



## EVALUATION OF RETENTION AND MICROBIAL COLONIZATION OF TWO DIFFERENT DENTURE BASE MATERIAL

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### **ABSTRACT**

**The aim of this study:** Is to evaluate the effect of conventional heat cure acrylic resin denture base and thermoplastic resin base on the retention and bacterial colonization in complete dentures.

**Material and methods:** Twenty completely edentulous patients were selected and randomly grouped into two equal groups: group I received maxillary and mandibular complete dentures made of conventional heat-cured acrylic resin, group II received maxillary and mandibular complete dentures made of thermoplastic resin. The retention mean values were measured for mandibular dentures for both groups at time of denture insertion and after 3 months by using the universal testing machine. Assessment of microbial colonization in the two denture base materials was done after 3 and 6 months from denture insertion. The isolation of microorganisms was done with gamma sterilized disposable swabs from tuberosity area under the fitting surface of the maxillary dentures, each swab cultivated in three culture media. The tested microorganisms are *Candida albicans* (*C. albicans*), *Streptococcus mutans* (*S. mutans*), *Staphylococcus aureus* (*S. aureus*) and mixed *Streptococcus* and *Candida*.

**Results:** The thermoplastic resin denture showed statistically significant higher mean of retention value than conventional heat cure denture at insertion and after 3 months (P value <0.000). The microbial colonization results revealed significant difference between group I and group II in the count of *C. albicans*, *S. mutans* and mixed *Streptococcus* after 3 months and 6 months. The thermoplastic resin base (group II) showed decreased microorganism count than group I (heat cured acrylic resin base) while *S. aureus* count showed no statistically significant difference between the two groups.

**Conclusion:** Thermoplastic resin denture bases showed better retention and less microbial colonization than conventional heat cure acrylic resin. Further studies are recommended to evaluate the long-term quality of retention and biological properties of thermoplastic resin complete dentures and its impact on patients' satisfaction.

**KEY WORD:** Denture base material, Retention, and Bacterial colonization

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## INTRODUCTION

Over the centuries, a variety of materials have been used for denture construction. The historic developments of these materials have led it to the times, when the dentures were carved from stone, ivory, bone and wood to the latest polymers<sup>(1)</sup>. The ideal denture base material should possess several key attributes, like biocompatibility, good esthetics, high bond strength with available denture teeth, radio- opacity, ease of repair, and should possess adequate physical and mechanical properties<sup>(2,3)</sup>.

Acrylic resins have been widely used due to their acceptable esthetics, and desirable characteristics such as easy handling, good thermal conductivity, low permeability to oral fluids, and color stability. A major problem of this material can be seen as the dimensional change during processing, frequently due to the polymerization shrinkage<sup>(4)</sup>. Therefore thermoplastic resins were introduced into the market as an alternative to the use of conventional acrylic resins in the construction of complete, and partial removable dentures that exhibited higher dimensional and color stability, excellent esthetics combined with favorable physical properties and easy processing characteristics<sup>(5)</sup>.

The two most commonly used molding techniques for denture base resins are injection molding and compression molding. The injection molding processing method for the denture fabrication leads to less Polymerization shrinkage and produces a more accurate denture than the compression molding process<sup>(6,7)</sup>.

Many studies in the past have been conducted on the properties of denture base material in order to find the best material for denture. Lack of retention and stability of the complete denture, specially mandibular denture, is a common problem among edentulous denture wearers, which may be due to poor ridge relationship, reduced neuromuscular coordination, inadequate quality and quantity of available bone and alveolar mucosa<sup>(8,9)</sup>. Effective retention can be attained by the close mucosal

contact of the denture base that is affected by dimensional stability and accuracy of the denture base<sup>(10)</sup>.

It is also known that, one of the clinical problems associated with acrylic resin denture base is the adhesion of dental plaque formed by oral microorganisms due to the polymethylmethacrylate (PMMA) has high water absorption capacity, surface porosities and materia alba configuration<sup>(11)</sup>. Denture plaque is a source of infection and is responsible for a variety of clinical problems, including abutment caries, denture stomatitis<sup>(12)</sup>. More importantly, the virulent factor released from denture plaque is known to be a significant risk factor for opportunistic infections and aspiration pneumonia especially in elderly people and in individuals who are immunocompromised<sup>(13)</sup>. *Streptococcus mutans* is regarded as a major etiological agent of dental caries<sup>(14)</sup> while *Candida* is a common bacterium that may cause *Candida*-associated denture stomatitis<sup>(15)</sup>. Thus, a restorative material that possesses antibacterial properties and inhibits bacterial growth around the restoration would be desirable. However, other problems must not be overlooked, that are the mechanical properties should not be weakened and the cytotoxicity should not be enhanced by the addition of the antibacterial agents. Therefore, the choice of specific antibacterial agents is important for incorporation into PMMA<sup>(16)</sup>.

This study was conducted to compare between the conventional heat cure acrylic resin denture base and thermoplastic resin base on the retention of mandibular complete denture and bacterial colonization on maxillary denture.

## MATERIAL AND METHODS

After the approval of the local Research Ethics Committee, 20 patients of both genders aged 50 to 60 years old, were selected from the Outpatient Clinic of Prosthodontic Department, Faculty of Dental Medicine for Girls Al-Azhar University. The patients were selected to be free from systemic

and oral diseases, especially those that might affect retention as diabetes mellitus. All the patients had well formed maxillary ridge and mandibular ridge with no undercuts to eliminate its effect on retention, and both ridges covered with firm mucoperiosteum. All patients had Angle's class I and free from any TMJ disorder. All patients were in good oral hygiene. Smoker patients or those receiving any medication such as antifungal, antibiotic and antidepressant drugs in the 6 months preceding the study were excluded. All patients informed about practical steps of this study and signed approval consent. Patients were randomly and equally divided into two treatment groups:

- Group I: Each patient in this group received maxillary and mandibular complete dentures made of conventional heat-cured acrylic resin.
- Group II: Each patient in this group received maxillary and mandibular complete dentures made of thermoplastic resin.

### **Prosthetic procedures.**

For each patient, maxillary and mandibular preliminary impression were made using irreversible hydrocolloid impression material. Zinc oxide eugenol paste was used to make final impression after border molding with green stick compound. Impressions were disinfected, boxed and poured to obtain master casts on which trial acrylic denture bases with wax occlusion rims were constructed. Maxillary cast was mounted on semi adjustable articulator (Hanau model H, Teledyne Buffalo, New York, USA) using maxillary face bow (Hanau engineering company, Inc., Buffalo, New York), centric jaw relation was made using wax wafer technique, the lower cast was mounted on the articulator using the centric jaw relation record taken from the patient at the predetermined vertical dimension of occlusion. A protrusive record was taken to adjust horizontal condylar guidance of the articulator while the lateral condylar guidance was adjusted according to Hanau formula. Cross-linked acrylic resin teeth

were selected according to the patient demands (age, sex, arch dimension, etc.). Setting-up of the artificial teeth cross-linked was carried out. Waxing up was done, and the waxed up denture was checked in the patient mouth.

For group I, the dentures were made from conventional heat cure acrylic resin (Acron Duo, Associated Dental Products Ltd., Kemdent, Purton, Swindon, Wiltshire, UK), in which, flasking, wax elimination, processing, deflasking, finishing and polishing of the dentures were done following the conventional routine method.

For group II, the dentures were made from thermoplastic resin (Vertex ThermoSens Rigid, Vertex-Dental B.V., Zeist, Netherland). The artificial acrylic resin teeth were mechanically attached to this type of denture base material. The mechanical means of retention were prepared in artificial teeth before setting as follows: Circumferential groove in the cervical area around the neck of the anterior and posterior teeth were prepared, and mesio-distal groove was made in the base of both anterior and posterior teeth.

After completing the arrangement of the artificial teeth; the waxed-up dentures were tried in-patient's mouth. Special type of flask (Vertex ThermoFlask, Vertex-Dental B.V., Zeist, Netherland) was used for flasking the waxed-up denture. According to the manufacturer's instruction, during flasking, the injection channels were attached as close as possible to the injection opening of flask. The waxed-up dentures were prepared with the set-up teeth to being finally invested in the flask. The flask was placed in boiling water for wax elimination. Now the mold is ready for injection of thermoplastic resin. The thermoplastic resin was supplied in the form of crystals put in a cartilage.

The thermoplastic dentures were processed with the injection molding technique (Vertex ThermoJect 22, Vertex-Dental B.V., Zeist, Netherland) (figure 1), that is started by melting the thermoplastic resin crystals then injection of the material into the

denture mould of the flask through the sprue channel. Once the injection was completed the system stopped for minutes and the flask was removed from the machine. The denture deflashed, then finished and polished by using thermal resin finishing burs at low speed, pumice, and finally buffing was done to add a very high luster. After that they were ready for insertion.

### Retention test

The retention measurements for mandibular denture were carried-out and recorded at time of the prostheses insertion and three months later by using Nexygen testing machine (Model LRX-plus, Lloyd instruments Ltd, Fareham, UK) which composed of Attachment part, Chin rest and universal testing machine with a computer software package which deliver a vertical dislodging force at a speed of 30mm/min to the denture. The patient was instructed to sit in an upright position and keep his chin firmly seated on a chin support. The bar was rigidly connected to the denture and the attachment part of the universal machine was adjusted. The device was subjected to a slowly increasing vertical load until the denture was totally out of place. The load at dislodgment manifested by an audible sound tuck and confirmed by a sharp drop at load-deflection curve recorded using computer software

and this value was recorded in Newton. The test was repeated five times to obtain 5 records, the mean of which was calculated. (Fig. 2)

### Microbial colonization test

For each patient, the isolation of microorganisms was done with gamma sterilized disposable swabs. Swabs were taken from the tuberosity area of the palate under the fitting surface of the maxillary denture, immediately before the insertion of the denture, after 3 and after 6 months from insertion by vigorous strokes over the mucosa from distance of 1 to 2 cm for 30 second. The patients were informed not to change their usual cleaning habits and not to take out the dentures the 4 h prior to the sampling.

Each swab cultivated in three culture media. Sabouraud agar; these media selective for isolation of fungi, MacConkys agar; which is differential media for isolation of gram negative microorganism and blood agar base No.2; which is an improved blood Agar Base processing enhanced nutritional properties suitable for the cultivation of pathogens and other micro-organism. The tested microorganisms are *Candida albicans* (*C. albicans*), *Streptococcus mutans* (*S. mutans*), *Staphylococcus aureus* (*S. aureus*) and mixed *Streptococcus* and *Candida*.



Fig. (1) The injection molding machine



Fig. (2) Measuring retention using universal testing machine

Sterile tubes were filled with sterile saline. Five sterile tubes were used for each sample. The first tube was filled with 1000µL of sterile saline, and the other four were filled with 900µL Each swab was incubated immediately in the first sterile tube containing 1000µL of sterile saline for 15 minutes, and followed by successive dilutions at 1/10, 1/100, 1/100, and 1/10000

A sample of 100µL of each tube was plated on the agar medium. All the microorganisms were cultured for 24 h at 37 °C. The anaerobic bacteria were cultured in anaerobic containers. The number of colonies on the surface of the agar gels were counted (Colonies forming units per sample) (CFU)/sample) and the number of microorganisms per ml was calculated by multiplying the number of colonies (CFU) by the actual dilution of the cell suspension. The mean difference between the microorganisms counts before insertion of the denture and, after 3 and 6 months was calculated, tabulated and statistically analyzed.

**Statistical analysis**

Data were collected, revised and entered to the Statistical Package for Social Science (SPSS Inc., Chicago, IL, USA) version 20. The data were presented as number, percentages, mean and standard deviations (SD). Comparison between two independent groups with quantitative data and parametric distribution were done by using independent t-test while paired groups were done by using paired t-test. Spearman correlation coefficients were used to assess the relation between two quantitative parameters. The p-value was considered significant at the level of < 0.05.

**RESULTS**

**I. Results of retention**

The effect of type of denture base material on the retention mean values is shown in (table 1, Fig 3).

TABLE (1): Comparison between retention mean values of the group I (conventional heat cure acrylic resin denture) and group II (thermoplastic resin denture)

Time	Group I (10 cases)		Group II (10 cases)		Independent t-test	
	Mean	SD	Mean	SD	t	P-value
At insertion	6.33	1.71	12.35	2.93	5.611	0.000
3 months	7.54	1.67	14.34	2.98	6.295	0.000
Paired t-test	t	2.264	2.130			
	P-value	0.029	0.039			

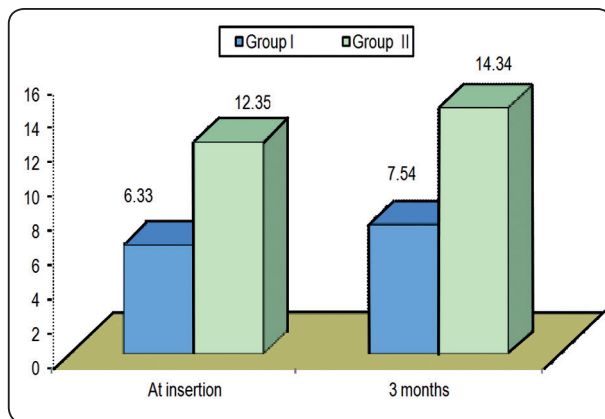


Fig. (3) Bar chart representing comparison between retention mean values of group I (conventional heat cure acrylic resin denture) and group II (thermoplastic resin denture).

The comparison of the retention force mean values of the group I (conventional heat cure acrylic resin denture) and group II (thermoplastic resin denture) revealed a statistically significant difference between them at insertion as well as after three months of insertion. The thermoplastic resin denture showed statistically significant higher mean of retention values than conventional heat cure denture (P value <0.000).

**1. Results of microbial colonization**

The comparison between the mean difference (difference between microbial count after 3 months and 6 months from the microbial count before the insertion of dentures) and standard deviation of organisms count CFU/ml in-group I (conventional heat cure acrylic resin) and II (thermoplastic resin) was represented in table 2 and Fig. 4, 5.

Statistical analysis of the results revealed significant difference between group I and group II in the count of *C. albicans*, *S. mutans* and mixed *Streptococcus* and *Candida* after 3 months and 6 months. The thermoplastic resin base (group II) showed decreased microorganism count than group I (heat cured acrylic resin base) while *S. aureus* count showed no statistically significant difference between the two groups.

TABLE (2): The comparison between the mean difference and standard deviation of microorganisms count CFU/ml x 10<sup>2</sup> in group I and II after 3 and 6 months

Microorganisms	Group I	Group II	Independent t-test	
	no. = 10	no. = 10	t	p-value
<b>After 3 months</b>				
<i>S. aureus</i>	4.4±1.3	3.8±0.98	1.165	0.259
<i>S. mutans</i>	3.5±0.72	2.40±0.93	2.958	0.008
<i>C. albicans</i>	4.2±1.3	2.6±0.73	3.394	0.002
Mix (strept, candida)	4±1.4	2.3±0.52	3.600	0.003
<b>After 6 months</b>				
<i>S. aureus</i>	5.4±1.7	4.0±1.5	1.953	0.066
<i>S. mutans</i>	5.5±1.8	2.75±0.57	4.606	0.000
<i>C. albicans</i>	16.7±35.8	4.2±12.3	10.442	0.000
Mix (strept, candida)	5±1.87	3.5±0.77	2.346	0.031

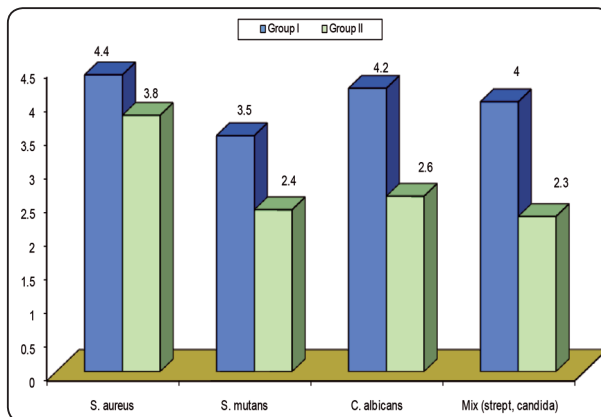


Fig. (4) Bar chart representing comparison the mean difference of microorganisms count CFU/ml x 10<sup>2</sup> in group I and II after 3 months.

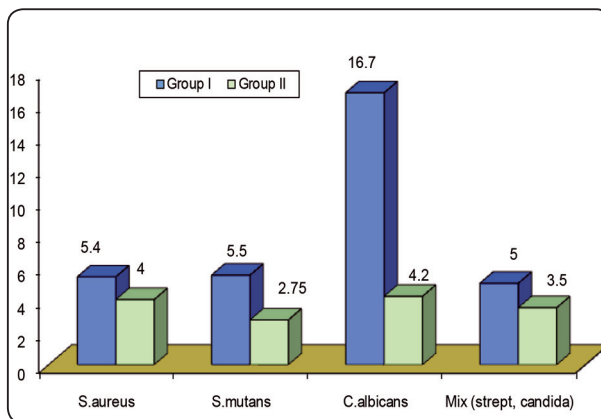


Fig. (5) Bar chart representing comparison the mean difference of microorganisms count CFU/ml x 10<sup>2</sup> in group I and II after 6 months.

**DISCUSSION**

The provision of a retentive and stable denture can be considered the principal goal of general dental practitioners when dealing with complete dentures specially for the mandibular denture. Research findings indicate that retention and stability of complete dentures are strong determinants of patients' satisfaction with newly delivered dentures<sup>(17)</sup>.

In this study, the comparison between retention mean values at insertion and three months of the conventional heat cured acrylic resin and thermoplastic resin mandibular dentures revealed

that, the thermoplastic resin showed statistically significant higher retention mean values, this may be attributed to the dimensional changes and resulting distortion, and due to water sorption of the conventional resin in comparison to thermoplastic resin that did not stop after three month, and the fact that, the physical properties of the thermoplastic resin, which allowed effective engagement with the lingual pouch undercut and close adaptation to the supporting tissues<sup>(18-20)</sup>.

Despite the relatively old age of the thermoplastic denture base materials, reports to evaluate its impact on denture retention are scarce. Antonelli and Hottel<sup>(21)</sup> reported that, the use of thermoplastic resin flanges constructed stable, retentive, well adapted, and comfortable complete dentures bases. Singh et al<sup>(20)</sup> found that thermoplastic denture bases produced better patient satisfaction and comfort, compared to conventional acrylic resin denture bases.

The oral cavity of healthy individuals with or without teeth may be colonized by yeast and bacteria coexisting in a relationship of commensalism<sup>(22)</sup>. Denture wearing and deficient denture hygiene are the predisposing factors for increasing the number of microorganisms in the oral cavity. So, the bacterial colonization increases and becomes more pathogenic, acting as a potential source of infection<sup>(23)</sup>.

In this study, swabs were taken from the upper dentures by rubbing once for a distance of 1 or 2 cm, however, to ensure a more accurate and standardized isolation procedure. The rubbing was done for a specified period of time 30 seconds<sup>(24)</sup>. The swabs were taken from the fitting surface of the denture that is considered an irregular surface as it usually shows micro pits and micro porosities that harbor microorganisms and are difficult to be removed by mechanical methods. The swabs were taken specifically from the tuberosity area of palatal fitting surface of the denture, as they are the most suspected areas for bacterial colonization<sup>(25)</sup>.

The intra-individual variations in the bacterial counts were very important. For that reason it was decided to calculate the mean difference between the microorganisms counts before insertion of the denture and, after 3 and 6 months<sup>(26)</sup>.

The results of this study showed that there was a gradual increase in the CFU values of the total microorganisms count during the time of using the dentures. For these patients this gradual increase may be because the mouth dealt with them as a foreign body and reacting by increasing the CFU of total microorganisms. Although, the increase of colonization of microorganisms during the follow up period might also be due to surface roughness of the materials that should slight increase in the mean values from the beginning of wearing the dentures, as aging process promotes the surface roughness of the denture base material.

In this study, the thermoplastic resin base showed decreased microbial colonization than the conventional heat cured acrylic resin base, this may be attributed to, the biocompatibility of these material as they are proved by many authors to have few or no residual monomer<sup>(27-29)</sup>. Which enabled this material to be a substitute for PMMA especially with patients who are allergic to monomer. This biological advantage might be due to their way of processing that occurs by injection of pre-polymerized resins under temperature and pressure without chemical reaction<sup>(30,31)</sup>.

Rougher surfaces contribute to microbial colonization and biofilm formation. Bacterial and fungal species have more of a propensity to adhere to rough denture base materials. And it was found that, the thermoplastic resins have less surface roughness than PMMA even when they are subjected to the same polishing techniques<sup>(32,33)</sup>.

The acrylic denture fitting surface cannot be mechanically polished and thus presents irregularities and microscopic pores that facilitate bacterial and fungal colonization, the denture may cause trauma to the palatal mucosa, which reduces

tissue resistance to infection and increases mucosa permeability, contributing to the passage of toxins produced by the microorganisms<sup>(34)</sup>

Newton<sup>(35)</sup> showed that although glazing dentures fitting surface did not prevent bacterial colonization, it favored plaque removal while the glaze layer was intact for one month. After three months of denture use, glaze cracks developed and created micro retentive areas that increased plaque accumulation.

After 6 months the most prominent microorganism was *Candida albicans*, specially in heat cured acrylic denture base, this in agreement with Ribeiro et al.<sup>(22)</sup> that found *Candida* (65.5%) more than *Strep. Mutans* and *Staph. Aureus* on dentures. Also, Baena- Monroy et al.<sup>(36)</sup> showed the presence of *Candida albicans* on the internal surface of complete dentures. *Candida albicans* is a well-known etiologic agent at denture stomatitis. This inflammatory disorder affects approximately 60% of denture wearers and causes inflammation of the oral mucosa in close contact with the denture<sup>(37)</sup>.

Denture use can reduce the salivary flow, which interferes with the physiological cleaning function of the tongue and creates a favorable environment for microbial survival<sup>(38)</sup>. In addition to this, *Candida* cells seem to have affinity with denture acrylic<sup>(39)</sup>, favoring the adhesion of epithelial cells to the oral mucosa and the onset of denture stomatitis<sup>(40)</sup>.

## CONCLUSION

Within the limitations of this study it was concluded that, thermoplastic resin denture bases showed better retention and less microbial colonization than conventional heat cure acrylic resin. Further studies are recommended to evaluate the long-term quality of retention and biological properties of thermoplastic resin complete dentures and its impact on patients' satisfaction.

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