INTRODUCTION

Polymethyl methacrylate (PMMA) is the most popular material in dentistry especially as a denture base material since 1937. PMMA has some advantages like ease of manipulation, repair and polish, low cost and acceptable esthetic properties. However, polymerization shrinkage, poor wear resistance, inadequate mechanical properties, and residual monomer content are the main limitations of the material. Polyamide denture base material (nylon) that has been described in 1950, is an alternative to the acrylic resins in special situations such as repeated denture fracture and allergies. Surface roughness and surface free energy play a key role for the bacterial adhesion on to intra-oral hard surfaces and also for following dental plaque accumulation process. It was reported in several studies that rough surfaces are more inclined to bacterial adhesion and plaque accumulation than smooth surfaces. Dental restorations with rough surfaces are also more prone to staining and discoloration which may lead to reduced esthetics and acceptability of the material since 1937. Polymethyl methacrylate (PMMA) (Meliodent; Acron MC) and 1 polyamide (Deflex) denture base materials, coated with a sealant agent (Palaseal) and divided into 4 groups according to overnight cleaning procedures: distilled water (control), 5% sodium hypochlorite (NaOCl) and two different sodium perborate (Corega; Rapident). The surface roughness values were measured with a profilometer before (R_a) and after 90 days immersion in denture cleaners (R_a). Significant differences were found, between the R_a values of 5% NaOCl applied Acron MC, Deflex and also Rapident applied Deflex groups. Denture cleaning procedures had no significant effects on the quantity of Candida albicans.

Keywords: Denture base material, Denture cleaners, Surface sealant agent, Surface roughness, Candida albicans

This study investigated the effect of denture cleansers on the surface roughness and Candida albicans adherence of surface sealant agent coupled denture base materials. One hundred and twenty specimens were fabricated from 2 polymethyl methacrylate (PMMA) (Meliodent; Acron MC) and 1 polyamide (Deflex) denture base materials, coated with a sealant agent (Palaseal) and divided into 4 groups according to overnight cleaning procedures: distilled water (control), 5% sodium hypochlorite (NaOCl) and two different sodium perborate (Corega; Rapident). The surface roughness values were measured with a profilometer before (R_a) and after 90 days immersion in denture cleaners (R_a). Significant differences were found, between the R_a and R_1 values of 5% NaOCl applied Acron MC, Deflex and also Rapident applied Deflex groups (p<0.05). Denture cleaning procedures had no significant effects on the quantity of Candida albicans.

Efficacy of denture cleaners on the surface roughness and Candida albicans adherence of surface sealant agent coupled denture base materials

Ayşegül KÖROĞLU1, Onur ŞAHIN1, Doğu Ömür DEDE2, Şule Tuğba DENIZ3, Nurdan KARACAN SEVER4 and Serkan ÖZKAN5

1 Department of Prosthodontics, Faculty of Dentistry, Bulent Ecevit University, Zonguldak, Turkey
2 Department of Prosthodontics, Faculty of Dentistry, Ordu University, Ordu, Turkey
3 Department of Prosthodontics, Faculty of Dentistry, Birün University, Istanbul, Turkey
4 Department of Microbiology, Faculty of Veterinary Medicine, Ankara University, Ankara, Turkey
5 Department of Orthodontics, Faculty of Dentistry, Ordu University, Ordu, Turkey

Corresponding author, Ayşegül KÖROĞLU; E-mail: aysegulozturk151@yahoo.com

Color figures can be viewed in the online issue, which is available at J-STAGE.
of sealant agent coupled denture base materials. The null hypothesis of this study was that the application of different denture cleaners or the type of denture base materials would not affect the surface roughness and Candida concentration values.

MATERIALS AND METHODS

Two PMMA and one polyamide denture base materials, three denture cleansers and also one surface sealant agent were used in this study (Table 1). Forty disk-shaped (10×2 mm) specimens were prepared for each denture base material according to the manufacturers’ instructions. Specimens were prepared as follows: (i) Meliodent-20 min/100°C in boiling water bath; (ii) Acron MC-3 min/500 W by microwave energy; (iii) Deflex-15 min/280°C by micro injection molding system. Before the polishing procedures, all specimens were ground-finished (100 revs/min, during 15 s) with 400 grit silicon carbide abrasive paper on a sanding machine (Phoenix Beta, Buehler) under water cooling. Then the specimens were coated with Palaseal surface sealant agent with a soft brush in an even thin layer in one direction avoiding air bubble formation. Twenty seconds later, the Palaseal coated specimens were polymerized for 90 s in a light polymerizing unit (Dentacolor XS, HeraeusKulzer). All specimens were ultrasonically cleaned in distilled water (Hygosonic, Dürr Dental) for 10 min, rinsed and dried with oil-free air. The specimens of each base material group were then randomly divided into 4 subgroups (n=10) through the types of denture cleansers.

The surface roughness (Rₐ) of each specimen was measured by using a contact profilometer (Perthometer M2, Mahr) before immersion in denture cleaners and recorded as Rₐ₀. Profilometer's resolution was 0.01 µm, the interval (cut-off length) was 0.8 mm, transverse length was 5.5 mm and stylus speed was 1 mm/s. For each specimen 3 measurements were performed and the arithmetic mean values were used for the statistical analysis.

The denture base materials were then subjected to cleaning procedures. The specimens of control groups were immersed in a container with 200 mL of distilled water at room temperature. The test specimens were immersed in a container; with 200 mL of 5% sodium hypochlorite (NaOCl) solution; 200 mL distilled water with one Corega tablet; 200 mL distilled water with one Rapident tablet. Immersions were made to simulate daily hygiene routine for 3 months (90 days). Therefore to simulate nocturnal immersion (overnight) each 24 h corresponded to 3 immersions of 8 h per day. The cleaning solutions were changed three times a day. Each specimen was washed for 5 s and dried before immersed in a new solution. Then the surface roughness measurements were repeated for each specimen and recorded as Rₐ₁.

Prior to Candida albicans contamination and adhesion assay, each specimen was ultrasonically cleaned in distilled water (Hygosonic, Dürr Dental) for 10 min, rinsed and dried with oil-free air. Candida albicans (ATCC 90128) strain was cultured aerobically at 37 ºC for 24 h on Sabouraud dextrose agar (SDA) (Oxoid). After 24 h of culture, Candida albicans strain was centrifuged (ThermoScientific SL8, Germany) at 6,000 rpm for 15 min and resultant cell pellets were washed three times with phosphate-buffered saline solution (PBS, pH 7.4) before being resuspended in PBS and adjusting to a final concentration of 0.5 MacFarland solution (1×10⁶ cfu/mL). Each test specimen was

<table>
<thead>
<tr>
<th>Product</th>
<th>Code</th>
<th>Component</th>
<th>Manufacturer</th>
<th>Lot Number</th>
</tr>
</thead>
<tbody>
<tr>
<td>Meliodent</td>
<td>HC</td>
<td>Conventional heat cure polymethyl methacrylate</td>
<td>Bayer UK</td>
<td>012373 12JUN170</td>
</tr>
<tr>
<td>Acron MC</td>
<td>MC</td>
<td>Microwave cure polymethyl methacrylate</td>
<td>GC</td>
<td>1212141 1112142</td>
</tr>
<tr>
<td>Deflex</td>
<td>TP</td>
<td>Polyamide</td>
<td>Nuxen</td>
<td>10616 CL 29</td>
</tr>
<tr>
<td>Palaseal</td>
<td>—</td>
<td>Methyl methacrylate, tris(2-hydroxyethyl)-isocyanurate-triacrylate, acrylatedepoxyoligomer, acrylates, acrylatedpolysiloxane</td>
<td>Heraeus-Kulzer</td>
<td>531</td>
</tr>
<tr>
<td>5% NaOCl</td>
<td>—</td>
<td>Sodium hypochlorite</td>
<td>Prime Dental</td>
<td>0507201-7</td>
</tr>
<tr>
<td>Corega</td>
<td>—</td>
<td>Potassium monopersulfate, sodium bicarbonate, sodium lauryl sulfoacetate, sodium perborate monohydrate, sodium polyphosphate</td>
<td>GlaxoSmithKline</td>
<td>HM2N</td>
</tr>
<tr>
<td>Rapident</td>
<td>—</td>
<td>Potassium persulfate, sodium carbonate, anhydrous citric acid, Sodium perborate monohydrate, sodium percarbonate copolymer, sodium lauryl sulfate, magnesium stearate, mint-apple flavor, sage-peppermint extract</td>
<td>L'Angelica</td>
<td>214507056</td>
</tr>
</tbody>
</table>
inoculated using 1 mL of *Candida albicans* suspension and incubated for 24 h at 37 °C in a horizontal shaker. After the adhesion phase, the nonadherent cells were removed from the specimen by washing three times with PBS. Subsequently specimens were transferred into 1 mL of PBS and vortexed for 60 s to resuspend all present microorganisms. 0.1 mL of this resuspensions were plated on Sabouraud dextrose agar and incubated aerobically in a horizontal shaker at 37°C for 48 h. After the incubation period, colonies were counted by the eye counting method and the logarithm of colony forming units (CFU) per millimeter (log cfu/mm) was then calculated\(^{18}\).

Data were statistically analyzed (SPSS 20.0 V, SPSS, Chicago, IL, USA). Firstly, a normal distribution was obtained for variables by Levene's test of homogeneity. Then the Ra and CFU results were separately analyzed by 2-way ANOVA test to evaluate the effects type of denture base material, cleaning procedure and their interactions. The mean Ra and CFU values were compared by Tukey's HSD multiple comparison test. The pairwise comparisons of Ra_0 and Ra_1 values were also compared by Independent Sample \(t\) test. Significance was evaluated at \(p<0.05\) for all tests.

**RESULTS**

According to the 2-way ANOVA results, while the type of denture base material and cleaning procedure were significant on CFU, these variables and their interactions also significant on Ra values (\(p<0.05\)) (Table 2). The mean Ra_0 and Ra_1 values and standard

**Table 2** Two-way ANOVA results for comparison of surface roughness (Ra) and *Candida* concentration values (cfu/mm)

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Source</th>
<th>SS</th>
<th>df</th>
<th>MS</th>
<th>F</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ra</td>
<td>Denture base material (A)</td>
<td>197,841,781.667</td>
<td>2</td>
<td>98,920,890.833</td>
<td>34.387</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td>Cleaning procedure (B)</td>
<td>36,447,976.667</td>
<td>3</td>
<td>12,149,325.556</td>
<td>4.223</td>
<td>0.007</td>
</tr>
<tr>
<td></td>
<td>A×B</td>
<td>107,631,518.333</td>
<td>6</td>
<td>17,938,586.389</td>
<td>6.236</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Error</td>
<td>Total</td>
<td>310,684,520.000</td>
<td>108</td>
<td>2,876,708.519</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>0.115</td>
<td>2</td>
<td>0.057</td>
<td>7.864</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>0.153</td>
<td>3</td>
<td>0.051</td>
<td>6.982</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>cfu/mm</td>
<td>A×B</td>
<td>0.040</td>
<td>6</td>
<td>0.007</td>
<td>0.917</td>
<td>0.486</td>
</tr>
<tr>
<td>Error</td>
<td>Total</td>
<td>0.787</td>
<td>108</td>
<td>0.007</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td></td>
<td>2.333</td>
<td>120</td>
<td>—</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
</tbody>
</table>

\(^*p<0.05\) indicates significant difference.

**Table 3** Mean surface roughness values (µm) and standard deviations at time 0 (Ra_0) and 90 days (Ra_1) for all groups

<table>
<thead>
<tr>
<th>Denture Base Materials</th>
<th>Cleaning Procedure</th>
<th>Ra_0</th>
<th>Ra_1</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean</td>
<td>SD</td>
<td>Sig</td>
</tr>
<tr>
<td>HC</td>
<td>Cont</td>
<td>0.045</td>
<td>0.015</td>
</tr>
<tr>
<td></td>
<td>NaOCl</td>
<td>0.112</td>
<td>0.167</td>
</tr>
<tr>
<td></td>
<td>Corega</td>
<td>0.065</td>
<td>0.024</td>
</tr>
<tr>
<td></td>
<td>Rapident</td>
<td>0.043</td>
<td>0.022</td>
</tr>
<tr>
<td>MC</td>
<td>Cont</td>
<td>0.055</td>
<td>0.040</td>
</tr>
<tr>
<td></td>
<td>NaOCl</td>
<td>0.044</td>
<td>0.016</td>
</tr>
<tr>
<td></td>
<td>Corega</td>
<td>0.067</td>
<td>0.018</td>
</tr>
<tr>
<td></td>
<td>Rapident</td>
<td>0.079</td>
<td>0.047</td>
</tr>
<tr>
<td>TP</td>
<td>Cont</td>
<td>0.074</td>
<td>0.022</td>
</tr>
<tr>
<td></td>
<td>NaOCl</td>
<td>0.084</td>
<td>0.027</td>
</tr>
<tr>
<td></td>
<td>Corega</td>
<td>0.063</td>
<td>0.027</td>
</tr>
<tr>
<td></td>
<td>Rapident</td>
<td>0.066</td>
<td>0.053</td>
</tr>
</tbody>
</table>

\(^*The multiple comparisons of denture base material/cleaning procedure combination groups by Tukey HSD test were shown as small letters and pairwise comparisons (Independent Sample \(t\) test) among sinter groups shown as capital letters.**

\(^*The values having same letters are not significantly different (\(p>0.05\).\)

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\(812\) *Dent Mater J* 2016; 35(5): 810–816
deviations (SD) of denture base material/cleaning procedure combination groups are shown in Table 3.

The $R_a$ values (0.041 to 0.197 $\mu$m) of all test groups were smaller than plaque accumulation threshold level (0.20 $\mu$m). The Tukey’s multiple comparisons showed that there was no significant difference between the $R_a$ values of test groups ($p>0.05$). When the same cleaning procedure applied denture base resins compared, significant differences was obtained between the HC and TP resin specimens that immersed in Rapident.

![Fig. 1](image1.png)

Fig. 1 Mean CFU values (±SD) of test groups. The multiple comparisons of denture base material/cleaning procedure combination groups by Tukey HSD test were shown as small letters.

![Fig. 2](image2.png)

Fig. 2 *Candida albicans* colonies of denture cleansers applied HC, MC and TP denture base materials.
The initial surface roughness values (Ra0) of all tested results, no significant difference was found between physical properties of dentures with smoother surface, sealant agent coupling (Palaseal) may improve the applied groups, which has incubated with base material groups (>0.05) (Fig. 2). Between different cleansing procedures applied denture p<0.05). However there was no significant difference in Fig. 1. Statistically significant difference was observed -test. paired sample t-test. =0.04) resin groups and applied MC (p=0.044) and TP (p=0.04) resin groups and the other test groups (p<0.05). However there was no significant difference between different cleansing procedures applied denture base material groups (p>0.05) (Fig. 2).

DISCUSSION

The null hypothesis of this study was rejected. The type of denture base material and cleaning procedure were both significant on the surface roughness and Candida forming unit values.

The surface irregularities on denture base materials may act as a reservoir of infection and increase the possibility of hosting microorganisms even after the cleaning of dentures19. Rough surfaces make easier the penetration of bacterial and fungal cells on the denture base resins20. Coating the surface of the denture base material with sealant agents, which are recommended to improve surface smoothness of restorations through filling the micro-fissures and micro-defects, is an alternative application compared to conventional polishing with pumice and polishing pastes. However, surface sealant agents have some limitations, such as; low abrasion resistance, weak retention to the underlying material, exhibiting poor surface quality resulting from failure of spread that depends on probable high viscosity12,21,22. Palaseal surface sealant agent selected for this study is a light-cured sealer that adhere to all denture materials and has high resistance to solvents23. Sesma et al.24 reported that sealant agent coupling (Palaseal) may improve the physical properties of dentures with smoother surface, decrease plaque adhesion and bacterial colonization, which results in good oral hygiene. Consistent with this results, no significant difference was found between the initial surface roughness values (Ra0) of all tested denture base materials in present study, which had been coated with Palaseal sealant agent before. In addition, all of the Ra0 values were lower than the threshold level of 0.2 µm, indicated by Bollen et al.25.

Denture care is an important process that conduces to oral health and denture longevity16,17. Unclean dentures and poor oral hygiene are usually predisposing factors for Candida associated denture stomatitis18. Denture cleaning methods with soaking the dentures in a solution of some chemical agents, have advantages of efficient disinfection and easy usage when compared to the mechanical methods15,25. However it is an important criterion that the cleaners have no adverse effect on the physical and mechanical properties of denture base materials and artificial teeth while effective on removing the organic and inorganic deposits; bactericidal and fungicidal26,27. In the present study, distilled water was determined as control since it is indicated for complete denture overnight immersions17,29. NaOCl solution that also used in the present study in the concentration of 5%, dissolves mucin, organic substances and shows antimicrobial activity by the action of hydroxyl ions and chloramination20,29. Several studies have reported satisfactory results for the antimicrobial activity of NaOCl solutions with different concentrations on denture base materials30-32. However, NaOCl has some disadvantages on denture base materials such as discoloration and an increase in surface roughness, depending on the concentration and immersion time17,33-35. It was also reported that NaOCl solutions may cause structural changes in the polymer matrix of acrylic resins. Thus consequently induce the deterioration on the surface layer and increase the roughness16,37. Consistent with this results, using 5% NaOCl denture cleaner was caused a significant increase on the surface roughness of the MC and TP denture base materials in the present study. However, 5% NaOCl also had no signficant effect on the surface roughness of HC resin group disagreeing with the study results of Ma et al.38.

Alkaline peroxide effervescent denture cleansers may also be used for denture overnight immersions15,17. When an effervescent tablet dissolved in water, alkaline peroxide solution forms by the decomposition of sodium perborate. Oxygen bubbles that released from the solution enable the mechanical cleaning as well as chemical cleaning17,29. In the present study, while one of the tested sodium perborate (Corega) denture cleaner had no significant effect the surface roughness of base materials, the other one (Rapident) was significantly effected the surface roughness of polyamide denture base material (p<0.05). Although both cleaning materials’ active ingredient was sodium perborate, this result may be caused by the difference in the composition of the cleansers. The result for conventional heat-polymerized acrylic resin is in agreement with the study findings of Peracini et al.39 and Paranhos et al.17.

Immersion times of the cleaning solutions that used in this study vary in different studies15,33,40. In the current study, the cleaning procedure simulated the nocturnal immersion (overnight) for 90 days. Dentists usually advise patients the overnight immersion, to remain their oral tissues in relax and to clean the dentures during sleeping period17.

Candida albicans has an ability to form biofilms on denture base materials and to cause denture-induced stomatitis41,42. This study evaluated the effectiveness of three denture cleaning agents on the surface characteristic and also adhesion of Candida albicans. Compared with HC and MC denture base material groups, the adherence and also quantity of Candida albicans was significantly higher on the TP denture base material groups. Several studies reported that the surface roughness can affect the microbial colonization of dentures18,43. In accordance with this results, surface
roughness values of TP resin groups were higher but not significant than other base material groups. In the current study, this situation may be responsible from the higher adherence ratios of TP denture base material groups. It was also resulted from present study that, denture cleaning procedures had no significant effect on the quantity of Candida albicans. This results may be depend on the fact that the surface roughness of all resin groups before and also after cleaning procedures were below the plaque accumulation threshold level of 0.2 μm².

This study has some limitations. The specimens were polished by only one surface sealant agent. Under the same experimental conditions by using different sealant agent applications, denture cleaners and microorganisms, long term in vivo and in vitro investigations should be made.

CONCLUSIONS

It was concluded from the results of the present study is that the sealant agent coupling may provide sufficient surface texture for all tested denture base materials. While the sodium perborate (Rapident) denture cleaner may be destructive for sealant agent coupled TP base material, the 5% NaOCl solution was for both MC and TP base materials. On the other hand, none of the denture cleaning procedures could significantly affect the quantity of Candida albicans on denture base materials.

ACKNOWLEDGMENTS

The authors report no conflict of interest. The authors alone are responsible for the content and writing of the paper.

REFERENCES


