals in the ECG

The period of time from the onset of the P wave to the beginning of the QRS complex is termed the **PQ-interval** or the **PR-interval**, which normally ranges from 0.12 to 0.20 seconds in duration. This interval represents the time between the onset of atrial depolarisation and the onset of ventricular depolarisation. An isoelectric interval from the end of P-wave to the beginning of QRS complex that precedes the depolarisation of the ventricle is called the **PR-segment**. The **ST-segment** is between the end of S-wave and the beginning of T-wave. This period is the time at which the entire ventricle is depolarised and roughly corresponds to the plateau phase of the ventricular action potential. The **QT-interval** is measured from the beginning of the QRS complex to the end of the T-wave. The QT-interval as well as the corrected QT-interval (QT_c) are important in the diagnosis of long QT syndrome and short QT syndrome. The most commonly used method for correcting the QT interval for rate is the one formulated by Bazett. Bazett's formula is

$$QT_c = \frac{QT(s)}{\sqrt{RR(s)}}$$

where: QT_c - the QT-interval corrected for rate

QT - the QT-interval measured in seconds

RR - the interval from the onset of one QRS complex to the onset of the next QRS complex, measured in seconds.

The **QT-interval** represents the time for both ventricular depolarisation and repolarisation to occur and therefore roughly estimates the duration of an average ventricular action potential. This interval can range from 0.35 to 0.43 seconds depending upon heart rate.

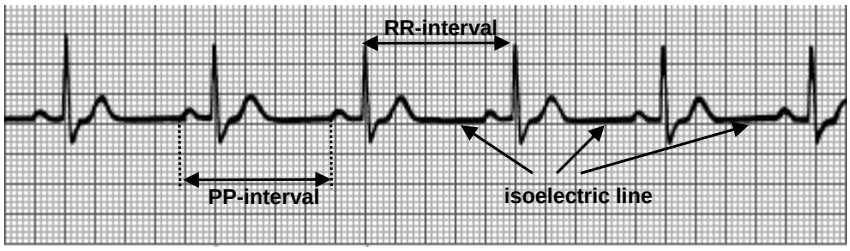


Fig. 8 Normal ECG. Here, there is one QRS-complex per second, so the heart rate is 60 beats/min.

The **RR-interval** represents the duration of ventricular cardiac cycle (an indicator of ventricular rate) and the **PP-interval** – the duration of atrial cycle (an indicator or atrial rate) (Fig. 8).

Reading the ECG

Paper speed, time

ECG paper is divided into small 1 mm, and large 5 mm squares (Fig.8 and 9).

1 large square = 5 small squares

When ECG paper runs at 25 mm per sec

- 1 small square = 1/25 = 0.04 sec = 40 ms
- 1 large square = 1/5 = 0.2 sec = 200 ms
- 5 large squares = 1 sec
- 300 large squares = 1 min

When ECG paper runs at 50 mm per sec

- 1 small square = 1/50 = 0.02 sec = 20 ms
- 1 large square = 1/10 = 0.1 sec = 100 ms
- 10 large squares = 1 sec
- 600 large squares = 1 min

Thus, precise measurements can be made of ECG intervals and the HR.

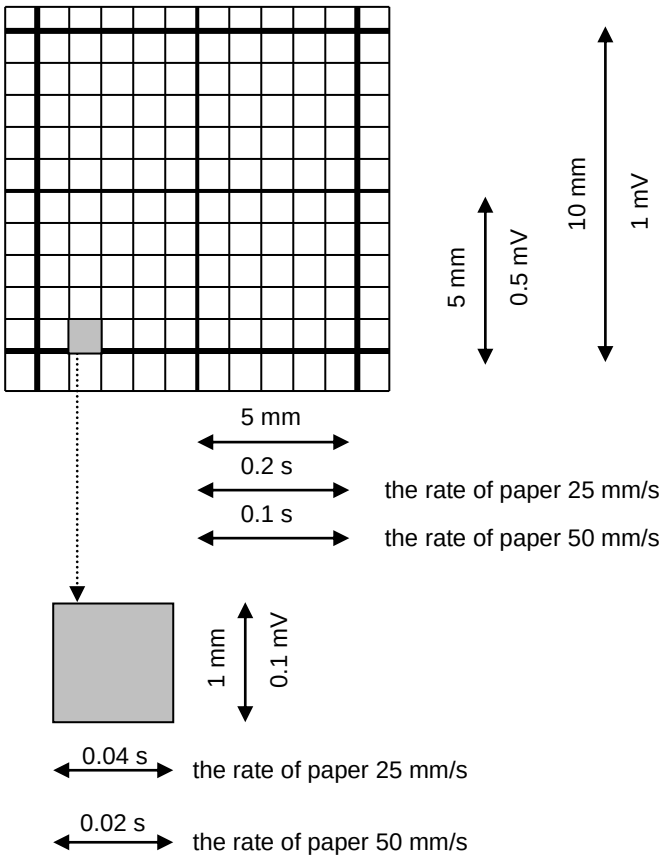
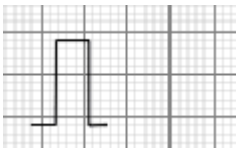


Fig. 9 Enlarged fragment of ECG paper.

The voltage recorded (Fig. 10) from the leads is also standardised on the paper where **1 mm = 0.1 mV** (or between each individual block vertically). This results in:

- 5 mm = 0.5 mV (or between 2 dark horizontal lines)
- 10 mm = 1.0 mv (this is how it is usually marked on the ECG's)



10 mm = 1 mV
1 mm = 0.1 mV

Fig. 10 The voltage recorded on the ECG paper.

The ways to interpret the heart rate (HR)

A. The basic way to interpret the rate is quite simple. You take the duration between two identical points of consecutive ECG waveforms such as the RR duration (Fig.7). Take this duration and divide it into 60. The resulting equation would be:

Heart Rate = 60 : (RR-interval in seconds)

For example: the RR-interval duration is 0.75 s, and then the heart rate is:
60 : 0.75 = 6000 : 75 = 80 bpm.

B. ***The Rule of 1500 (or 3000):*** You take the length of the RR-interval or the PP-interval in millimetres (Fig.7) and use the equation:

When the rate of paper is 25 mm/s – **1500 : (RR-interval in mm)**
When the rate of paper is 50 mm/s – **3000 : (RR-interval in mm)**

For example: the RR-interval is 20 mm, so for the rate of paper 25 mm/s, the heart rate is 1500 : 20 = 75 bpm.

C. An easy and quick way to estimate the heart rate: As seen in the figure 11 below, **when the rate of paper is 25 mm/s** and QRS complexes are 1 large square apart the rate is 300 bpm. 2 large squares apart...150 bpm, etc. So if you memorize these simple numbers you can estimate the heart rate at a glance!

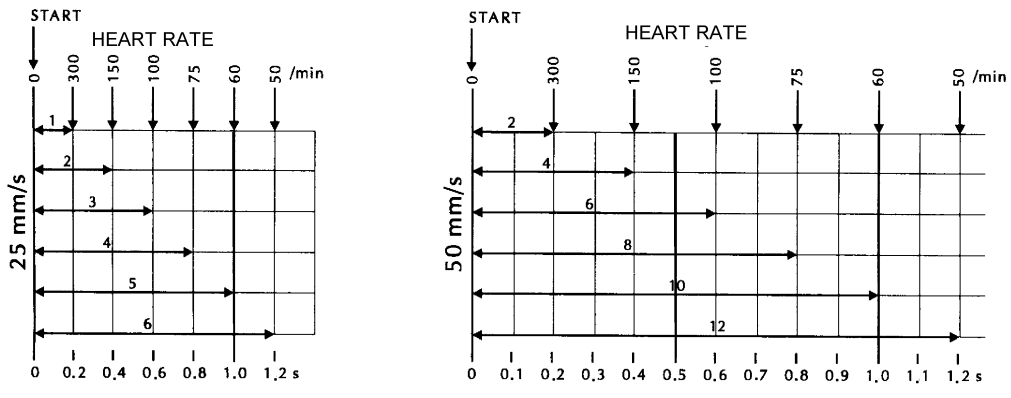


Fig.11 Relationship between duration of RR-interval and heart rate.

Locate the QRS *(the big spike)* complex that is closest to a dark vertical line. Then count either forward or backwards to the next QRS complex. For each dark vertical line you pass, select the next number off the mnemonic "300-150-100-75-60-50" to estimate the rate in **bpm**.

For 50 mm/s you have to double the count number of large squares between consecutive beats.

Remember that this is merely an estimate. You should use real measurements to determine the exact HR

D. ***The Rule of 300 (or 600):*** Count number of large squares between consecutive beats (RR-interval) (Tab. 1)

- When the rate of paper is 25 mm/s

  Divide 300 by number of large squares in RR- interval

- When the rate of paper is 50 mm/s

  Divide 600 by number of large squares in RR- interval

Tab.1 Relationship between the number of large squares covered by the RR-interval and the heart rate.

the rate of paper is 25 mm/s		the rate of paper is 50 mm/s	
number of large squares (5mm) of RR-interval	HR in bpm	number of large squares (5mm) of RR-interval	HR in bpm
	300 : 1 = 300	2	600 : 2 = 300
2	300 : 2 = 150	3	600 : 3 = 200
3	300 : 3 = 100	4	600 : 4 = 150
4	300 : 4 = 75	5	600 : 5 = 120
5	300 : 5 = 60	6	600 : 6 = 100
6	300 : 6 = 50	17	600 : 7 = 86
7	300 : 7 = 43	8	600 : 8 = 75
8	300 : 8 = 38	9	600 : 9 = 67
9	300 : 9 = 33	10	600 :10 = 60
10	300 :10 = 30	11	600 :11 = 55
		12	600 :12 = 50
		13	600 :13 = 46
		14	600 :14 = 43
		15	600 :15 = 40

The Mean Electrical Axis

The electrical activity of the heart at any instant in time can be represented by a vector. The **mean electrical axis** of the heart is the summation of all the vectors occurring in a cardiac cycle. Since the QRS interval caused by ventricular depolarisation, represents the majority of the electrical activity of the heart, we can approximate the mean electrical axis by looking only in this interval.

Because the body is mostly a saline solution (Fig. 12A) that conducts electricity extremely well, Einthoven found he could pretend that the leads were at the shoulders and the navel rather than the ankle and wrists. Thus, an equilateral triangle (Einthoven's Triangle) is an adequate representation of the spatial orientation of the leads.

For our purposes, we will replace Einthoven's Triangle with a set of axes at 60° angles with each other (Fig 12B). This triaxial system is shown in the next figure. This triaxial diagram is easier to use than Einthoven's Triangle, but it maintains the same polarity and orientation of the leads with respect to each other.

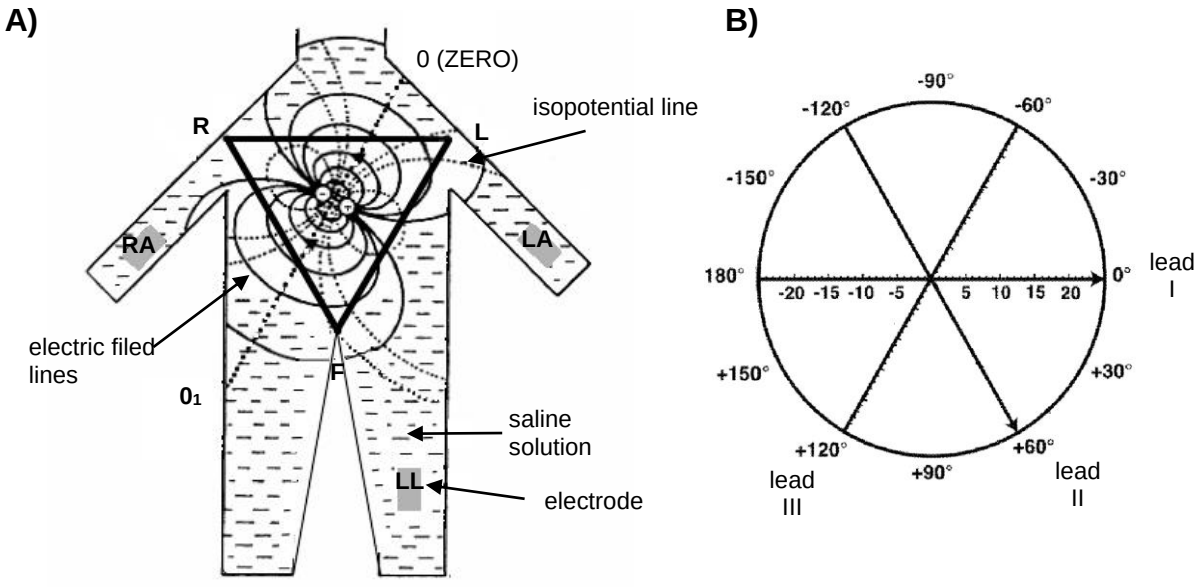


Fig. 12 **A)**: The heart model (according to Massie) with the Einthoven's Triangle. Linia $0\text{-}0_1$ – the line of zero potential, RA – Right Arm, LA – Left Arm, LL – Left Leg. **B)**: This triaxial diagram with a set of axes at 60° angles with each other.

The **normal electrical axis** of the QRS complex is from -30 degrees to +110 degrees. Any value of the QRS mean electrical axis more negative than -30 degrees is considered a pathologic **left axis deviation** (LAD). Any value of the

QRS mean electrical axis more positive than 110 degrees is considered a pathologic **right axis deviation** (RAD). A QRS mean electrical axis between -90 degrees and 180 degrees is **no man's land** and not usually seen clinically.

PRACTICAL PART

Explain the problems:

1. Define:

- Resting potential

- Depolarisation

- Repolarisation

- Hyperpolarisation

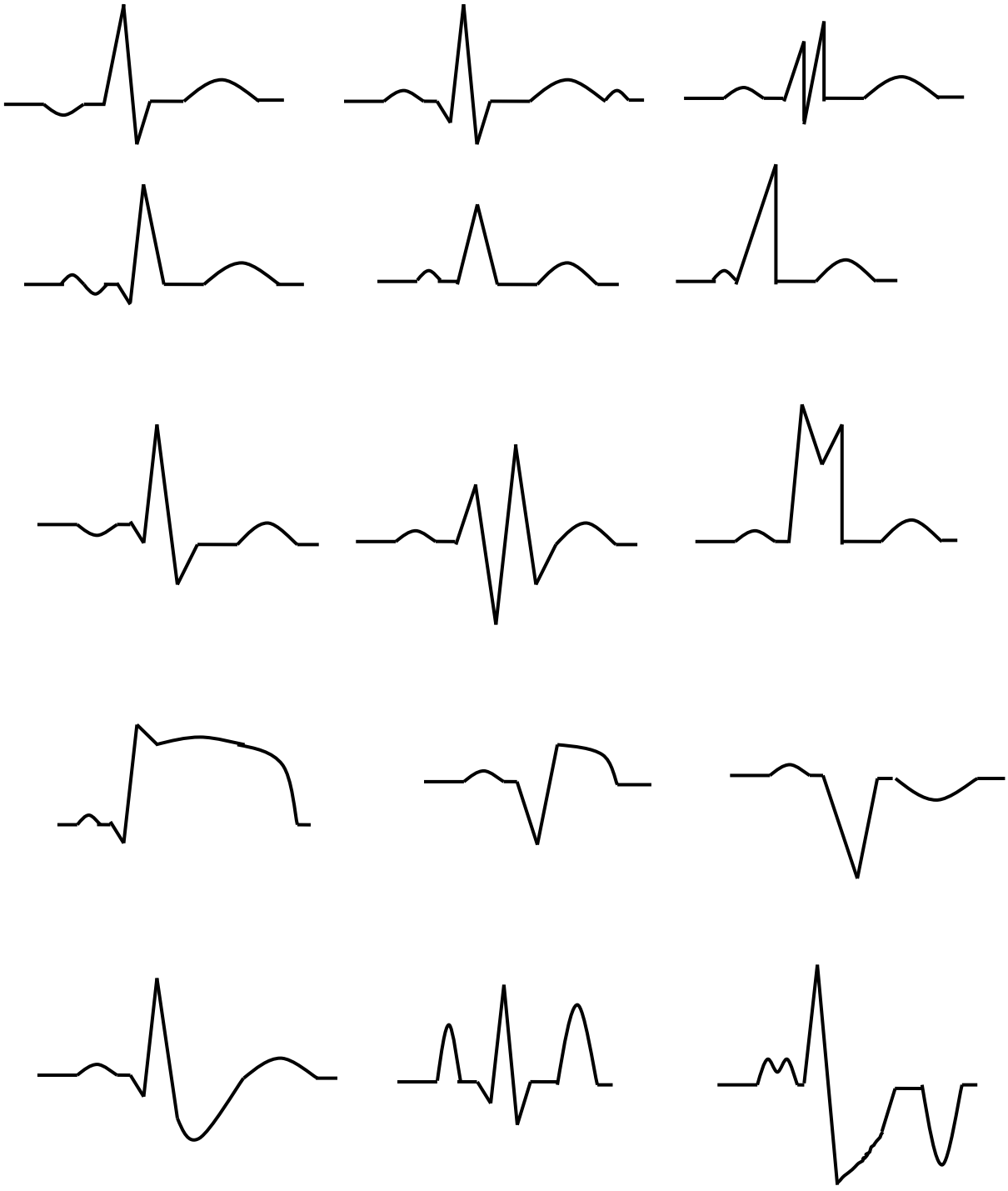
- Action potential

2. Summarise the action potential in non-pacemaker cells (phases, ion channels, channel conductance).

2. Summarise the action potential in pacemaker cells (phases, ion channels, channel conductance).

3. Define the bipolar and unipolar limbs.

4. Name the waves in the ECGs below



EXPERIMENTAL PART

Part A

Objective*:* Observe and record an ECG and identify its characteristics (waves, rhythm, heart rate, etc).

Materials: an ECG device, electrodes and an oscilloscope.

Procedure of making an ECG recording:

1. The patient must lie down and relax (to prevent muscle tremor).
2. Proper **electrode placement** is essential in order to acquire accurate EKG electrodes. The following are some general guidelines for skin preparation:
    - Shave hair away from electrode placement site (not necessarily at the lab).
    - Rub site briskly with alcohol pad.
    - Rub site with 2cm x 2cm gauze or swab.
    - Place electrode. Be sure that the electrode has adequate gel and is not dry.

2. Connect up the limb electrodes, making certain that they are applied to the correct limb.
3. Calibrate the record.
4. Record the 12 standard leads – three or four complexes are sufficient for each

Stick the ECG recording in here

Data and observations

1. Calculate the heart beat using all known methods (you have to be able explain methods used).

2. According to ECG, calculate:

a) The duration of: PP-interval (s)……..…and RR-interval (s)……
Identify the basic rhythm:
Regular rhythm ☐
Sinus Rhythm: ☐, other ☐

b) The duration of (place results in the table)
- P wave norm: 0.04 – 0.12 s In the II lead
- PQ-segment norm: 0.04 – 0.10 s
- PQ-interval norm: 0.12 – 0.20 s
- QRS complex norm: 0.06 – 0.10 s
- QT_c-interval norm: 0.35 – 0.43 s.

counting QT_c-interval.

Duration of QT in seconds: ……. Duration of RR-interval in seconds:……

Write down Bazett's formula:

Count the duration of QT_c.

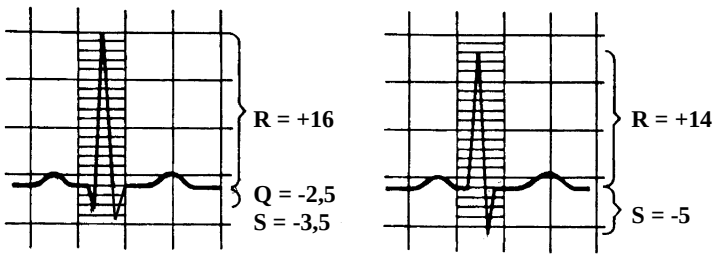
	Length in mm (rate of paper 25 mm/s)	duration (s)
P wave		
PQ-segment		
PQ-interval		
QRS complex		
QT_c-interval		

3. Calculate the mean electrical axis of the QRS complex.

Procedure

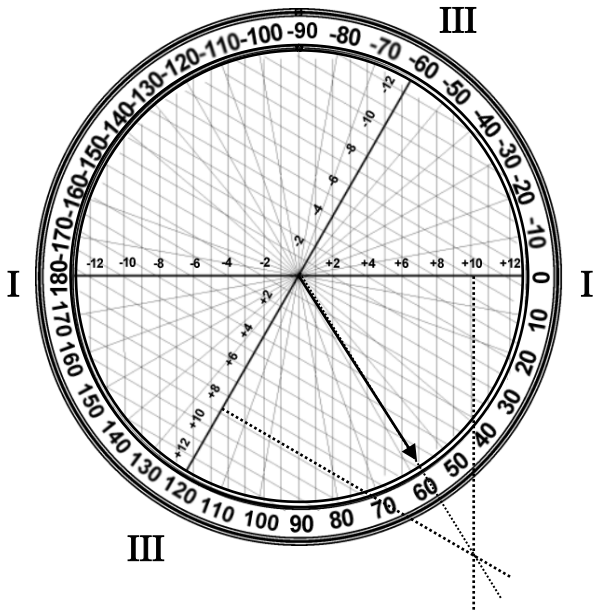
Geometric method for calculating mean electrical axis (according to Scheidta)

1. To calculate the mean electrical axis of the QRS complex in this method, standard leads I and III are used. The vectorial sum of the deflections of the QRS complex for each lead is calculated in millimetres. **Remember**, to get the height of the QRS complex, we measure the height of R above the isoelectric line and subtract the depth of Q and S below the isoelectric line.
    a. The sum of QRS in mm in lead I: Q + R + S = (-2,5 mm) + (+16 mm) + (-3,5 mm) = +10 mm
    b. The sum of QRS in mm in lead III: Q + R + S = 0 mm + (+14 mm) + (-2,5 mm) = +9 mm



2. Perpendiculars are drawn at plotted points on respective vectorial reference lines.
    a. The point corresponding to this sum is then located on lead I and a perpendicular is dropped from **lead I.**
    b. The same is done for **lead III.**

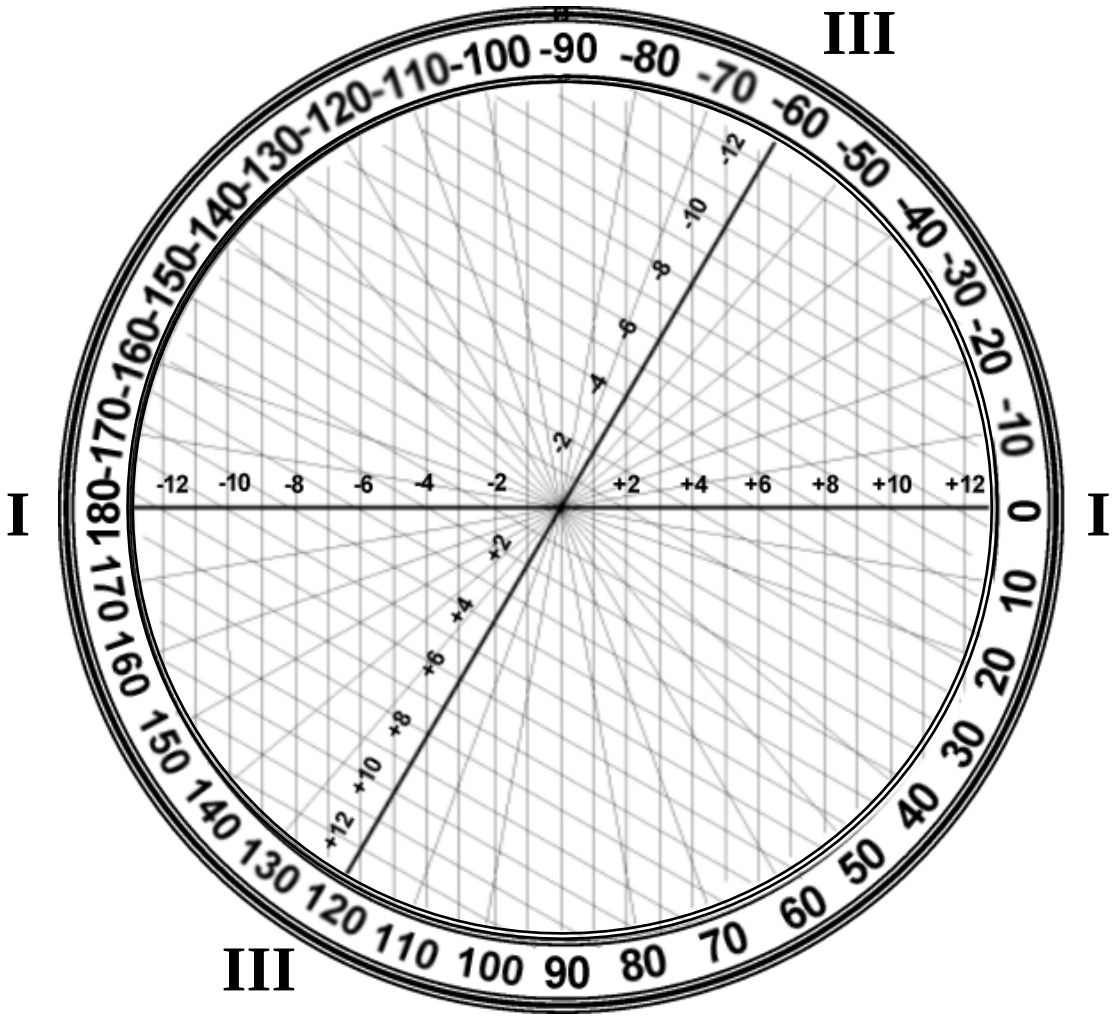
Let's say that we found the height of the QRS complex in a lead I ECG to be 10 mm; we would draw a perpendicular line passing through +10 on the lead I side (perpendicular to lead I) of the triangle like this! Now, let's say that we calculated the height of the QRS complex as 9 mm; we draw a second line passing through +9 on the lead III side (perpendicular to lead III) of the triangle like this!



3. A line is then drawn from the centre of the grid through the point of intersection of the two perpendicular lines to obtain the mean electrical axis. The vector just drawn summarises the net direction of electrical activity in the examining heart. In this case, the mean electrical axis of the QRS complex is **60 degrees** which is within the normal range

Data and observations

Lead I:	Lead III
Q:	Q:
R:	R:
S:	S:
The sum of QRS in mm =	The sum of QRS in mm =



The α angle is: …….

The assessment of mean electrical axis (underline the proper one):
normal electrical axis; a pathologic left axis deviation (LAD); a pathologic *right axis deviation* (RAD); *no man's land*

The date	Student's name and surname	Lab assistant signature

Part B

Objective*:* The purpose of this laboratory activity is to measure a person's electrocardiogram (ECG) when the person is at rest, and compare it to the ECG of the same person after mild exercise and how it affects the amount of carbon dioxide in person's breath.

Materials: ScienceWorkshop™ Interface, EKG Sensor (CI-6539), EKG electrodes (included with sensor), pH Sensor, beakers, 250 ml, wash bottle, buffer solutions, high pH and low pH, distilled water, straw (for blowing into the water)

Procedure:

In this activity, the EKG sensor will measure the electrical current associated with the heart's contractions. The *ScienceWorkshop* program records and displays the electrocardiogram (heart voltage signals). The *ScienceWorkshop* program automatically calculates heart rate based on the peaks and valleys in the EKG trace.

The EKG during rest will be compared to the EKG after mild exercise.

The pH Sensor measures the change in the pH of water that the same person will blow through before and after mild exercise. The carbon dioxide gas in the person's breath will dissolve in the water and turn it into a weak acid.

Computer setup, sensor calibration and equipment setup are detaily described in the "*ScienceWorkshop* ECG Manual".

Data Table: Interval Analysis

Item	**Time** (resting ECG)	**Time (**EKG After Mild Exercise**)**
P wave	sec	sec
PQ-interval	sec	sec
PQ-segment	sec	sec
QRS complex	sec	sec

QT-interval	sec	sec

Data Table: Record the mean heart rate at rest and after mild exercise. Also record the Min (minimum) value of pH at rest and after mild exercise.

Trial	Mean heart rate (bpm)	Minimum pH
At rest		
Post-exercise		

Answer Questions

1. Compare your values for the PQ, QRS, and QT intervals for the EKG at rest to the values for the PQ, QRS, and QT intervals for the EKG after mild exercise. How do the time intervals for the EKG after mild exercise compare to the time intervals for the EKG at rest? What could explain the differences, if any?

3. What is the influence of exercise on the amount of carbon dioxide gas in your exhaled breath?

4. How do you explain the pH value in the water before exercise to the pH value in the water after exercise?

The date	Student's name and surname	Lab assistant signature

NOTES

2.5 Lab exercise

THE MEASURMENT OF THE BLOOD FLOW VELOCITY

THEORY

Sound waves which are within the range of normal hearing are of a frequency of about 20 to 20,000 Hertz (Hz). Sound with a frequency over 20 kHz is called ultrasound. Human hearing can't go beyond about 18,000 – 20,000 vibrations per second, or 18 - 20 kHz (too fast producing a sound to high to hear). Sounds in the range 20 -100 kHz are commonly used for communication and navigation by bats, dolphins, and some other species. Much higher frequencies, in the range 1-20 MHz, are used for medical ultrasound. Such sounds are produced by ultrasonic transducers, which operate either by the inverse piezoelectric effect or the magnetostrictive effect.

Inverse piezoelectric effect

The word piezo is Greek for "push". The effect known as piezoelectricity was discovered by brothers Pierre and Jacques Curie in 1880. Piezoelectricity literally means "pressure electricity" and may be defined as the electric polarisation produced by a mechanical strain in crystals (that belong to certain classes of crystals). The polarization produced is proportional to the strain and changes sign with it. This is called the **direct piezoelectric effect**.

Closely related to it is the **inverse effect** (sometimes called the **reciprocal** or **converse effect**), whereby a piezoelectric crystal becomes strained, when electrically polarised, by an amount proportional to the polarising field. Both effects are manifestations of the same fundamental property of the crystal and they are both reversible.

Crystals which acquire a charge when compressed, twisted or distorted are said to be piezoelectric. Quartz (SiO_2) demonstrates this property and is extremely stable. Quartz crystals are used for watch crystals and for precise frequency reference crystals for radio transmitters. Rochelle salt (potassium sodium tartrate; KNa; $(C_4H_4O_6) \cdot 4H_2O$) produces a comparatively large voltage upon compression

and was used in early crystal microphones. Barium titanate ($BaTiO_3$), lead zirconate ($PbZrO_3$), and lead titanate ($PbTiO_3$) are ceramic materials which exhibit piezoelectricity and are used in ultrasonic transducers. The standard piezoelectric material for medical imaging processes has been lead zirconate titanate (PZT). Ultrasonic medical imaging typically uses ultrasound frequencies in the range 1-20 MHz.

Magnetostriction

Magnetostriction it is one of the magnetic properties which accompanies ferromagnetism where ferromagnetic material respond mechanically to magnetic fields. For applied magnetic fields in AC electrical devices such as transformers, the maximum length change happens twice per cycle, producing the hum. Magnetostriction is used to produce high intensity ultrasonic vibrations in the 20-40 kHz range either as a sound source or as ultrasonic waves in liquids which can act as a cleaning mechanism in ultrasonic cleaning devices.

Ultrasound is widely used in diagnostics, because of the fact that ultrasound propagation is strongly dependent on the properties of the propagation medium. The ability of the ultrasonic wave to travel through any medium is restricted by the properties of that medium, such as acoustic impedance, the speed of sound in certain tissues.

Acoustic impedance (Z) is a function of the elasticity and density of a particular tissue. The acoustical impedance of the medium is determined by the equation

$$\mathrm{Z} = \sqrt{\rho \cdot \mathrm{B}} = \mathrm{v} \cdot \rho$$

where: Z - the acoustical impedance,

ρ - the density of media

B - a bulk modulus

v - the propagation velocity of ultrasound.

Unit for the acoustical impedance is therefore [Z] = kg m^{-2} s^{-1}.

Materials with high Z transmit sound faster than others, but do not allow for continued compression by the impending wave. Some common acoustic

impedance values are given in table 1. There is almost complete reflection at the boundary between soft tissue and air and also when a beam travelling through soft tissue encounters bone. In contrast, the reflection at a boundary between different kinds of soft tissues is quite small, leaving most of the energy to travel across the boundary and into deeper tissues of the body. Reflection at the boundaries between different kinds of tissues is a rather idealised situation. It is the backscattering of ultrasound as it travels through tissues that is generally much more relevant to the imaging process.

Material	Propagation of speed, *c* (m s^{-1})	Characteristic impedance, Z (10^6 kg m^{-2} s^{-1})
Air	330	0.0004
Blood	1570	1.61
Brain	1540	1.58
Fat	1450	1.38
Liver	1550	1.65
Muscle	1590	1.70
Skull bone	4000	7.80
Soft tissue (mean values)	1540	1.63
Water	1480	1.48

The propagation velocity of ultrasound ***v*** depends on many parameters e.g. density of the media, elastic properties of the media and temperature. In solid media the propagation velocity can be calculated from

$$\mathrm{v} = \sqrt{\frac{\mathrm{B}}{\rho}}$$

where: *B* - a bulk modulus

ρ - the density of the medium

When the propagation media is water, main parameters affecting the propagation velocity are temperature of the water and the amount of foreign substances e.g. salt in water. The pressure of water has also a small influence on

propagation velocity. We can get the following theoretical equation for the propagation velocity

$$v = \frac{1}{\sqrt{\rho \cdot \beta_{ad}}}$$

where: ρ - the density of water

β_{ad} - the adiabatic compressibility.

To obtain information about a medium, the wave is propagating through the medium as a disturbance of the particles. Due to the presence of the ultrasound wave, the particles will oscillate around their mean positions. The ultrasound wave interacts with the particles and is reflected, absorbed or scattered. Ultrasounds are reflected from the boundaries between materials of nearly the same density. The reflected or scattered wave can be received by a transducer. After processing the received signals, information about the medium and its properties can be obtained.

Two main concepts are commonly used in medical ultrasound systems: the first concept is called the **continuous wave system**, where one transducer emits a continuous ultrasound wave, and a second transducer receives the reflected and scattered echo signal. The second concept is based on using a single transducer which switches between emitting and receiving ultrasound waves. This concept is called the **pulsed wave system**. For measuring blood flow, it would be much easier to use the first concept because the Doppler equation can be applied directly.

Doppler Effect

When a vehicle with a siren passes you, a noticeable drop in the pitch of the sound of the siren will be observed as the vehicle passes. This is an example of the Doppler Effect (Fig. 1). An approaching source moves closer during period of the sound wave so the effective wavelength is shortened, giving a higher pitch since the velocity of the wave is unchanged. Similarly the pitch of a receding sound source will be lowered.

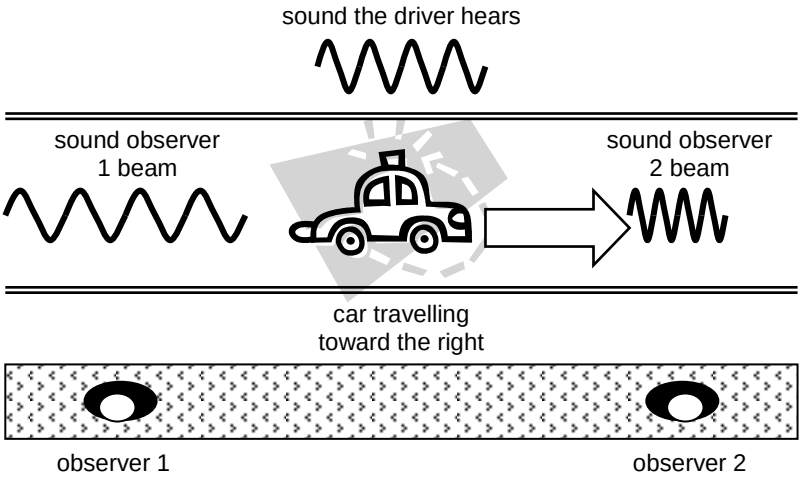


Fig. 1 Doppler Effect: The person behind the car hears a lower tone than the driver because the car is moving away. The person in front of the car hears a higher tone than the driver because the car is approaching.

When the observer is stationary and the source is receding, the frequency observed f_o is:

$$f_o = \left[\frac{v}{v + v_s} \right] f_s$$

When the source is approaching the frequency observed f_o is:

$$f_o = \left[\frac{v}{v - v_s} \right] f_s$$

where: f_o – frequency observed

f_s – frequency of the source

v – velocity of the sound

v_s – velocity of the source

When emitting an ultrasound wave into a medium containing blood vessels, the emitted energy will be received by either the same transducer (**pulsed wave**) or by another transducer (**continuous wave**).

To obtain a Doppler signal from a fluid, the fluid must contain scattering particles, which in the case of blood are the blood cells. The size of a red blood cell is about 2 x 7 μm, which means that the scatterering objects are much smaller than the wavelength of the ultrasound. Hence, a diffuse scattering of the

ultrasound will occur (Rayleigh scattering). The scattering from tissues surrounding the heart and vessels usually gives a much larger signal (20 to 40 dB) than that from blood in motion. The velocity of tissue motion is usually much lower than that of blood. This contribution can therefore be suppressed by high-pass filtering. In recent years ultrasound contrast agents (consisting of gas-filled shells) have been introduced to increase the blood flow signal.

Continuous wave ultrasound probe detection (Fig. 2)

Continuous wave Doppler ultrasound is a widely used non invasive diagnostic technique to evaluate cardiovascular disorders. Continuous wave ultrasound probe techniques involve an ultrasound signal being transmitted into the skin, and the difference in frequency reflected being measured. Continuous wave Doppler instruments detect blood flow velocity using the Doppler Effect by means of continuous wave transmission of ultrasound into the tissues. The backscattered ultrasound signal is detected and amplified by the instrument as an audio frequency signal. Transmission is continuous, therefore continuous wave Doppler

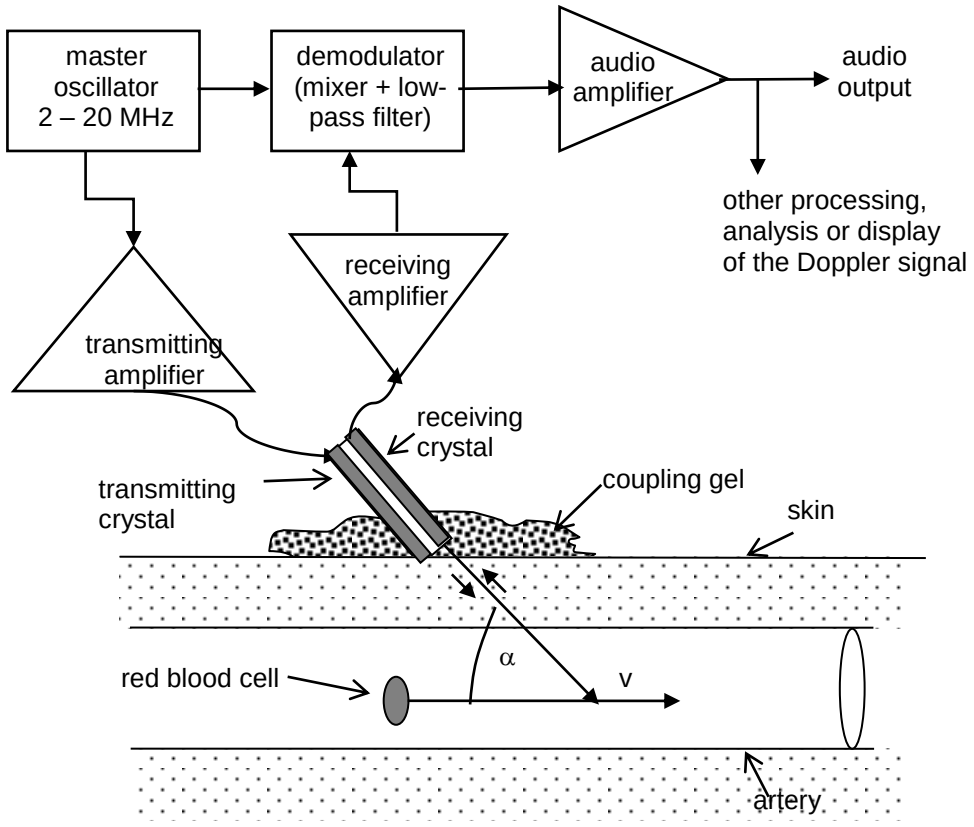


Fig. 2 Flow velocity components illustrated on a single moving red blood cell (scatterer).

instruments have no depth resolution. However, continuous wave methods are extremely simple and able to detect high velocities. A block diagram of a continuous wave Doppler system is depicted in Fig. 2. Continuous wave Doppler probes are constructed using two identical crystals. One insonates the moving media when excited by the oscillator, a radio frequency signal generator. The other detects back-scattered ultrasound signal and converts it into an electrical signal. This electrical signal is amplified by the radio frequency amplifier if necessary, and the frequency-shifted audio signals are demodulated by means of the mixer. The mixer is an electronic device that basically multiplies two incoming signals and produces an output proportional to the amplitudes of the input signals. The output of the mixer has two main frequency bands; f_t+f_r and f_t-f_r. A low-pass filter, which forms the product detector with the mixer, filters out the frequency band containing high frequency signals. The remaining signals are the frequency shifted Doppler signals. These are amplified by the audio amplifier and may be presented audibly via a speaker or processed for further interpretation.

The **continuous wave** method makes use of the **Doppler Effect** to calculate the velocity of the objects causing the scattering. An ultrasound beam is sent toward a moving object. The beam hits the object and returns to the receiver with a Doppler shifted frequency carrying information about the velocity of the object. The Doppler shift frequency f_d of an ultrasound signal presents the difference between the transmitted frequency f_t and received frequency f_r, and is given by

$$f_d = f_t - f_r = \frac{2v \cdot f_t \cdot \cos\alpha}{c}$$

where: f_d - the Doppler shift frequency

f_t - frequency transmitted by an ultrasound transducer

f_r - frequency received by an ultrasound transductor

v - the velocity of the target (a red blood cell)

c - the velocity of sound in the medium (1500 to 1600 m/s in soft tissue and usually set to 1540 m/s)

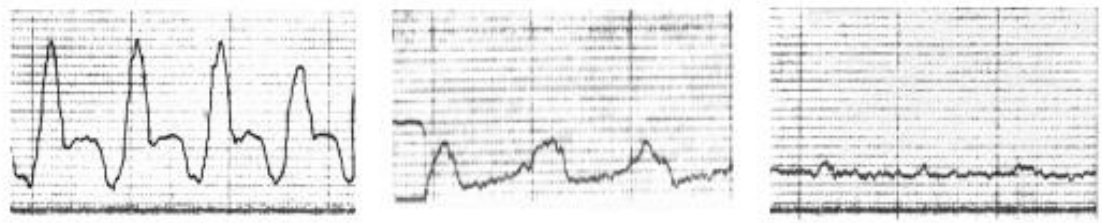
α - the angle between the ultrasound beam and the direction of a target motion (or skin)

Note that the factor of two appears in the equation because the moving object “hears” a Doppler shift, as does the observer of the scattered ultrasound.

The velocity of sound in the medium and the transmitted frequency are known and the angle between the ultrasound beam and the direction of the target's motion can be determined. In this case, the velocity of the target can be found from the expression:

$$v = \frac{f_d \cdot c}{2 f_t \cdot \cos\alpha}$$

Ultrasonic flowmetry is a commonly used technique for measuring blood velocity in the peripheral arteries. As a result, it can also be used to determine the amount of stenosis, or narrowing, in an artery, since a narrow passage means higher blood velocity (Fig. 3).



A) B) C)

Fig. 3 Pulse-volume recording waveform of a femoral artery a) normal b) narrowed by atherosclerosis c) obstructed.

Spectral Doppler waveform measurements

Doppler waveform analysis is often used as a diagnostic tool in the clinical assessment of disease. The complex shapes of Doppler waveforms can be described by relatively simple waveform indices, which have been used to evaluate foetal health and organ blood flow. Common indices are the **pulsatility index** (PI or Gosling index), **resistance index** (RI or Pourcelot Index), and **systolic diastolic ratio** (S/D) (Fig. 5).

The pulsatility index (Fig. 4) quantifies the shape of the velocity waveform and is defined as the peak-to-peak amplitude of a waveform divided by the average amplitude over the cardiac cycle.

$$PI = \frac{(v_{max} - v_{min})}{v_{av}}$$

where: v_{max} – the maximum velocity

v_{min} - the minimum velocity

v_{av} – the time average peak velocity

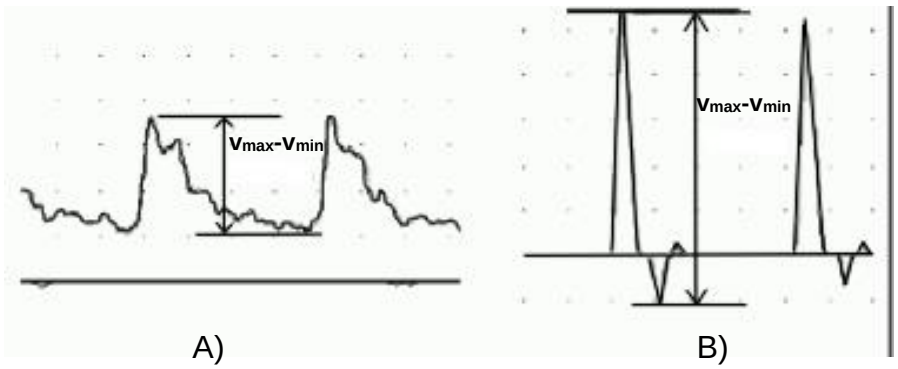


Fig. 4 The method of measurement v_{max} - v_{min} during one-way flow (A) and two-way flow with a return wave (B).

A PI below 5 corroborates an acceptable flow waveform. A PI less than 1 shows an severe narrowing of the artery and a PI of more than 5 is often considered indicative of technical error.

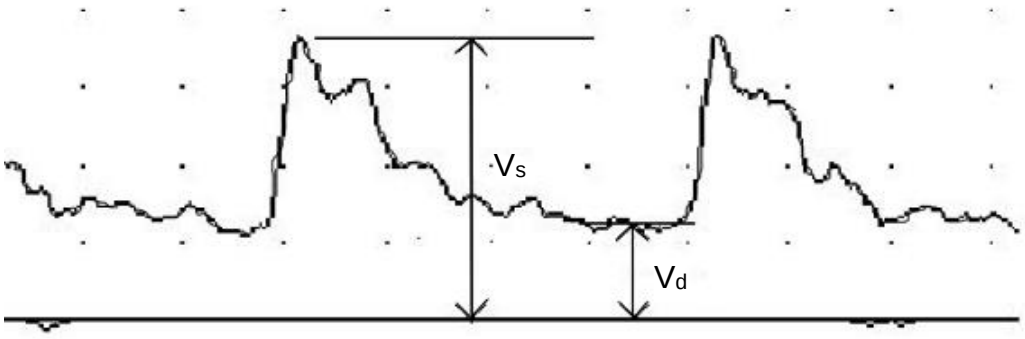


Fig. 5 The method of measurement v_s and v_d

$$RI = \frac{(v_s - v_d)}{v_s}$$

$$S/D = \frac{v_s}{v_d}$$

where: v_s – the maximum systolic velocity

v_d - the minimum diastolic velocity

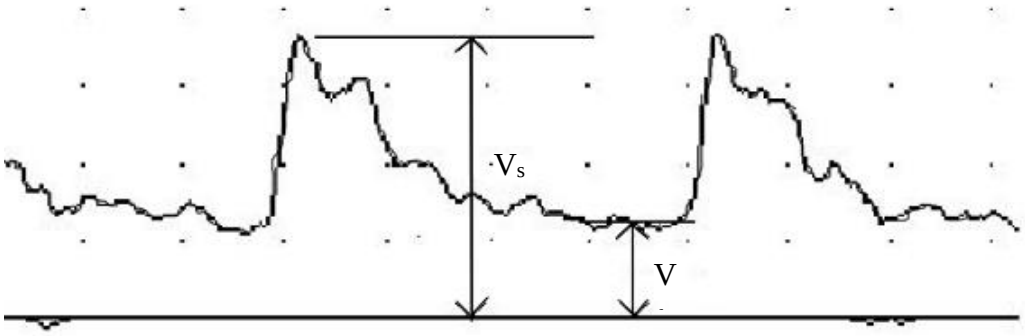


Fig. 5 The method of measurement v_s and v_d

Dimensionless variables such as the PI and RI are not dependent on the insonation angle and are calculated for each flow velocity. These parameters are considered to indicate the peripheral vascular resistance.

Biophysical effects of ultrasound

Because ultrasound is energy there have been many concerns about the safety of ultrasound. Biophysical effects of ultrasound are traditionally separated into thermal and non-thermal (mechanical) effects.

Non-thermal effects are divided into cavitation and other mechanical effects:

- *Cavitation* may be more specifically defined as the formation of tiny gas bubbles in the tissues as the result of ultrasound vibration.
- *Other mechanical effect:*

– *Acoustic streaming* is as a major effect of insonation and is described as localised liquid flow in the fluid around the vibrating bubble.
– Mast cell *degranulation*
– *Increased* membrane permeability
– *Small oscillation* of particles due to the movement of ultrasound waves through tissues.
– *Free radical formation* as a potential source of cell damage with ultrasound.
– Ultrasound may causes growth of new blood vessels (*angiogenesis*)
– *Stimulation of fibroblast proliferation* with ultrasound
– *Changes* in the plasma membrane and in intracellular organelles such as lysosomes and mitochondria.

Thermal effects (minimum 10 min - 2.0 watts - 1 MHz) are those due to **heating** and are accepted as including increased metabolic activity and blood flow and an analgesic effect on nerves. Beneficial effects of heating are exploited in ultrasound **therapy**:

- increased blood flow which promotes healing,
- produces a pain killing effect which can reduce muscle spasm, and promote normal function,
- softens fibrous tissues and scar tissue which are formed during healing
- resolution of inflammatory infiltrates, e
- extensive heating will destruct tissues, and high-intensity focused ultrasound may be used in tumour ablation.
- heat also has a teratogenic effect.

Major uses of ultrasound in diagnostic

Ultrasound is useful for diagnostic procedures including: obstetric (related to pregnancy) and non-obstetric uses for ultrasound. Specialized ultrasound imaging procedures include: echocardiography, Doppler echocardiography, ultrasound for biopsy

Here is a short list of some uses for ultrasound:

Obstetrics and gynaecology

- measuring the size of the foetus to determine the due date
- determining the position of the foetus
- checking the position of the placenta
- seeing the number of foetuses in the uterus
- checking the sex of the baby (if the genital area can be clearly seen)
- checking the foetus's growth rate by making many measurements over time
- detecting ectopic pregnancy, the life-threatening situation in which the baby is implanted in the mother's Fallopian tubes instead of in the uterus
- monitoring the baby during specialised procedures - ultrasound has been helpful in seeing and avoiding the baby during amniocentesis (sampling of the amniotic fluid with a needle for genetic testing).
- seeing tumours of the ovary and breast

Non-obstetric uses for ultrasound

- scanning of the brain in a newborn
- examining of the internal organs, including the appendix in possible appendicitis (rupture of the appendix)
- scanning of the liver to determine cirrhosis (liver inflammation) and liver cysts (fluid filled sacs), abscesses (infections), or tumours (masses of cells)
- locating gallstones in the gallbladder or bile ducts
- scanning of the pancreas for cysts, tumours, or pancreatitis (inflammation of the pancreas)
- evaluating the eyes
- scanning of the kidneys for congenital defects, tumours, and hydronephrosis (swelling of the kidney due to urine outflow obstruction)
- scanning of the thyroid gland, breasts, bladder, testicles, ovaries, spleen, and eyes for cysts, tumours, or foreign objects
- identification of the cause of enlargement of the abdomen or other organs

Echocardiography

Echocardiography is a specialized type of ultrasound that is used to look at the action and function of the **heart**. This type of ultrasound is a major **diagnostic** technique that can detect structural and some functional abnormalities of the heart wall, valves, and large blood vessels. Blood flow across the valves can also be measured.

Other diagnostic uses of echocardiography include:

- Detection of congenital heart disease
- Detection of problems with the large blood vessels
- The presence of a blood clot in one of the chambers of the heart

Doppler echocardiography

Doppler echocardiography is a recently developed ultrasound technique that indirectly measures the flow velocity of blood as it passes through the heart. It is used in assessing malfunctioning valves in **aortic stenosis** (narrowing of the

aortic valve opening) or **mitral insufficiency** (failure of the mitral heart valve to close properly) and in assessing patients with **congenital heart disease**.

Ultrasound for biopsy

Another increasingly common use for ultrasound is in conjunction with fine-needle biopsy (inserting a very thin and hollow needle into an organ to remove tissue or fluid for examination). Ultrasound provides a real time, moving image. So, the image is valuable in helping to guide the needle accurately to a specific spot.

PRACTICAL PART

Answer the questions:

1. Define “infrasound” and "ultrasound".

2. What are the effects of infrasound?

3. What are the effects of ultrasound?

EXPERIMENTAL PART

Part A

Objective: Calculating the velocity of the blood in the arteries.

Materials: a vascular flowmeter UDP-10, a coupling gel (used during ultrasound examinations. The gel fills the space between the transducer and the skin and prevents reflection by trapped air.)

Procedure

1. Plug the transductor jack into the Doppler unit at the plug with the “GŁOWICA” symbol (Fig. 6).
2. Switch on the Doppler unit by pressing “SIEĆ” key and press the artery key (“TĘTNICE”)
3. Place some gel on your wrist (place your index and middle finger over the underside of your opposite wrist, below the base of the thumb) and obtain a signal for arterial blood flow (from the radial artery). The Doppler shift frequency returning to the transductor is processed and we can observe an indicator swing on the A or B meters. If the transductor is placed in the direction of the blood flow we observe a swing on the A meter, when the transductor is positioned opposite the blood flow direction we observe a swing on the B meter.
4. Listen to the signal. Record a Doppler shift frequency and the change in heart rate.
5. Adjust position as necessary to achieve a signal that is as loud as possible.
6. Perform some mild exercises: i.e. 20 knee bends within 20 seconds or 10 press-ups. Record a Doppler shift frequency and the change in heart rate.
7. Perform a Valsalva manoeuvre, which is a forceful exhalation against a closed glottis. The increased pressure that develops within the thorax decreases venous return and increases heart rate. Record a Doppler shift frequency and the change in heart rate.

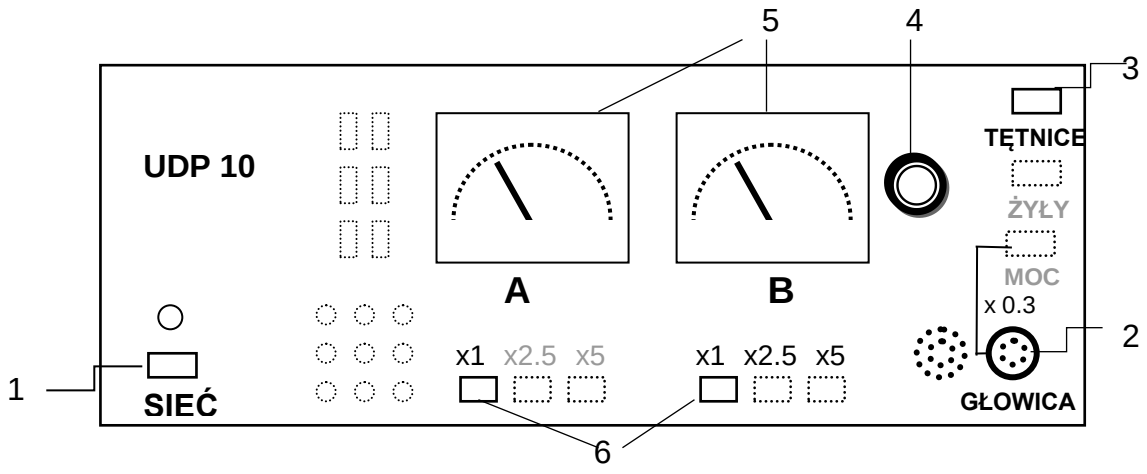


Fig. 6 Ultrasonic Doppler flowmeter UDP 10 front panel. 1 - the power switch; 2 - the transductor jack; 3 - the artery key; 4 – the knob of sound intensity; 5 - the A or B meters; 6 – the frequency multiplier.

8. Calculate the velocity of the blood in the artery, assuming *c* is 1570 m/s, the frequency transmitted is 8 MHz.

Data and observations

a) write down the equation you need

……

b) write down the known data

The name of the given data	symbol	value
the frequency transmitted [Hz]		
the velocity of sound in the medium [m/s]		

c) Complete the table with proper data. All calculations write down below the table

The place or the method of measurement	A Doppler shift frequency [Hz]	α	cos α	The velocity of the blood [cm/s]
The radial artery				
The radial artery after mild exercise				
The radial artery after a Valsalva manoeuvre				

The date	**Student's name and surname**	**Lab assistant signature**

Part B

Objective: Observing and describing pulse-volume recordings and the waveform parameters.

Materials: a vascular flowmeter UDP-5R, a coupling gel, a computer, computer software UDP 1.29.

Procedure

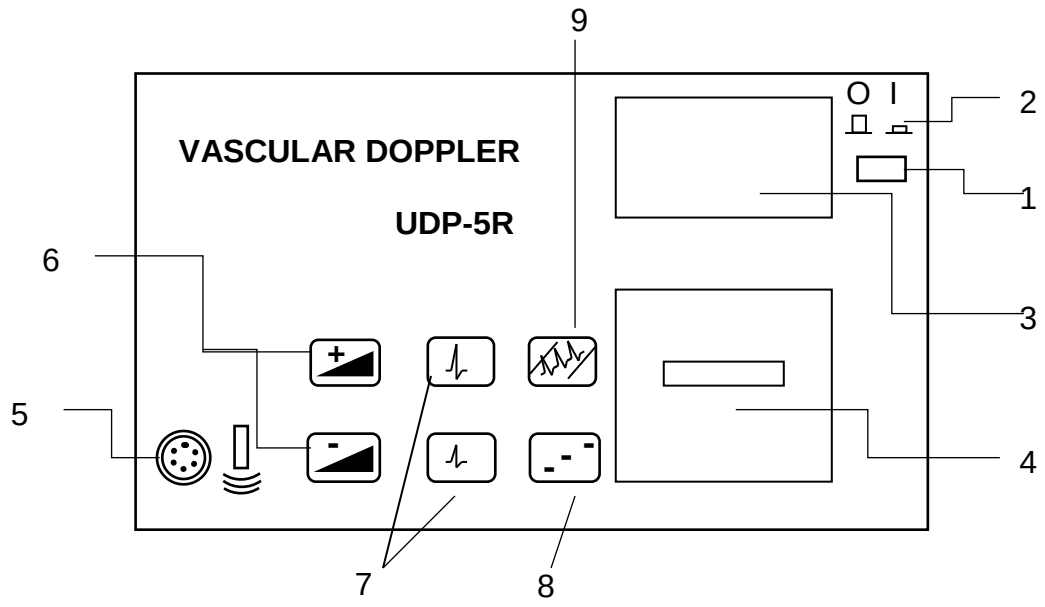


Fig. 7 Ultrasonic Doppler flowmeter UDP-5R front panel. 1- the power switch; 2 – position of the power switch; 3 – the thermal registering device; 4 - the door of a thermal paper container; 5 - the transductor jack; 6 - sound intensity keys; 7 –flow velocity curve keys for changing the registration range; 8 – “0” line change key; 9 – “start-stop” registration key.

1. Plug the transductor jack into the vascular Doppler flowmeter at the plug with  the symbol (Fig. 7).

2. Switch on the vascular Doppler by pressing the **O I** key.
3. Place some gel on your wrist (place your index and middle finger over the underside of your opposite wrist, below the base of the thumb) and obtain a signal for arterial blood flow (from the radial artery). Observe the signal.

4. Perform a Valsalva manoeuvre, which is a forceful exhalation against a closed glottis. The increased pressure that develops within the thorax decreases venous return and increases heart rate. Record the change in heart rate.
5. Occlude blood flow by pressing your finger on the artery inside your elbow. Observe the resulting signal.
6. Release finger and observe the resulting signal.
7. Place some gel on the inside of your elbow, just below where you have been occluding blood flow. Move the probe to obtain a signal for venous blood flow while avoiding the pulsating arterial signal. Observe the signal.
8. Occlude blood flow again. Observe the signal.
9. Release finger and observe the resulting signal.
10. Inhale deeply, hold your breath briefly, and observe the resulting signal.

Data and observations

Describe and explain what happens in step from 7 to 11.

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NOTES

2.6 Lab exercise

THE CIRCULATORY SYSTEM – THE FUNDAMENTALS OF MOTION OF FLUIDS

THEORY

Arterial blood pressure is the force exerted by the blood on the wall of a blood vessel as the heart pumps (contracts) and relaxes. **Systolic** blood pressure is the degree of force when the heart is pumping (contracting). The **diastolic** blood pressure is the degree of force when the heart relaxes.

How to measure blood pressure

• Invasive measurement (direct measurements) is the gold standard for blood pressure measurement.

• Non-invasive measurements (indirect measurements) are conducted without surgery, and are performed on an out-patient basis (the auscultatory method, the oscillometric method, alternative methods)

Invasive Catheterisation Method

The blood pressure in an artery or vein is measured by the direct insertion into the vessel of a cannula, which is a small glass or plastic tube containing saline solution plus an anticlotting agent.

The Auscultatory Method

In 1905, Korotkoff described the auscultatory sounds which became the foundation for the auscultatory technique (Fig.1). This is the most common method of blood pressure measurement today because of patient safety, comfort, and cost.

Real fluids in motion always exhibit some effects of frictional or viscous forces. Viscosity is readily defined by considering a simple experiment. Figure 3 shows two flat plates separate by a thin fluid layer. If the lower plate is held fixed, a force is required to move the upper plate at a constant speed. This force is needed to overcome the viscous force due to the liquid. The force F is observed to be

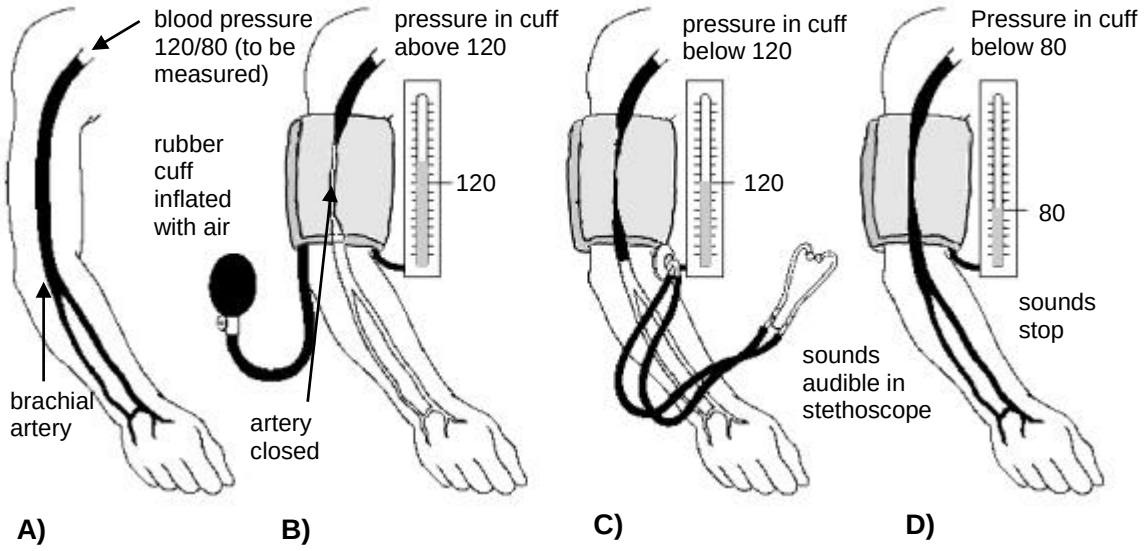


Fig.1 Conventional sphygmomanometry: technique of auscultatory blood pressure measurement. The unit of pressure is mmHg.

proportional to the area of the plates A and the velocity of the upper plate Δv, and inversely proportional to the plate separation Δx:

$$F = \eta S \frac{\Delta v}{\Delta x}.$$

The proportionality constant η (eta) is called the **viscosity**. From this equation the S.I. unit of viscosity is

$$[\eta] = \frac{Ns}{m^2} = \frac{kg}{ms}$$

The fluid in contact with the moving plate in Fig.2 has the same velocity as the plate. The fluid layer just below moves slightly more slowly, and each successive layer lags a bit more. This layer structure or **laminar flow** is the kind of streamline

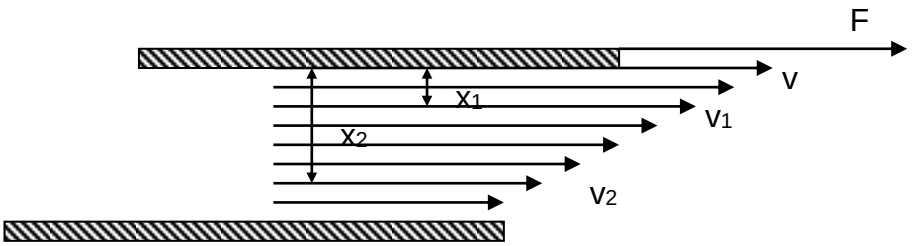


Fig. 2 Viscous forces.

flow characteristic of viscous fluids at low velocities. In a tube of radius R, the laminar flow rate is proportional to the pressure gradient and R^4:

$$Q = \frac{\pi R^4}{8\eta l}\Delta p$$

The formula for Q is called Poiseuille's law. It indicates that high viscosity leads to low flow rates, which is reasonable. The velocity of the fluid is a maximum at the centre of the tube and decreases gradually in successive lamina to zero at the walls.

When the fluid velocity is increased sufficiently, the flow changes its character and becomes **turbulent**. In order to determine whether the flow is laminar we use empirical rules and relationships developed from experimental studies. One of these rules states that the value of a dimensionless quantity called the Reynolds number N_R determines whether the flow is turbulent or laminar. Consider a fluid of viscosity η and density ρ. If it is flowing in a tube of radius R and has an average velocity v, then the Reynolds number is defined by

$$N_R = \frac{2\rho \cdot v \cdot R}{\eta}$$

In tubes, it is found experimentally that if

$N_R < 2000$, flow is laminar

$N_R > 3000$, flow is turbulent

$2000 < N_R < 3000$, flow is unstable (may change from laminar to turbulent or vice versa).

Or considering the velocity we can say that: If the velocity of a fluid is increased past a critical point, the laminar flow is disrupted. In a cylindrical pipe the critical flow velocity v_c above which the flow is turbulent, is given by

$$v_c = \frac{N_R \cdot \eta}{\rho \cdot R}$$

The examined person (the patient) sits down and rests their arm on a table so the brachial artery is level with the heart. This is important when monitoring blood pressure, as pressure is proportional to height ($\Delta p = \rho g \Delta h$). Through most of the circulatory system the blood flow is laminar. A sphygmomanometer cuff is wrapped around the patient's upper arm, just above the elbow and the cuff is

'pumped- up' to a pressure of 180mmHg, compressing the brachial artery hence causing the artery to collapse once the systolic pressure (the maximum pressure exerted by the blood against the wall of the brachial artery when the heart beats) has been exceeded.

At the point where the pressure of the cuff is greater then the systolic pressure, the artery has collapsed thus, there is no flow of blood through the brachial artery (Fig.1B and 3).

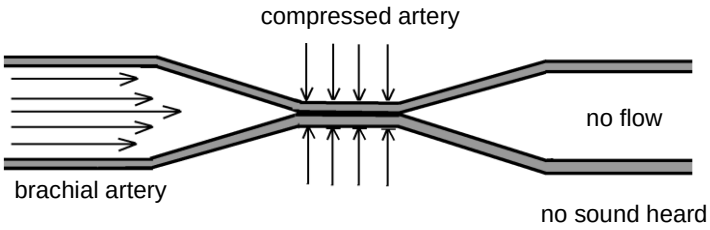


Fig. 3 Cuff inflated until brachial artery compressed and blood flow stopped.

The valve on the pump is loosened slowly to allow the pressure of the sphygmomanometer cuff to decrease. The velocity of blood can be increased for instance by the compressing the artery (since the velocity is inversely proportional to the radius of the tube). Once the systolic pressure is reached (approximately 120 mmHg in the 'normal' case), the brachial artery opens causing turbulent blood flow. The turbulent flow is characterized by eddies and whirls disrupting the laminar flow which cause vibrations against the artery walls (Fig.4). These noises

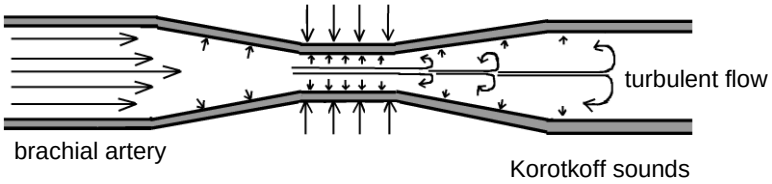


Fig. 4 Slowly release pressure in a cuff: turbulent flow trough a compressed artery sets up vibrations that are heard as Korotkoff sounds

are called Korotkoff sounds (named after their discoverer) and can be heard through a stethoscope as the pressure exerted onto the brachial artery falls. The

blood flow through the brachial artery increases steadily, until the pressure of the sphygmomanometer cuff falls below the diastolic pressure (the pressure between successive heart beats, the low pressure), approximately 80 mmHg. This is the point where the blood flow through the artery is laminar (Fig.5).

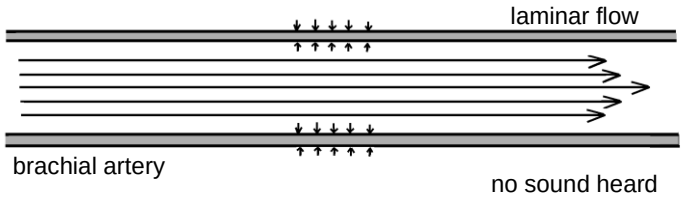


Fig. 5 The point when all sounds have disappeared and where the blood flow through the artery is laminar again.

The Korotkoff sounds

The Korotkoff sounds are the sounds heard through the stethoscope as the pressure cuff deflates. The sounds are first heard when the cuff pressure equals the systolic pressure, and cease to be heard once the cuff has deflated past the diastolic pressure. It is generally accepted that there are five phases of Korotkoff sounds. Each phase is characterized by the volume and quality of sound heard (Fig.6).

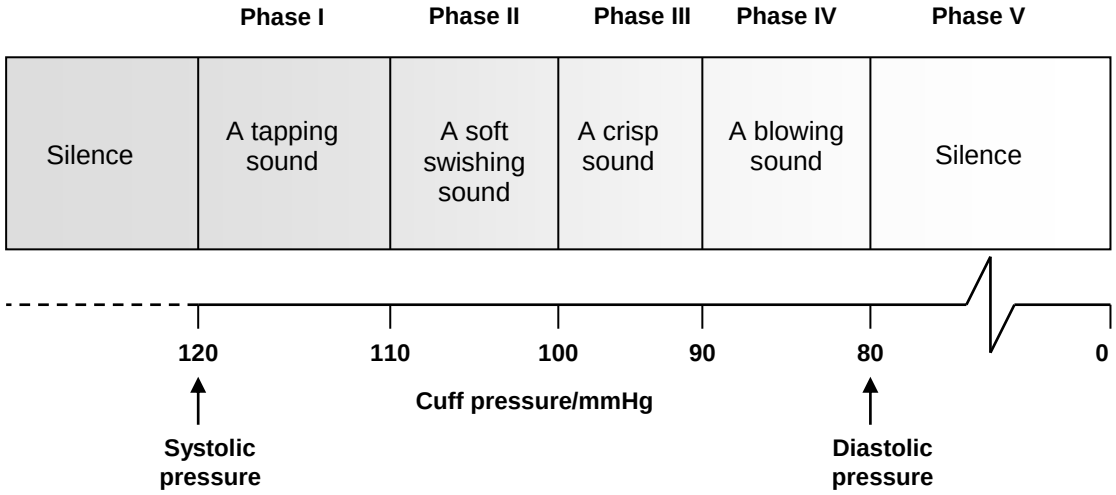


Fig. 6 The phases of Korotkoff sounds.

- Phase I - The first appearance of faint, repetitive, clear tapping sounds which gradually increase in intensity for at least two consecutive beats is the systolic blood pressure.
- Phase II - A brief period may follow during which the sounds soften and acquire a swishing quality. In some patients sounds may disappear altogether for a short time (an auscultatory gap).
- Phase III - The return of sharper sounds, which become crisper to reach, or even exceed, the intensity of phase I sounds.
- Phase IV - The distinct abrupt muffling of sounds, which become soft and blowing in quality.
- Phase V - The point at which all sounds finally disappear completely is the diastolic pressure.

Pressure measurement equipment

The mercury sphygmomanometer (Fig. 7A) is a simple and accurate device, which can be easily serviced, but there are rightly concerns about the toxicity of mercury for individuals using mercury sphygmomanometers, and for those who have to service them. Aneroid sphygmomanometers (Fig.7B) register pressure through a bellows and lever system, which is mechanically more intricate than the mercury reservoir and column. The jolts and bumps of everyday use affect their accuracy; they lose accuracy over time, usually leading to falsely low readings with the consequent underestimation of blood pressure. They are therefore less

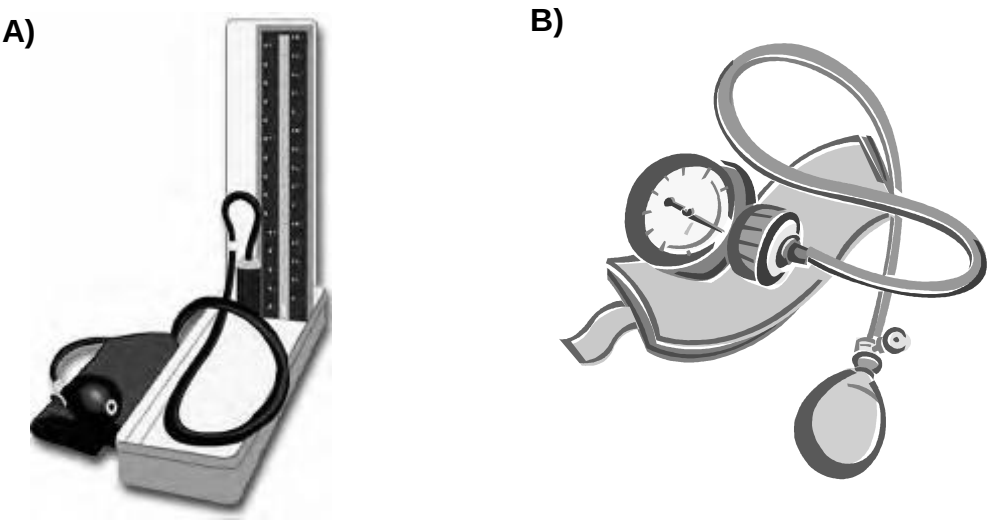


Fig. 7 A mercury sphygmomanometer – A); an aneroid sphygmomanometer – B).

accurate in use than mercury sphygmomanometers. When calibrated against a mercury sphygmomanometer a mean difference of 3 mmHg is considered to be acceptable; however, 58% of aneroid sphygmomanometers have been shown to have errors greater than 4 mmHg, with about one third of these having errors higher than 7 mmHg.

The Oscillometric Method

The term "oscillometric" refers to any measurement of the oscillations caused by the arterial pressure pulse. These oscillations are the direct results of the coupling of the occlusive cuff to the artery. This method allowed blood pressure measurement of critical care and intensive care (ICU) patients with muted Korotkoff sounds. These devices do not use microphones. Therefore, cuff placement and external noise are not significant problems. These devices are sensitive to patient movement and do not allow measurement validation.

Unlike auscultatory techniques, which measure systolic and diastolic, but estimate, mean arterial pressure, oscillometric devices measure the mean but estimate systolic and diastolic. An air-filled cuff is wrapped around the patient's upper arm. The cuff is inflated to occlude the brachial artery. As the cuff is allowed to deflate, pressure data is recorded by the device.

Alternative Methods

The majority of monitors today are either oscillometric or auscultatory in nature. However, there are other types of devices. Some monitors employ both auscultatory and oscillometric methods, using one method as a primary measurement and the other for verification, to minimize the inherent disadvantages of each.

The infrasound technique attempts to improve on the auscultatory method by detecting low frequency Korotkoff vibrations below about 50 Hz, including sub-audible vibrations.

Ultrasound techniques are also used in combination with other methods. Values recorded by ultrasound can be very operator dependent. A blood pressure cuff is applied to the limb being tested. It is inflated until the pulsatile flow is no longer heard. Then the pressure is released from the cuff until the flow returns.

Arterial tonometry uses a very different approach. The artery is flattened by applying pressure non-invasively to squeeze the artery against bone. The applied pressures required to maintain the flattened shape are recorded. This is accomplished by using an array of sensors, each of which measures pressure.

The role of gravity in the circulation

When humans evolved to the point where they spent a significant amount of time standing upright, a number of changes in the circulatory system where required. Of particular importance is the system used to return blood from the lower extremities to the heart. Humans have adapted to the problem of moving blood upward a large distance against the force of gravity. Animals that have not, such as snakes, eels, and even rabbits, will die if held head upwards.

Figure 11 shows what is observed if a person's large arteries are cannulated. In the reclining position, the pressure everywhere are almost the same, The small pressure drop between the heart and the feet or brain is due to the viscous forces. However, the pressures at the three points are quite different in the standing person, reflecting the large difference in their heights.

Since viscous effects are small, we can use Bernoulli's equation to analyze this situation.

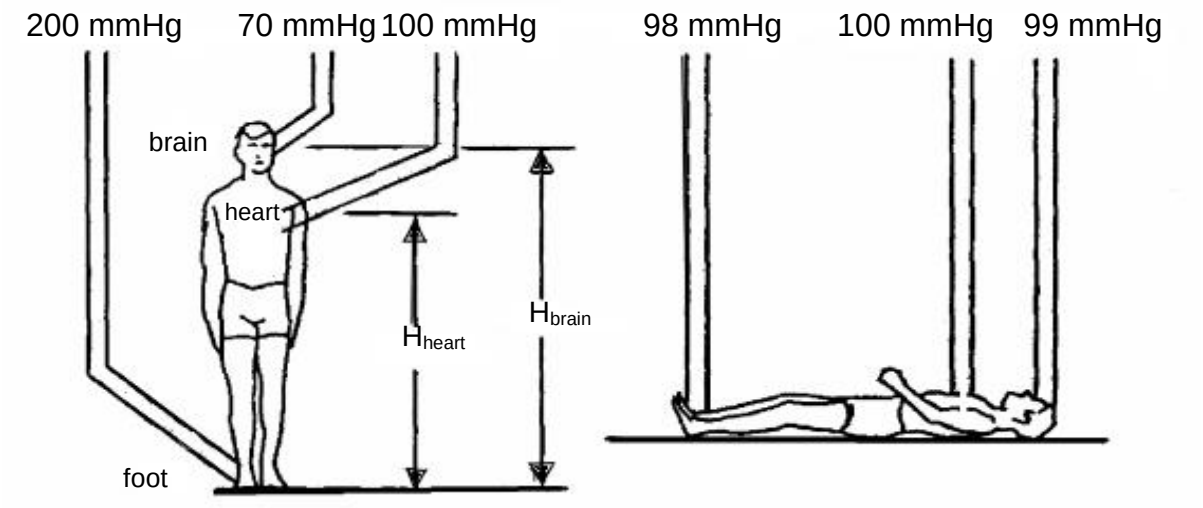


Fig. 11 Schematic view of the results of cannulation of arteries in various parts of the human body, when standing and reclining.

$$p + \rho gh + \frac{1}{2}\rho v^2 = const$$

The velocities in the three arteries are small and roughly equal, so the $\frac{1}{2}\rho v^2$ term can be ignored. Hence the gauge pressures at the heart P_H, at the foot P_F and the brain P_B are related by

$$P_F = P_H + \rho g h_H = P_B + \rho g h_B$$

where: ρ = 1.0595 x 10^3 kg/m^3 (the density of blood)

g = 9.8 m/s^2 (acceleration of gravity)

P_H - the gauge pressures at the heart when standing [Pa]

P_F - the gauge pressures at the foot [Pa]

P_B - the gauge pressures at the brain when standing [Pa]

h_H – the height of the heart [m]

h_B - the height of the brain [m]

PRACTICAL PART

Answer the questions:

1. Define the following:

The equation of continuity

The Bernoulli equation

diastolic blood pressure

systolic blood pressure

Korotkoff sounds

2. What factors can change your blood pressure?

EXPERIMENTAL PART

Objective*:* Students analyse the amount of pressure using different methods.

Materials: manual pressure cuff/stethoscope/manometer; a vascular flowmeter;

Procedure

Part A

An Auscultatory Method (Riva-Rocci Method)

1. Clean the earpiece and the chestpiece of the stethoscope with alcohol wipes.
2. Have your lab partner roll up a sleeve on the left arm and sit in a chair (or lie flat) so that the upper arm is level with the heart.
3. Squeeze the blood pressure cuff to expel any air from it. Close the valve on the bulb.
   Wrap and fasten the cuff around the lab partner's upper arm at least one inch above the elbow. The cuff should fit evenly and snugly. The gauge should be vertical and at your eye level.
4. Place the earpiece of the stethoscope in your ears. Place the chestpiece of the stethoscope at the bend of the elbow over the brachial artery.

5. Begin to inflate the cuff by squeezing forcefully on the bulb. Inflate the cuff to about 180 -200 mm Hg. You should not be able to hear a pulse when you press down gently on the chestpiece of the stethoscope.
6. Slowly release the valve until the needle on the gauge drops by 2-3 mm per sec.
7. Note the point on the scale where you begin to hear a pulse (Korotkoff sounds).. Record this measurement in the data table as the systolic pressure.
8. As the cuff continues to deflate, listen closely for the point at which the pulse can no longer be heard. Record this as the diastolic pressure in the data table.
9. Open the valve all the way to deflate the cuff completely. Remove it from the arm. Remove the stethoscope and clean the earpiece and the chestpiece with alcohol wipes.
10. Change places with your partner. Record the values of the average blood pressure on the data sheet.

Part B

A Palpation Method

NOTE

DO NOT use your thumb to check a casualty's pulse because you may confuse your pulse beat with that of the casualty.

1. Hand pump rapidly to 200 mm Hg to apply enough pressure in the cuff to occlude arteries. Note loss of sound by palpation of the radial artery (place your index and middle finger over the underside of your opposite wrist, below the base of the thumb).
2. Slowly bleed air out of the cuff with valve on pump bulb (~ 3 mmHg/s).
3. Record systolic pressure as you simultaneously hear the return of the radial pulse (Korotkoff sounds).

Part C

A Doppler Method

1. Hand pump rapidly to 200 mm Hg to apply enough pressure in the cuff to occlude arteries. Note loss of sound by palpation of the radial artery (place

your index and middle finger over the underside of your opposite wrist, below the base of the thumb).

2. Slowly bleed air out of the cuff with valve on pump bulb (~ 3 mmHg/s).
3. Record systolic pressure as you simultaneously hear the return of the radial or brachial pulse using an ultrasound probe for sound detection.
4. Record diastolic pressure as soon as the Korotkoff sounds disappear.

Part D

An automatic method

1. Place cuff around arm and press the START button.
2. Record systolic and diastolic pressure.

Data and observations

Pressure conversion

760 mmHg = 101.3 kPa

1 mmHg = 133.322 Pa

1 mmHg = 1.33322 hPa

1 Pa = 7.5 x10^{-3} mmHg

A MEASUREMENT METHOD	RR in mmHg (Systolic/diastolic)	RR in kPa (Systolic/diastolic)	HART RATE/min
An Auscultatory Method			
A Palpation Method			
A Doppler Method			
An Automatic Method			

The date	Student's name and surname	Lab assistant signature

Part E

Objective: Students calculate the amount of pressure at the foot artery (P_F) and brain artery (P_B).

Materials: a measuring scale

Procedure

1. Write down the relation among pressure at the foot artery (P_F), brain artery (P_B) and at the heart (P_B)

2. Fill out the data in the table below

KIND OF DATA	Data	Units
ρ (the density of blood)		kg/m^3
g (acceleration of gravity)		m/s^2
P_H - the gauge pressure at the heart when standing		Pa
h_H – the height of the heart		m
h_B - the height of the brain		m

3. Calculate the amount of pressure at the foot artery (P_F) and brain artery (P_B).

KIND OF DATA	Pressure in [Pa]	Pressure in [mmHg]
P_F - the gauge pressure at the foot		
P_B - the gauge pressure at the brain when standing		

Calculations:

The date	Student’s name and surname	Lab assistant signature

NOTES

ASSIGMENTS FOR RADIOACTIVITY

3.1. Lab Exercise - Principles of dosimetry.

1. Atom and its components.
2. Isotopes and radioisotopes – how they are produced ?
3. Transmutations of the nuclei.
4. The transmutation theory – its mathematical form and chart. Decay constant and half-life.
5. Activity – formula and units.
6. Kinds of ionising radiation.
7. Principles of dosimetry (exposure, absorbed dose, effective dose).
8. Sources of exposure.

3.2. Lab Exercise - Interaction of photons and charged particles with matter.

1. Photoelectric effect, the Compton scattering, pair production.
2. The law of attenuation. Half thickness.
3. The Linear Energy Transfer. The interaction of particles with matter.
4. Bremsstrahlung.
5. Materials used for stopping ionising radiation: alpha, beta, neutrons and gamma photons.

3.3. Lab Exercise - The methods of detection of ionising radiation.

1. Gas filled detectors.
2. Scintillation detector and its assemblies.
3. Types of scintillators.
4. Applications of scintillation detectors in medicine.
5. Semiconductor detectors.
6. Measuring of radiation energy – spectrometry.
7. The Anger gamma camera.
8. Computed Tomography – generations.
9. Random nature of the decay process.
10. Standard Deviation and relative error.
11. Error of the complex measurement (with background).

3.1 Lab Exercise

RADIOACTIVITY. PRINCIPLES OF DOSIMETRY.

THEORY

The atom is the smallest particle of an element. It consists of a positively charged nucleus surrounded by negatively charged electrons, which move in orbits similar to the way in which planets move round the sun. The positive charge on nucleus exactly balances the total negative charge of all the surrounding electrons when the atom is neutral.

The nucleus consists of two types of particle very strongly held together – protons and neutrons. A proton carries one unit of positive charge. A neutron has no charge and the mass of a neutron is very slightly more than the mass of a proton.

The number of protons (and the corresponding number of electrons) determines the element (atomic number, **Z**). So if a proton is removed from nucleus (or added to) the nucleus of an atom is no longer the same element.

Chemical elements can create one, two or more forms differing only in atomic weight, which are called isotopes. The difference is due entirely to the addition or subtraction of neutrons from the nucleus. The number of neutrons (**N**) determines the isotope of the element (**Z**). Mass number (**A=Z+N**) determines the number of nucleons in an atom.

Many elements have several stable isotopes (tin has 10). Every element can be made to have radioactive isotopes (radioisotopes) by adding or removing neutrons to the nucleus. The easiest way of producing many neutron-induced nuclear transformations is to place the material to be irradiated within a nuclear reactor within which it can be subjected to intense neutron flux .

A few elements have naturally occurring radioisotopes.

A radioactive nucleus is a nucleus having a certain probability (λ - the decay constant) to undergo a transmutation, either by the emission of:

1/ a positively charged ${}^{4}_{2}\mathrm{He}$ - nucleus (called α-radiation),

2/ a negatively charged electron e^{-} (called β^{-}-radiation),

3/ a positively charged electron (positron) e^{+} (called β^{+}-radiation)

4/ by capturing a negatively charged atomic electron E_c,

5/ by spontaneous fission SF(called fission fragments).

The mass difference between the neutral atoms before and after the transmutation is found back under form of the mass and the kinetic energy of the emitted particles, the recoil energy of the emitting nucleus and the γ- radiation energy.

A neutrino ν or an anti-neutrino $\bar{\nu}$ is also emitted during the β-process. These are neutral, weightless particles carrying also part of available energy so that the β-particles have a continuous energy distribution.

All these transmutations affect the atomic number **Z** and neutron number **N** as summarised in *Table 1*.

Table 1. The transmutations of the nuclei.

Radioactivity	Emitted particle	Change of Z	Change of N	Change of A
α	He^4 nucleus	Z-2	N-2	A-4
β^-	$e^- (+\bar{\nu})$	Z+1	N-1	A
β^+	$e^+ (+\nu)$	Z-1	N+1	A
EC	$(+\nu)$	Z-1	N+1	A
SF	fragments			

These radioactive phenomena may leave the daughter nucleus in an excited state. It may lose its excitation energy in (mostly) three different ways:

1/ by γ-emission,

2/ by internal conversion,

3/ by particle emission (neutrons and protons).

In the case of internal conversion the nucleus gives its excitation energy to an atomic electron which is ejected from its shell (γ-ray emission is always accompanied by internal conversion).

When a nuclear state has a measurable life-time it is called an isomeric state. An isomeric state may either decay by particle emission, by electron capture or by more or less converted isomeric γ-transition **(IT**) to a lower excited state.

One or several decay modes are possible for one and the same nucleus, each having its own transition probability: λ_1, λ_2, λ_3 ..... The observed transition probability λ is given by the sum: $\lambda = \lambda_1 + \lambda_2 + \lambda_3 +$

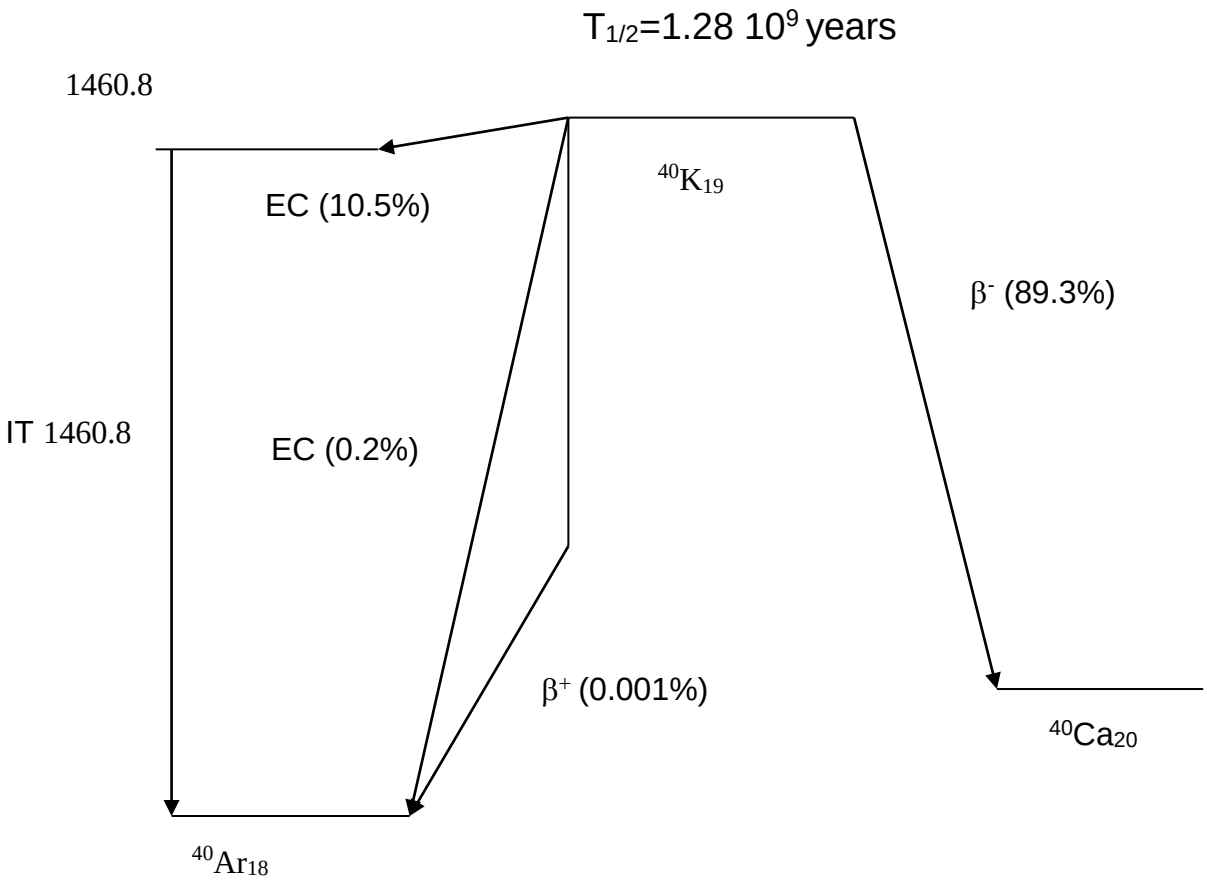


Fig. 1. Decay of the naturally occurring radio-isotope ^{40}K

Figure 1 shows the decay modes of K_{19}^{40} – isotope decaying by β_+ β_- and the capturing of an electron.

The transformation theory states that the number of atoms of a radioactive element which decay at any instant is proportional to the number of present atoms. In mathematical form it becomes:

$$\frac{dN}{dt} = -\lambda \cdot N \qquad (1)$$

where: dN/dt - is the instantaneous rate of decay – activity,

λ - is the decay constant (characteristic of the radioactive substance), a probability to undergo a nucleus transmutation,

N – is the number of atoms present.

The minus sign indicates that the number of atoms is decreasing.

If N_0 is the number of atoms originally present then the solution of the formula (1) is:

$$N = N_0 \cdot e^{-\lambda \cdot t} \qquad (2)$$

where: e = 2.72 is the base of Naperian logarithms,

t – decay time,

N - number of atoms present.

Half-life is the time for half of the nuclei in a sample to decay. It is used as an indication of the rate of decay. Half-life ranges from a fraction of second for some (man-made) isotopes to 4510 million years for Uranium 238. The radioactive materials used in diagnostic medicine have half-lives ranging from a few hours to a few weeks.

The relation between the decay constant, λ, and the half-life, $T_{1/2}$, is established as follows.

For a time equal to the half-time the equation (2) becomes:

$$\frac{N_0}{2} = N_0 \cdot e^{-\lambda \cdot T_{1/2}} \qquad (3)$$

then:
$$\frac{1}{2} = e^{-\lambda \cdot T_{1/2}} \qquad (4)$$

and:
$$\lambda \cdot T_{1/2} = \log_e 2 = 0.693 \qquad (5)$$

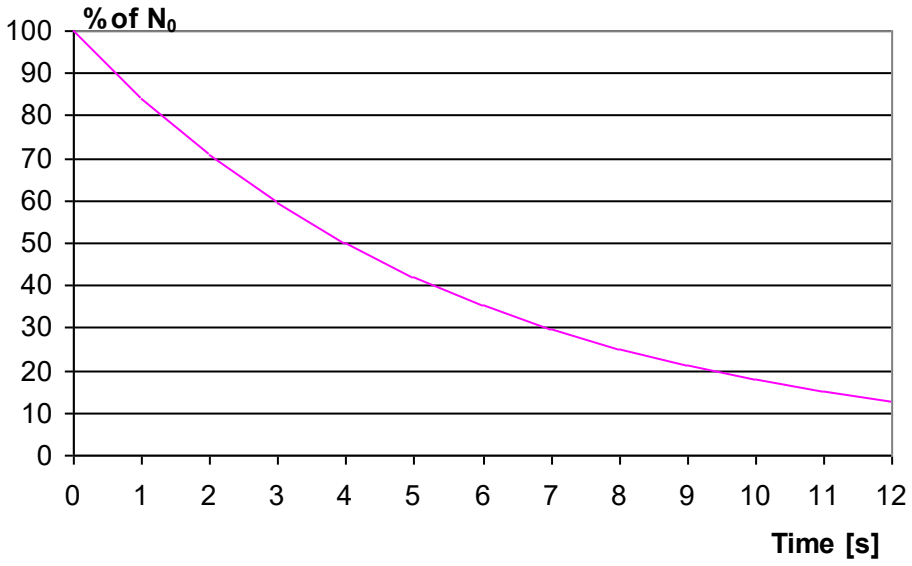


Fig. 2. Decay of radioactive substance ($T_{1/2}$ = 4s)

Figure 2 shows the example of percentage changes of the numbers of nuclei (N) as a function of decay time (t). In the first 4 seconds ($T_{1/2}$) will decay 50% of N_0, in next 4 seconds - 50% of 50% of N_0 etc....

After 10 half-lives the initial number of nuclei (N_0) is reduced by a factor of 1000.

Effective half-life

In medicine and biology to describe a process of losing the radioisotopes from a body not only in radioactive decay, but by biological processes too, we use the term effective half-life. The effective half-life (T_{eff}) is obtained by combining the biological half-life (T_b) and the radioactive (physical) half-life (T_p) according to the formula:

$$\frac{1}{T_{eff}} = \frac{1}{T_b} + \frac{1}{T_p} \qquad (5a)$$

From *Formula 5* we have:

$$\lambda \cdot T_{1/2} = \log_e 2 = 0.693$$

so: $$\frac{1}{T_{1/2}} = \frac{\lambda}{0.693}$$

then: $$\frac{\lambda_{eff}}{0.693} = \frac{\lambda_b}{0.693} + \frac{\lambda_p}{0.693}$$

and, finally: $\lambda_{eff} = \lambda_b + \lambda_f$ (5b)

The effective half-life is shorter than either the biological or physical half-life. This happens because both processes are depleting the supply of the radionuclide.

Radioactive equilibrium

If a substance is radioactive, its disintegration product is often radioactive, so that a radioactive series is built up. There are three main natural radioactive series: the thorium series, the uranium-radium series and the actinium series.

The other series, man-made, is the plutonium series. Each series ends at stable isotope of lead.

When a mother and a daughter substance are in radioactive equilibrium, there are as many atoms of the mother substance as of the daughter substance disintegrating per second. If N_1 and N_2 are the numbers of atoms of the two substances present, and λ_1 and λ_2 are their decay constants, then radioactive equilibrium is when:

$$\lambda_1 \cdot N_1 = \lambda_2 \cdot N_2 \qquad (6)$$

This means that the number of atoms of more quickly decaying substance (which has the greater value of λ) is less than that which decays more slowly.

The activity A is the disintegration rate of a radioactive material (*Formula 1*). It can be expressed as:

$$A = \lambda \cdot N = \frac{\ln 2}{T_{1/2}} \cdot N \qquad (7)$$

where: λ - the decay constant [s^{-1}]

N – number of atoms in the sample

$T_{1/2}$ – half-life [s]

It is measured in Becquerels [Bq] defined as follows:

1 Bq = 1 disintegration per second [1 Bq = 1 s^{-1}]

An earlier unit of activity was 1 Curie [Ci] defined by the following relationship:

1 Ci = $3.7 \cdot 10^{10}$ disintegrations per second

One gram of radium Ra-226 has $3.7 \cdot 10^{10}$ disintegrations per second (approximately) so its activity is 1 Ci.

If there are *n* moles in the sample, then the number of atoms in the sample is $N = n \cdot N_A$ where $N_A = 6.023 \cdot 10^{23}$ [mol^{-1}] – Avogadros number - is the number of atoms in a mole.

The activity of the sample is: $A = \lambda \cdot n \cdot N_A = \frac{\ln 2}{T_{1/2}} \cdot n \cdot N_A \qquad (8)$

Calculating activity on the basis of measurements

The results of measurements of the radioactive samples (frequency of pulses I) depend on many factors: the efficiency of the detector used, the kind and energy of the emitted radiation, the shape of both the detector and the sample, the density of the sample and others. When we are to calculate the activity of the sample, we should use the standard activity (A_s) for determining efficiency calibration of the to the detector. If both samples, the standard and the measured sample, are similar with reference to the above mentioned factors, we can use the following formula:

$$A_x = \frac{I_x}{I_S} \cdot A_s \qquad (9)$$

where: A_x – the activity of the measured sample (unknown) [Bq]

A_s – the activity of the standard (known) [Bq]

I_x – the frequency of pulses from the measured sample [pulses per sec]

I_s – the frequency of pulses from the measured standard [pulses per sec]

Ionising radiation has sufficient energy to ionise atoms or molecules. An atom is ionised when one or more electrons are separated from the atom. Binding energy of electrons is more than 10 eV (one electron-volt is the energy gained by a particle having one elementary charge /1e = $1.6 \cdot 10^{-19}$C/ moving between two surfaces with a potential difference of one volt (1V) in vacuum).

The principal kinds of ionising radiation are:

a/ gamma rays and X-rays

b/ beta particles (high speed electrons or positrons)

c/ alpha particles (high speed He-4 nuclei)

d/ neutrons (uncharged particles, mass nearly equal to H-1 atoms)

These kinds of radiation differ in: a/ methods of production

b/ interaction with atoms

c/ penetration ability through matter

Gamma rays and X-rays consist of tiny packets of energy known as photons which travel with the speed of light. They have identical properties and differ by the mechanisms which produce them. They have no mass or electric charge.

Alpha particles, beta particles and neutrons are basic components of the atoms and all possess mass.

The energy carried by ionising radiation is measured in units of kilo-electron-volts (keV) and mega-electron-volts (MeV) and is much greater than the amount of energy necessary to cause ionisation of an atom, therefore the ionising radiation produces a large number of ionisations.

Radiation is emitted by all radioactive substances. The decay of radioactive atoms is the source of gamma rays, some X-rays, alpha and beta particles. Neutrons are released in the process of interaction of high energy rays or particles with atomic nuclei, and when the nuclei of certain elements are caused to split (nuclear fission).

X-rays are produced by absorption of high-speed charged particles, usually electrons. The most common source of X-rays is an X-ray tube in which a beam of electrons is directed toward a metal target. Ultra-short X-rays (E > MeV) are produced with the use of accelerators producing high-energy electrons (E ~ dozens MeV).

Dosimetry

Exposure – is the ability of the radiation beam to ionise air.

$$X = \frac{\Delta Q}{\Delta m} \qquad \frac{charge\ of\ 1\ sign}{mass\ of\ air} \qquad (10)$$

when: X – exposure [$C \cdot kg^{-1}$]

ΔQ – charge of 1 sign ions occurs at the site of energy transfer of the photon beam in air [C]

Δm – mass of air [kg]

It is a historical quantity. By convention, applied only to photon beams. Practical to measure only for E < 3 MeV.

Pre-SI unit of exposure – 1 Roentgen (R) = $2.58 \cdot 10^{-4}$ C kg^{-1}

Absorbed dose – is the amount of energy absorbed per unit of mass of absorber.

$$D = \frac{\Delta E}{\Delta m} \qquad (11)$$

where: D – absorbed dose [Gy]

ΔE – energy of radiation imparted to element of irradiated material [J]

Δm – the mass of the element of material [kg]

The unit of absorbed dose is the joule per kilogram, it is called gray [Gy] (1 Gy = 1J/1kg).

Most radiations exposures cause different absorbed doses in different parts of the human body. Absorbed doses from different types of radiation have different biological effectiveness, and the organs and the tissues in the body have different sensitivities.

It is useful to combine the absorbed doses from different types of radiations to provide a further quantity called equivalent dose.

Equivalent dose – in a human tissue or organ is the absorbed dose weighted by a radiation weighting factor Q that ranges from 1 for low LET radiation to 20 for alpha particles. The unit of equivalent dose is called sievert (Sv).

$$H_t = D_t \cdot Q \qquad (12)$$

where: H_t – the equivalent dose [Sv]

D_t – the absorbed dose [Gy]

Q – radiation weighting factor (earlier Quality Factor)

Various organs and tissues in the body differ in their response to exposure to radiation. To allow for this the effective dose is used.

Effective dose – is the sum of equivalent dose in each tissue or organ multiplied by a tissue weighting factor H_t over the whole body. The unit of effective dose is sievert (Sv).

$$H_{ef} = \sum_t H_t \cdot w_t \qquad (13)$$

where: H_{ef} – the effective dose [Sv]

H_t – the equivalent dose [Sv]

w_t – the tissue weighting factor

The effective dose is an indicator of the total detriment due to stochastic effects in the exposed individual and his or her descendants.

The tissue weighting factors for some human organs:

reproductive organs – 0.25, red marrow – 0.12, stomach – 0.12, lungs – 0.12, liver – 0.05, thyroid – 0.03, skin – 0.01, surface of bones – 0.01.

Dose rate – is a dose divided by a time of irradiation (the formula includes all kinds of doses), for example:

$$\dot{D} = \frac{D}{\Delta t} \quad (14)$$

where: $\dot{D}$ - absorbed dose rate [Gy s^{-1}]

D – absorbed dose [Gy]

Δt – time of irradiation [s]

Levels of exposure

1/ Exposures of natural sources

Table 2 shows typical average annual effective doses in adults from the principal natural sources. The cosmic ray dose rate depends on height above sea level and latitude – annual doses in areas of high exposure are about five times average. The terrestrial gamma-rays dose rate depends on local geology with a high level being about 10 times the average. The dose from radon and its decay products depends on local geology and housing constructions and use, with the dose in some regions being about 10 times the average.

Table 2. Annual effective doses to adults from natural sources

Source of exposure	**Annual effective dose [mSv]**
Cosmic rays	0.39
Terrestrial gamma rays	0.46
Radionuclides in the body except radon Rn-222	0.23
Radon Rn-222 and its decay products	1.3
Total	2.4

2/ Terrestrial radiation

In terrestrial materials there are some long-life radionuclides with half-lives comparable with the age of the earth. In terms of dose the principal radionuclides are: K -40 (half-life:$1.28 \cdot 10^9$ years}, Th-232 (half-life:$1.41 \cdot 10^{10}$ years) and U-238

(half-life:$4.47 \cdot 10^9$ years). The thorium and uranium radionuclides head the series of several radionuclides which contribute to human exposure. Exposure to gamma rays from natural radionuclides occurs outdoors and indoors. The absorbed dose rate (outdoors) per unit activity concentration of radionuclides can be calculated as follows:

$$\dot{D} = C_K f_K + C_{Th} f_{Th} + C_{Ra} f_{Ra} \qquad (15)$$

where: $\dot{D}$ - the absorbed dose rate (nGy h^{-1})

f_K - the K-40 dose factor, estimated as 0.0414 (nGy h^{-1} per Bq kg^{-1})

f_{Th} - the Th-232 series dose factor, estimated as 0.623 (nGy h^{-1} per Bq kg^{-1})

f_{Ra} - the Ra-226 (sub series of U-238) dose factor, estimated as 0.461 (nGy h^{-1} per Bq kg^{-1})

C_K, C_{Th}, C_{Ra} – the value of specific activity, respectively (Bq kg^{-1})

When comparing the indoor to outdoor averages, it is seen that the overall effect of surrounding building materials is to increase the dose rate by 50%.

The absorbed dose (D) in the time (t) per unit activity concentration of radionuclides can be calculated as follows:

$$D = \dot{D} \cdot \Delta t \qquad (16)$$

where: $\dot{D}$ - The absorbed dose rates per unit of mass (nGy h^{-1} kg^{-1})

Δt – the time of “irradiation”(h)

3/ Medical exposures

Radiation is used in diagnostic examinations and in treatments.

X-ray procedures account for about 90% of the radiation dose to the population from all artificial sources of radiation. In all of these procedures exposure is to a part of the body, not to the whole body.

Typical levels of patient effective dose from common X-ray examinations are expressed in *Table 3.*

Table 3. Typical levels of patient effective dose from X-ray examinations.

Examination	**Effective dose [mSv]**	**Equivalent period of natural radiation**
Extremities (arms, legs etc)	<0.01	1.5 days
Dental	0.02	3 days
Chest	0.04	1 week
Skull, mammography	0.1	2 weeks
Hip	0.3	2 months
Abdomen	1.4	8 months
CT head	1.8	10 months
Intravenous urography	4.6	2 years
CT chest	8.3	4 years

Medical examinations providing information on the functioning of specific organs are also performed by administering to patients small amounts of radioactive materials (more than 90% of medical procedures employ Tc-99m) - the average effective dose is of the order 1 mSv.

The equivalent dose to individual patients undergoing radiotherapy is very much higher than in diagnosis – typically about 60 Sv to the tumour-bearing tissue over a period of 6 weeks.

Lethal effective dose is about 7 Sv (whole body exposition).

PRACTICAL PART

EXPERIMENTAL PART A

Objective

you will calculate activity of an unknown sample using a standard

you will calculate the mass of previously found radioisotope in a sample

Materials

system for measuring gamma rays with scintillation well-type detector and lead shield

standard Cs-137 sample

unknown sample (polluted with Cs-137)

electronic calculator

Procedure *(Table 1)*

1/ Measure the background of a measuring system with mass equivalent of a sample in 5 minutes

2/ Measure the number of pulses from an unknown sample in 5 minutes

3/ Calculate activity of an unknown sample (use *Formula 9*)

4/ Calculate the number of atoms of Cs-137 in unknown sample (use *Formula 7*)

$$A = \lambda \cdot N \rightarrow N = \frac{A}{\lambda}$$

5/ Calculate the mass of Cs-137 in the sample

We calculate the mass of Cs-137 in the sample with the use of Avogadros number.

$N_A = 6.023 \cdot 10^{23}$ [mol^{-1}] – Avogadros number - is the number of atoms in a mole.

The mole of Cs-137 is 137grams, so the mass of $6.023 \cdot 10^{23}$ atoms of Cs-137 is 137 grams.

$$m = \frac{137 \cdot N}{N_A} \text{ (grams)}$$

Data and observations

Table 1

Activity of the standard	A_S (Bq)	5000
Half-life of Cs-137	years	30.07
Decay constant of Cs-137	s^{-1}	$7.17 \cdot 10^{-10}$
Result of measuring or calculation	**Symbol or formula and unit**	**Value**
Background (measured)	N_B (pulses per 5 minutes)	
Standard (measured)	N_S (pulses per 5 minutes)	
Sample (measured)	N_X (pulses per 5 minutes)	
Net standard	$N_S - N_B$ (pulses per 5 minutes)	
Net sample	$N_X - N_B$ (pulses per 5 minutes)	
Frequency of pulses per sec - net standard	$I_S = \frac{N_S - N_B}{5 \times 60}$ (s^{-1})	
Frequency of pulses per sec - net sample	$I_x = \frac{N_X - N_B}{5 \times 60}$ (s^{-1})	
Activity (A_X) of the sample	$A_X = \frac{I_X}{I_S} \cdot A_S$ (Bq)	
Number (N) of atoms of Cs-137 in the sample	$N = \frac{A_X}{\lambda}$	
Mass (m) of Cs-137 in the sample	$m = \frac{137 \cdot N}{N_A}$ (g)	

Questions and conclusions

EXPERIMENTAL PART B

Objective

you will calculate activity of natural radioisotopes Ra-226, Th-232 and K-40 in a sample

you will calculate an absorbed dose rate from gamma rays outdoors

you will calculate an annual effective dose from gamma rays indoors. Assume that the examined material was used for building the house

Materials

sample for measurement (raw building material)

Marinelli container

mass equivalent of sample in Marinelli container

scale

calibrated system for measuring gamma rays with NaI scintillation detector and 3-channel analyser MAZAR

lead shield

electronic calculator

Procedure *(Table 2)*

1/ Find the net mass of the sample using the scales

1/ Measure the background of measuring system with mass equivalent of sample in 2000 seconds

2/ Measure the sample in 2000 seconds

3/ Calculate the specific activity of Ra-226, Th-232 and K-40 in the sample with the use of the MAZAR analyser

4/ Calculate the annual absorbed dose from gamma rays outdoors (*Formula 15 and Formula 16*)

5/ Calculate the annual absorbed dose from gamma rays indoors (50% more than outdoors)

Data and observations

Radioisotope	**Dose factor f** (nGy h^{-1} per Bq kg^{-1})
K_{40}	0.0414
Th_{232}	0.623
Ra_{226}	0.461

Table 2

Result of measuring or calculation	**Symbol or formula and unit**	**Value**
Mass of container	m_c (kg)	
Gross mass of sample	m_g (kg)	
Net mass of sample	m (kg)	
Background (measured)	N_B (pulses per 2000 sec)	
Pulses from sample (measured)	N_K (pulses per 2000 sec)	
	N_{Th} (pulses per 2000 sec)	
	N_{Ra} (pulses per 2000 sec)	
Specific activity of sample (calculated)	C_K (Bq/kg	
	C_{Th} (Bq/kg)	
	C_{Ra} (Bq/kg)	
The absorbed dose rate (outdoors)	$\dot{D} = C_K f_K + C_{Th} f_{Th} + C_{Ra} f_{Ra}$ (nGy/h)	
The annual absorbed dose (outdoors)	$D = \dot{D} \cdot \Delta t$ (μGy)	
The annual absorbed dose (indoors)	$D_{in}=1.5 \cdot D$ (μGy)	

Questions and conclusions

The date	**Student's name and surname**	**Lab assistant signature**

3.2 Lab Exercise

INTERACTION OF PHOTONS AND CHARGED PARTICLES WITH MATTER

THEORY

When the ionising radiation passes through matter, it loses energy in radiation, ionisation and photonuclear interactions. The following categories of radiation are of interest to us: gamma (γ) and X-rays, charged particles (alpha (α) particles, beta (β) particles and protons) and neutrons.

Gamma rays and X-rays are absorbed by collisions with electrons to which their energy is transferred (photoelectric effect and Compton scattering) and in the reaction $h\nu \rightarrow e^- + e^+$, which can take place in the Coulomb field of a nucleus (pair production). These electrons subsequently lose their energy in ionising events, which are relatively widely spaced.

Photoelectric effect is the process in which a photon collides with a bound electron causing ejection of the electron from the atom. The photon is absorbed by the atom.

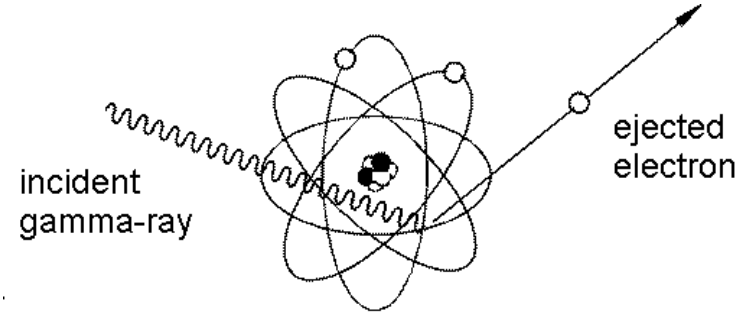


Fig.1. Photoelectric effect.

The electron energy (E_e) is equal to photon energy ($h\nu$) minus the binding energy (E_B)

$$E_e = h\nu - E_B \qquad (1)$$

The presence of the atom is required to conserve momentum because it is impossible to balance both momentum and energy for a photon completely absorbed by a free electron. After a photoelectric interaction, the atom is left in an

excited state due to vacancy created when the electron is ejected. The atom relaxes by electronic transition from a higher orbital to the vacant orbital. The energy difference can appear as fluorescence or a characteristic x-ray photon, or it can be transferred to a higher orbital electron which then is ejected from the atom as an Auger electron.

For tissues, since the binding energies are small, almost all the energy goes to the photoelectron. The photoelectric probability increases strongly with Z of material and decreases at higher energies.

In incoherent Compton scattering the photon collides with a (free or weakly bound) electron, losing energy to the electron and scattering in a different direction.

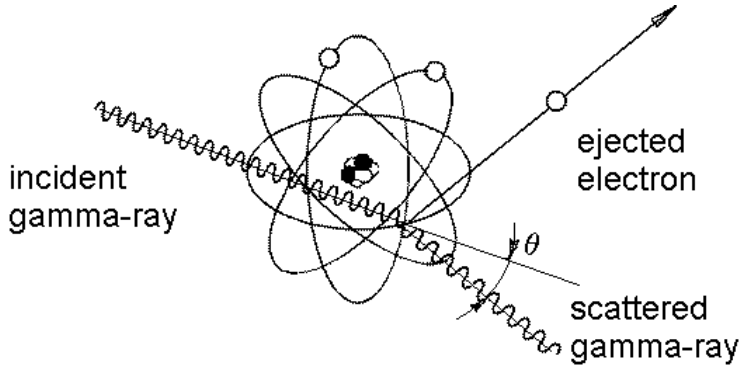


Fig. 2. The Compton scattering.

The energy of electron and the energy of scattered photon are given by:

$$h\nu' = h\nu\left[1 + (h\nu / m_e c^2)(1 - \cos\theta)\right]^{-1} \qquad (2)$$

$$E_e = h\nu - h\nu' \qquad (3)$$

where: $h\nu$ - incident photon energy

$h\nu'$ - scattered photon energy

E_e - electron energy

m_e - electron mass

c - velocity of light

θ - angle between incident and scattered gamma ray directions

For θ = 0 there is no energy transfer, while $\theta = 180^0$ there is maximum energy transfer. The electron is never scattered backwards.

When bound electrons are considered, the incoherent scattering cannot take place unless the energy given to the electron exceeds binding energy. Therefore

incoherent scattering is prohibited for small incident photon energy, for small θ, with participation by inner shell electrons being lost first because of their higher binding energy.

Pair production is the process by which a positron – electron pair is created when a photon interacts in the Coulomb field of a nucleus. At least 1.022 MeV = $2m_ec^2$ of a photon energy is required to create a positron - electron pair.

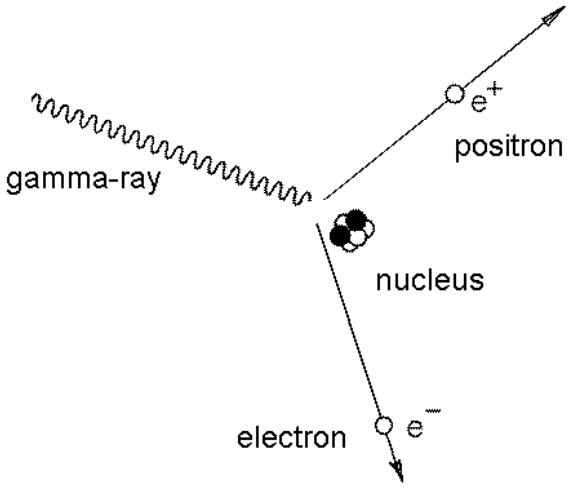


Fig. 3. Pair production.

Energy conservation: $E_e+E_p = h\nu + 2m_ec^2$

where: E_e, E_p – electron and positron energy

$h\nu$ - photon energy

$2m_ec^2$ – energy equivalent of mass of electron and positron

At rest the positron annihilates with electron creating two photons of 0.511 MeV emitted at 180°.

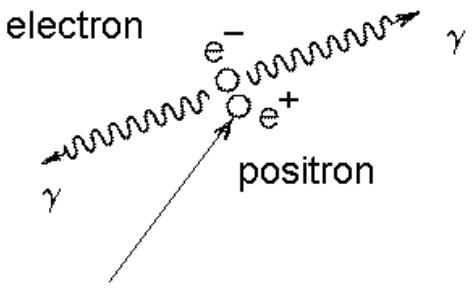


Fig. 4. Annihilation electron with positron

The pair production can also take place in the Coulomb field of an electron, termed historically “triplet” production, since there are three particles in motion (the two that are created and the original electron). The threshold energy for triplet is $4m_ec^2$. Triplet production is a small fraction of the pair production and is usually included with it.

Table 1 shows the distribution of the types of photon interactions in water in function of energy of photons.

Table 1. Types of photon interaction in water

$h\nu$	% energy transferred		
(keV, MeV)	Compton	Photo	Pair
10 keV	0.1	99.9	0.0
30	6.8	93.2	0.0
50	37.2	62.8	0.0
100	89.6	10.4	0.0
200	99.0	1.0	0.0
500	99.9	0.1	0.0
1 MeV	100.0	0.0	0.0
1.5	99.9	0.0	0.1
2	99.3	0.0	0.7
5	89.6	0.0	10.4
10	71.9	0.0	28.1
20	49.3	0.0	50.7
50	24.6	0.0	75.4
100 MeV	13.3	0.0	86.7

The probability of interacting with matter in one of these three processes can be expressed as an attenuation coefficient μ. The attenuation coefficient for a beam of gamma rays is related to the number of gamma rays removed from the beam, either by absorption or scattering. For all the three processes the total attenuation coefficient μ is the sum of the three partial attenuation coefficients:

$$\mu = \mu_{photo} + \mu_{Compt} + \mu_{pair} \qquad (4)$$

The attenuation coefficient is often called absorption coefficient.

Figure 5 shows the relative importance of three major types of gamma-ray interaction with the matter. The lines show the values of atomic number *Z* and gamma ray energy $h\nu$ for which the two neighbouring effects are just equal.

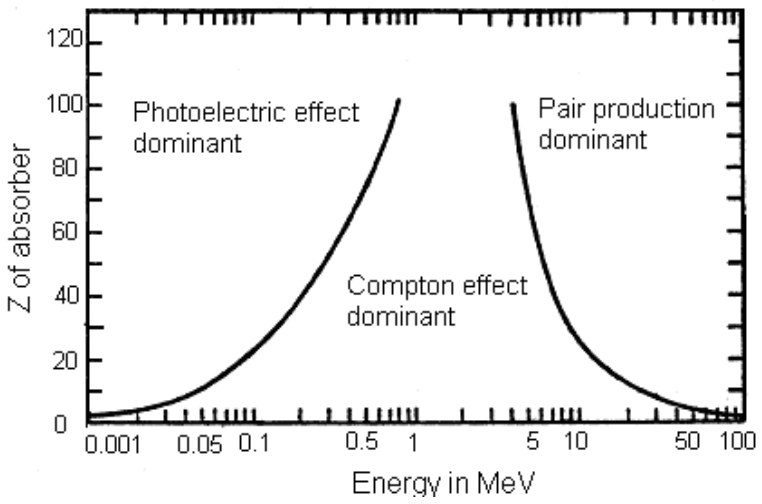


Fig. 5. The relative importance of three major types of gamma-ray interaction with matter

The law of attenuation

The number of photons removed from a beam "dn" of "n" primary photons on the distance-travelled "dx" is:

$$dn = -n \cdot \mu \cdot dx \qquad \text{or} \qquad dI = -I \cdot \mu \cdot dx \quad (5)$$

which integrates to:

$$n = n_0 \cdot e^{-\mu \cdot x} \qquad \text{or} \qquad I = I_0 \cdot e^{-\mu \cdot x} \quad (6)$$

where: n – final number of photons or I - final beam intensity

n_0 – an initial number of photons or I_0 - an initial beam intensity

μ - the total attenuation coefficient (m^{-1})

x – thickness of material (m)

For the <u>surface density</u> defined as "$\rho \cdot x$" the formula (6) is:

$$n = n_0 \cdot e^{-\frac{\mu}{\rho}(\rho x)} \qquad \text{or} \qquad I = I_0 \cdot e^{-\frac{\mu}{\rho}(\rho x)} \quad (7)$$

where: n – final number of photons, (I – final beam intensity)

n_0 – initial number of photons, (I_0 – initial beam intensity)

$\mu \cdot \rho^{-1}$ - mass attenuation coefficient ($m^2\ kg^{-1}$)

ρ - density of absorber ($kg\ m^{-3}$), $\rho \cdot x$ – surface density ($kg\ m^{-2}$)

Fig. 6. Percentage changes of intensity of photons γ in function of travelled distance in matter

It is useful to define a half-thickness $d_{1/2}$. It is the thickness for which the intensity

of a beam of parallel gamma rays given energy E is reduced by factor 2.

It is easily verified that:

$$d_{1/2} = \frac{\ln 2}{\mu} \qquad (8)$$

where: $d_{1/2}$ – half-thickness (m)

μ - the linear absorption coefficient (m^{-1})

ln2 = 0.693 – Napierian logarithm of 2

Gamma rays and X-rays will penetrate considerably greater distances than electrons possessing the same energy. Photons used in X-ray diagnosis will travel several centimetres into the body before an electron occurs. Some will penetrate the entire thickness of the human body without the collision and can therefore produce an anatomical image on a screen or recording device. The fraction of the photons that reach a given depth in the body increases as the photon energy increases.

The linear energy transfer (LET) is defined as the average amount of energy transferred to an absorber per unit distance along the path of radiation. This is also known as “stopping power”.

The LET is expressed as:

$$LET = \frac{dE}{dx} \qquad (8)$$

where: LET – linear energy transfer [keV micron^{-1}]

dE - energy loss [keV]

dx – distance travelled by radiation [micron = 10^{-6}m]

High values of LET signify that ionisations are produced much closer together in an absorber than is the case with low values of LET. LET is an important radiation characteristic because it determines the amount of damage produced by the absorption of a given amount of radiation energy.

Beta particles have energies up to few MeV. They lose not only their energy by ionisation and excitation of atoms, but also by emitting a continuous electromagnetic radiation called bremsstrahlung, which appears when a high energy electron is stopped in the Coulomb field of nuclei. The efficiency of bremsstrahlung (the probability for the electron to lose part of its energy by radiation) varies nearly as Z^2. The total amount energy dissipated under the form of bremsstrahlung is determined by the radiation yield, Y. Radiation yield is the average fraction of the electron’s kinetic energy that an electron radiates as bremsstrahlung if completely stopped. It is given by:

$$Y = \frac{A}{A+1} \qquad (9)$$

where: $A = 6 \cdot 10^{-4} \cdot Z \cdot E_k$

Z – the atomic number of the stopping material

E_k – the kinetic energy of the electrons (MeV)

To keep the bremsstrahlung low, low Z materials should be used for stopping beta particles. For example: a 2 MeV electron loses a fraction of Y=0.015 (1.5%) in Al (Z=13) and Y=0.09 (9%) in Pb (Z=82) by bremsstrahlung. The positron (β^+ particle) at rest will annihilate with an electron resulting in the emission of two gamma ray photons of 511 keV emitted in coincidence at 180^o of each other.

Electrons and beta particles will penetrate distances in the body ranging from a small fraction of millimetre up to a few centimetres depending on their energy. A 1 MeV electron has a range with water (or soft tissues) of 0.4 cm.

Alpha particles have a strong ionisation power and thus a very short range. They deposit their energy in very short distances and they are said to have a high LET (approximately 200 keV per micron in water). Alpha particles with a few MeV of energy are absorbed in:

- a few centimetres of air
- a sheet of paper
- a few contiguous cells

They will not penetrate the skin and only constitute a hazard in the case of intake of the radioactive material inside the body.

When the kinetic energy of alpha particle decreases to about 1 MeV, the particle acquires two electrons and becomes helium He^4 atom (and stops in a short distance after a few collisions).

Fast neutrons are also classified as high LET radiation because their interactions with matter produce heavy charged particles (fast protons) that have a high LET (approximately 40 keV per micron in water). However, the neutrons themselves carry no electrical charge, so they can travel relatively large distances between interactions and can penetrate many centimetres of the body – as do gamma rays and X-rays.

PRACTICAL PART

EXPERIMENTAL PART A

Objective

you will find a half-thickness ($d_{1/2}$) and calculate the attenuation coefficient (μ) and mass attenuation coefficient (μ_m) of gamma rays emitted by Co-60 (E_{av}=1.25MeV) for two absorbers: zinc and lead

Materials

- NaI scintillation gamma detector connected to pulse counting system
- lead shield
- source of gamma-rays (Co-60)
- absorbing discs (made of zinc and lead)
- electronic calculator

Procedure

1/ Measure the background of a measuring system in 5 minutes. Calculate the background for 1 minute. Write the results in *Table 1*

2/ Measure the number of pulses from uncovered source in 1 minute (3 times). Write the results in *Table 2*

3/ Measure the number of pulses from source covered in zinc absorber in 1 minute (3 times). Use different number of absorbing discs from 1 to 5. Write the results in *Table 2*

4/ Draw a chart of frequency of pulses as a function of the thickness of the absorber *(Chart 1)*

5/ Find a half-thickness $d_{1/2}$ of zincum (use the *Chart 1*)

6/ Calculate the attenuation coefficient (μ) and mass attenuation coefficient (μ_m). Write the results in *Table 3*

7/ Measure the number of pulses from source covered in lead absorber in 1 minute (3 times). Write the result in *Table 2*

8/ Calculate the attenuation coefficient (μ), the mass attenuation coefficient (μ_m) and half-thickness $d_{1/2}$ of lead. Write the results *in Table 4*

Data and observations

Table 1

Background N_B (pulses/5 minutes)	Background I_B (pulses /1 minute)

Table 2

Thickness of the absorber d (10^{-3} m)	Frequency of pulses I (minute^{-1})			Mean frequency of pulses I_M (minute^{-1})	Net mean frequency of pulses ($I_M - I_B$) (minute^{-1})
	I_1	I_2	I_3		
uncovered source					
Zn $_{1disc}$ =					
Zn $_{2discs}$ =					
Zn $_{3discs}$ =					
Zn $_{4discs}$ =					
Zn $_{5discs}$ =					
Pb $_{1disc}$ =					

Chart 1

$d_{1/2}$=.....(m)

$$\mu = \frac{\ln 2}{d_{1/2}} = \frac{0.693}{d_{1/2}} = \ldots\ldots\ldots\ldots\ldots\ldots (m^{-1})$$

$$\mu_m = \frac{\mu}{\rho} = \ldots\ldots\ldots\ldots\ldots\ldots (m^2 kg^{-1})$$

Table 3

Density of zinc ρ (kg m^{-3})	Attenuation coefficient μ (m^{-1})	Mass attenuation coefficient μ (m^2 kg^{-1})
$7.19 \cdot 10^3$		

Lead (Pb) Thickness of lead absorber d = ... (m)

$$\mu = \frac{\ln \frac{I_0}{I_x}}{d} = \ldots\ldots\ldots\ldots\ldots\ldots (m^{-1})$$

$$\mu_m = \frac{\mu}{\rho} = \ldots\ldots\ldots\ldots\ldots\ldots (m^2 kg^{-1})$$

$$d_{1/2} = \frac{\ln 2}{\mu} = \ldots\ldots\ldots\ldots\ldots\ldots (m)$$

Table 4

Density of lead ρ (kg m^{-3})	Attenuation coefficient μ (m^{-1})	Mass attenuation coefficient μ (m^2 kg^{-1})	Half-thickness of lead $d_{1/2}$ (m)
$13.6 \cdot 10^3$			

Questions and conclusions

EXPERIMENTAL PART B

Objective

you will calculate the attenuation coefficient (μ) and mass attenuation coefficient (μ_m) of beta particles emitted by Sr-90 for three absorbers: aluminium, copper and polyester (the radiographic film base)

Materials

- detector connected to pulse counting system
- lead shield
- radioactive source of β radiation
- micrometer screw
- foils of absorbers: aluminium, copper and polyester
- electronic calculator

Procedure

1/ Measure the background of a measuring system in 5 minutes. Calculate the background for 1 minute. Write the result in Table 5

2/ Measure the number of pulses from uncovered source in 1 minute (3 times). Write the results in Table 6

3/ Measure the thickness of absorbers and the number of pulses from source covered in each absorber: Al, Cu and polyester) in 1 minute (3 times). Write the results in Table 6

4/ Calculate the attenuation coefficient (μ) and the mass attenuation coefficient (μ_m). Write the results in Table 7

Data and observations

Table 5

Background N_B(pulses/5 minutes)	Background I_B(pulses /1 minute)

Table 6

Thickness of the absorber d (10^{-3} m)		Frequency of pulses I (minute^{-1})			Mean frequency of pulses I_M (minute^{-1})	Net mean frequency of pulses ($I_M - I_B$) (minute^{-1})
		I_1	I_2	I_3		
uncovered source						
Al						
Cu						
Polyester						

Table 7

Density of absorber ρ(kg m^{-3})		Attenuation coefficient $\mu = \frac{\ln \frac{I_0}{I_x}}{d}$ (m^{-1})	Mass attenuation coefficient $\mu_m = \frac{\mu}{\rho}$ (m^2 kg^{-1})
Aluminium	$2.7 \cdot 10^3$		
Copper	$9.96 \cdot 10^3$		
Polyester	$1.4 \cdot 10^3$		

Questions and conclusions

The date	**Student's name and surname**	**Lab assistant signature**

3.3 Lab Exercise

THE METHODS OF DETECTION OF IONISING RADIATION

THEORY

Gas filled detectors

The ability of atomic radiation to ionise a gas and provide a measurable electronic signal is used in gas counters. Gas counters were most popular in measuring radioactivity in the twentieth century because of their simplicity and flexibility of technology.

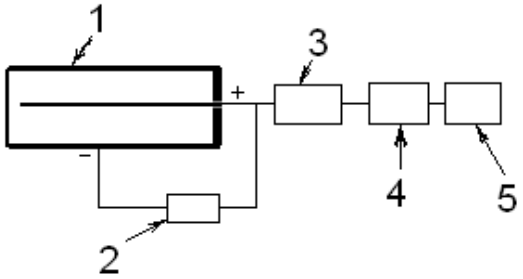


Fig. 1. Gas filled detector

A gas filled detector (*Figure1)* consists of a gas filled tube (1), a positive (+) anode wire and negative (-) cathode tube. Ionising radiation entering the tube, atoms of gas are ionised, producing positive and negative ions and electrons. If a potential is applied across the tube (voltage source – 2), the positive ions will migrate towards the cathode and electrons and the negative ions towards the anode (Coulomb attraction). The migration of electrons and ions to the electrodes result in a current flow or electrical charge, which can be amplified in an amplifier (3), analysed in the analyser of amplitude (4) and displayed in the scaler (5).

Only a fast charged particle (alpha, beta) can ionise gas inside the tube. Any neutral particles such as gamma ray or X-ray quanta and neutrons must first be converted into charged particles before detection can take place. This is done in the case of X-rays and gamma rays in three processes: photoelectric effect, Compton effect and pair production. Neutrons are converted by means of nuclear reactions in a component of the counter gas. A typical process is:

$$n_0^1 + He_2^3 \rightarrow p_1^1 + H_1^3 + h\nu$$

The number of ionising events inside the tube of gas detector depends on the electric field (and applied voltage).

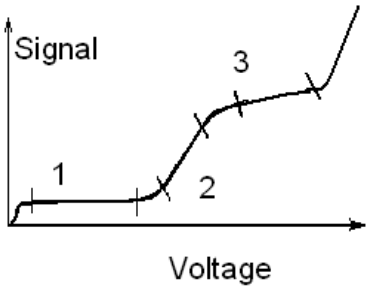


Fig. 2. The regions of the gas filled detectors.

Figure 2 shows how the amplitude of output signal changes with the increase of applied voltage. The electric field causes the electrons and ions created by the incident radiation to migrate to the collecting electrodes –ionisation (ion) chamber region **1**.

As the electric field is increased, the free electrons are accelerated and can achieve kinetic energy sufficient to cause further ionisations within the detector gas. The electrons freed by these ionisations are accelerated by the field and cause additional ionisations (gas amplification). First the number of secondary ionisation events is proportional to the number of primary ions -proportional counter region **2**. If the voltage is sufficiently high, each output pulse from the detector is the same for all energies of ionising radiation -Geiger-Müller counter region **3**. Gas amplification can provide a charge gain up to 10^6, which can give pulses of several volts. If the applied voltage is increased above the Geiger-Müller region, the gas goes into continuous discharge.

The Geiger-Müller counter needs suppressing of the secondary pulse caused when the large cloud of positive ions (excited noble gas) hits the cathode and releases electrons. This can be done by making the load resistor on the anode supply decrease the voltage or by using a few percent of a quench gas with lower ionisation potential whose charge exchanges with the noble gas ions and does not cause electron emission at the cathode. Chlorine is a commonly used quencher. Argon and helium are favourite filling gases. The time after each pulse until the quenching is complete (dead time) can be hundreds of microseconds, which limits the counter to low count rate applications. Geiger-Müller counters for measuring

particle's radiation are available with a thin mica window, permeable to the particles.

The applications of gas counters in bio-medicine:

- monitoring (dosimetry) and calibration of radioactive sources (ion chambers)
- detection of weak activities and contamination (Geiger-Müller counters)
- diagnostic imaging and autoradiography – still in research phase (with the use of the multi wire proportional counters MWPC):

a/ digital radiographic scanner

b/ angiography using an MWPC gamma camera

c/ Positron Emission Tomography PET

Scintillation detectors

In some crystals in the processes of energy loss radiation excites some atoms above the ground state, these atoms then decay back to the ground state. When this process emits photons we have scintillation.

In the scintillation detector the radiation interacting with a scintillator produces a pulse of light (proportional to the energy deposited within the crystal) which is converted to an electric pulse by a photomultiplier tube (PMT). The photomultiplier consists of a photocathode which converts light photons into low energy electrons, 10 or more dynodes that multiply the number of electrons striking them several times each and anode. The anode and dynodes are biased by a resistor dynode chain located in a plug-on tube base assembly. Total gain of the 12 stages of multiplier is 10^8. Next the pulse is amplified by preamplifier and amplifier and analysed by pulse height analyser. An amplitude of the final pulse is proportional to the number of photons and the energy of the measured radiation.

Complete assemblies of at scintillation detector are shown on *Figure 3*.

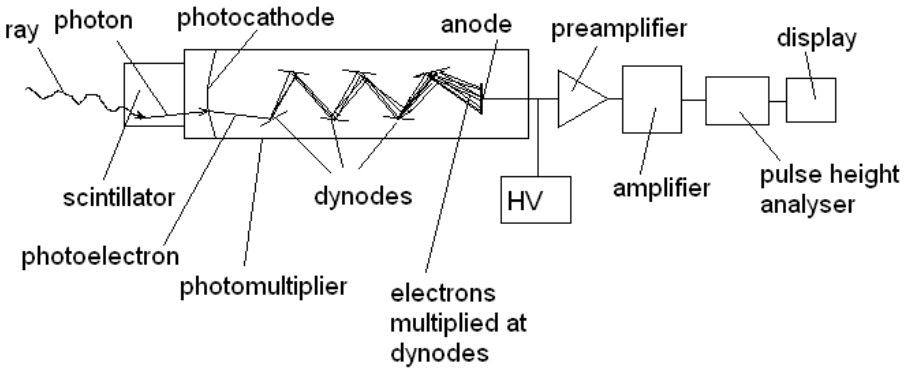
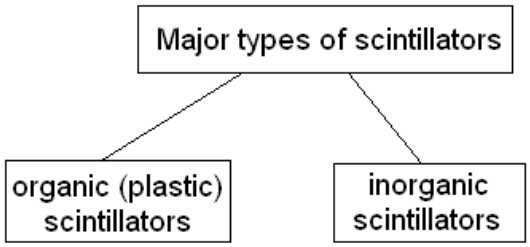


Fig.3. The scintillation detector and its assemblies

The properties of scintillation material required for good detectors are:

1) transparency 2) availability in large size

3) large light output proportional to rays energy.



The most frequently used inorganic scintillation crystal is sodium iodide doped with thalium - NaI(Tl). The different types are: ZnS, CsI(Tl). These crystals combine a good detection efficiency with a large light output for the detection of gamma rays.

Organic scintillators consist of a transparent host material (a plastic) doped with a scintillating organic molecule and are mostly used for the detection of beta and other particles. The others are: naphtalene, stilbene.

Liquid scintillators

Many samples include tritium and C^{14} - very important beta-emitting isotopes used in medical and biological researches and tests. They are sources of very low energy beta rays (19keV and 155keV) which are too low to detect with solid scintillators. The liquid scintillation detection involves mixing a liquid scintillator with the sample (processed in liquid form and transparent) and then observing the light pulses with photomultiplier tubes.

Semiconductor detectors (scintillation)

Another way to detect the scintillation light (instead of photomultiplier tube) is the use of silicon photodiode (PIN photodiodes are most widely used). The photodiode consists of a thin layer of silicon in which the light is absorbed and the free charge carriers (electron and holes) are created. When the photodiodes are optically coupled to a crystal a scintillation light pulse will generate a small charge pulse in the diode which can be observed after amplification. In contrast to photomultiplier tubes photodiodes do not require a high voltage but only a bias voltage of about 30 volts. Photodiodes are thin, rugged and insensitive to magnetic field. Due to the electronic noise which increases with an increasing surface area, the dimension of PIN photodiode is limited to a few square cm.

Applications of scintillation detectors in medicine:

- Gamma Cameras
- SPECT (Single Photon Emission Computed Tomography)
- PET (Positron Emission Tomography) - CT (Computed Tomography)

Semiconductor detectors

Semiconductor detectors are the solid detectors that operate like ionisation chambers, however the charge carriers are not electrons and ions as in the gas filled detectors, but electrons and holes (absences of the electrons). Major types of semiconductor materials used in construction of detectors are as follows: pure silicon (Si), high-purity germanium (Ge), germanium lithium-drifted (GeLi), and silicon lithium-drifted (SiLi).

Semiconductor detectors have a PIN diode structure in which the intrinsic region is created by depletion of charge carriers when a reverse bias is applied across the diode *(Figure 4)*. When photons interact within the depletion region, charge carriers (holes and electrons) are freed and are swept to their respective collecting electrode by the electric field. The resultant charge is integrated by a charge sensitive preamplifier and converted to a voltage pulse with an amplitude proportional to the energy of radiation.

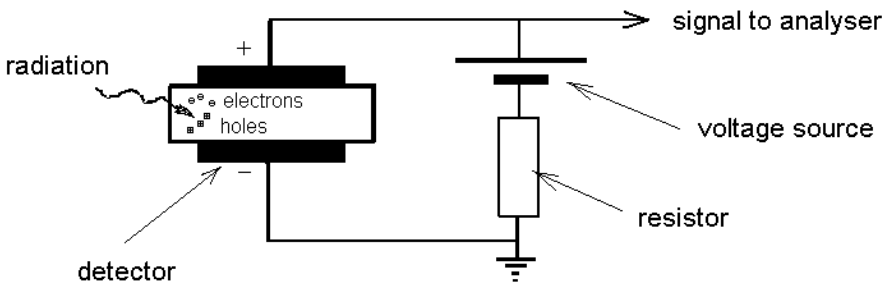


Fig.4. Semiconductor detector

Pulse height spectrometry

The basic principle of scintillation counting is that the light output of a scintillator is proportional to the deposited energy. Voltage pulses of a given height are counted following passage through a gamma ray spectrometer which consists of a preamplifier, an amplifier and a multichannel analyser (MCA). With a scintillation detector the energy of the incident radiation can be determined by measuring the pulse height distribution. The shape of a pulse height spectrum depends on the material of the scintillator, the dimensions of the crystal and the energy of radiation.

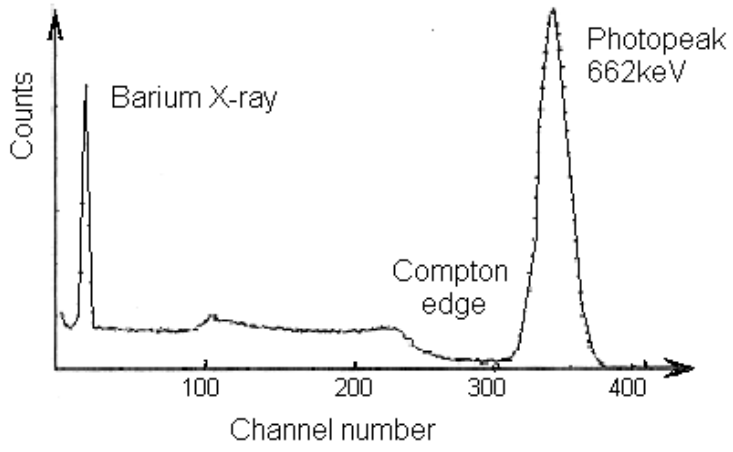


Fig. 5. Scintillation pulse height spectrum of Cs^{137} source - (NaI(Tl) crystal

The ability to discriminate between gamma rays with slightly different energy is characterised by the energy resolution which is defined as: the number of channels between two points at half the maximum intensity of the photopeak, divided by the channel number of the peak mid-point, multiplied by 100%.

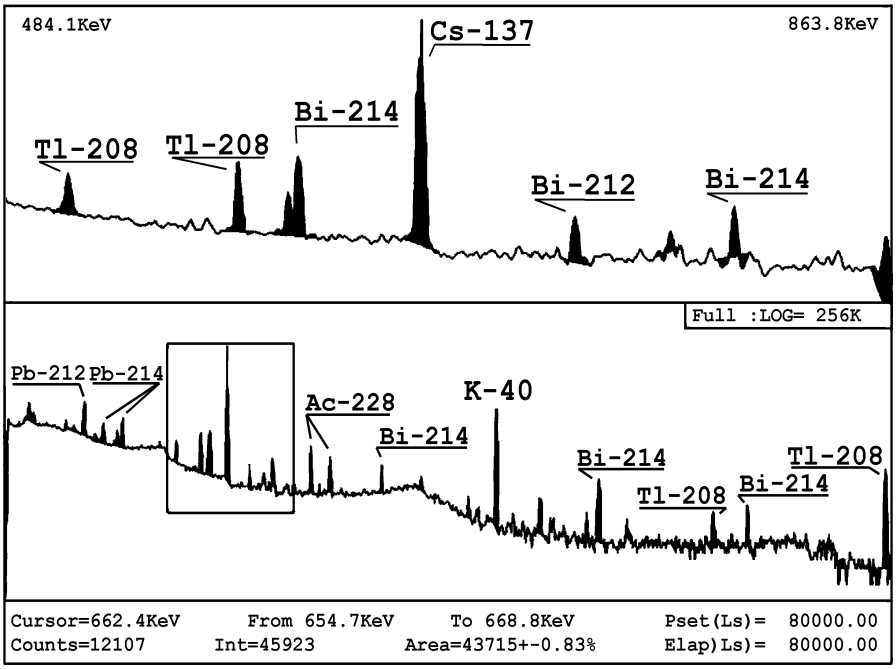


Fig. 6. Spectrum of multi-gamma rays source (semiconductor (Ge) detector, 15% relative efficiency) – print of S-100 interface software for analysis spectra

The best energy resolution *(Figure 6)* makes the semiconductor detectors excellent detectors for gamma spectroscopy – method of an acquisition and qualitative and quantitative analysis of gamma spectra for the identification of sources of gamma rays.

Medical radiation detectors

1/ Planar imaging in nuclear medicine – the Anger gamma camera

The Anger camera is most commonly used to image the distribution of radioactive tracer within human body. The device yields information regarding the energy and position of incident gamma photons. The data acquired can be used to form a single image of the distribution or a series of images which can be used to assess the time course of the tracer (static study and dynamic study).

The tracer consists of the pharmaceutical to which a radioactive material is chemically bonded (radiopharmaceutical). Many different radionuclides are used for this – but more then 90% of nuclear medicine procedures employ Technetium-99m, which is a gamma emitter (energy = 140keV, half-life = 6 hours).

The Anger camera *(Figure 7)* consists of a single crystal of NaI(Tl) (20-60 cm in size) and an array of photomultiplier tubes (30-150) coupled directly to the optical window of the crystal which are contained within a light-tight lead shield. The collimator which is positioned between the source and the crystal produces a geometrical relationship between the position of source and the position at which photons emitted from it strike the detector.

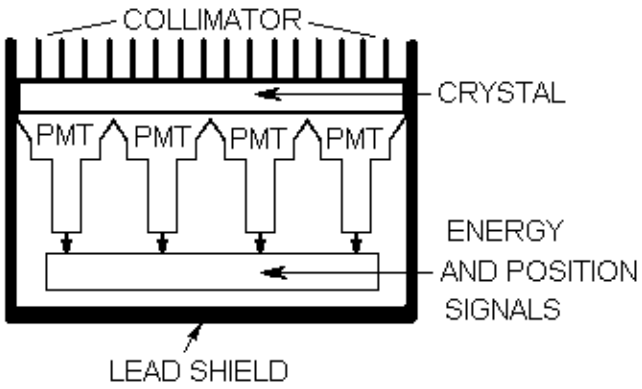


Fig. 7. The construction of an Anger gamma camera.

2/ X-ray imaging (Computed Tomography (CT))

All medical imaging systems represent the compromise between spatial resolution, low contrast detectability and radiation dose. During the diagnostic examination the detectability of a given tissue embedded within another depends on the energy of photons, the patient thickness, the thickness of structure of interest, the linear attenuation coefficient of the tissues and the detector characteristics.

The first clinically used CT system was installed in 1971. Since that year several developments have taken place in the system design. These changes have been brought about by the need for faster scan times (lower doses) coupled with a need for improved spatial resolution (viewing images of high spatial resolution lends confidence to the diagnosis).

The basic geometrical arrangement of the source, patient and detector are given in *Figure 8*.

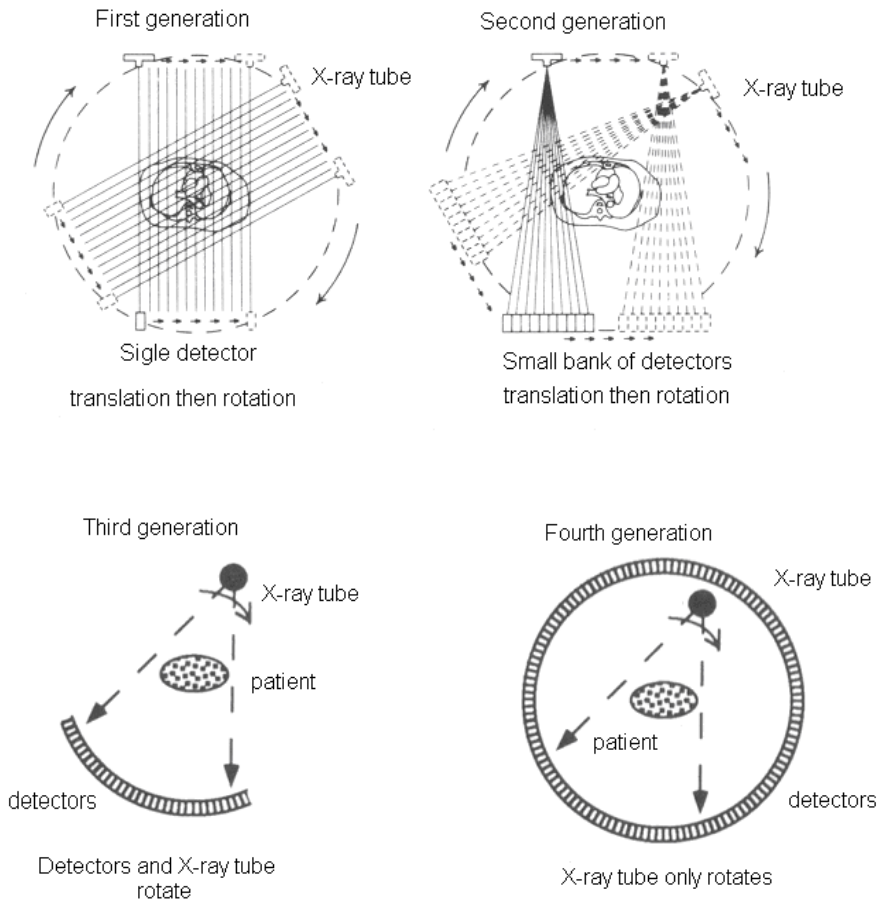


Fig. 8. The four generations of CT

The features of the different CT systems are presented in *Table 1*.

Table 1

Feature	First Generation	Second Generation	Third Generation	Fourth Generation
X-ray source	Single	Single	Single	Single
Detectors type	Single NaI(Tl)	NaI(Tl)	gas proportional chambers or scintillation with photodiode	scintillation with photodiode
Number of detectors	1	20 - 32	400-800	2000
Scan time	4.5-6 minutes	10-70 seconds	1-5 seconds	1-5 seconds

3/ Radiographic films

In diagnostic radiology photographic film is used for the direct imaging of X-rays and for recording the images produced by fluorescent screens and image intensifiers. In autoradiography a standard method of visualising particular molecular species in gel is to label it with beta emitter which then reveals its presence by exposing a photographic emulsion laid on top of it.

Counting statistics

Successive counts of a long-lived radioisotope will yield different counts, even though the sample geometry, equipment settings, etc. remain unchanged. The differences are, of course, due to the random nature of the decay process.

If we were to take a very large number of counts, the distribution of the recorded data would be quite accurately described by the normal distribution, or Gaussian curve. This is expressed as:

$$N(n) = \frac{k}{\sqrt{2\pi}} e^{\frac{-(n-\bar{n})^2}{2\sigma^2}} \quad (1)$$

where: k – number of measurements

n – any count

$\bar{n}$ - true mean count

N(n) – probability of obtaining a count of n

σ - Standard Deviation

Standard Deviation is defined as:

$$\sigma = \sqrt{\bar{n}} \quad (2)$$

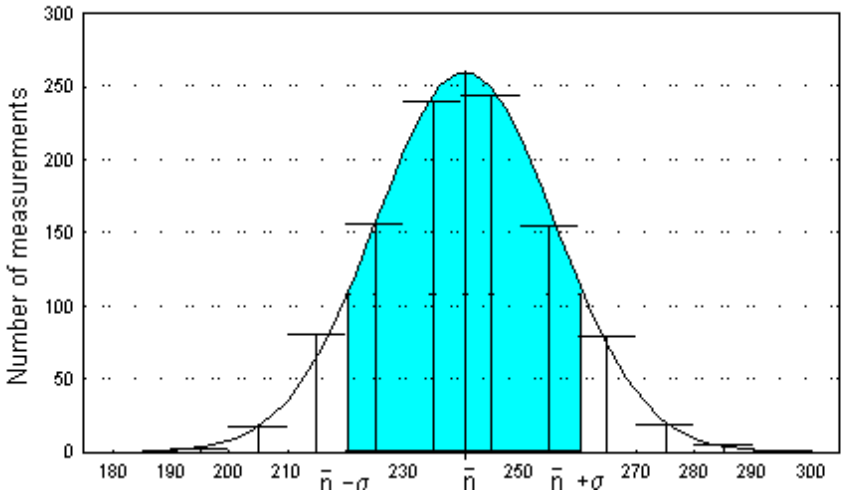


Fig. 9. The Gaussian curve (area of $\bar{n} \pm 1\sigma$ indicated)

The area under the curve, bounded by two coordinate values n_1 and n_2, is the probability of attaining a value of n between n_1 and n_2. The value under the entire curve is 1.

68.3% of the counts will be within ±1 standard deviation of the true mean, $\bar{n}$

95.5% will be within ±2 standard deviations of $\bar{n}$

99.7% will be within ±3 standard deviations of $\bar{n}$

In practice we often have the opportunity to take only one measurement of a sample. In this case, the single count becomes the only estimate of the mean, and the standard deviation is given by the following expression, from before:

$$\sigma = \sqrt{\bar{n}} = \sqrt{n} \qquad (3)$$

It is common practice to describe the precision of the measurements of the number of counts (n) by expressing Standard Deviation (σ) as a part and percent of n (relative error):

$$\varepsilon = \frac{\sigma}{n} \qquad (4)$$

$$\varepsilon_{\%} = \frac{\sigma}{n} \times 100\% \qquad (5)$$

Counts should always be reported as (n ± σ) and the confidence level specified. In this notation *n* is the counts measured, and *σ* defines the range of values having the given confidence level. As a convention, no specification of confidence level implies the 1 band - 68.3% confidence that the true mean lies within the range:

[n - σ] to [n + σ].

Equations (1) through (5) are applicable only if the dead time losses are negligible.

For *n* counts the standard deviation is $\sqrt{n}$, so the observed number of counts is: (n ± σ).

For the sum or difference the absolute errors add up in quadrature, so

$$Err^2(a \pm b) = Err^2(a) + Err^2(b) \qquad (6)$$

Applying these principles to the measured sample (T – total counts) and to the background (B – background counts in the same time interval), the net counts due to the sample (N) are:

$$N = T - B \qquad (7)$$

Then the Standard Deviation on N:

$$\sigma_N^2 = \sigma_T^2 + \sigma_B^2 = T + B \qquad (8)$$

and:

$$\sigma_N = \sqrt{T + B} \qquad (9)$$

So relative errors can be expressed as:

$$\varepsilon_N = \frac{\sigma_N}{N} = \frac{\sqrt{T + B}}{T - B} \qquad (10)$$

$$\varepsilon_{N\%} = \frac{\sigma_N}{N} \times 100\% = \frac{\sqrt{T + B}}{T - B} \times 100\% \qquad (11)$$

The equations (8) – (11) give the value of one SD or the 68% confidence level interval. Reporting an experimental result with its 95% confidence level (2σ) assumes that the particular measurement obeys a normal distribution, which in turn means that if the experiment were to be repeated, there would be a 95% chance that the new result would be within the quoted interval.

Example:
If the result of counting is 100 counts, then:
SD = 10 and 68% of the measurements would fall in the interval [90 -110] counts,
2SD = 20 and 95% of the measurements would fall in the interval [80 – 120] counts.
When we determine the activity of the sample, we use the count rate (I) which is expressed as a number of counts (n) divided by the time of measurement (t). Then we have:

$$I = \frac{n}{t} \qquad (12)$$

If we express the confidence level n±σ as $n \pm \sqrt{n}$ then, respectively, we have

$$I \pm \sigma_I \quad \text{and} \quad \frac{n}{t} \pm \frac{\sqrt{n}}{t} \quad \text{but} \quad \frac{\sqrt{n}}{t} = \frac{\sqrt{I \cdot t}}{t} = \sqrt{\frac{I}{t}}, \text{ so}$$

$$\sigma_I = \sqrt{\frac{I}{t}} \qquad (13) \qquad \varepsilon_I = \frac{1}{\sqrt{n}} \qquad (14) \qquad \varepsilon_{I\%} = \frac{100\%}{\sqrt{n}} \qquad (15)$$

The errors of net count rate we can express as follows:

$$I_N = I_T - I_B \qquad (16)$$

$$\sigma_{IN} = \sqrt{\sigma_{IT}^{2} + \sigma_{IB}^{2}} = \sqrt{\frac{I_T}{t_T} + \frac{I_B}{t_B}} \qquad (17)$$

$$\varepsilon_{IN} = \frac{\sigma_{IN}}{I_T - I_B} = \frac{\sqrt{\frac{I_T}{t_T} + \frac{I_B}{t_B}}}{I_T - I_B} \qquad (18)$$

$$\varepsilon_{IN\%} = \varepsilon_{IN} \times 100\% = \frac{\sqrt{\frac{I_T}{t_T} + \frac{I_B}{t_B}}}{I_T - I_B} \times 100\% \qquad (19)$$

PRACTICAL PART

EXPERIMENTAL PART

Objective

- you will calculate the Standard Deviation (σ) of ten results of measurements of the same radioactive sample using formula (2) and compare it with the value calculated by computer or scientific calculator

- you will make a histogram of values and find the statistical distribution of a hundred results of measurements of the same radioactive sample and calculate values of true mean and SD (using program “Statistica”)

- you will calculate and draw on the histogram intervals of 68% and 95% confidence levels

- you will calculate the probability of getting the result of one measurement in the intervals of 1σ and 2σ confidence levels

Materials

- NaI scintillation gamma detector connected to pulse counting system
- lead shield
- source of radiation
- PC with Statistica program
- scientific calculator

Procedure

1/ Measure the number of pulses from source in 10 seconds (10 times). Write the results in *Table 1*. Calculate the value of true mean and σ using the formula (2)

2/ Calculate the value of SD using PC program or scientific calculator. Write the results of calculation in *Table 1*

3/ Measure the number of counts from source in 10 seconds (100 times). Write the results in *Table 2*

4/ Use the results of measurements to generate a Gaussian distribution, as follows (use “Statistica” software):

- put the content of *Table 2* to one column of the program’s data table,

- calculate and write in *Table 3* the values of: Standard Deviation, the true mean, minimum and maximum,
- make a histogram of values (at least 100) for ten-second counts,
- calculate *(Table 4)* and draw on the histogram intervals of 68% and 95% confidence levels,
- count (using *Table 2*) how many measurements had the results in the intervals 1σ and 2σ. Put the results in *Table 4* as N_{σ} and $N_{2\sigma}$
- calculate (and write in *Table 4*) the probability of the result of one measurement falling in the intervals of 1σ and 2σ confidence levels by dividing the above calculated values by the total number of measurements (one hundred)

Data and observations

Table 1

Measurement number	Result (pulses)	True Mean $\bar{n}$	$\sigma = \sqrt{\bar{n}}$	SD computer calculated
1				
2				
3				
4				
5				
6				
7				
8				
9				
10				

Compare the two calculated values of Standard Deviation:

Table 2

Measurement		Measurement		Measurement		Measurement		Measurement	
Number	Pulses	Number	Pulses	Number	Pulses	Number	Pulses	Number	Pulses
1		21		41		61		81	
2		22		42		62		82	
3		21		43		63		83	
4		24		44		64		84	
5		25		45		65		85	
6		26		46		66		86	
7		27		47		67		87	
8		28		48		68		88	
9		29		49		69		89	
10		30		50		70		90	
11		31		51		71		91	
12		32		52		72		92	
13		33		53		73		93	
14		34		54		74		94	
15		35		55		75		95	
16		36		56		76		96	
17		37		57		77		97	
18		38		58		78		98	
19		39		59		79		99	
20		40		60		180		100	

Table 3

SD (σ)	True mean $(\bar{n})$	Minimum	Maximum

Place your histogram here

Table 4.

$\bar{n} - \sigma$	$\bar{n} + \sigma$	N_σ	$\bar{n} - 2\sigma$	$\bar{n} + 2\sigma$	$N_{2\sigma}$	$p_\sigma = \frac{N_\sigma}{100}$	$p_{2\sigma} = \frac{N_{2\sigma}}{100}$

Questions and conclusions