# Determination of the criteria of early caries diagnostics in children of different ethnic groups domiciled in areas biogeochemically deficient in fluorine and iodine\*

# Ustalenie kryteriów diagnozy wczesnych zmian próchnicowych u dzieci w różnych grupach etnicznych zamieszkujących obszary z niedoborem fluoru i jodu\*

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#### Abstract

Aim of the study. To determine the criteria for early diagnosis of carious lesions in children of different ethnic groups permanently exposed to conditions of fluorine and iodine deficiency. Materials and methods. Microbial environment, biochemical parameters and immunological status of saliva in 146 children aged 3-8 years were determined. The study subjects belonged to different ethnic groups. Statistical evaluation of the results was conducted. Results. Dependences of biochemical parameters of microbial flora and local immune characteristics of saliva were determined. Correlation of dependence between pH of saliva, content of ionized calcium and alkaline phosphatase of Ck = 0.825, p < 0.01 was also determined. Direct correlation dependence between pH and phosphorus content in the saliva from Ck = -0.876, p < 0.01 has been proven, which shows the inverse correlation dependence

## **KEYWORDS:**

children, caries, biogeochemical fluorine and iodine deficiency, microbial environment, saliva, oral microorganisms, biochemical parameters, local immunity

#### Streszczenie

Cel pracy. Ustalenie kryteriów dla diagnostyki wczesnych zmian próchnicowych u dzieci z różnych grup etnicznych zamieszkałych w regionach ubogich we fluor i jod. Material i metody. Określono środowisko drobnoustrojowe, parametry biochemiczne i status odpornościowyślinyu 146 dzieci w wieku 3-8 lat. Badani reprezentowali różne grupy etniczne. Przeprowadzono analizę statystyczną. Wyniki. Stwierdzono zależności między parametrami mikroflory a charakterystyką odpornościowa jamy ustnej. Zależność między pH śliny, zawartością jonów wapnia i fosfatazy zasadowej wynosiła Ck = 0.825, p < 0.01. Wykazano bezpośrednią zależność między pH a zawartością fosforu w ślinie Ck = -0.876, p <0.01, co dowodzi odwrotnej zależności między intensywnościa próchnicy a pH śliny Ck = -0.09, p<0.01. Wnioski. Wykazano następujące zależności: bezpośrednią – pomiędzy pH a zawartością fosforu;

#### **HASŁA INDEKSOWE:**

dzieci, próchnica, niedobór fluoru i jodu, środowisko drobnoustrojowe, ślina, drobnoustroje jamy ustnej, parametry biochemiczne, odporność miejscowa

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of intensity of caries and pH of saliva Ck = -0.09, p < 0.01. **Conclusions.** The following correlation dependences have been proven: direct – between pH and phosphorus content; inverse – between pH ionized calcium content and alkaline phosphatase; between the intensity of caries and pH; linear – between the concentration of Streptococcus sobrinus and intensity of caries; exponential – between sIgA concentration and Streptococcus sobrinus; microflora and pH, alkaline phosphatase and intensity of caries. odwrotną – pomiędzy pH jonów wapnia i zasadowej fosfatazy; pomiędzy intensywnością próchnicy a pH; liniową – pomiędzy stężeniem Streptococcus sobrinus a intensywnościa próchnicy; wykładniczą – pomiędzy stężeniem sIgA a Streptococcus sobrinus; mikroflorą a pH, fosfatazą zasadową a intesywnością próchnicy.

## Introduction

Much attention is given by leading researchers to caries etiology and methods of its prevention and treatment.<sup>1,7,8,10,12,16,17</sup> The role of microbial coenosis and biochemical parameters of saliva in the formation of caries and its progression has been established.<sup>4-6,11,13,15,20-22</sup> A significant factor in the formation and progression of caries is the environmental conditions in which the child resides, including biogeochemical deficiency of microelements such as fluorine and iodine. Transcarpathian region is an example of such an environment.<sup>3,5,9,10</sup>

Regarding the prevalence and intensity of caries in children who permanently live in conditions of biogeochemical fluorine and iodine deficiency epidemiological studies have been conducted and emergency indices of morbidity, namely, the prevalence of caries of deciduous teeth, that is 98.3  $\pm$  2.2%, and in permanent teeth 91.4  $\pm$  2.3 have been established.<sup>18,19</sup>

A sufficient number of scientific studies have been devoted to therapeutic and preventive measures.<sup>2,6,23-25</sup> However, the issue of preclinical diagnosis of early forms of caries is especially valid, and requires determination of effective parameters of correction, which will be presented in this study.

The aim is to determine the criteria for early diagnosis of caries in children of different ethnic groups permanently living in conditions of biogeochemical fluorine and iodine deficiency.

#### **Materials and methods**

For creation of prognostic model of formation and forecasting of lesions hard tissues of teeth were used to evaluate criteria identified in the course of the clinical and laboratory research, namely, the degree of active caries in 146 children of preschool and primary school age, the level of oral microbial coenosis, the degree of mouth's dysbiosis, levels of secretory immunoglobulin A in saliva, biochemical parameters of saliva, content of total and ionized calcium, phosphorus, alkaline phosphatase.

Biochemical parameters of saliva, such as total and ionized calcium, phosphorus, alkaline phosphatase were determined and pH measured using Dimension RxL Max Integrated Chemistry System (Siemens) apparatus by method of immunofluorescence.

Microbial environment was determined by selective culture of saliva on the following nutrient media: meat peptonic selective agar, semi-selective medium, semi-liquid environment, environment Endo, bismuth-sulfite agar, enterococcus agar, laktobak adar, bifidum, Bifidobacterium Agar for cultivation.

Determination of secretory IgA was carried out with "sandwich" variant of enzyme-linked immunosorbent assay by measuring the value of optical density of holes tablet content by vertical scanning on the photometer with wavelength of 450 nm. Compliance of statistical distribution empirical indicators according to theoretical normal Gaussian distribution was assessed using Kolmogorov-Smirnov criterion (K.-S.) and Shapiro-Wilks (W), with p – an indicator of reliability.

Also paired and Pearson's partial correlations (r) with reliability interval (p), based on absolute data were analyzed. Methods of nonparametric statistics – Spearman's rank correlation (rs) were also used.

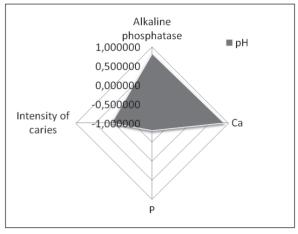
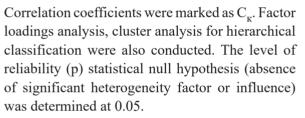


Fig. 1. Gradation image of correlation dependence of quantity alkaline phosphatase, phosphorus and ionized calcium from pH of saliva.



Interpretation calculations were performed on a personal computer using licensed software "MS Excel 7" operating system "Windows XP".

#### Results

Dependences of biochemical parameters of microbial flora and local immunity of saliva were determined. Figure 1 shows inverse correlation of dependence between pH of saliva, content of ionized calcium and alkaline phosphatase of Ck = 0.825, p<0.01.

Direct correlation dependence between pH and phosphorus content in saliva from Ck = -0.876, p <0.01 was also proven, which shows the inverse correlation dependence of intensity of caries and pH of saliva Ck = -0.09, p <0.01 (Fig. 2).

# Discussion

The analysis revealed the correlations of intercommunication dependence of concentrations of secretory immunoglobulin A and *Streptococcus sobrinus* content in saliva.

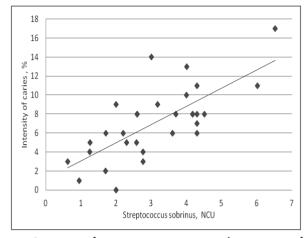


Fig 2. Diagram of scattering: caries intensity and concentration of Streptococcus sobrinus in saliva.

According to Figure 3, there is exponential dependence of concentrations of secretory immunoglobulin A and *Streptococcus sobrinus* content, which means that with increasing concentration of *Streptococcus sobrinus* the concentration of sIgA also increases; the maximum number of points is localized within average indices; the absence of microorganisms in the mouth corresponds to a decrease of sIgA concentration.

Figure 4 recreates gradation dependence of species diversity and quantity of microflora of saliva and intensity of caries, taking into account the pH and alkaline phosphatase.

Correlation dependency was revealed between the number of microorganisms and alkaline phosphatase levels, including: *Streptococcus sobrinus* (C $\kappa$ =0.21), *Enterococcus faecalis* (C $\kappa$ =0.39),*Escherichiacoli*(C $\kappa$ =0.2),*Enterococcus faecium* (C $\kappa$ =-0.4), *Proteus vulgaris* (C $\kappa$ =-0.65). *S.aureus* (C $\kappa$ =-0.63), *Pseudomonadas aeruginosa* (C $\kappa$ =-0.21). *Citrobacter Freundi* (C $\kappa$ =-0.2).

A direct relationship of caries intensity and concentration of *Streptococcus sobrinus* (C $\kappa$ =-0.57), *Enterococcus faecalis* (C $\kappa$ =0.55), *Escherichia coli* (C $\kappa$ =0.15), *Enterococcus faecium* (C $\kappa$ =0.12), *Proteus vulgaris* (C $\kappa$ =0.16), *S.aureus* (C $\kappa$ =0.26), *Pseudomonadas aeruginosa* (C $\kappa$ =0.19), *Citrobacter Freundi* (C $\kappa$ =0.11) was also proven. A

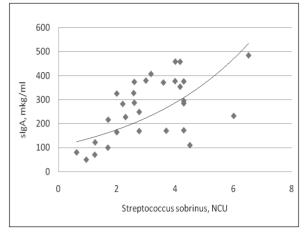


Fig. 3. Diagram of scattering slgA and Streptococcus sobrinus

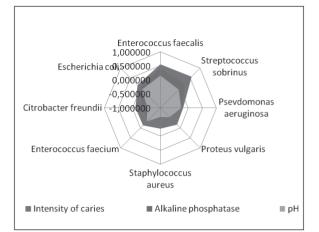


Fig. 4. Gradation image of correlation between saliva microflora and pH, alkaline phosphatase and intensity of caries.

similar direct correlation dependence was found between the number of microorganisms and pH level of saliva: *Streptococcus sobrinus* (C $\kappa$ =0.1), *Enterococcus faecalis* (C $\kappa$ =0.16), *Escherichia coli* (C $\kappa$ =0.1), *Enterococcus faecium* (C $\kappa$ =0.31), *Proteus vulgaris* (C $\kappa$ =0.53). *S.aureus* (C $\kappa$ =0.67), *Pseudomonadas aeruginosa* (C $\kappa$ =0.25), *Citrobacter Freundi* (C $\kappa$ =0.1).

The intensity of caries correlates with the number of *Streptococcus sobrinus* in saliva. Figure 2 shows observed linear dependence of the disease on the number of microorganisms in saliva: the higher the concentration of microorganisms, the higher the intensity of the incidence of caries. Therefore, *Streptococcus sobrinus* is the main cariogenic microorganism.

To create a prognostic model and predict hard dental tissues lesions, evaluation criteria, identified in the course of the clinical and laboratory research were used, namely, the level of dental caries's activity in preschool and early school age, the level of microbial coenosis in the mouth, the degree of dysbiosis in the mouth, the levels of secretory immunoglobulin A in saliva, biochemical parameters of saliva, content of total and ionized calcium, phosphorus, alkaline phosphatase. In all the children of the Romany ethnic group a significant increase of sIg A was found; increased concentrations of secretory IgA is a marker of violation of microbial coenosis of the oral cavity and indication of progression of caries.

# Conclusions

Established prognostic statistical model was based on the following correlation dependencies: direct – between pH and phosphorus content; inverse - between pH ionized calcium content and alkaline phosphatase; between the intensity of caries and pH; linear – between the concentration of Streptococcus sobrinus and intensity of caries; exponential - between sIgA concentration and Streptococcus sobrinus; microflora and pH, alkaline phosphatase and intensity of caries. Correlation dependence between the content of secretory immunoglobulin A and caries activity was determined, and with the increasing complexity of caries the activity sIg A concentration increased in direct proportion in children of mixed ethnic groups compared with those in healthy children.

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