

Physicochemical Properties and Bacterial Adhesion of Conventional and 3D Printed Complete Denture PMMA Materials: An *In Vitro* Study – Part I

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Received on: 02 November 2024; Accepted on: 17 December 2024; Published on: 27 January 2025

ABSTRACT

Aim: To evaluate and compare the surface morphology, wettability, roughness, and bacterial adhesion properties of polymethyl methacrylate (PMMA) materials fabricated by conventional methods and 3D printing for complete denture applications.

Materials and methods: Two PMMA materials were investigated: Conventionally processed (ProBase Hot) and 3D-printed (3DP) (V-Print Dentbase). Surface morphology ($n = 3$) was analyzed using scanning electron microscopy (SEM) and energy-dispersive X-ray spectroscopy (EDX). Surface roughness ($n = 10$) was measured using an optical profilometer. Wettability was assessed through contact angle measurements ($n = 6$) at 10, 30, and 60 seconds. Bacterial adhesion ($n = 9$) and biofilm formation ($n = 3$) were evaluated using *Escherichia coli* (*E. coli*) as a model organism, with quantitative bacterial counts and SEM analysis of bacterial morphology. Data were statistically analyzed.

Results: Scanning electron microscopy analysis revealed nanoparticles on the surface of 3DP samples, while EDX detected silicon in these samples, absent in conventional PMMA. 3D-printed surfaces exhibited significantly lower roughness ($1.05 \pm 0.32 \mu\text{m}$) compared to conventional surfaces ($20.46 \pm 6.71 \mu\text{m}$) ($p < 0.001$). Contact angle measurements showed that 3DP surfaces were more hydrophilic ($64\text{--}68^\circ$) than conventional surfaces (100°) ($p < 0.05$). Bacterial adhesion studies demonstrated more adherent bacteria on 3DP surfaces (92.5 ± 30.8) compared to the conventional surfaces (57.6 ± 12.5), but biofilm formation was observed only on conventional surfaces.

Conclusion: 3D-printed PMMA exhibited distinct surface characteristics compared to conventionally processed PMMA, including the presence of silicon nanoparticles, lower surface roughness, and higher hydrophilicity. While 3DP surfaces showed higher initial bacterial adherence, in contrast, they appeared to inhibit biofilm formation, which highlights the complex nature of bacterial interactions with these materials.

Clinical significance: Further clinical studies are needed to validate the results of this investigation and generate clinical translational data.

Keywords: 3D printing, Additive manufacturing, Bacterial adhesion, Complete denture, Polymethyl methacrylate, Surface roughness.

The Journal of Contemporary Dental Practice (2024): 10.5005/jp-journals-10024-3781

INTRODUCTION

Polymethyl methacrylate (PMMA) has been a cornerstone in dentistry for decades, particularly in the fabrication of complete dentures. Its widespread use can be attributed to its favorable properties, including biocompatibility, esthetic appeal, ease of manipulation, and cost-effectiveness. However, the advent of digital dentistry and additive manufacturing technologies has ushered in a new era of dental material fabrication, challenging the traditional methods of PMMA processing.¹ This paradigm shift has raised important questions about the comparative efficacy of conventionally processed and 3D-printed (3DP) PMMA materials, especially regarding their physicochemical properties and susceptibility to bacterial adhesion.² 3D printing technology has emerged as a promising alternative to conventional methods for fabricating dental prostheses, including complete dentures.³ The layer-by-layer additive manufacturing process offers advantages such as increased precision, reduced material waste, and the ability to create complex geometries easily. However, the adoption of any new technology in healthcare must be predicated on robust scientific evidence demonstrating its safety, efficacy, and potential benefits over existing methods.⁴ The physicochemical properties of dental materials play a crucial role in determining their clinical performance and longevity. Surface characteristics such as roughness, wettability, and morphology are essential

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How to cite this article: Khoury P, Kharouf N, Etienne O, *et al.* Physicochemical Properties and Bacterial Adhesion of Conventional and 3D Printed Complete Denture PMMA Materials: An *In Vitro* Study – Part I. *J Contemp Dent Pract* 2024;25(11):1001–1008.

Source of support: Nil

Conflict of interest: Dr Mutlu Ozcan is the Editorial board members of this journal and this manuscript was subjected to this journal's standard review procedures, with this peer review handled independently of these Editorial board members and their research group.

as they influence the material's mechanical properties and its interaction with the oral environment.⁵ Bacterial adhesion and subsequent biofilm formation on dental materials are of paramount concern in prosthodontics. The oral cavity, with its warm, moist environment and regular nutrient supply, provides an ideal habitat for microbial growth.⁶ Denture surfaces can serve as reservoirs for microorganisms, potentially leading to oral infections, denture stomatitis, and other health complications. Various factors influence bacteria's adhesion to PMMA surfaces. Understanding how these factors differ between conventionally processed and 3DP PMMA materials is crucial for predicting their clinical performance and developing strategies to minimize bacterial colonization.⁷ The comparison between conventional and 3DP PMMA materials extends beyond surface characteristics to encompass their overall physical and mechanical properties.⁸ Conventionally processed PMMA, typically fabricated through heat-curing or self-curing methods, has a well-established track record in clinical use. Its properties, strengths, and limitations are well documented in the literature. In contrast, 3DP PMMA materials are relatively new, and their long-term clinical performance is still being evaluated. These materials often incorporate proprietary formulations and may exhibit different polymerization characteristics than their conventional counterparts.^{1,2} Surface roughness, a critical parameter in dental materials, is quantified using optical profilometry.^{9,10} This non-contact method provides accurate surface topography measurements, allowing for calculating parameters such as arithmetic mean roughness (Ra). The comparison of roughness values between conventional and 3DP PMMA surfaces is crucial, as it directly impacts not only the material's mechanical properties but also its susceptibility to bacterial colonization and biofilm formation.¹¹

Several tests could be performed to evaluate the surface energy of a material, including contact angles analysis to assess their behavior in the oral environment.¹²

The choice of *Escherichia coli* (*E. coli*) as a model organism, while not typically associated with oral infections, is a useful proxy for understanding bacterial behavior on these surfaces.¹³

The objective of this study was to compare the physicochemical properties and bacterial adhesion characteristics of PMMA materials used in complete denture fabrication, focusing on those produced by conventional methods and 3D printing processes. Specifically, the study aimed to evaluate and contrast the surface morphology, wettability, roughness, and adhesion of *Candida albicans* (*C. albicans*) between conventionally fabricated and 3DP PMMA materials. The null hypothesis proposed that there would be no significant differences in these properties between the two fabrication techniques.

MATERIALS AND METHODS

Materials

This study used two types of PMMA materials: A 3D-printable resin and a conventional heat-curing PMMA (Table 1). This *in vitro* study was conducted in 2023 in collaboration between Strasbourg/France and Beirut/Lebanon.

Sample Preparation

Specimen Design and Preparation

Two virtual designs of a cylinder were created using computer-aided design software (Dental Cad, Exocad GmbH, Germany) to simulate a denture base: (1) one design of a cylinder measuring 20 mm in diameter and 5 mm in height $n = 16$ were used for the wettability and roughness tests; (2) the second design of a cylinder measuring 5 mm in diameter and 3 mm in height ($n = 15$) were used for scanning electron microscopy (SEM) and bacterial adhesion tests.

These two designs were then converted into a stereolithography (STL) file for production.

A total of 31 specimens were fabricated for each group according to the manufacturing technique.

3D-printed Group: The 3DP group was fabricated using additive manufacturing technology. The PMMA denture base was printed using V-print Dentbase (VOCO, Germany) on an LCD 3D printer (DentraTec, Lebanon). The manufacturing process began with importing the STL files of the denture base into slicing software (Chitobox). Following printing, all samples were cleaned and post-cured following the manufacturer's instructions. This post-curing process was carried out using a dedicated post-curing device (Dentmate, Taiwan).

Conventional Heat-cured (CHC) Group: The CHC group, fabricated using traditional denture manufacturing techniques. The designed denture base was 3D printed using Castable Wax 40 resin (Formlabs, Germany), creating a precise wax prototype. Following traditional prosthodontic procedures, the wax assemblies were invested in dental stone and conventionally flashed. The wax was then eliminated through a boil-out process, creating molds for the final denture bases. Promolux conventional heat-polymerized PMMA (Merz Dental, Germany) was packed into these molds and processed according to the manufacturer's recommended heat-curing cycle. This hybrid approach, combining modern 3D printing for the wax prototype with conventional heat-curing techniques, allowed for the production of 31 specimens that maintained consistency with the digital designs while representing traditional denture fabrication methods.

Table 1: Chemical compositions, preparation methods, and manufacturers of conventional and 3DP materials

Material	Composition	Curing method	Manufacturer
V-print dentbase (printed group)	Liquids: <ul style="list-style-type: none"> Aliphatic urethane dimethacrylate Bis-EMA Triethylene glycol dimethacrylate Diphenyl (2,4,6-trimethyl benzoyl) phosphine oxide 	Light curing by layer	Voco GmbH, Germany
ProBase hot (conventional group)	Powder: PMMA Liquid: MMA	Heat curing 10 hours at 80°C	Ivoclar Vivadent AG, Liechtenstein

Surface Morphology Analysis

Surface morphology was examined using an SEM (FEI Company, Eindhoven, The Netherlands) at 10 kV with magnifications of $\times 500$ and $\times 2000$. Three samples (5 mm in diameter and 3 mm in height) of each group were cleaned with 100% ethanol in an ultrasonic bath for 3 minutes, mounted on SEM stubs, and sputter-coated with gold palladium. Energy-dispersive X-ray spectroscopy (EDX) was employed to identify the chemical composition of each surface, with an acquisition time of 50 seconds and a working distance of 10 mm.

Surface Roughness Measurement

Surface roughness was assessed using an optical profilometer (InfiniteFocus SL, Bruker Alicona, Graz, Austria) at $\times 10$ magnification, with horizontal and vertical resolutions of 2 and 5 μm , respectively, on 10 samples of each group (20 mm in diameter and 5 mm in height). The arithmetic mean roughness (Ra) was determined across five distinct areas for each sample, following ISO 4287 guidelines. Each Ra measurement was conducted along a profile of at least 18 μm using a sawtooth pattern perpendicular to the surface relief and a cut-off filter (Lc) of 2,500 μm .

Contact Angle Measurement

Six specimens from each group (20 mm in diameter and 5 mm in height) were tested. Wettability was evaluated using a contact angle device (Biolin Scientific, Espoo, Finland). A 5 μL drop of distilled water was placed on each sample, and the contact angle was measured using a horizontal camera. Measurements were taken at 10, 30, and 60 seconds to assess changes in wettability over time.

Bacterial Adhesion Analysis

Bacterial Culture

Escherichia coli (SCC1) was cultured in LB (Lysogeny broth) (Difco™, Le Pont de Claix, France) according to the manufacturer's instructions.¹⁴ The bacterial suspension was adjusted to an optical density (OD_{600}) of 0.3 for all experiments.

Quantitative Bacterial Adhesion

Nine samples from each group (5 mm diameter, 2 mm height) were sterilized using an Auto sterilizer FLASH VBX25D (BRC SAS, Bouaye, France) for 30 minutes. Each sample was placed in a well of a 24-well plate with 2 mL of bacterial suspension. The plates were incubated for 24 hours at 37°C with constant stirring at 120 rpm. After incubation, the medium was gently removed, and each disc was transferred to a sterile 15 mL falcon tube containing 3 mL of phosphate-buffered saline (PBS) (Dominique Dutscher, Bernolsheim, France). The tubes were sonicated for 10 minutes and vortexed for 1 minute to detach adhered bacteria. Each suspension's optical density (OD_{600}) was measured to quantify bacterial adhesion.

Bacterial Morphology Analysis

Three additional samples from each group were prepared following the same procedure as stated above. After 24 hours of incubation, the samples were gently rinsed with distilled water for 10 seconds and fixed with 0.05 M glutaraldehyde in 4% cacodylate buffer for 4 hours. The samples were then rinsed three times with 4% cacodylate buffer (5 minutes each) and dehydrated in a graded ethanol series (35, 50, 70, and 95%) for 3 minutes each.

Final drying was performed using hexamethyldisilane (HMDS). Samples were transferred from 95% ethanol to a 1:1 solution of HMDS for 10 minutes, then to 100% HMDS twice for 10 minutes each. Due to its high toxicity, HMDS steps were carried out in a fume hood overnight. After drying, the samples were sputter-coated with gold-palladium alloy (20/80) and observed under SEM to analyze *E. coli* biofilm morphology, attachment, and density. Six images from each group were analyzed using ImageJ (version 1.54 g, Wayne Rasband, Maryland, USA) to quantify the number of attached bacteria.

Statistical Analysis

Data were analyzed using SigmaPlot release 11.2 (Systat Software, Inc., San Jose, CA, USA). The Shapiro–Wilk test was used to assess data normality. Significant differences in bacterial adhesion, roughness, and contact angle between groups were determined using the Kruskal-Wallis test (one-way analysis of variance on ranks) with the *post-hoc* Tukey test for multiple comparisons. The statistical significance level was set at $\alpha = 0.05$.

RESULTS

Surface Morphology

Scanning electron microscopy revealed distinct morphological characteristics for the conventional and 3DP PMMA surfaces (Fig. 1). Both materials exhibited surface porosity, a common feature in polymer-based dental materials. However, the nature and distribution of these porosities differed between the two groups. The conventional PMMA surfaces displayed a relatively homogeneous topography with randomly distributed pores. These pores likely result from the heat-curing process, where trapped air or volatile components can create small voids in the material.

In contrast, the 3DP PMMA surfaces exhibited a unique feature: Nanoparticles on the surface. These nanoparticles, likely a result of the specific formulation of the printable resin, could have significant implications for the material's properties.

By using energy-dispersive X-ray spectroscopy (EDX) analysis, both conventional and 3DP samples showed solid signals for carbon (C) and oxygen (O), the primary constituents of PMMA. Interestingly, the 3DP surfaces also revealed the presence of silicon (Si). This silicon content could be attributed to silica nanoparticles in the printable resin, which might serve as reinforcing agents or stabilizers.

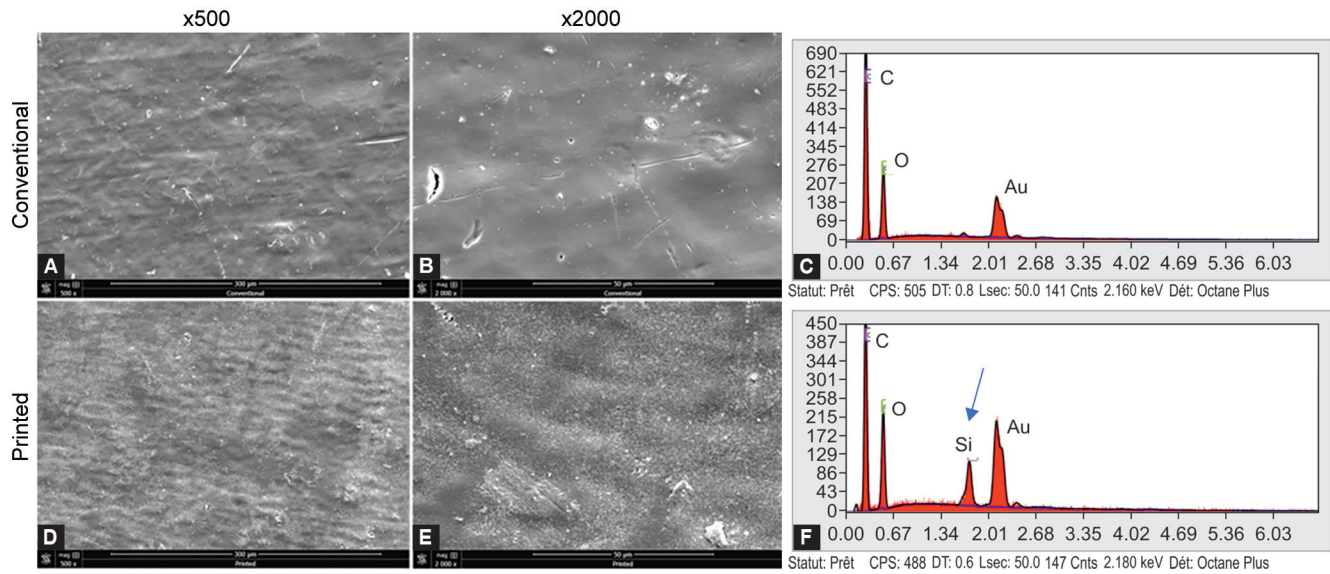
The detection of gold (Au) on both surfaces results from the sputter coating process used in SEM sample preparation and does not reflect the inherent composition of the materials.

Surface Roughness

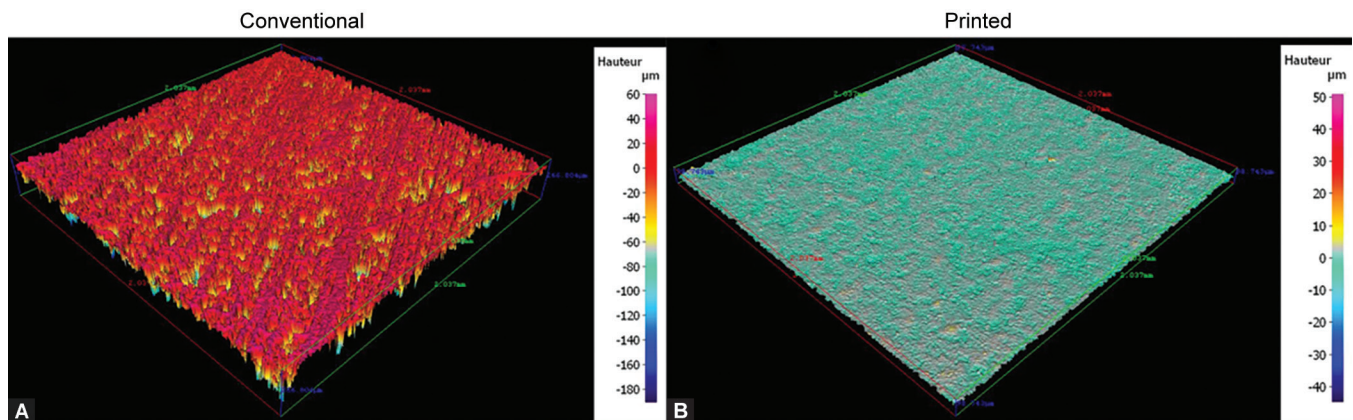
Surface roughness analysis revealed significant differences between the conventional and 3DP PMMA materials (Fig. 2). The conventional PMMA surfaces exhibited substantially higher roughness values ($20.46 \pm 6.71 \mu\text{m}$) compared to the 3DP surfaces ($1.05 \pm 0.32 \mu\text{m}$), with the difference being statistically significant ($p < 0.001$). This marked difference in surface roughness can be attributed to the distinct manufacturing processes.

Wettability

Contact angle measurements provided insights into the wettability of the PMMA surfaces (Fig. 3). The conventional PMMA group



Figs 1A to F: Scanning electron microscopy images at $\times 500$ and $\times 2000$ magnifications demonstrated the morphology of (A and B) conventional; (D and E) Printed materials surfaces. EDX spectrum demonstrated the presence of O and C for the (C) conventional group as well as Si for the (F) printed group. Au is the signal of the sputter coating process



Figs 2A and B: 3D micrographs taken by a profilometer demonstrated the roughness of both sample surfaces. (A) Conventional; (B) Printed

demonstrated consistently high contact angles at all-time points: $100 \pm 1.2^\circ$ at 10 seconds, $100 \pm 1.08^\circ$ at 30 seconds, and $100 \pm 1.08^\circ$ at 60 seconds. These values indicate a highly hydrophobic surface. In contrast, the 3DP PMMA surfaces exhibited significantly lower contact angles: $68 \pm 4.5^\circ$ at 10 seconds ($p = 0.002$), $65 \pm 4.1^\circ$ at 30 seconds ($p < 0.001$), and $64 \pm 3.7^\circ$ at 60 seconds ($p = 0.002$). These lower contact angles suggest a more hydrophilic surface than the conventional PMMA.

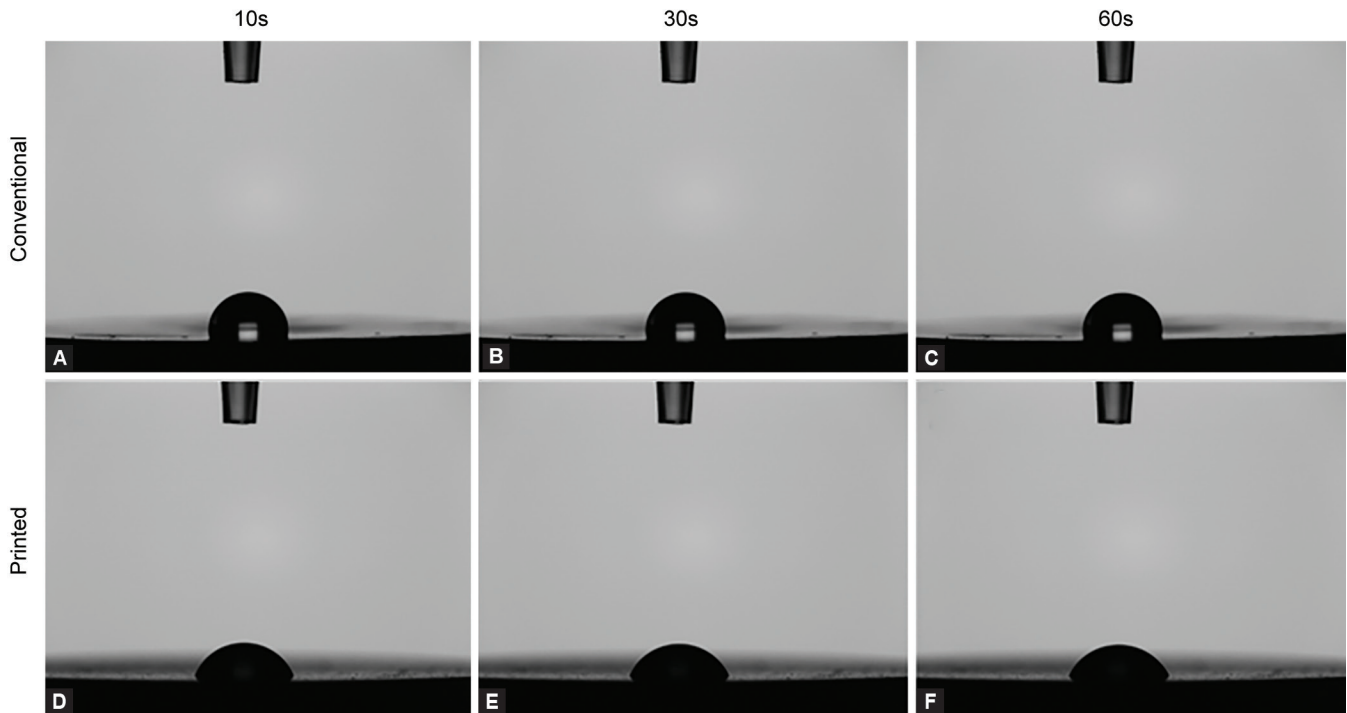
Bacterial Activity

Scanning electron microscopy analysis revealed intriguing differences in bacterial morphology and adhesion patterns between the conventional and 3DP surfaces (Figs 4 and 5). On the 3DP surfaces, qualitatively larger bacterial sizes were observed compared to the traditional group. Quantitative analysis using ImageJ software showed a higher number of bacteria on the 3DP surfaces (92.5 ± 30.8) compared to the conventional ones (57.6 ± 12.5), with the difference being statistically significant ($p = 0.028$) (Table 2). The optical density (OD) measurements of

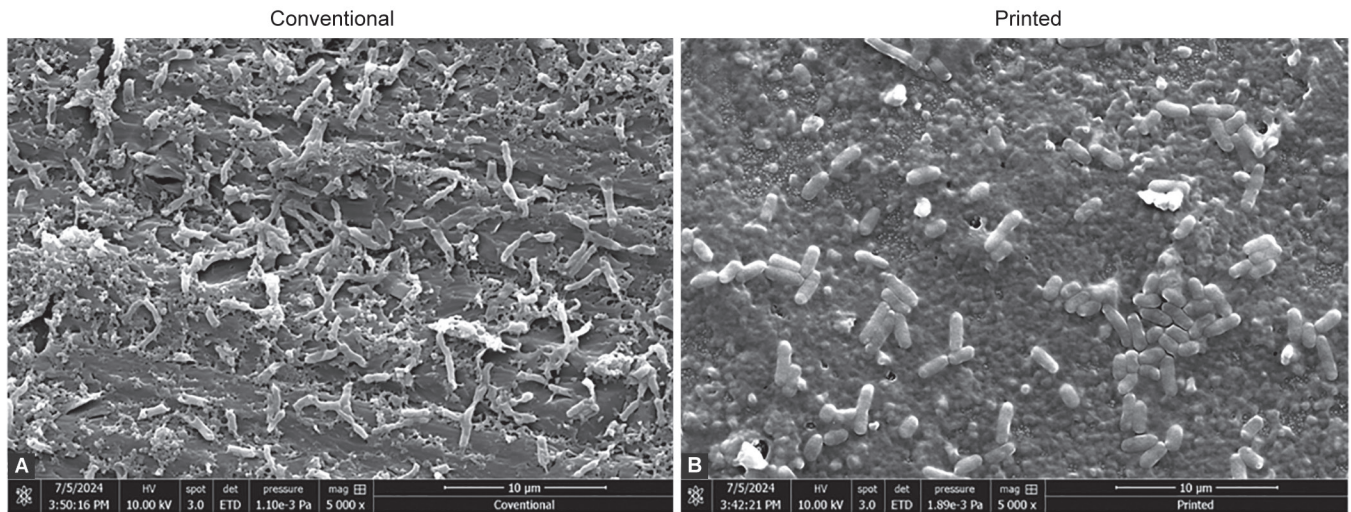
the bacterial medium showed no statistically significant difference between the conventional (0.018 ± 0.017 nm) and 3DP (0.012 ± 0.013 nm) groups ($p = 0.479$).

DISCUSSION

The SEM analysis revealed distinct morphological differences between PMMA surfaces fabricated by conventional methods and 3D printing. Both manufacturing procedures showed some porosity on the surface, which could affect the materials' mechanical integrity and adhesive performance when used in clinical settings.¹⁵ Notably, the 3DP samples displayed nanoparticles on the surface, increasing surface area and enhancing bonding potential with other dental materials.¹⁶ This could be advantageous for applications requiring adhesive solid interfaces. Furthermore, the EDX analysis indicated the presence of Si on the 3DP surfaces, which was absent in the conventionally fabricated PMMA. The presence of Si may impart additional thermal stability and biocompatibility, which could improve long-term performance.¹⁷ While C and O detections were



Figs 3A to F: Contact angles of the profile of 5 μL water drop onto both material surfaces. (A to C) Conventional material; (D to F) Printed material at different time points after 10, 30, and 60 seconds of deposition



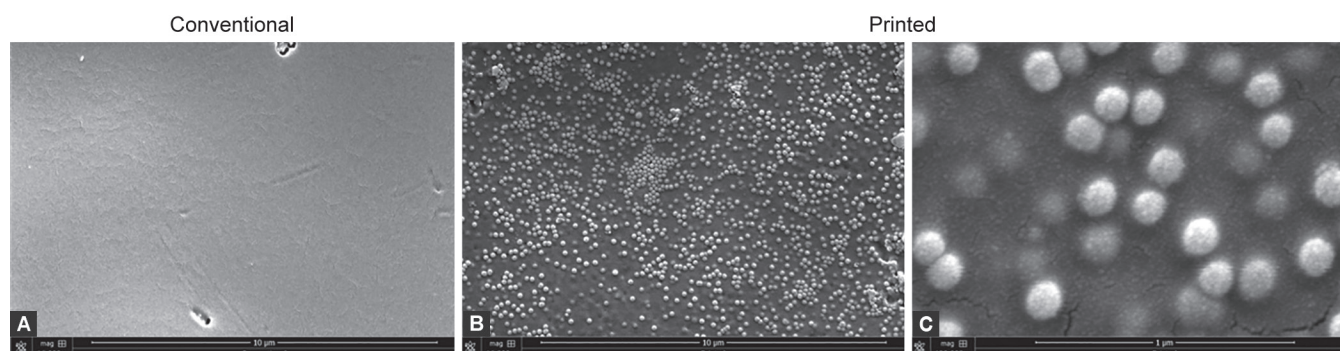
Figs 4A and B: Scanning electron microscopy images demonstrated the bacterial adhesion, density and accumulation onto the surface of (A) Conventional; (B) Printed materials after 24 hours of incubation at 37°C

consistent across both surfaces, the detection of Au was attributed to the sputter-coating process employed for SEM imaging.¹⁸ These discrepancies emphasize the potential for 3D printing to develop PMMA materials with unique surface properties that may benefit dental applications, such as enhanced bonding and durability.¹⁶ Further research is needed to explore the clinical implications of these findings.¹⁷

The comparison between conventional and 3DP PMMA surfaces showed a significant difference in surface roughness values. The conventional surfaces had notably higher roughness values compared to the smoother 3DP surfaces. This difference can be

attributed to the distinct fabrication techniques used for each. Conventional fabrication methods, which involve manual polishing and curing, often result in more surface irregularities due to the less controlled nature of the finishing process.¹⁹ In contrast, 3D printing technologies, such as SLA and digital light processing (DLP), offer more precision in layer deposition, creating surfaces with lower roughness and fewer surface defects.²⁰

Surface roughness is an important element that influences the clinical performance of dental materials, specifically bacterial colonization, wear resistance, and esthetic.²¹ Materials with higher surface roughness, such as commonly produced PMMA, may be



Figs 5A to C: High magnifications of SEM images demonstrate the surface characteristics of (A) Conventional (16000 \times); (B and C) Printed (16000 \times and 120000 \times , respectively)

Table 2: Contact angle, roughness, and medium OD of both materials

Materials	Conventional			Printed			Statistical analysis
	10s	30s	60s	10s	30s	60s	
Contact angle ($^{\circ}$)	100 \pm 1.2	100 \pm 1.08	100 \pm 1.08	68 \pm 4.5	65 \pm 4.1	64 \pm 3.7	<0.05
Roughness (μ m)		20.46 \pm 6.71			1.05 \pm 0.32		<0.05
Medium OD (nm)		0.018 \pm 0.017			0.012 \pm 0.013		0.479
Bacterial count		57.6 \pm 12.5			92.5 \pm 30.8		0.028

more susceptible to plaque deposition and biofilm formation, increasing the risk of subsequent caries or periodontal disease.²²

Conventional PMMA processing involves mixing powder and liquid components and heat curing. This process can lead to inherent surface irregularities due to monomer evaporation, polymerization shrinkage, and the presence of residual monomers.^{19,20} In contrast, the significantly lower roughness of 3DP surfaces suggests a more controlled manufacturing process. The layer-by-layer deposition in 3D printing, combined with liquid resin, likely results in a smoother surface finish.^{21,22} The presence of nanoparticles, as observed in SEM, doesn't significantly increase surface roughness, suggesting they are well-integrated into the polymer matrix.

Smoother surfaces, like those made by 3D printing, can reduce bacterial adhesion and improve the long-term clinical success of restorations.²³ Furthermore, smoother surfaces are often associated with better esthetic outcomes and enhanced patient comfort, as they feel less abrasive and are easier to maintain.²⁴ Given these possible advantages, the adoption of 3DP materials in dentistry may increase not just clinical durability, but also patient happiness. However, more research is needed to determine how these changes in surface roughness affect mechanical qualities such as fracture resistance and wear durability.²⁵

The observed higher contact angle values for the conventional PMMA group than the 3DP group at various time points (10, 30, and 60 seconds) indicate a significant difference in the surface wettability between the two fabrication methods. The conventional group consistently demonstrated higher contact angles compared to the 3DP group, which showed significantly lower contact angles. This indicates that the conventionally fabricated surfaces were more hydrophobic than the 3DP surfaces. As measured by the contact angle, surface wettability is crucial in determining how well a material can interact with adhesives, bonding agents, and other surface treatments used in dentistry. Higher contact angles correlate with greater hydrophobicity, impacting the material's adhesion properties and overall clinical performance.²⁶ The lower contact angle values

observed in the 3DP group suggest that these surfaces are more hydrophilic, which could enhance bonding with hydrophilic adhesive systems and improve the longevity of restorations.²⁷

The differences in contact angles between the two groups could be attributed to the variations in surface roughness and the inherent material properties of the PMMA processed through different manufacturing techniques. The conventional method, which often leads to a rougher surface, can increase surface energy and, consequently, a higher contact angle.²⁸ On the other hand, 3D printing, which produces smoother surfaces, may lead to lower surface energy and, thus, a reduced contact angle.²⁹ Interestingly, no significant differences were observed in the contact angle within the same group across the different time points, indicating that the surface characteristics remained stable over the observation period. This stability is crucial for consistently applying adhesives and bonding agents in clinical settings, ensuring predictable and reliable results.³⁰ Overall, the lower contact angles observed in the 3DP PMMA surfaces highlight the potential advantages of 3D printing in dentistry, particularly in applications where strong bonding is critical. However, the implications of these findings on long-term clinical outcomes warrant further investigation. The stability of contact angles over time (10, 30, and 60 seconds) within each group ($p > 0.05$) indicates that both materials maintain their wetting characteristics, at least over the short term. This stability is essential for predicting how the materials interact with oral fluids and dental adhesives. The difference in wettability between the two materials is striking and could have significant clinical implications. The more hydrophilic nature of the 3DP PMMA might promote better adaptation to oral tissues and improved patient comfort. It could also enhance the material's ability to retain a salivary film, which is crucial for lubrication and protection against wear. However, increased hydrophilicity might also lead to higher water sorption, potentially affecting the material's dimensional stability and mechanical properties over time. The balance between these factors needs careful consideration in clinical applications.

The printed group has silica particles included in its chemical composition, which could play an essential role in the hydrophile of these surfaces and play a stabilizer role, as mentioned by the manufacturer. Silica is hydrophilic by nature because of the presence of hydroxyl groups (-OH) attached to the silica particles' surface.³¹ Scanning electron microscopy revealed notable differences in bacterial morphology and adhesion between the 3DP and conventional surfaces. The SEM images showed larger-sized bacterial colonies on the printed surfaces than the traditional group. This observation indicates that surface roughness and texture significantly influence bacterial colonization and biofilm formation. Higher roughness levels, often found in 3DP surfaces, can provide more surface area and niches for bacteria to adhere to, potentially leading to increased bacterial growth.³²

Quantitative analysis using ImageJ further supported these findings, showing more bacteria on the printed surfaces than conventional ones. This suggests that the structural characteristics of 3DP materials, which often exhibit increased surface irregularities and porosities, can enhance bacterial colonization.³³ In contrast, being smoother and more homogeneous, conventional surfaces demonstrated the onset of biofilm formation without extensive bacterial adhesion or proliferation. The size and distribution of these pores can influence the material's mechanical properties and interaction with the oral environment. This difference in bacterial morphology could indicate altered growth patterns or stress responses induced by the surface characteristics of the 3DP material. The presence of nanoparticles or the more hydrophilic nature of these surfaces influences bacterial behavior at the microscopic level.

Given their lower surface roughness, this higher bacterial count on 3DP surfaces is counterintuitive. It suggests that factors other than surface roughness, such as surface chemistry or the presence of nanoparticles, play a significant role in bacterial adhesion. Interestingly, while more bacteria were observed on the 3DP surfaces, the conventional surfaces showed signs of early biofilm formation. This observation indicates that the traditional PMMA might provide a more favorable environment for bacterial colonization and biofilm development, despite having fewer individual bacteria attached.

Despite the higher bacterial count, the absence of visible bacterial attachments on the 3DP surfaces is particularly intriguing. While these surfaces may allow for initial bacterial adhesion, they inhibit the progression to biofilm formation. This could be due to the material's surface properties, such as its higher hydrophilicity or the presence of nanoparticles, which might interfere with the bacterial communication necessary for biofilm development. This suggests that while there are differences in bacterial adhesion and morphology, the overall bacterial growth in the surrounding medium is similar for both materials. These findings highlight the complex nature of bacterial interactions with PMMA surfaces and underscore the need for a multifaceted approach in designing dental materials that can resist bacterial colonization and biofilm formation.

The initiation of biofilm formation was observed on the conventional surfaces. At the same time, minimal bacterial attachment was noted on the printed surfaces, highlighting a critical difference in how these materials interact with bacteria. Conventional surfaces may have characteristics that favor the early stages of biofilm development, such as greater wettability or smoother surfaces that initially support bacterial adhesion.³⁴

In contrast, the 3DP surfaces, despite their higher roughness, might exhibit properties that limit early bacterial colonization, such as more hydrophobicity or less favorable surface chemistry for initial bacterial adhesion.^{35,36} These observations underscore the importance of surface treatment and material selection in managing bacterial adhesion and biofilm formation in dental applications. Further research into optimizing surface properties and antimicrobial treatments for both types of materials could enhance their clinical performance and longevity.

This study addresses an important area by comparing conventional and 3DP PMMA, which is increasingly relevant given the rise of additive manufacturing in dentistry, and provides a comprehensive insight into surface morphology, composition, and wettability with a quantitative and qualitative bacterial analysis. This *in vitro* study's limitations depend on its ability to replicate complex intraoral situations, such as variations in saliva, temperature, and multi-species biofilm interactions as only *E. coli** was used as a test organism, which does not represent the diversity of oral microbiota that interacts with denture materials *in vivo*.

Further studies should be performed to evaluate the adhesion of several bacteria, such as *S. mutans* and *C. albicans*. In addition, an aging period should be performed to evaluate the effect of aging on the bacteria's adhesion and surface characteristics, including morphological and roughness changes, which could play an important role in biocompatibility and bacteria colonization. This article presents part I of the comparison between two PMMA materials fabricated by conventional methods and 3D printing for complete denture applications. In part II, the mechanical properties, including the bond strength and failure modes, of these materials will be evaluated.

Long-term studies are needed to evaluate the wear characteristics, mechanical properties, and *in vivo* performance of 3DP dentures. Additionally, investigation is warranted into optimizing the surface properties of 3DP materials to enhance their antibacterial properties while maintaining favorable physical characteristics.

CONCLUSION

3D-printed PMMA surfaces exhibited unique characteristics, including nanoparticles, lower surface roughness, and higher hydrophilicity than conventional PMMA. These properties offer advantages regarding material bonding, patient comfort, and potentially reduced long-term biofilm formation. However, the higher initial bacterial adherence on 3DP surfaces, coupled with the apparent inhibition of biofilm formation, highlights the complex nature of bacterial interactions with these materials.

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