STUDY OF CYTOLOGICAL PROFILE OF MUCOUS MEMBRANE OF PROSTHETIC BED IN PATIENTS, USED BY DIFFERENT ADHESIVE AGENTS FOR FIXATION OF COMPLETE REMOVABLE PLATE PROSTHESES

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Abstract. In the dental market of adhesive agents for fixing removable prostheses there are different forms of release: pastes, creams, gels, films, powders of different composition and different manufacturers. In most cases, the choice of one or another means is determined by the patient’s own opinion. However, there are researches that indicate a violation of the microflora and the deterioration of the hygienic condition of the oral cavity, the occurrence and development of prostatic stomatitis in people who use adhesive agents to improve the fixation of removable prostheses. Therefore, in our opinion, it is important to develop recommendations for dentists on the choice of adhesives in accordance with the clinical, cytological and microbiological profiles of the tissues of the prosthetic bed of patients.

Aim. Study the characteristic features of the prosthetic bed mucosa (PBM) according to the results of its cytological identification in persons using different adhesive agents, and on the basis of the obtained results to recommend the criteria for their selection.

Materials and methods. In the clinic of the Department of Dentistry of Postgraduate Education of Ivano-Frankivsk National Medical University, 106 smear were obtained from 98 patients who used CRLD for 2-3 years. Among them, 72 people used adhesive pastes to improve fixation (group II): «Corega» adhesive paste was used by 30 people (Iia); «Lakalut» 20 (Iib) and «Protefix» 22 people (Iic). The third group consisted of 26 patients who used complete removable laminar dentures (CRLD) without the use of adhesive agents. In addition, 15 people with intact dentitions without dental and general somatic pathology were examined, in which 30 smear-impressions from the mucous membrane of the upper and lower jaws (group I) were obtained.

Cytological research was performed in the Educational and Scientific Laboratory of Morphological Analysis of IFNMU. Impression smear-impressions stained with «Leikodyf» reagent (manufactured by «Erba Lachema» (Czech Republic)) and hematoxylin and eosin were examined using a Micros Austria MC 300 light microscope at x200 and x400 magnification and photographed with a Toup Cam 5.1 M UHCCD C- Mount Sony with Toup Tek Photonics AMA075 adapter using Toup View 3.7 software.

Results. In the examination of the PBM in patients of these groups it was found that in group II (22) 30.6% had first-made CRLD, and remade - 69.4% (50). In the third group, 30.8% (8) people used CRLD made for the first time, and 69.2% (18) repeatedly. The use of adhesive agents to improve the fixation of CRLD reduces the intensity of the processes of regenerative-plastic insufficiency and polymorphic cell infiltration. Revealed by the predominance of ortho- and parakeratosis in persons who did not use adhesive agents to improve the fixation of the first manufactured CRLD; the phenomenon of increased desquamation of epithelial cells, the presence of cells of other layers of the epithelium in persons who used CRLD made repeatedly. In the smears of patients using «Protefix» and «Lakalut» pastes, a decrease in the intensity of keratinization of epitheliocytes was observed, compared with groups I, Iia, III. In the smears of persons who used the adhesive agents «Corega» noted moderate intensity of keratinization of epitheliocytes, anisocytosis, anisocarasia, pyknosis of the nucleus.

Conclusions. According to the results of cytological identification, adhesive agents «Protefix» and «Lakalut» can be recommended for primary orthopedic treatment and «Corega» for complex clinical conditions in repeated orthopedic treatment of CRLD.

Keywords: adhesive agents, cytological research, removable prostheses, mucous membrane of the cell process and parts.

Introduction. The dental market of adhesive agents for fixing removable prostheses is filled with numerous pastes, creams, gels, films, powders of different composition and different manufacturers. According to a survey of patients who use full CRLD, in 59.9% of cases the choice of adhesive is made personally by everyone directly in the pharmacy/store, 19.3% - guided by advertising, only 8.4% follow the recommendations provided in a dental clinic, and most importantly for all - reliable fixation of the prosthesis [1]. However, there are researches that indicate the acceleration of atrophy of the bone tissue of the jaws and the development of oral diseases such as prostatic stomatitis, candidiasis, imbalance of the oral microflora, poor hygiene in patients using adhesive agents to improve fixation to removable prostheses [2, 3, 4, 5, 6].
It is known about the negative effect of CRLD base on the, which is manifested by intensification of keratinization processes, thinning of the upper layer and increasing permeability of the PBM [7, 8].

**Rationale for the research.** Therefore, in our opinion, it is important to intensify the role of orthopedist in the selection of adhesive agents and provide clear recommendations for clinical, cytological and microbiological profiles of prosthetic tissues of patients [9].

Information on the dynamics of changes in PBM in patients who use adhesive agents to improve fixation needs further study, in particular, to be able to choose an adhesive agent according to the cytological profile.

**The aim of the research.** Study the characteristic features of the PBM according to the results of its cytological identification in persons using different adhesive agents, and on the basis of the obtained results to recommend the criteria for their selection.

**Materials and methods.** In the clinic of the Department of Dentistry of Postgraduate Education of Ivano-Frankivsk National Medical University, 106 smear – impressions were obtained from 98 patients who used CRLD for 2-3 years. Among them, 72 people used adhesive pastes to improve fixation (group II): in particular, «Corega» adhesive paste was used by 30 people (IIa); «Lakalut» 20 (IIb) and «Protefix» 22 people (IIc). The third group consisted of 26 patients who used CRLD without the use of adhesive agents. In addition, 15 people with intact dentitions without dental and general somatic pathology were examined, in which 30 smear-impressions from the mucous membrane of the upper and lower jaws (group I) were obtained.

Collection of material for cytological examination was performed in the clinic of the Department of Dentistry of Postgraduate Education IFN MU according to the classical method of obtaining a smear-impression. Cytological research was performed in the Educational and Scientific Laboratory of Morphological Analysis of IFN MU. Impression smear-impressions stained with «Leikodyfo» reagent (manufactured by «Erba Lachema» (Czech Republic)) and hematoxylin and eosin were examined using a Micros Austria MC 300 light microscope at x200 and x400 magnification and photographed with a Toup Cam 5.1 M UHCCD C- Mount Sony with Toup Tek Photonics AMA075 adapter using Toup View 3.7 software.

**Research results.** Obtaining smear-impressions was preceded by examination of PBM in patients of these groups. Among the surveyed persons of group II (22), 30.6% had first-time CRLD, and 69.4% had re-manufactured ones (50). In the third group, 30.8% (8) people used CRLD made for the first time, and 69.2% (18) repeatedly. In the second group, the state of the PBM corresponded to the first class according to Suple, in 46.2% (12) - the second and 23.1% (6) - III class.

In the research of smear-prints of group I, single epitheliocytes of the surface layer of the multilayered epithelium were observed. Ordinarily, the volume of the nuclei of these cells is much smaller than the volume of their cytoplasm. When stained with hematoxylin and eosin, well-defined basophilic nuclei with nucleoli and fine-grained cytoplasm are traced (Fig. 1).

In addition to epitheliocytes in the smear visualize lymphocytes that provide local immunity, and representatives of the resident microflora of the oral cavity (Fig. 2).

During light-optical research in the fields of view, representatives of the leucocyte series are identified (Fig. 3).

Some of the researched samples are characterized by the presence of cells in which the boundaries of the nuclei are vaguely visualized. In such epitheliocytes the color of the nuclei does not differ from the cytoplasmic (Fig. 4, Fig. 5).

In general, in the smear-prints of group I we note the moderate intensity of keratinization processes, filling the cytoplasm with keratohyalin inclusions. In some places, cells with compacted small nuclei are observed, which correlates with signs of karyopyknosis (see Fig. 5).

In patients who have used CRLD and adhesive agents to improve the fixation of «Corega», «Lakalut» and «Protefix» a characteristic feature of smear-prints of the mucous membrane of the prosthetic bed is polymorphism. Thus, in group Ia, epitheliocytes with signs of violation of differentiation are observed, which may lead to a decrease in the barrier function of the epithelium of the PBM. In some cases, cells of different sizes with different chromatophilia of the cytoplasm and nuclei are observed, among which cells with compacted nuclei are visualized. Single epitheliocytes contain more than one nucleus in the cytoplasm (Fig. 6).

In such cells the phenomena of anisocytosis, anisocariosis, nuclear pyknosis, multinuclearity are observed (Fig. 7).

Indicators of cytograms of the prosthetic bed in individual clusters of cells show the development of intracellular vacuolation (Fig. 8).

![Fig. 1. Structural features of the epithelium of smear-impression in patients of group I: 1 - nucleus, 2 - cytoplasm, 3 - non-nuclear epitheliocyte. Coloring: hematoxylin and eosin. Coll.: x400.](image-url)
Fig. 2. Structural features of the epithelium of smear-impression of the PBM in patients of group I: 1 - epitheliocyte, 2 - epitheliocyte nucleus, 3 - microbiota, 4 - lymphocyte. Coloring: «Leykodyf». Coll.: x200.

Fig. 3. Structural features of the epithelium of smear-impression in patients of group I: 1 - segmental neutrophilic granulocyte, 2 - epitheliocytes of the mucous membrane. Coloring: hematoxylin and eosin. Coll.: x400.

Fig. 4. Structural features of the epithelium of smear-prints in patients of group I: 1 - pyknotic nucleus, 2 - epitheliocytes of the mucous membrane, 3 - leukocytes. Coloring: hematoxylin and eosin. Coll.: x400.

Fig. 5. Structural features of the epithelium of smear prints in patients of group I: 1 - epitheliocyte, 2 - epitheliocyte nucleus, 3 - cytoplasm. Coloring: «Leykodyf». Coll.: x400.

Fig. 6. Structural features of the epithelium of smear-prints in patients of group IIa, who use removable prostheses and adhesive agents «Corega»: 1 - epitheliocyte, 2 - epitheliocyte nucleus, 3 - cytoplasm. Coloring: «Leykodyf». Coll.: x200.

Fig. 7. Structural features of the epithelium of smear-prints of the PBM in patients of group IIa, who use removable prostheses and adhesive agents «Corega»: 1 - pyknotic nuclei, 2 - cytoplasm of the PBM epitheliocytes. Coloring: hematoxylin and eosin. Coll.: x400.
Mucosal immunity of prosthetic bed tissues is provided by the representation of leukocytes (Fig. 11).

Irregularly shaped surface layer cells with a large number of weakly basophilic cytoplasm, in which pyknotic nuclei and inhomogeneous dispersed fine-grained inclusions are observed (Fig. 9).

Individual smear-imprints show non-nuclear polygonal cells with sharply basophilic cytoplasm, which allows them to be classified as those that have undergone intensive keratinization processes (Fig. 10).

The same tendency is noted at coloring by hematoxylin and eosin. Many epitheliocytes of irregular shape with the phenomena of anisokariosis are noticeable in the fields of view (Fig. 13).

Immature epithelial cells are small, round shape with a narrow rim of basophilic cytoplasm and, accordingly, a high nuclear-cytoplasmic ratio are quite rare (Fig. 14).

The phenomena of cytological pathology of the epithelium of the mucous membrane of the prosthetic bed are observed. Against the background of homogeneous weakly basophilic cytoplasm, hyperchromic small compacted nuclei are visible (Fig. 15).
The cytograms of the impressions were taken from the PBM of patients of subgroup IIb who used «Protefix» show similar morphological changes. In particular, epitheliocytes are characterized by polymorphism of cell populations (Fig. 16).

The shape of the cells is changed due to small protrusions and intussusception of the plasmalemma. Their cytoplasm is stained from weakly basophilic to pronounced basophilia. At the same time, fine-grained grain dominates. The nuclei are moderately compacted (Fig. 17).

In some fields of vision epitheliocytes are visualized, in the nuclei of which there are figures of mitosis (Fig. 18).
In this group of observations dystrophic changes prevail, the manifestation of which is the above-mentioned granularity of the cytoplasm (Fig. 19).

When coloring smear-imprints with hematoxylin and eosin, a pool of cells with pyknosis of nuclei is visualized when the contours of the plasmolemma are blurred (Fig. 20).

Orthokeratosis was observed in the PBM smear-imprints of patients who used CRLD without adhesive agents and was group III, and parakeratosis in some samples. Signs of regenerative-plastic insufficiency and polymorphonuclear infiltration are more pronounced. When collecting the material, the epithelial layers were easily exfoliated, which led to multicellular samples. Such cytological complexes in light-optical examination are represented, respectively, by numerous polymorphic cells. These groups of epitheliocytes mostly number more than 12-18 cells (Fig. 21).

In pleomorphic populations of epitheliocytes, nonnuclear keratinous scales are observed, as well as cells from other layers of the epithelium with deformed nuclei. Their cytoplasm contains dispersed granules, which are unevenly distributed (Figs. 22, 23). In fig. 22 microbial inclusions and complexes are well visualized in the fields of view.
The presence of leukocyte cells indicates the phenomenon of nonspecific antimicrobial protection (Fig. 23, Fig. 24).

Discussion. For the maximum identification of the epithelium of the PBM in the smear-prints, different dye systems were used, which allowed to analyze the features of their structural components when using adhesive agents and without their use. Hematoxylin and eosin - for visualization of nuclei and cytoplasm of cells, «Leukodyf» - for additional identification of leukocytes.

The greatest changes in the structural features of epitheliocytes were observed in group II, in particular, in IIa - compacted pyknotic nuclei, more than one nucleus in a cell, non-nuclear polygonal cells with sharply basophilic cytoplasm; subgroups IIb and IIc are characterized by small compacted nuclei and pycnosis of nuclei. In the third group, on the other hand, the presence of non-nuclear keratinized epitheliocytes and numerous polymorphic groups of epitheliocytes were noted - more than 12-18 cells in stratified epithelial layers.

It should be noted leukocyte activity in subgroup IIa, compared with IIb and IIc, however, visually in the field of view of leukocytes was not more than in the smears of patients of groups I and III.

The cytological picture of the PBM in the examined patients of group II is consistent with our experimental researches of integrated indicators of antimicrobial activity of fixing creams, which showed the highest effectiveness of «Lakalut Dent Minto» (72.0), much lower «Protefix Hypoallergenic» (32,17) and «Corega Fresh taste» (31.78) points [12]. However, we believe that the adhesion of microorganisms on the epitheliocytes of the PBM in patients using various means to improve fixation, still needs additional clinical study.

Comparing the obtained cytograms with the results of clinical dental examination, we can trace more similar changes in the processes of keratinization in primary orthopedic treatment and morphologically diverse - from areas of intense keratinization to excessive exfoliation of epitheliocytes as a manifestation of the inability of epithelial cells to undergo complete differentiation, with repeated orthopedic treatment. This is especially true in patients who have not used means to improve the fixation of removable prostheses.

The use of adhesive agents to improve the fixation of CRLD helps to ensure an even distribution of the occlusal load on the cell process and the part that, in turn, maintains capillary blood flow for optimal blood supply to bone tissue. At the same time, it helps to reduce the coefficient of friction in the interaction between the base of the prosthesis and the supporting mucous membrane, and, accordingly, prevents injury to the PBM [10, 11, 13].

Conclusions:
1. The use of adhesive pastes to improve the fixation of removable prostheses reduces the intensity process of regenerative-plastic insufficiency and polymorphically cellular infiltration.
2. In patients who did not use adhesive agents to improve the fixation of removable prostheses in the cytological profile, two types can be distinguished: with a predominance process of ortho- and parakeratosis in persons who used CRLD made for the first time, and with increased epithelial cell desquamation, the presence of cells in other layers in persons who used CRLD, made repeatedly. Both types are characterized by the presence of leukocytes, which may indicate an additional antimicrobial mechanism in areas of the mucous membrane, not protected by the stratum corneum.
3. According to the results of cytological identification of the mucous membrane of prosthetic bed tissues in smear-prints of patients using «Protefix» and
В ПАЦІЄНТІВ, ЩО КОРИСТУЮТЬСЯ ПЗПП, виготовленими повторно, у 15 осіб без знімних протезів, у 26 пацієнтів, що не користувалися, а також у 15 осіб без знімних протезів. УДК 616.311+616-08+615.242+616.314-77

ВИВЧЕННЯ ЦИТОЛОГІЧНОГО ПРОФІЛЮ СЛИЗОВОЇ ОБОЛОНКИ ПРОТЕЗНОГО ЛОЖА В ПАЦІЄНТІВ, ЩО КОРISTUЮТЬСЯ РЗІНИМИ ADГEЗIVNIMI ZASOBAMI ДЛЯ ФІКСAЦІЇ Pовних ЗНІMних ПЛASTИНОКовИХ ПРОТЕЗів

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Резюме. Використання засобів для покращення фіксації повних знімних пластикових протезів (ПЗПП) забезпечує надійну естетичну, жувальну, мовленню функцій зубоцелепної системи. Проте залишаються несформованими рекомендації щодо вибору адгезивних засобів залежно від стану тканини протезного ложа.

Мета: вивчити характерні ознаки слизової оболонки протезного ложа (СОПЛ) за результатами її цитологічної ідентифікації в осіб, що користуються різними адгезивними засобами, та на основі отриманих результатів рекомендувати критерії вибору.

Матеріали та методи: проаналізовано цитологічну картину СОПЛ 72 осіб, що користувалися адгезивними засобами для покращення фіксації ПЗПП, та 26 паціїнтів, що не користувалися, а також 15 осіб без знімних протезів. Результати. Використання адгезивних засобів для покращення фіксації ПЗПП зменшує інтенсивність процесів регенераторно-пластичної недостатності і поліморфоклітинної інфільтрації. Виявлено переважання процесів орто- та паракератозу в осіб, що не використовували адгезивні засоби для покращення фіксації вперше виготовлених ПЗПП, явища підвищеної десквамації епітеліоцитів, присутність клітин інших шарів епітелію в осіб, що користувалися ПЗПП, виготовленими повторно. У мазках пацієнтів, що користувалися пастами «Протефікс» та «Лакалут», спостерігали зниження інтенсивності кератинізації епітеліоцитів, порівняно з 1, 2, 3 групами. У мазках осіб, що користувалися адгезивним засобом «Корега», відзначали помірно виражену інтенсивність кератинізації епітеліоцитів, анізоцитоз, анізокаріоз, пікноз ядер.

References:
1. Yohei Okazaki, Yasuhiko Abe, Kana Dainobu, Shogo Iwaguro, Ryoji Kato, Kazuhiro Tsuga. A web-based survey of denture adhesive use among denture wearers 40 years of age and older. Journal of Oral. Science. 2021; 63(1):98-100.
2. Papadiochou S, Emmanouil I. Papadiochos I. Denture adhesives: a systematic review. J Prosth Dent. 2015; 113(5):391-7.
3. Bartlett D, Carter N, Felton D, Goffin G, Kawai Y, Muller F, Polyzois G, Waals A. White paper on guidelines for the use of denture adhesives and their benefits for oral and general health: Oral Health Foundation. 2019; 6:11-5.
4. Quiney D, Nishio Ayre W, Milward P. The effectiveness of adhesives on the retention of mandibular free end saddle partial dentures: an in vitro study. J Dent. 2017; 62:64-71.
5. Leite AR, Mendoza-Marin DO, Paleari AG, Rodriguez LS, Roccia AA, Policastro VB, Compagnoni MA, de Souza RF, Pero AC. Crossover clinical trial of the influence of the use of adhesives and their benefits for oral and general health: Oral Health Foundation. 2019; 6:11-5.
6. Mykhaylenko TM. Oral cavity hygiene in persons using adhesive products to improve the fixation of removable dentures constructions. J Pharma Innov. 2015; 3(12):64-67.
7. Kalashnikov DV, Kindiy DD, Korol DM, Kindiy VD. Analysis of denture base impact on the denture foundation area tissues. Svit medytsyny ta biolohii. 2020; 1(71):58-62.
8. Veiga N, Herdade A, Diniz L, Brites B, Pinto S, et al. Oral lesions associated with removable prosthesis constructions. J Pharma Innov. 2015; 3(12):64-67.
9. Tun Min Bo, Yohei Hama, Norihisa Akiba and Shunsuke Minakuchi Utilization of denture adhesives and the factors associated with its use: a crosssectional survey. BMC Oral Health. 2020; 20:194-202.
10. Junning Chen, Rohana Ahmad, Wei Li, Michael Swain, Qing Li. Biomechanics of oral mucosa. J Royal Society Interface. 2015 Aug 6; 12(109):1-20.
11. Priya Nirmish Deo, Revati Deshmuk Pathophysiology of keratinization. J Oral Maxillofac Pathol. 2018 Jan-Apr; 22(1):86-91.
12. Redushko YuV, Dmytryshyn TM, Kutsyk RV. Portiwniaja protimotkrobnukh vlastivostei riznych adhezyvnykh zasobiv dla fiksatsii povnykh znimnych plastynkovykh proteziev. Visnyk problem biolohi i medytsyny. 2018; 1;374-380.
13. Mennatallah MM, Essam AA, Nagla MH, Nermine RA. Comparative study of silicone-based soft liner versus versacryl liner for mandibular complete denture cases (randomized clinical trial). Egyptian dental journal. 2021; 67(1):651-60.
Висновки. За результатами цитологічної ідентифікації, адгезивні засоби «Протефікс» та «Лакалут» можемо рекомендувати за умов первинного ортопедичного лікування, а «Корега» за складних клінічних умов при повторному ортопедичному лікуванні ПЗПП.

Ключові слова: адгезивні засоби, цитологічне дослідження, знятні протези, слизова оболонка коміркового відростка та частини.

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