

Microbiological approaches to caries risk determination in children

Oksana I Godovanets¹, Anastasiia V Kotelban^{2*}

To Cite:

Godovanets OI, Kotelban AV. Microbiological approaches to caries risk determination in children. Medical Science 2022; 26:ms304e2316.
doi: <https://doi.org/10.54905/disssi/v26i125/ms304e2316>

Authors' Affiliation:

¹Doctor of Medical Sciences, Professor, Head of the Department of Pediatric Dentistry, Bukovynian State Medical University, Ukraine; <https://orcid.org/0000-0002-1889-3893>

²Associate Professor of the Department of Pediatric Dentistry, Bukovynian State Medical University, Ukraine; <https://orcid.org/0000-0001-8266-3454>

*Corresponding author

Associate Professor of the Department of Pediatric Dentistry, Bukovynian State Medical University, Ukraine
Email: kotelban_anastasiia@bsmu.edu.ua

Peer-Review History

Received: 26 May 2022
Reviewed & Revised: 29/May/2022 to 20/July/2022
Accepted: 21 July 2022
Published: 26 July 2022

Peer-review Method

External peer-review was done through double-blind method.

URL: <https://www.discoveryjournals.org/medicalscience>



This work is licensed under a Creative Commons Attribution 4.0 International License.

ABSTRACT

The aim is to evaluate the microbial factor in the development of caries of temporary teeth. 6-year-children were examined at Bukovyna region (n=73). As cariogenic microflora detection, we were used the CRT bacteria kit (Ivoclar Vivadent, Liechtenstein). We found (<10⁴) CFU of streptococci at a low level of caries intensity in the 55.55% of children. In the case of a high level of caries intensity, (>10⁶) CFUs of streptococci were shown in 46.67% of children and (10⁵ - 10⁶) CFUs of streptococci in 33.33% of children. Regarding lactobacilli, the low level of intensity of caries is characterized by the concentration (<10⁴) of CFU and high level – (10⁴ - 10⁵) CFU of lactobacilli. Thus, we found a high concentration of the main cariogenic microorganisms in children with caries of temporary teeth. As the level of caries intensity increases, there is a probable increase in the titer of streptococci and lactobacilli.

Keywords: dental health, caries, CRT bacteria kit, *Streptococcus mutans*, *Lactobacillus salivarius*.

1. INTRODUCTION

Since ancient times dental caries has been the undisputed leader among all dental diseases. The key to its development, regardless of age, is the oral microflora (Shakovets and Terekhova, 2015). It can be aerobic and anaerobic microorganisms, as various types of fungi, viruses, protozoa. Among them the most common are facultative anaerobes, in particular α -hemolytic streptococci. The second ones are lactobacilli. Mostly oral microorganisms are non-pathogenic, but the other ones are conditionally pathogenic. Its composition can change under the influence of various adverse factors that reduce the body's defense mechanisms. As a result, there are quantitative and qualitative shifts in the population of microorganisms in the oral ecosystem. It means that it exists clearly relation in the ecosystem "microorganisms – oral cavity – external factors" that are determined all sequence of caries development (Petrushanko et al., 2013). That's why microflora of the oral cavity is divided into cariogenic and non-cariogenic. And it remains unresolved question: how specific are "cariogenic" microorganisms for the disease? According to Marsh and Percival (2006), the "aggressiveness" of

cariogenic bacteria is determined by the actual environmental situation in plaque.

In addition, most epidemiological studies have shown that the high level of *Str. mutans* and *Lactobacillus* in the oral cavity are associated with a high prevalence of dental caries (Ranganathan and Akhila, 2019; Lemos, 2019; Al-okbi et al., 2022). It is well known that these microorganisms have a high degree of adhesion to tooth enamel and have significant acid-producing function of teeth (Priya et al., 2015). Therefore, it needs further epidemiological research, the search for improved diagnostic methods and new treatment and prevention programs.

Aim

Assess the microbial factor in the development of caries of temporary teeth by determining the titer in the oral cavity of the main cariogenic microorganisms.

2. MATERIALS AND METHODS

For this purpose, 73 children aged 6 living in Bukovyna were examined at the clinical base of the Department of Pediatric Dentistry at the Chernivtsi Regional Children Clinical Hospital (Chernivtsi) from April 2021- January 2022. The children were divided into 2 groups: I – examined with caries of temporary teeth of varying intensity and II – dental healthy. To determine the level of intensity of dental caries, the LIC index was used (Leus, 2009), according to which low, medium and high levels were determined. The titer of cariogenic microflora was determined in dental healthy children and children with different caries intensity according to the CRT bacteria kit test (Ivoclar Vivadent, Liechtenstein) according to the manufacturer's instructions. To obtain reliable results, parents of children were warned to refrain from eating, drinking, brushing their teeth for at least 1 hour before taking saliva samples. The number of colonies of *Streptococcus mutans* and *Lactobacillus salivarius* was estimated by comparing the results with the standard scale of the CRT bacteria kit.

Statistically evaluated the probability of the obtained results in the case of normal distribution of both samples by Student-Fisher test, in other cases – U-Wilkson for independent samples and T-Wilkson test for dependent samples using the program "STATISTICA 6.1" ("StatSoftInc") № AGAR909E415822FA (Stirling, 2013).

3. RESULTS AND DISCUSSION

As a result of our research, the intensity of caries of temporary teeth was 3.78 ± 0.32 points, which corresponds to the average level (table 1). According to the LIC index, we have identified three main levels of caries intensity of temporary teeth: low, medium and high. The largest number of children (38.29%) with a low level of caries intensity and an index value of 1.58 ± 0.19 points. Medium (3.04 ± 0.45 points) and high (6.72 ± 0.32) levels were found in 29.78% and 31.91% of respondents, respectively. In most developed countries there is a similar trend: 20% of those surveyed account for 80% of all carious lesions (Shakovets and Terekhova, 2015).

Table 1 The level of intensity of caries of temporary teeth in children ($M \pm m$), points

Indicator	dmf (n=47)	LIC		
		low (n=18)	middle (n=14)	high (n=15)
	3.78 ± 0.32	1.58 ± 0.19	3.04 ± 0.45	6.72 ± 0.32

The leading factor in the development of dental caries, regardless of age, is the microbial factor. It is well known that bacteria of the species *S. mutans* play an important role in initiating carious lesions of the teeth. After all, they together with *S. sobrinus* convert sucrose into lactic acid (Dianawati et al., 2020). Because of this process, the acidic environment of the oral cavity causes a decrease in the resistance of the enamel. In addition, the authors claim that *S. mutans* is responsible for creating a retention niche for the colonization of lactobacilli and possibly other cariogenic bacteria (Caufield et al., 2015). In the vast majority of children, cariogenic bacteria in the oral cavity, in particular *S. mutans*, appear after the eruption of deciduous teeth. It is interesting that this streptococcus is found in children with and without caries (Ghazal et al., 2018).

According to Skatova (2010), determining the amount of these bacteria in saliva, along with other clinical data of the patient, is an important component of assessing the level of risk of caries occurrence. While determining the amount of *S. mutans*, we found a probable increase in the titer of its colonies in children with different levels of caries intensity compared to dental healthy children (table 2). With a low level of caries intensity in the vast majority (55.55%) of children found ($<10^4$) CFU of streptococci, in the third of the examined – ($10^4 - 10^5$) CFU of streptococci, 5.56% – ($10^5 - 10^6$) CFU of streptococci and also in 5.56% – ($> 10^6$) CFU of streptococci. For the average level of caries intensity, the most characteristic is the number of colonies of microorganisms with a

concentration of ($10^5 - 10^6$) CFU in 57.14% of subjects and ($> 10^6$) CFU – in 21.42%. In the case of a high level of caries intensity, ($> 10^6$) CFU of streptococci were shown in 46.67% of children and ($10^5 - 10^6$) CFU of streptococci in 33.33% of children.

According to Skatova et al., (2010) at the titer of these microorganisms ($> 5 \times 10^5$) CFU / ml of saliva in patients there is a high risk of developing carious lesions in the future. The obtained results by the author and our results reflect the same trends, because in the vast majority (54.38%) of our surveyed children with medium and high levels of intensity found ($> 5 \times 10^5$) CFU of streptococci. According to Petrushanko et al., (2013), under conditions of high activity of the carious process, the population density of *S. mutans* increased in 20 times. These results are comparable to ours and confirm the decisive role of streptococci in the occurrence and deepening of carious dental lesions in children. Regarding the levels of growth of streptococcal colonies, scanty growth prevailed at low levels of caries intensity, moderate – at medium and abundant – at high.

Table 2 The content of *Streptococcus mutans* in the oral cavity of children depending on the degree of caries intensity, %

Number of colonies, CFU	level of caries intensity			dental healthy, n=26
	low, (n=18)	middle, (n=14)	high, (n=15)	
$<10^4$	55.55 % (10)*	7.12 % (1)*	6.67 % (1)*	88.46 % (23)
$10^4 - 10^5$	33.33 % (6)*	14.32 % (2)*	13.33 % (2)*	7.69 % (2)
$10^5 - 10^6$	5.56 % (1)	57.14 % (8)*	33.33 % (5)*	3.85 % (1)
$>10^6$	5.56 % (1)	21.42 % (3)	46.67 % (7)	–

Notes. 1. * – the difference between the rates of children with caries of temporary teeth and dentistically healthy is probable ($p < 0,05$).

Regarding the amount of *Lactobacillus salivarius*, there is also a significant difference in the performance of children with carious and intact teeth (table 3).

Table 3 The content of *Lactobacillus salivarius* in the oral cavity of children depending on the caries intensity, %

Number of colonies, CFU	level of caries intensity			dental healthy, (n=26)
	low, (n=18)	middle, (n=14)	high, (n=15)	
$<10^4$	44.44% (8)*	50.00% (7)*	26.68% (4)*	96.15% (25)
$10^4 - 10^5$	38.89% (7)*	21.42% (3)*	53.33% (8)*	3.85% (1)
$10^5 - 10^6$	16.67% (3)	21.42% (3)	13.33% (2)	–
$>10^6$	0	7.16% (1)	6.66% (1)	–

Notes. 1. * – the difference between the rates of children with caries of temporary teeth and dental healthy is probable ($p < 0,05$).

The low level of intensity is characterized by the following titers of lactobacilli colonies: ($<10^4$) CFU – in 44.44% of subjects, ($10^4 - 10^5$) CFU – in 38.89%, ($10^5 - 10^6$) CFU – in 16.67%. No one child was inoculated with these microorganisms more than 10^6 CFU. Under medium conditions, half of the children were diagnosed with (10^4) CFU of lactobacilli, 1 child (7.16%) – more than 10^6 CFU, the rest – equally, respectively ($10^4 - 10^5$) CFU and ($10^5 - 10^6$) CFU. Regarding the high level of intensity, in the vast majority of children (53.33%) ($10^4 - 10^5$) CFU of the lactobacilli, in the fourth part – ($<10^4$) CFU, in 13.33% – ($10^5 - 10^6$) CFU and in 6, 66% – more than 10^6 CFU. Low and medium levels of intensity is characterised by insignificant growth of colonies of microorganisms and for high – moderate.

By Petrushanko et al., (2013), it was identified *Lactobacillus spp.* in the amount of 4.1 ± 0.05 lg CFU / ml in 43% of people with intact teeth. At dmf > 6 *Lactobacillus spp.* in the amount of 4.9 ± 0.18 lg CFU / ml, which is probably higher than in dental healthy children. The results of the authors also indicate an increase in the titer of lactobacilli in patients with carious lesions and their impact on the development of carious processes in the temporary teeth. However, Dianawati (2020) had an interesting study. It was determined level of *S. mutans* and *S. sobrinus* by different intensities of dental caries. The authors did not find a statistical difference between the detection of *S. mutans* and *S. sobrinus* and between high and low levels of caries. The authors believe that carious lesions of the first molars are due to other factors, including bacterial virulence, diet, environmental influences, socio-demographic or socio-economic aspects, level of knowledge and behavior.

4. CONCLUSION

We found a greater number of major cariogenic microorganisms in children affected caries of temporary teeth compared to dental healthy children. As for streptococci, in the vast majority of children with caries (76.6%) $> 10^4$ CFU of microorganisms were detected, while in dental healthy children the same number of colonies - only in 12.54% of examined. Lactobacilli with an amount of $> 10^4$ CFU was detected in 59.57% of children with caries of temporary teeth and only in 3.85% of dental healthy ones. With increasing caries intensity, there is a probable increase in the titer of streptococci and lactobacilli: at low caries intensity in the vast majority (55.55% of surveyed) found ($< 10^4$) CFU of streptococci and the same number of lactobacilli in 44.44% of children. Regarding the average level of caries intensity, 57.14% of the examined revealed ($10^5 - 10^6$) CFU of streptococci and 50.00% of children ($< 10^4$) CFU of lactobacilli. The high level of caries intensity is characterized by ($> 10^6$) CFU of streptococci in 46.67% of children and ($10^4 - 10^5$) CFU of lactobacilli in 53.33% of children.

Ethical approval

The research proposal was approved by the by the Medical Ethics Committee of Bukovynian State Medical University, Chernivtsi, Ukraine (registration number: 21.04.22/123).

Authors' contribution

concept and design of the study, editing – Oksana I. Godovanets; concept and design of the study, collection of material, processing, text writing – Anastasiia V. Kotelban.

Funding

This study has not received any external funding.

Conflicts of interest

The authors declare that there are no conflicts of interests.

Data and materials availability

All data associated with this study are present in the paper.

REFERENCES AND NOTES

- Al-okbi MA, Kosyrev T, Katbeh I, Tuturov N, Voeykova O, Birukov A. Plaque deposition around conventional and self-ligating brackets in patients undergoing orthodontic treatment. *Medical Science* 2022; 26:ms44e2059. doi: 10.54905/disssi/v26i120/ms44e2059
- Caufield PW, Schön CN, Saraithong P, Li Y, Argimón S. Oral lactobacilli and dental caries: a model for niche adaptation in humans. *J Dent Res* 2015; 94(9):110–18. doi: 10.1177/0022034515576052.
- Dianawati N, Setyarini W, Widjiastuti I, Ridwan RD, Kuntaman K. The distribution of *Streptococcus mutans* and *Streptococcus sobrinus* in children with dental caries severity level. *Dent J (Majalah Kedokteran Gigi)* 2020; 53(1):36–39. doi: 10.20473/j.djmk.v53.i1.p36-39.
- Ghazal T, Levy SM, Childers NK, Carter DJ, Caplana JJ, Warren J, Cavanaugh JE. Mutans Streptococci and Dental Caries: A New Statistical Modeling Approach. *Caries Res* 2018; 52:246–52. doi: 10.1159/000486103.
- Lemos JA. The biology of *Streptococcus mutans*. *Gram-Posit. Pathog* 2019; 1:435–48. doi: 10.1128/9781683670131.ch27.
- Leus PA. Clinical index assessment of dental status: textbook. Minsk: BGMU; 2009. pp 60.
- Petrushanko TO, Chereda VV, Loban HA. Qualitative composition of the oral microbiocenosis of young people with different caries intensity. *Svit medytyny ta biolohii* 2013; 1:57–59.
- Priya A, Kumar CBM, Valliammai A. Usnic acid deteriorates acidogenicity, acidurance and glucose metabolism of *Streptococcus mutans* through downregulation of two-component signal transduction systems. *Sci Rep* 2015; 4:366–74. doi: 10.1038/s41598-020-80338-6.
- Ranganathan V, Akhila CH. *Streptococcus mutans*: has it become prime perpetrator for oral manifestations? *J Microbiol Exp* 2019; 7(4):207–13. doi: 10.15406/jmen.2019.07.00261.
- Shakovets NV, Terexova TM. The incidence of dental caries in young children and its relationship with various risk factors. *Profilaktychna ta Dytiacha Stomatologia* 2015; 1(12):38–42.
- Skatova EA, Makeeva MK, Shakariants AA. Practical aspects of caries risk determination. *Problemy stomatologii* 2010; 3:13–18.
- Stirling WD. Textbooks for Learning Statistics: Public CAST e-books. 2013.