

Antifungal Action of Some Natural Oils on Acrylic Resin Denture Base Materials

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الخلاصة

الاهداف: تهدف الدراسة الى تقييم بعض الزيوت الطبيعية كمضادات للمبيضات البيضاء مادة الطقم الاكريليكية ، المواد وطرق البحث: المواد المستخدمة تشمل زيت عباد الشمس، زيت حبة السوداء، زيت السمسم، زيت الكتان ، وزيت الزنجبيل ، مقارنة بمحلول النستاتين. بلغ العدد الكلي للعينات (70 عينة) بابعاد (2×10×10) ملم ، نصف عدد عينات مادة الاكريلك لكل مجموعة كان ذو تصلب حراري والنصف الثاني ذو تصلب بارد، وقد غمرت في هذه المخاليل لمدة ثمان ساعات بعد ان تم تحضيرها مع خلايا المبيضات البيضاء لمدة (48) ساعة. **النتائج:** اختبرت الدراسة فاعلية هذه المخاليل على القضاء على خلايا المبيضات البيضاء مقارنة بالنستاتين و الماء المقطر، وقد تم استعمال التحليلات الإحصائية الآتية (التحليل الوصفي و تحليل التباين واختبار تي) وذلك عند مستوى معنوية (0.05)، حيث وجد فرق معنوي واضح بين الزيوت المستعملة والماء المقطر . **الاستنتاجات:** نستنتج من هذه الدراسة إن كل الزيوت الطبيعية المستعملة كمنظفات المادة قاعدة الطقم الاكريليكية ناجحة في الاختبارات التي أجريت للقضاء على خلايا المبيضات البيضاء عند التغطيس لمدة ثمان ساعات.

ABSTRACT

Aims: The study aims to evaluate the antifungal action of some natural oils (sun flower oil, sesame oil, nigella sativa, flax oil and ginger oil) in relation to nystatin suspension on acrylic resin denture base materials. **Materials and method:** The total number of specimens were seventy, they were (10×10×2mm). Half of the specimens were prepared from heat cured acrylic resin and the other half prepared from cold cured acrylic resin denture base materials, for each group they were immersed for 8hr in these oils after they had been infected with *Candida albicans* and incubated for 48hr. This study compared antifungal efficiency of sun flower oil, sesame oil, nigella sativa, flax oil and ginger oil. The statistical tests used were one way analysis of variance test, Dunnett t-test. **Results:** There were significant differences between all tested oils and distilled water in relation to antifungal action at ($P=0.05$). **Conclusions:** All the tested natural oils were effective antifungal agent.

Key words: Natural oils, *Candida albicans*, acrylic resin

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INTRODUCTION

Denture stomatitis is the most common infectious disease affecting the palatal mucosa and is highly prevalent in denture wearers, mainly characterized by the presence of *Candida albicans* ^(1,2). *Candida* species are responsible for the most frequently encountered opportunistic fungal infection ⁽³⁾. The adherent material "denture deposit" is unaesthetic in appearance, unpleasant in terms of tactile sensation, taste and malodor "halitosis" which is related to microorganisms that release volatile sulfur compound (H₂S) ⁽⁴⁾.

Traditional treatment modalities of denture stomatitis include the use of antifungal agents and modification of the prosthesis to receive a denture liner, where the most widely used antifungal agent was

nystatin^(5,6). It is more accepted to use natural denture cleanser solutions such as sunflower or sesame oil for their oil pulling action that prevent teeth decay, oral malodor, bleeding gum, dryness of throat and lips, and for strengthening of teeth, gums and jaws ⁽⁷⁾.

The use of natural products as disinfectants or denture cleansers is greatly advantageous over using systemic approach by antibiotics or local approach with synthetic products or some oral antibiotics. Al-Haroni⁽⁸⁾ showed that orally directed therapies against bacteria is superior to the use of broad spectrum antibiotics. The advantages of using natural products as denture cleansers include: safety and biocompatibility, has no chance to develop bacterial resistance ⁽⁹⁾, effective as fungicidal and

bactericidal agents, and has anti-tumor, anti-oxidant, anti-inflammatory, anti-bacterial activity, and stimulate the immune system⁽¹⁰⁻¹⁹⁾, in addition to their low cost and availability in mostly every house.

Oil pulling with sunflower and sesame oil has been used extensively for many years in India to prevent teeth decay, oral malodor, bleeding gums, dryness of throat and cracked lips, and for strengthening the teeth, gums, and jaws. They are effective mouth washes and especially effective on *Streptococcus mutans* in plaque and saliva of children when used for 10-15 min. The viscous oil turns thin and milky white. It is claimed that they activates enzymes and draws the toxins out of the blood. The oil should not be swallowed as it contains bacteria and toxins. Oil pulling therapy should be followed by tooth brushing⁽⁷⁾. Recently many researchers improve that the essential natural oils has antifungal, antiviral, antibacterial and anti amoebic action, including nigella(*nigella sativa*), sesame(*Sesamum indicum*) , flax(*Linum usitatissimum*) and ginger oil(*Zingiber officinale*)⁽¹³⁻²¹⁾, and some of these oils were safe and biocompatible materials⁽²²⁾.

MATERIALS AND METHODS

The total number of specimens were (seventy), prepared from heat cured acrylic resin and cold cured acrylic resin denture base materials, specimens were of (1cm×1cm×2mm) according to Webb⁽²³⁾. Specimens preparation of acrylic denture base material (Cold and Heat Cured) are the same, whereas the hard elastic foil of 2mm thickness were cut into plastic specimens of (1cm×1cm×2mm). Flasking was done by the conventional method. Packing accomplished according to the manufacture instruction. Then curing for the heat cured acrylic resin specimens were carried out by placing the clamped flask in the thermostatically controlled water bath for (1 hr at 74°C then 1/2 hr at 100°C), according to the manufacture instruction. After the completion of curing flasks were allowed to bench cool for 30minuts. The acrylic specimens were removed from their stone moulds. Any flashes of excess resin material were removed from the specimens by using acrylic bur. The speci-

mens were stored in distilled water at 37°C in the incubator for 7 days for conditioning. According to Williams⁽²⁵⁾ all specimens were sterilized by autoclave at 15 pound/inch²/121°C for 15 minutes. The specimens for each group were immersed for 8hrs in the tested oils (sun flower oil, sesame oil, nigella sativa, flax oil and ginger oil)[Emad factory/Mosul/Iraq] after they had been infected with *Candida albicans* and incubated for 48hr. The bacteriological procedure were accomplished according to Kazazoglu⁽²⁴⁾, by using the standardized Candidal cell suspensions (600×10⁶ CFU/ml), that is equal to macfarland standard bacteriological solution tube no.2. The procedure involve preparing the MacFarland Standard Bacteriological Solution (tube No.2) that composed of 0.2 ml. Barium Chloride of 1% and 9.8 ml. H2SO4 of 1%.(prepare new culture of pure *C. albicans* (so it will be fresh and in the active face),mix loop full *C. albicans* for several times with sterile distilled water to prepare a bacterial suspension matching MacFarland Standard Bacteriological Solution tube No.2. by using U.V.spectrophotometer (CECIL),then put 1 ml. of the prepared bacterial suspension in a screw capped bottles then immerse one acrylic specimen in each one, then incubated for 24hrs at 37°C, where after incubation we take 0.01 ml. of the bacterial suspension and plated on Sabourid agar for counting of *C. albicans* colonies after incubated for 24hrs at 37 °C . (to check the count of viable species only).After that remove each acrylic specimen from their screw capped bottles by using sterile twizzer then place each specimen in a screw capped bottle containing (1ml) of one of the tested oils for 8hrs, then take 0.01ml of solutions from each screw capped bottle and plated on sabourid agar for counting of colonies as (CFU/ml) after incubated for 24hrs at 37°C. (Plating and counting of *Candida albicans* was done for 0.01 ml of each tested oil to facilitate and simplify bacterial counting).

RESULTS

The mean ,number of samples, and standard deviation for the antifungal action of these oils on cold and heat cured acrylic

resin denture base materials were shown in Table (1) which show that cold cured acrylic resin inharber *C. albicans* more than the heat cured acrylic resin ,that is

due to the fact that cold cured acrylic resin has more surface roughness than heat cured acrylic resin ^(26 - 31).

Table (1) Descriptive statistics for disinfection of acrylic denture base material from *C. albicans*

| Treats | Acrylic | Mean(CFU/ml) | N | S. D |
|------------------|------------|--------------|----|-----------|
| D.W | Heat cured | 79881.20 | 5 | 9049.290 |
| | Cold cured | 81120.40 | 5 | 39183.575 |
| Nystatin | Heat cured | 1442.40 | 5 | 450.128 |
| | Cold cured | 2938.80 | 5 | 582.212 |
| Sunflower | Heat cured | 22.20 | 5 | 6.380 |
| | Cold cured | 46.80 | 5 | 11.032 |
| Sesame | Heat cured | 13.60 | 5 | 3.782 |
| | Cold cured | 22.60 | 5 | 4.393 |
| Nigella | Heat cured | 40.00 | 5 | 11.979 |
| | Cold cured | 58.40 | 5 | 6.804 |
| Flax | Heat cured | 9.40 | 5 | 4.506 |
| | Cold cured | 24.80 | 5 | 9.418 |
| Ginger | Heat cured | 24.60 | 5 | 10.479 |
| | Cold cured | 47.20 | 5 | 11.735 |
| Total | Heat cured | 11633.34 | 35 | 28443.497 |
| | Cold cured | 12037.00 | 35 | 31630.897 |

S.D: standard deviation, N: number of samples

The one way analysis of variance (Table 2) showed that at P=0.05 there were

significant differences between treats,

Table (2) Analysis of variance for disinfection of acrylic denture base material from *C. albicans*

| | Sum of Squares | df | Mean Square | F | Sig. |
|-----------------------|-----------------|----|----------------|--------|------|
| Between Groups | 55046840695.143 | 6 | 9174473449.190 | 89.188 | .000 |
| Within Groups | 6480579478.800 | 63 | 102866340.933 | | |
| Total | 61527420173.943 | 69 | | | |

df :degree of freedom

Which results from the significant differences between all tested oils and

D.W(control) shown in Dunnett t-test (Table 3).

Table (3) Dunnett (2-sided) t- test for disinfection of acrylic denture base material from *C. albicans*

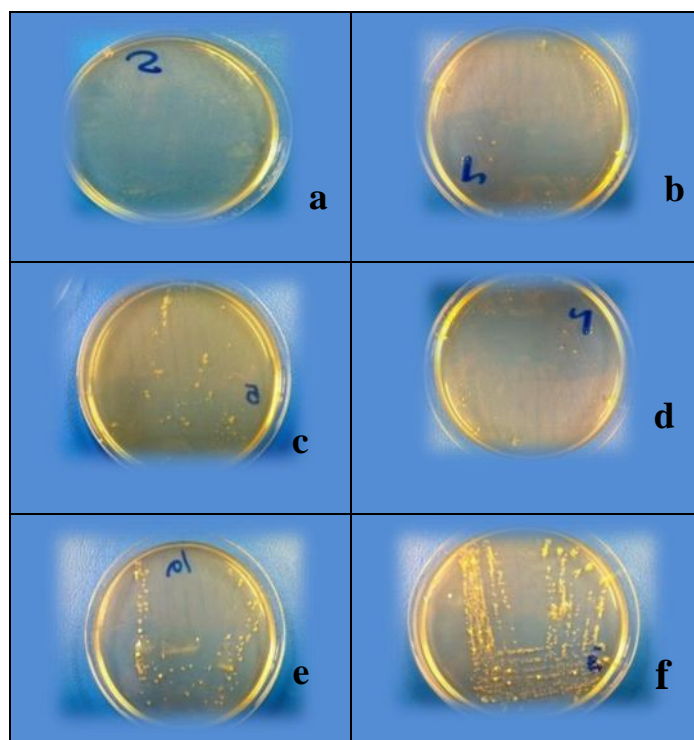
| | (I) treats | (J) treats | Mean Difference (I-J) | S. E | Sig. | 95% Confidence Interval | |
|----------------|------------------|------------|-----------------------|----------|------|-------------------------|-------------|
| | | | | | | Lower Bound | Upper Bound |
| Dunnett | Nystatin | D.W | -78310.20 | 4535.776 | .000 | -90275.76 | -66344.64 |
| | Sunflower | D.W | -80466.30 | 4535.776 | .000 | -92431.86 | -68500.74 |
| | Sesame | D.W | -80482.70 | 4535.776 | .000 | -92448.26 | -68517.14 |
| | Nigella | D.W | -80451.60 | 4535.776 | .000 | -92417.16 | -68486.04 |
| | Flax | D.W | -80483.70 | 4535.776 | .000 | -92449.26 | -68518.14 |

S.E: standard error

DISCUSSION

All tested oils were effective in disinfection of cold and heat cured acrylic resin from *C. albicans*. This was agreed with different researchers for the antifungal action of these oils⁽³²⁻³⁷⁾, the mechanism of action of these oils may be due to its viscosity that act by oil pulling mechanism, saponification, or emulsification action^(7,14), this may be caused by their high content of poly unsaturated fatty acids, palmitic acid, stearic acid, oleic acid, li-

noleic acid, α -linolenic acid, gingerol^(18,21), flax oil and sesame oil had strong antifungal action that may result from their content of the triply unsaturated omega-3 fatty acid, Sesamin, and sesamol⁽¹⁵⁻¹⁷⁾. So, the results of this study showed that all the tested oils were effective as a fungicidal agent, this agreed with Lima⁽³⁷⁾ who stated that when yeast <1000 CFU/ml of saliva considered as no growth. Figure (1) Compare the antifungal differences between the tested oils and nystatin.



Figures (1): Compare the antifungal differences between the tested oils (a- flax oil ,b- sesame oil ,c-sun flower oil, d- ginger oil and e-nigella sativa oil) and f- nystatin

CONCLUSIONS

All the tested natural oils were effective antifungal agents of heat and cold cured acrylic resin denture base materials when immersed for 8hrs in these oils.

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