9. Niu C, Zhang P, Wei L, Dong J, Xu C, Yang Q. et al. The global burden of hypertension and its epidemiological impacts on adolescents and young adults: projections to 2050. Front Cardiovasc Med. 2025 Oct 24;12:1619445. doi: 10.3389/fcvm.2025.1619445.
10. Puteikis K, Ažukaitis K, Dadurkevičienė D, Mikučionytė D, Simanauskas K, Šileikienė V. et al. Cognitive Outcomes in Young Adults with Primary Arterial Hypertension: The Role of Cardiovascular Risk Factors and Hypertension-Mediated Organ Damage. Medicina (Kaunas). 2024;60(8):1353. doi: 10.3390/medicina60081353.
11. Vithalapara PK, Ramana NP, Chavada NM, Babriya DG. Prevalence and Risk Factors of Hypertension in Young Adults: A Cross-Sectional Study. Journal of Heart Valve Disease. 2025 Jun;30(6):69-72.
12. Yao Z, Tasdighi E, Dardari ZA, Jha KK, Osuji N, Rajan T. et al Association Between Cigarette Smoking and Subclinical Markers of Cardiovascular Harm. J Am Coll Cardiol. 2025;85(10):1018-1034. doi: 10.1016/j.jacc.2024.12.032.

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SCREENING FOR RISK FACTORS FOR THE DEVELOPMENT OF CARIES

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Dental caries remains a widespread chronic disease requiring effective risk assessment strategies. The purpose of the study was to identify risk factors for caries development and progression in young adults. The study involved 26 dental students aged 19–20 years, divided into three groups: caries-free, compensated caries, and subcompensated caries. Complex examination included assessment of salivary flow rate, viscosity, and pH, and plaque age using indicating tablets, as well as microscopic analysis of Gram-stained dental plaque. The results showed that salivary flow rate and pH (6.0–7.0) were within physiological norms for all groups. However, patients with caries exhibited increased salivary viscosity and a prevalence of mature dental plaque (>48 hours). Microbiological analysis revealed a dominance of Gram-positive cocci, morphologically similar to Streptococcus species, in the plaque of caries-active individuals. It is concluded that local factors, such as salivary viscosity and biofilm maturity, are critical determinants of risk even when general salivary parameters are normal.

Key words: dental caries, risk factors, saliva, salivary viscosity, dental plaque, oral microbiota.

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СКРИНІНГ ФАКТОРІВ РИЗИКУ РОЗВИТКУ КАРІЄСУ

Карієс зубів залишається поширеним хронічним захворюванням, що вимагає ефективних стратегій оцінки ризику. Метою дослідження було визначити фактори ризику виникнення та прогресування карієсу у молодих людей. У дослідженні взяли участь 26 студентів-стоматологів віком 19–20 років, розподілених на три групи: без карієсу, з компенсованою та субкомпенсованою формами карієсу. Комплексне обстеження включало оцінку швидкості слиновиділення, в'язкості, pH, давності нальоту з використанням індикаторних пігулок та мікроскопічний аналіз зубного нальоту, забарвленого за Грамом. Результати показали, що швидкість слиновиділення та pH (6,0–7,0) були в межах фізіологічної норми у всіх групах. Однак у пацієнтів з карієсом виявлено підвищену в'язкість слини та переважання зрілого зубного нальоту (>48 годин). Мікробіологічний аналіз виявив домінування грампозитивних коків, морфологічно подібних до видів Streptococcus, у нальоті осіб з активним карієсом. Зроблено висновок, що локальні фактори, такі як в'язкість слини та зрілість біоплівки, є критичними детермінантами ризику навіть за нормальних загальних параметрів слини.

Ключові слова: карієс зубів, фактори ризику, слина, в'язкість слини, зубний наліт, мікробіоценоз порожнини рота.

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Dental caries remains one of the most common chronic noncommunicable diseases in the world and continues to be a significant public health problem, despite significant progress in preventive and restorative dentistry [2, 10]. According to current epidemiological data, caries affects individuals of all age groups and is accompanied by significant functional, psychosocial and economic consequences [9]. The current understanding of caries defines it as a biofilm-mediated, diet-modulated, multifactorial, and dynamic disease characterized by alternating processes of demineralization and remineralization of dental hard tissues [8, 9, 10].

Saliva plays a key role in maintaining oral homeostasis and protecting against caries through its buffering properties, antimicrobial activity,

mechanical cleansing, and participation in enamel remineralization [2, 4]. Disturbances in salivary flow rate, viscosity, and pH can alter the ecological balance in the oral cavity, promoting the accumulation and maturation of dental biofilm and increasing susceptibility to cariogenic agents [2, 4]. Even minor changes in salivary properties can significantly affect the course of caries, especially in young individuals exposed to behavioral and nutritional risk factors [2, 3].

Dental biofilm is a highly organized microbial community, the composition and metabolic activity of which are closely dependent on environmental conditions, particularly the characteristics of saliva [6, 7]. The dominance of acidogenic and aciduric microorganisms, primarily Streptococcus mutans and related species, is closely associated with the initiation and progression of caries [6, 9]. The presence of mature dental biofilm is considered a key factor in caries risk, making it appropriate to assess the age of dental plaque and its microbial composition in clinical studies [7].

Current approaches to the management of caries patients emphasize individual risk assessment and early detection of biological factors, rather than just the diagnosis of cavitation lesions [10]. In this context, a comprehensive assessment of the functional state of saliva and oral microbiological characteristics is necessary to predict caries risk and implement personalized preventive measures [2, 9, 10]. However, data on the relationship between salivary flow, plaque maturity, and microbiological characteristics in the population remain limited.

Therefore, further study of salivary parameters and oral microbiological characteristics in relation to the risk of caries development is relevant and necessary to improve evidence-based preventive approaches and support personalized patient management strategies in modern dental practice.

The purpose of the study was to identify local risk factors for the development of carious lesions in patients, as well as the risk of progression of existing lesions.

Materials and methods. The initial patient selection, direct observation, and analysis of the data obtained were conducted at the Poltava State Medical University in Poltava.

The material for microbiological and clinical research comprised the results of a comprehensive dental examination of 26 students aged 19–20 who were in the 3rd year of the Faculty of Dentistry at Poltava State Medical University. And belong to the category of fully capable individuals capable of independently providing informed voluntary consent. In this study, vulnerable populations, as defined by the Good Clinical Practice (GCP) classification, were not included [1, 10, 11].

All those surveyed provided informed consent in writing to participate in the study. The formation of the study groups was in accordance with generally accepted principles of clinical dental research [9, 10]. No participants dropped out of the study, were lost to follow-up, or crossed over between groups during the study period, so the size of the analyzed sample remained identical to the original distribution.

Permission from the local bioethics committee for this study was not obtained, as it was conducted from October to December 2024 as part of student research work. References were made to National regulatory documents of Ukraine: Constitution of Ukraine (Articles 28, 32 – the right to respect for dignity and inviolability of private life); Fundamentals of Ukrainian legislation on healthcare; Law of Ukraine “On Protection of Personal Data”; Order of the Ministry of Health of Ukraine No. 690 dated 23.09.2009 “On approval of the Procedure for conducting clinical trials of medicinal products” (regarding compliance with GCP principles) [7].

The distribution of the surveyed students into groups based on the forms of caries was carried out based on a comprehensive clinical and epidemiological assessment of the intensity and activity of the carious process. The intensity of caries was determined using the DMFT (Decayed, Missing, Filled Teeth) index, in accordance with the World Health Organization's recommendations set out in Oral Health Surveys: Basic Methods.

The examination was performed according to the standard WHO clinical and epidemiological protocol using a dental mirror and probe under artificial lighting. Caries intensity was determined as the sum of the component values DMFT for each subject [11].

When forming groups, the total DMFT value, the structure of the index (ratio of the components of DMFT), the number of active carious lesions, and the presence of complications were taken into account. The compensated form was determined under the conditions of the predominance of filled teeth and the minimum number of active lesions; subcompensated – in the presence of a moderate amount of active caries and a balanced ratio of the index components; decompensated – with the dominance of the K component, high intensity of caries and the presence of clinical signs of process progression [8].

Patients were divided into 3 groups: the first group included 10 people without carious lesions of hard dental tissues; the second group included 9 students with compensated (caries intensity (CIV) up to 4 units); the third group included 7 students with subcompensated caries (caries intensity (CIV) more than 4 units).

Clinical examination included visual assessment of the condition of the oral mucosa and functional activity of the minor salivary glands of the lower lip mucosa. To obtain objective data, the lower lip was turned outward, the mucosa was thoroughly blotted with a sterile bandage, and, under sufficient lighting, the appearance of saliva droplets from the ducts of the minor salivary glands was observed.

Secretion was assessed by the time of saliva appearance: 1 – the appearance of droplets after more than 60 seconds was considered as reduced salivation, 2 – after less than 60 seconds, as normal secretory activity [2, 3].

The consistency of the oral fluid was also assessed using tweezers during functional rest. 1 point – The presence of sticky foamy residues or pronounced foaminess with bubbles indicated increased saliva viscosity, 2 points – while clear watery saliva corresponded to normal parameters. The assessment was carried out according to the criteria of physicochemical properties of saliva described in the literature [4].

The acid-base state of oral fluid (pH) was determined using indicator strips dipped into unstimulated saliva samples and compared with the resulting color to the manufacturer's standard color scale. The description of the oral fluid pH scale is provided below.

Standardized pH color scale:

– 1 point – pH 5.0 – deep orange: indicates severe acidosis of the oral cavity (critical level for enamel demineralization).

– 2 points – pH 5.5 – light orange/ochre: threshold value for enamel demineralization ("critical pH").

– 3 points – pH 6.0 – yellow-brown: moderate acidity; observed with carbohydrate consumption or reduced salivation rate.

– 4 points – pH 6.5 – yellowish-green: slight acidity; often borders on the lower limit of the physiological norm.

– 5 points – pH 7.0 – green: neutral environment; optimal state of oral fluid for remineralization processes.

– 6 points – pH 7.5 – dark green: slightly alkaline reaction; characteristic of high stimulated saliva secretion.

– 7 points – pH 8.0 – blue-green: alkaline reaction; may occur with an excess of mineral salts or specific diets.

The method is simple, reproducible, and widely used in dental clinical and laboratory research [4, 9].

For microbiological analysis, dental plaque was collected from the tooth surfaces with a sterile swab. The obtained material was applied to a glass slide, where it was fixed by air drying and stained by Gram's method using gentian violet (LLC Pharmaktiv, Ukraine) for primary staining, Lugol's solution (LLC Pharmaktiv, Ukraine) for fixation, ethyl alcohol for differentiation, and magenta (LLC Pharmaktiv, Ukraine) for contrast staining. Our studies allowed us to determine only the group affiliation of microorganisms – gram-positive or gram-negative. After staining, the smears were examined under a light microscope to detect and pre-identify microorganisms, according to standard diagnostic microbiology techniques [1, 6].

The age of dental plaque was determined using special indicator tablets. Absence of plaque was scored as 1 point, the appearance of pink or red color indicated fresh plaque as 2 points, while blue or purple color indicated the presence of old plaque as 3 points, formed for at least 48 hours, which is consistent with modern ideas about plaque biofilm and its clinical significance [6, 7].

Results of the study and their discussion. According to the results of the clinical and microbiological examinations, in most of the examined individuals, the rate of salivation was within the physiological norm. Salivary droplets from the minor salivary glands of the mucous membrane of the lower lip were observed within 60 seconds, in both students without carious lesions and individuals with compensated and subcompensated forms of caries. No signs of severe hyposalivation were detected in the studied groups. Saliva and plaque status indicators are given in Table 1.

Table 1

Characteristics of risk factors in the study groups

Indicator	Group 1 (n=10)	Group 2 (n=9)	Group 3 (n=7)
Salivation rate, s (M±m)	51.6±1.7	53.6±1.8	51.0±1.3
pH value (M±m)	6.5±0.17	6.4±0.18	6.6±0.20
Thick saliva, % (abs.)	30.0 % (3)	22.2 % (2)	57.1 % (4)
Old plaque, % (abs.)	60.0 % (6)	66.7 % (6)	71.4 % (5)
Gram-positive bacteria, % (abs.)	60.0 % (6)	77.8 % (7)	100.0 % (7)

Statistical analysis demonstrated no significant intergroup differences in salivation rate and acid-base status of oral fluid ($p > 0.05$). The average secretion rate ranged from 51 to 54 seconds, indicating preserved functional activity of the minor salivary glands in all subjects. The data suggest that in young individuals with initial or compensated forms of caries, salivation is not a leading risk factor.

At the same time, the analysis of the qualitative characteristics of saliva demonstrated certain differences between the groups. In group 3 (subcompensated form of caries), the frequency of detecting thick, viscous saliva was 57.1 %, almost twice that of the similar indicators in the group without caries and the group with a compensated course. Increased saliva viscosity can reduce its self-cleaning function, slow the elimination of microorganisms, and promote the longer retention of dental plaque on the enamel surface. Thus, even with a normal secretion rate, changes in the rheological

properties of saliva can play a significant role in the formation of a cariogenic situation.

The study of the acid-base state showed that the pH values in all groups were within 6.0–7.0, corresponding to the physiological range. The absence of significant differences between the groups indicates that the buffer properties of saliva were preserved in the examined individuals. However, it should be taken into account that the measurement was carried out under resting conditions, without functional load; the obtained values do not reflect dynamic changes in acidity, which can be observed after eating and have greater clinical significance for the initiation of demineralization processes in hard dental tissues.

Microscopic examination of Gram-stained dental plaque smears revealed a predominance of Gram-positive coccal microflora in all groups. In group 3, the detection frequency of Gram-positive bacteria was 100 %, which was statistically higher than the corresponding indicators in other groups. Morphologically, microorganisms were represented mainly by cocci arranged in chains, a characteristic of the genus Streptococcus. Taking into account the clinical picture and cariogenic potential, the presence of Streptococcus mutans can be assumed; however, the Gram-staining technique used does not allow for accurate species identification.

A moderate positive correlation was found between the presence of gram-positive bacteria and old dental plaque (r=0.56), as well as between bacterial contamination and increased saliva viscosity (r = 0.40). This indicates that mature dental biofilm is conducive to the accumulation and proliferation of cariogenic microorganisms. With increasing plaque age, the structural organization of the biofilm develops, a matrix of extracellular polysaccharides forms, and the microbial consortium's resistance to mechanical and chemical influences increases.

The results of the age-indicator tablets showed that some respondents, especially those with caries, had old dental plaque that appeared blue or purple. In group 3, this figure was 71.4 %, which may indicate insufficient personal hygiene or reduced effectiveness of the oral cavity's self-cleaning mechanisms. At the same time, even in the group without clinical manifestations of caries, the proportion of people with old plaque was quite high (60 %), indicating a potential risk of future pathological processes.

In general, the results confirm the multifactorial nature of the carious process. In the studied cohort of young individuals, the key role was played not by quantitative indicators of salivation or average pH level, but by qualitative characteristics of oral fluid and the state of dental biofilm. Increased saliva viscosity, the presence of mature plaque, and a high frequency of detection of gram-positive microflora collectively contribute to a local decrease in pH within the biofilm and the initiation of enamel demineralization.

It is important to note that the lack of significant differences in baseline salivary parameters between groups may be attributable to the subjects' relatively young age and the absence of systemic pathology. In older age groups or in the presence of somatic diseases, changes in salivary secretion may be more pronounced and have a greater impact on caries development.

The results obtained indicate that in the majority of the examined students, the functional activity of the minor salivary glands of the lower lip mucosa remained within the physiological norm, regardless of the presence of carious lesions. The absence of signs of pronounced hyposalivation in young people is consistent with data from modern studies, which indicate preservation of the secretory function of the salivary glands in the absence of systemic diseases and drug load [2, 3].

At the same time, the detected increase in the viscosity of oral fluid in some individuals with caries can be considered an unfavorable local factor that contributes to the retention of dental plaque and the disruption of the natural self-cleaning processes of the oral cavity. Similar changes in the physicochemical properties of saliva have been reported in the literature to indirectly affect the formation of cariogenic biofilm [4, 6].

The pH values of the oral fluid across all studied groups were within the physiological range, with no statistically significant differences between individuals with intact teeth and those with initial caries. This confirms the proposition that the acid-base balance of saliva at a young age is relatively stable, and that its local fluctuations are primarily due to microbial biofilm activity rather than to the overall pH of saliva [4, 9].

Microbiological examination of dental plaque showed a predominance of gram-positive cocci, consistent with current understanding of the key role of Streptococcus mutans in the pathogenesis of caries. The more frequent detection of old plaque in individuals with caries and its correlation with greater microbial contamination support the concept of mature biofilm as the main reservoir of cariogenic microorganisms [1, 7].

Thus, the results of the study indicate that, even with preserved indicators of salivation rate and oral fluid pH, increased saliva viscosity and the presence of outdated dental biofilm are associated with higher microbial contamination. This creates prerequisites for the formation of a cariogenic situation and progression of the carious process. The obtained data emphasize the need for a comprehensive assessment of risk factors, including analysis of both the physicochemical properties of saliva and the state of the dental biofilm, to enable timely prevention and individualized hygiene recommendations.

Limitations. During the research, we faced limitations in planning due to the limited variety of reagents, stemming from their low availability and high cost, as well as resource constraints in research activities.

Conclusions

1. In the majority of examined students, the salivation rate and pH values of oral fluid corresponded to the physiological norm, regardless of the presence of carious lesions of hard dental tissues.

2. In individuals with compensated and subcompensated forms of caries, an increase in saliva viscosity was more often observed, which may contribute to the formation and retention of dental plaque.

3. Microbiological analysis of dental plaque revealed the dominance of gram-positive coccal microflora, characteristic of cariogenic biofilm, with a higher prevalence in individuals with old dental plaque.

4. The presence of mature dental biofilm has been associated with increased microbial contamination and can be considered as one of the leading risk factors for the development and progression of caries.

5. The results obtained confirm the feasibility of a comprehensive assessment of the oral cavity condition, taking into account the properties of saliva and the characteristics of the dental biofilm for early detection of caries risk and individualization of preventive measures.

References

1. Council for International Organizations of Medical Sciences (CIOMS). International Ethical Guidelines for Health-related Research Involving Humans, Guideline 15: Research involving vulnerable persons and groups. Geneva: CIOMS; 2023.
2. Dawes C, Wong DTW. Role of saliva and salivary diagnostics in the advancement of oral health. J Dent Res. 2019;98(2):133-141. doi: 10.1177/0022034518816961.
3. Fejerskov O, Nyvad B, Kidd EAM. Dental Caries: The Disease and Its Clinical Management. 4th ed. Oxford: Wiley Blackwell; 2023.
4. Humphrey SP, Williamson RT. Saliva: diagnostic and protective functions. Dent Clin North Am. 2020;64(2):291-305. doi: 10.1016/j.cden.2019.11.002.
5. Lamont RJ, Koo H, Hajishengallis G. The oral microbiota: dynamic communities and host interactions. Nat Rev Microbiol. 2018;16(12):745-759. doi: 10.1038/s41579-018-0089-x.
6. Marsh PD, Zaura E. Dental biofilm: ecological interactions in health and disease. J Clin Periodontol. 2017;44(S18): S12-S22. doi: 10.1111/jcpe.12679.
7. Ministry of Health of Ukraine. Clinical guideline based on evidence "Dental caries" (Clinical Guideline KN 2024-869). Kyiv: Ministry of Health of Ukraine; 23 May 2024. Available at: https://www.dec.gov.ua/wp-content/uploads/2024/05/2024_kn_kariyes_zubiv.pdf.
8. Pedersen AML, Sorensen CE, Proctor GB, Carpenter GH. Salivary secretion in health and disease. J Oral Rehabil. 2021;48(8):955-969. doi: 10.1111/joor.13189.
9. Samaranayake LP. Essential Microbiology for Dentistry. 5th ed. Edinburgh: Elsevier; 2018.
10. Schwendicke F, Splieth C, Breschi L, et al. When to intervene in the caries process? An expert Delphi consensus statement. Clin Oral Investig. 2020;24(10):3315-3321. doi: 10.1007/s00784-020-03269-6.
11. World Medical Association. Declaration of Helsinki: ethical principles for medical research involving human subjects. JAMA. 2013;310(20):2191-2194. doi:10.1001/jama.2013.281053.

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