journal of the mechanical behavior of biomedical materials 134 (2022) 105390

Contents lists available at ScienceDirect

Journal of the Mechanical Behavior of Biomedical Materials

journal homepage: www.elsevier.com/locate/jmbbm

Effect of simulated brushing and disinfection on the surface roughness and color stability of CAD-CAM denture base materials

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A R T I C L E I N F O

Keywords:
CAD-CAM
Color stability
Denture base
Disinfection
Simulated brushing
Surface roughness

A B S T R A C T

Purpose: To evaluate the effect of simulated brushing and chemical disinfection on the surface roughness and color stability of CAD-CAM denture base materials and to compare with those of a heat-cured denture base material.

Material and methods: Disk-shaped specimens (Ø 10mm × 2 mm) were prepared from 3 CAD-CAM denture base resins (AvaDent, Ava; Merz M-PM, Merz; Polident d.o.o, Poli) and a heat-cured polymethylmethacrylate resin (Promolux, Conv) (n = 30). After polishing, baseline surface roughness (R_a) and color coordinates were measured. The measurements were repeated after 20000 cycles of simulated brushing, and the specimens were divided into 3 groups according to disinfection protocol (distilled water, 1% sodium hypochlorite (NaOCl), and effervescent tablet) (n = 10). After 9 cleaning cycles over a period of 20 days, R_a and color coordinates were remeasured. Color differences (ΔE_{00}) were calculated by using CIEDE2000 formula. One-way analysis of variance (ANOVA) and pairwise t-tests were used to analyze R_a and ΔE_{00} data, while repeated measures ANOVA test was used to compare baseline, after brush, and after disinfection R_a values ($\alpha = 0.05$).

Results: Brushing did not affect the R_a values of tested materials ($P \geq .08$). Both before and after brushing, Merz and Conv had higher R_a values than Poli and Ava ($P < .001$). Among disinfectants, effervescent tablet led to the lowest R_a for Merz ($P = .003$) and the highest R_a for Poli ($P \leq .039$). Only NaOCl resulted in significant differences among the R_a of materials ($P < .001$), as Merz and Conv had higher R_a values than Poli ($P \leq .002$). Repeated measures ANOVA revealed that effervescent tablet disinfection of Merz led to lower R_a values than those of baseline and after brushing ($P \leq .042$). After brushing, Ava and Conv had higher ΔE_{00} values compared with Merz and Poli ($P \leq .015$). When compared with other disinfection protocols, effervescent tablet led to higher ΔE_{00} values for Merz and Poli ($P < .001$). Significant differences were observed among materials when NaOCl was used; Conv had higher ΔE_{00} values than Ava and Merz ($P = .004$).

Conclusions: Brushing did not increase the surface roughness of materials. Disinfection protocol's effect on the surface roughness varied. The effect of brushing on the color of materials varied; color of Ava and Conv was affected from brushing more than the other materials. Color stability of materials varied depending on the disinfection protocol. Effervescent tablet caused higher color change with Merz and Poli compared with other disinfectants**.** NaOCl led to small color change for Poli, Ava, and Merz materials.

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https://doi.org/10.1016/j.jmbbm.2022.105390
Received 14 June 2022; Received in revised form 13 July 2022; Accepted 17 July 2022
Available online 21 July 2022

1. Introduction

Various types of materials are available for the fabrication of a complete denture, yet polymethylmethacrylate (PMMA) is still preferred due to its easy manipulation and repair, polishability, biocompatibility, and low cost (Al-Dwairi et al., 2019; Alp et al., 2019; Atalay et al., 2022; Kul et al., 2021; Kurt et al., 2018; Porwal et al., 2017). Computer aided design-computer aided manufacturing (CAD-CAM) technologies have enabled the fabrication of complete dentures by using prepolymerized PMMA discs (Al-Qarni et al., 2020). These materials display improved physical properties, no polymerization shrinkage, and tissue adaptation compared with the heat-cured PMMA (Arslan et al., 2018; Raszewski, 2020).

Regardless of the manufacturing technique or the type of the PMMA used, denture cleansing is essential considering the microporosities and surface irregularities on a denture's surface. Inadequate oral hygiene may lead to biofilm adherence and subsequent denture stomatitis (Masetti et al., 2018). It is possible to eliminate the biofilm chemically, mechanically, or by a combination of these methods (Costa et al., 2021). Among denture hygiene methods, brushing is widely preferred, being an inexpensive and simple method (Sorgini et al., 2015). However, combining brushing with immersion in chemical solutions is recommended to increase the effectiveness of cleansing (Tsutsumi-Arai et al., 2022). These chemical solutions have antimicrobial properties (Costa et al., 2021) and various disinfectants, including sodium-hypochlorite (NaOCl) and alkaline peroxides, are currently available (Masetti et al., 2018; Polychronakis et al., 2018). However, chemical methods have shortcomings including bleaching of the denture, corrosion, increased surface irregularities, unpleasant taste, and oral tissue reactions (Costa et al., 2021; Jaiswal et al., 2018; Polychronakis et al., 2018). Considering that a previous study has reported 0.2 μm as the clinically acceptable threshold for surface roughness (Alp et al., 2019), disinfectants may impair the longevity of a complete denture due to increased roughness.

Previous studies have evaluated the effect of brushing (Alfouzan et al., 2021c, 2022; Chang et al., 2021; de Freitas Pontes et al., 2016; Shinawi, 2017; Sorgini et al., 2012) and chemical disinfection (Arruda et al., 2015; Fernandes et al., 2013; Goiato et al., 2013; Kurt et al., 2018; Lira et al., 2012; Ozyilmaz and Akin, 2019; Paranhos Hde et al., 2013; Porwal et al., 2017; Rocha et al., 2021) on the surface roughness and color denture base materials. However, number of studies on surface roughness (Alfouzan et al., 2021a) and color stability (Alfouzan et al., 2021b) of CAD-CAM denture base materials after when brushing and disinfection are combined is limited. In addition, those studies (Alfouzan et al., 2021a, 2021b) were limited to 1 year of simulated tooth brushing, which is a relatively short period of time considering the clinical use of a complete denture. Thus, the present study aimed to investigate the effect of long-term brushing and subsequent disinfection on the surface roughness and color change of 4 different PMMA denture base materials (3 CAD-CAM PMMA and 1 conventional heat-cured PMMA). The null hypotheses were that i) material type would not affect the surface roughness at any time interval, ii) brushing would not affect the surface roughness of tested PMMA resins, iii) disinfection would not affect the surface roughness of tested PMMA resins, iv) disinfection protocol would not affect the surface roughness within each type of PMMA, v) material type would not affect the color change at any time interval, vi) brushing would not affect the color change of different PMMAs, and vii) disinfection protocol would not affect the color change within each type of PMMA tested.

2. Materials and method

2.1. Specimen preparation

Number of specimens in each group was based on previous studies, which have reported significant differences (Alfouzan et al., 2021a, 2021b). One hundred and twenty disk-shaped specimens (Ø 10mm × 2 mm) were fabricated from 4 denture base materials (3 different CAD-CAM PMMAs; Merz M-PM, Merz Dental GmbH, Lütjenburg, Germany (Merz); AvaDent, Global Dental Science, Tilburg, Netherlands (Ava); Polident d.o.o, Polident, Volčja Draga, Slovenia (Poli), and a heat-cured PMMA resin; Promolux, Merz Dental GmbH, Lütjenburg, Germany (Conv)) ($n = 30$). A cylinder (Ø = 10 mm) was designed in standard tessellation language (STL) format on a software (Meshmixer v3.5.474, Autodesk Inc, San Rafael, CA, USA) for the fabrication of CAD-CAM PMMA resin specimens. This STL file was used to mill (Wieland Zenotec mini, V6.12.04, Wieland Dental + Technik GmbH & Co. KG, Pforzheim, Germany) cylinder-shaped specimens. A precision cutter (Vari/cut VC-50, Leco Corporation, St Josephs, MI, USA) was used to wet-slice PMMA resin cylinders into 2 mm-thick specimens. To fabricate conventional heat-cured PMMA resin specimens, wax patterns (Ø 10 mm × 2 mm) were prepared and further heat processed at 74 °C for 8 h by using the flask-press-pack technique (Gruber et al., 2021). After deflasking, all specimens were trimmed and further smoothened with 600-grit silicon carbide abrasive papers under running water. Final dimensions of the specimens were measured with a digital caliper (Model number NB60, Mitutoyo American Corporation, Providence, RI, USA) to ensure a uniform thickness of 2 ± 0.03 mm (Alp et al., 2019).

All specimens were conventionally polished using a slurry of coarse pumice in water (Pumice fine, Benco Dental, Pittston, PA) for 90 s at 1500 rpm and a polishing paste (Fabulustre, Grobet USA, Carlstadt, NJ, USA) was applied for an additional 90 s for fine polishing (Sahin et al., 2016). Prior to roughness and color measurements, all specimens were cleaned in distilled water for 10 min (Eltrosonic Ultracleaner 07–08, Eltrosonic GmbH, Wiesbaden, Germany) and dried with paper towels.

2.2. Surface roughness analysis

Surface roughness (R_a) of each specimen was measured by using a non-contact optical profilometer equipped with an H0 sensor (FRT MicroProf 100, Fries Research & Technology GmbH, Bergisch Gladbach, Germany) (Livas et al., 2020). The parameters of the profilometer were 5.5 mm of tracing length, 0.8 mm of cut-off Lc value, 1 μm of resolution, and 1000/mm of pixel density (Livas et al., 2020). For each specimen, a total of 6 linear traces (3 vertical and 3 horizontal) that were 1 mm apart from each other were measured and averaged by using a software (Mark III, Fries Research & Technology GmbH, Bergisch Gladbach, Germany) (Livas et al., 2020).

2.3. Color coordinate measurements

A digital spectrophotometer (CM-26d, Konica Minolta, Tokyo, Japan) was used to measure the color parameters (L*, a*, b*) defined by the Commission International de l'Eclairage (CIE). This spectrophotometer uses CIE Standard (2°) human observer characteristics in its color estimations and has an 8-mm aperture. Color measurements were performed on a gray backing and saturated sucrose solution was used for the optical contact between the specimens and the backing. A single operator (M.S.P.) performed all measurements in a humidity and temperature-controlled room with day light. The spectrophotometer was calibrated before each measurement. Three measurements were made for each specimen and these values were averaged. The CIEDE2000 formula was used to calculate the color differences (ΔE_{00}) among materials. The parametric factors of K_L, K_C, and K_H were set to 1 (Alp et al., 2019).

2.4. Simulated brushing and disinfection

Following baseline measurements, all specimens were subjected to artificial brushing for 20000 cycles (40000 strokes), replicating approximately 4 years of denture cleaning (Chang et al., 2021). Each brushing cycle consisted of 2 strokes (back and forth linear movement) with 1,5 Hz frequency. Brushing machine was programmed to apply a vertical load of 200 g during the movement of the brushes. Six FDA-certified tooth brushes were mounted to an automatic brushing machine (Bürstmaschine linear LR1; Syndicad Engineering, Munich, Deutschland) (Flury et al., 2017) with their bristles facing directly the specimen surface. A soap slurry consisting of 3 parts of distilled water and 1 part of ground alkali-free soap (Sibonet pH 6.5, Burnus GmbH, Darmstadt, Germany) by weight (Viana Í et al., 2021) was used as the cleaning medium. Brushes and soap slurry were renewed after each 10000 cycles (Sulaiman et al., 2020). Specimens were removed from their holders, rinsed, and air-dried after brushing was completed.

Before disinfection, specimens were randomly divided into 3 subgroups (Excel; Microsoft Corp, Seattle, WA, USA) according to disinfection protocol applied, which were 1% NaOCl, denture cleanser alkaline peroxide effervescent tablet (Goiato et al., 2013), and distilled water (control) ($n = 10$). In 1% NaOCl groups, specimens were immersed in a 200-mL beaker that was filled with 1% NaOCl solution for 10 min (Goiato et al., 2013). In effervescent tablet groups, an effervescent tablet (Corega tablet, Block Drug Company, Jersey City, NJ, USA) was dissolved in a 200-mL beaker filled with 40 °C tap water and specimens were immersed for 15 min as recommended by the manufacturer. Specimens of the control group were stored in distilled water 37 ± 2 °C. Beakers were used to ensure that specimens were in complete contact with the disinfectant solutions (Atalay et al., 2022).

To simulate 180 days of denture cleaning, 9 cleaning cycles were completed over a period of 20 days (Atalay et al., 2022). Following each cycle, specimens were removed from the beakers and rinsed for 30 s under running tap water (Goiato et al., 2013). Each cleaning cycle was performed by using a freshly prepared solution and the cleaning process was repeated. Specimens of 1% NaOCl and effervescent tablet groups were stored in distilled water at 37 ± 2 °C in between cycles, similar to those of the control group (Goiato et al., 2013). After all disinfection cycles were completed, specimens were ultrasonically cleaned for 15 min and air-dried. R_a and ΔE_{00} measurements were repeated after brushing and disinfection protocols.

2.5. Statistical analysis

One-way analysis of variance (ANOVA) and pairwise t-tests were used to analyze baseline and after brush R_a values ($n = 30$), to compare R_a within materials depending on disinfection protocol, and amongst materials when the same disinfection protocol was used ($n = 10$). Each material's baseline, after brush, and after disinfection R_a values were compared by using repeated measures ANOVA. One-way ANOVA and pairwise t-tests were used to analyze after brush ΔE_{00} data ($n = 30$), to compare ΔE_{00} within materials depending on disinfection protocol, and amongst materials when the same disinfection protocol was used ($n = 10$). All analyses were performed by using a software (SPSS v25.0, IBM, Armonk, NY, USA) at a significance of $\alpha = 0.05$. ΔE_{00} values were further evaluated in terms of perceptibility and acceptability by using previously described 50% perceptibility (1.72 units) and 50% acceptability (4.08 units) thresholds for acrylic resin denture base materials (Ren et al., 2015).

3. Results

3.1. Surface roughness results

Descriptive statistics of baseline and after brushing R_a values for each material are listed in Table 1. Baseline ($P = .89$) and after brush ($P = .21$) R_a of Merz and Conv was similar, and was greater than the R_a of other materials ($P < .001$). However, the difference between Ava and Poli was nonsignificant before ($P = .51$) and after ($P = .2$) brushing. Brushing did not increase the R_a of the materials ($P \geq .08$).

Within the materials tested, after brushing, disinfection protocol only affected the R_a of Merz ($P = .001$) and Poli ($P = .015$). For Merz, effervescent tablet resulted in the lowest R_a ($P = .003$). The difference between distilled water and NaOCl was nonsignificant ($P = .94$). For Poli, effervescent tablet resulted in the highest R_a ($P \leq .039$), whereas the difference between distilled water and NaOCl was nonsignificant ($P = .67$). Among the materials, even though the ANOVA results ($P = .04$ for distilled water, $P < .001$ for NaOCl, and $P = .027$ for effervescent tablet) showed that disinfection protocols affected the R_a values, pairwise comparisons revealed significant differences only when NaOCl was used. Merz and Conv showed similar Ra values ($P = .46$) that were higher than Poli ($P \leq .002$). Ava showed similar Ra to the other materials ($P \geq .09$) (Table 2).

Repeated measures ANOVA results have shown significant differences among baseline, after brushing, and after disinfection R_a values of Merz when effervescent tablet was used ($P = .002$), and of Ava for every disinfection protocol ($P \leq .041$) (Table 3). For Merz, baseline and after

Table 1
Change in surface roughness (mean ± standard deviation) within materials based on brushing and disinfection protocol. Total surface roughness values were used to compare surface roughness of materials at baseline and after brushing.

Material	n	Baseline	After brushing	After Disinfection	Disinfection Protocol
Ava	10	0.19 ± 0.07^{a}	0.24 ± 0.09^{a}	0.41 ± 0.28^{a}	Distilled water
	10	0.19 ± 0.08^{a}	0.23 ± 0.08^{b}	0.29 ± 0.11^{c}	NaOCl
	10	0.19 ± 0.04^{a}	0.18 ± 0.04^{a}	0.21 ± 0.07^{a}	Tablet
Total	30	0.19 ± 0.06^{A}	0.22 ± 0.08^{A}		
Merz	10	0.41 ± 0.07^{a}	0.33 ± 0.09^{a}	0.39 ± 0.12^{a}	Distilled water
	10	0.38 ± 0.09^{a}	0.40 ± 0.11^{a}	0.40 ± 0.1^{a}	NaOCl
	10	0.34 ± 0.1^{b}	0.38 ± 0.1^{b}	0.21 ± 0.11^{a}	Tablet
Total	30	0.38 ± 0.09^{B}	0.37 ± 0.1^{B}		
Poli	10	0.23 ± 0.09^{a}	0.18 ± 0.04^{a}	0.21 ± 0.08^{a}	Distilled water
	10	0.22 ± 0.12^{a}	0.19 ± 0.09^{a}	0.17 ± 0.07^{a}	NaOCl
	10	0.16 ± 0.04^{a}	0.21 ± 0.12^{a}	0.40 ± 0.28^{a}	Tablet
Total	30	0.20 ± 0.09^{A}	0.19 ± 0.09^{A}		
Conv	10	0.38 ± 0.11^{a}	0.36 ± 0.09^{a}	0.38 ± 0.11^{a}	Distilled water
	10	0.36 ± 0.09^{a}	0.34 ± 0.09^{a}	0.36 ± 0.14^{a}	NaOCl
	10	0.39 ± 0.07^{a}	0.33 ± 0.09^{a}	0.41 ± 0.22^{a}	Tablet
Total	30	0.37 ± 0.09^{B}	0.34 ± 0.09^{B}		

*Different superscript lowercase letters indicate significate differences in rows, while uppercase letters indicate significant differences in columns ($P < .05$).

Table 2
Mean surface roughness values and standard deviations for materials after disinfection ($n = 10$).

Disinfection Protocol	Material			
	Ava	Merz	Poli	Conv
Distilled water	0.41 ± 0.28^{aA}	0.39 ± 0.12^{bA}	0.21 ± 0.08^{aA}	0.38 ± 0.11^{aA}
NaOCl	0.29 ± 0.11^{aAB}	0.40 ± 0.10^{bB}	0.17 ± 0.07^{aA}	0.36 ± 0.14^{aB}
Tablet	0.21 ± 0.07^{aA}	0.21 ± 0.11^{aA}	0.40 ± 0.28^{bA}	0.41 ± 0.22^{aA}

*Different superscript letters represent significant differences (Lowercase letters for columns and uppercase letters for rows) ($P < .05$).

Table 3
Repeated measures ANOVA results of surface roughness data within each material (n = 10).

Material	Disinfection Protocol	F value	*P* value
Ava	Distilled water	4.234	.031
	NaOCl	11.553	<.001
	Tablet	3.851	.041
Merz	Distilled water	1.764	.2
	NaOCl	0.622	.548
	Tablet	8.901	.002
Poli	Distilled water	1.849	.186
	NaOCl	0.764	.48
	Tablet	4.992	.019
Conv	Distilled water	0.184	.834
	NaOCl	0.499	.615
	Tablet	1.101	.354

*P < .05 indicate significant differences among the initial, after brushing, and after disinfection surface roughness values within material.

brushing R_a values were similar (P = .32), and higher than the R_a of after effervescent tablet disinfection ($P \leq .042$). However, paired sample t-tests revealed significant differences only when NaOCl was used for the disinfection of Ava; baseline R_a was the lowest ($P \leq .037$) and the R_a after disinfection was the highest ($P \leq .037$) (Table 1).

3.2. Color difference results

One-way ANOVA revealed significant differences among the ΔE_{00} values of materials after brushing (df = 3, F = 8.332, $P < .001$), which were 1.21 ± 0.6 for Ava, 0.62 ± 0.36 for Merz, 0.85 ± 0.64 for Poli, and 1.24 ± 0.62 for Conv (n = 30). Ava and Conv had similar ΔE_{00} values (P = .88) that were higher than those of Merz (P < .001) and Poli (P ≤ .015). The difference between Merz and Poli was nonsignificant (P = .12).

Table 4 summarizes the descriptive statistics of overall ΔE_{00} values of each material after disinfection, while Fig. 1 illustrates the ΔE_{00} in between each time interval. Within the materials tested, disinfection protocols affected the ΔE_{00} values of Merz ($P < .001$) and Poli ($P < .001$). Disinfection by using effervescent tablet resulted in the greatest ΔE_{00} in both materials ($P < .001$), whereas the differences between storing in distilled water and NaOCl were nonsignificant (P = .72 for Merz and P = .80 for Poli). Significant differences among the materials were observed when NaOCl (P = .012) and effervescent tablet ($P < .001$) were used. When NaOCl was used, Ava and Merz had similar ΔE_{00} values (P = .93), which were lower than that of Conv (P = .004). The difference between Poli and other materials was nonsignificant (P ≥ .058). When effervescent tablet was used, the differences between Merz and Poli (P = .56), and Ava and Conv (P = .37) were nonsignificant. In addition, Merz and Poli had significantly higher ΔE_{00} than Ava and Conv (P < .001). Change in mean color coordinate values (L*, a*, and b*) of each material over gray backing is given in Fig. 2.

Table 4
Mean ΔE_{00} values and standard deviations for color change after disinfection compared with baseline (n = 10).

Disinfection Protocol	Material			
	Ava	Merz	Poli	Conv
Distilled water	1.17 ± 0.72^{aA}	0.85 ± 0.40^{aA}	1.03 ± 0.42^{aA}	1.27 ± 0.54^{aA}
NaOCl	0.68 ± 0.38^{aA}	0.70 ± 0.48^{aA}	0.93 ± 0.45^{aAB}	1.36 ± 0.60^{aB}
Tablet	1.04 ± 0.81^{aA}	4.68 ± 1.50^{bB}	4.39 ± 1.37^{bB}	1.49 ± 0.58^{aA}

*Different superscript letters represent significant differences (Lowercase letters for columns and uppercase letters for rows) (P < .05).

4. Discussion

Material type had a significant effect on R_a values at different time intervals. Therefore, the first null hypothesis was rejected. Baseline R_a values of Ava and Poli were significantly lower than Merz and Conv. In addition, Ava and Poli had R_a values similar to or slightly higher than clinically acceptable threshold of 0.2 μm before and after brushing.

The only significant effect of brushing on R_a was observed when baseline, after brushing, and after disinfection R_a values of Ava-NaOCl pair were evaluated. However, considering that this difference may be related to the randomization of the samples after brushing and the fact that the mean difference between baseline and after brushing R_a values of this pair is 0.04 μm, the authors believe that brushing did not significantly affect the R_a values. Thus, the 2nd null hypothesis was accepted. This result is in line with de Freitas Pontes et al.'s (2016) and Lira et al.'s (2012) studies. Contrarily, other studies have shown that brushing increased the surface roughness of denture base materials (Chang et al., 2021; Shinawi, 2017; Sorgini et al., 2012, 2015); it should be noted that brushing was performed by using dentifrice in those studies (Chang et al., 2021; Shinawi, 2017; Sorgini et al., 2012, 2015). In addition, a previous study has reported increased R_a of Poli after 40000 strokes of brushing with dentifrice (Shinawi, 2017), which may indicate the possible effect of dentifrice. However, to the authors' knowledge, no study has compared the R_a of denture base materials when brushed with soap slurry and dentifrice. Therefore, future studies are needed to substantiate this hypothesis.

Disinfection had a significant effect on the R_a values of PMMA resins. Therefore, the 3rd null hypothesis was rejected. A previous study investigated the R_a of 3 denture base materials (heat-cured PMMA, prepolymerized PMMA, and light-polymerized PMMA) after brushing, immersing in different solutions, and thermocycling (Alfouzan et al., 2021a). The authors (Alfouzan et al., 2021a) have concluded that disinfection protocols did not affect the surface roughness of heat-cured PMMA, whereas NaOCl disinfection of prepolymerized PMMA led to a higher R_a when compared with effervescent tablet disinfection. In the present study, Merz and Poli were significantly affected by the disinfection protocol considering the final R_a values within and between the materials tested. Even though both materials are prepolymerized PMMAs, effervescent tablet disinfection of Poli and NaOCl disinfection of Merz led to the highest R_a of these materials within disinfection protocols. In addition, only NaOCl resulted in a significant difference among test groups. These results may be associated with the fact that disinfectants interact with denture base materials differently (Arruda et al., 2015). Depending on the concentration of the disinfectant, the amount of chemical components leached from denture base materials may vary (Alfouzan et al., 2021a). In addition, water absorption of PMMA resins, which deteriorates their physical properties due to hydrolytic degradation (Alfouzan et al., 2021a) might have caused this difference among the tested materials and disinfection protocols.

Fourth null hypothesis was rejected as disinfection protocol had a significant effect on R_a values within PMMAs tested. When the R_a values measured at different time intervals within each material were concerned, it was observed that consecutive brushing and disinfection gradually increased the R_a of Ava when NaOCl was used. However, effervescent tablet disinfection of Merz led to its lowest R_a within different intervals. Nevertheless, other than Ava-effervescent tablet, Merz-effervescent tablet, Poli-distilled water, and Poli-NaOCl pairs, every material-disinfection protocol pair had R_a values higher than 0.2 μm. Sorgini et al. (2015) concluded that 0.5% NaOCl disinfection did not have a significant effect on after brush R_a of PMMA, which may be associated with the differences in tested materials, number of strokes (40000 vs 178000), and concentration of NaOCl. Considering the contradicting results between the present and Sorgini et al.'s (2015) studies, and the fact that combined effect of brushing and disinfection is scarcely investigated, studies that involve different materials, brushing parameters, and disinfection protocols are needed.

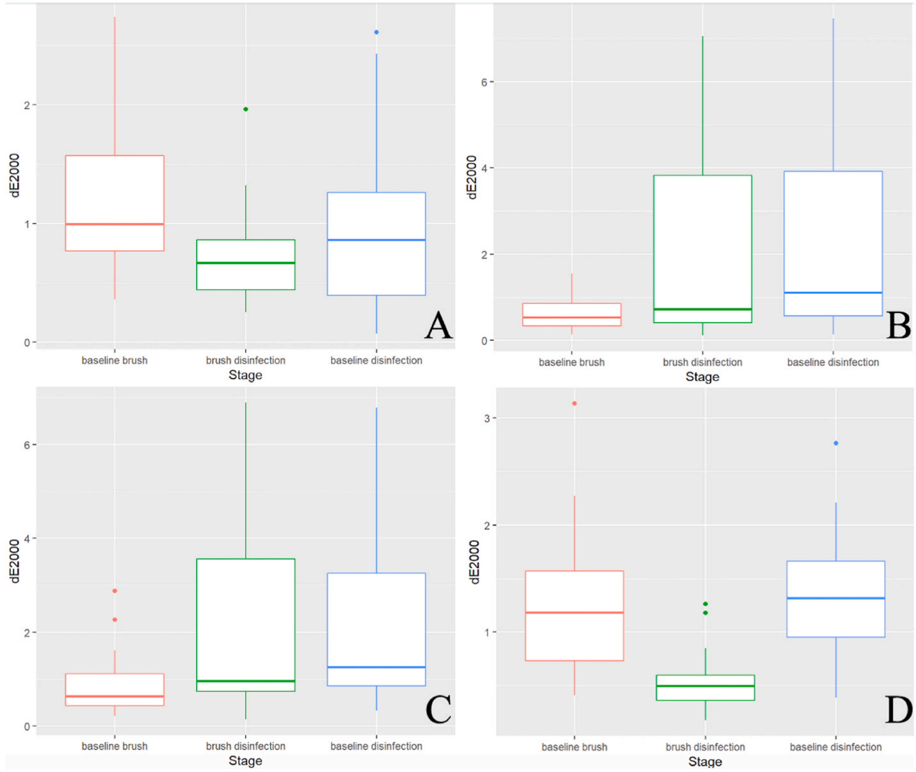

Fig. 1. Box plots of the ΔE_{00} values of each material in between different time intervals (A: Ava; B: Merz; C: Poli; D: Conv.

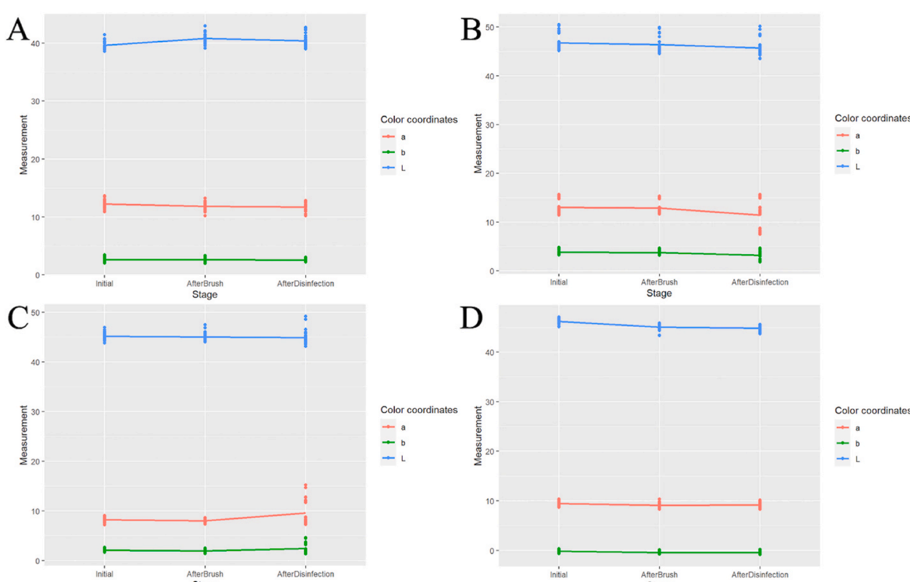

Fig. 2. Line graphs of change in color coordinates of each material in between different time intervals (A: Ava; B: Merz; C: Poli; D: Conv).

Material type had a significant effect on ΔE_{00} values at different time intervals, while brushing affected ΔE_{00} values among the PMMA resins tested. Therefore, the 5th and 6th null hypotheses were rejected. After brushing, Ava and Conv had higher ΔE_{00} values than Merz and Poli. However, all ΔE_{00} values after brushing were smaller than the perceptibility threshold of 1.72.

Disinfection protocol significantly affected the ΔE_{00} values of PMMA resins tested, which led to the rejection of the 7th null hypotheses. After disinfection, while both chemical disinfection protocols caused significant differences in ΔE_{00} values among the materials tested, only effervescent tablet disinfection of Merz and Poli resulted in the greatest color change within those materials. In addition, Merz and Poli showed greater color change than clinically acceptable threshold of 4.08 when effervescent tablet was used, while every other material-disinfection protocol pair had imperceptible color changes. These results may be interpreted as, when effervescent tablet for Ava and NaOCl for Poli is used, the maintenance of complete dentures made of Ava and Poli may be easier to sustain. However, this interpretation needs further in vivo corroboration.

Similar to the results of the present study, Alfouzan et al. (2021b) reported that effervescent tablet disinfection of prepolymerized PMMA caused higher color change than 5.25% NaOCl disinfection, whereas disinfection protocols did not affect the color stability of heat-cured PMMA. The effervescent tablet used in the present study contains chemicals that cause oxidation and its alkaline nature may explain this adverse effect on the color stability of denture base materials (Arruda et al., 2015) However, the authors (Alfouzan et al., 2021b) have also stated that the differences between heat-cured PMMA and prepolymerized PMMA were nonsignificant, regardless of the disinfectant used. This contradiction between the present study and Alfouzan et al.'s (2021b) study may be related to the differences in the materials tested as well as the color difference formulae used.

Merz and Poli showed a similar pattern of color stability as both materials had lower color change after brushing than after disinfection (Fig. 1B and C). As for the other materials, brushing led to a greater color change than disinfection (Fig. 1A and D). Some of the color coordinates of the materials tested changed throughout the procedures (Fig. 2). Lightness of Ava increased after brushing and showed a slight decrease after disinfection (Fig. 2A). In addition, lightness evidently decreased throughout the procedures for Merz (Fig. 2B) and after brushing for Conv (Fig. 2D). Merz and Poli (Fig. 2C) showed the greatest differences in redness, as a* values of Merz increased, whereas those of Poli decreased. None of the materials tested showed a notable change in yellowness throughout the procedures. However, considering that no perceptibility and acceptability thresholds have been defined for these parameters, it is critical to interpret these results carefully.

Even though the number of specimens were based on similar previous studies (Alfouzan et al., 2021a, 2021b) and significant differences were observed among the materials and disinfection protocols tested, the absence of a priori power analysis could be considered as a limitation. In vitro design of the present study is a limitation as possible deteriorating effects of intra oral conditions due to temperature changes were not simulated. In addition, duration of the chemical disinfection protocols was relatively short considering that 6 months do not replicate long-term use of complete dentures and sole effect of chemical disinfection on the tested parameters has not been investigated. Another limitation of the present study was that the tested prepolymerized PMMAs were limited to certain brands. Even though these materials are widely available and were tested in previous studies (Al-Dwairi et al., 2019; Alp et al., 2019; Arslan et al., 2018; Atalay et al., 2022; Gruber et al., 2021), other materials may lead to different results. Also, specimens of the present study were prepared to be compatible with the brushing machine and the spectrophotometer used. Therefore, no international standard was followed for the dimensions of the specimens. Finally, combined effect of brushing and chemical disinfection on other material properties such as surface hardness, flexural strength, water sorption, and bacterial accumulation should be investigated in future studies to broaden the knowledge on the limitations of the tested CAD-CAM PMMAs.

5. Conclusions

Within the limitations of this in vitro study, the following conclusions could be drawn:

1. Brushing did not increase the surface roughness of materials tested.
2. Disinfection protocol's effect on roughness varied for Merz and Poli. Effervescent tablet resulted in low surface roughness for Merz and high surface roughness for Poli compared with other disinfectants' effects.
3. After disinfection with NaOCl, surface roughness of Merz and Conv was higher than that of Poli.
3. Within material and disinfection protocol, surface roughness of Merz decreased after effervescent tablet disinfection. Surface roughness of Ava increased after NaOCl disinfection.
4. The effect of brushing on the color of materials varied. The color of Ava and Conv was affected from brushing more than the other materials.
5. Color stability of materials varied depending on the disinfection protocol. Effervescent tablet caused higher color change with Merz and Poli compared with other disinfectants.

CRediT authorship contribution statement

Gülce Çakmak: Methodology, Investigation, Conceptualization. **Mustafa Borga Donmez:** Writing – original draft. **Canan Akay:** Methodology, Investigation. **Sevda Atalay:** Investigation, Conceptualization. **Marcella Silva de Paula:** Methodology, Investigation, Conceptualization. **Martin Schimmel:** Visualization, Validation, Supervision. **Burak Yilmaz:** Writing – original draft, Visualization, Validation, Supervision.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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