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Laboratory Methods Of Studying Children With Facial Developmental Faults

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ABSTRACT	Facial malformation in children is not only a medical but also a social problem all over the world. At the end of the 20th century, over the past two decades, there was a 2-fold increase in the frequency of birth of children with this pathology. In the structure of mortality of children in perinatal and early childhood, congenital defects occupy a significant place. And congenital orofascial defects are among the five most common intrauterine malformations.	
		malformations of the face, laboratory tests, children, congenital.

Keywords:

malformations of the face, laboratory tests, children, congenital, pathology, jaw

Relevance. One of the most relevant areas of research in the study of the development of ARGN is the study of the immunodeficiency state in expectant mothers. These factors can disrupt not only the formation of the maxillofacial region of the fetus, but also change the process of embryogenesis of many other organs. Early diagnosis of congenital malformations of the face, development and use of modern surgical methods of treatment, improvement of rehabilitation and early prevention of disability in children remains the leading direction of scientific research.

This research work corresponds to the tasks set in the State programs: "The Year of Harmoniously Developed Generation". approved by the decree of the President of the Republic of Uzbekistan dated January 27, 2010 No. PP-1271; "On the State Program for the Early Detection of Congenital and Hereditary Diseases for the Prevention of the Birth of People with Disabilities from Childhood for the Period of 2013-2017" dated March 12, 2013 No. PP-1235, and "Year of a Healthy Child" No. PP-2133 approved by the resolution of the President of the Republic of Uzbekistan of February 19, 2014

Teething is a physiological stage process characterized by the appearance of milk teeth, then permanent teeth in children. Teething is its axial movement from a non-functional position in the jawbone to a functional occlusion. The dynamics of this process depends on the degree of formation of the root, periodontium and is closely related to the development and growth of the craniofacial complex.

Depending on the operating mechanism of the delay in the eruption of the tooth, impaction aneruption and are classified: impact (collision) is a delay in the eruption of a tooth associated with the presence of a mechanical obstacle. The reason for this pathology may be a lack of space in the dentition against the background of crowding, the presence of a mucous barrier, supernumerary teeth and others; aneruption (aneruption - absence of eruption, absence of eruption) is a primary violation of the process of eruption of nonankylosed teeth with complete or partial absence of growth. It is known that dental buds appear in the fetus at about 6 weeks of intrauterine life. For another 1.5 months, the process of bone mineralization takes place.

During this period (up to the 13th week of pregnancy), the fetus from the mother's body takes calcium, phosphorus, protein and other substances necessary for its teeth. During this period, various diseases, unhealthy diet, a woman's intake of certain medications (for example, antibiotics of the tetracyclines group) causes anomalies in the number and shape of dental crowns in the fetus, and disrupts the strength and color of their enamel. By the time the baby is born, the crowns of 20 milk teeth are located deep in the alveolar processes of the jaws in a fully formed state. The process of their eruption is a kind of gradual increase in their volume and pushing outwards, in which they overcome the resistance of the bone tissue, mucous membrane. During this time, the gums become swollen and tender.

Teeth erupt in a certain sequence, the order of teething is disrupted in some diseases: rickets; genetic syndromes; lack of bookmarks of dental germs as a result of a complicated course of pregnancy; endocrine pathology. Various factors influence the eruption of deciduous teeth. The main importance in the process of teething is the human genotype, its constitution, while the role of various external environmental factors cannot be excluded.

The authors believe that children of older parents have teeth erupting earlier than children of younger parents. In the firstborn, teeth begin to erupt earlier than in the second and third children, in girls earlier than in boys, there is a direct relationship between the degree of prematurity of the child and the timing of the eruption of milk teeth. The peculiarities of the course of pregnancy in the mother also affect the physiology of teething.

Violations of the processes of eruption and change of teeth can be noted with pathology of the pituitary gland, refusal of breastfeeding, frequent acute respiratory diseases, pneumonia, sepsis of the newborn.

At the present stage, it is still significant to study the age-sex and regional characteristics of the eruption of permanent teeth as an important indicator of the biological maturity and health status of children. This is necessary when planning, implementing measures to prevent violations of biological development. Physiological and pathological aspects of teething and formation.

This study is based on the results of our own clinical, laboratory and instrumental observations.

Criteria for the inclusion of patients in the study:

- the presence of a clinical diagnosis of malformations of the face;

- parental consent to the child's participation in the study.

- boys and girls under 6 years old;

Criteria for excluding patients from the study: - children with serious illnesses.

In this regard, we have set the following goal

The purpose of the study is to carry out clinical and laboratory methods for the study of children with facial defects

Materials and research methods. This work is based on clinical and laboratory studies of 110 children with facial defects in the department of pediatric maxillofacial surgery of the regional hospital in Bukhara. This study is based on the results of our own clinical, laboratory and instrumental observations.

Criteria for the inclusion of patients in the study:

- the presence of a clinical diagnosis of malformations of the face;

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- boys and girls under 6 years old;

Criteria for excluding patients from the study:

- children with serious illnesses.

All examined 150 children were divided into 2 groups: comparison group 1 consisted of 40 (26.7%) without facial malformations, the main group - of 110 (73.3%) with facial malformations.

In the comparison group, there were 1.67 times more boys than girls, and 1.97 times more in the main group.

In children of the main group, cleft of the upper lip and palate was noted in 11 (10.0%), cleft of the hard and soft palate - 30 (27.3%), cleft of the upper lip - 52 (47.3%), cleft of the soft palate - 9 (8.2%), cleft palate - 1 (0.9%), anomaly of the upper jaw - 5 (4.5%), hemangioma of the lip - 2 (1.8%) cases. The immune status was studied in 110 children in children with malformations of the face using indicators of cellular and humoral namely: determination of the immunity, percentage of lymphocytes; determination of number of T- and **B-lymphocytes:** the determination of a subpopulation of Tlymphocytes - T-helpers and T-suppressors; the phenotype of immunocompetent cells was determined using monoclonal antibodies CD3, CD4, CD8, CD25, CD95 and others produced by Sorbent-LTD by the method of F. Yu. Garib et al. (1988).

For the study, blood was taken from the cubital vein in an amount of 3 ml in saline with heparin. The blood is diluted in a ratio of 1: 3 and layered on a mixture of ficoll-verografin with a solution density ingredient of 1.077 for the isolation of lymphocytes by sedimentation. The resulting mixture was centrifuged at 1500 rpm for 30 minutes. The resulting lymphocyte ring was aspirated into a clean tube and washed with saline twice. The volume of the solution was brought to 1 ml, and then the number of lymphocytes in the Goryaev chamber was determined, the number should be from 40 to 60 in 12.5 squares. Preparation: T-, B-, H-, S-, NK-systems.Human blood was taken, erythrocytes were washed, and a 50% erythrocyte suspension was prepared. Prepared 0.3% solution of chromium chloride in physiological saline. Mixed in a clean test tube 50 µl of 50% ervthrocyte mass, 50 µl of 0.3% chromium chloride solution and 3 µl of the corresponding monoclonal antibodies. The tube was shaken for 2-3 minutes, centrifuged for 2-3 minutes and the resulting suspension was washed with 3 ml of 0.9% sodium chloride solution for 10 minutes three times. The supernatant was decanted, and then the volume of the solution was brought to 1.25 ml the mixture was resuspended.We and conducted a comprehensive study of the state of the immune status in patients with ASLT. The complex of studies included the study of general humoral immunity by determining the level of serum immunoglobulins A, M and G and cellular immunity by determining the number **T-lymphocytes** of and their subpopulations.Determination of the level of

serum immunoglobulins A, M and G and immunoglobulins in the peripheral blood was carried out by the method of simple radial immunodiffusion according to the method of Mancini Get.al. (1965)using standard monospecific antisera of Russian production. When determining the total number of Tlymphocytes and their main subpopulations, the total number of B-lymphocytes and natural killer cells in the peripheral roof, we used the method of rosette formation (Garib F. Yu. Et al., 1995; Zalalieva M.V., 2004). To formulate the reaction of rosette formation, erythrocyte diagnostics were preliminarily prepared using formalinized human erythrocytes 1 (0) of the + group, which were loaded with Rh monoclonal antibodies using 0.3% chlorine chromium.

Venous blood was collected on heparin (24 U / ml), layered on a density gradient of ficollverographin (1.077 g / ml), and centrifuged at 1500 rpm for 30 min. The lymphocytes obtained in the interphase were transferred into a clean test tube, washed three times with medium 199 by centrifugation at 1000 rpm for 10 min, and the cell concentration was adjusted to 2 million / ml. Cell viability was determined in the trypan blue test. Usually, the cell suspension contained at least 95% of viable lymphocytes.

At the same time, blood was taken from a finger to determine the total number of leukocytes and calculate the leukocyte formula, which was necessary for the subsequent determination of not only the relative, but also the absolute number of immunocompetent cells. In the wells of the plate for immunological reactions, 100 µl of a suspension of lymphocytes and the corresponding erythrocyte diagnosticum were poured, the mixture was precipitated by centrifugation at 1000 rpm for 5 minutes and incubated for 60 min at + 4 ° C. The reaction was stopped by adding an equal amount of glutaraldehyde (final concentration 0.06%, + 4 ° C, 20 min). The contents of the wells were shaken out with a sharp movement, the wells were filled with distilled water and stored until the results were counted (from 2-3 days to 1 week).

Results and discussion. On the day of counting, the contents of the wells were shaken out, an equal amount (100 μ l) of Zadorozhny-Dozmorov dye was poured into the wells to stain the nuclei of lymphocytes, the cells were resuspended, a "crushed drop" preparation was prepared, and the number of rosette-forming cells (ROC) was counted in relation to free lymphocytes using an increase biological microscope 7 × 40.

We counted 200 lymphocytes, isolating ROK from them, then found the average value in percent. For ROC, a lymphocyte with 3 or more erythrocytes tightly attached to its surface was taken.

Conclusions.

1. In children of the main group, cleft of the upper lip and palate was noted in 11 (10.0%), cleft of the hard and soft palate - 30 (27.3%), cleft of the upper lip - 52 (47.3%), cleft of the soft palate - 9 (8.2%), cleft palate - 1 (0.9%), upper jaw anomaly - 5 (4.5%), lip hemangioma - 2 (1.8%) cases

2. Also in our research we used the method of paired sera, in which specific IgG and IgM are determined using the ELISA method. The principle of the method is that the patient's blood is taken twice within 10-14 days. After that, these 2 samples are simultaneously examined under the same conditions for the presence of specific antibodies using ELISA. An increase or decrease in the titers of specific immunoglobulins of the IgG and IgM class makes it possible to draw an appropriate conclusion and make a diagnosis.

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