STUDY REGARDING THE ADHERENCE OF ORAL MICROBIOTA ON COMPOSITE PLATING MATERIALS

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Abstract: The paper set to carry out a study about the behavior of the 3 types of composite materials (*Solidex*, *Ceramage*, *Gradia*) in the oral environment, by investigating the relationship between bacterial micro-flora and marginal periodontal tissue (assessing the degree of microorganisms adherence on the surface of these plating materials).
Results thus obtained showed a significant difference between the two groups: study group (teeth reconstructed with composite materials) and control group (natural teeth). Although prosthetic parts were finished and polished to allow a smooth self-cleaning surface and subjects have used additional methods of oral hygiene (dental thread and mouthwash), the prosthetic surface presented a far greater number of microorganisms compared with the surface of natural counterparts teeth thus, in general, values were ten times higher in favor of the prosthetic surface compared with the natural control teeth

INTRODUCTION

Making fixed dental restorations includes, besides the restorative therapy itself, the development of a correct diagnosis and a therapeutic plan, a potential periodontal therapy to maintain proper relationships with tissues other than natural teeth (prosthetic pieces of various materials), the prevention of subsequent failure and especially a specific health education in the field (1, 2, 8).

Fixed dental prosthesis is achieved through a succession of clinical and technological phases that are developed by collaboration between professional dentists and dental technicians. Conducting their sequence of stages is specific for each type of fixed dentures, according to their particularities and the materials they are made of (8).

The progress of sampling techniques, cultivations, molecular biology and especially genetics, have enabled scientists to observe that teeth and oral mucosa is colonized by a flora composed of about 350-400 different bacterial species.

The indigenous micro biota differs, quantitatively and qualitatively, from one region of the organism to another, according to hosting and self regulating conditions that are established between the micro-resident microorganisms (those who have already colonized, persistent) and the micro-floating ones (new contaminants, variable and transitory for hours, days or weeks).

Morph-histological and functional, the oral cavity offers the microorganisms various hosting conditions different from other body cavities: lips, teeth, gingival liquid, lingual and oral mucosa, the palate, and saliva.

Hosting conditions for these biotopes vary a lot under the report of Eh, pH, available nutrients, presence or absence of receptors for certain microbial liants, forces of microorganisms removal (e.g., secretion of salivary or crevicular fluid, mastication forces, the action of abrasive food, oral hygiene).

The oral cavity offers hosting conditions for the following types of organisms: for the palatal mucosa are constant: *Streptococcus oralis*, *Streptococcus sanguis*, *Haemophilus spp*, *Lactobacillus spp*, *levurs* and variable: *S. salivarius*, *Corynebacterium spp*, *Neisseria spp*, *Moraxella spp*; anaerobe microorganisms are present in small numbers, beta-hemolytic streptococci (placed on the palate soft vale, palatal-lingual and palatal-fairings mucosa).

At the jugal mucosa one can constantly find, *Streptococcus oralis* and *S. sanguis*, and variably *S. salivarius*, levurs, *Haemophilus spp*, *Neisseria spp*; on the tongue one can find *Streptococcus salivarius*, *S.oralis*, *Haemophilus spp*, *Stomatococcus mucilaginosus*, *Candida albicans*, and on the lips, that make the transition between skin and the oral cavity, mainly S.*epidermidis* and oral *streptococci*(7).

These microorganisms have an obviously related to age dynamic (newborn and infant dental previously eruption, temporary and final dentition, edentulations, prosthetic etc.).

Consequently, the oral micro biota varies from one site to another and by different moments, being dominated by bacteria and levurs; only in the absence of oral hygiene and in conditions of promiscuity, significantly, protozoa appear, such as *Entamoeba gingivalis* and *Trichomonas tenax*.

A relatively small number of these micro-organisms which in terms of theory are able directly or indirectly to destroy periodontal tissues - expressed in terms of virulence - pathogenesis power.

The pathogenic power of the bacteria is represented by antigens action, the enzymes production and bacterial degradation products, and adding to them, the production of endo-and exotoxine.

All these bacterial products behave as irritants that induce changes to the periodontal tissues.

In the context of the above, the paper set to carry out a study about the behavior of the 3 types of composite materials (*Solidex*, *Ceramage*, *Gradia*) in the oral environment, by investigating the relationship between bacterial microflora and marginal periodontal tissue (assessing the degree of microorganisms adherence on the surface of these plating materials).

MATERIAL AND METHODS

We conducted a study which included a lot of 3 subjects aged between 25 and 35 years who had metal-composite dental prosthetic pieces, with an average age of 2 years.

The first patient (P_1) presented a metal-composite prosthesis at the 1.4 tooth. The plating material was represented by *Solidex* and 2 years old. The first sample was taken from the vestibular surface of the tooth and the second one from the vestibular surface of 2.4.

The second patient (P_2) presented a dental prosthesis at 1.2, 1.1, 2.1, 2.2, 2.3. The plating material was represented by *Gradia*, and it was cemented 1 year and seven months before. The first sample from this patient was taken off the vestibular surface of 2.3, while the second one, used as control, was collected from the vestibular area of the 1.3.

The third patient (P3) from whom we collected the samples presented a dental metal-composite prosthesis at 1.5, 1.6, 1.7 teeth. The first sample was taken from the vestibular surface of 1.5, and the second one from 2.5. In this case the plating material was represented by *Ceramage* and the prosthesis was cemented 2 and half years ago.

For each case, after achieving the prosthetic result, a post-prosthetic treatment has been applied, consisting of: indications given to the patient regarding the use of the prosthesis and regular check-ups every 6 months in order to asses oral hygiene, device adaptation to oral conditions and the verification of mental, mechanical, functional and biological prosthetic adjustment.

We have conducted 2 tests for each patient: the first sample was collected from the surface of the prosthetics (P_xa), and the second one, was a sample from the natural surface of the homologous tooth (P_xb), in order to refer results to the same surface.

Sampling was conducted 2 hours after regular oral hygiene (under normal individual conditions of hygiene) using sterile swabs

After collection the buffer was immersed in the transport environment and immediately sent to the laboratory for examination (4,5).

The first stage of the laboratory analysis has been obtaining the initial suspension made for each sample using an agitator (Bio Vortex v1) for 2 minutes.

To determine the number of microorganisms the method of plate cultivation was used; the method uses the colony developed after inseminating agar areas in Petri plates, with dilutions obtained from the analyzed sample.

From the last dilution tubes, inseminations were made on agars (to asses the total number of bacteria), on MRS (for lactobacilli) and then Columbia-agar-blood (for *streptococci*, *staphylococci* and *Corynebacterium*), and incubated for 48 hours at 37^0C.

Colony counting was done with the naked eye under magnifying glass to enlarge, the best dilution was considered to be the one from which a number of 50-100 colonies developed on the plate.

The number of germs in a initially examined product was calculated by formula: **$UFC/ml=ax10^n/V$**, in which: UFC = colony-forming units, a = average number of colonies developed by insemination; 10 = coefficient of dilution, n = number of the dilution in which the insemination was made (taken with changed sign), V = volume (in ml) of suspension used to inseminate.

The bacteria examination in colored smears has taken the following steps: displaying (preceded by mixing portions of a colony in a drop of serum physiological), drying at room temperature, warmth and color fixation (Gram coloration).

Preparations were examined under a Nikon Eklipse 600 microscope, and photographs were obtained by a 100 x objective (with oil immersion), with a Nikon Cool Pen digital camera, with 1600 x 1200 dpi resolution.

RESULTS AND DISSCUTIONS

Bringing a foreign device in the oral environment will increase the oral microbiota by the apparition of new retentive areas where they can develop new environmental niches. In addition, the placing of the prosthesis edges under the gingival margin creates irritation by retaining bacterial plaque in the cement irregularities from the joint's periphery (3).

A person that wears the device should be made aware of the risks that such treatment rises and should be taught that the last phase of treatment is not cementing, but maintaining oral hygiene and repeated inspections made at regular check-ups.

Prognosis of a prosthetic treatment depends directly on bacterial plaque control. However, the most important aspect in the control of bacterial plaque is patient motivation. When the motivation is absent, no material, no matter how sophisticated, will obtain the expected results.

Of major importance in maintaining oral hygiene is the strict microfilm and plaque control, microfilm formed in the crevicular area. If the patient does not maintain a perfect oral hygiene any prosthetic treatment is compromised by installing periodontal disease.

Bacterial plaque may cover any part of the tooth, but is especially cervical located. Because it’s adherence to the dental structures, removing it can be made only by proper means of oral hygiene

Maintaining the health status in the area were the tissues that support and/or come in contact with a fixed prosthesis, represents the objective of the tertiary prophylaxis. The concept of prevention for prosthetic restorations in general aims for maintaining the eutroficity status for as long as the tissues come in contact with the prosthesis, which in turn should have a very low biological, physical or chemical influence.

It is known that dental plaque, deposits and plaque colonize the surfaces of both natural and artificial teeth.

Following this study we found that oral microorganisms are found in greater number on the surface of prosthetic parts than on the surface of natural counterparts teeth, the number varying from one subject to another.

Numerical estimate of oral microbiota:

Laboratory analysis result of samples collected from patients with metal-composite prosthetic devices showed the broad differences between the number and type of microorganisms that have colonized the artificial surfaces compared to natural ones. Of course, in these cases a number of factors that have influenced the results were taken into account, factors unrelated to the premises of our study, such as patient age, prosthetic work, individual oral hygiene, and individual dexterity.

Results were presented comparing the investigated artificial tooth and the natural counterpart one, present on the arch and used as a control. These results were reported to a surface unit for the tested area, in order to provide data repeatability, but also to create a certain system of reference.

Results thus obtained showed a significant difference between the two groups: study group (teeth reconstructed with composite materials) and control group (natural teeth). Although prosthetic parts were finished and polished to allow a smooth self-cleaning surface and subjects have used additional methods of oral hygiene (dental thread and mouthwash), the prosthetic surface presented a far greater number of microorganisms compared with the surface of natural counterparts teeth thus, in general, values were ten times higher in favor of the prosthetic surface compared with the natural control teeth (Fig 1 -3).

The greatest number of lactobacilli was found on the *Solidex* surface, and from the total microorganisms identified in this study, the largest percentage of retained bacteria was obtained by *Ceramage* (58%), followed by *Solidex* (7%) and *Gradia* (6%).

The lowest rate of surface adherence has been observed for the *Gradia* plating material, a reality that empowers us to say, once again, that the enhanced dental research in the field of composite materials will offer new materials with increased performance that will erase the disadvantages related to their behavior under constant submission to oral environment factors.

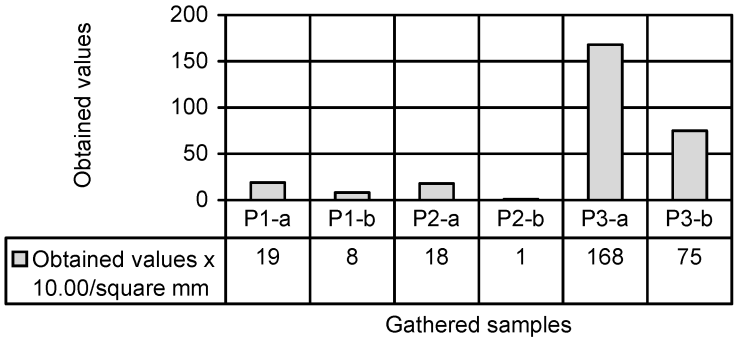

Fig. 1 - The total number of adherent bacteria on dental composite prosthetic parts, with an average age of 2 years

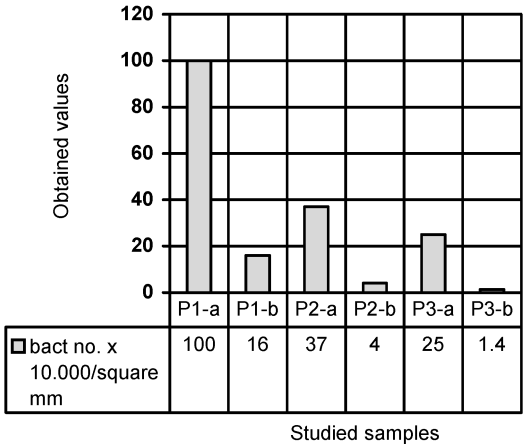

Fig. 2 - Number of lactobacilli adherent on dental composite prosthetic parts, with an average age of 2 years

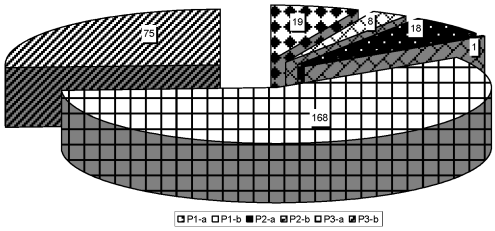

Fig. 3 - Percentage distribution of oral microorganisms on different dental composite prosthetic surfaces, with an average age of 2 years

Morphological characterization of oral microbiota components

Through a smear examination under the microscope, the following morphological types of bacteria were identified (Table 1):

- **coci** displayed in chains - streptococci (oval shaped cells, with a diameter of 0,8-1 μm immobile, unsporulated, uncapsulated) and in a bunch display - staphylococci (spherical when they are isolated or ovoid, flattened on a side when they are grouped, with diameter between 0.8 and 1 μm the bunch display is typical in the case of cultures developed on solid medium-agars) - (fig 4-5)
- **bacili** - isolated (cylindrical bacteria in a stick-like form, the longitudinal diameter is several times larger than the transverse, immobile, unsporulated, uncapsulated) or grouped (more rarely seated in palisades or forming chains displayng angles between them forming letters of the alphabet: L, M, N, V, etc.) - (fig.6).

Table No.1-Morphological characteristics of isolated microorganisms

No.	Morphological type	Gram coloration	Bacteria display	Other characteristics
1	coci	G +	Streptococci (chains)	
2	coci	G +	Staphilococi (group)	
3	bacilli	G +	isolated	Cells grouped in the form of letters of the alphabet
4	streptococci	G +	chains	
5	streptococci	G +	chains	
6	bacilli	G +	isolated	

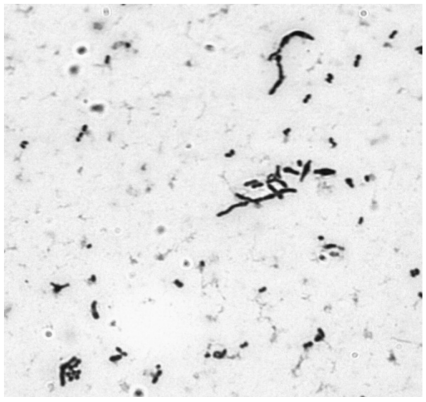

Fig. 4 - Image representing chains deployment of the streptococci

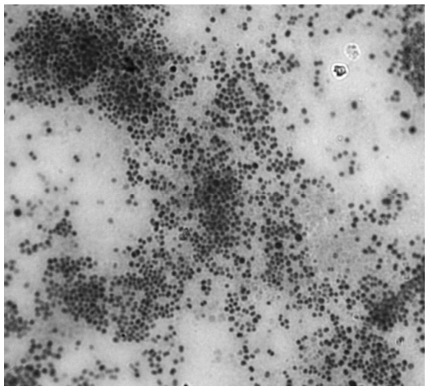

Fig.5 – *Staphilococi* -group deployment.

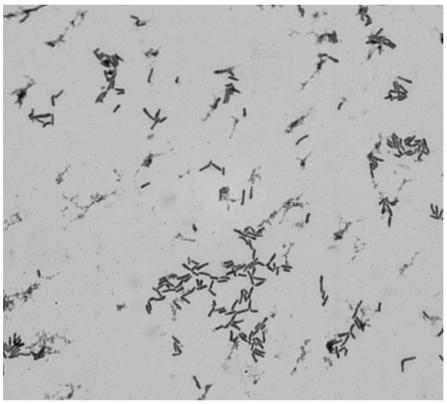

Fig. 6 – Bacilli microscopic image

As a result, although in the technological development process of a metal- composite dental prosthesis a series of mechanical processing actions are included (actions that aim to achieve a perfect smooth area), over their life time usage the surface status changes; as a result, it becomes rough and thus retentive for oral micro-flora and, in advanced usage stages, retentive for food, in the case when this issue is doubled by an inadequate oral hygiene.

As a result, regardless of the finishing and polishing degree of the composite plating material, over time, its surface suffers alterations, becoming increasingly rough and retentive for bacteria.

CONCLUSIONS

Despite the latest spectacular evolution of composite materials, we can say that their introduction in the oral environment produces an imbalance. The magnitude of this distortion depends on the resin processing degree and on material properties.

As in any type of dental prosthetics the problems appear in a long-term results analysis, when material aging phenomenon appears, and when bio-compatibility decrease.

Proper hygiene, restoration surface status close to the physiological one is obtained by two ways: self-cleaning and professional cleaning. For the biological integration to be achieved more easily, the dental technician must take into account and implement the restoration principles that ensure a smooth self-cleaning surface. These rules refer to:

- Correct design of the physiognomic component;
- Rational modulation of the inter-dental spaces;
- The design of the intermediary parts, and their relation to the edentulous ridge;
- The avoidance of concave, retentive areas;
- Finishing and polishing to a glass like status of the metallic and plating component.

Judging in the light of the benefits of these materials (lower price cost, less technology, compared to ceramic materials), we have the certainty that in the future, their drawbacks will be successfully overcome by the manufacturers.

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