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FEATURES OF BASIC INDICATORS OF ORAL FLUID IN CHILDREN AFTER HEMATOPOIETIC CELL TRANSPLANTATION (pilot study)

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SUMMARY

Relevance. Treatment of patients with refractory forms of acute lymphoblastic leukemia causes a number of complications from the organs and tissues of the oral cavity. It can be both early complications — cytotoxic reactions with the development of oral mucositis, and late, leading to a violation of the mineralization of hard tooth tissues, and to underdevelopment of the teeth. Oral fluid plays an important role in maintaining oral homeostasis. One of the most important functions that it performs is maintaining the physiological balance of the processes of re- and demineralization of tooth enamel. Since demineralization processes begin in childhood at higher pH values than in adults, when the hydrogen index decreases below 6.4, demineralization processes begin to prevail over remineralization, and the solubility of hydroxylapatite crystals which enamel consists of increases.

The **objective** was to assess the basic indicators of oral fluid and oral hygiene indices in this group of patients.

Methods and materials. During the examination of the oral cavity, the intensity of caries and hygiene indices were determined. For the study, oral fluid was collected, followed by determination of its viscosity and hydrogen index.

Results. These patients showed a lower level of oral fluid hydrogen index and a lower level of oral hygiene compared to healthy children. In addition, during the study, it was found that there is a direct relationship between the pH of the oral fluid and the time elapsed since the hematopoietic cell transplantation.

Conclusion. The detected changes are factors that predispose to an increase in the intensity of caries, and require early preventive measures.

Keywords: oral fluid, hydrogen index, oral fluid viscosity, hematological malignancies

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RELEVANCE

Leukemia is a malignant disease of blood cells characterized by unregulated proliferation of leukocytes in the bone marrow, peripheral blood and extramedullary regions [1]. In the structure of oncological morbidity in Russia, hematological malignant neoplasms occupy the 6th place (4.8 %). In the age group of patients from 0 to 29 years old, among the malignant neoplasms, mortality from tumors of the lymphatic and hematopoietic systems dominates (29.3 %) [2].

It should be noted that the components of the structure of pediatric oncological morbidity are represented, most often, by 7 — 8 nosologies, the leading ones of which are diseases of the lymphatic system and tissues producing blood cells. So, based on the data of I. V. Bondar (2002) [3], in the structure of the oncological incidence of children in the Russian Federation, the first place belongs to hemoblastoses (52.8 %), followed by neoplasms of the central nervous system (13.2 %), kidneys (7.6 %), skeleton (5.8 %) and soft

tissues (4.1 %). Acute lymphocytic leukemia (36.2 %), lympho- and reticulosarcoma (24.7 %), lymphogranulomatosis (16.4 %) and other acute leukemia (16.4 %) have a high incidence among blood tumors. Moreover, girls get sick less often than boys.

In the Russian Federation, among malignant tumors of the hematopoietic system and lymphoid tissue, children account for 38 – 40 % of leukemia. The frequency of occurrence of acute lymphoblastic leukemia in children is 4:100 000 with a ratio of 1:1.3 for girls and boys [4].

Today, with the use of programmed chemotherapy, 5-year relapse-free survival ranges from 77 to 92 %. A particular problem is the treatment of patients from the high-risk group, who are currently able to achieve complete clinical and hematological remission in 50 % of cases only with hematopoietic cell transplantation in combination with the use of chemotherapeutic drugs [5].

Treatment of patients with acute leukemia involves the use of high doses of methotrexate, cytarabine, asparaginase, imatinib, cyclophosphamide, etc. [6]. In more complex cases, modern chemotherapy programs are used, including nucleoside analogues, such as fludarabine and nelarabine, followed by hematopoietic cell transplantation in patients with refractory forms of acute lymphoblastic leukemia [7].

Because of cytostatic therapy causes a number of complications from organs and tissues of the oral cavity, the interest of pediatric dentists in the problem of treatment and rehabilitation of patients suffering from acute lymphoblastic leukemia is justified. Children undergoing treatment for lymphoblastic leukemia need to be monitored by a dentist, primarily because the drugs they receive can cause various cytotoxic reactions with the development of oral mucositis.

The use of high doses of cytotoxic drugs leads not only to damage to the mucous membrane of the oral cavity, as many researchers indicate [8 – 10], but also to a violation of the mineralization of hard tooth tissues, and to underdevelopment of teeth [10].

The system of various factors of local nonspecific oral immunity in leukemia and the pathogenetic mechanism of mucositis development have been studied quite extensively [11 – 17].

Violations of the composition and properties of the oral fluid are a factor in the development of various diseases of the teeth and periodontium in children suffering from this pathology [15].

According to I. V. Postnova (2003), in patients with acute lymphoblastic leukemia, there is a significant increase in the index of intensity of dental caries (DMFT). Also, the hygiene index according to Fedorov – Volodkina in these patients becomes almost twice as high as the norm. Thus, in children with acute lymphoblastic leukemia, the level of oral hygiene is significantly reduced, which also leads to an increase in the intensity of dental caries.

Oral fluid plays an important role in maintaining oral homeostasis. One of the most important functions that it performs is maintaining the physiological balance of the processes of re- and demineralization of tooth enamel. The correct implementation of this function is especially relevant in childhood during the maturation of enamel, i. e. immediately after teething. In childhood, demineralization processes begin at higher pH values than in adults [18]. With a decrease in pH below 6.4, demineralization processes begin to prevail over remineralization, and the solubility of the hydroxylapatite crystals, which enamel consists of increases [19].

After a literature review, most studies relate to children in the acute stage of the disease and undergoing polychemotherapy [6, 7, 9]. Since there is not so much data on the dental status of children suffering from leukemia and undergoing hematopoietic stem cell transplantation (HSCT), a study of this group of patients seems relevant.

A feature of this category of patients is the presence of an acute or chronic graft-versus-host reaction (GVHR) [20]. In these patients, there are no symptoms of leukemia in the oral cavity, such as hyperplastic gingivitis and ulcerative necrotic stomatitis. This is due to the fact that these patients are in remission after HSCT. They do not have manifestations of the underlying disease, but there are all manifestations of the activation of the transplant's immune system against normal cells of the patient, as well as against leukemia cells (the so-called action – graft-versus-leukemia effect).

The objective was to evaluate basic indicators of oral fluid and oral hygiene indices in this group of patients.

METHODS AND MATERIALS

Patients were divided into two groups. Group 1 consisted of 15 children aged 4 to 16 years, suffering from hemoblastoses and undergoing bone marrow transplantation. The ratio of boys to girls was 7/8. Group 2 included 9 practically healthy children aged 4 to 14 years, of which 4 were girls and 5 were boys. Due to the small sample of patients in the pilot study, age groups were not disaggregated.

For examination of patients, generally accepted clinical methods were used: history taking, objective dental examination with determination of DMFT and OHI-S Green-Vermillion.

The oral fluid viscosity was determined by the method of T. L. Redinova (1989) using a 2 ml micropipette. Mixed saliva was collected in the morning, before eating and brushing your teeth, in sterile plastic graduated tubes immediately before the study. Previously, the pipette was calibrated on distilled water taking into account the expired water in 5 seconds set by the stopwatch. Having installed the micropipette in an upright position, 1 ml of saliva was taken into it,

Table 1

The values of hydrogen index and oral fluid viscosity in the surveyed groups

Indicator	Group 1		Group 2	
	girls	boys	girls	boys
pH	(6.70±0.14)	(6.93±0.33)	(7.22±0.22)	(7.12±0.09)
Viscosity	(1.55±0.15)	(1.47±0.17)	(1.34±0.10)	(1.29±0.05)
p	≥0.05		≥0.05	

Table 2

Values of DMFT and Green-Vermilion hygiene index in the surveyed groups

Indicator	Group 1		Group 2	
	girls	boys	girls	boys
DMFT	(4.5±0.81)	(3.29±0.65)	(2.75±1.59)	(3.60±1.15)
OHI-S	(1.2±0.25)	(1.66±0.21)	(0.7±0.17)	(0.78±0.12)
p	≥0.05		≥0.05	

followed by measurement of the saliva expired over the same period of time.

The oral fluid viscosity was determined in relative units by the formula: $S_v = V_w \times W_v/V_s$, where V_w was the volume of expired water (ml), V_s was the volume of expired saliva (ml), S_v was the saliva viscosity (centipoises, cP), W_v – water viscosity (cP). We measured the saliva viscosity several times for each sample with the calculation of average values. Laboratory studies were performed no later than 60 min after collection of the material.

The pH of the oral fluid was determined by the potentiometric method three times for each sample, calculating the average value. We used a laboratory ionomer I-160MP, a silver chloride electrode EVL-1M3.1 «polumikro» (GZIP). The activity of the concentration of H + ions was measured using a measuring transducer and a set of electrodes.

The research materials were statistically processed using parametric analysis. Statistical analysis was performed using the STATISTICA 13.3 program.

When comparing the average values in normally distributed sets of quantitative data, the Student's t-test was calculated.

The obtained values of Student's t-test were evaluated by comparison with critical values. Differences in indicators were considered statistically significant at a significance level of $p<0.05$.

RESULTS AND DISCUSSION

A comparative analysis of the pH and oral fluid viscosity did not reveal significant differences between boys and girls in the survey groups. The results of measurements of pH and oral fluid viscosity are presented in table 1.

The results of the values of the Green – Vermilion hygiene index and DMFT in the examined groups are presented in table 2.

The average pH and oral fluid viscosity, DMFT and the Green-Vermilion hygiene index for the groups are summarized in table 3.

Thus, in the pilot study, it was not possible to identify statistically significant differences in the parameters of the oral fluid viscosity and DMFT in both groups of patients. All values corresponded to the average age norm.

When determining the OHI-S (Green – Vermillion), it was found that despite the fact that in both groups the index indicators were within satisfactory hygiene (0.7 – 1.6 points), the differences between the groups were statistically significant (Student's t-test was 2.91; $p = 0.00836$).

When analyzing the results of pH measurement of the oral fluid, it was found that in children who underwent hematopoietic cell transplantation due to lymphoblastic leukemia, the indicator was significantly lower than in their practically healthy peers.

Differences in the level of this trait in the compared groups are statistically significant (Student's t-test was 3.01; $p = 0.006685$). In addition, during the study it was found that there is a direct relationship between the pH of the oral fluid and the time elapsed since the hematopoietic cell transplantation (figure).

Trend of changes in the pH of the oral fluid after bone marrow transplantation in patients from group 1

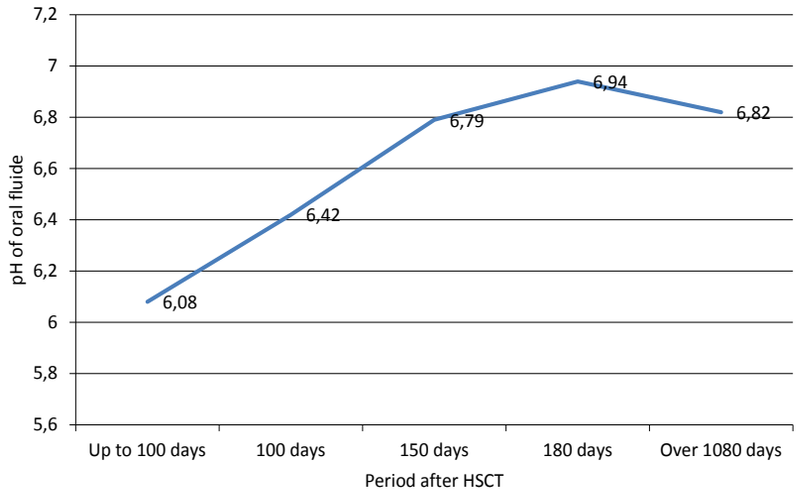
Thus, in children who underwent hematopoietic cell transplantation, the oral fluid hydrogen index, according to our studies, was initially lower than in healthy ones. Its level was normalized not earlier than

Table 3

Average indicators of the studied parameters in the groups

Indicator	Group 1	Group 2
pH	(6.70±0.12)	(7.17±0.10)
Viscosity	(1.54±0.10)*	(1.31±0.05)*
DMFT	(4.07±0.58)*	(3.22±0.84)*
OHI-S	(1.41±0.16)	(0.81±0.13)

* – differences are not statistically significant ($p \geq 0.05$).



Trend of changes in the pH of the oral fluid after bone marrow transplantation in patients from group 1

140 days after surgery. Most likely, this was not associated with the transplantation, but with the appointment of those drugs that this group of patients was forced to take and with the peculiarities of the course of GVHR. Treatment of leukemia is difficult to tolerate by children, has side effects, expressed in the development of gingivitis, xerostomia, caries.

Assessing the prevalence and intensity of caries in our groups, we did not see that the cariogenic situation in children with leukemia was worse than healthy children, which contradicts the literature. At the same time, we found a statistically significant decrease in the level of the hydrogen indicator of the oral fluid in children with hemoblastoses compared with a group of healthy patients of the same age group. According to literature data, pH = 6.2 units is the critical value of the hydrogen index at which the structural properties of saliva are violated [18]. When the pH of the oral fluid is shifted to the acid side, the activity of proteinases, including bacterial origin, increases. This contributes to the development of tooth demineralization and periodontal damage.

In our opinion, the absence in the observation group of a high intensity of damage to the hard tooth tissues, despite the low pH of the oral fluid, was due to the fact that all the patients were scanned by the dentist before the operation of HSCT, as well as the fact that most of the children included in group 1, were examined by us in the early stages after transplantation. According to the Order of the Ministry of Health of the Russian Federation dated December 20, 2012 «№ 1279н» «On the approval of the standard of specialized medical care after allogeneic bone marrow transplantation (examination and correction of treatment)» (Registered in the Ministry of Justice of the Russian Federation on February 25, 2013 «№ 27306»), dental care is not provided, but at the examination stage, it is assumed examination of the oral cavity 1 time in 201 days. Nevertheless, the data obtained by us cannot be ignored as evidence of the potential danger of developing major dental diseases in this category of patients, which, in our opinion, requires a review of the timing of the follow-up for a dentist in children with this group of diseases.

CONCLUSION

1. The cariogenic situation in children suffering from leukemia and undergoing hematopoietic cell transplantation in the early period after the intervention does not differ significantly from their healthy peers.

2. Treatment of the underlying disease (hemoblastosis) creates the prerequisites for the development of major diseases of the oral cavity. Given the age of patients, it is necessary to conduct regular individual preventive measures aimed at improving oral hygiene and lowering the acid solubility of tooth enamel.

3. In our opinion, it is important to establish the rules for monitoring and conducting preventive dental measures for children who have undergone HSCT for leukemia, starting from the first weeks after transplantation and for the entire rehabilitation period, taking into account adverse conditions for the maturation of enamel erupted teeth.

Conflict of interest

Authors declare no conflict of interest.

Compliance with ethical principles

The authors confirm that they respect the rights of the people participated in the study, including obtaining informed consent when it is necessary, and the rules of treatment of animals when they are used in the study. Author Guidelines contains the detailed information.

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