https://africanjournalofbiomedicalresearch.com/index.php/AJBR
Afr. J. Biomed. Res. Vol. 28(2s) (February 2025); 705-711
Research Article

The Influence of Tea Tree Oil Incorporation on Candida Albicans Adhesion, Surface Hardness and Transverse Flexural Strength of High Impact Acrylic Denture Base Material

Mohammed Hameed. Hassan[1*], Aseel Mohammed Al-Khafaji[2]

[1*,2]*Department of Prosthodontics, College of Dentistry, University of Baghdad, Baghdad, Iraq.*

***Corresponding Author:** Mohammed Hameed. Hassan
**Department of Prosthodontics, College of Dentistry, University of Baghdad, Baghdad, Iraq,*
E-mail: Mohammed.Hasan2401m@codental.uobaghdad.edu.iq

ABSTRACT
Background: Cleaning dentures was an important procedure in order to decrease the spread of infections, improve patient health, increase the longevity of dentures, and general quality of life; as a result, it was essential to select the right cleanser that, in addition to was efficient and, even after prolonged use, had no negative effects on the denture base resin's qualities. Tea Tree oil (TTO) is the essential oil which has antifungal, antioxidant and antibacterial properties. this study aimed to evaluate the effect TTO on *Candida albicans* adhesion, surface hardness and transversal flexural strength property of high impact acrylic denture material after incorporation with TTO at different concentration. Methods: A total of 80 specimens of high impact acrylic incorporation with different concentration of TTO were used for *Candida albicans* adherence, surface hardness and transverse flexural strength tests. They are divided into three groups, 30 specimens for *Candida albicans* adherence test, 25 specimens for surface hardness and 25 specimens for transverse flexural test. After that categorized into subgroups: (control, 6% tea tree oil, 9% ,12% and 15%) of tea tree oil incorporation with high impact acrylic, 5 specimens for each group and 5 specimens of 1.4% nystatin were used as positive control to decide which concentration of tea tree oil is to be used in *Candida albicans* adherence test. A spectrophotometer was used to measure optical density in order to identify *Candida* albicans adhesion test. to measure transvers flexural strength Three-points bending test was carried out utilizing an Instron universal testing apparatus and to measure surface hardness used shore D Indenter. At a significance level of 0.05, the Shapiro-Wilk test, one-way ANOVA, and post hoc Tukey's test were used to analyze the data. Results. TTO had a statistically highly significant effect on *Candida albicans* adherence with different concentration of TTO (6%,9%,12% and 15%) when compared with negative control. For positive control (1.4%nystatin) showed a statistically non-significant difference ($P > 0.05$) between (6% and 9% TTO) and positive control. For transverse strength have a significant effect ($P < 0.05$) between control and (6% ,9% and 15%), with the exception of the non-significant difference between 12% and control. However, the surface hardness test found a difference between 6% and 9% and control and 6% TTO that was statistically non-significant ($P > 0.05$). with the exception of the non-significant difference between 12% and control. However, the surface hardness test found that was statistically non-significant ($P > 0.05$) between) 6% and 9%) and (control and 6% TTO).

KEYWORDS: Tea Tree oil, High-impact Polymer, Polymethylmethacrylate, Antifungal Efficacy, Surface hardness, Transverse flexural strength

***Author of Correspondence E-mail:** Mohammed.Hasan2401m@codental.uobaghdad.edu.iq*

Received 20/01/2025, Acceptance 07/02/2025
DOI: https://doi.org/10.53555/AJBR.v28i2S.6767

Introduction

Polymethylmethacrylate (PMMA) is the recommended polymer for dentures because of its low toxicity, low water absorption, and aesthetically pleasing appearance(1). To increase strength, a rubber compound was added to a PMMA polymer to create high-impact strength acrylics(2) .

About 68% of acrylic resin dentures break within a few years This happens when the denture accidentally falls on a hard surface or fractures from the intense strain of chewing(3). Butadiene-styrene rubber particles are added to the powder of these denture foundation materials. A methacrylate group is grafted onto the rubber to create a covalent link between the particles and the polymer network(4) .

People who wear dentures are at a higher risk of developing denture stomatitis, which is primarily defined by the presence of *Candida albicans* and can lead to the development of candidiasis, PMMA permits the growth of microorganisms and food accumulation. Therefore, cleansing dentures is necessary(5). Traditional methods of treating denture stomatitis involve administering antifungal medications, of which nystatin was the most commonly used(6).

Some oils that Extract from medicinal plants are utilized in biomaterials as a natural substitute that works well and has strong antifungal and antibacterial qualities(7). Consequently, it offers a viable alternative to antifungal drugs and addresses their limitations, primarily regarding penetration and chemical interaction inside biofilms, including the extracellular polymeric substance(8) .The primary benefits of using natural plant extracts, aside from their financial worth, were their safety, biocompatibility, and lack of adverse effects (8).Research is being done on the therapeutic use of essential oils. Tea tree oil (TTO) is utilized in PMMA because it has antibacterial, antifungal, and antioxidant properties(9, 10).

TTO was extracted from the leaves of Melaleuca alternifolia, a native plant in Australia TTO was composed of a variety of compounds, primarily hydrocarbons of monoterpene and sesquiterpene and their alcohols. according to several investigations TTO offered antiseptic and antibacterial properties(11). The current study aimed to investigate the *Candida albicans* adhesio*n*, Surface hardness and transversal flexural strength of high-impact polymethylmethacrylate acrylic after incorporation it with TTO.

Materials and Methods

In this study, 80 specimens of the high-impact PMMA (Veracril® / Opti-cryl high impact, Newstetic, Colombia) were made. The obtained specimens were categorized into three groups depending on the type of test utilized: *C. albicans adherence*, surface hardness and transversal flexural strength. Each test consisted of 25 specimens except *C. albicans adhesion* tes*t* have 30 specimens (5 specimen for nystatin as appositive control, then separated into five categories based on the different concentration of T.T.O used.

The polymer, monomer, and T.T.O amounts have been determined using a digital balance of 0.001 g to weigh powder and monomer and a micropipette to measure liquids. Afterwards, the polymer and monomer were blended using a powder-to-liquid ratio of (2:1) by weight according to manufacturer's instructions of high impact acrylic(12) (13)

On the other hand, the experiment groups were manufactured using the same acrylic resin and combined with differing concentrations of 100% pure Tea Tree oil (NOW®/U. S. A) 6% 9%,12% and 15%. Using a tiny electric hand mixer, the required amount of TTO was measured in a dry, clean glass beaker, with a micropipette, reduced from the volume of the monomer, and then combined with the monomer for one minute. After adding this mixture to the acrylic powder, which was thoroughly mixed, the curing process suggested by the manufacture(12).

Packing

The polymer and monomer were mixed following the company's instructions. Once the resin became plastic, it was placed into the mold within the lower half of the flask, inside a hydraulic press, the whole object was compressed, including its upper portion and a cover secured over the bottom half. An initial press was made to achieve a uniform distribution of the substance. After that, a flask was taken out of the press to enable the removal of excess material. Then, retake in hydraulic press and pressure gradually increased until it reached 100 kg/cm2.The flask was securely closed once more, and a final pressing is performed at 150 kg/cm2 pressure to guarantee that the vertical dimension remains unchanged according to manufacturer instructions. The flask was taken out of the press, put in the clamp, and closed firmly.

Curing of high-impact PMMA

According to the company's recommendations, water was used to cure the flask and clamp. At first the cold water was used to immerse the flask, and the water's temperature was increased to 74 ˚C and maintained at (74 ˚C) for 90 minutes.

After that, the water temperature was raised to (100 ˚C) for (30 minutes). The flask was then removed from the water bath and placed in the air at (23 ˚C) for (30 minutes). Finally, the flask was immersed in cold water at (23 ˚C) for (15 minutes)(12).

Classifying of specimens

80 specimens in all were made and divided up into three sets.

FIRST SET: *Candida albicans adhesion* test: Thirty specimens in all have been divided up into six groups, with five specimens in each group.

1. Control: Preserved in distilled water (negative control).
2. 1.4%nystatin: incorporation with high impact acrylic (positive control for antifungal test).
3. 6% TTO: Incorporation with high impact acrylic.
4. 9% TTO: Incorporation with high impact acrylic.
5. 12%TTO: Incorporation with high impact acrylic.
6. 15%TTO: Incorporation with high impact acrylic.

SECOND SET: Transverse flexural test: Five groups of five specimens each were developed from a total of 25 specimens.
1. Control: Preserved in distilled water (negative control).
2. 6% TTO: Incorporation with high impact acrylic.
3. 9% TTO: Incorporation with high impact acrylic.
4. 12%TTO: Incorporation with high impact acrylic.
5. 15%TTO: Incorporation with high impact acrylic.

THIRD SET: Surface hardness test: Five groups of five specimens each were developed from a total of 25 specimens**.**
1. Control: Preserved in distilled water (negative control).
2. 6% TTO: Incorporation with high impact acrylic.
3. 9% TTO: Incorporation with high impact acrylic.
4. 12%TTO: Incorporation with high impact acrylic.
5. 15%TTO: Incorporation with high impact acrylic.

General test specimen preparation
For Candida. albicans adhesion test: Specimen's Preparation: The plastic disks were constructed with dimensions of 10 mm in diameter and 2 mm in thickness. An autoclave was used to sterilize each specimen for 15 minutes at 15 psi and 121 °C(14). Ten milliliters of sterile tryptic soy broth (TSB) containing Candida albicans was used to infect each specimen separately. The samples were then cultured for twenty-four hours at 37 °C in an aerobic environment. The

Analysis of statistics

Transverse flexural readings, individual surface hardness readings, and adhesion measurements for Candida albicans were calculated and assembled. SPSS (IBM SPSS version 20) was used to determine each group's mean and standard deviation. One-way ANOVA and Tukey HSD were used for evaluating the outcomes of each group. The significance level has been set at $P < 0.05$.

tubes' turbidity was modified on day two to match McFarland tube No. 5, which has a turbidity of 107 organisms/ml (14). Following a saline wash, the samples had been immersed in a solution of denture cleanser(15) .In accordance with the specimen groups for 10 minutes. After that, the samples were cleaned and stained with crystal violet. The adherent Candida albicans was then removed by gently rinsing them with sterile DW and submerging them for three minutes in three milliliters of 96% ethanol. The adhering Candida albicans was analyzed at 540 nm using a spectrophotometer adjusted to 0.5(16) .

For the transverse strength: dimensions of the specimen were (65 mm x 10 mm x 2.5 mm) according to (ADA,1999)(17) .Plastic patterns with the precise dimensions were made using a laser-cutting machine. Following the manufacturer's instructions (water/powder ratio of 25 mL/100 g), the type 4 dental stone (Zhermack®, Italy) was prepared and put into the flask's lower section after it had been painted with separating media (IZO-SOL, Zhermack, Italy). The mold and plastic design were then inserted into the stone and let to harden. Following the stone setting, the whole surface was painted with a separating substance, including, stone, and plastic designs. Following that, the flask's top half was placed over its bottom half, filled with the new stone mixture, covered, and left to solidify. The two portions of the flask were then separated, and the patterns were removed.

For surface hardness: According to ADA specification No.12 (ISO 1567, 1999), the specimens used for the surface hardness test had the following the measurements for length, width, and thickness are 65 x 10 x 2.5 mm, appropriately. Before the test was established, the specimens were stored for 48 hours at 37°C in distilled water. The surface hardness test was performed using a shore D hardness durometer.as.

Results.

Table 1 reports the Candida albicans adhesion test findings for each group. Examining the stained specimens from each group, it was discovered that the group control had the highest mean value (0.2112), followed by the experimental groups while the group (15%) had the lowest mean value (0.0654).

Table 1: Descriptive statistics for candida adherence test

Sample	N	Mean	Std. Deviation	Std. Error	Minimum	Maximum
control	5	.21700	.004637	.002074	.211	.223
nystatin1.4%	5	.08700	.002915	.001304	.083	.090
6%tea tree oil	5	.08380	.002775	.001241	.081	.088
9%tea tree oil	5	.08280	.003114	.001393	.080	.088
12%tea tree oil	5	.07420	.002950	.001319	.072	.079
15%tea tree oil	5	.06540	.003578	.001600	.060	.069
Total	30	.10170	.053040	.009684	.060	.223

One-way ANOVA showed highly significant result when comparing between groups (P < 0.05) as demonstrated in **Table 2**:

Table 2: ANOVA test of candida adherence test

	Sum of Squares	D f	Mean Square	F	P-value
Between Groups	.081	5	.016	1416.117	.000
Within Groups	.000	24	.000		
Total	.082	29			

* =Significant at p<0.05, ^=not significant at p>0.05

*. The mean difference is significant at the 0.05 level.

Table 3: Multiple Comparisons of candida adherence between groups **Tukey's Honestly** showed had non-significant differences (P > 0.05) between (nystatin1.4%, 6% TTO and9% TTO) and (6% and 9%TTO) whereas other groups had significant differences (P < 0.05).

Table 3: Significant Difference (Tukey's HSD)

(I)Group	(J)group	Mean Difference(I-J)	SE	P-value
Control	nystatin1.4%	.130000*	.002143	.000
	6%tea tree oil	.133200*	.002143	.000
	9%tea tree oil	.134200*	.002143	.000
	12%tea tree oil	.142800*	.002143	.000
	15%tea tree oil	.151600*	.002143	.000
nystatin1.4%	6%tea tree oil	.003200	.002143	.672
	9%tea tree oil	.004200	.00214	.393
	12%tea tree oil	.012800*	.002143	.000
	15%tea tree oil	.021600*	.002143	.000
6%tea tree oil	9%tea tree oil	.001000	.002143	.997
	12%tea tree oil	.009600*	.002143	.002
	15%tea tree oil	.018400*	.002143	.000
9%tea tree oil	12%tea tree oil	.008600*	.002143	.006
	15%tea tree oil	.017400*	.002143	.000
12%tea tree oil	15%tea tree oil	.008800*	.002143	.000

*. The mean difference is significant at the 0.05 level.

The results of the transverse flexural strength test for each group are reported in Table 4.

Table 4: Descriptive statistics for transvers flexural strength among groups

Sample	N	Mean	SD	SE	Minimum	Maximum
control	5	84.00	1.871	.837	82	86
6%tea tree oil	5	96.00	2.345	1.049	92	98
9%tea tree oil	5	114.20	2.280	1.020	112	117
12%tea tree oil	5	86.20	1.483	.663	84	88
15%tea tree oil	5	69.40	4.827	2.159	65	76
Total	25	89.96	15.331	3.066	65	117

Table 5: ANOVA test for transvers flexural strength .

	Sum of Squares	Df	Mean Square	F	P-value
Between Groups	5482.160	4	1370.540	172.612	.000
Within Groups	158.800	20	7.940		
Total	5640.960	24			

* =Significant at p<0.05, ^=not significant at p>0.05

One-way ANOVA test produced a highly significant result when comparing between groups (P < 0.05) Table 5.

Table 6: Multiple Comparisons of transvers flexural strength between groups **Tukey's Honestly** showed had non-significant differences (P > 0.05) between (Control, 12% TTO) whereas other groups had a significant difference (P < 0.05).

Table 6: Significant Difference (Tukey's HSD) Tukey HSD

(I)group	(J) group	Mean Difference (I-J)	Std. Error	Sig.
Control	6%tea tree oil	-12.000*	1.782	.000
	9%tea tree oil	-30.200*	1.782	.000
	12%tea tree oil	-2.200	1.782	.732
	15%tea tree oil	14.600*	1.782	.000
6%tea tree oil	9%tea tree oil	-18.200*	1.782	.000
	12%tea tree oil	9.800*	1.782	.000
	15%tea tree oil	26.600*	1.782	.000
9%tea tree oil	12%tea tree oil	28.000*	1.782	.000
	15%tea tree oil	44.800*	1.782	.000
12%tea tree oil	15%tea tree oil	16.800*	1.782	.000

The results of surface hardness test are listed in Table 7 for every group. It was discovered that the group control had the highest mean value (81.64), The experimental groups were next, and the group (15%) had the lowest mean value (63.12).

Table 7: Descriptive statistics for surface hardness test.

sample N	Mean	Std. Deviation	Std. Error	Minimum	Maximum
control	5	81.64	.757	.339	81	83
6%tea tree oil	5	78.98	.861	.385	78	80
9%tea tree oil	5	74.80	1.093	.489	73	76
12%tea tree oil	5	70.86	.723	.323	70	72
15%tea tree oil	5	63.12	2.032	.909	60	65
Total	25	73.88	6.735	1.347	60	83

One-way ANOVA showed highly significant result when comparing between groups ($P < 0.05$) as demonstrated in Table 8.

Table 8: ANOVA test for surface hardness test

	Sum of Squares	Df	Mean Square	F	P-value
Between Groups	1059.860	4	264.965	185.031	000
Within Groups	28.640	20	1.432		
Total	1088.500	24			

* =Significant at p<0.05, ^=not significant at p>0.05

Table 9: Multiple Comparisons of surface hardness test between groups Tukey's Honestly showed had non-significant differences ($P > 0.05$) between (control and 6%) and (6% and 9%TTO) whereas other groups had significant differences ($P < 0.05$).

Table 9: Significant Difference (Tukey's HSD)

(I)group	**(J) group**	**Mean Difference (I-J)**	**Std. Error**	**Sig**
control	6%tea tree oil	1.32000	.84010	.531
	9%tea tree oil	2.82000*	.84010	.023
	12%tea tree oil	6.78000*	.84010	.000
	15%tea tree pol	18.52000*	.84010	.000
6%tea tree oil	9%tea tree oil	1.50000	.84010	.409
	12%tea tree oil	5.46000*	.84010	.000
	15%tea tree pol	17.20000*	.84010	.000
9%tea tree oil	12%tea tree oil	3.96000*	.84010	.001
	15%tea tree pol	15.70000*	.84010	.000
12%tea tree oil	15%tea tree pol	11.74000*	.84010	.000

*. The mean difference is significant at the 0.05 level.

Discussion

One of the most used foundation materials for dentures is acrylic resin. However, it has a number of disadvantages, including poor mechanical properties and the adherence of *C. albicans* to denture acrylic resin materials is an essential initial step in the successful colonization and development of an infection(5, 18). Because of TTO have individual characteristics, its works well as a disinfectant, especially when applied in the right amounts(19). Tea tree oil's ability to cause damage with the permeability barrier of microbial membrane structures give it antimicrobial properties(20) .causing them to release components from their cytoplasm, considering that essential oils

could get through the cytoplasmic membrane due to their lipophilicity, Additionally, TTO and terpinen-4-ol may penetrate fungal organelle membranes and cause damage(20) .

The ongoing research showed a significant different between control and the three test (Candida *albican adherences test,* flexural strength test and hardness strength test) all test the p-value<0.05.

TTO significantly reduced C*andida albican adherences* when added to High impact acrylic resin at different concentrations (6%, 9%, 12%and 15%), Decrease in number of *Candida albican adherences* when increase concentration of TTO, the lowest mean value seen in (15%) of TTO.

Compressive, tensile, and shear strengths are combined to form the transverse (flexural) strength, which is a direct indicator of the material's stiffness and fracture resistance. Comparing the transverse strength of high impact acrylic resin (control) and TTO additive groups at four different concentrations (6%, 9%, 12%, and 15%) was another goal of the study. The results demonstrated that the TTO group's transverse strength was higher at 6% and 9% than that of the control group and the other two concentrations (12% and 15%), which might be explained by the TTO's potential elastomeric properties to PMMA (20).Another possible reason for such an effect is that the maximum saturation of the matrix formation between high impact acrylic and oil occurred at concentrations of 6% and 9%.

According to the study's findings, acrylic resin's hardness decreases with increasing TTO. concentration So, 12% and 15% of TTO has a large decrease in hardness when compare with hardness of 6% and 9% concentration which are the most appropriate used in main study. TTO coating polymer particles may be the cause of this phenomenon since it reduces the quantity of monomer to polymer conversion, leaving a significant amount of residual monomer and because a plasticizing effect, the latter negatively impacts the mechanical characteristics(21) .

Conclusion

Within the limitations of this study, we can say that the adding of TTO to High impact acrylic resin at different concentration resulted in a -significant decrease in number of *candida albican* adherence and the greatest decrease in concentration of 15%. The result of transverse flexural strength of incorporation high impact acrylic with (6% and 9% TTO) lead to increase transverse flexural strength and improved properties of high-impact acrylic, the mean of group (6% and 9%) shown in Table 4, were the other groups', suggesting a decline in the transverse flexural strength. while the surface hardness had non-significant differences (P > 0.05) between (control and 6%) and (6% and 9%TTO).

Conflict of interest

The authors of this study did not disclose any potential conflicts of interest.

References

1. Noori ZS, Al-Khafaji AM, Dabaghi. Effect of tea tree oil on candida adherence and surface roughness of heat cure acrylic resin. Journal of Baghdad College of Dentistry. 2023;35(4):46-54.
2. Meng TR Jr, Latta MA. Physical properties of four acrylic denture base resins. J Contemp Dent Pract. PMID: 16299611. 2005 Nov 15;6(4):93-100.
3. Abdul HNG, Khairi AW. The Effect of Silica Particles Derived from Rice Husk on Some Mechanical Properties of a High Impact Heat Cure Acrylic Resin. Journal of medicinal and chemical sciences**.** 2023.
4. Noori ZS, Al-Khafaji AMJJoE. Evaluation of the Effect of Tea Tree Oil Denture Cleanser on the Properties of Dental Polymers.University of Baghdad Journal of Engineering . 2024;30(11):39-49.
5. Karaagaclioglu L, Can G, Yilmaz B, et al. The adherence of Candida albicans to acrylic resin reinforced with different fibers. Journal of Materials Science. 2008;19:959-63..
6. Dias AP, Samaranayake LP, Lee MT. Miconazole lacquer in the treatment of denture stomatitis: clinical and microbiological findings in Chinese patients. Clinical oral investigations. 1997 Mar;1:47-52.
7. Hammed SS, Al-Khafaji AM. Tea Tree Oil Effect on Dimensional Change and Detail Reproduction of Addition Silicon Impression Material. Dentistry 3000. 2024 Oct 7;12(2).
8. Kanathila H, Bhat AM, Krishna PD. The effectiveness of magnesium oxide combined with tissue conditioners in inhibiting the growth of Candida albicans: An: in vitro: study. Indian Journal of Dental Research. 2011 Jul 1;22(4):613.
9. Carson CF, Riley TV. Antimicrobial activity of the essential oil of Melaleuca alternifolia. Letters in applied microbiology. 1993 Feb 1;16(2):49-55.
10. Noumi E, Snoussi M, Bakhrouf A. In vitro effect of Melaleuca alternifolia and Eucalyptus globulus essential oils on mycelia formation by oral Candida albicans strains. Afr. J. Microbiol. Res. 2010 Jun 18;4(12):1332-6.
11. Mondello F, De Bernardis F, Girolamo A, Salvatore G, Cassone A. In vitro and in vivo activity of tea tree oil against azole-susceptible and-resistant human pathogenic yeasts. Journal of Antimicrobial Chemotherapy. 2003 May 1;51(5):1223-9.
12. Al-Khafagi KS, Mahmood WS. Assessment of the surface hardness of high-impact polymethylmethacrylate following long-term dipping in clove oil solution. (Humanities, social and applied sciences) Misan Journal of Academic Studies. 2024 Dec 25;23(52):113-21.
13. Pillai CK. Regional Research Laboratory (CSIR), Thiruvananthapuram, India. Handbook of Engineering Polymeric Materials. 1997 Jul 25:411
14. Al-Shammari SS, Abdul-Ameer FM, Bairam LR, Al-Salihi Z. The influence of lemongrass essential oil addition into heat cured acrylic resin against

Candida albicans adhesion. Journal of Baghdad College of Dentistry. 2023 Sep 15;35(3):67-75.
15. Volety S, Shetty PP, Kumar K, Shetty G. Antifungal effects of herbal extracts and fluconazole on heat-polymerized acrylic denture base resin as denture cleanser: An in vitro study. The Journal of Contemporary Dental Practice. 2021 Feb 1;22:162-5.
16. Taylor R, Maryan C, Verran J. Retention of oral microorganisms on cobalt-chromium alloy and dental acrylic resin with different surface finishes. The Journal of prosthetic dentistry. 1998 Nov 1;80(5):592-7.
17. Nandal S, Ghalaut P, Shekhawat H, Gulati MS. New era in denture base resins: a review. Dental Journal of Advance Studies. 2013 Dec;1(03):136-43.
18. Dhir G, Berzins DW, Dhuru VB, Periathamby AR, Dentino A. Physical properties of denture base resins potentially resistant to Candida adhesion. Journal of Prosthodontics. 2007 Nov;16(6):465-72.
19. Yahya YK, Al-Khafaji AM. Studying the Effect of Tea Tree Oil on the Surface Hardness, and Detail Reproduction of Type III Dental Stone.Journal of natural and appliaed sceinces. 2024; 6, Vol: 99,100.
20. Cox SD, Mann CM, Markham JL, Bell HC, Gustafson JE, Warmington JR, Wyllie SG. The mode of antimicrobial action of the essential oil of Melaleuca alternifolia (tea tree oil). Journal of applied microbiology. 2000 Jan 1;88(1):170-5.
21. Faltermeier A, Rosentritt M, Müssig et al. Orthopedics D. Acrylic removable appliances: Comparative evaluation of different postpolymerization methods. American Journal of Orthodontics and Dentofacial Orthopedics. 2007;131(3):301. e16-. e22.