The study of the base resin impact on planktonic growth of microorganisms of oral origin

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ABSTRACT

Aim: To study the main differences in the intensity of growth of microbial strains of oral origin when they are cultivated in the presence of base resins that are widely used in the clinic of prosthetic dentistry.

Materials and Methods: 3 types of base resin samples were used to evaluate the impact on the growth of microorganisms. Strains of opportunistic pathogens representing the facultative anaerobic transient microflora of the oral cavity were used for the research. Microbial cultures were isolated from the oral mucosa of the patients with removable dentures. The optical density of the culture was determined using a multi-mode photometer. Control growth of the cultures was evaluated under similar cultivation conditions in test tubes with a nutrient medium without resin samples.

Results: All resins reduced the intensity of polyantibiotic-resistant *S. aureus* MRSA growth compared to control samples. The impact on *S. epedermidis* growth was different. Thus, Vertex and Breflex reduced the growth intensity insignificantly, and Villacryl increased it to the level of 1.104 ± 0.026 CU in comparison with glass constituting 1.178 ± 0.033 CU. All studied resins inhibited the growth of polyantibiotic-resistant S. aureus MRSA. Villacryl resin had the least impact on the growth of S. epedermidis. Villacryl resin inhibited *C. albicans* growth only by 13.70%, and the other two resins inhibited it by 25% on average.

Conclusions: The conducted microbiological studies showed the diverse influence of base materials on such important indicators as the optical density of cultures grown in the presence of base resins and the growth inhibition index of cultures.

KEY WORDS: removable dentures, base resin, microbial cultures

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INTRODUCTION

The prevalence and intensity of dental diseases remains at a high level despite the implementation of a significant number of treatment and prevention programs aimed at reducing their level. This leads to edentulousness and the occurrence of a significant percentage of pathological and structural-functional changes in the maxillofacial system. According to the WHO, partial edentulousness is observed in 75% of the world's population [1]. Ukrainian adult population's need for the prosthodontic treatment in case of partial edentulousness constitutes 57.5 people per 1000 examined [2].

The prosthodontic treatment of patients with partial edentulousness should prevent and remove the consequences of the edentulousness, namely an atrophy of the maxillofacial skeleton and muscles, and is aimed at a person's general improvement and prolongation of the active period of their life [3, 4].

The prosthodontic measures during the treatment of patients with edentulousness consist in the justifi-

cation of the choice of the optimal dental prosthesis, the material of the dental prosthesis and the patient's treatment approach [5].

Most often, an orthopedic surgeon chooses any standard dental prostheses and materials that are constantly used in modern practice [6]. Until this point, we have mostly mentioned removable partial dentures made of acrylic, that is, made of plastics, but nowadays dentures made of thermoplastic are very popular [7]. Thermoplastics are a type of plastic based on thermoplastic polymers. Thermoplastics soften under the influence of high temperatures and become viscous fluid, and return to a solid form upon cooling [8]. Despite the positive results of these materials, most patients try to avoid the use of removable dentures [9].

AIM

To study the main differences in the intensity of growth of microbial strains of oral origin when they are cultivat-



Fig.1. Optical density of cultures (CU) grown in the presence of resins.

| Table 1. C | Optical density | of cultures | (CU) grown in | the presence of resins |
|------------|-----------------|-------------|---------------|------------------------|
|------------|-----------------|-------------|---------------|------------------------|

| | S. aureus MSSA | S. aureus MRSA | S.epidermidis | S. pyogenes (β Str A) | S. dysgalactiae ssp. equisimilis (β Str G) | S. gordonii | S. oralis | S. sanguinis | C. albicans | C. tropicalis |
|-----------|----------------|----------------|---------------|--------------------------|---|--------------|--------------|--------------|--------------|---------------|
| Vertex | 0.751±0.066 | 1.010±0.087* | 0.953±0.035* | 0.744±0.070* | 1.224±0.067* | 0.886±0.050* | 0.947±0.021* | 0.579±0.055* | 1.130±0.015* | 1.124±0.012* |
| Breflex | 1.130±0.027* | 1.107±0.026* | 0.975±0.089* | 0.686±0.038* | 1.116±0.036* | 0.757±0.039* | 0.592±0.032* | 0.577±0.038* | 1.154±0.055* | 1.120±0.011* |
| Villacryl | 0.794±0.053 | 1.176±0.018* | 1.104±0.026 | 0.778±0.020* | 1.310±0.102* | 0.769±0.030* | 0.588±0.093* | 0.557±0.180* | 1.316±0.033* | 1.002±0.023* |
| Glass | 0.821±0.086 | 1.461±0.035 | 1.178±0.033 | 1.356±0.049 | 1.580±0.076 | 1.408±0.041 | 0.731±0.010 | 1.445±0.014 | 1.525±0.046 | 1.454±0.042 |
| Control | 0.770±0.012 | 1.479±0.017 | 1.179±0.028 | 1.390±0.014 | 1.553±0.046 | 1.412±0.021 | 0.740±0.011 | 1.461±0.011 | 1.530±0.030 | 1.448±0.030 |

* significant differences at p < 0.05.

ed in the presence of base resins that are widely used in the clinic of prosthetic dentistry.

MATERIALS AND METHODS

This research was conducted according to the WMA Declaration of Helsinki – "Ethical Principles for Medical Research Involving Human Subjects" and approved by the Ethics Committee of the Ivano-Frankivsk National Medical University (protocol No. 131/22 dated November 24, 2022).

3 types of base resin samples such as Vertex poly methyl methacrylate, Breflex polyamide and Villacryl polymethacrylate were used to evaluate the impact on the growth of microorganisms.

In total, the results of microbiological studies of prostheses of 55 patients were studied. 3 groups of patients were formed. The group with prostheses, where the base resin was Vertex poly methyl methacrylate, consisted of 18 patients; Breflex polyamide – 16 patients; Villacryl polymethacrylate – 21 patients.

Ready resin samples for the experiment had the form of plates with a thickness of 2 mm and an area of 1 cm². Glass plates of similar size were used as controls. The study and control samples were placed in a sealed cellophane package and sterilized by X-ray irradiation at a dose of 0.44 mGy for 1.540 s.

Strains of opportunistic pathogens representing the facultative anaerobic transient microflora of the oral cavity were used for the research: *Streptococcus pyogenes* (Group A β -hemolytic streptococcus), *Streptococcus dysgalactiae ssp. equisimilis* (Group G β -hemolytic streptococcus), methicillin-susceptible *Staphylococcus*



Fig.2. Growth inhibition index (%) in the presence of resins (in relation to glass).

| Table 2. | Growth inhibit | on index (%) |) in the presence | of resins (in relation | ı to glass) |
|----------|----------------|--------------|-------------------|------------------------|-------------|
|----------|----------------|--------------|-------------------|------------------------|-------------|

| | S. aureus MSSA | S. aureus MRSA | S.epidermidis | S. pyogenes (βStr A) | S. dysgalactiae ssp. equisimilis (β Str G) | S. gordonii | S. oralis | S. sanguinis | C. albicans | C. tropicalis |
|-----------|----------------|----------------|---------------|----------------------|---|-------------|-----------|--------------|-------------|---------------|
| Vertex | 8.53 | 30.87* | 19.10* | 45.13* | 22.53* | 37.07* | -29.55* | 59.93* | 25.90* | 22.70* |
| Breflex | -37.64* | 24.23* | 17.23* | 49.41* | 29.37* | 46.24* | 19.02* | 60.07* | 24.33* | 22.97* |
| Villacryl | 3.29 | 19.51 | 6.28 | 42.63* | 17.09* | 45.38* | 19.56* | 61.45* | 13.70* | 31.09* |

* significant differences at p < 0.05.

aureus (MSSA), methicillin-resistant S. aureus (MRSA) with associated resistance to fluoroquinolones, macrolides, tetracyclines and aminoglycosides, methicillin-susceptible Staphylococcus epidermidis; yeast-like fungi Candida albicans and Candida tropicalis as well as the mitis group a-hemolytic streptococci (Streptococcus oralis, Streptococcus sanguinis, Streptococcus gordonii) as the main representatives of the resident microflora of this biotope. Microbial cultures were isolated from the oral mucosa (prosthetic bed, gingival pockets) of the patients with removable dentures with manifestations of prosthetic stomatitis and identified on the basis of morphological, cultural properties and biochemical microtests "STAPHYtest 16", "STREPTOtest 16" (Lachema, Czech Republic) and VITEK 2 GP and VITEK 2 YST test systems (Biomerieux, France) using the VITEK 2 Compact analyzer.

The study sample was placed in a test tube with 2.0 ml of nutrient Brain Heart Infusion Broth (HiMedia

Laboratories Pvt. Ltd., India) with the addition of 1% glucose, previously freshly inoculated with test strains of microorganisms at a final concentration of 1×10⁴CFU/ml. Cultures were cultivated for 24 hours at a temperature of 37°C with constant stirring by means of an MR-1 shaker (SIA BIOSAN, Latvia) with a stirring frequency of 20 times/ min. 5 portions of planktonic culture with a volume of 200 µl were taken into the cells of a polystyrene tablet after the cultivation. The optical density (OD) of the culture was determined using a multi-mode photometer for Synergy™ NTX S1LFTA microplates (BioTek Instruments, Inc., USA) at a wavelength of 495 nm using Gen5™ Data Analysis Software. Control growth of the cultures was evaluated under similar cultivation conditions in test tubes with a nutrient medium without resin samples.

Based on the results of the experiment, the growth inhibition indices of test cultures of microorganisms were calculated for each sample (1):

$$GI(\%) = 100 - \frac{OD_{experiment} \times 100}{OD_{control}}$$
(1)

Statistical processing of the obtained data was presented in the form of mean values of measurements \pm standard deviation for three independent experiments. Results were processed using a two-sample t-test, differences were considered statistically significant at p<0.05.

RESULTS

We analyzed the differences in the growth intensity of microbial strains of the oral origin in the course of their cultivation in the presence of the resins widely used in a clinic of prosthetic dentistry (Table 1 and Fig. 1).

All resins reduced the intensity of polyantibiotic-resistant S. aureus MRSA growth compared to control samples. The impact on S. epedermidis growth was different. Thus, Vertex and Breflex reduced the growth intensity insignificantly, and Villacryl increased it to the level of 1.104 ±0.026 CU in comparison with glass constituting 1.178 ± 0.033 CU (p < 0.05). All samples reduced the growth intensity of Group A β-hemolytic streptococci S. Pyogenes and Group G S. dysgalactiae ssp. Equisimilis, moreover a sample of Breflex resin did it to the greatest extent (p < 0.05). When studying the representatives of the oral cavity microflora, namely α-hemolytic streptococci, the most significant growth inhibition influenced by the samples of all three basic resins was observed in the cultures of S. gordonii and S. sanguinis. The growth rate of S. gordonii culture constituted 53.7% in the presence of Breflex and Villacryl resins and 62.9% in the presence of Vertex resin in relation to the growth intensity in test tubes with glass as the control material (p < 0.05). The growth rate of S. sanguinis culture constituted 40% in the presence of Vertex and Breflex base resins and 38.6% in relation to glass (p < 0.05).

Somewhat different picture was observed when *S.* oralis cultures were studied. Thus, Vertex stimulated its growth up to 0.947 \pm 0.021 CU relative to glass (0.731 \pm 0.010 CU) (p < 0.05), and the other two resins reduced it. Vertex and Breflex resins equally reduced the growth intensity of yeast-like fungi *C. albicans*, *C. tropicalis*. Villacryl base resin caused a more significant growth inhibition of *C. tropicalis* than *C. albicans*.

The growth inhibition index is a more informative criterion characterizing the impact of base resins on microbial cultures growth. It was calculated in relation to the control material, namely glass (Table 2 and Fig. 2). All studied resins inhibited the growth of polyantibiotic-resistant *S. aureus MRSA*: Vertex – by 30.87%, Breflex by 24.23% and Villacryl by 19.51% compared to glass (p < 0.05). Villacryl resin had the least impact

on the growth of *S. epedermidis* (inhibition by 6.28% compared to glass) (p < 0.05). The growth rates of *S. pyo-genes* (β Str A) in the studied resins were approximately at the same level and ranged from 49.41% to 42.63% (p < 0.05). Villacryl base resin inhibited *S. dysgalactiae ssp. equisimilis* (β Str G) growth by 17.09% (p < 0.05). *S. gordonii* growth was inhibited by Vertex base resin most of all. S. oralis culture growth was stimulated by Vertex by 29.59% (p < 0.05), however it was inhibited by other resins, on the contrary (by 19% compared to glass). Resins did not show any significant difference on the growth inhibition index of *S. sanguines* (p < 0.05).

Important results were obtained regarding the impact of resins on *yeast-like fungi cultures*. Thus, Villacryl resin inhibited *C. albicans* growth only by 13.70% (p < 0.05), and the other two resins inhibited it by 25% on average (p < 0.05). Villacryl resin inhibited the growth intensity of *C. tropicalis* most of all, namely by 31.09% (p < 0.05).

DISCUSSION

Dental implantation has become widespread at the current stage of the dentistry development. This treatment is aimed at restoring a patient's chewing efficiency and aesthetic comfort [10]. Unfortunately, dental implantation is not always possible immediately after tooth extraction and requires some time and appropriate preparation of the oral cavity [11].

In order to prevent bone tissue atrophy and preserve the supporting teeth, we offer temporary removable dentures for patients with edentulousness who are scheduled for dental implantation. This problem is quite relevant and has become the objective of our research, namely, the determination of the material that should be used for temporary removable dentures manufacturing in order to prevent the development of dysbacteriological complications and the development of the maxillofacial skeleton diseases, which will eventually become contraindications to implantation surgeries.

As is well known, the wrong choice of structural material is the cause of microbial imbalance, which leads to the accumulation of pathogenic microflora, which leads to a shortening of the period of use of prostheses, and in many cases to the refusal of their use [12, 13].

Scientists have proven that as the period of use of partial removable lamellar prostheses increases, the negative impact of the bases of the prostheses on the microflora of the oral cavity increases, regardless of the construction material. This is an excellent environment for increased reproduction of microorganisms, primarily for *C. albicans* and *E. Coli* [14].

Colonization by fungi of the genus *C. albicans* was observed in 22.3%-30% of the examined patients in

patients who use removable prostheses and steadily increased to 41.5% with increasing duration of use [15].

A number of scientists claim that along with the massive insemination of the mucous membrane of the oral cavity in elderly patients who use removable orthopedic structures, the number of microflora increases, among which dominate staphylococci in 90.5%, lactobacilli in 76.2% and yeast-like fungi in 57.6% of cases [16].

The obtained data on the optical density of microbial cultures characterized the intensity of their planktonic growth.

Comparing the base resins impact on the growth of *S. aureus MSSA*, the most intense growth was observed in the presence of Breflex resin (it constituted 1.130 ± 0.027 CU). Growth indicators of Vertex and Villacryl resins were at the same level, even slightly lower than the corresponding indicators compared to the control samples made of glass constituting 0.82 ± 0.086 CU and the control amounting 0.77 ± 0.012 CU.

The conducted microbiological studies showed different impact of base resin samples on the growth inhibition of antibiotic-sensitive *S. aureus MSSA*. Vertex inhibited growth by 8.53% in relation to glass, and Breflex, on the contrary, sharply stimulated the culture

growth by 37.64%. The impact of Villacryl resin was the closest to the control indices in comparison with glass.

The period of adaptation is a very difficult period for patients and dentists, so the obtained research results contribute to the improvement of the selection of structural elements of partial removable lamellar prostheses in the treatment of partial edentulousness.

CONCLUSIONS

The conducted microbiological studies showed the diverse influence of base materials on such important indicators as the optical density of cultures grown in the presence of base resins and the growth inhibition index of cultures.

According to the obtained results, such basic resins as Breflex, Vertex increased the optical density of microbial cultures and could not be recommended for the manufacture of temporary removable laminar dentures.

The index of cultures growth inhibition and the impact on the optical density provided an opportunity to recommend Villacryl base resin as a base material for the manufacture of temporary removable laminar dentures.

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CONFLICT OF INTEREST

The Authors declare no conflict of interest

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