Studies for antifungal activity of selected concrete sealers on white cement panels

Rajesh K. Verma, Deepa Devi

***Abstract* -- The aim of the present study was to assess efficacy of different concrete sealers against building fungi. The 0.5 ml of seven concrete sealers (Liquid Sealer LS-S, Magik impregnator, WEB-CBX, RIK- SEAL Medium Gloss, KONEX WRA - 2318, Evercrete DPS, La Guard PWC) and essential oil of peppermint and eucalyptus were tested for their antifungal activity against *Rhizopus stolinifer, Penicillium* chrysogenum and *Mucor racemosus* by poison food technique up to 120 hrs. The individual concrete sealers, mixture of evercrete DPS sealer with peppermint and eucalyptus oils were applied on white cement panel to evaluate their antifungal activity against *Mucor racemosus* up to 21 days and mixed culture of *Rhizopus stolinifer, Penicillium* chrysogenum and *Mucor racemosus* up to 4 weeks. The essential oils of peppermint, eucalyptus and WEB-CBX exhibited significant antifungal activity against *M. racemosus* on petri plate method. The peppermint oil, WEB-CBX, Magik Impregnator exhibited significant antifungal activity against *R. stolinifer*. However, peppermint oil, WEB-CBX, Evercrete DPS with or without eucalyptus oil exhibited significant antifungal activity against *P. chrysogenum*. The evercrete DPS with peppermint oil, WEB-CBX, Magik Impregnator and La Guard PWC exhibited significant antifungal activity on treated white cement panels in both *Mucor racemosus* and mixed culture of three fungi. This is the first report for antifungal activity of concrete sealers applied on the white cement panels against *R. stolinifer*, *P.* chrysogenum and *M. racemosus* in India. These findings would be useful to search new alternative of antifungal coating for concrete and building industries.**

***Index Terms* -- Antifungal activity, Bio-deterioration, Concrete sealers, Essential oil, Fungi.**

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I. INTRODUCTION

Microorganisms – bacteria, cyanobacteria, fungi, algae, and lichens – are liable to grow on building materials. They will not grow on building materials if there are no nutrients or carbon sources [1]. Biological activity contributes to deterioration of building material, and its interaction with physico-chemical mechanisms is responsible for long term deterioration [2]-[3]. Fungi are among the most harmful organisms associated to bio-deterioration of organic and inorganic materials [4]-[5]. Mortar and concrete are the most widely used building materials all over the world as they are cheap, easily available and convenient to cast [6]. Cement plays an essential role in concrete works behaviour, because it provides its mechanical resistance [7]. Concrete is mostly used to cast foundations, floors, and other horizontal structures and will hold soil, dirt, and dust better than vertical surfaces [8].

The fungi play an important role in the deterioration of concrete. Under humid conditions, the fungi biofilm was encouraged to grow on the surface of the normal concrete specimens and allowed to slowly degrade the concrete because of its interaction with the products of microbial metabolism [9]-[10]. It was reported that the oxalic acid excreted during the growth of fungal film interact with the concrete and form insoluble calcium complexes like calcium oxalate [11]. Fungi produce a wide range of organic acids like, acetic, oxalic and glucuronic acids [12]. They can also etch the concrete surfaces and favour penetration of fungal hyphae deep into the concrete which can result in enlargement of damaged area and increase in porosity [13]. Discoloration of concrete or paint on walls or nearby surfaces is usually an indication of presences of mould colonies. Of all the biological factors, fungi are responsible for the 80% of total building materials degradation [14]. Fungal growth in damp or water-damaged buildings has adverse effects on both the occupants and the buildings. The *Penicillium chrysogenum* and *Aspergillus versicolor* are the most common fungal species reported from water-damaged buildings [6]. Air sampling is unreliable because it favors fungi that produce large quantities of small, dry spores, such as *Aspergillus* spp., *Cladosporium* spp., and *Penicillium* spp. [15]. Besides, fungal diversity is different in outdoor air [16], in indoor air [17], and in house dust [18] compared with each other and with moldy building materials [19].

The presence of *Aspergillus niger, Ascotricha charatarum, Fusarium solani, Aspergillus spp, Penicillium spp., Cladosporium spp., Phoma spp., Stachybotrys spp. Ascospora spp., Curvularia spp, Alternaria* spp. were reported in indoor walls of the residential/ commercial constructions exposed to seepage [20]. The *Aspergillus flavus* 30.77%, *Penicillum chrysogenum* 21.27%, *Alternaria* 14.48%, *Aspergillus niger* 9.05%, Mucor 6.33%, *Rhizopus* 5.43%, *Fusarium* spp. 4.07%, *Trichoderma* 2.71%, Unknown spp. 2.26% were reported from religious, educational and industrial buildings of Haridwar

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[14]. The average daily hyphal growth of fungi rates were 4 and 16 times higher in the living rooms and in the bedrooms of the UGH user homes, respectively. Viable spores are always present in dwellings and germination and subsequent hyphal development can occur if the nutrients, temperature and moisture availability are suitable [21]. Fungi are essential to survival of global ecology but they may pose a significant threat to the occupants health when they grow in buildings. Fungal growth in buildings is influenced by source of infection, nutrient, temperature, relative humidity, oxygen, etc. and affects indoor air quality [22]. In air and surface samples from hospitals, offices and residential buildings of Roorkee, India, *Alternaria*, *Aspergillus*, *Penicillium*, *Rhizopus* and *Trichoderma* were found present in wall of all types of buildings. However, *Aspergillus*, *Geotrichum*, *Mucor*, *Penicillium* and *Rhizopus* were found in indoor environment [23].

Detection and species identification of all fungi present in a moldy building are the first step toward resolving the cause and effect of building-related illness (sick building syndrome). Dieldrin, pentachlorophenol (PCP) and tributylin oxide (TBTO) are most widely used to control fungal decay in buildings [24]. These synthetic fungicides are removed from markets because of their harmful effects on the environment and in effectiveness due to development of fungi resistance. Concrete sealers are applied to concrete to protect it from surface damage, corrosion, and staining. They either block the pores in the concrete to reduce absorption of water and salts or form an impermeable layer which prevents such materials from passing. The successful application of concrete sealer into reinforced concrete surface protect it from ingress of water, soluble salts and other contaminants [25]. Commercially available concrete sealers are based on silicon, silane, silicate, siliconate, acrylic, epoxy, urethane, etc. However, these were not assayed for their antifungal activity against fungi occurring in building. Eucalyptus and peppermint oils were found effective against eleven filamentous fungi [26]. Menthol was found to be individual aroma responsible for the antifungal activity of peppermint essential oil [27]. The peppermint essential oil exhibited strong antifungal activities against the examined fungi at concentrations ranging from 0.12 to 8.0 μL/mL [28]. Antifungal activities of essential oil and methanolic extracts of peppermint were demonstrated [29]-[32]. The essential oils from three *Eucalyptus* species showed significant antifungal activity against *Sclerotium rolfsii*, *Fusarium solani* [33]. The essential oil of *Eucalyptus* possesses good antifungal potential against eleven fungal strains [34]-[35]. The papers deals to evaluate selected concrete sealers (Liquid Sealer LS-S, Magik impregnator, WEB-CBX, RIK- SEAL Medium Gloss, KONEX WRA - 2318, Evercrete DPS, La Guard PWC) with or without essential oils of *Eucalyptus globulus* and *Mentha piperita* against three building fungi to determine their antifungal activity by poison food technique and applying on the white cement panels.

II. EXPERIMENTAL PROCEDURE

A. Building Fungi

The *Rhizopus stolinifer*, *Penicillium* chrysogenum and *Mucor racemosus* were isolated from the surface and indoor air of building, respectively [23] and have been maintained on potato dextrose agar (Himedia) in Building Pests and Mycology Laboratory, Environmental Science and Technology Group, CSIR-Central Building Research Institute, Roorkee, India.

B. Essential oils

The fresh leaves of *Eucalyptus globulus* (Myrataceae) and *Mentha piperita* (Lamiaceae) were collected / purchased from plants / superstore for essential oil extraction. The plant materials were identified and voucher specimens were submitted in the herbarium of Building Pests and Mycology Laboratory, Environmental Science and Technology Group, CSIR-Central Building Research Institute, Roorkee, India. Plant materials were thoroughly washed twice with distilled water and subjected to Clevenger's hydro –distillation apparatus to isolate essential oils. The isolated essential oil was dehydrated with anhydrous sodium sulphate (100 mg ml^{-1}) and kept on stand for half an hour. There after, upper layer containing dehydrated essential oil was collected with the help of micropipette and stored in clean glass vial at 4^oC [36].

C. Concrete Sealers

The concrete sealers were purchased directly from their manufactures / suppliers of India. The selected concrete sealers and their specific feathers and symbols are presented in Table-1.

D. Antifungal activity on Petri plates

All the concrete sealers (Liquid Sealer LS-S, Magik Impregnator, WEB-CBX, RIK- SEAL Medium Gloss, KONEX WRA - 2318, Evercrete DPS, La Guard PWC), essential oils of *Eucalyptus globulus* and *Mentha piperita* were individually evaluated for their fungi toxic activity against *Rhizopus stolinifer*, *Penicillium chrysogenum* and *Mucor racemosus* by adopting food poisoning technique into a petridish [37]-[38]. For each fungal isolate, a conidial spore suspension of 10^6 spores ml^{-1} was prepared and petridish were needle-inoculated in the centre. The potato dextrose agar (Himedia) nutrient medium was poisoned with 0.5 ml of test concrete sealer or essential oil. The radial growth was measured in millimeters after incubated at 28 ± 0.5^0C and 85% relative humidity up to 120 hours in triplicate.

E. Antifungal activity on white cement panels

White cement test panels (150×65×5 mm) were prepared by mixing a ratio of 1:1 of white cement and sand. The slurry of white cement and sand was poured on the wooden mould to cast cement panels. White cement test panels were cured for

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28 days before antifungal studies. The white cement panels were dried and divided into two equal parts. After the partition, one side of the panel was labeled as UT (Untreated) and the other side was labeled by the name of concrete sealer abbreviation. The evercrete DPS was separately mixed with 5 % essential oil of peppermint and eucalyptus oils. All the panels were treated by painting brush on treated part of panels with appropriate concrete sealers with or without essential oils. The second coat was applied after 24 hours from first coat.

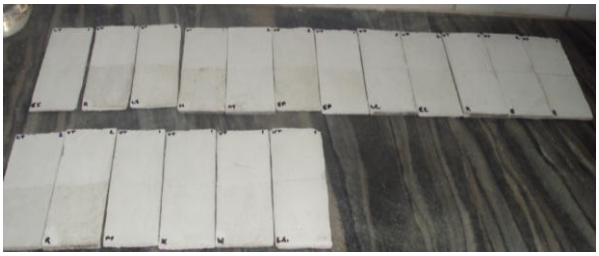
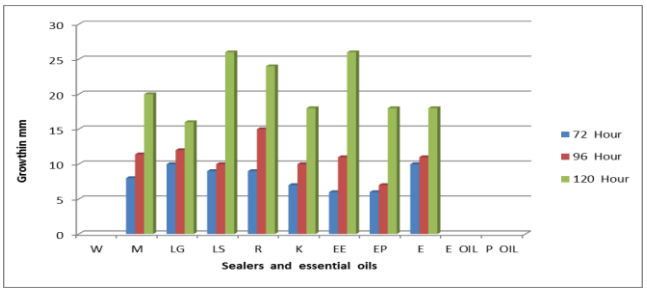


Fig.1. Concrete sealer coated white cement panels for antifungal study

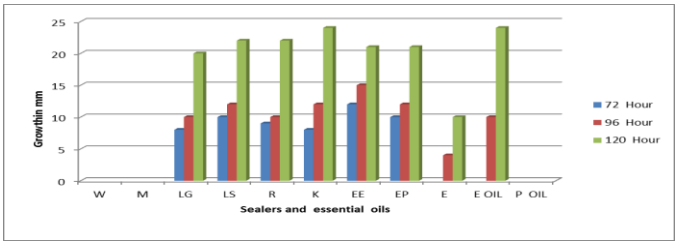
The potato dextrose agar medium was prepared and autoclaved at 121^0C for 15 min. Poured the 200 ml of culture media into each sterile petri plates (200x20 mm) and allowed to solidify. The coated panels were exposed to UV radiation for 120 min. to sterilize them. The white cement panels were placed on the center of petri plates over the solidified medium. The inoculation of fungal species was done using conidial spore suspension of 10^6 spores ml^{-1} of *Mucor recemosus* and mixed culture of *Rhizopus stolinifer*, *Penicillium chrysogenum* and *Mucor racemosus*. The plates were incubated at 28 ± 0.5^0C and 85% for 21 days for mixed fungi culture and 28 days for *Mucor racemosus* to check antifungal activity of concrete sealers in duplicate.

III. RESULTS AND DISCUSSION

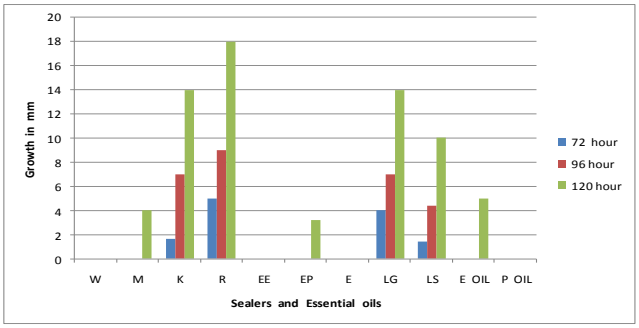
Fungi are a natural part of our environment and play an important in decomposition of organic matter. They can grow on almost any building material if there is enough moisture available and cause damage to the structure [39]-[40]. Antifungal activity of various concrete sealers and essential oils of *Eucalyptus globulus* and *Mentha piperita* against *Mucor racemosus*, *Rhizopus stolinifer* and *Penicillium chrysogenum* are presented in the Graph 1 to 3.



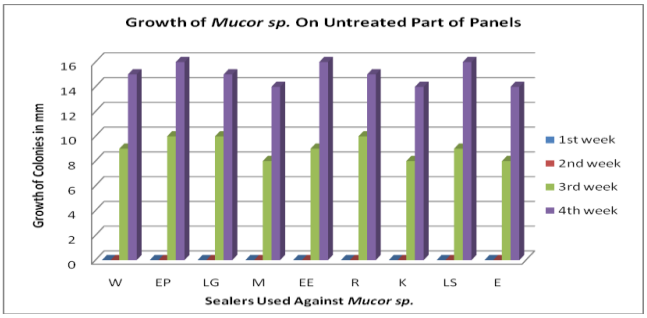
Graph 1: Antifungal activity of tested concrete sealers and essential oils against *Mucor racemosus.*



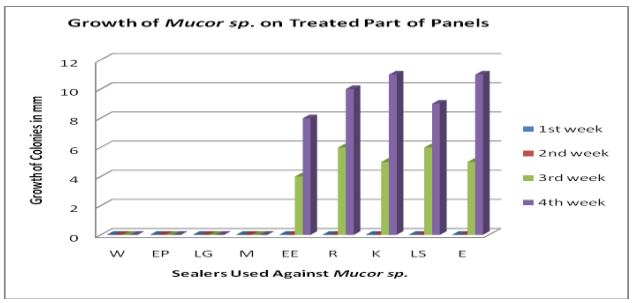
Graph 2: Antifungal activity of tested sealers and essential oils against *Rhizopus stolinifer.*



Graph 3: Antifungal activity of tested concrete sealers and essential oils against *Penicillium chrysogenum.*



Graph 4: Diameter of *Mucor recemosus* on untreated part of white cement panels.



Graph 5: Diameter of *Mucor recemosus* on treated part of white cement panels.

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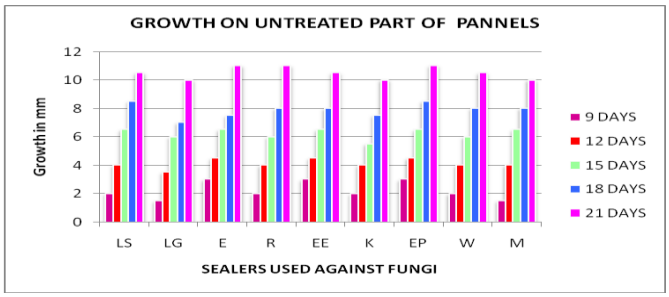
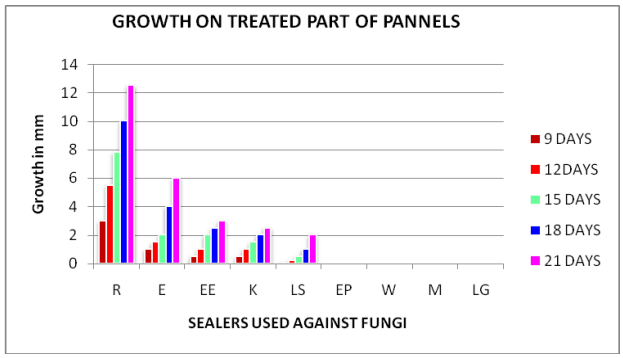


Graph 6: Diameter of fungal colonies on untreated part of white cement panels.



Graph 7: Diameter of fungal colonies on treated part of white cement panels.

From Graph 1, it is shown that WEB – CBX (W) exhibited significant antifungal activity on *Mucor racemosus* after incubation of 72 - 120 hrs. However, other concrete sealers (Liquid Sealer LS-S (LS), Magik impregnator (M), RIK- SEAL Medium Gloss (R), KONEX WRA – 2318 (K), Evercrete DPS (E), La Guard PWC (LG) exhibited moderate / least antifungal activity.

From Graph 2, it is shown that WEB-CBX (W), Magic Impreganator (M) exhibited significant antifungal activity on *Rhizopus stolinifer* after incubation of 72 – 120 hrs. However, all other concrete sealers (Liquid Sealer LS-S (LS), RIK- SEAL Medium Gloss (R), KONEX WRA – 2318 (K), Evercrete DPS (E), La Guard PWC (LG) exhibited moderate / least antifungal activity.

From Graph 3, it is shown that WEB-CBX (W), Evercrete DPS (E) and Evercrete with eucalyptus oil (EE) exhibited significant antifungal activity on *Penicillium chrysogenum* after incubation of 72 - 120 hrs. However, Evercrete DPS with peppermint oil (EP) and Magic Impreganator (M) exhibited moderate antifungal activity, while Knoex WRA (K), Liquid Sealer (LS), La Guard (LG) and Rik Seal medium gloss (R) exhibited least antifungal activity.

From Graph 1 to 3, it is shown that essential oils of Eucalyptus and Peppermint oils exhibited significant antifungal activity on *Rhizopus stolinifer*, *Penicillium chrysogenum* and *Mucor racemosus* after incubation of 72 – 120 hours. No fungal growth was appeared in petri plates treated with both essential oils.

The fungal growth of *Mucor recemosus* on untreated and treated parts of white cement panels are presented in Graph 4 and 5. From Graph 4, it is recorded that in the case of WEB – CBX (W), Evercrete DPS with peppermint oil (EP), La Guard (LG), Magik Impregnator (M), Evercrete with Eucalyptus oil (EE), Rik – seal medium gloss (R), Konex WRA (K), Liquid Sealer (LS) & Evercrete (E) the growth of *Mucor recemosus* were appeared as 15 mm, 16 mm, 15 mm, 14 mm, 16 mm, 15 mm, 14 mm, 16 mm and 14 mm, respectively.

From Graph 5 it is shown that there is no fungal growth was appeared on white cement panels coated with WEB-CBX (W), La Guard (LG), Magik imperegnator (M) and Evercrete DPS with Peppermint oil (EP). However, little growth of fungus was appeared in cases of evercrete DPS with eucalyptus oil (EE), Rik seal medium gloss (R), Knoex WRA (K), Liquid sealer (LS) and evercrete (E). The diameter of grown *Mucor recemosus* were appeared as 8 mm, 10 mm, 11 mm, 9 mm and 11 mm, respectively at the end of 4 weeks. It is shown that when peppermint oil was added to the Evercrete DPS (E), the growth of *Mucor recemosus* gets inhibited completely as compared to evercrete DPS.

The diameters of mixed fungal colonies grown on untreated and treated parts of the panels are presented in Graph 6 and 7. After comparison of both graphs (6 & 7), we observed that WEB-CBX (W), La Guard PWC (LG), Magic impregnator (M), Evercrete DPS with peppermint oil (EP) exhibited highest antifungal activity against mixed culture of fungi.

From Graph 6 and 7 it is shown that WEB-CBX (W), La Guard (LG), Magik imperegnator (M) and Evercrete DPS with Peppermint oil (EP) exhibited highest antifungal activity on white cement panel after the incubation of 21days in case of mixed fungi culture. Liquid sealer (LS), Konex- WRA (K) and Evercreat-DPS with Eucalyptus oil (EE) exhibited moderate antifungal activity, while Evercrete-DPS (E) exhibited least antifungal activity.

Evercrete DPS with peppermint oil (EP), WEB-CBX (W), La Guard (LG) and Magik impregnator (M) showed complete inhibition of fungal colonies on white cement panels in case of mixed culture of fungi and *Mucor recemosus*. It was concluded that no fungal growth was occurred on the treated part of white cement panels after 21 days. The concrete sealers WEB-CBX (W), La Guard PWC (LG), Magic impregnator (M), Evercrete DPS with peppermint oil (EP) were found effective to prevent the fungal (*Rhizopus stolinifer*, *Penicillium* chrysogenum and *Mucor racemosus*) growth on white cement panels incubated in BOD incubator.

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Table.1 Selected concrete sealer with their specific feathers, dilutions and symbols.

S.N.	Product	Specific Feature	Dilution	Symbol
1	Liquid Sealer LS-S	• Silane • Apply on old construction • UV stable • Maintains natural appearance • Clear	Dilutable 10 times with thinner	LS
2	Magik Impregnator	• Acrylic • Excellent stain proof property • UV stable • Transparent • Good resistance against atmospheric condition	Without Dilution	M
3	WEB-CBX	• Silane • Water repellent • Suited to restoration work • Clear appearance	Dilute only if necessary with distilled water	W
4	RIK- SEAL Medium Gloss	• Acrylic • UV resistant • Eco-friendly • Block penetration of dust, oil and chemicals • Clear appearance	Without Dilution	R
5	KONEX WRA - 2318	• Silane • Reduces water permeability • Prohibits fungal growth and foul smell • Clear appearance	Dilutable 14 times with water	K
6	Evercrete DPS	• Silicate • Penetrate upto 3 cm • Protects against chloride ion ingress • Anti-carbonation coating	Without Dilution	E
7	Evercrete DPS with Eucalyptus oil	• Silicate • Penetrate up to 3 cm • Protects against chloride ion ingress • Anti-carbonation coating • Anti- Fungal	Without Dilution	EE
8	Evercrete DPS with Peppermint oil	• Silicate • Penetrate up to 3 cm • Protects against chloride ion ingress • Anti-carbonation coating • Anti- Fungal	Without Dilution	EP
9	La Guard PWC	• Clear • Water Repellent	Without Dilution	LG

REFERENCES

1. M. Chauhan and P. Singh, "Antifungal evaluation of medicinal plants to control the growth of mould fungi on building material, World Journal of Pharmaceutical Research," vol. 8, pp. 1531-1541, 2014.
2. V. Wiktor, F. De Leo, C. Guyonnet, R. Grosseau and E. Garcia-Diaz, "Accelerated laboratory test to study fungal biodeterioration of cementitious matrix," International Biodeterioration and Biodegradation, vol. 63, pp. 1061-1065, 2009.
3. C. Saiz-Jimenez, "Biodeterioration vs Biodegradation the role of microorganisms in the removal pollutants deposited on historic building," International Biodeterioration and Biodegradation, vol. 40, pp. 225-232, 1997.
4. C. Urzì and P. Albertano, "Studying phototrophic and heterotrophic microbial communities on stone monuments," Methods in Enzymology, vol. 336, pp. 340-355, 2001.
5. C. Urzì, F.De Leo, "Sampling with adhesive tape strips: an easy and rapid method to monitor microbial colonization on monument surfaces," Journal of Microbiological Methods, vol. 44, pp.1–11, 2001.
6. R. K. Verma, L. Chaurasia, V. Bisht and M. Thakhur, "Bio-mineralization and bacterial carbonate precipitation in mortar and concrete," Bioscience and Bioengineering, vol. 1, pp. 5-11, 2015.
7. E. Guillon,"Durabilité des matériaux cimentaires : modelisation de l'influence des équilibres physico-chimiques sur la microstructure et les propriétés mécaniques résiduelles," PhD thesis, Ecole Normale Supérieure de Cachan, Cachan, France 2004.
8. B. Andersen, J. C. Frisvad, Ib. Sondergaard, Ib. S. Rasmussen and L. S. Larsen, "Association between fungal

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species and water damaged building materials," Applied and Environmental Microbiology, vol. 77, pp. 4180-4188, June 2011.
9. R.P. George, S. Ramya, D. Ramachandran, U. Kamachi Mudali, "Studies in biodegradation of normal concrete surface by fungus Fusarium sp.," Cement and Concrete Research, vol. 47, 8-13, 2013.
10. M. Fomina, V.S. Podgorsky, S.V. Olishevska, V.M. Kadoshnikov, I.R. Pisanska, S. Hillier, G.M. Gadd, "Fungal Deterioration of Barrier Concrete used in Nuclear Waste Disposal," Geomicrobiology Journal, vol. 24, pp. 643–653, 2007.
11. H.G.M. Edward, N.C. Russel and M.R.D. Seaward, "Calcium oxalate in lichen biodeterioration studies using FT-Raman Spectroscopy," Spectrochim. Acta A, vol. 53 pp. 99–105, 1997.
12. M.V. Dutton, C.S. Evans, "Oxalate production by Fungi: its role in pathogenicty and ecology in the soil environment," Candian Journal of Microbiololgy, Vol. 42 pp. 881–895, 1996.
13. J.D. Gu, T.E. Ford, N.S. Berke, R. Mitchell, "Fungal degradation of Concrete," in: W. Sand (Ed.), DECHEMA Monographs, vol. 133, Biodeterioration and Biodegradation, International Biodeterioration and Biodegradation Symposium, VCH Verlags Gesellschaft, Frankfurt, Germnay, pp. 135–142, 1996.
14. M. Chauhan and P. Singh, "Building deterioration fungi as biocontamint," Asian Journal of Experimental Biological Science, vol. 3, pp 209-213.
15. R. A. Samson, J. Houbraken, U. Thrane, J. C. Frisvad and B. Andersen, "Food and indoor fungi," 1st edition. CBS-KNAW Fungal Diversity Centre, Utrecht, Netherlands, 2010.
16. L. Larsen and S. Gravesen, "Seasonal variation of outdoor airborne viable microfungi in Copenhagen," Denmark. Grana vol. 30**,** pp. 467–471, 1991.
17. S. Gravesen, **"**Identification and quantification of indoor airborne micro- fungi during 12 months from 44 Danish homes," Acta Allergol, vol. 27, pp. 337 -354, 1972.
18. M. Wickman, S. Gravesen, S. L. Nordvall, G. Pershagen, and J. Sundell, "Indoor viable dust-bound microfungi in relation to residential characteristics, living habits, and symptoms in atropic and control children," Journal of Allergy Clinical Immunology, vol. 89**,** pp. 752–759, 1992.
19. S. Gravesen, P.A. Nielsen, R. Iversen, and K. F. Nielsen, "Microfungal contamination of damp buildings—examples of risk constructions and risk materials," Environmental Health Perspect, vol. 107, pp. 505–508, 1999.
20. J.R. Chunduri, "Indoor fungal populations inhabiting cement structures-remedial measures," IOSR Journal of Environmental Science, Toxicology and Food Technology, vol. 8, pp. 19-24, April 2014.
21. M. Boulic, R. A. Phipps, M. Cunningham, D. J. Cleland, P. Fjallstrom, K. Abe and P Howden-Chapman, "Heater choice, dampness and mould growth in 26 New Zealand homes: A study of propensity for mould growth using encapsulated fungal spores," Building, vol. 5, 149-162, 2015.
22. M. Kumar and R. K. Verma, "Fungi diversity, their effects on building materials, occupants and control-a brief review," Journal of Scientific and Industrial Research, vol. 69, pp. 657-661, 2010.
23. R. K. Verma and L. Chaurasia, "Studies on building fungi and their control with selected phytochemicals," in Annual Report *(2008-2009)* (CSIR-Central Building Research Institute, Roorkee India), pp. 9-13, 2009.
24. R. K. Verma, L. Chaurasia and S. Katiyar, "Potential antifungal plants for controlling building fungi," Natural Product Radiance, vol. 7, pp. 374-387, 2008.
25. R. K. Verma and Ajay Chourasia, "Protection of bio-deteriorated reinforced concrete using concrete sealers," International Journal of Materials Chemistry and Physics, In press 2015.
26. S. Pattnaik, V.R. Subramanyam and C. Kole, "Antibacterial activity of ten essential oils in vitro'" Microbias, vol. 86, pp. 237-246, 1996.
27. A. E. Edris and E. S. Farrag, "Antifungal activity of peppermint and sweet basil essential oils and their major aroma constituents on some plant pathogenic fungi from the vapor phase," Nahrung, vol. 47, pp. 117.121, 2003.
28. M. J. Saharkhiz, M. Motamedi, K. Zomorodian, K. Pakshir, R. Miri and K. Hemyarr, "Chemical composition, antifungal and antibiofilm activities of essential oil of *Mentha piperita*," ISRN Pharmaceutics doi: 10.5402/2012/718645, 2012.
29. S. Kizil, N. Has¸imi, V. Tolan, E. Kilinc¸, andU. Y¨uksel, "Mineral content, essential oil components and biological activity of two mentha species (*M. piperita L., M. spicata L.*)," Turkish Journal of Field Crops, vol. 15, no. 2, pp. 148–153, 2010.
30. A. K. Tyagi and A. Malik, "Liquid and vapour-phase antifungal activities of selected essential oils against *Candida albicans*: microscopic observations and chemical characterization of cymbopogon citratus," BMC Complementary and Alternative Medicine, vol. 10, article 65, 2010.
31. S. Behnam, M. Farzaneh, M. Ahmadzadeh, and A. S. Tehrani, "Composition and antifungal activity of essential oils of *Mentha piperita* and *Lavendula angustifolia* on post-harvest phytopathogens," Communications in Agricultural and Applied Biological Sciences, vol. 71, pp. 1321–1326, 2006.
32. N. Khaledi, P. Taheri and S. Tarighi, "Antifungal activity of varoius essential oils against Rhizoctonia solani and Macrophomina phaseolina as major been pathogen,' Journal of Applied Microbiology, vol. 118, pp. 704-717, 2014.
33. N. S. Kotterachchi, A. Sammani, D. B. Kelaniyangoda and R. Samarasekara, "Antifungal activity of essential oils of Ceylon Eucalyptus species for the control of *Fusarium solani* and *Sclerotium rolfsii*," Archives of Phytopathology and Plant Protection, vol. 45, pp. 2026-2025, 2012.
34. M. Riddhi, M. Patel and Y. T. Jasrai, "Antifungal potency of Eucalyptus globules essential oil against important plant pathogenic fungi," CIBTech Journal of Microbiology, vol. 4, pp. 42-52, 2015.
35. R. Mahato, S. Awasthi, S. Kumar and T. Bhatnagar, "A comparative study of effect of essential oil and alchoholic extract of Eucalyptus leaves on medically important bacterial and fungal isolates, International Journal of Current Microbiology and Applied Sciences, vol. 4, pp. 504-508, 2015.
36. A. Kumar, R. Shukla , P. Singh, C.S.Prasad and N.H. Dubey, "Assessment of *Thymus vulgaris* L essential oil as a botanical preservative against post harvest fungal infection of food commodities," Innovative Food Science Emerging Technology, vol. 9, pp. 575-580, 2008.
37. R. K. Grover and J.D. Moore, "Toximetric studies of fungicides against the brown rot organisms, *Sclerotinia*

ISSN: 2278 – 7798

fructicola and *S. laxa"* Phytopathology, vol. 52, pp. 876-879, 1962.
38. R. H. Garber and B. R. Houston, "An inhibitor of *Verticillium albo-atrum* in cotton seeds," Phytopathology, vol. 49, pp. 449-450, 1959.
39. R. K. Verma, L. Chaurasia and M. Kumar, "Antifungal activity of essential oils against selected building fungi," Indian Journal of Natural Product Research, vol. 2, pp. 448-451, 2011.
40. R. K. Verma, L. Chaurasia and M. Kumar, "Fungal deterioration in buildings," Biozone International Journal of Life Sciences, vol. 3, pp. 420-429, 2011.

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