



IJCRR

Section: Healthcare

ISI Impact Factor
(2019-20): 1.628IC Value (2019): 90.81
SJIF (2020) = 7.893

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"Comparative Evaluation of Antifungal Effect of Origanum Oil Solution and Povidone Iodine Aqueous Solution on Scleral Resin: An *In-Vitro* Study"

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ABSTRACT

Aim: To compare and evaluate the antifungal effect of origanum oil and povidone-iodine aqueous solution on scleral resin.

Introduction: Ocular prosthesis being a part of maxillofacial prosthodontics aims to restore and repair the ophthalmic cavity. An important prerequisite for a prosthesis is the eradication of plaque accumulation due to bacteria. To neglect probable infections in the patient's ophthalmic cavity a naturally available antifungal agent was tested in this study.

Methods and Material: In this study, 40 custom made scleral resin disks (20 in each group) immersed in 2 different solutions were studied for their antifungal activity. The evaluation of this property was done by calculating the zone of inhibition around the disks propelled in the specific agar media.

Statistical Analysis: Descriptive and analytical statistics were done. The data is represented in mean and standard deviation. The normality of continuous data was analysed by the Shapiro-Wilk test. As the data followed a normal distribution, parametric tests were used to analyse the data. The independent sample t-test and paired sample t-test were used to check to mean differences. The level of significance was kept at $p < 0.05$. The software used was SPSS (Statistical Package for Social Sciences) Version 24.0 (IBM Corporation, Chicago, USA)

Results: The antifungal efficacy of the origanum oil solution was found to be significantly higher than the povidone-iodine solution.

Conclusion: This study concluded that origanum oil is a potent antifungal against *C. Albicans*. This encourages the researcher to assess the effectiveness of *O. vulgare* oil in other forms of systemic and superficial fungal infections as well as to investigate its broad-spectrum outcome against other pathogenic manifestations which also include malignancies.

Key Words: Ocular prosthesis, Antifungal, Origanum oil, *C. Albicans*, SDA, Maxillofacial, Prosthetics

INTRODUCTION

Ocular prosthesis is a part of maxillofacial prosthodontics that aims to restore and repair the ophthalmic cavity which aims in beautifying the compromised patients face. It also aids to improve the psychology and social development of the patient thus enhancing the quality of life.¹

The ocular prosthesis has seen exponential growth for a decade. This development is due to improved materials, enhanced postoperative management and the rising patient requirement. A diversity of materials are being used nowadays for ocular prostheses.²

As this prosthesis has an intimate and dynamic relationship with the ocular surface it carries a threat of infection throughout their life and has a major effect on the esthetics of the patient esthetics. Proper function and retention act as an important factors in the success of this type of prosthesis.²

An important prerequisite for a prosthesis is the eradication of plaque accumulation due to bacteria. To neglect probable infections in the patient's ophthalmic cavity, the ocular prosthesis should be removed on a routine basis and disinfection should be done; after which it can be inserted again.³

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ISSN: 2231-2196 (Print)

ISSN: 0975-5241 (Online)

Received: 08.02.2021

Revised: 12.03.2021

Accepted: 05.04.2021

Published: 24.10.2021

AIM

To compare and evaluate the antifungal effect of origanum oil and povidone-iodine aqueous solution on scleral resin

OBJECTIVES

- Evaluation of the antifungal effect of origanum oil solution on scleral resin
- Evaluation of the antifungal effect of povidone-iodine aqueous solution on scleral resin
- To compare and determine the efficacy of antifungal property of origanum oil solution with the povidone-iodine aqueous solution on scleral resin

STUDY DESIGN AND METHODOLOGY

MATERIALS USED

1. Scleral resin disks
2. Povidone-iodine aqueous solution
3. Origanum oil solution

PREPARATION OF SAMPLE AND GROUPS-

- Preparation of scleral resin disks (40) using a custom mould was done and was divided into two groups (Group A (20)–scleral resin to be immersed in origanum oil solution Group B(20)–scleral resin to be immersed in povidone-iodine aqueous solution) [Fig.1]

METHOD TO CHECK ANTIFUNGAL PROPERTY

- The fungal suspension containing strains of *C. Albicans* was prepared.[Fig.2] Scleral resin disks of both the groups (A and B) were immersed in the fungal suspension for the desired period after which the disks were incubated for 24hrs for the organisms to grow

Phase I

- A sterile swab was taken from the surface of these incubated disks and cultured in Sabouraud's dextrose agar (SDA) and incubated at 37°C. The disks of group A were immersed in an origanum oil solution(60%) and the disks of group B in 10% povidone-iodine aqueous solution(betadine) [Fig.3]

Phase II

- After 15 minutes of immersion of the disks of both groups in the two solutions, the antifungal potential of the testing solutions was evaluated by Agar well diffusion method. For this evaluation, previously cultured Petri dishes containing Sabouraud's dextrose agar (SDA) which were cultured using the swab technique for *C. Albicans* were taken. After this, the plates containing agar were left for drying. Using sterile cork borer 3 wells or cups were made. The disks which were immersed in the solution were inserted in the wells of the inoculation of the particular media agar

plates. [Fig.4] The plates were kept on hold for 10 min so that essential oils get diffused. After which incubation of these plates was carried out at 37°C for 48 h. Finally, after the incubation period, examination of plates detected the existence of clear zones of growth inhibition surrounded the wells which contained the testing solutions, which pointed to their efficacy against *C.albicans*. [Fig.5] The zone of inhibition was recognized by measurement of the good diameter (in millimetres) in one plane using the Vernier scale.[Table 1].

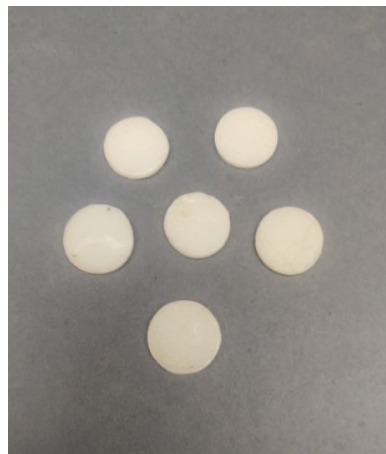


Figure 1: Custom Made Scleral Resin Disks.

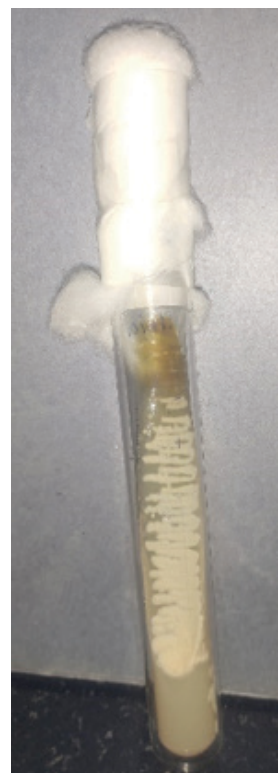


Figure 2: Candidal Growth Used for Broth Preparation.



Figure 3: Scleral Resin Disk Immersed in Povidone Iodine solution and Origanum Oil Solution.

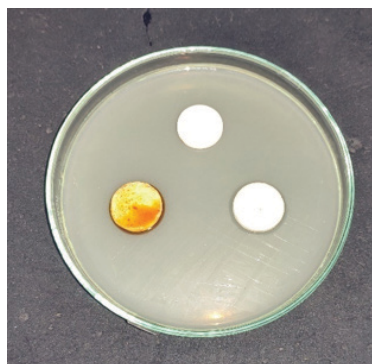


Figure 4: Scleral Resin Disks Immersed in Two Solutions Along with a Control Group Disk Propelled in Sda Plates inoculated With Candida.



Figure 5: Scleral Resin Disks Immersed in Two Solutions Along with a Control Group Disk Propelled in SDA Plates inoculated with Candida Showing Zone of Inhibition.

Table 1: Diameter of the inhibition zone calculated by vernier calliper in millimetres (mm).

Sample no.	Povidone Iodine aqueous solution (mm)	Origanum oil (mm)
1	17	>40
2	16	>40
3	11	>40
4	12	32
5	17	34
6	16	>40
7	18	>40
8	7	22
9	10	>40
10	12	18
11	11	>40
12	17	>40
13	16	36
14	12	38
15	17	>40

STATISTICAL ANALYSIS

Descriptive and analytical statistics were done. The data is represented in mean and standard deviation. The normality of continuous data was analysed by the Shapiro-Wilk test. As the data followed a normal distribution, parametric tests were used to analyse the data. The independent sample t-test and paired sample t-test were used to check to mean differences. The level of significance was kept at P<0.05.

Software: SPSS (Statistical Package for Social Sciences) Version 24.0 (IBM Corporation, Chicago, USA)

OBSERVATION AND RESULTS

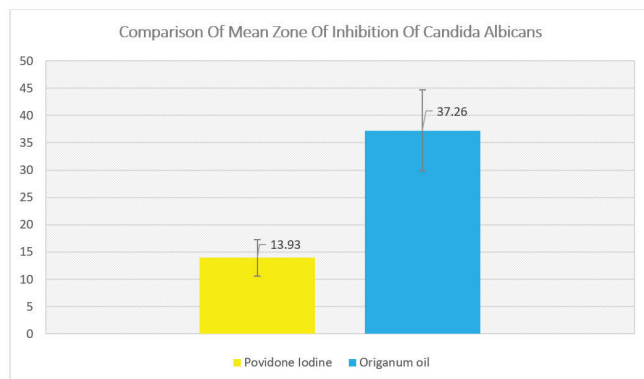
Output Tables:

Table 2: Comparison of the mean zone of inhibition of candida Albicans by origanum oil solution and povidone-iodine aqueous solution on scleral resin

Groups	N	Mean	S.D.	S.E.	M.D.	95% C.I.	t-value	P-value [#]
Povidone Iodine	15	13.93	3.36	0.86	-23.33	-27.65--19.00	-11.053	<0.001 [†]
Origanum oil	15	37.26	7.44	1.92				

[#]P-value derived from independent sample t-test; [†]significant at P< 0.05

The mean zone of inhibition of candida Albicans by origanum oil solution and povidone-iodine aqueous solution on the scleral resin was compared. It was found that there was a highly statistically significant difference in the mean zone of inhibition of *candida albicans* ($P < 0.001$). The mean zone of inhibition of povidone iodine aqueous solution (13.93 ± 3.36) was significantly lesser than that of origanum oil solution (37.26 ± 7.44). [Table.2]



Graph 1: Comparison of the mean zone of inhibition of candida Albicans by origanum oil solution and povidone-iodine aqueous solution on scleral resin.

DISCUSSION

Nowadays, drug resistance for multiple drugs in the human pathogenic microbes are observed at a high rate this is a result of excessive use or abuse of commercially available antimicrobial drugs. Resistant strains of *Candida albicans* are becoming a reason for major health concerns, and thus new antifungal agents are required to overcome this issue. This situation has encouraged the researchers to develop some new and effective antimicrobial agents that have the potential to replace the regimens currently used.

For a long time, medicinal plants are playing a vital role in promoting healthy mankind. Traditional medicines have always been economical and readily available sources of management in the principal health care system of indigent communities and countries. More than 80% of the people in the developing countries make use of traditional medicines reason being the non-affordability of the marketed medicines available for health care and also because these unconventional medicines are more suitable from a cultural and spiritual perspective.⁸

In the presenting study, the in vitro antifungal actions of Origanum essential oil, as well as povidone iodine aqueous solution in opposition to *C. albicans*, were compared. Thus resulting in a significant impact on the growing trend in the usage of herbal products in medicine.

C. Albicans is a dimorphic, yeast-like fungus residing as a commensal in the mucocutaneous areas in the body like the skin, the vagina and the intestine of humans. It can lead to infections under altered physiological and pathological circumstances such as “infancy, pregnancy, diabetes, prolonged broad spectrum antibiotic administration, steroidal chemotherapy and acquired immune deficiency syndrome”.⁹

The Origanum oil commonly known as Ajwain is a perennial creeper cultivated in most parts of India. The essential oil extracted from *O. Vulgare* has an inhibitory outcome on fungal species of clinical importance.^{10,11} Therefore, this study aimed to compare the antifungal activities of *O. Vulgare* essential oil and povidone-iodine aqueous solution on *C. Albicans*.

To check the efficacy as well as analysing the antimicrobial action of origanum oil against povidone-iodine solution an accepted and effective method of Agar well diffusion was used.¹² The clear zone [area of inhibition] by the above technique for origanum oil was found to be >40 mm which was comparatively higher than the povidone iodine aqueous solution. Thus a conclusion was driven that *C. Albicans* was most sensitive for *O. Vulgare* oil when compared to the povidone-iodine aqueous solution.

Manohar et al.⁶ in which *C. Albicans* was found to be sensitive to *O. Vulgare* at a concentration of 0.2% using broth micro-dilution method. Another study reported by Adams et al.¹³ found that higher antifungal properties are seen in Origanum oil due to its main content being carvacrol^{6,14} and are responsible for the antifungal and antibacterial activities. They cause alterations in the cell membrane configuration ensuing in the interruption of the permeability barrier of microbial membrane formation.⁶

In the patients with an ocular prosthesis, advice to clean the prosthesis with soap solution is given to maintain its hygiene.¹⁵ In this study, an antifungal solution of *O. vulgare* oil which can aid in maintaining the hygiene of the prosthesis was found to be a more effective antifungal agent against *C. Albicans* when compared to the povidone-iodine aqueous solution. This study helps in promoting traditional medicine form of modern dentistry, as *O. Vulgare* essential oil is easily accessible, economically feasible, culturally acceptable and less toxic.

CONCLUSION

This study encourages the researcher to assess the effectiveness of *O. Vulgare* oil in other forms of systemic and superficial fungal infections as well as to investigate its broad-spectrum outcome against other pathogenic manifestations which also include malignancies. Further research may be performed to examine the detailed mechanism of the action

of *Origanum* oil in various metabolic behaviour in humans, which will, in turn, be helpful to mankind.

ACKNOWLEDGEMENT

Authors acknowledge the immense help received from the scholars whose articles are cited and included in references of this manuscript. The authors are also grateful to authors/editors/publishers of all those articles, journals and books from where the literature for this article has been reviewed and discussed.

Source of Funding: None

Conflict of Interest: None

Authors' Contribution: All the authors have contributed to the planning, implementation and analysis of the research study and its presentation in the form of the manuscript.

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