

10.SUMMARY

Oxidative stress (SO) is a pathophysiological phenomenon involving the overproduction of reactive oxygen species (ROS) in the body during the oxidation of compounds with the participation of cytochrome P-450, transport of electrons in the respiratory chain or an oxygen explosion. SO is one of the initiators of cell apoptosis, leading to weakening of the skin's function and premature aging. ROS are physiologically a source of energy in oxygen cells. The parameter indicating the oxidative processes occurring in the cell is the total oxidative status (TOS). When cells over-produce superoxide anion radicals, large amounts of hydrogen peroxide are formed, which cannot be reduced, and can cause a series of radical chain reactions leading to oxidative damage to proteins, lipids and DNA.

The skin is the organ that connects the body to the external environment. Under the influence of internal and environmental factors, it undergoes constant reconstruction processes. The slowing down of regeneration mechanisms, resulting from aging, gives symptoms in the form of reduced skin firmness and elasticity. An important element responsible for the appearance and proper functions of the skin is the right diet. The eating habits of the last few decades in society have largely departed from the recommendations of dietitians. During this time, increased consumption of carbohydrates and animal fats was noted. Currently, for several years, the trend of healthy eating and "fit" life has been observed. A very important element of a balanced diet is the right amount of protein provided with food, especially the essential amino acids, whose rich and valuable source is whey. Whey protein concentrate (WPC-80 preparation) contains essential, exogenous amino acids in the form of leucine, isoleucine and valine as well as growth factors and cytokines. Scientific studies have shown that whey protein concentrates have a very wide spectrum of activity (antioxidant, anti-cancer, hypotensive, central, antibacterial, antiviral, immunomodulatory, anabolic, repair). However, there is no information available on the effect of amino acid-enriched diets on biochemical parameters and the behavior of oxidative processes that damage proteins, lipids and DNA in the skin.

Therefore, the purpose of the experiment was to undertake studies assessing the effect of various amino acid-enriched diet variants on selected oxidation products of proteins, lipids and genetic material of intact skin of old rats.

The research material was downloaded as part of an earlier project implemented at UMB, which obtained IKE approval. The study material will be shaved dorsal skin taken from 40 Wistar rats with a body weight of 350-450 g, divided into 5 groups of 8 individuals each:

group I - control,

group II - WPC-80 preparation at a dose of 0.3 mg / kg bw for 7 days,

group III - WPC-80 preparation in a dose of 0.5 mg / kg bw for 7 days,

group IV - WPC-80 preparation at a dose of 0.3 mg / kg for 14 days,

group V - WPC-80 preparation at a dose of 0.5 mg / kg for 14 days.

After collection, the material was placed in deep freeze in liquid nitrogen, and after 24 hours placed in a low-temperature freezer at - 80°C until the day of the determination. On the day of biochemical determination, the samples will be washed with ice-cold PBS and then weighed. The samples will be cut into smaller fragments, suspended in PBS, placed in an ice bath and homogenized in a knife homogenizer. The resulting suspension will be sonicated. The homogenates will be centrifuged and the supernatant fluid obtained will be used for testing.

Methods

The following determinations will be made using enzyme immunoassays:

1. total oxidative status (TOS),
2. concentrations of lipid peroxidation products: 4-hydroxynonenal complexes with proteins (4-HNE protein adduct) and 8-isoprostanes (8-isoP),
3. concentration of protein oxidation products: advanced protein oxidation products (AOPP),
4. concentrations of DNA oxidation products: 8-hydroxy-2'-deoxyguanosine (8-OHdG) and
5. concentration of all skin proteins (BC).

Assays will be carried out in duplicates. After performing the experiments based on the results obtained, a selection of tests will be made to perform a statistical analysis in the Statistica software.

Conclusions

1. Different concentrations of WPC-80 and different times of its administration do not affect the overall oxidative status of healthy rat skin.
2. Administration of WPC-80 at a dose of 0.5 mg / kg for 14 days significantly reduces the value of lipid protein and DNA oxidation products.
3. The presence of lipid protein and DNA oxidation products in skin tissue indicates the induction of oxidative stress during the life of the animals in the research laboratory.
4. The use of a 0.5 dose in the diet for 14 days could protect against stress associated with natural, daily functioning.

