

Football players' saliva crystallogenic property dynamics during different periods of the competition cycle

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Abstract:

The development of informative non-invasive methods for diagnosing the body reserve capabilities is important and necessary in sports practice to improve athletes' performance. *Research aim:* is to study the crystallogenic characteristics of oral fluid in different periods of the training and competitive cycle. *Research materials and methods.* 36 highly qualified athletes-football players (Russia) took part in the research project. All subjects had a single intake of oral fluid at different stages of the annual macrocycle: during the vacation period, in the middle of training camps, at the beginning, middle and end of the sports game season. Oral fluid intake (1 ml of saliva) was carried out in the morning athletes did not perform intense physical activity and did not experience psycho-emotional stress. The salivary fluid crystallization was analyzed using the method of classical crystallogeny. The obtained results interpretation was carried out using a parametric assessment of the characteristics of the biological medium crystallogenesis. The estimated parameters included crystallizability, the index of the structurality level, and the degree of the facies destruction, the marginal protein zone severity and the digital integral facies index. *Research results.* There was a significant decrease in the number of complex dendritic structures, the presence of single small crystals, significant structural damage and multiple fractures in the samples of the microslide at the beginning of training camps and at the end of the competitive season. This was manifested in the qualitative and quantitative transformation of the crystallogenic picture in the biological fluid, including changes in the digital integral facies index. These changes also depend on the individual functional reserves of the studied individuals, so, the fact confirms the need for their monitoring during different periods of the sports season. *Conclusions.* The conducted research demonstrated the informativeness and reliability of the biocrystalline testing method results, which allows predicting the state of tension of regulatory mechanisms affecting the performance of athletes.

Key Words: oral fluid, biocrystalline testing, adaptation to physical loads, saliva, annual macrocycle

Introduction

Human sports activity is accompanied by significant changes in morphofunctional, structural-metabolic and adaptive systems of the body (Nuutila et al., 2017). In sports medicine, an important area of medical control is the development of accessible and simple methods for a comprehensive assessment of athletes' reserve capabilities state (Amap et al., 2018; Tummala et al., 2018; Goodman et al., 2018; Vynohradov et al., 2021).

The use of non-invasive methods of monitoring the athlete's body adaptive reserves in the training process allows increasing his/her physical performance (Reneker, 2018; Lucas de Albuquerque Freire et al., 2020), make adjustments to the training process (Budzyn et al., 2018; Mirzaev, 2017), choose effective recovery measures and procedures (Chiriac Paul Bogdan et al., 2021) and prevent injuries during sports activities (Mardiana et al., 2021).

Maintaining the internal environment constancy in various conditions of physical activity requires human adaptive reserves consumption (Guzii et al., 2021; Bocharin et al., 2021a). Various methods are used to

assess athletes' reserve capabilities. This is a method for assessing vegetative status based on a study of heart rate variability (Aparecida Maria Catai et al., 2020; Bocharin et al., 2021b), study of variability of systemic hemodynamics parameters in different conditions of physical exertion (Martusevich et al., 2022a), registration of hormonal shifts in the student's body (Saad et al., 2020; Foretic et al., 2020). These methods make it possible to assess the adaptive response of the whole organism, while the metabolic status of persons engaged in sports activities is not sufficiently taken into account (Kokornaczyk et al., 2021). Some studies of oxidative metabolism are carried out mainly in the biochemical analysis of skeletal muscle activity (Bishop et al., 2019). The impact of regular physical exertion on metabolic processes in the body in the conditions of training camps during the competitive and off-season cycles has not been studied enough.

In this regard, attention should be paid to a promising modern direction in the field of sports medicine, namely, the crystalloscopic method of studying metabolic status (Dessau, Modis, 2011; Jordanishvili A.K., 2019). It is based on the study of one's own crystal-forming ability and the initiative potential of human biological fluid. It can be serum and blood plasma and salivary fluid. This method is needed for use as an integral test capable of comprehensively assessing the level of athletes' of any qualification functional reserves and metabolic status of their body (Hooper et al., 2015). Its practical implementation does not require laboratory conditions and the availability of expensive equipment on the basis of a medical institution. Testing is non-invasive, which additionally characterizes its prospects for studying the physical and chemical properties of the oral fluid of people who are engaged in physical culture and sports.

There are not enough reports in the scientific literature about the study of crystalloscopic saliva samples from highly qualified football players. We believe that our research will increase the amount of knowledge on this issue and will allow purposefully making adjustments to the training process of highly qualified athletes to improve the effectiveness of their performances at competitions.

Research aim is to study the crystalloscopic characteristics of oral fluid in different periods of the training and competitive cycle.

Material & methods

The research project consisted of a dynamic survey of 36 (aged 20-25) highly qualified football athletes who trained at the sports club in the Nizhny Novgorod region (Russia). The average sports experience was 9.4 ± 1.2 years. Before the start of the examination, all athletes gave written consent to participate in the research experiment. The work carried out does not violate the principles of the organization of biomedical research, according to the materials of the Helsinki Declaration of 2008.

In the morning, all subjects had a single intake of oral fluid (1 ml) into sterile dry test tubes at different periods of the training and competition cycle before performing intense physical exertion: during the vacation period, in the middle of training camps, at the beginning, middle and end of the sports season.

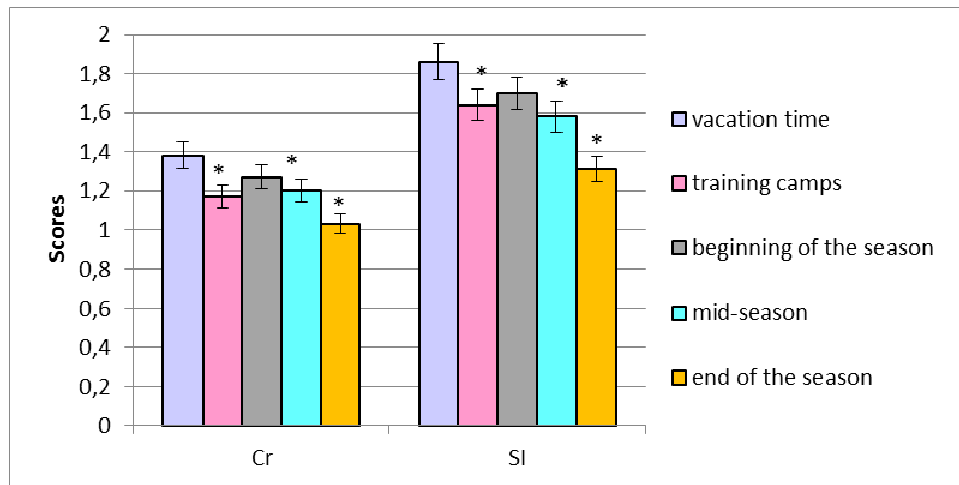
Individual features of salivary fluid crystallization were studied in the laboratory, with the performance of a dehydration test by classical crystalloscopy. The method used makes it possible to evaluate the crystal-forming properties of the biological fluid and obtain diagnostically significant facies (crystallized samples) and establish the presence of individual components in the analyzed substrate (Martusevich et al., 2022b).

The obtained data interpretation was carried out in points using static methods for assessing the characteristics of the crystallogenesis of the biological medium. At the same time, we used an algorithm for the quantitative description of the oral fluid crystallization, which included the following parameters: the structural index (SI), reflecting the composition of the sample elements and the crystallizability level (Cr). The latter is an indicator of the crystallogenesis process activity. In addition, we used semi-quantitative criteria in assessing the crystal-forming properties of biological fluid.

The degree of facies destruction (DDF), which is an integral indicator of the crystallogenesis process correctness in a biological fluid, and the severity of the marginal protein zone (EZ), which indicates the relative content of elements with a high molecular weight (protein) in the biological medium were studied. At the same time, we developed a new criterion allowing visualizing the overall assessment of the crystallogenic properties of a biological fluid, namely the integral facies index (DIFI). Statistical processing of the obtained data was performed in the software package Microsoft Excel 2003 and Statistica 6.1.

Results

Analysis of the crystalloscopic test results found that professional football players show a variation in the physical-chemical properties and component composition of the oral fluid depending on the stage of the training and competitive cycles. To study the adaptive reaction of the athletes' body, the indicators of the degree of crystallizability (Cr) and the structural index (SI) of the oral fluid were determined, Figure 1.



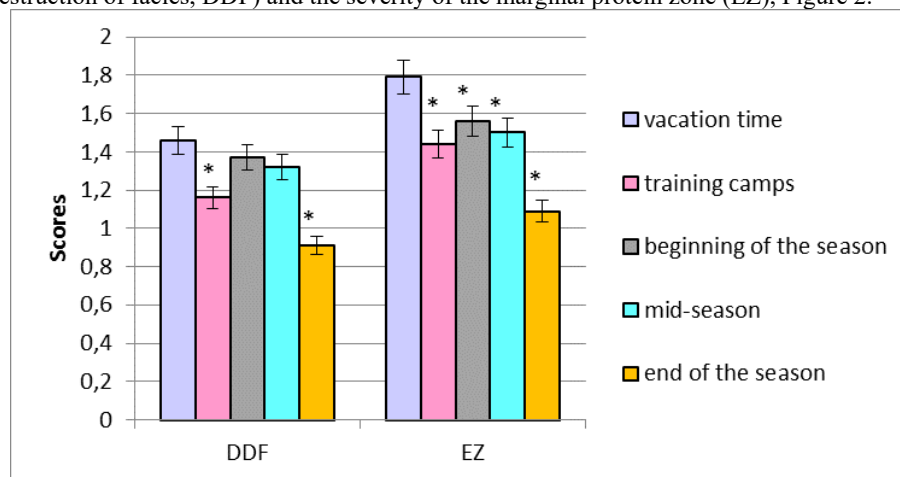
Note: * - significant differences between indicators, $p < 0.05$

Fig. 1. Monitoring of the crystallizability level and the structural index of the athletes' oral fluid samples in different periods of the annual macrocycle

It was found that the indicator of the activity of the players' own crystallogenesis of the oral fluid shifts to the disadaptive side by 25.4% (1.38 ± 0.02 points) at the beginning of the pre-competitive training camps and by the end of the playing season is 1.03 ± 0.01 points, $p < 0.05$. Crystallogenesis is characterized by the presence of single small crystals (in the field of view not > 10 units). At the beginning of the football playing season, there is a relative normalization of cellular formations, which may be associated with the restorative mechanisms of the body regulatory systems. In the middle of the playing season and by its end, there are no crystallization centers, which is accompanied by a significant decrease in the number and size of «crystal islands» in the obtained oral fluid samples.

A similar trend was observed when determining the index of microslides structurality. Before the start of the sports season, the presence of a dendritic component in the facies was noted, which was estimated at 1.86 ± 0.03 points. At the beginning of training camps, in the middle and at the end of the season, mainly amorphous formations and single dendritic bodies were observed, with a slight stabilization of this factor during the observation period at the beginning of the sports season. By the end of the football season, a 29.6% decrease in the structural index of the athletes' oral fluid samples was found to 1.31 ± 0.02 points, compared with the beginning of the season (1.86 points), $p < 0.05$. This fact indicates that intensive training during training camps and competitions have a significant negative impact on the functional state of the football players' body, causing a change in the saliva composition. This is manifested in an increase in the number of bodies of amorphous formation in the dried facies of oral fluid samples.

We have studied the parameter that characterizes the correctness of the crystallogenesis process course, when exogenous and endogenous factors are summed up in the formation of the dehydration process (the level of the degree of destruction of facies, DDF) and the severity of the marginal protein zone (EZ), Figure 2.



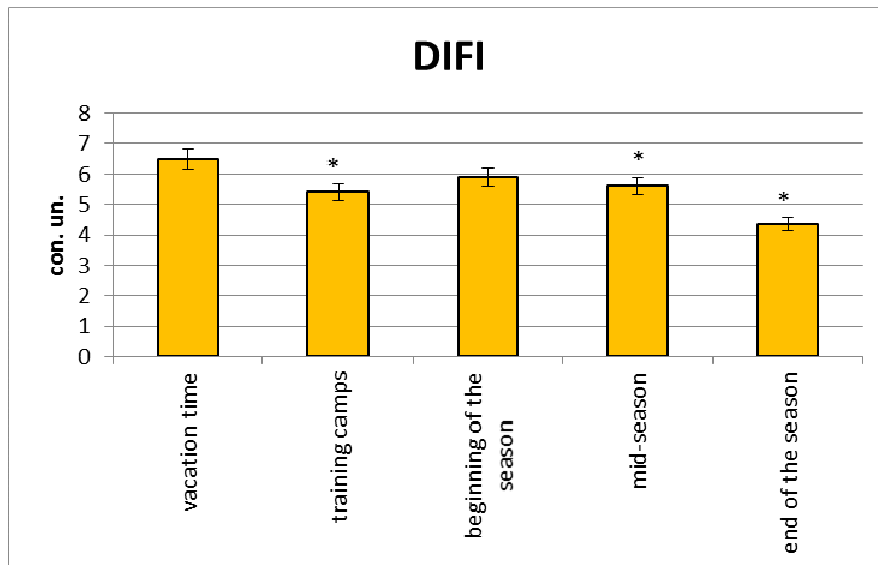
Note: * - significant differences between indicators, $p < 0.05$

Fig. 2. Monitoring of the degree of facies destruction level and the severity of the athletes' marginal protein zone of oral fluid samples in different periods of the annual macrocycle

At the beginning of training camps, the degree of facies destruction was 1.46 ± 0.04 points and decreased by 61.3% to 0.91 ± 0.01 points by the end of the competitive season, $p < 0.05$. During these periods, a large number of destroyed or altered structures and a violation of their integrity were noted in the samples. At the same time, after the athletes passed the training cycle during the pre-competitive training camps, there was a partial inactivation of destruction signs of elements in the biological fluid. Microslides of football players' dried oral fluid had qualitative criteria of peculiarities.

At the beginning of the pre-competitive training camps and especially at the end of the sports and competition season, deep faults of the marginal zone appeared. At the same time, the absence of a clear exposure of the marginal protein zone was observed throughout the facies of the dried specimens. At the beginning and middle of the sports season, the relative alignment of the marginal zone was noted, including that which had numerous faults. By the end of the game season, the value of the EZ indicator decreased by 36.2% (from 1.71 ± 0.05 to 1.09 ± 0.04 points), $p < 0.05$.

The digital integral facies index (DIFI) reflects the general changes in the metabolic properties of the biological fluid in athletes in different functional states and is calculated on the basis of the above quantitative and qualitative indicators (Figure 3).



Note: * - significant differences between indicators, $p < 0.05$

Fig. 3. Monitoring of the athletes' oral fluid samples digital integral facies index level in different periods of the annual macrocycle

The highest value of the DIFI index (6.49 ± 0.13 conventional units) was established during the athletes' vacation period. Further, at all stages of observation, we noted a decrease in the values of the DIFI indicator. By the end of the football season, the indicator decreased by 49.5% (relative to the DIFI indicator during the athletes' vacation) and amounted to 4.34 ± 0.03 conventional units, $p < 0.05$. Analysis of the decrease in DIFI results by the end of the sports season indicates the appearance of signs of the athletes' body regulatory systems significant tension against the background of training during sports camps and the development of a borderline state in them, close to the breakdown of adaptive reserves by the end of the competitive period.

As a result of the research, data indicating the possibility of using the oral fluid crystalloscopy method to determine the adaptive reserves of football players in different periods of the annual macrocycle were obtained. We believe that this type of medical and pedagogical control is necessary for the correction of training and competitions, taking into account metabolic changes in the athletes' body and for the relief of maladaptation conditions in them.

Discussion

Performing physical loadings causes diverse morphological and functional changes in the athlete's body. Diagnostics of morphofunctional changes in the human body is used in sports and clinical medicine to assess reserve capabilities and tolerance to physical exertion (Aparecida Maria Catai et al., 2020; Bocharin et al., 2021b; Martusevich et al., 2022a).

In the practice of sports activity, the problem of intersystem relations in the human body is particularly relevant (Belskaya et al., 2018). At the same time, an important aspect facing scientists in the study of physiological systems is the assessment of persons' performing significant physical activity adaptive capabilities

in terms of volume and intensity. Similar loads are registered in game sports among highly qualified athletes. A particularly important problem in such conditions of training and performances at competitions is the development of simple, affordable and effective means of diagnosing the reserve potential of athletes.

About 30 years ago, a crystalloscopic method for studying metabolic status was proposed, which became widely used in medical and sports practice (Dessau, Modis, 2011; Jordanishvili A.K., 2019). This method makes it possible to evaluate the adaptive response of the whole organism taking into account the metabolic status of persons engaged in sports activities (Martusevich et al., 2022b).

The value of a body's reserve capabilities comprehensive assessment increases with the use of diagnostic techniques at different stages of the training and competition cycle as a monitoring of athletes' readiness to perform physical loads. The materials obtained by us from the study of the highly qualified football players' oral fluid showed the dynamics of changes in their adaptive capabilities. In our project, the analysis of the athletes' oral fluid samples integral facies index results at different stages of the training and competitive cycles indicates that the highest reserve capabilities of the athletes' body are registered at the beginning of the competitive stage, the lowest reserve capabilities are noted at the end of the playing season.

Analysis of the results of other parameters of dried oral fluid samples crystallization study indicates that during the vacation period, athletes had a relatively good level of adaptation. Significant tension of regulatory mechanisms was noted at the start of training camps and in the middle of the season. During the end of the competition, athletes had a condition close to maladaptation disorders, which is confirmed by shifts in crystallization parameters, the index of the level of structurality, the degree of facies samples destruction, the degree of marginal protein zone severity and the digital integral facies index. Our results are consistent with the data of other authors who examined the oral fluid of athletes in other sports (Russo Krauss I, et. al., 2013; Vahdatahar E, et. al., 2021).

The development of non-invasive and informative tests to study the adaptive status of an organism is an effective method of evaluating the initial prediction of an individual athlete's performance results. From these positions, the results of human biological fluids own crystallization according to the scheme presented by us allow obtaining a reliable forecast about the functional status of persons professionally engaged in playing sports.

Conclusions

After the research project, it was found that the method of biocrystalline testing proved the informativeness and reliability of predicting the state of athletes' regulatory mechanisms tension results in different periods of their annual macrocycle. Monitoring of the athletes' oral fluid indicators showed the presence of significant tension of regulatory mechanisms at the beginning of training camps and in the middle of the football season. By the end of the competition period, the players had a condition close to maladaptation disorders. This is confirmed by a decrease in crystallization parameters by 25.4%, the index of the structurality level by 29.6%, the degree of facies samples destruction by 61.3%, the degree of the marginal protein zone EZ severity by 36.2% and the digital integral index of the facies DIFI by 49.5%. The presence of a qualitative and quantitative transformation of the crystalloscopic picture of the biological fluid in the digital integral facies indicator has been established.

During the beginning of training camps and at the end of the competition season, there is a significant decrease in the number of complex dendritic structures, the presence of single small crystals, large structural failures and multiple fractures in the microspecimen samples. These changes also depend on the individual functional reserves of the body of the persons' contingent selected for the study; therefore, monitoring is grounded at different periods of the sports season.

Conflicts of interest. The authors declare no conflict of interest.

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