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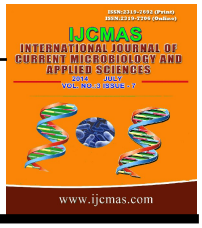

Original Research Article

Alternative control techniques against fungal colonization for preserving monument Deterioration

Preeti Bhatnagar[1*] and S.K.Jain[2]

[1]College of Life Sciences, CHRI, Gwalior (MP), India
[2]Department of Microbiology Vikram University, Ujjain (M.P.), India
**Corresponding author*

A B S T R A C T

A magnificent palace like Gwalior fort, in Madhya Pradesh, India has survived from medieval period. Microorganisms and other deteriorating agent can modify the composition & structure of artifact. Fungi are complex communities of microorganisms that damage historic monuments. It is preferable to eliminate biological growth for conservation reasons and maintain the original status and integrity of the heritage site [13,16]. Relatively little research has been conducted on antifungal treatments for stone. Thus present study was taken to determine the efficacy of natural products against monument associated fungi so as to determine their antifungal potential to prevent colonization of these fungi. In this study samples were collected from Gwalior fort in Madhya Pradesh, India. The antifungal activity of twelve different essential oil of *Pluchea lanceolata*, *Carum copticum, Ocimum sanctum, Mentha piperata, Allium sativum, Tagetes minuta, Cinnamomum zeylanicum, Pinus pinaster, Eucalyptus globulus, Syzygium aromaticum, Mentha arvensis, Cedrus deodara* were assayed against *Alternaria , Aspergillus, Curvularia Penicillium , Fusarium* isolated from the tested monument site. Results indicate that the antifungal activity of the essential oils is different. Essential oils from *Cinnamomum zeylanicum, Eucalyptus globulus,* and *Cedrus deodara* were found to be inhibitory for all of test fungi. These oils showed higher efficacy against all the tested fungi than the control. On the other hand *Pluchea lanceolata, Allium sativum, Tagetes minuta, Mentha arvensis* oils were seems to be poor in this respect since no inhibition zone was observed. *Mentha piperata* oil was highly effective against *Curvularia sp,* while moderately active against *Penicillium sp, Fusarium sp, and A. nidulans* however *Alternaria sp was* resistant to tested oil. *Syzygium aromaticum o*il showed to have significant activity against all tested fungi but more effective against *Alternaria* & *A. nidulans*. *Ocimum sanctum* oil was also active against all fungi but most effective against *Curvularia sp*.

Keywords

Essential oils, Antifungal activity, Deterioration, Conservation.

Introduction

The biodeterioration of archaeological monuments by microorganisms is a world wide problem thus adequate interventions must be taken to stop or at least slow down the process of biodeterioration. Any biocide intended for use on historic monuments and

rock sites must be not only effective against biological growths but also not damage stone material [4, 13]. The product should be chosen on the basis both of the organisms to be eliminated and of its highest efficacy at lowest dosage [3].

The antimicrobial properties of volatile aromatic oils from plants against both plant and human pathogenic microorganisms have been recognized since antiquity [2, 10]. The present study is designed to determine if these properties may also be useful for the control of stone monument associated fungi. This concern has help to look for other solutions to synthetic pesticides.

Materials and Methods

Alternaria sp, A. nidulans, Curvularia sp, Penicillium sp, Fusarium sp isolated from Gwalior fort, India was selected as test fungi.

The Twelve essential oil of *Pluchea lanceolata, Carum copticum, Ocimum sanctum, Mentha piperata, Allium sativum, Tagetes minuta, Cinnamomum zeylanicum, Pinus pinaster, Eucalyptus globulus, Syzygium aromaticum, Mentha arvensis, Cedrus deodara* obtained from the Department of pharmacy, Jiwaji University, Gwalior, India, were used in this study.

Anti-microbial assays were performed on plates containing Mueller-Hinton agar supplemented with 2% glucose and 0.5 µg methylene blue per ml [15]. Concentration of test mold inoculum is adjusted spectrophotometrically to contain $0.4x10^4$ to $5x10^4$ CFU/ml by OD determination at 530 nm. For *Aspergillus* sp. the OD was adjusted to 0.09-0.11; for *Fusarium* sp. the OD was adjusted to 0.15-0.17; and for *Alternaria* sp. the OD was adjusted to 0.2. Tween 80 (0.05%) was added as wetting agent in the preparation of each inoculum. Antifungal activity of the essential oils was tested using the disk diffusion method [1].

Inhibitory zones measured after 48 hrs at 28 ± 2^0C. A set of control was also run in same way for each test organism using Amphotericin B (Table1.1).

Results and Discussion

Eight essential oils were found to be inhibitory for the mycelial growth of all test fungi. Essential oils from *Cinnamomum zeylanicum, Eucalyptus globulus,* and *Cedrus deodara* were found to be inhibitory for all of test fungi. While *Pluchea lanceolata, Allium sativum, Tagetes minuta, Mentha arvensis* were seems to be poor in this respect. *Mentha piperata* oil was highly effective against *Curvularia sp,* while moderately active against *Penicillium sp, Fusarium sp, and A. nidulans* however *Alternaria sp was* resistant to tested oil. *Syzygium aromaticum o*il showed to have significant activity against all tested fungi but more effective against *Alternaria* & *A. nidulans*. *Ocimum sanctum* oil was also active against all fungi but most effective against *Curvularia sp*. Compared to the inhibitor control, amphotericin B, some of the essential oils produced larger zones of inhibition around the test fungi (Table1.1).

Essential oils and their constituents have a long history of applications as antimicrobial agents, but their use as stone preservatives has not yet been reported. The antifungal activity of the volatile oils found different, depending on the mould type and the oil used. Essential oils from *Cinnamomum zeylanicum*, Eucalyptus, and *Cedrus deodara* were found to be inhibitory for all of test fungi [8]. The antimicrobial effect of eucalyptus essential oil was reported on the mycelial growth and toxigenesis of *Penicillium aurantiogriseum* and *Penicillium viridicatum* [7].

Table.1.1 Antimicrobial activity of essential oils against fungi.

S.no	Essential oils	Fungal Cultures (Zone diameter in mm)*				
		Penicillium sp	*Fusarium sp*	*Curvularia sp*	*Alternaria sp*	*Aspergillus nidulans*
1	*Pluchea lanceolata*	R	R	R	R	R
2	*Carum copticum*	14	60	55	25	33
3	*Ocimum sanctum*	18	30	28	22	19
4	*Mentha piperata*	12	12	17	R	10
5	*Allium sativum*	R	R	R	R	R
6	*Tagetes minuta*	R	R	R	R	R
7	*Cinnamomum zeylanicum*	57	57	90	52	90
8	*Pinus pinaster*	35	37	47	14	40
9	*Eucalyptus globulus*	34	28	54	22	31
10	*Syzygium aromaticum*	28	12	31	90	90
11	*Mentha arvensis*	R	R	R	R	R
12	*Cedrus deodara*	39	49	29	24	68
13	Control	21	R	31	27	19

*Values are mean of three replicates, R=resistant

Mentha oil was not found effective against environmental isolates though it is reported as active antifungal agent against otomycotic fungi [9]. The essential oil of *Carum copticum* was found to be effective against *Fusarium & Curvularia* sp [17].

Clove oil was found to be inhibitory against assayed fungi. The antimicrobial activity of clove oil has been attributed to the presence of some active constituents in the oil [5,12].

Pinus pinaster, Ocimum sanctum oil was found to be active against all the test fungal isolates. These results are in high accordance with previous researches on fungi isolated from stone [14,11,18].

The vapors of Peppermint oil and two of its major constituents (menthol & menthone) were previously reported against *Sclerotinia* in a closed system[6].

This study reveals that many essential oils possess antifungal activity against monument associated fungi. These findings support the application of essential oils for surface treatment or vapor exposure of stone to prevent mold infestation. As a final conclusion of this research, the uses of oils can be recommended to be used as sporicidal agents in biocidal formulations.

References

1.Bauer AW, Kirby W M M, Sherris J.C, Turck M, 1966, Antibiotic susceptibility testing by a standardised single disk method. *Am J Clin Path*;45:493-496

2.Burt, S. (2004). Essential oils: their antibacterial properties and potential applications in foods – a review. *Int J Food Microbiol* 94: 223–253

3.Caneva, G., Nugari, M.P., Pinna D. and

Salvadori, O. 1996.*Controllo del degrado biologico*. Fiesole: Nardini.

4.Ciarallo, A., Festa L., Piccioli, C. and Raniello, M. 1985. Microflora action in the decay of stone monuments. In *Vth International Congress on Deterioration and Conservation of Stone*, Vol. 2: 607-616. Lausanne: Presses Polytechniques Romandes.

5.Duke, J.A., 1985 Handbook of medicinal herbs.CRC Press,Inc.,Boca Raton,Fla.

6.Edris A.E., and Farrag E.S. 2003. Antifungal activity of peppermint and sweet basil essential oils and their major aroma constituents on some plant pathogenic fungi from the vapor phase. Nahrung. 47(2):117-121.

7.Ezziyyani M, Lamarti A, Tantaoui-Elaraki A; Khaddor M 2006. Antifungal Activity of Three Essential Oils on Growth and Toxigenesis of *Penicillium aurantiogriseum and Penicillium viridicatum, Journal of Essential Oil* Research vol. 18, 5, 586-589

8.Giordani, R., P. Regli, J. Kaloustian, C. Mikail, L. Abou and H. Portugal. 2004. Antifungal effects of various oils against Candida albicans. Potentiation of antifungal action of amphotericin B by essential oil from Thymus vulgaris. Phytother. Res., 18(12): 990-995.

9.Jain S K, Agrawal S C.,1995.Antimycotic activity of some essential oils. *Asian journal of experimental Sciences,9,1&2,1-3*

10.Kordali S, Kotan R, Mavi A, Cakir A, Ala A, Yildirim A,2005.Determination of the chemical composition and antioxidant activity of the essential oil of Artemisia dracunculus and of the antifungal and antibacterial activities of Turkish Artemisia absinthium, A. dracunculus, Artemisia santonicum, and Artemisia spicigera essential oils, *J Agric Food Chem*, 53; 24, 9452-8

11.Krauze-Baranowska M. and Wiwart M. (2002), Antifungal activity of biflavones from *Taxus baccata* and *Ginkgo biloba*. Z. Naturforsch. **58c**, 65Ð69.

12.Matan N, Rimkeeree H, Mawson A J, Chompreeda P, Haruthaithanasan V, Parker M**.**2006, Antimicrobial activity of cinnamon and clove oils under modified atmosphere conditions. *Int J Food Microbiol*, Mar 15;107(2):180-5.

13.May, E., Lewis, F.J., Pereira, S., Taylor, S., Seward, M.R.D. and Allsopp, D. 1993. Microbial deterioration of building stone – a review. *Biodeterioration Abstracts*, *CAB International* 7(2): 109-123.

14.Mironescu M., Georgescu C., Oprean L .2009.Comparative sporicidal effects of volatile oils . *Journal of Agroalimentary Processes and Technologies*.*1 5*(3), 361-365.