

Influence of Physical Activity on Concentration of Macro- and Microelements in Physically Active Students' Hair

Kazys Milasius¹, Marija Peciukoniene¹, Liudmila Loseva², Olga Tsvunchyk², Tatsjana Krupskaya², Slavamir Anufrik² and Vecheslav Maksimovich²

1. Department of Sport Teaching Methods, Lithuanian University of Educational Sciences, Vilnius LT-08106, Lithuania

2. Physico-Technical Faculty, Yanka Kupala State University of Grodno, Grodno 230023, Belarus

Abstract: The aim of the work is to evaluate the influence of physical activity character on concentration of macro- and microelements in hair of physically active students. 127 students of I-IV year of Physical Education program at Lithuanian University of Educational Sciences participated in the research, aged from 20 to 24. They were split into three groups. The first (I) group ($n = 48$) consisted of endurance sport athletes (skiers, biathlon athletes, middle and long distance runners, rowers and cyclists). The second group (II) ($n = 61$) was made up of the basketball, football, handball and volleyball players. The third group (III) consisted of combat sports (wrestling and boxing) athletes ($n = 18$). For establishing the concentration of macro- and microelements in hair, x-ray fluorescence method was applied, using CEP-1 ELVAX (MP.MN 3730-2011) device. The results showed that the hair of the students of Physical Education program, practicing endurance sports possessed more calcium, copper, manganese in comparison to the athletes practicing other sports. Concentration of the main elements—zinc, iron, selenium and chromium in the hair of combat sports representatives was statistically significant. Concentration of potassium was also the greatest in this group of the investigated. However, this group of athletes also possessed the greatest values of toxic elements (plumbum, arsenic) in their hair, comparing to the athletes of endurance sport and players. In their hair, hydrargyrum concentration was significantly greater comparing to the values of this element in players' hair. The greatest mean values of cadmium were established in players' hair.

Key words: Macro- and microelements, students of physical education program, hair.

1. Introduction

Contemporary training loads raise rather high requirements for athletes' body functional systems. The studies proved the maintenance of a proper concentration of vitamins, macro- and microelements in human body during persons' physical activity to be essential, as these elements regulate metabolic processes and body adaptation to training loads [1]. Minerals participate in cells regeneration and fermentation processes, endocrine glands activities; they regulate acid base balance, water metabolism, metabolism processes in muscles and head brain, as

well as in oxygen transfer. All minerals participate in metabolism processes either directly or indirectly, so they must be constantly supplemented via food intake [2, 3]. Athletes feel greater need for macro- and microelements, as due to great training loads the body loses salts. Normal course of vital processes is often hindered not only by the lack of minerals, but by their surplus or imbalance as well [4, 5].

In many cases, balance of electrolytes is being restored by using various sport beverages, while all necessary minerals should be consumed with food [6, 7]. Regarding significance of these elements to maintain body homeostasis, it is necessary to supply it by necessary elements. Thus quick obtainment of information on the amounts of certain elements, their surplus or lack in physically active persons becomes

Corresponding author: Kazys Milasius, Dr. Habil., professor, research fields: sport physiology, organism adaptation to physical loads, metabolism, sport training.

very important.

Concentration of minerals can be evaluated directly; by studying tissues, body fluids, nails and hair [8], and indirectly; by studying person's nutrition. One of the perspective methods to gain such information is a study of chemical elements concentration in hair using x-ray fluorescence method. Based on this method, scientists lately keep focusing their attention on macro- and microelements concentration studies in hair, together with blood and urine studies [9-12]. Hair is a formation of epidermis, which has become one of excretory body part during the process of evolution. Hair stands in the second place after bone marrow as a metabolic tissue of the body and reflects cells metabolism like any other tissue does. Hair is an informative substrate, which performs the functions of minerals bailment and accumulation. Hair studies allow establishing concentration of various bio elements in human body, as well as their surplus or lack. Besides, hair itself is able to excrete toxic elements from body. Analysis of hair composition allows establishing the lack of various elements in body (Ca, K, Mg) [13]. Hair grows rather rapidly between 0.2-0.5 mm per day, thus bearing information not only on current situation of a body, but on a previous period as well. Based on scientific studies [14-16], concentration of minerals in hair actually reflects the status of these elements in body and serves as an integral index of minerals metabolism. Legitimacy and effectiveness of hair use for ecological and toxicological analysis is proved by international coordinating programs, carried out by International atomic energetic service (TATENA). Hair is a favorable bio-substance for such studies and has numerous advantages: a sample is taken without damage for an athlete, storage of a bio-substance does not require a special equipment, and it does not deteriorate for an unlimited period of time, while information about the hair content is obtained in 30-40 min [17].

Considering the named facts, this method is useful in selecting athletes, making prognosis of their functional

abilities, allocation of training load, as hair studies provide information on body provision by vitally important microelements.

The review of literature sources dealing with macro- and microelements concentration in hair shows that this problem is being under analysis in the aspects of age, gender, geographical region of the investigated, nutrition and addiction as well [18-23].

Among numerous publications about the role of necessary for a human macro- and microelements, scientific works dealing with the impact of training load on the concentration of these elements in hair or blood serum are found [24-26]. However, the studies dealing with the question of physical load character influence on concentration of macro- and microelements in physically active persons' hair are not enough yet.

The aim of the work is to evaluate the influence of physical activity character on concentration of macro- and microelements in hair of physically active students of Physical Education program at Lithuanian University of Educational Sciences.

2. Methods

2.1 Participants

127 students of I-IV year of Physical Education programme at Lithuanian University of Educational Sciences participated in the research, aged from 20 to 24. They were split into three groups. The first (I) group ($n = 48$) consisted of endurance sport athletes (skiers, biathlon athletes, middle and long distance runners, rowers and cyclists). Their average height was 182.6 ± 0.8 cm, body mass is 75.7 ± 1.4 kg, BMI is 22.7. The second group (II) ($n = 61$) was made up of the basketball, football, handball and volleyball players. Their average height was 187.9 ± 1.1 cm, body mass is 81.9 ± 1.0 kg, BMI is 23.3. The third group (III) consisted of combat sports (wrestling and boxing) athletes ($n = 18$). Their average height was 182.7 ± 1.6 cm, body mass is 80.9 ± 2.8 kg, BMI is 24.2.

2.2 Experimental Design

The study was carried out during the spring semester, in April-May. The training load, carried out by the investigated, was the content of Physical Education program at the university, its amount being 6-10 weekly hours and training sessions of their sports (8-12 weekly hours).

2.3 Apparatus

For establishing the concentration of macro- and microelements in hair, x-ray fluorescence method was applied, using CEP-1 ELVAX (MP.MN 3730-2011) device; the research performed in Yanka Kupala State University of Grodno, Belarus. Technology of the samples collection and preparation is presented in Fig. 1. Agreement of the investigated to participate in the research was received before the start of the procedure. The hair sample was taken from the crown part of head; its mass was 0.5-0.6 g.

2.4 Statistical Analyses

The data of the research were processed applying mathematical statistical methods. Arithmetic mean (\bar{X}), standard error ($\sigma_{\bar{x}}$) and standard deviation (s.d.) were calculated. For evaluation of the group difference indices, method of dispersion analysis (ANOVA) was applied, calculating Fischer F criterion for dependent variables [27].

3. Results

Analysis of the results of investigation demonstrated that concentration of the main macro- and microelements in our investigated Physical Education program students' hair was within the limits of norms, as Table 1.

Established concentration of calcium in the hair of the investigated was of the following values according to their performed physical activity: endurance sports athletes is $659.01 \pm 45.41 \mu\text{g/g}$; players is $469.35 \pm$

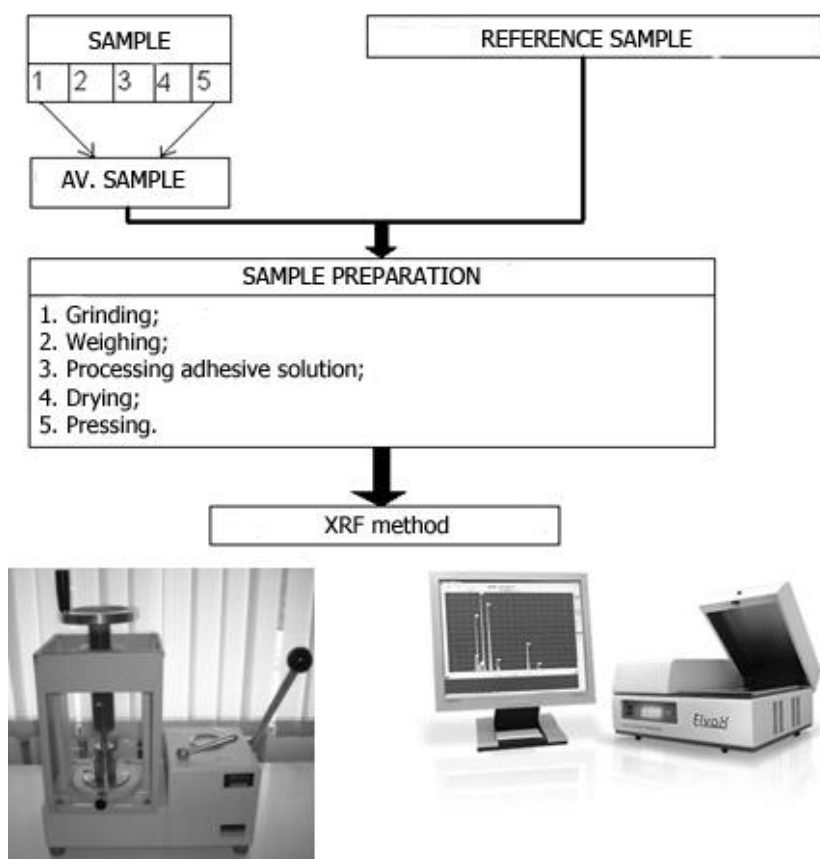


Fig. 1 XRF method. Stages of sample preparation.

Influence of Physical Activity on Concentration of Macro- and Microelements in Physically Active Students' Hair

Table 1 Macro- and microelements concentration in the hair of physically active students of Physical Education program at Lithuanian University of Educational Sciences ($\mu\text{g/g}$).

Indices	Main elements							Toxic elements				
	Ca	Zn	K	Fe	Cu	Se	Mn	Cr	Pb	Hg	As	Cd
Norm min-max	550-1700	120-200	70-170	10-25	9-30	0.3-1.2	0.5-2.0	0.5-5.0	0.0-5.0	0.0-2.0	0.0-2.0	0.0-1.0
Group I Endurance sports												
\bar{X}	659.01	172.10	81.85	16.09	29.51	0.55	1.44	2.14	2.06	0.53	0.23	0.38
$S\bar{x}$	45.41	7.04	7.91	1.10	3.25	0.03	0.11	0.16	0.15	0.04	0.04	0.03
S	314.62	48.81	54.82	7.65	22.55	0.17	0.78	1.11	1.03	0.29	0.23	0.20
Group II Players												
\bar{X}	469.35	184.84	85.15	12.44	18.51	0.49	1.12	2.43	2.24	0.48	0.20	0.43
$S\bar{x}$	32.52	7.88	9.10	0.69	1.42	0.02	0.08	0.16	0.14	0.03	0.02	0.03
S	251.92	61.02	70.49	5.38	10.97	0.16	0.65	1.21	1.03	0.24	0.16	0.23
Group III Combat sports												
\bar{X}	489.33	241.19	90.14	18.25	18.37	0.66	1.41	2.98	2.34	0.66	0.25	0.42
$S\bar{x}$	47.47	40.81	15.11	1.67	1.69	0.05	0.13	0.32	0.27	0.05	0.05	0.07
S	201.40	173.13	64.09	7.10	7.18	0.22	0.56	1.34	1.13	0.21	0.18	0.30
Significance of difference												
I-II F	12.110	1.382	0.071	8.427	11.039	3.418	5.622	1.683	0.779	1.236	0.672	1.713
P	0.001	0.242	0.790	0.004	0.001	0.067	0.020	0.197	0.380	0.269	0.414	0.193
I-III F	4.516	6.435	0.273	1.085	4.197	5.098	0.030	6.696	0.918	2.767	0.078	0.452
P	0.037	0.014	0.603	0.302	0.044	0.027	0.863	0.012	0.342	0.101	0.781	0.504
II-III F	0.095	3.790	0.072	13.836	0.002	13.766	2.784	2.728	0.125	7.948	1.165	0.029
P	0.759	0.056	0.789	0.000	0.960	0.000	0.099	0.103	0.724	0.006	0.284	0.866

32.52 $\mu\text{g/g}$; combat sports athletes is 489.33 \pm 47.47 $\mu\text{g/g}$. Concentration of calcium was statistically greater in the hair of endurance sports athletes comparing to the values of the players ($P < 0.001$) and combat sports athletes ($P < 0.037$).

The research demonstrated the greatest values of zinc concentration in combat sports athletes' hair is 241.19 \pm 40.8 $\mu\text{g/g}$, while the less values were established in the hair of endurance sports athletes is 172.10 \pm 7.04 $\mu\text{g/g}$ ($F = 6.435$, $P < 0.05$). Potassium concentration in the hair of the investigated was within the limits of norms, no statistically significant differences between the groups were found. Iron concentration in the players' hair was significantly less than in the hair of the other investigated students ($P < 0.004$ and $P < 0.001$), and was at the lower limit of the norm (12.44 \pm 0.69 $\mu\text{g/g}$). For the students of Lithuanian University of Educational Sciences, carrying out Physical Education program and practicing endurance sports, considerably greater

copper concentration was found is 29.51 \pm 3.25 $\mu\text{g/g}$ (significance of difference p between the I-II groups was < 0.001 ; P between I-III groups was < 0.044). Concentration of selenium was statistically greater in the hair of our investigated athletes of combat sports is 0.66 \pm 0.05 $\mu\text{g/g}$, comparing amounts of this element in players ($P < 0.004$) and endurance sports athletes' hair ($P < 0.067$). The research demonstrated significant greater concentration ($P < 0.02$) of manganese in endurance sports athletes' hair (1.44 \pm 0.11 $\mu\text{g/g}$), comparing to the players' values. Chrome concentration was the greatest in the hair of combat sport athletes is 2.98 $\mu\text{g/g}$, and this value had statistically significant difference ($P < 0.012$) comparing to the athletes practicing endurance sports.

The results of investigation on toxic elements demonstrated that greater values of plumbum were found in combat sports athletes' hair; however, this concentration did not exceed the limits of norms, and differences between the groups of the investigated did

not have statistical difference (Table 1).

Concentration of hydrargyrum, arsenic and cadmium in the hair of the investigated was within the limits of norms, while the differences between the groups were not statistically significant.

4. Discussion

Calcium, one of the main macroelements, is a compound part of bones, it participates in muscle contraction, serves as a mediator for neuro processes, is important for blood coagulation, forces breakdown processes of ATP, regulates activeness of ferments. Lack of calcium is followed by osteoporosis, decrease in density of bones, weakening of muscle contraction, muscle spasms, and arrhythmias. However, the deposits of calcium in body are not constant. Several hundred milligrams of calcium are removed from body with excrements and urine daily, so these losses must be supplemented with daily food intake. Paschal et al. [10] having carried out investigation on relation between metal concentration and age in the USA citizens' hair established that concentration of calcium in hair increases until 12-14 years of age, and becomes stable afterwards. Analogous data was received by Koziolec et al. [28] in their study of macro- and microelements concentration in the hair of young North Poland seaside dwellers. The latter study demonstrated greater concentration of calcium in girls than in boys' hair. The data of our study show an increase of calcium concentration in athletes' hair due to aerobic training load.

Zinc is a component of ferments, it participates in metabolic processes, synthesis of proteins and nucleic acids, as well as in immune reactions, participates in blood production. Długaszek et al. [29] in their study having investigated impact of environment and age on the state of minerals in male and female hair, found greater concentration of zinc in women's hair. Based on their obtained data, mean concentration of zinc in the hair of 1038 women and men, split into five age groups, reached 161 $\mu\text{g/g}$. In the hair of our

investigated third group students, representatives of combat sports, this concentration reached $241.19 \pm 40.81 \mu\text{g/g}$. This fact demonstrates that prevailing force exercises for these group athletes during their training sessions stimulate more active metabolism of zinc. According to Contiero and Folin [11], concentration of zinc in body is very important for increasing body mass; this is rather urgent in combat sports, making corrections on body mass according to requirements for weight categories.

Potassium participates in nervous impulse transmission, maintains homeostasis, and regulates muscles excitability. In case of hypokalemia, such symptoms as muscle weakness and their spasms, lack of appetite and heart rate decrease are felt. Lubkowska [25] in her investigation of Polish students, studying Physical Education program at *University of Szczecin*, established that average concentration of potassium in the hair of women was 13.52 $\mu\text{g/g}$, while of men is 107.5 $\mu\text{g/g}$. Concentration of potassium in the hair of Lithuanian students, studying at Lithuanian University of Educational Sciences, was in average from 81.85 to 90.14 $\mu\text{g/g}$, depending on the character of physical activity they used to perform.

Iron is necessary for carry of oxygen, enters into composition of hemoglobin and myoglobin, participates in immune reactions, plays important role in growth, blood and ferments production. Deficiency of iron is shown in anemia, fatigue, absence of immunity to infectious diseases, decreased working efficiency. There are many factors of influence on iron reserves in athletes' body: hard training workouts, loss of iron with urine and perspiration, insufficient amount of iron, consumed with food. According to Jeukendrup and Gleeson [1], metabolism of iron and its intake can be damaged by sporting activity the person is engaged into. Thus, observation of proper balance of iron is very important task for athletes practicing various sports.

Copper participates in the processes of growth, split of glucose and glycogen, it increases activeness of lipolysis, stimulates iron absorption and synthesis of

hemoglobin. Deficiency of copper manifests in weakened immune system, anemia and demineralization of bones. The results of Gonzalez et al. [21] study on minerals concentration of Canary Islands inhabitants' hair showed that greater concentration of copper and iron was found in the hair of persons consuming more vegetables and fish products, while concentration of copper was significantly less in the hair of persons suffering from obesity and overweight. Inverse correlation was found for those persons between the amounts of copper and their body mass index ($r = -0.21$, $P = 0.043$). The same authors [30], having performed research on the hair of the Spanish students, studying at La Laguna University, found in average 41.35 mg/kg copper in their hair with rather great difference between minimum and maximum value (7.79-109.55 mg/kg). Concentration of copper in our investigated students of Lithuanian University of Educational Sciences ranged from 10.30 to 91.70 $\mu\text{g/g}$.

Selenium acts against degeneration of tissues, caused by deficiency of tocopherol (vitamin E). It is essential for the immune system and acts as a very strong antioxidant, protecting from radiating and chemical carcinogens. Deficiency of selenium in human body results in heart failure, muscle work disorder, decreased reproduction, formation of depression features [22]. The results of our research showed greater values of selenium in the hair of athletes practicing power sports.

Manganese is a catalyst of energy metabolism, helps to maintain bones composition and participates in the synthesis of fats. Deficiency of manganese is followed by violation of metabolism of cholesterol, slackened growth of hair and nails, worse tolerance for glucose, appearance of hormones changes which might be the reason of infertility, together with the weight loss. Superfluous amount of manganese manifests itself in ailment of general character, weakness, quick fatigue, memory weakening, headache, sleep disorders, etc. [1].

Chromium participates in energy metabolism

processes; it is a component of cholesterol and acts as an activator of ferments, enhances permeability of nervous impulses. Its deficiency blocks glucose intake and decreases effectiveness of insulin activity. Following our research data, the hair of combat sports athletes developing force during their training workouts, collects more chromium. As a matter of fact, the study of Afridi et al. [20] showed that the hair of persons suffering from skin diseases possesses greater concentration of chromium than the hair of healthy subjects. Otag et al. [26] found greater concentrate of chromium in the blood serum of elite athletes developing aerobic capacity comparing to the results obtained from combat sports athletes. Nevertheless, our research demonstrates statistically greater values ($P < 0.012$) of chromium in combat sports representatives' hair comparing to the values of athletes practicing endurance sports.

Numerous publications, dealing with the problem of concentrate of toxins in human hair, state that this concentration depends on environmental pollution, consumed food [12] and age [10, 23]. Otag et al. [24] states that amount of toxic elements can limit the result of an athlete and be of influence on his health. Literature review on this question does not provide one opinion about influence of toxic elements concentration on physically active persons' working efficiency, thus the further researches in investigating concentration of these elements in persons' hair should be continued [29].

5. Conclusions

X-ray fluorescence method is a perspective and informative way for establishing concentration of macro- and microelements in physically active persons and elite athletes' hair during different stages and periods of their preparation. The data of these studies can be used in assisting and optimizing athletes' nutrition and training process.

The results of our performed research demonstrated that the hair of the students of Physical Education

program, studying at Lithuanian University of Educational Sciences and practicing endurance sports possessed more calcium, copper, manganese in comparison to the athletes practicing other sports. Concentration of the main elements such as zinc, iron, selenium and chromium in the hair of combat sports representatives was statistically significant. Concentration of potassium was also the greatest in this group of the investigated. However, this group of athletes also possessed the greatest values of toxic elements (plumbum, arsenic) in their hair, comparing to the athletes of endurance sports and players. In their hair, hydrargyrum concentration was significantly greater comparing to the values of this element in players' hair. The greatest mean values of cadmium were established in players' hair.

References

- [1] Jeukendrup, A., and Gleeson, M. 2010. *Sport Nutrition: An Introduction to Energy Production and Performance (2nd ed.)*. USA, Champaign, IL: Human Kinetics.
- [2] Benardot, D. 2000. *Nutrition for Serious Athletes*. USA, Human Kinetics.
- [3] Woolf, K., and Manore, M. 2007. "Micronutrient Important for Exercise." In *Nutrition and Sport*, edited by MacLaren, D. Edinburgh: Churchill Livingstone Elsevier, 119-36.
- [4] Hunt, S., and Groff, J. 1990. *Advanced Nutrition and Human Metabolism*. MN: West: St. Paul Publishing Company, 264-348.
- [5] Maughan, R. J. 1999. "Role of Micronutrients in Sport and Physical Activity." *British Medical Journal* 55 (3): 683-90.
- [6] Clarkson, P. 1991. "Minerals: Exercise Performance and Supplementation in Athletes." *Journal of Sports Sciences* 9: 91-116.
- [7] Clarkson, P. 2000. "Trace Minerals." In *Nutrition in Sport*, edited by Maughan, R. Oxford: Blackwell Science: 339-55.
- [8] Hopps, H. C. 1977. "The Biologic Bases for Using Hair and Nail for Analyses of Trace Elements." *Science of the Total Environment* 7 (1): 71-89.
- [9] Gordon, G. F. 1985. "Sex and Age Related Differences in Trace Element Concentrations in Hair." *Science of the Total Environment* 42: 133-47.
- [10] Paschal, D. C., Di Pietro, E. S., Philips, D. L., and Gunter, F. W. 1989. "Age Dependence of Metals in Hair in a Selected U. S. Populations." *Environmental Research* 48 (1): 17-28.
- [11] Contiero, E., and Folin, M. 1994. "Trace Elements Nutritional Status, Use of Hair as a Diagnostic Tool." *Biological Trace Element Research* 40 (2): 151-9.
- [12] Kuangfei, L., Yaling, X., Xuefeng, L., Zuoli, W., Bukkens, S., Tommaseo, M., and Paoletti, M. 1999. "Metallic Elements in Hair as a Biomarker of Human Exposure to Environmental Pollution: A Preliminary Investigation in Hubei Province." *Critical Reviews in Plant Sciences* 18 (3): 417-28.
- [13] Mikulewicz, M., Chojnacka, K., Gedrange, T., and Górecki, H. 2013. "Reference Values of Elements in Human Hair: A Systematic Review." *Environmental Toxicology and Pharmacology* 36 (3): 1077-86.
- [14] Pavlov, I. V., Agadzhanian, N. A., Alisievich, V. I., and Chekhovskikh, M. M. 1989. "Comparative Study of the Macro- and Microelemental Composition of the Hair on the Head in Inhabitants of Various Regions." *Human Physiology (in Russian)* 15 (1): 154-61.
- [15] Radomska, K., Graczyk, A., Konarski, J., and Adamowicz, B. 1991. "Evaluation of Macro- and Micro-Element Content in the Human Body Determined by Hair Analysis." *The Polish Medical Weekly* 46 (24-26): 461-3. (in Polish)
- [16] Noguchi, T., Itai, T., Kawaguchi, M., Takahashi, S., and Shinsuke, T. 2012. "Applicability of Human Hair as a Bioindicator for Trace Elements Exposure." *Interdisciplinary Studies on Environmental Chemistry—Environmental Pollution and Ecotoxicology*: 73-7.
- [17] Skalnyj, A. V. 2003. "Reference Values of Chemical Elements Concentration in Hair, Obtained by Means of ICP-AES Method." *Trace Elements in Medicine* 2 (1): 7-11. (in Russian)
- [18] Sturaro, A., Parvoli, G., Doretti, L., Allegri, G., and Costa, C. 1994. "The Influence of Color, Age and Sex on the Content of Zinc, Copper, Nickel, Manganese, and Lead in Human Hair." *Biological Trace Element Research* 40 (1): 1-8.
- [19] Kałuzka, J., Jeruszka, M., and Brzozowska, A. 2001. "Iron, Zinc and Copper Status in the Elderly Living in Warsaw District Determined by Hair Analysis." *Annals of the National Institute of Hygiene* 52 (2): 111-8. (in Polish)
- [20] Afridi, H. I., Kazi, T. G., Jamali, M. K., Kazi, G. H., and Shar, G. Q. 2006. "The Status of Trace and Toxic Elements in Biological Samples (Scalp Hair) of Skin-Disease Patients and Normal Subjects." *Turkish Journal of Medical Sciences* 36 (4): 223-30.
- [21] Gonzalez-Reimers, E., Martin-Gonzalez, M. C., Galindo-Martin, L., Duran-Castellon, M. C., Aleman-Valls, M. R., Velasco-Vázquez, J., Gonzalez-Perez, J. M., and Barroso-Guerrero, F. 2008. "Hair Zinc, Copper and Iron: Relationships with Quality

Influence of Physical Activity on Concentration of Macro- and Microelements in Physically Active Students' Hair

- of Diet, Tobacco Smoking and Nutritional Status.” *Trace Elements and Electrolytes* 25 (1): 35-40.
- [22] Momcilovic, B., Moroviic, J., Prejac, J., Skalnaya, M. G., and Ivicic, N. 2008. “Relationship of Iodine, Selenium and Copper in the Hair and Whole Blood of Depressed Human Subjects.” *Trace Elements and Electrolytes* 25 (4): 195-8.
- [23] Grabeklis, A. R., Lakarova, E. V., Eisazadeh, S., and Skalny, A. V. 2011. “Sex Dependent Peculiarities of Some Important Element Ratios in Hair of Schoolchildren.” *Trace Elements and Electrolytes* 28 (2): 88-90.
- [24] Qian, Q., Chiao, Z., Wu, Y., and Tian, J. 1990. “Study of Trace Elements in Hair of Athletes by Synchrotron Radiation X-Ray Fluorescence Analysis.” *Hejishu* 14 (8): 493-6.
- [25] Lubkowska, A. 2009. “Concentration of Selected Elements in Hair of Healthy Individuals with Increased Physical Activity.” *Trace Elements and Electrolytes* 26 (4): 145-9.
- [26] Otag, A., Hazar, M., Gürkan, A. C., and Okan, I. 2014. “Responses of Trace Elements to Aerobic Maximal Exercise in Elite Sportsmen.” *Global Journal of Health Science* 6 (3): 90-6.
- [27] Gonestass, E., and Strielčiūnas, R. 2003. *Applied Statistics*. Kaunas: LKKA. (in Lithuanian)
- [28] Koziolec, T., Drybańska-Kalita, A., Hornowska, I., and Sałacka, A. 1996. “Levels of Calcium, Magnesium, Zinc, Copper and Iron in Hair of Children and Adolescents.” *Medical Research: Journal of Polish Medical Society* 1 (2): 150-4. (in Polish)
- [29] Długaszek, M., Skrzeczanowski, W., and Kaszczuk, M. 2014. “Age-Related Mineral Status of Females and Males Hair in Human Health Risk Assessment.” *Trace Elements and Electrolytes* 31 (3): 131-40.
- [30] González-Reimers, E., Martín-González, C., Galindo-Martín, L., Aleman-Valls, R., González-Pérez, J. M., Jorge-Ripper, C., Elvira-Cabrera, O., and Quintero-Platt, G. 2014. “Hair Copper in Normal Individuals: Relationship with Body Mass and Dietary Habits.” *Trace Elements and Electrolytes* 31 (2): 67-72.