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CYTOLOGICAL INDICATORS OF BUCCAL EPITHELIUM CELLS REGARDING PHYSICAL ACTIVITY AND COMPLIANCE WITH THE DRINKING REGIME OF CERTAIN GROUPS OF YOUNG PEOPLE

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The study of the buccal epithelium also plays an important role in modern approaches to preventive medicine, in assessing the risk of various pathologies, including pathological conditions associated with the drinking regimen. The purpose of the study was to determine the cytogenetic features of buccal epithelium cells in athletes of strength sports (sambo) and team sports (volleyball) in the period between competitions with a normal training regimen, to compare with a control group of young people who do not go in for sports, and to study water consumption regimes. In the studied groups and assess the relationship with cytological and morphometric parameters. 3 homogeneous groups were formed for a comparative assessment of the cytogenetic parameters of the buccal epithelium. In total, 27 athletes of power sports (judo), 25 athletes of team sports (volleyball) and 45 representatives of the control group took part in the study. Evaluation of cytogenetic parameters revealed statistically significant patterns: Cells containing micronuclei were found 3.85 times more often in the power group and 4.3 times more often in the game group than in the control group. The frequency of cells with protrusions generally corresponded to the trend towards the absence of statistically significant differences between strength and game sports and the presence of significant differences compared to the control group.

Keywords: buccal epithelium, water consumption regimen, athletes, power sports, team sports, cytogenetic changes.

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Introduction. One of the urgent tasks of medicine is the use of non-invasive methods for the early diagnosis of diseases. Buccal epithelium is also used as a material for non-invasive methods. According to modern data, the buccal epithelium is a sensitive "mirror" that allows you to identify many factors that affect human health. The study of the buccal epithelium also plays an important role in modern approaches to preventive medicine, in assessing the risk of various pathologies, including pathological conditions associated with the drinking regimen [1, 4].

A correlation was determined between laboratory parameters: cholesterol in obese children, total protein in healthy children and the number of binuclear cells. There is a 10x increase in the number of cells with condensed chromatin in obese children [1].

A number of studies have shown differences in the frequency of occurrence of various cytogenetic disorders in healthy individuals of different nationalities, gender, and age. To a greater extent, there is a different ratio of the number of cells with micronuclei, protrusions of the "broken egg" type, nuclei with perinuclear vacuoles, protrusions of the "notched" type, protrusions of the "tongue" type, and other disorders in the studied samples. Apparently, this is due to the adaptive abilities of the body to new environmental conditions, the individual charac-

teristics of the body, hormonal activity, general physical fitness, the state of the nervous system, and the processes of humoral regulation [3, 4, 5].

Various ecotoxins, smoking, alcohol and negative factors of the labor process have a significant impact on the state of the nuclei of buccal epithelium cells. Thus, pesticides cause activation of the processes of proliferation and destruction [6], almost always there is a statistically significant increase in the number of micronuclei in workers in hazardous industries and residents of regions with chemical industries [7, 8, 9].

The dynamics of changes in buccal epithelium cells in power sports athletes at various stages of the competitive process was studied. Correlations between the level of various types of aggression and other indicators of the psychophysiological state of athletes have been revealed, and complex models for the regulation of these changes have been proposed [10]. The results obtained can be considered markers of the effectiveness of preparation for competitions, and the low invasiveness of the micronucleus test allows you to control these processes in dynamics.

Based on the above trends, we set the goal of the study - to determine the cytogenetic features of buccal epithelium cells in athletes of strength sports (sambo) and team sports (volleyball) in the period between competitions with a nor-

mal training regimen, to compare with the control group of young people, non-athletes, as well as to study the water consumption regimes in the studied groups and evaluate the relationship with cytological and morphometric parameters.

Materials and methods. 3 homogeneous groups were formed for a comparative assessment of the cytogenetic parameters of the buccal epithelium. In total, 27 athletes of power sports (judo), 25 athletes of team sports (volleyball) and 45 representatives of the control group took part in the study.

All groups included males aged 19 to 25 years. Before the start of the study, everyone completed a questionnaire. All respondents claimed that they did not have bad habits, chronic diseases, complaints from the dentoalveolar system. All respondents also claimed to have been in Barnaul for the past year or more. All respondents stated that they train in the period between competitions in the usual way, which includes 3 workouts per week, lasting 2 astronomical hours each.

Representatives of the control group met the indicated criteria, but did not go in for professional sports and did not attend the gym on a regular basis, however, as students of a higher educational institution, they attended a general physical training class once a week as part of the main health group (2 academic hours).

The screening questionnaire about the drinking regimen included questions about the characteristics of water consumption ("tap", "bottled"), knowledge about the drinking regimen ("I know and observe", "I don't know", "I know and do not observe"), signs of dehydration ("I feel thirsty in the morning constantly", "I feel thirsty in the morning periodically", "I don't feel thirsty in the morning"). Further comparison of water consumption regimens was carried out between athletes in general and the control group.

The selection of the buccal epithelium was carried out with a wooden spatula, from the mucous surface of the oral cavity and applied to a glass slide with further fixation over the flame of an alcohol burner and stained with methylene blue. Visual microscopy was performed at a magnification of 100x10, digital microscopy and morphometry using a TouPCam 3.2 megapixel video camera, at least 1000 cells were viewed in each preparation.

Based on the sample sizes, the Shapiro-Wilk test was used to assess the compliance with the normality of the data distribution. To detect the absence or presence of differences between the indicators in the studied groups, the Kruskal-Wallis test was used; if statistically

significant differences were found, post-hoc intergroup comparisons were carried out using the Mann-Whitney test. In a simple paired comparison, the significance level was taken equal to $p = 0.05$; in cases of paired comparisons between three groups, the critical significance level was taken equal to $p = 1 - 0.95^{1/3} = 0.017$. In the text and tables, data are presented as the median and the values of the first and third quartiles in parentheses. The calculations were carried out in IBM SPSS Statistics 23.0.

Results. At the first stage of the study, a screening assessment of the drinking regime was carried out on the basis of a questionnaire. It turned out that in the group of athletes 71% use bottled water and only 29% tap water, in the control group these values correspond to 57% and 43%, respectively. To the question about compliance with the drinking regime, the answers were distributed as follows: athletes - "I know I comply" - 89%, "I don't know" - 2%, "I know I do not comply" - 9%, the control group - "I know I comply" - 11%, "I don't know" - 56%, "I know I don't comply" - 33%. In the group of athletes, when asked about the signs of dehydration, the answers were distributed as follows - "I feel thirsty in the morning all the time" - 12%, "I feel thirsty in the morning periodically" - 31%, "I don't feel thirsty in the morning" - 57%; in the control group, respectively - "I feel thirsty in the morning all the time" - 22%, "I feel thirsty in the morning periodically" - 40%, "I do not feel thirsty in the morning" - 38%.

Next, we compared the values of the area of the cytoplasm and the nucleus in the cells of the buccal epithelium of athletes of all categories and the control group. Morphometry in the group of athletes showed that the median area of the cytoplasm corresponds to 3621.3 (2911.2-4887.3) μm^2 , and the area of the nucleus is 61.3 (56.8-66.6) μm^2 . In the control group, the median area of the cytoplasm corresponds to 3017.8 (2613.7-4113.3) μm^2 , and the area of the nucleus is 58.9 (53.1-68.3) μm^2 . It was statistically reliably established that the median of the cytoplasm area in the group of athletes is greater than in the control group, which may be due to better conditions for observing the drinking regimen. There were no statistically significant differences between the medians of the core area.

The obtained results testify to the positive effect of the pedagogical work of the coaching staff, counting on high achievements, including through the fight against dehydration and timely replenishment of microelement deficiencies, however, to

clarify the role of bottled water, additional research is required on the composition and additives used, such as zinc, selenium, iodine, fluorine, etc.

The data in the control group indicate the need to intensify sanitary and educational work among different groups of young people not associated with sports, organize lectures, schools for a healthy drinking regime.

Earlier studies supplemented the results organically, so in [11, 14] it was shown that men's total daily water intake was about 0.25 l less than the recommended adequate intake, while women's intake was about the same as the adequate intake. On average, men and women aged 60 years and older, non-Hispanic black men and women, Hispanic men and women, men and women with low physical activity, and men with moderate physical activity consumed less than the adequate daily norm, which also indicates an indirect positive effect of physical activity on the maintenance of internal environmental constancy in athletes. Also of interest for practical implementation are studies [12, 13, 15], where it was found that an increase in water consumption can contribute to the prevention of overweight. It can be said that a balance between adequate physical activity, a full drinking regime and lifestyle is an effective mechanism of health saving in all age groups.

At the second stage of the study, before conducting intergroup comparisons, we assessed the type of distribution for each of the studied parameters, since the sample did not exceed 50 cases for each of the groups, the Shapiro-Wilk test was used, for all values except for the frequency of cells with an atypical nucleus ($p=0.057$) and the frequency of cells with protrusions of the "tongue" type ($p=0.066$), it took values of $p<0.05$, which indicates non-normal types of distribution in the studied samples. Thus, further, we used nonparametric criteria to establish differences between the studied groups.

Evaluation of cytogenetic parameters revealed statistically significant patterns: Cells containing micronuclei were found 3.85 times more often in the power group and 4.3 times more often in the game group than in the control group. The frequency of cells with protrusions generally corresponded to the trend towards the absence of statistically significant differences between strength and game sports and the presence of significant differences compared to the control group. Within this indicator, a number of features stand out, so the frequency of cells with protrusions of the "bubble" type is more

common in the power group than in the game and control groups. The frequency of cells with protrusions of the "broken egg" type is 2 times more common in the power group and 2.4 times more common in the game group than in the control group. When pairwise comparing the frequency of cells with an atypical nucleus, it turned out that such changes are more typical for representatives of team sports (7.64 times more often than in the control group and 2.37 times more often than in the game group). The integral indicator of the cytogenetic effect corresponded to the general trend and generally showed that statistically significant differences were found between the control group and the strength and play groups (see Table 1). It can also be noted that in the group of athletes of both power and game groups, rare anomalies such as "many micronuclei" (see Fig. 1) and "atypically shaped core" (see Fig. 2) were more common, statistically significant differences compared to the control group could not be established due to the low frequency of observation of these changes. The results obtained are generally consistent with existing studies and are primarily due to reactive processes in the body - a systematic increase in the level of cortisol, inflammatory mediators, changes in circadian rhythms due to participation in competitions in different time zones and climatic zones, however, rather high values of the frequency of occurrence of micronuclei in all groups, including the control group, require an additional assessment of environmental factors and an assessment of the role of nutrition, drinking regimen, and environmental factors.

Grade features indicators of proliferation of cells of the buccal epithelium made it possible to determine that the main differences were represented (see Table 2) by an increase in the frequency of occurrence of cells with two or more nuclei in the game group, cells with double nuclei and circular notches in the power group (2.01 times more often than in the control group and 1.84 times more often than in the game group), which ultimately led to statistically significant differences between all the groups studied, so the integral proliferation index is highest in the game group, the second place is the power group and the last is the control group. These changes may reflect the constant processes occurring in the body of athletes - increased testosterone levels, chronic stress, some of the changes are also due to the background level of environmental factors.

Evaluation of indicators of early and late destruction of the cell nucleus (see

Cytogenetic parameters, %

Indicators	Power, group 1 (n=27)	Gaming, group 2 (n=25)	Control group, group 3 (n=45)
Cells with micronuclei	7.36 (4.5-7.9)	8.22 (6-10)	1.91*** (0-2.75)
cells with protrusions	2.82 (1-3)	2.22 (1-4)	1.73*** (1-2)
Cells with vesicular protrusions	2.09** (0-3)	1.44 (0-3)	1.18 (1-2)
Cells with "broken egg" protrusions	0.18 (0-1)	0.22 (0-1)	0.09*** (0-1)
Cells with tongue protrusions	0.55 (0-1)	0.56 (0-1)	0.45 (0-1)
Cells with an atypical nucleus	1.45* (1-2)	3.44* (2-4)	0.45* (0-1)
Integral indicator of cytogenetic action	10.18 (5.5-12.5)	10.44 (7-11)	3.64*** (1-6)

Note: * $p < 0.017$ changes are significant between all groups in a pairwise comparison (1-2, 2-3, 1-3), ** $p < 0.017$ changes are reliable in pairs of groups 1-2 and 1-3, *** $p < 0.017$ changes are reliable in pairs of groups 1-3 and 2-3

Table 3) did not reveal significant differences, only the frequency of cells with perinuclear vacuole in the game group was statistically significantly higher in the game group than in the power and control groups (5.56 times), these changes can be explained by active preparation for participation in competitions at the regional level.

Evaluating the results obtained, it seems relevant to identify the role of individual trace elements such as potassium, calcium, magnesium in the formation of indicators of mental and physical performance, correction of cytogenetic, proliferative apoptotic changes in the nuclei of buccal epithelial cells. Ways to additionally compensate for micronutrient deficiencies may include both changes in dietary behavior and the consumption of specialized bottled waters enriched with essential substances.

Conclusions:

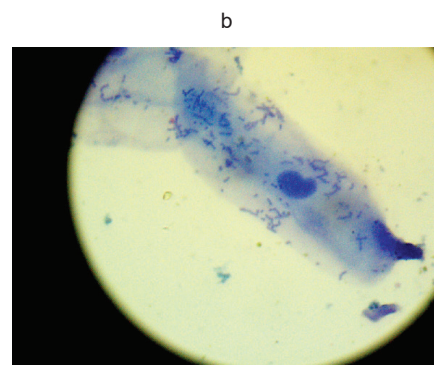
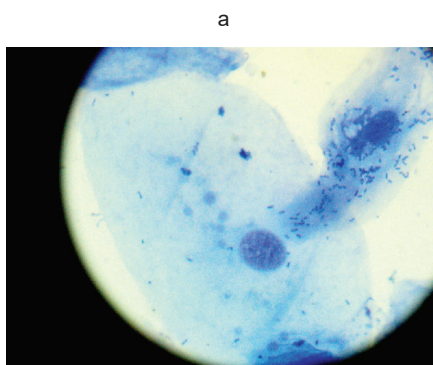
1) The features of water consumption of athletes in game and power sports were determined in comparison with the control group and related changes in cytological parameters;

2) Athletes in the conditions of preparation for competitions are under stress and the risk of increasing the level of genome instability, determined by cytogenetic and proliferative indicators;

3) The control of cytological parameters of the buccal epithelium seems to be a promising method for assessing the effectiveness of training;

4) An assessment of the cytological landscape of the representatives of the control group is necessary to clarify the contribution of other risk factors for genomic instability due to regional characteristics;

5) It is necessary to conduct ad-



Microscopic picture of anomalies of the buccal epithelium of the type "many micronuclei" (a), type "atypical nucleus" (b)

Table 2

Proliferation indices, %

Клетки	Power, group 1 (n=27)	Gaming, group 2 (n=25)	Control group, group 3 (n=45)
Cells with two or more nuclei	2.00 (1-2.5)	3.67** (2-5)	1.82 (0.5-2)
Cells with double nuclei and circular notches	2.00*** (0-3.5)	1.44 (0-3)	1.18 (0-1.5)
Integral proliferation index	4.00* (2-6)	5.11* (3-8)	3.00* (1-4.5)

Note: * $p < 0.017$ changes are significant between all groups in a pairwise comparison (1-2, 2-3, 1-3), ** $p < 0.017$ changes are reliable in pairs of groups 1-3 and 2-3, *** $p < 0.017$ changes are reliable in pairs of groups 1-2 and 1-3

Table 3

Indicators of early and late destruction of the cell nucleus, %

Indicators	Power, group 1 (n=27)	Gaming, group 2 (n=25)	Control group, group 3 (n=45)
Cells with perinuclear vacuole	0.36 (0-0.5)	2.00** (0-3)	0.36 (0-0.5)
Cells with chromatin condensation	4.64* (1-7)	19.00* (2-33)	1.73* (1-2.5)
Cells with nuclear vacuolization and onset of karyolysis	7.64 (4.5-10)	8.89 (3.25-9.33)	4.00 (1.5-6)
Cells with karyopyknosis	4.73 (2-9)	5.89 (5-8)	2.55 (1-4.5)
Cells with karyorrhexis	3.36 (1.5-4)	4.22 (1-4.5)	2.36 (0.5-4)
Cells with complete karyolysis	60.73 (38.5-75)	68.44 (41-134.5)	51.73 (21-63.75)
Apoptotic index	73.82 (51-83)	99.56 (35-154.5)	58.73 (25.5-71)

Note: * $p < 0.017$ changes are significant between all groups in a pairwise comparison (1-2, 2-3, 1-3), ** $p < 0.017$ changes are reliable in pairs of groups 1-2 and 2-3

ditional studies on large sample sizes to build predictive models such as "environmental risk factors - social risk factors - lifestyle - cytogenetic changes" .

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