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In vitro Study of the Materials of the Components of the Implant Overdentures

DANIELA MIHAELA MEGHEA[1], CRISTINA TEODORA PREOTEASA[2]*, ELENA PREOTEASA[1], MIHAELA MARIN[1], CATALINA MURARIU MAGUREANU[1]
[1] Carol Davila University of Medicine and Pharmacy, Faculty of Dental Medicine, Department of Prosthodontics, 37 Dionisie Lupu Str., Bucharest, Romania
[2] Carol Davila University of Medicine and Pharmacy, Faculty of Dental Medicine, Department of Scientific Research Methodology and Ergonomics, 37 Dionisie Lupu Str., Bucharest, Romania

***Abstract**: To analyze the implant overdentures components as following: morphology and composition aspects of several small diameter systems, evaluating both the implant and the retention system (1); biomechanical characteristics of some of the polymers used for manufacturing the overdenture base (2). An experimental in vitro study was carried out through optic microscopy to analyze the metallic structure of dental implants components and anchor systems. The marketed products analyzed were mini[1]SKY (Bredent) and 3M ESPE MDI Mini dental implant (3M ESPE). A Zeiss microscope with Kohler illumination was used. Samples from polymeric materials commonly used for overdenture based (i.e. Duracryl and Superacryl) were analyzed by 3 point bending test by Tira device. In the case of the mini[1]SKY system (Bredent), the dental implant has the typical structure of marketed pure titanium that went through thermomechanical processing, resulting in a higher rate of granulation, and the matrix has a completely different structure, as it is a monophasic structure specific to materials that crystalize in the cubic system of the stainless steels. In the case of the 3M ESPE MDI system, the implant and the matrix have a similar microstructure, specific to a Ti-6Al-4V beta alloy annealed with an extremely fine rate of granulation. Duracryl and Superacryl samples fractured variable (frequently close to the midline) and were similar in regard to the value of breaking force. Knowledge of the materials of the components of implant overdenture guide their selection from the point of view of biocompatibility, resistance and ensuring denture retention.*

***Keywords**: dental implants, attachment system, in vitro, edentulism*

1. Introduction

Nowadays, the demographic changes including population aging, through the increase of elderly people, with its specific aspects, the occurrence of extended or complete edentulism at older age, but also the befits brought about by prosthetics through the use of implants, led to overdenture turn into an optimum prosthetic solution for elderly patients who are partially or completely edentulous [1]. The long-term evaluation of implant overdenture and associated complications and with various implant retention systems, can supply useful information for the doctor in order to select the type of anchorage system (materials and appropriate design for overdenture) [2].

An important aspect of implant overdentures' components as far as biomaterials are concerned is their behavior in the oral environment, directly related to their structure and composition. One of the mostly used methods to characterize materials, especially when aiming at a deep analysis of their structure and morphology, is the microscopic analysis. For the macroscopic characterization of materials, as well as for the faults or artefacts on their surfaces, a frequently used method is the stereo-microscopic analysis [3]. Within the study, there are two characterization methods applied to the selected implant and retention systems, used in overdenture, namely stereomicroscopy and optical reflected light microscopy.

**email: cristina.preoteasa@umfcd.ro*

The purpose of the study was to analyze the implant overdentures components as following: construction, morphology and composition aspects of several small diameter systems of implants used in overdenture, evaluating both the implant and the retention system [1]; biomechanical characteristics of some of the polymers used for manufacturing the overdenture base [2].

2. Materials and methods

An *in vitro* study was conducted, following the analysis of the components of implant overdentures.

The implant and their retention systems were analyzed through optical microscopy. With the purpose of this analysis, two mini implant options were selected that were meant to be used in overdentures, belonging to the small diameter implant systems (a diameter smaller than 3 mm), screw-type designs, rough surface, acid-treated, monoblock, similar design (spherical abutment joined to the implant), with O-ring retention system. The implant is inserted into the bone, being self-tapping. The part of the abutment that comes into contact with the O-ring remains outside the bone, being positioned supra-gingival. The metal ring/the matrix, belonging to the retention system, is being fixed into the basis of the overdenture. The marketed products submitted to analysis were mini^{1}SKY (Bredent) and 3M ESPE MDI Mini dental implant (3M ESPE) (Figure 1). The initial naming of the 3M ESPE MDI was dental mini implant (Sendax MDI, IMTEC), being the first implants of this kind used in overdenture treatment. They were developed more than 30 years ago and certified in 1997, by Victor I. Sendax, in the USA, and later on introduced in the dental implant procedures, in April 1999. The processing of these implants by 3M ESPE took place in 2008 [4, 5].

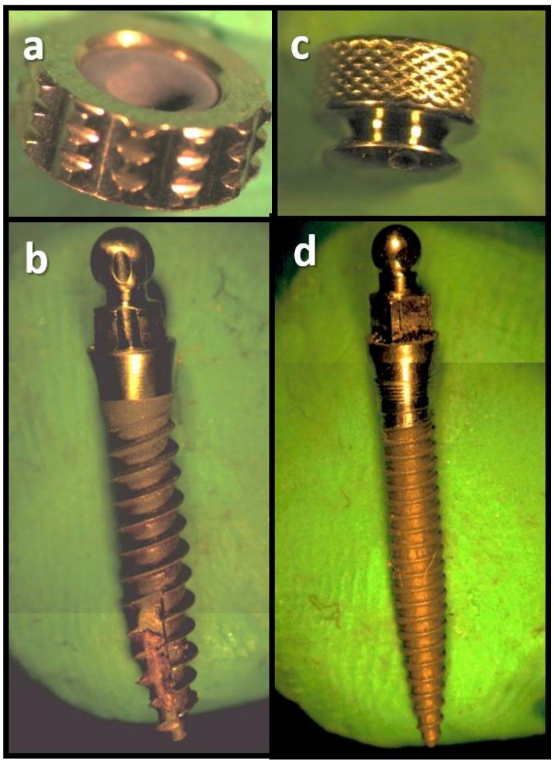

Figure 1. Dental implant systems used in the study a) metal ring of the O-ring retention system from mini^{1}SKY b) mini^{1}SKY implant c) matrix of the O-ring retention system from 3M ESPE MDI d) 3M ESPE MDI implant

In order to analyze the metal components, samples were taken, by detaching material from both the endo-bone implant and the retention system. The sampling site should be chosen so that the sample is representative of the material under investigation, it matches the purpose of the research and contains the crystal structure and possible variations in structure as well. The samples were taken as follows: one sample from each type of endo-bone implant (2 samples); one sample from each type of metal matrix (2 samples).

The subsequent preparation of the samples taken is aimed at obtaining a flat and extremely smooth surface for macro and microscopic analysis in reflected light and this is carried out according to the guidelines of the standard 4203-74 [6, 7]. Sample preparation consists of the following operations: sampling, polishing, dry grinding, final polishing, etching with specific reagents, using advanced preparation methods that are related to the nature of the material, in terms of mechanical properties, i.e. hardness and plasticity. The parameters defining the preparation method are: type of abrasive disc or abrasive paper, type of sample, pressing force, time. The abrasive grit size should be chosen according to the quality of the surface to be prepared, initially as fine as possible, and the jump to the next step, in terms of grit size, as large as possible in order to shorten the preparation time. The cooling and lubricating liquid is dependent on the nature of the material prepared and on the current stage of preparation. The pressing force of the samples on the surface of the abrasive disc is important. A force that is too high can lead, by strong local heating, to the thermal distortion of the structure, while a force that is too low results into an unacceptable increase in grinding time. This clamping force is determined according to the nature of the material, the total area of the surface to be sanded and the time required for sanding. Too long sanding times can lead to artefacts such as surface relief or ovalisation of the sample.

The metallographic samples were prepared on Buehler equipment. Thus, metallographic samples were cut using a special B 2211 disc, specific to titanium alloy and the cutting was carried out using coolant, in order to avoid heating of the sample and modification of the material structure. The fluid used (ABRASIVE CUTTING FLUID) contains distilled water and lubricant. The embedding of metallographic samples was performed in BAKELITE powder on a Buehler - SIMPLIMET 1000 AUTOMATIC MOUNTING PRESS (Figure 2), the working parameters used are given in Table 1.

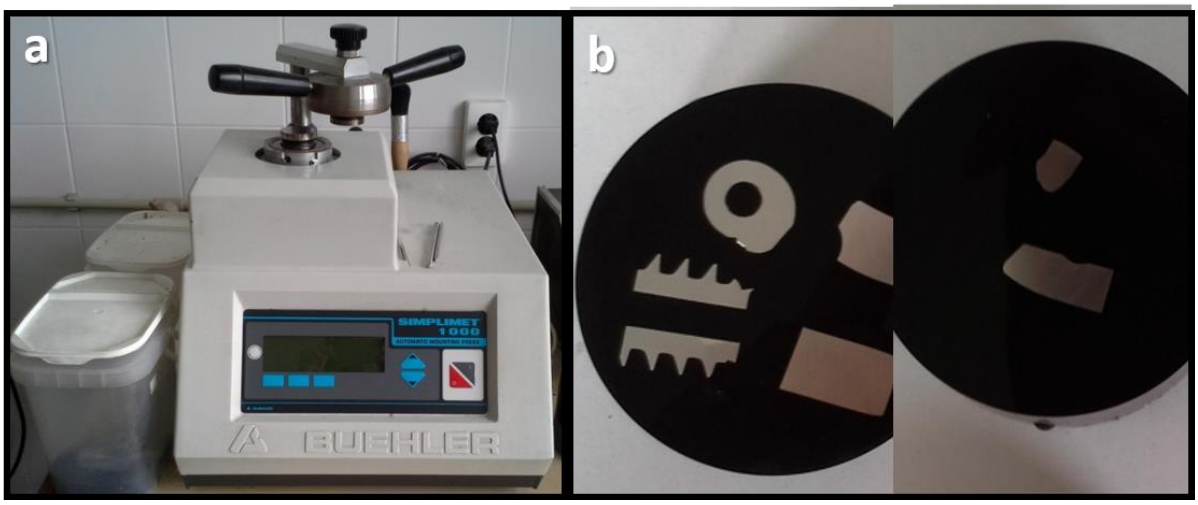

Figure 2. Sample preparation a) the Buehler SIMPLIMET 1000 AUTOMATIC MOUNTING PRESS on which the samples were embedded; b) the appearance of the metallographic samples after embedding

Table 1. Characteristics of the embedding machine

Parameter	Value
Working pressure	300 bar
Preheating pressure	10 bar
Working temperature	150^0C
Heating/cooling time	8 min/3 min

After embedding, the samples were subjected to the polishing process on a Buehler VECTOR automatic machine with a platen having the characteristics detailed in Table 2, comprising of several polishing stages and the device used for polishing can be seen in Figure 3.

Figure 3. The Buehler VECTOR machine where the samples were polished

Table 2. Sample polishing - parameters

Parameters	Stage 1	Stage 2	Stage 3	Stage 4	Stage 5
Sample diameter (mm)	30	30	30	30	30
Turntable speed (rpm)	250	200	150	120	120
Head speed (rpm)	60	60	60	60	60
Working time (min)	6	5	10	10	10
Textile medium	TEXMET	TEXMET	TEXMET	TEXMET	TEXMET
Abrasive - METADI type suspension with different particle diameters (μm)	15	9	3	1	0.05
Relative head/flat rotation	Opposite direction	Opposite direction	Opposite direction	Same direction	Same direction
Pressing force (N/test)	15	15	15	5	5

Subsequently, the sample surface was attacked with chemical reagents. This procedure is generally carried out with solutions of acids (specific combinations of acids) and aims at highlighting the structural constituents of the metal sample. The chemical attack was carried out using Keller Reagent (190mL distilled water, HNO_3 5mL, HCl 3mL, HF 2mL) or Kroll Reagent (92mL distilled water, HNO_3 6mL, HF 2mL). When the reagent attacks the grain boundaries, the chemical attack dissolves the crystalline grains differentially, depending on the nature of the phases and their crystallographic orientation. The chemical reagent can form deposits of constituents on the surface of the crystalline grains, which cause them to color. Thus, nitric acid forms oxide layers of varying thickness on the surface of the grains, ranging in color from light yellow (thin layer) to dark brown (thick layer). Chemical attack is carried out either by immersing and shaking the sample in the reagent or by wiping the surface of the sample with a cotton swab soaked in the reagent until the mirror shine disappears. The sample is then washed in water or alcohol and dried by blotting on filter paper or using a hot air jet. The duration of the attack varies from a few seconds to a few minutes depending on the chemical composition and on the structure of the sample material. Anisotropic constituents (which crystallize in systems other than the cubic system) can be revealed by microscopic analysis under polarized light. Non-metallic inclusions can be observed after polishing the samples and the structural constituents after chemical attack.

A Zeiss stereomicroscope with Kohler illumination was used for the stereomicroscopy analyses, which allows the investigation of non-transparent objects using reflected, natural or electric light, with a magnification power ranging from 10x to 80x (Figure 4). An objective lens with a magnification power of 20x was used together with a 10x eyepiece. The images were captured using an Olympus camera with a 4x zoom. The light reflected from the object (sample surface) passes through the objective, which forms a magnified and flipped intermediate image. This image is then magnified by the eyepiece, forming an image visible to the eye or a real image projected onto a monitor. By examining a suitably prepared material by optical microscopy, one can observe the phases that make up the material, the grains, the distribution of phases and grains, their nature, the size of the crystals, etc. Large magnifications have been chosen in the presentation of the results in order to highlight the structural aspects as clearly as possible (100x and 500x).

Figure 4. The optical microscope used

Some of the materials used for the manufacturing of the implant overdenture base were used, namely Duracryl and Superacryl (Spofa). These materials are both polymers (Polymethyl methacrylate), are available in powder and liquid format. From a clinical usage perspective the main difference between these is that the first is a self-cured material (it is frequently used in the dental office for clinical application, e.g. repairing the fractured overdenture base, fixation of the matrix in the overdenture base, relining), while the second one is a heat-cured material (it is used in the dental laboratory mainly for manufacturing the base of removable dentures, implant overdentures included). From each material five sample of 80X64X4 mm were made accordingly to the manufacturer recommendations and tested. The Tira test device was used, being applied a 3point bending test, the speed used being 2 mm/min.

3. Results and discussions

In the case of the mini^{1}SKY system, the endossseous implant has a structure typical of pure commercial titanium that has undergone thermomechanical processing, with a larger grain size (Figure 5). The matrix of the mini^{1}SKY system has a completely different structure, with a single-phase microstructure typical of materials crystallizing in the cubic system, specific to stainless steels (Figure 6). As a retention system for the overdenture on implants, Bredent also has another variant, namely a gold matrix, but in this case the metal ring of the O-ring system, the commonly used variant, was analyzed.

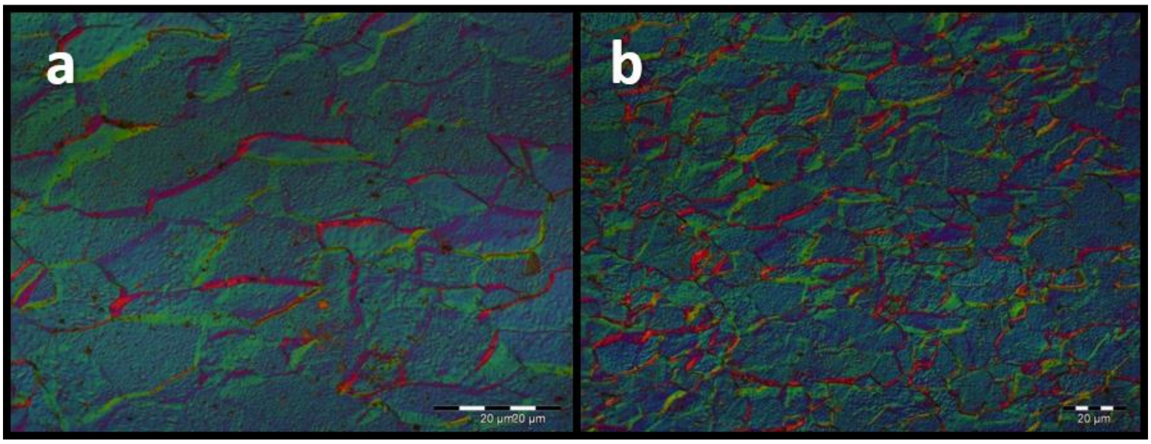

Figure 5. Light microscopy image of the sample taken from the mini[1]SKY implant, magnification 500x, acid attack with Kroll Reagent, immersion 15s, image obtained in differential phase contrast a) cross section; b) longitudinal section

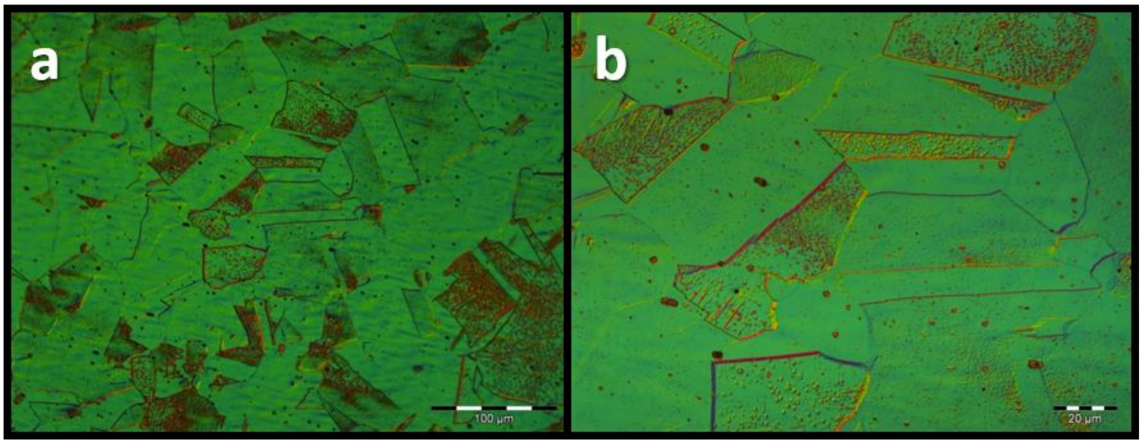

Figure 6. Optical microscopy image of the sample taken from the matrix (metal ring) of the mini[1]SKY system, magnification 500x, chemical attack with Kroll Reagent, immersion 15s, image obtained in differential phase contrast a) cross section; b) longitudinal section

In the case of the 3M ESPE MDI system, the microstructure of the implant is specific for a beta-annealed Ti-6Al-4V alloy with an extremely fine grain size that gives the implants exceptional mechanical strength. No serious differences were observed between the cross-section and the longitudinal section after the attack with Keller Regent (Figure 7). For the matrix system of 3M ESPE MDI system, the same type of structure with very fine grain as in the case of the implants is observed (Figure 8), probably the materials used in the realization of the endosseous implant and attachment system are similar, unlike the mini[1]SKY system where differences are observed between the microstructures of the component materials (implant-matrix). Considering the direct relationship of the implant with the bone, in both cases the composition of the endosseous implant is based on titanium as biomaterial, either as pure titanium (mini[1]SKY) or as titanium alloy (3M ESPE MDI).

Stereomicroscopic metallographic analyses of mini[1]SKY and 3M ESPE MDI systems demonstrated differences between the compositions of the two types of mini-implants, as well as between the corresponding retention systems. Differences were also found between the components of the same system (implant and retention system), as this was observed for the system supplied by Bredent.

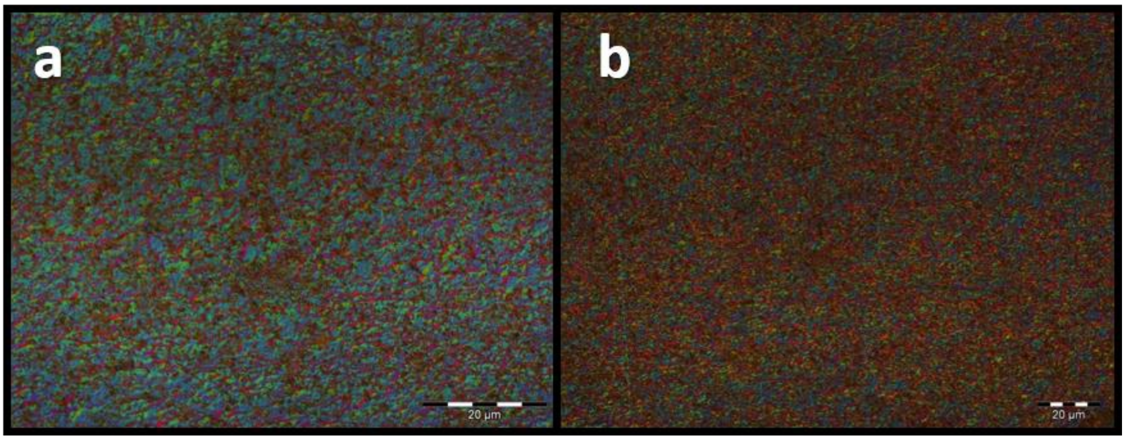

Figure 7. Optical microscopy image of the sample taken from the 3M ESPE MDI implant, magnification 500x, attack with Keller Reagent, immersion 20s, image obtained: a) cross section, image obtained in bright field; b) longitudinal section, image obtained in dark field

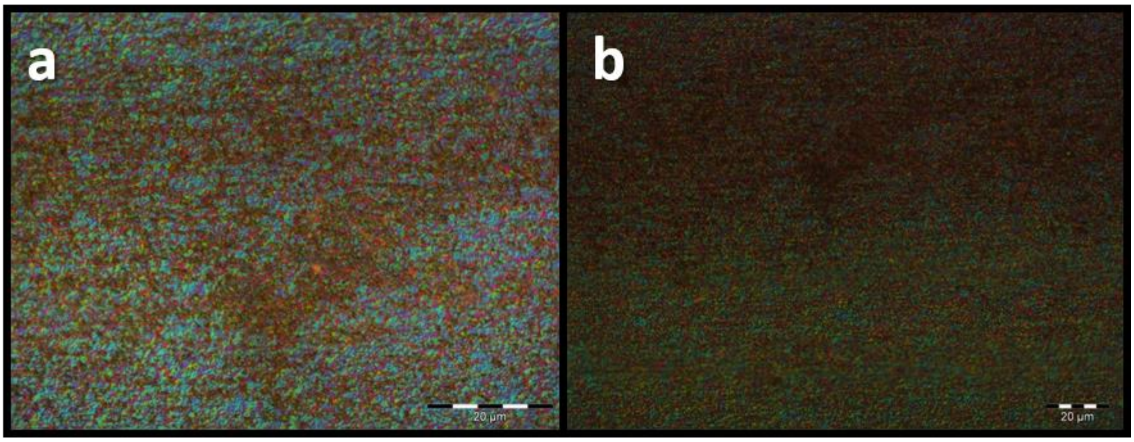

Figure 8. Optical microscopy image of the sample taken from the matrix of 3M ESPE MDI, Keller Reagent, 20s immersion, image obtained: a) cross section, image obtained in bright field; b) longitudinal section, image obtained in dark field

Implants from 3M ESPE MDI system have a microstructure typical of a beta annealed Ti-6Al-4V alloy with an extremely fine grain size that gives the implants an exceptional mechanical strength. As for the matrices of the retention systems of these manufacturers, the annealing heat treatment probably led to a completely lamellar microstructure formed by large colonies of co-oriented alpha lamellae between the beta grains, which are bounded by the boundaries of the alpha grains, spectacular structures typical of these types of alloys. From the images obtained on these systems, a peculiar care can be observed in their thermo-mechanical processing, as the resulting structures are almost free of imperfections, which favors the very good behavior and clinical tolerance of these biomaterials. The images also show that the aligned alpha wafers are separated by thin beta remnant ribs. These ribs are likely to be ineffective barriers against sliding. However, the colonies have different crystallographic orientations and therefore would be expected, like beta grain boundaries, to act as barriers against slipping. In fact, boundaries between colonies and remnant beta grains are not always effective barriers against slipping.

Mini[1]SKY implants have a structure typical of pure commercial titanium that has undergone thermomechanical processing. Thermomechanical processing of pure commercial titanium is usually performed completely in the alpha phase domain and the main microstructural objective is to adjust the grain size to the desired level. Due to its good deformability the deformation of titanium is carried out at relatively low temperatures (200-300°C) and even at room temperature (cold deformation).

Moreover, the desired grain size can be adjusted by choosing the temperature for the recrystallization treatment. For example, very small grain sizes visible in the microstructures obtained in the case of 3M ESPE MDI implants can be reached over time by recrystallization at around 700-750°C, and larger grain sizes can be obtained by recrystallization at around 900°C.

In addition to oxygen and grain size, the crystallographic texture of the hexagonal alpha phase developed during the deformation step, unaltered significantly by recrystallization, is a third important parameter for the mechanical properties of titanium.

Regarding the microstructures obtained on samples from the retention system matrixes, it can be clearly seen that the mini[1]SKY system matrix presents a completely different structure from the implant and the other retention system (from 3M ESPE), as a monophasic microstructure characteristic of materials crystallizing in the cubic system, by the appearance in their structure of deformation macules, specific to stainless steels, of which the metal ring is made. The macules, resulting from the cold deformation of the crystal under the action of external forces, will behave differently when attacked with metallographic reagents.

Comparing the mechanical properties of fully equiaxial microstructures presented over time in literature with those of two-phase microstructures, in general terms, it is important that in the case of strong crystallographic texturing the slip length in fully equiaxial microstructures is much larger than the grain size, which can especially reduce the resistance against fatigue failure (characteristic of this type of application). In other words, the volume fraction of the alpha phase in two-phase microstructures should be kept below about 50%, in order to avoid extensive clustering of beta titanium grains.

The Polymethyl methacrylate samples of Duracryl and Superacryl fractured variable, most of them closely to the midline, some of them paramedian (Figure 9). The value of breaking force was similar and not statically significant between the two products, even so range of the values for Duracryl was higher than for Superacryl (Table 3). These results suggest that overdenture fracture, a known and relative frequent complication of overdenture treatment, is not related to the type of acrylic (e.g. with different curing method) used. In removable denture there are many risk factors for denture’s fracture, and these need to be carefully analyzed and integrated in the treatment plan. In implant overdenture in particular one must consider that usage of attachment system relates to a decrease thickness of overdenture base at that level, and also that the progressive ridge resorption leads in time to the support of overdenture base mainly on the implants, as both of these are factors for the fracture of overdenture base. In this regard, carefully treatment planning and recall appointments are recommended for ensuring a good long term outcome.

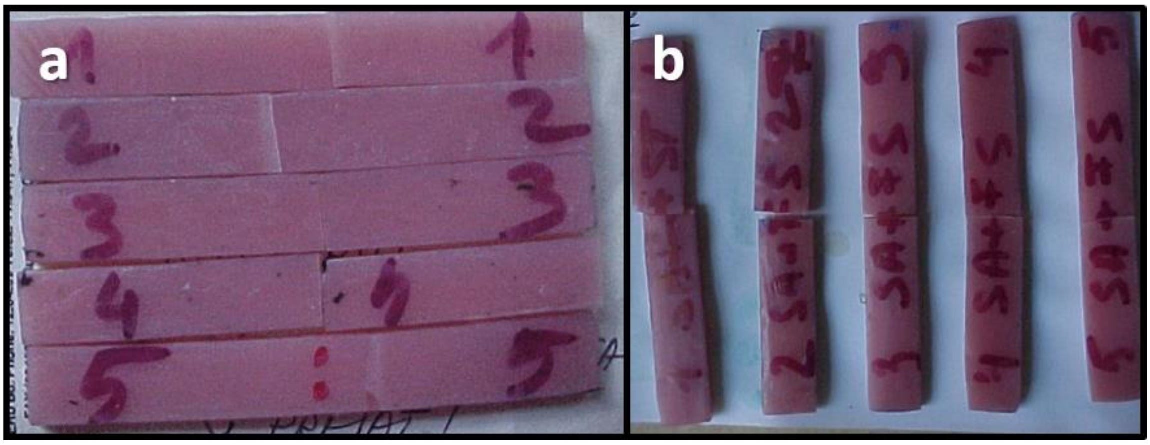

Figure 9. Line fracture for Polymethil methacrylate materials used for overdenture base a) Duracryl b) Superacryl

Parameters	Duracryl	Superacryl	p
Mean (N)	150.64	149.12	0.754
Median (N)	150	158.60	
Minim (N)	114.00	115.60	
Maxim (N)	190.10	173.50	

Table 3. Breaking force of Polymethil methacrylate materials used for overdenture base

Factors that enter into the selection process of dental materials in general, including implant and overdenture retention systems, are their biomechanical characteristics [8], as well as their properties that support their biocompatibility and mechanical strength [9, 10], aspects that are well documented for titanium and titanium alloys. The success of prosthetic treatments in general, and of implant treatments in particular depends on many factors, such as bacterial biofilm formation, which is different depending on material type [11]. Along with these very important characteristics for tolerance and durability of treatment, extensive previous clinical experience, cost of implants and other factors influencing the choice of implant system and overdenture retention are also involved. In these high performance medical applications, fatigue strength or creep resistance and other mechanical properties make the choice of implants in terms of material, size and design a very difficult issue. Titanium is well known for its mechanical strength, corrosion resistance and overall chemically inert behavior, however, in these applications, titanium is not the only choice. Therefore, competing materials such as titanium alloys, zirconium oxide, sometimes gold or stainless steel (for retention systems) are also alternatives being considered. Since pure titanium is usually more expensive, the life cycle costs of titanium implants, determined by durability, become an important aspect of the final decision on the selection of the material of which the components of retention systems are made, as the higher or lower hardness may affect the other component in terms of its morphology and dimensions and their role in the retention of the prosthesis.

Reduced diameter implants show good clinical performance, with a survival rate of over 90%, higher in the mandible than in the maxilla, associating good patient satisfaction and adequate mastication with this treatment option [12, 13]. Research reported in the literature indicates different success and survival rates of dental implants [14], which may also be related to the particularities of the studies carried out, the prosthetic treatments, and the particularities of the implant systems used, which are available in a wide range. Fracture of implants is a common complication, which may occur during insertion [15] or afterwards [1], and may be related to the design of this particular type of implant, but also to the material of which it is made. The particularities of the attachment system are also important and may influence the success of the treatment, with different variants associating for example different marginal bone resorption [16]. Consequently, given the many differences between small-diameter implant systems used in overdenture, it is important to know their structural features and to see to what extent this information explains the observed clinical behavior, which will contribute to the improvement of existing variants and prosthetic treatments.

4. Conclusions

Aspects related to the biocompatibility, structure, resistance to breakage or wear of the components of implant overdentures are important to be known as explaining clinical aspects and useful in the complex process of their selection between different available alternatives. The structure and surface of implants are important in the bone integration process, as biomaterials. Retention systems must ensure and maintain retention over time in relation to the implant component, by preserving the morphology of the interrelated components and by preserving the space necessary for the interposition of non-metallic components that play a role in retention (rubber ring, silicone matrix, Teflon, etc.) and that can be replaced in the event of their exhaustion or reduction in retention of the prosthesis. The variants of single-piece implants with O-ring retention systems and polymeric materials analyzed are among the most widely used in overdenture, with mini-implants offering a number of advantages in their use through minimally invasive techniques with immediate loading, and retention systems offering good retention of the prosthesis with minimal complications and easy to repair. Considering the higher risk of fracture of the overdenture base, a careful analysis of available space, of material thickness, of selecting most appropriate polymeric material must be conducted.

References

1.PREOTEASA, E., IMRE, M., PREOTEASA, C.T., A 3-year follow-up study of overdentures retained by mini-dental implants, *Int J Oral Maxillofac Implants*, 29 (5), 2014, 1170-1176.
2.ANDREIOTELLI, M., ATT, W., STRUB, J.R., Prosthodontic complications with implant overdentures: a systematic literature review, *Int J Prosthodont,* 23(3), 2010, 195-203.
3. BHATAVADEKAR, N.B., GHARPURE, A.S., BALASUBRAMANIUM, N., SCHEYER, E.T., In Vitro Surface Testing Methods for Dental Implants-Interpretation and Clinical Relevance: A Review, *Compend Contin Educ Dent*, 41(3), 2020, e1-e9.
4. SINGH, R.D., RAMASHANKER, CHAND, P., Management of atrophic mandibular ridge with mini dental implant system, *Natl J Maxillofac Surg*, 1(2), 2010, 176-178.
5.GLEIZNYS, A., SKIRBUTIS, G., HARB, A., BARZDZIUKAITE, I., GRINYTE, I., New approach towards mini dental implants and small-diameter implants: an option for long-term prostheses, *Stomatologija*, 14(2), 2012, 39-45.
6. RUSANESCU, C.O., RUSANESCU, M., JINESCU, C., BIRIS, S.S., Laser Hardening Influence of Metal Surfaces, *Mater. Plast.*, **55**(2), 2018, 184-187.
7.SARACIN, A., VOICU GH SARACIN, I., PANDIA, O., Determination of use of dental materials and stomatological frees on stand. In *Proceedings of the Conference "Engineering for Rural Developing"*, Jelgava, Latvia, 22–24 May 2019.
8.FEURDEAN, C.N., BABTAN, A-M., IONEL, A., ILEA, A., BUHATEL, D., SAVA, A., MOLDOVAN, M., SAROSI, C., VESA, S., CAMPIAN, R.S., Hybrid Dental Composites Biomechanical Properties, *Mater. Plast.*, **57**(3), 2020, 70-80.
9. TOPOR. G., EARAR, K., CIUCA, I., BERBECE, S., STEFANESCU, V., BEZNEA, A., GRIGORE, C.A., MARIS, M., FOTEA, S., ONISOR, C., FEIER, R., IORDACHE, C.M., GURAU, G., Mechanical Properties of Bis-GMA/HEMA Resin Composite with Addition of Silicon Dioxide Nanoparticles, *Mater. Plast,* **58**(1), 2021, 257-264.
10.DIACONU-POPA, D., VITALARIU, A., TATARCIUC, M., FRATILA, D., Studies on the Mechanical Parameters of Denture Base Acrylic Resins, *Mater. Plast.*, **57**(4), 2020, 360-365.
11.BOLAT, M., BOSINCEANU, D.G., SANDU, I.G., BOSINCEANU, D.N., SURLARI, Z., BALCOS, C., SOLOMON, O., VITALARIU, A., Comparative study on the degree of bacterial biofilm formation of dental bridges made from three types of materials, *Mater. Plast.*, **56**(1), 2019, 144-147.
12 LEMOS, C.A., VERRI, F.R., BATISTA, V.E., JÚNIOR, J.F., MELLO, C.C., PELLIZZER, E.P., Complete overdentures retained by mini implants: A systematic review, *J Dent*, 57, 2017, 4-13.
13.PREOTEASA, E., ONCESCU MORARU, A.M., MEGHEA, D., MURARIU MAGUREANU, C., PREOTEASA, C.T., Food Bolus properties in relation to dentate and prosthetic status, *Healthcare*, 10(9), 2022, 1596, https://doi.org/10.3390/healthcare10091596
14.VI, S., PHAM, D., DU, Y.Y.M., ARORA, H., TADAKAMADLA, S.K., Mini-Implant-Retained Overdentures for the Rehabilitation of Completely Edentulous Maxillae: A Systematic Review and Meta-Analysis, *Int J Environ Res Public Health*, 18(8), 2021, 4377, doi: 10.3390/ijerph.
15.SCHWINDLING, F.S., SCHWINDLING, F.P., Mini dental implants retaining mandibular overdentures: A dental practice-based retrospective analysis, *J Prosthodont Res*, 60(3), 2016, 193-198.
16.AUNMEUNGTONG, W., KUMCHAI, T., STRIETZEL, F.P., REICHART, P.A., KHONGKHUNTHIAN, P., Comparative Clinical Study of Conventional Dental Implants and Mini Dental Implants for Mandibular Overdentures: A Randomized Clinical Trial, *Clin Implant Dent Relat Res*, 19(2), 2017, 328-340.

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