Research Article

Elucidating the impact of incorporating grapefruit skin seed particles into three-dimensional printed denture base resin on Candida Albicans adhesion and surface roughness

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Received date: 05-12-2024
Accepted date: 01-02-2025
Published date: 15-09-2025

Article DOI

Abstract: Background: Denture stomatitis is a common problem in patients wearing dentures. To improve people's health and prevent infection spread, it is essential to select suitable fillers into the denture base material, which should not only be effective in term of durability and mechanical properties, but also do not exert unfavorable effects on the oral tissue. Objectives: The purpose of this study was to determine the effects of adding grapefruit skin seed particles (GSSPS) at different concentrations on the adhesion of Candida albicans and the surface roughness of Three-Dimensional (3D) printed denture base resin. Materials and Methods: Sixty specimens were digitally printed with 30 disks distributed for the Candida albicans adhesion test and another 30 samples for the surface roughness test. Samples in each test were divided into three groups (n=10) for each GSSPS percentage (0 wt%, 5 wt%, and 7 wt%). Spectrophotometer was chosen to determine the optical density of Candida albicans and profilometer was used to measure surface roughness. Results: There was significant decrease in both the Candida albicans adhesion and the surface roughness of 3D printed denture base resin after addition of GSSPS. Scanning electron microscopy proved the presence of an evenly distributed GSSPS particles, atomic force microscopy proved the decrease in surface roughness when the concentration of GSSPS increased and energy dispersive X-ray spectroscopy (EDX) showed the presence of new elements. Conclusion: Within the limitation of this study, addition of GSSPs to 3D printed denture base resin has a significant effect on decreasing the adhesion of the Candida albicans and the surface roughness, this leads to decrease the opportunity to present denture stomatitis.

Keywords: Three-Dimensional printing, 3D printed denture resin, grapefruit skin seed particles, flavonoids, Candida albicans, surface roughness

Introduction

Denture base materials have been the focus of development for decades due to their inherit drawbacks and the fact that denture fabrication requires multiple clinical and laboratory steps. The advancement in digital dentistry and additive manufacturing led to the development of 3D-printed denture base resin [(1)]. However, this resin still has several disadvantages, such as vulnerability to microbial colonization and its inadequate physical and mechanical characteristics [(2)].

Oral candidiasis is commonly diagnosed in patients who has been wearing dentures. In denture related stomatitis, *Candida albicans* was the most commonly isolated species. This yeast adheres readily onto the oral prostheses and also medical catheters [3].

Candida albicans is gram-positive microorganisms. It is oval or round shape as seen by microscope, lives in 37°C temperature in a humid condition and at acidic environment of about 6-6.2 pH [4]. *Candida albicans* in specific, is a polymorphic yeast, that has the ability to penetrate host tissue causing skin and mucosal infection [5].

The properties of the denture base material, such as the surface roughness can significantly affect the occurrence of denture stomatitis (DS). Many oral microorganisms especially those responsible for periodontal disease, caries, and DS, still survive in the mouth forming colonies if they adhere to the non-shedding surface, so increased surface roughness facilitates candida adherence and has a harmful impact on the oral health [6]. To achieve a smooth denture base surface, it is crucial to finish and polish the denture surfaces well using various abrasives [7].

The use of natural products in the treatment of human disease has been widely advocated for many years [8]. The principal benefits of using natural plant extracts are attributed by its biocompatibility, safety, minimal adverse effects, and their economic value [9].

There are many antifungal products that have been added to polymethyl methacrylate (PMMA) for example, Henna (Lawsonia inermis, natural inexpensive plant [2]. Other products were used to decrease DS by immersion of the denture into an antifungal solution like eugenol [10] or adding fillers to soft denture lining material like tea tree oil [11].

Grapefruit seed extract, was known to be rich in polyphenolic compounds; flavonoids, which are a kind of polyphenol consisting of about 80% polyphenols [12]. Flavonoids have an inhibitory impact on some *Candida spp*., gram-negative and gram-positive bacteria, by a number of mechanisms [13,14]. Grapefruit seed extract was reported to possess powerful antifungal and antimicrobial properties [14,15].

Therefore, the objective of this study is to evaluate the ability of grapefruit skin seed particles (GSSPs) to prevent the attachment of *Candida albicans* to the 3D printed denture base resin and evaluate their effect on the surface roughness. The null hypotheses supposed that there have been no effects on *Candida albicans* attachments or the surface roughness of 3D-printed denture base resin reinforced with GSSPs.

Materials and Methods

Grapefruit skin seed particles (GSSPs) production

The mature grapefruits (Duncan type) were bought from local market in Baghdad. A sharp blade was used to cut it into pieces, and the seeds were collected manually. The seeds peels were extracted with the same blade. Then, tap water was used to wash the peels, and they were dried in the oven for 24 hours at 40°C [16]. After that, using a mixer blinder, the seeds peels were ground and a planetary ball mill (NQM-0,4 MODEL PLANTARY BALL MILL, MTI corporation, USA) was used to make it in micro size [17] and finally high-quality stainless-steel sieve (25 μm, Diameter 200mm, ASTM No. 500, MANIKARN 8 inch) was used to obtain the final GSSPs powder. 2g from this powder was used to determine particle size by use the particle size analyzer (90Plus Brookhaven, software Ver 5.34) which confirmed the effective particulate diameter was 6327nm.

Mixing procedure of GSSPs with 3D-printed denture base resin

3D-printed denture base resin (Optiprint Laviva) from Dentona Germany Company which had a light pink color was used in this study. The resin was poured into autoclavable (250 ml) amber glass bottle, the temperature increased to 60°C for 30 minutes to reduce its viscosity, and the micro-particles were gradually added to the liquid resin while it was continuously stirred with a magnetic stirrer (78-1 magnetic heating stirrer, India) at room temperature for 8 hours to obtain a homogeneous mixture that was ready for printing (according to manufacture instructions).

Specimens' preparation

The sample size was determined using G*Power software (3.1.9.7; Heinrich-Heine-Universität Düsseldorf) with the following parameters: alpha: 0.05, power: 0.80, effect size f: 0.6. A total of 30 specimens were used for each test. For the Candida albicans adherence test, 30 disks with dimension of 6 mm in diameter and 2 mm in thickness were prepared, and for the surface roughness test, 30 specimens were printed with dimensions of (12*12*3mm) according to ISO (20795-1:2013).

The specimens were designed using (Autodesk, San Rafael, Meshmixer, CA, USA) and the resultant standard triangle language (STL) files were sent to Pre Form software (Formlabs, Somerville, MA, USA) for processing and support structure design and finally printed using digital light processing (DLP) printer (Microlay Versus 385, EU). The Z-axis thickness of each slice was set to 50 micrometer and it took seven seconds to print per slice [18].

After the printing, the residual resin was removed with a clean disposable spatula. Then the build platform was removed from the printer. The printed specimens were cleaned in an ultrasonic bath filled with 99% isopropyl alcohol for 5 minutes. Compressed air was used to dry the samples, followed by a glycerol painting and cured 20 minutes to complete the polymerization by ultraviolet light (Creality UW-01, United Kingdom). The support structures were removed using a low-speed handpiece. Subsequently, the specimens were polished using 120 grit sand paper (Norton) and then washed with distilled water. All specimens were measured with digital Vernier (China) to ensure the dimension of the specimens. The samples were stored in distilled water for 24 hours at 37 °C prior to the testing [19].

Characterization of GSSPs within the 3D-printed denture base resin

Scanning electron microscopy (SEM)

Three square specimens (n=1) for control, 5 wt% and 7 wt% GSSPS were used to make SEM for surface topography by cutting small pieces from the samples surface and then coated them with gold before scanning them with the (Philips Quanta250 electron microscope).

Atomic force microscopy (AFM)

Three square specimens(n=1) for control, 5 wt% and 7 wt% GSSPS were used in this technique to identify surface texture and average thickness using AFM (coreAFM 2023, Nanosurf, Liestal Switzerland)

Energy- dispersive X- ray spectroscopy (EDX)

Three square specimens (n=1) for control, 5wt% and 7wt% GSSPS, EDX is a nondestructive technique used with SEM to give quantitative and qualitative analysis; it was were used to identify the elements and determine the percentages of them without cause any damage to the sample (EDX-8000 shimadzu, Japan) was used.

Candida albicans adhesion test

Isolation of Candida albicans

Candida albicans were collected from the mouth of patient suffering from DS in Baghdad Teaching Hospital using a sterile cotton. Subsequently, it was inoculated in sabouraud dextrose agar (SDA) [20], under aerobic conditions at 37 °C for 24 hours. After that, it was stored at 4°C in between experiments [21].

Identification of Candida albicans

Morphological assessment:

At 37 °C after 48 hours, a creamy, smoothy and pasty *Candida albicans* colonies developed on SDA [21].

Microscopical assessment:

A glass slide was used to blend a drop of normal saline with a small amount of a single isolated colony to produce an evenly distributed solution. After that the slide was left at room temperature for drying. Then, it was passed over a Bunsen flame several times, followed by staining using Gramme's Method. Light microscope was used to examine the prepared specimen, and the *Candida albicans species* appeared oval or round in morphology with longitudinal hyphae [22].

Germ tube formation:

To create the solution, an individual lope inoculum of cell yeast was obtained and blended with serum (0.5 ml), a drop from this suspension was placed on a slide and examined under a light microscope to check for the formation of germ tubes [23].

Biochemical identification

The VITEK 2 device was used to identify the unknown microorganisms. Before the testing, the suspension was prepared by turbidity sterile saline with 2.0 McFarland standard, which was determined by DensiChek instrument. The VITEK ID-YST card was filled automatically by this suspension, then sealed and incubated for 18 hours at 35.5°C in the VITEK 2 device. At every fifteen minutes, the optical density (OD) reading was taken automatically [24].

Testing procedure of Candida albicans adherence

The disks were sterilized using autoclave at 15 psi, 121°C for 15 minutes. Then, Candida albicans culture were exposed to the disk specimens which equivalent to 0.5 McFarland standards for 24 hours. After removal, they were washed with phosphate-buffered saline to remove the unattached candida, and then, they were dried with crystal violet for 20 minutes. The disks were placed separately for 3 minutes in 3 ml of ethanol alcohol (96%) and vibration by vortex. This solution was examined by verifying the optical density with spectrophotometer under 540 w [25].

Surface roughness

The surface roughness test was examined using a portable roughness tester (TR 220, Beijing, Time High Technology Ltd., China). The mean reading for three measurements was taken for each sample in various

directions to calculate the average surface roughness. The stylus of the profilometer was passed over 11 mm in length to record the surface irregularities [26,27].

Statistical Analysis

Descriptive statistics were used for the Candida albicans adherence and surface roughness tests that involve the mean, standard deviation, and confidence interval for the mean. The one-way ANOVA parametric test was utilized as the data was normally distributed and after running a Brown-forsythe test, Games-Howell's and Bonferroni multiple comparisons test was used to evaluate the differences among the groups. Statistical significance was at P value 0.05. All the collected data were analyzed using the GraphPad prism 9 statistics software.

Materials and Methods

SEM results

SEM images showed the presence of scattered pores in the control sample (Fig. 1A, 1B) which reduced when GSSPs were added (Fig. 1D, 1F). These made the surface of the 3D-printed denture base were more regular and compact compared with control group. In addition, the presence of GSSPs filled the voids presence in 3D-printed denture base, which eventually making the surface smother (Fig. 1C, 1E). The GSSPs of appeared evenly distributed in 3D- printed denture base resin without obvious particles agglomeration (Fig. 1D, 1F).

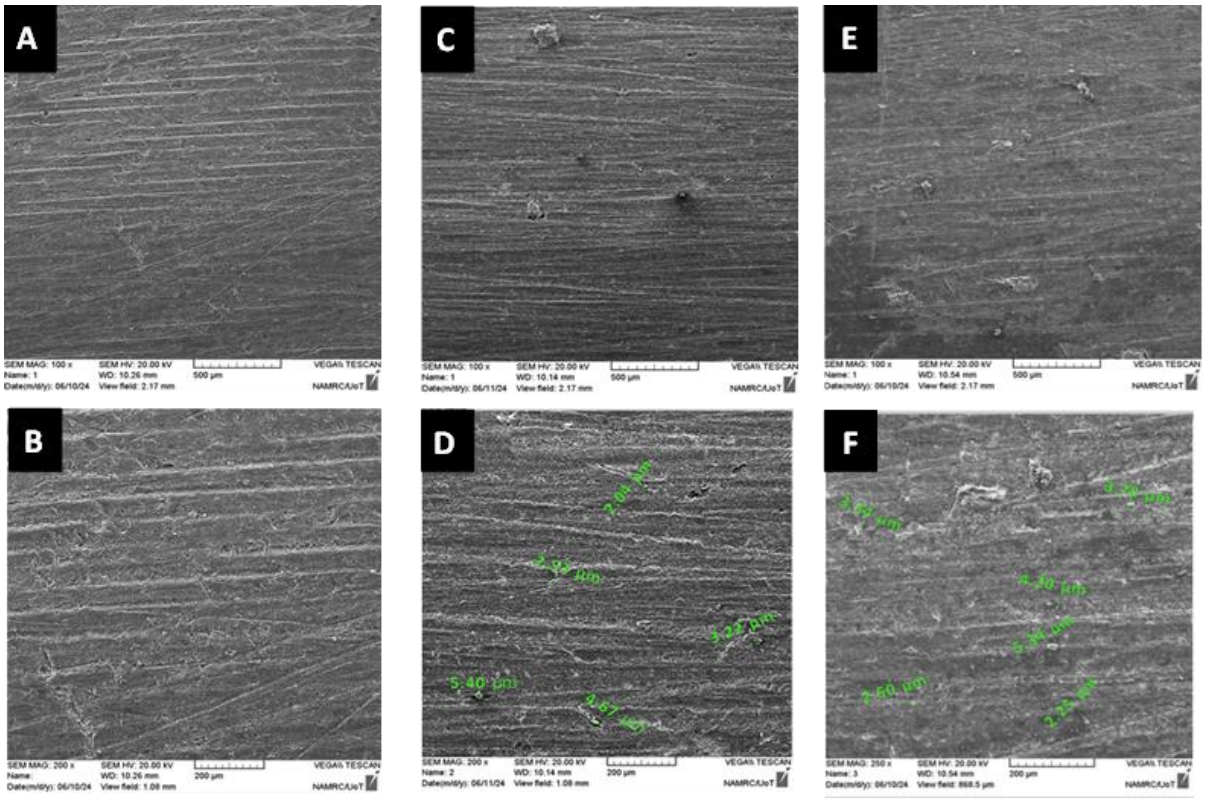

Figure 1: Scanning electron microscopy images (A) for control sample in MAG 100 and (B) MAG 200, (C) for 5 wt% sample in MAG 100 and (D) MAG 200, (E) for 7 wt% sample in MAG 100 and (F) MAG 200.

AFM results

Fig. 2A and Table 1 showed the results obtained from the AFM for the control group. Sa (arithmetic mean height) refers to the essential measurement of surface roughness, as it provides a general idea of the extent of the height and depression of the terrain on the surface of the sample. It was observed that its value equal to 222.7 nm, which indicates that the surface has a very high roughness. Sz (Maximum height) represents the difference between the highest peak and lowest valley on the surface and was used to determine the maximum range of elevations and depressions. Sz value recorded was 1506 nm, indicating that the surface has large peaks and valleys. Ssk (Skewness) evaluates the uniformity of the height distribution. If he found that its value was close to zero, the distribution is balanced between peaks and valleys. Sku (Kurtosis) measures the sharpness of peaks and troughs. A Sku value close to 3 indicates that the surface has a relatively even distribution between peaks and valleys. The results of the AFM test for the 5% and 7% samples are showed a clear difference in values, as shown in Figs. 4B and 4C and Table 1.

Table 1: The statistical roughness coefficients of control, 5% and 7% GSSPs samples

Height Parameters	Control group value	5% by weight group value	7% by weight group value
S_q (Root-mean-square height)	255.1 nm	277.5 nm	207.1 nm
S_{sk} (Skewness)	0.0343	-0.1392	-0.2868
S_{ku} (Kurtosis)	2.976	2.932	3.039
S_p (Maximum peak height)	800 nm	835.3 nm	608 nm
S_v (Maximum pit depth)	706 nm	999.9 nm	743 nm
S_z (Maximum height)	1506 nm	1835 nm	1351 nm
S_a (Arithmetic mean height)	222.7 nm	180 nm	166.3 nm

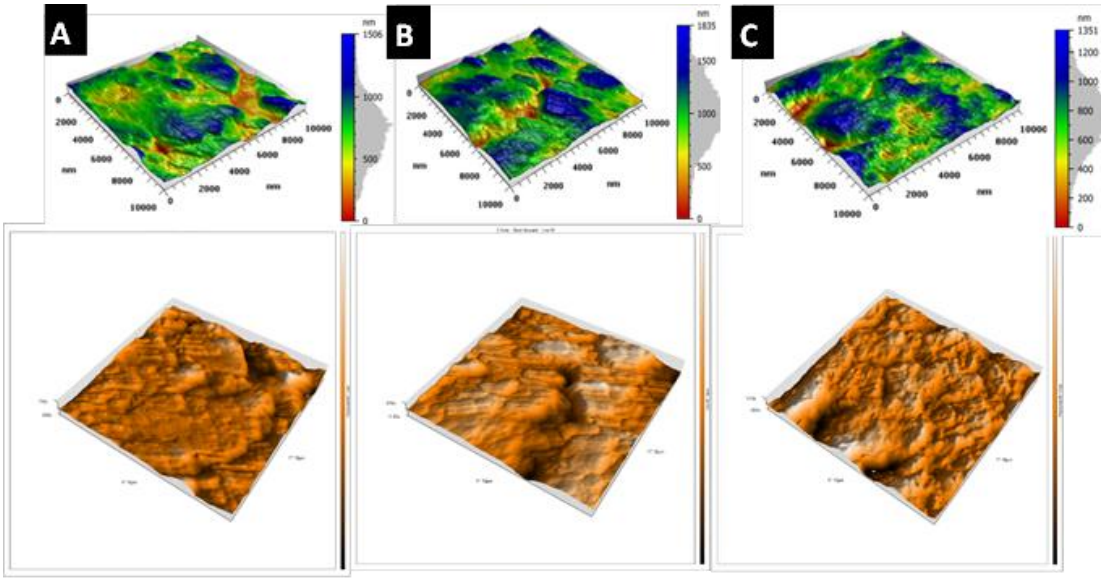

Figure 2: (A) 3D topographic analysis using AFM of the control sample. (B) 3D topographic analysis using AFM of the 5% sample. (C) 3D topographic analysis using AFM of the 7%. Blue color indicated valley's depth while red color indicated the peak's height, the colors between them (from up to the down) in this parameter showed surface roughness

Energy- dispersive X- ray spectroscopy result

Fig. 3A showed EDX results for 3D printed denture base without addition of GSSPs. Formation of FeKa in 5 wt% and CeLg3 in 7 wt% indicates the presence of new elements from GSSPS (figs. 2B and 2C). In addition, changes in concentrations and intensity of elements indicated the presence of grapefruit skin seed particles throughout the groups (Table 2). Such intensity of SiKa in control, 5 wt% and 7 wt% which was equal to 4.0624, 3.516 and 7.6150 respectively and BaLa which was equivalent to 9.6157, 24.2190 and 9.8782 for control, 5 wt% and 7 wt% respectively.

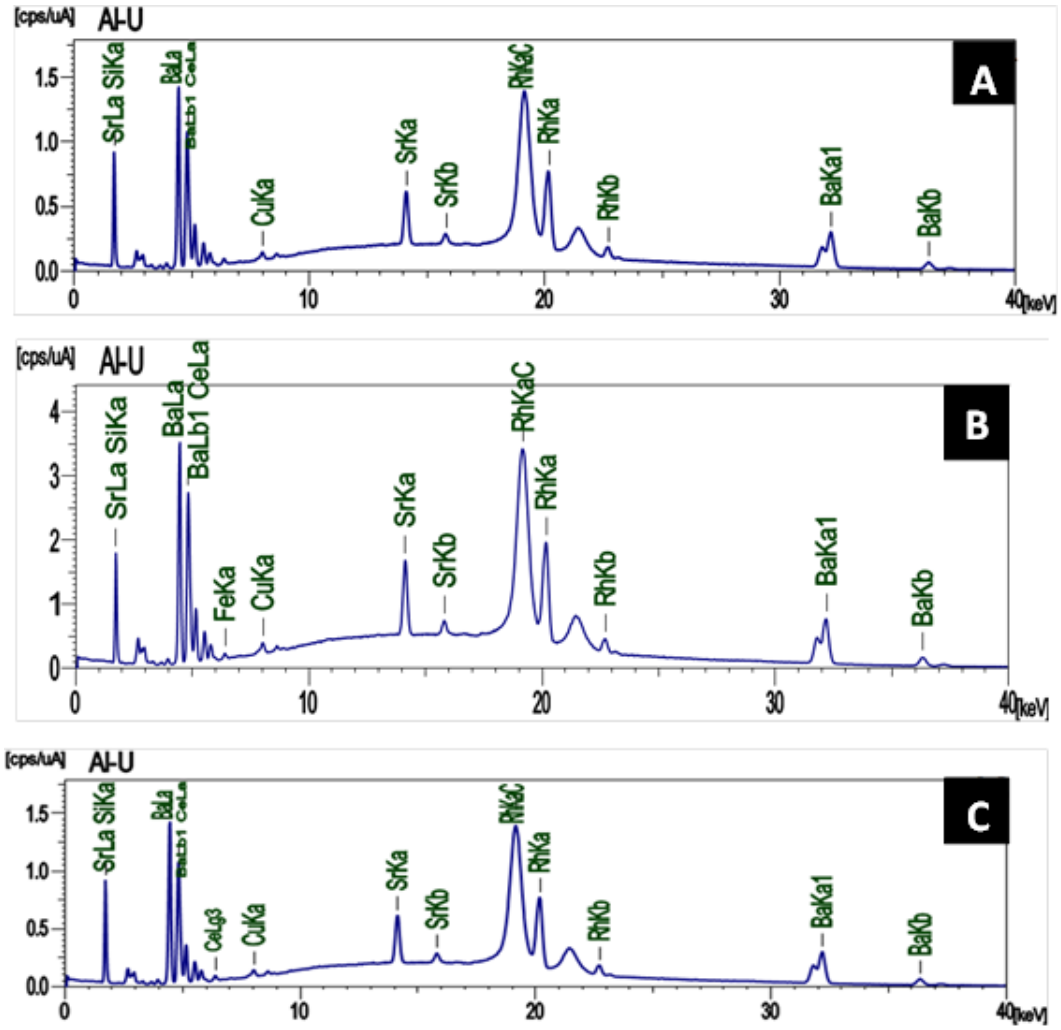

Figure 3: EDX spectroscopy for (A) control sample, (B) 5% GSSPs samples, (C) 7% GSSPs samples.

Candida albicans adherence test

The results shown that the highest mean value was 0.0945 CFU/mL for the control group, which decreased to 0.0261 CFU/mL for the 7 wt% group, which represents the lowest mean value with standard deviation of 0.003064 CFU/mL and 0.004122 CFU/mL respectively (confidence interval at 95%). The one-way ANOVA table represent a significant difference ($P < 0.0001$) on the candida adherence on denture base added with different percentages of GSSPs as presented in Table 3. Games-Howell's multiple comparisons test was used to evaluate the differences between group, there were significant difference between all groups as shown in fig. 4.

Regretfully, despite the clear and precise instruction demonstrating by the orthodontists to patients and their career, the orthodontic appliance breakage (represented by bracket failure) still representing the most frequently recorded orthodontic problem as can be seen in figure 4 [23,27-29]. In another study, retightening or replacement of the archwires were also reported as an orthodontic emergency in with a percentage of 13% of the archwire problems [30,31].

Table 2: Quantitative result for (A) control sample, (B) 5% GSSPs samples, (C) 7% GSSPs samples.

(A) Results for control sample						
Analyte	**Result**		**Std. Dev.**	**Calc. Proc**	**Line**	**Intensity**
Si	79.997	%	[0.768]	Quan-FP	SiKa	4.0624
Ba	17.617	%	[0.116]	Quan-FP	BaLa	9.6157
Ce	1.451	%	[0.010]	Quan-FP	CeLa	1.0662
K	0.401	%	[0.034]	Quan-FP	K Ka	0.1566
Sr	0.212	%	[0.003]	Quan-FP	SrKa	4.6735
Ca	0.176	%	[0.017]	Quan-FP	CaKa	0.1271
Cu	0.066	%	[0.004]	Quan-FP	CuKa	0.5244
Ag	0.049	%	[0.007]	Quan-FP	AgKa	0.3070
Zn	0.030	%	[0.003]	Quan-FP	ZnKa	0.2808
(B) Result for 5 wt% sample						
Si	75.713	%	[0.829]	Quan-FP	SiKa	7.6150
Ba	20.933	%	[0.140]	Quan-FP	BaLa	24.2190
Ce	2.240	%	[0.015]	Quan-FP	CeLa	3.4798
K	0.375	%	[0.039]	Quan-FP	K Ka	0.3160
Sr	0.304	%	[0.004]	Quan-FP	SrKa	12.9293
Ca	0.132	%	[0.019]	Quan-FP	CaKa	0.2046
Ag	0.101	%	[0.010]	Quan-FP	AgKa	1.2092
Cu	0.094	%	[0.005]	Quan-FP	CuKa	1.4623
Fe	0.077	%	[0.007]	Quan-FP	FeKa	0.6245
Zn	0.033	%	[0.004]	Quan-FP	ZnKa	0.6033
(C) Result for 7wt% sample						
Si	78.435	%	[0.790]	Quan-FP	SiKa	3.5716
Ba	19.351	%	[0.127]	Quan-FP	BaLa	9.8782
Ce	1.858	%	[0.013]	Quan-FP	CeLa	1.2753
Sr	0.253	%	[0.003]	Quan-FP	SrKa	4.9606
Cu	0.071	%	[0.004]	Quan-FP	CuKa	0.5019
Zn	0.032	%	[0.004]	Quan-FP	ZnKa	0.2703

Table 3: Descriptive statistics and one-way ANOVA table of Candida albicans adherence test.

	Control	**5%**	**7%**	**F**	**P value**
Number of values	10	10	10	1224	P<0.0001
Mean	0.094	0.051	0.026		
Std. Deviation	0.003	0.002	0.004		
Minimum	0.090	0.049	0.021		
Maximum	0.099	0.054	0.033		
Lower 95% CI of mean	0.092	0.050	0.023		
Upper 95% CI of mean	0.097	0.053	0.029		

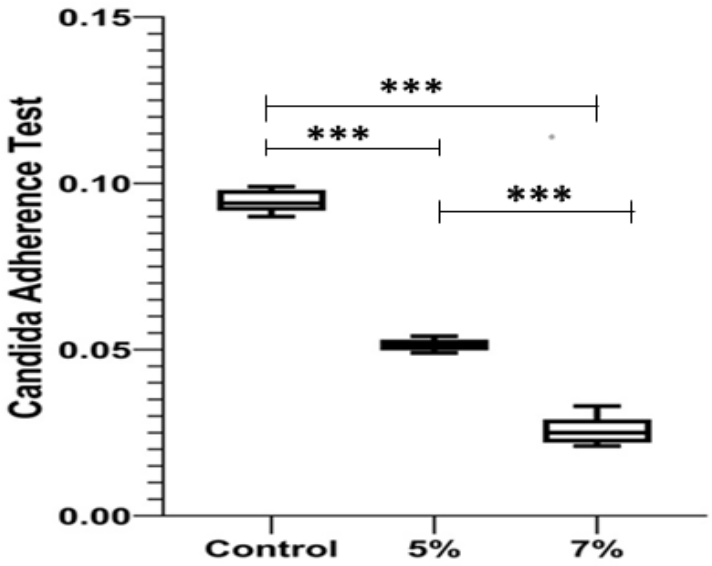

Figure 4: Box plot for Candida albicans adherence test comparing the control group with 5% and7% by weight groups. *** refers to a P value as <0.0001

Surface roughness

The highest mean value was 1.677 μm for the control group, which decreased to 0.8835μm for the 7 wt% group, which represents the lowest mean value with standard deviation 0.1937 μm and 0.1981 μm respectively (confidence interval at 95%). The one-way ANOVA table represented a significant difference P < 0.0001 in the surface roughness as shown in Table 4. Games-Howell's multiple comparisons test was used to evaluate the differences between group, there were significant difference between all groups as shown in fig 5.

Table 4: Descriptive statistics and one-way ANOVA table for surface roughness test.

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	Control	5%	7%	F	P value
Number of values	10	10	10		
Mean	1.68	1.09	0.88		
Std. Deviation	0.19	0.15	0.20		
Lower 95% CI of mean	1.54	0.98	0.74	51.38	P<0.0001
Upper 95% CI of mean	1.81	1.19	1.02		
Minimum	1.43	0.84	0.68		
Maximum	1.98	1.27	1.31		

Discussion

Addition of GSSPs to 3D-printed denture base resin have a significant effect on the reduction of *Candida albicans* and surface roughness, therefore, the null hypothesis was rejected. 3D -printed denture base resin is a new material, so there are inadequate studies assessing its mechanical, biological, and physical properties.

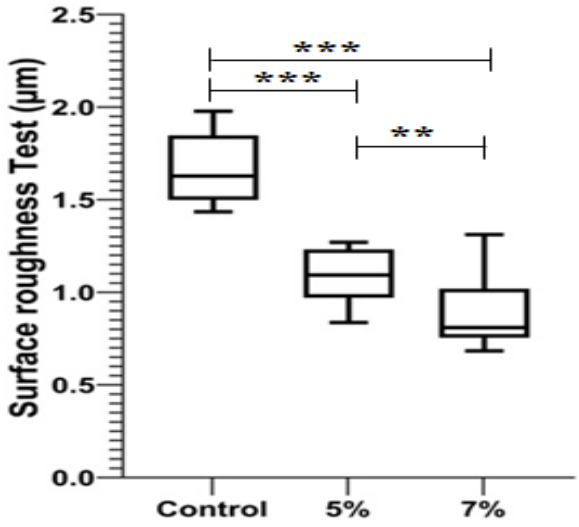

Figure 5: Box plot for surface roughness test comparing the control group with 5% and7% by weight groups. *** refers to a P value as <0.0001, ** refers to a P value as 0.0532

Because of that, in this article, direct comparison with 3D-printed denture base resins is limited and the interpretations were compared to studies other closely related materials like PMMA. The utilization of 3D printing denture base resin increasing because of its accurate details and time saving, but they are some disadvantages and challenges compared to heat cured polymerized resin [1]. With the purpose of improving oral hygiene for patients wearing dentures, many researchers have indicated an addition of antifungal agents to acrylic resin denture base, which serve as an effective method to control oral infection [28]. *Candida albicans* adherence to the denture fitting surface which in intimate contact with the oral tissue causing the development of DS especially in patients who wear dentures continuously particularly through their sleep at night [29]. The development of DS is particularly more eminent in immunocompromised and elderly patients. This was due to the microbiota shifts towards favoring more *Candida spp*., in turn causing DS [30]. The goal of adding GSSPs is to eliminate pathogens that cause DS as they exhibited fungicidal activity against *Candida albicans*.

In this study Spectrophotometer device was chosen to measure the optical density and to determine the amount of *Candida albicans*, and the results revealed there were significant decreases in the adherence of *Candida albicans* to 3D-printed denture base resin reinforced with GSSPs when compared the control group. So, the percentage of GSSPs inversely proportional with *Candida albicans* adherence. Because GSSPs contain flavones and isoflavones which work as efflux pump inhibitor and flavanols which attack cell wall and cause damage, also presence of phenolics making an alteration in permeability of cell membrane because of their lipophilic nature [31].

This result is in agreement with Nawasrah et al., who found that the addition of Henna to PMMA contributed to reduction in the adhesion of *Candida albicans* [2]. At the same time, it also affecting the surface roughness with added Henna caused increasing surface roughness. This may be return to the loose attachment of Henna from acrylic which makes it easy to be removed through finishing [32]. However, these results disagree with our current study which found the addition of GSSPs to 3D printed denture base resin showed a statistically significant decreased in surface roughness. This may be attributed to inclusion of particles between the resin matrix to fill the microgaps which decreased the irregularities and voids found on the surface samples, as seen in SEM images.

Furthermore, profilometer is a good device to measure the surface roughness of restorative materials, which provides measurements that can be compared and evaluated statistically [26]. Increase surface roughness

encourages adhesion and accumulation of plaque bacteria, staining, and facilitate *Candida albicans*, which has a negative effect on the aesthetics of dentures and promote DS due to that, it's very important to enhance the material and polish the surface to have a smooth surface and limit the development of plaque[28,30].

In this study Profilometer exhibit high mean value for surface roughness for control group (1.677 μm), as the concentration of GSSPs increased the mean value decreased to 1.089 μm and 0.8835 μm for 5 wt% and 7 wt% respectively.

This current study was in contrary with Fatalla [33] who found that there is no significant difference after addition of Polyester micro-particle powder to PMMA, and Hazim et al., who found there was no significant difference after adding Tellurium oxide micro-particles to PMMA[34].
Finally, 3D-printed denture base resin is new material and there are no previous studies about also adding GSSPs to 3D printed denture base resin or similar materials. So, caution should be taken when interpreting the results as it was affected by the size of GSSPs and type of equipment used. In addition, select suitable concentration of GSSPs is very important to obtain good results.

Conclusion

Within the limitation of this study, addition of GSSPs to 3D printed denture base resin has a significant effect on decreasing the adhesion of the *Candida albicans* and the surface roughness. The highest values of candida adhesion and surface roughness were observed at the control group. When the concentration of GSSPs increased, the values of candida adhesion and surface roughness were decreased, the lowest values were observed at 7% group this leads to decrease the opportunity to present denture stomatitis.

Conflict of interest

The authors report no conflicts of interest.

Author contributions

All authors contributed in designing the study, collecting the data, interpreting the results, writing up the draft of the manuscript, reviewing and finalizing the manuscript.

Acknowledgement and funding

Self- funded research. The authors would like to sincerely thank University of Baghdad for its continued support.

Informed consent

None

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توضيح تأثير دمج جزيئات بذور قشر الجريب فروت في راتنج قاعدة الأسنان الصناعية المطبوعة ثلاثية الأبعاد على التصاق المبيضات البيضاء وخشونة السطح.

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المستخلص:

الأهداف :التهاب الفم الناتج عن أطقم الأسنان هو مشكلة شائعة لدى المرضى الذين يرتدون أطقم الأسنان. لتحسين صحة الناس ومنع انتشار العدوى، من الضروري اختيار الحشوات المناسبة في مادة قاعدة أطقم الأسنان، والتي لا ينبغي أن تكون فعالة من حيث المتانة والخصائص الميكانيكية فحسب، بل وأيضًا لا تمارس تأثيرات غير مواتية على الأنسجة الفموية. كان الغرض من هذه الدراسة تحديد آثار إضافة جزيئات بذور قشر الجريب فروت (GSSPS) بتركيزات مختلفة على التصاق المبيضات البيضاء وخشونة سطح راتنج قاعدة أطقم الأسنان المطبوعة ثلاثية الأبعاد. المواد والطرق: تم طباعة ستين عينة رقميًا مع توزيع 30 قرصًا لاختبار التصاق المبيضات البيضاء و 30 عينة أخرى لاختبار خشونة السطح. تم تقسيم العينات في كل اختبار إلى ثلاث مجموعات (ن = 10) لكل نسبة 0 GSSPS٪ بالوزن، 5٪ بالوزن، و 7٪ بالوزن). تم اختيار مطياف ضوئي لتحديد الكثافة البصرية لفطريات المبيضات البيضاء وتم استخدام مقياس البروفيليمتر لقياس خشونة السطح. النتائج: كان هناك انخفاض كبير في كل من التصاق فطريات المبيضات البيضاء وخشونة سطح راتنج قاعدة طقم الأسنان المطبوع ثلاثي الأبعاد. بعد إضافة GSSPS أثبت المجهر الإلكتروني الماسح وجود جزيئات GSSPS موزعة بالتساوي، وأثبت المجهر الذري للقوة انخفاض خشونة السطح عندما زاد تركيز GSSPS وأظهر مطياف

الأشعة السينية المشتتة للطاقة (EDX) وجود عناصر جديدة. الاستنتاج: ضمن حدود هذه الدراسة، فإن إضافة GSSPs إلى راتنج قاعدة طقم الأسنان المطبوع ثلاثي الأبعاد له تأثير كبير على تقليل التصاق فطريات المبيضات البيضاء وخشونة السطح، مما يؤدي إلى تقليل فرصة حدوث التهاب الفم الناتج عن طقم الأسنان.