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Comparative Evaluation of Antimicrobial Efficacy of *Ocimum sanctum (Tulsi)* and *Punica granatum* (Pomegranate Peel) Extracts as Herbal Denture Cleansers in Geriatric Denture Wearers: An *in vivo* Screening Study

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ABSTRACT

Introduction: Denture cleansing is an important procedure to prevent the accumulation of plaque. Patients often also use household cleansers as it can be easily made, safe to use and of low cost. Ocimum sanctum and Punica granatum are well known for its therapeutic uses. However, studies documenting its use in Prosthodontics and its effect on complete dentures are rare.
Aims and Objectives- To evaluate and compare the change of microbial count on the tissue surface of the maxillary denture on immersion in Ocimum sanctum (Tulsi) extract and Punica granatum (Pomegranate peel) extract solution after 1 day and 1 week of complete denture insertion.
Materials And Methods- Aqueous solution of Ocimum Sanctum and Punica Granatum extract solutions were prepared by cold extraction method. A total of 30 complete denture patients were selected. Patients were divided into three groups:
Group A: 10 patients were given distilled water as control for denture immersion.
Group B: 10 patients were given formulated Ocimum sanctum extract solution for denture immersion.
Group C: 10 patients were given formulated Punica granatum extract solution for denture immersion.
Swab were taken from each group from the tissue surface of upper denture and sent for microbial analysis post one day and one week of usage.
Results-The microbial load of the dentures reduced significantly by the usage of extract solutions.
Conclusion- Ocimum sanctum (Tulsi) extract solution was slightly more effective than Punica granatum (Pomegranate peel) extract solution.

Keywords- Ocimum sanctum, Punica granatum, oral flora, denture cleanser, complete dentures.

INTRODUCTION

Oral well-being is an impression of one's general wellbeing, influencing the capacity of a person to chew and phonate, and contributes altogether to a feeling of certainty and prosperity. Oral wellbeing status decays with age and accordingly the requirement for removable prostheses increments.

With age, wearing removable dental prosthesis causes a change in the oral microflora. S. *aureus*, S. *mutans*, C. *albicans*, *Diphtheroids*, *Veillonella* and *Acinetobacter* are a part of the normal flora of the edentulous patient that remains unaltered by denture wearing. While E.coli, Klebsiella, Moraxella Branhamella begins to be seen after denture wearing. For specific people, this new condition is in

charge of the advancement of a specific condition i.e. denture related stomatitis. [1-4]

Removable dentures are in constant contact of skin and mucosal surfaces. Hence, the entrapped food in the denture material becomes a nidus for the growth of micro-organisms. Moreover, in the wake of the COVID- 19 era, it becomes all the more important to ensure proper hygiene maintenance of the denture. [5] Denture cleansing is an essential method to keep the amassing of plaque. This can be ensured by simple practices of cleaning it with water diluted hand washes, or powders and dentifrices. But, cleaning with dentifrices has been proved to be abrasive to acrylic teeth and denture bases. The other immersion types of cleansers have also been proved to be harmful on the plastic or metal parts of the removable prosthesis. [6] Adding to this, if not water washed properly, the cleanser can be ingested causing damage not only oral mucosa, skin but may also effect gastrointestinal system, making the elderly more prone to systemic conditions. [7]

Although many commercial chemical denture cleansers are accessible, a perfect denture cleanser ought to be easy to utilize, successfully expel natural and inorganic matter from denture surface, have bactericidal and fungicidal properties and be compatible with all denture base materials. [8] Patients can likewise utilize herbal denture cleansers as it can be effectively made, safe to utilize and of minimal effort.

Clinical reviews have demonstrated that Punica granatum (Pomegranate peel) containing mouthwash may battle dental plaque and tartar development by repressing the exercises of the microorganisms that cause plaque. Furthermore, pomegranate mixes have mitigating properties that may help calm the tissues. Research shows that pomegranate peel separate stifles the capacity of microorganisms to adhere to the surface of the tooth. [9-11]

Tulsi (Ocimum sanctum) is a little plant, sub-shrub which has numerous uses. Ayurveda mentions the importance of medicinal uses of it. Biting of its leaves are considered very viable for the ulcer and contaminations in the mouth. Tulsi has been tried against a wide array of micro-organisms like *Candida albicans, Staphylococcus aureus, Streptococcus mutans* etc. [12]

Despite O. sanctum and P. granatum are notable for its therapeutic utilizes, its utilization in Prosthodontics and its impact on complete dentures is constrained. Therefore, the aim of the study is to evaluate and compare the effectiveness of O. sanctum (Tulsi) extract, P. granatum (Pomegranate peel) extract solutions on the microbial count in complete denture patients on the first day and seventh day of denture insertion.

The study was conducted to evaluate and compare antimicrobial efficacy of Ocimum sanctum (Tulsi) extract and Punica granatum (Pomegranate peel) extract denture immersion solutions on day 1 and day 7 after denture insertion in complete denture patients as an effective denture cleanser.

MATERIALS AND METHODS

The study was conducted at J.S.S Dental College and Hospital, JSS Academy of Higher Education and Research, Mysuru, Karnataka, India over a period of 6 months from August 2018-jan 2019. Before the start of the study clearance was obtained from the institutional ethical review board of the Dental College Hospital (DCH/IEC/2018-2019(2)/4).

The study was done in 3phases:-

Phase 1: Preparation of extracts (Fig 1,2)

Fresh leaves of the green variety of Ocimum sanctum (Linn.) were plucked from the local areas of Bannimantap, Mysuru and were taxonomically identified at JSS college of Pharmacy Mysuru. Collected plants were washed altogether and chopped into small pieces, conceal dried and grinded into powdered form.

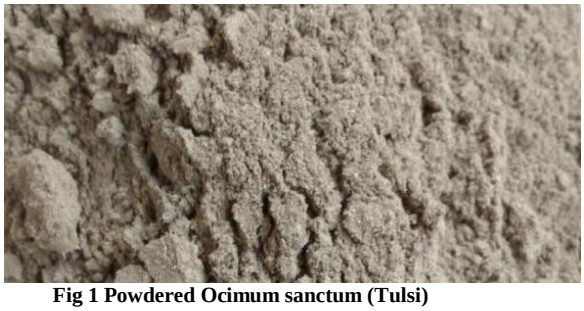

Fig 1 Powdered Ocimum sanctum (Tulsi)

Fig 2 Powdered Punica granatum (Pomegranate peel)

The pomegranate (Punica granatum) organic products were gathered from nearby market of Bannimatap, Mysuru. In the wake of washing, the peel was isolated from the mesocarp and flushed with refined water. Peel was then dried in hot air oven at 40^{O}C for 48 hours. After drying, the peels were granulated by blender and powdered type of plant test was acquired.

The coarse powder of Ocimum Sanctum and Punica Granatum were extracted with water by cold maceration (Fig 3) [11] and the marc (the damp solid material) was again extracted with water. The process was repeated four times. It was then subjected to filtration with Whatman filter paper to obtain a clear filtrate. The filtrate so obtained was reduced at a low temperature of less than 40°C to obtain a solid residue of Ocimum sanctum (Linn.) extract and Punica granatum extract and the filtrates were combined, distilled and evaporated. The extract was stored at 4°C for further testing.(Fig 4)

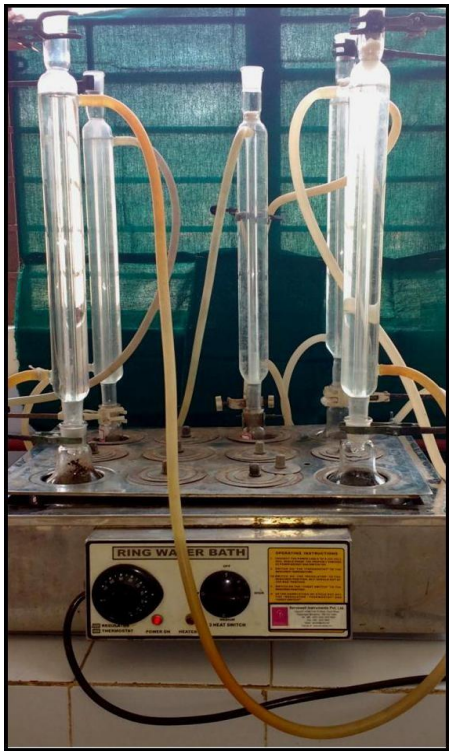

Fig 3 Cold Maceration Process

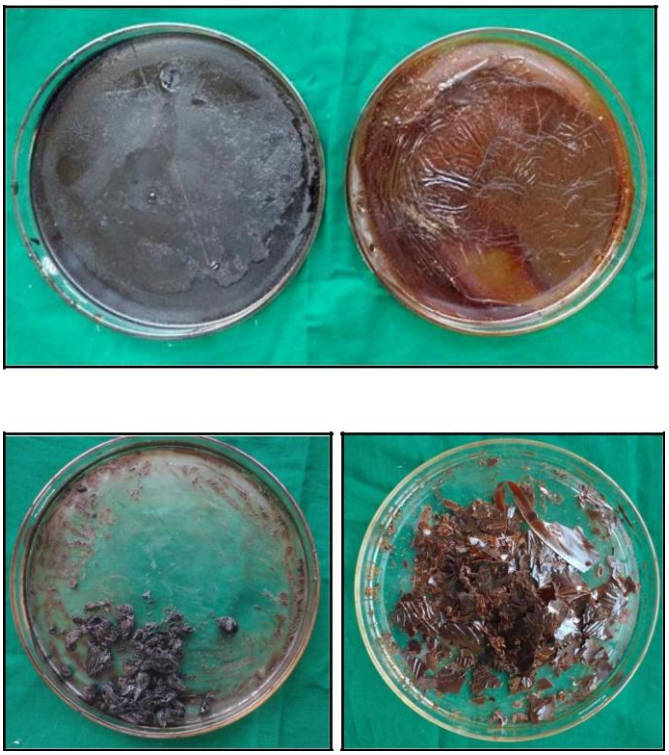

Fig 4 Ocimum sanctum, Punica granatum Extract

The percentage yield of Ocimum sanctum and Punica granatum extract was found to be 9.1 % and 12.7% respectively. For example: From 200 gms of Ocimum sanctum and Punica granatum powder dissolved in 1 litre of distilled water, 18.2 g and 25.4 g of extract (residue) was obtained, and thus the yield was 9.1 % and 12.7% w/v respectively (fig 4).

Phase 2:MIC (Minimum Inhibitory Concentration) determination of the extracts.

MIC determinations of the extracts were done at the Department of Microbiology. MIC determination was done by Broth Macro dilution method as per NCCLS guidelines.

Preparation of BHI (Brain Heart Infusion) broth-was carried out as per the standard protocol. E.coli ATCC 25923 (gram negative bacilli) and Staphylococcus aureus ATCC 25922 (gram positive cocci) were taken as the Quality Control (QC) organisms. These are the opportunistic microorganisms found in the oral cavity. The colonies were suspended with a loop and the growth transferred to the broth. The broth was incubated at 35–37 °C for minimum 2 hours until the growth reached a turbidity equal to or greater than that of a 0.5 McFarland standard.

The BHI broth was transferred into 9 test tubes of 1ml each .1 ml of prepared extract solutions of different concentrations was mixed with it.

The Ocimum sanctum and Punica granatum extract were diluted with distilled water to obtain 9 different concentrations (2%, 4%, 10%, 20%, 25%, 40% 50%, 75%, 100%) of the extract.

After 24 hrs, with the help of inoculating loop the solution was sub-cultured into blood agar plates and labelled according to the concentration. After each streaking the loop was heated till it became red hot for sterilization of the loop. After streaking, the agar petri dishes were placed in an incubator at 37^{o}C for 24 hours. The concentration at which agar plate did not show any visible growth was considered as minimum inhibitory concentration. For Ocimum sanctum and Punica granatum, the MIC was found to be 40% and 50% respectively.

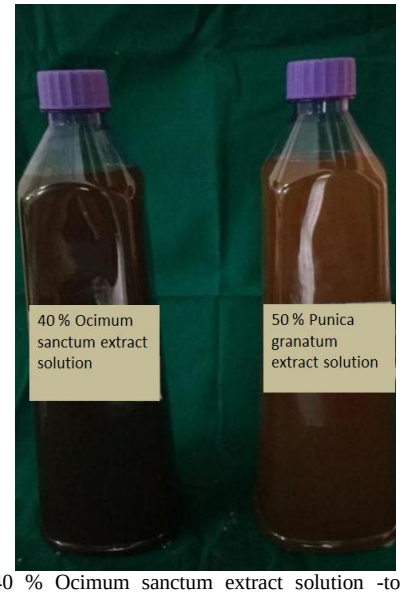

Fig 5: 40 % Ocimum sanctum extract solution -to Group B patients 50 % Punica granatum extract solution - to Group C patients

Phase 3: To determine the antimicrobial efficacy of the extracts in complete denture patients

Source of Data:

A total of 30 complete denture patients were selected from the outpatient section in the Department of Prosthodontics and Crown & Bridge, JSS Dental College and Hospital, for the study based on the inclusion criteria-Patients who gave informed consent, Edentulous patients wearing dentures for not less than 1week, Patients in the age group of 45-65years All the participants were informed about the study objectives and methodology and informed written consent was obtained from them in vernacular language.

Patients were divided into three groups:

Group A: 10 patients were instructed to use given distilled water as control for denture immersion.

Group B: 10 patients were instructed to use given formulated Ocimum sanctum extract solution for denture immersion.

Group C: 10 patients were instructed to use given formulated Punica granatum extract solution for denture immersion.

Patients were instructed not to use any chemical disinfectant to clean the dentures. Patients were instructed to wear the dentures whole day on the day of denture insertion

and were recalled the following day for swab collection.

1 Day after Denture Insertion: sterile swab were dispensed from the sterile test-tube and rolled on the tissue surface of the upper denture for 3-4 times. Swab were placed back in the sterile test-tube and marked, and sent for microbial analysis.

After Day 1, patients were instructed to wear denture only during the day and to remove their denture before going to the bed and immersing the denture into their respective solutions i.e. distilled water / Ocimum sanctum/ Punica granatum extract solution.

Patients were instructed to soak their dentures overnight in the given solution in a closed container provided for the purpose. Patients were instructed to continue the protocol for next 6 days and replenish the solution after each night.

1 Week After Denture Insertion: swab were collected again by using a sterile cotton swab from the tissue surface of the upper denture and sent for microbial analysis

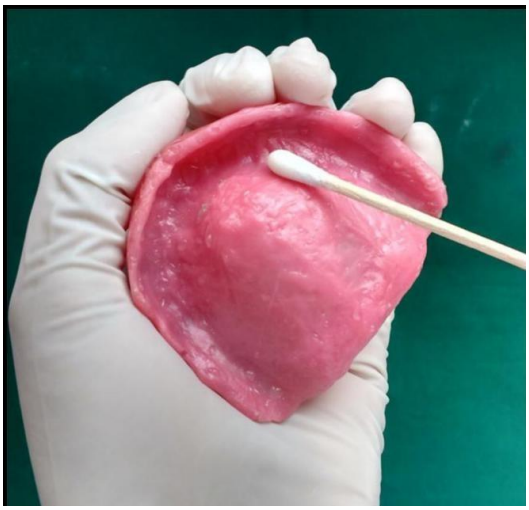

Fig 6: Swab taken from tissue surface of the denture at Day 1 and Day 7 after denture insertion

All the patients from each group were recalled after 1 day and a week of denture insertion. Swab were taken for each group from the tissue surface of upper denture and sent for microbial analysis. The swabs were cultured in blood agar media and incubated at 37^{o}C for 24 hours. Then microbial count was recorded in CFU/ml. After collecting the data, results were tabulated, statistically analysed and compared. (fig 6)

MICROBIOLOGICAL EVALUATION (Fig 7 a&b ,8 a&b ,9 a&b)

After the collection the swab was processed in the Department of Microbiology, J.S.S. Medical Hospital, JSS Academy of Higher Education and Research, Mysuru, as follows-

The labelled swabs were streaked on the petridish containing blood agar medium using a loop. Thereafter, the agar petri dishes were placed in an incubator at 37^{o}C for 24 hours.

After 24 hours the inoculated colony forming units were counted

After collecting the data, results were tabulated, statistically analyzed and compared.

Numbering of the patients was done to avoid selection bias during evaluation which included three digit coding which was also followed even for the samples collected from all the three groups at different days of interval.

First digit denoted the main group of the patients. A (patients using distilled water as denture soaking agent) or B (patients using Ocimum sanctum extract as denture soaking agent) or C (patients using Punica granatum extract as denture soaking agent).

Fig 7a

Fig 7b

Fig 7a: Microbial load at day 1 after complete denture insertion for group a
Fig 7b: Microbial load at day 7 after complete denture insertion for group a

Fig 8a

Fig 8b

Fig 8a: Microbial load at day 1 after complete denture insertion for group b
Fig 8b: Microbial load at day 7 after complete denture insertion for group b

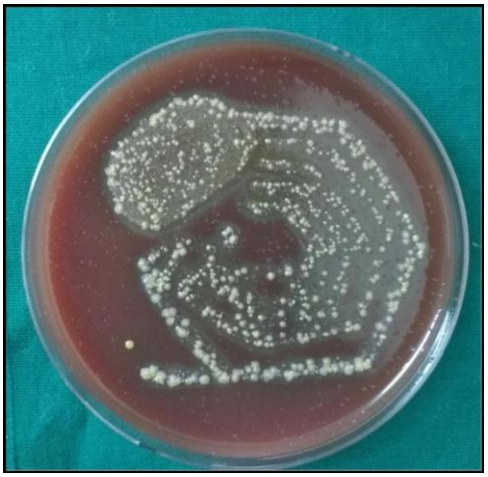

Fig 9a

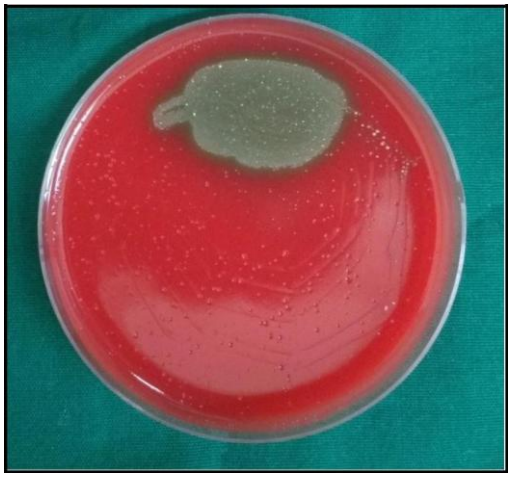

Fig 9b

Fig 9a: Microbial load at day 1 after complete denture insertion for group c
Fig 9b: Microbial load at day 7 after complete denture insertion for group c

Eg ; B (19) a

B-indicates that patient is using Ocimum sanctum extract as denture soaking agent
19 -indicates that it is the 19th patient of the group B
a- Indicates that the swab sample is collected on 1st day after complete denture insertion.

RESULTS

The following statistical methods were applied in the present study: - Descriptive statistics were used to describe the basic features of the data in a study. They provide simple summaries about the sample and the measures, Independent samples 'test-The independent samples 't' test compared the means of two independent groups in order to determine whether there is statistical evidence that the associated population means are significantly different, Paired samples 't'test- The Paired Samples '*t'* Test compared two means that are from the same individual, object, or related units, One Way Anova-determined the statistical significant differences between the means of both the groups.

The statistical operations were done through SPSS (Statistical Presentation System Software) for Windows, version 22 (2015).

In the present study, on comparision of microbial count (CFU/ml) reduction among the patients using distilled water (Group A) which was 6.6000CFU/ml SD(1.07) on day 1, The mean cfu/ml reduction on usage Ocimum sanctum extract solution (Group B) showed 6.05 cfu/ml reduction with SD(0.97), The mean cfu/ml reduction on usage of Punica granatum extract solution (Group C) was 7.3 cfu/ml with SD (1.25) as denture soaking agent which was statistically significant. (Table1)After one week of usage of denture cleansing solution, the mean CFU reduction in Group A as 4.8CFU/ml with SD OF 1.13, Group B showcased mean fungal reduction of 3.0CFU/ml with SD of 1.41, and Group C showed significant reduction of 3.7CFU/ml with SD of 2.35 (Table 1)

Table 1: Descriptive Statistics for Group A, B, C

Groups	Day 1		Day 7	
	Mean	Standard Deviation	Mean	Standard Deviation
Group A	6.6000	1.07497	4.8000	1.13529
Group B	6.5000	0.97183	3.0000	1.41421
Group C	7.3000	1.25167	3.7000	2.35938
Total	6.8000	1.12648	3.8333	1.82101

Independent samples "t" test(Table 2 ,3&4) between Group A and Group B was done to evaluate the mean values of microbial count among Group A and Group B denture wearers on day 1 and on day 7 after denture insertion. Difference in the antimicrobial efficacy between the two groups at day 1 was not statistically significant (p*>**0.05) having both groups microbial counts statistically the same. Difference in the antimicrobial efficacy between the two groups at day 7 is statistically significant (p*<**0.05), with participants in group B with higher value of microbial inhibition.

Table 2:- Independent Samples 'T' Test Between Group A and Group B

Duration	t	Df	Sig. (2-tailed)	Mean Difference
Day 1	0.218	18	0.830	0.10000
Day 7	3.139	18	0.006	1.80000

Table 3: Independent Samples 'T' Test between Group B and Group C

	t	Df	Sig. (2-tailed)	Mean Difference
Day 1	-1.596	18	0.128	-0.80000
Day 7	-0.805	18	0.431	-0.70000

Table 4: Independent Samples 'T' Test between Group A and Group C

	t	Df	Sig. (2-tailed)	Mean Difference
Day 1	-1.342	18	0.196	-0.70000
Day 7	1.329	18	0.201	1.10000

On comparison of mean values of microbial count reduction among Group B and Group C denture wearers on day 1 and on day 7 after denture insertion revealed no significant statistical difference and microbial counts for both the groups are statistically same on both days.

The mean values of microbial count reduction among Group A and Group C denture wearers on day 1 and on day 7 after denture insertion revealed no statistically significant difference (p*>**0.05), indicating that microbial counts for both the groups are statistically same on both days.

Paired samples 't' test shows that the mean microbial count measurements recorded in Group A, B and C on first day

were higher than that on the seventh day after treatment.

This result was statistically significant (p*<**0.05) among all the pair of days interval. (Table 5,6 &7 graph 1)

Group A showed 27.27% reduction in the bacterial count (CFU/ml) after 7 days of complete denture insertion in distilled water as an overnight denture soaking agent.

Table 5: Paired Samples 'T' Test of Group A

Comparison	Mean	Std. Deviation	Std. Error Mean	T	Df	Sig. (2-tailed)
day1 vs day7	1.80000	1.22927	0.38873	4.630	9	0.001

Table 6: Paired Samples'T' Test of Group B

Comparsion	Mean	Std. Deviation	Std. Error Mean	T	Df	Sig. (2-tailed)
day1 vs day7	3.50000	1.50923	0.47726	7.334	9	0.000

Table 7: Paired Samples 'T' Test of Group C

Comparison	Mean	Std. Deviation	Std. Error Mean	T	Df	Sig.(2-tailed)
day1 vs day7	3.60000	2.27058	0.71802	5.014	9	0.001

Group B showed 53.84% reduction in the bacterial count (CFU/ml) after 7 days of complete denture insertion in distilled water as an overnight denture soaking agent.

Group C showed 49.31% reduction in the bacterial counts (CFU/ml) after 7 days of complete denture insertion in distilled water as an overnight denture soaking agent. Graph 4 represents the comparison of the microbial count (CFU/ml) of the 3 groups' i.e. Group A, Group B and Group C on day 1 after complete denture insertion and day 7 after complete denture insertion.

One way Anova revealed no statistically significant Difference in the antimicrobial efficacy of the three groups at day 1 and day 7 (p*> **0.05) (Table: 8)

Table 8: One Way Anova for Group A, B and C on Day 1 and Day 7

Between Groups	Sum of Squares	df	Mean Square	F	Sig.
Day 1	3.80	2	1.90	1.55	0.23
Day 7	16.47	2	8.23	2.79	0.08

DISCUSSION

Human oral cavity micro-flora plays a significant role in maintaining a healthy oral mucosa in normal dentulous and even edentulous denture users. The oral cavity contains about 700 bacterial and fungal species of which more than half have not yet been isolated and differentiated. [14]

The oral deposits and microorganisms that adhere to a dental prosthesis bring about several untoward effects. Firstly, the adherent material itself is unaesthetic in appearance and unpleasant in terms of tactile sensation, taste, and odor. The person with an unclean denture is likely unaware of the unpleasant smell and taste of the prosthesis because the sensory receptors accommodate to it. [15] Secondly, there are problems posed by the abundance of the microbial population supported by unclean dentures. [16] Metabolic by-products and exotoxins in the deposits can be irritating to oral tissues. Denture plaque can also become calcified if not removed thoroughly and regularly. Calculus is also readily stained by tobacco, tea, coffee, certain medications (particularly iron supplements), and numerous other ingested materials.

Denture cleanliness is essential to prevent malodor and poor esthetics. The accumulation of plaque/calculus and consequent deleterious effects on the mucosa. Dentures can be cleaned mechanically, chemically, or by a combination of the two. Although many commercial chemical denture cleansers are available, patients also often use household cleansers because of its ease of availability and low cost. The popular household cleansers are vinegar, diluted vinegar, soda, salt, and bleach. [17-21] Tulsi, scientifically

known as Ocimum sanctum, is a time-tested premier medicinal herb. It is a plant of Indian origin, and used in Ayurvedic medicine since ancient times. Literature review reveals that the antimicrobial property of Tulsi has been tested against a variety of microorganisms like Candida albicans, Staphylococcus aureus, enteric pathogens, Klebsiella, Escherichia coli and Proteus. It has also demonstrated antigonorrheal efficacy against multiresistant strains of Neisseria gonorrhea and clinical isolates of beta lactamase-producing methicillin-resistant Staphylococcus aureus. [22]

The pomegranate with the botanical name of Punica granatum Linn. is a fruit-bearing deciduous shrub or small Asian tree from the family of Punicaceae growing between 5 to 8 meters tall. Review of literature showed that some researchers such as Vasconcelos et al [22] also reported that extracts of Punica granatum peel in different concentrations were effective against S. epidermidis, S.aureus, S. mutans, S. sanguinis and S. salivarius. [23-26]

The impetus for the study was the non-availability of literature about the antimicrobial activity of Ocimum sanctum (Tulsi) and Punica granatum (Pomegranate peel) in the field of prosthodontics and in complete denture patients. Therefore, this study has been based on the antimicrobial property of Ocimum Sanctum (Tulsi) and Punica granatum (Pomegranate peel) to be used as denture immersion solution in complete denture patients.

The Ocimum sanctum and Punica granatum extract solutions were prepared by cold extraction process. Minimum Inhibitory Concentration of the extracts were determined by Broth macrodilution method and was found to be 40 % for Ocimum sanctum extract and 50 % for Punica granatum extract.

Previous studies showed extracts were prepared with ethanol or methanol as a solvent and had been tested for in vitro purpose. As this is in vivo study and to be used by complete denture patients as denture immersion solution, the extracts were prepared in an aqueous base.

A total of 30 complete denture patients were selected for the study based on the inclusion criteria. Patients were divided into 3 groups-one using distilled water (which was taken as control) as denture immersion solution, one using 40 % Ocimum sanctum extract solution as denture immersion solution and last one using 50 % Punica granatum extract solution as denture immersion solution. Patients were instructed not to use any chemical disinfectant to clean the denture. Patients were advised not to use mechanical method of denture cleansing. All the patients from each group were recalled after 1 day and a week of denture insertion. Swab were taken for each group from the tissue surface of upper denture and sent for microbial analysis.

The results in the present study demonstrated a statistically significant ($p < 0.05$) decrease in the microbial count (mean CFU/ml) in all three groups.

The control group had an average of 10^7 microbial counts on day 1 and reduced on an average of 10^5 on day 7 after complete denture insertion using distilled water. Group A showed 27.27% reduction in the bacterial count (CFU/ml) after 7 days of complete denture insertion in distilled water as an overnight denture soaking agent.

The Group B had an average of 10^7 microbial counts on day 1 and reduced on an average of 10^3 on day 7 after complete denture insertion using the Ocimum sanctum extract solution. Group B showed 53.84% reduction in the bacterial count (CFU/ml) after 7 days of complete denture insertion in Ocimum sanctum extract solution as an overnight denture soaking agent.

The Group C had an average of 10^7 microbial counts on day 1 and reduced on an average of 10^4 on day 7 after complete denture insertion using the Punica granatum extract solution. Group C showed 49.31% reduction in the bacterial counts (CFU/ml) after 7 days of complete denture insertion in

Punica granatum extract solution as an overnight denture soaking agent.

The results thus obtained were in agreement with the hypothesis that Ocimum sanctum (Tulsi) and Punica granatum (Pomegranate peel) could cause reduction in microbial count of the dentures. But the complete eradication of the microbes was not observed. Study needs to be done to see the effect of these solutions along with mechanical method of denture cleansing as well. As this study was done only for 7 days, long term clinical studies to check the efficacy might be useful to further evaluate the antimicrobial action of ocimum sanctum (tulsi) extract solution and punica granatum (pomegranate peel) extract solution on the dentures and to see its long term effects on denture properties.

Hence it may be concluded that Ocimum Sanctum (Tulsi) and Punica Granatum (Pomegranate peel) may represent a promising herbal denture cleanser for complete denture wearers. Its inexpensive and safe use can benefit the edentulous elderly patients. Easy availability makes it convenient for long term usage.

CONCLUSION

Within the limitation of this in vivo study, the following observations were made;

- Significant change in the microbial count (CFU/ml) from the palatal tissue surface of the dentures of complete denture patients occurred by overnight soaking of the dentures in formulated Ocimum sanctum (Tulsi) and Punica granatum (Pomegranate peel) extract solution and the reduction continued till the 7^{th}day.
- Among the two, Ocimum sanctum (Tulsi) extract solution was slightly more effective in being an antimicrobial agent (53.84% reduction in microbial count) than Punica granatum (Pomegranate peel) extract solution (49.31% reduction in microbial count)
- Thus, it can be concluded from this study that, Ocimum sanctum (Tulsi) and Punica granatum (Pomegranate peel) are equally effective and have a good antimicrobial property which can be used as a herbal denture cleanser. Inexpensive and easy availability makes them promising denture cleanser solutions. As this study was done only for 7 days, long term clinical studies to check the efficacy might be useful to further evaluate the antimicrobial action of Ocimum sanctum (Tulsi) extract solution and Punica granatum (Pomegranate peel) extract solution on the dentures and to see its long term effects on denture properties and an appropriate usage regimen can be formulated. Research can be also be done to see its effect on dentulous patients. Nonetheless, with the detailed knowledge obtained, we can use Ocimum sanctum (Tulsi) extract solution and Punica granatum (Pomegranate peel) as an effective herbal denture cleanser which can aid towards the development of potential therapeutic agents with better patient compliance.

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Competing Interest:- None.

List of Abbreviations:

% : Percentage
& : and
/ : Per
µ : Micron
ANOVA : Analysis of Variance
CFU : Colony Forming Units
Df : degree of freedom
etc : et cetera.
Fig : Figure
Gm : gram
Hrs : hours
MIC : Minimum Inhibitory Concentration
ml : milliliter
mm : millimeter
^{o}C : degrees Centigrade
SD : Standard deviation
Sec : seconds

REFERENCES

1. Barbeau J, Séguin J, Goulet JP, de Koninck L, Avon SL, Lalonde B, Rompré P, Deslauriers N. Reassessing the presence of Candida albicans in denture-related stomatitis. Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology. 2003 Jan31;95(1):51-9.
2. Harrison Z, Johnson A, Douglas CW. An in vitro study into the effect of a limited range of denture cleaners on surface roughness and removal of Candida albicans from conventional heat-cured acrylic resin denture base material. J Oral Rehabil. 2004 May; 31(5):460-7.
3. Kanli A, Demirel F, Sezgin Y. Oral candidosis, denture cleanliness and hygiene habits in an elderly population. Aging Clin Exp Res. 2005Dec;17(6):502-7.
4. Jagger DC, Harrison A.Denture cleansing-the best approach. Br Dent J 1995;178:413-17.
5. Veres EM, Wolfaardt JF, Hnizdo E. Denture cleansers: Part II--A survey of instructions given by dentists on denture cleansing. The Journal of the Dental Association of South Africa 1985 Oct 1;40(10):585-9.
6. American Dental Association. Denture cleansers. Council on Dental Materials, Instruments, and Equipment. J Am Dent Assoc. 1983;106:77-9.
7. Evelyna A, Sutanto D, Hasianna ST, Azaria C, Wahyudianingsih R. Cytotoxicity Test of Chitosan Solution as Acrylic Denture Cleansers Solution. InJournal of Physics: Conference Series 2019 Mar (Vol. 1175, No. 1, p. 012280). IOP Publishing.
8. Kukreja BJ, Dodwad V. Herbal mouthwashes-A gift of nature. Int J Pharma Bio Sci. 2012Apr;3:46-52.
9. Vasudevan DM, Kedlaya R, Deepa S, Ballal M. Activity of Ocimum sanctum (the traditional Indian medicinal plant) against the enteric pathogens. Indian journal of Medical Sciences. 2001 Aug;55(8):434-8.
10. Carolina Yoshi Campos Sugio1, Maria Gabriela Robles Mengoa1, Anna Clara Gurgel G, Amanda Aparecida Maia Neves G, Thaís Marchini de O, Karin Hermana N. Use of Natural Products in the Prevention and Treatment of Denture Stomatitis. 2020 - 1(5) OAJBS.201-206
11. Pinni J, Avula J.S.S, Mukthineni S, Bandi S, Gokul T. Evaluation of Anticariogenic Efficacy of pomegranate (Punica Granatum) Pericap Extract as natural Mouth rinse: An In Vitro and In vivo study. Bio med Pharmacol J.2018;11(4):
12. Handa SS, Khanuja SPS, Longo G,Rakesh DD(2008) Extraction Technologies for Medicinal and Aromatic Plants, (1stedn), no. 66. Italy: United Nations Industrial Development Organization and the International Centre for Science and High Technology.
13. Marsh PD, Martin MW. Oral microbiology, 3rd Edn, Chapman and Hall, London,1992
14. Hyde RJ. Feller RP. Sharon IM. Tongue brushing, dentifrice, and age effects on taste and smell. Journal of Dental Research. 60(10):1730-4, 1981.
15. Loesche WJ. Bromberg J. Terpenning MS. Bretz WA. Dominguez BL. Grossman NS. Langmore SE. Xerostomia, xerogenic medications and food avoidances in selected geriatric groups. Journal of the American Geriatrics Society. 43(4):401-7, 1995.
16. Anthony DH, Gibbons P. The nature and behaviour of denture cleansers. J Prosthet Dent1958;8:796-810
17. Kimball HD. Factors to be considered in the control and elimination of chronic tissues soreness beneath dentures. J Prosthet Dent.1954; 4:298.
18. Anthony DH, Gibbons P. The nature and behaviour of denture cleansers. J ProsthetDent.1958.
19. Oral hygiene devices and aids in oral health: Guide to dental materials and devices, 8th edition. Chicago, 1976, American Dental Association, p24.
20. Abelson DC. Denture plaque and denture cleansers. The Journal of prosthetic dentistry. 1981 Apr30;45(4):376-9.
21. Shokeen P, Bala M, Sing M, Tandon V. In vitro activity of eugenol, an active component from Ocimum sanctum, against multiresistant and susceptible strains of Neisseria gonorrhea. Int J Antimicrob Agents.2008; 32:172-9.
22. Vasconcelos LC, Sampaio FC, Sampaio MC, Pereira MD, Higino JS, Peixoto MH. Minimum inhibitory concentration of adherence of Punica granatum Linn (pomegranate) gel against S. mutans, S. mitis and C. albicans. Brazilian Dental Journal. 2006; 17(3):223-7.
23. Nara Ligia Martins Almeida, Luiz Leonardo Saldanha, et al. Antimicrobial activity of denture adhesive associated with *Equisetum*

giganteum- and *Punica granatum*-enriched fractions against *Candida albicans* biofilms on acrylic resin surfaces, 2018; Biofouling: 34:1, 62-73.
24. Jurenka JS (2008) Therapeutic applications of pomegranate (Punica granatum L.): a review. Altern Med Rev 13(2): 128-144.
25. Madugula P, Reddy S, Koneru J, Rao AS, Sruthi R, et al. (2017) "Rhetoric to Reality"-Efficacy of Punica Granatum peel extract on oral candidiasis: an in vitro study. J Clin Diagn Res 11(1): ZC114-ZC117.
26. Bassiri-Jahromi S, Katiraee F, Hajimahmoodi M, Mostafavi E, Talebi M, et al. (2015) In vitro antifungal activity of various persian cultivars of Punica granatum L. Extracts against Candida species. Jundishapur J Nat Pharm Prod 10: e19754.

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