

Comparative efficacy of tea tree oil nanoemulgel and clove oil nanoemulgel against *Candida albicans*

Dear Sir,

Candida, a type of fungus, can cause infections, which can be either local or systemic. The human body hosts small amounts of this fungus under normal conditions. However, problems arise when it begins to multiply and creates an overgrowth. Although more than 150 species of *Candida* exist, the majority of infections are caused by a species called *Candida albicans*. The prognosis for candida infections is often very good. In general, the condition is not serious and can be easily treated.^[1]

The majority of synthetic therapeutic agents used to treat candida infections suffer from various disadvantages such as they are toxic, expensive, and have to be administered frequently.^[2] To overcome the drawbacks of synthetic antifungal agents, the use of plant-derived products as disease control agents has been explored. These phytotherapeutics are reported to have low mammalian toxicity, less environmental effects, and wide public acceptance.^[3]

Plant-derived essential oils have been extensively used as natural antimicrobial agents, with applications ranging from pharmaceutical industry to food sector and cosmetic sector. These natural oils effectively inhibit the growth of a wide range of microorganisms, with fewer side effects as compared to synthetic antimicrobials available. Among the different essential oils explored, clove oil has been widely investigated due to its popularity, availability, and high essential oil content.^[4] Clove oil has been reported to possess strong antifungal activity against opportunistic fungal pathogens such as *C. albicans*. The essential ingredient responsible for its antifungal activity is eugenol.^[5]

Besides clove oil, tea tree oil (TTO) is another essential oil, whose therapeutic properties have come under increasing scrutiny both *in vitro* and *in vivo*. It is a volatile essential oil derived from *Melaleuca alternifolia*. It has been used extensively for its antimicrobial properties; TTO is incorporated as the active ingredient in many topical formulations for treating cutaneous infections, besides being marketed over the counter in Australia,

Europe, and North America for the treatment of various ailments.^[6]

In the present study, an effort has been made to compare the antifungal efficacy of both TTO and clove oil after incorporating them in nanoemulgel formulations. The results obtained in cup–plate microbiological assay were compared with those obtained for placebo gel containing carbopol 934P alone.

For the study, TTO, clove oil, and carbopol 934P were purchased from Sigma Aldrich Pvt. Ltd. (Bengaluru, India). Transcutol P was obtained as a gift sample from Gattefosse (Saint-Priest, Cedex, France). Tween 20 and polyethylene glycol were purchased from the Central Drug House, New Delhi, India. All other chemicals and reagents were of analytical grade and procured from Merck (Mumbai, India) and S. D. Fine Chem. (Mumbai, India). *C. albicans* MTCC No. 227 was procured from the Institute of Microbial Technology, Chandigarh, India.

The TTO nanoemulgel and clove oil nanoemulgel were prepared using aqueous titration method by incorporating carbopol 934P as a gelling agent. Plain carbopol 934P gel was made by accurately weighing carbopol 934P and dissolving in distilled water to get 2% w/v concentration.

In vitro antifungal activity using cup and plate method was done as per the procedure given by Maebashi *et al.* and Vijaya *et al.* From the *C. albicans* suspension (1×10^7 cfu/ml), 50 μ l suspension was taken and spread on Sabouraud dextrose agar plates aseptically with the help of sterile cotton swab. It was allowed to dry at room temperature with the lid closed. Then, three wells (3 mm diameter) were punched using sterile core borer into the agar medium and filled with TTO nanoemulgel (1 g), clove oil nanoemulgel (1 g), and plain carbopol 934P gel (1 g), respectively. The plates were then incubated at 28°C for 18–24 h and zone of inhibitions calculated.^[7,8] The results were expressed as mean \pm standard deviation (S.D.). The data obtained from various groups were statistically analyzed using GraphPad InStat 3 (GraphPad Software, San Diego, California), using

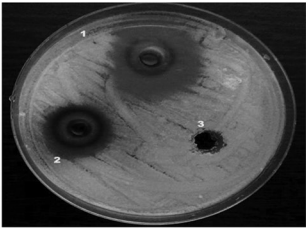
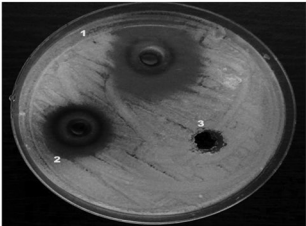
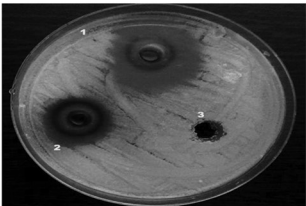
Incubation Time (hrs)	Observations of Zone of Inhibition for different formulations		
	<i>Candida albicans</i> (MTCC No. 227)		
24			
48			
72			
Where, 1 = Tea tree oil Nanoemulgel, 2 = Clove oil Nanoemulgel, 3 = Placebo carbopol 934 P Gel			

Figure 1: Observations of zones of inhibition for different formulations evaluated against strain of *Candida albicans* at different time intervals

two-tailed paired *t*-test. $P \leq 0.05$ was considered statistically significant.

The zone of inhibition for TTO nanoemulgel (38 ± 1.4 mm) was found to be significantly higher ($P \leq 0.05$) as compared to that of clove oil nanoemulgel (21 ± 1.5 mm) and plain carbopol 934P gel (00 mm). The larger zone of inhibition for TTO-loaded nanoemulgel could be attributed to more pronounced antifungal activity of TTO as compared to clove oil. As per reports in literature, TTO inhibits the growth of fungus by inhibition of synthesis of ergosterol, a component required for maintaining the integrity of the fungal cell walls. The results are given in Figure 1 and Figure 2.

It can thus be concluded that nanoemulgel of TTO significantly increases the antifungal activity of TTO against *C. albicans* as compared to clove oil nanoemulgel and carbopol 934P gel and therefore can be used successfully for the treatment of candidiasis.

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Conflicts of interest

There are no conflicts of interest.

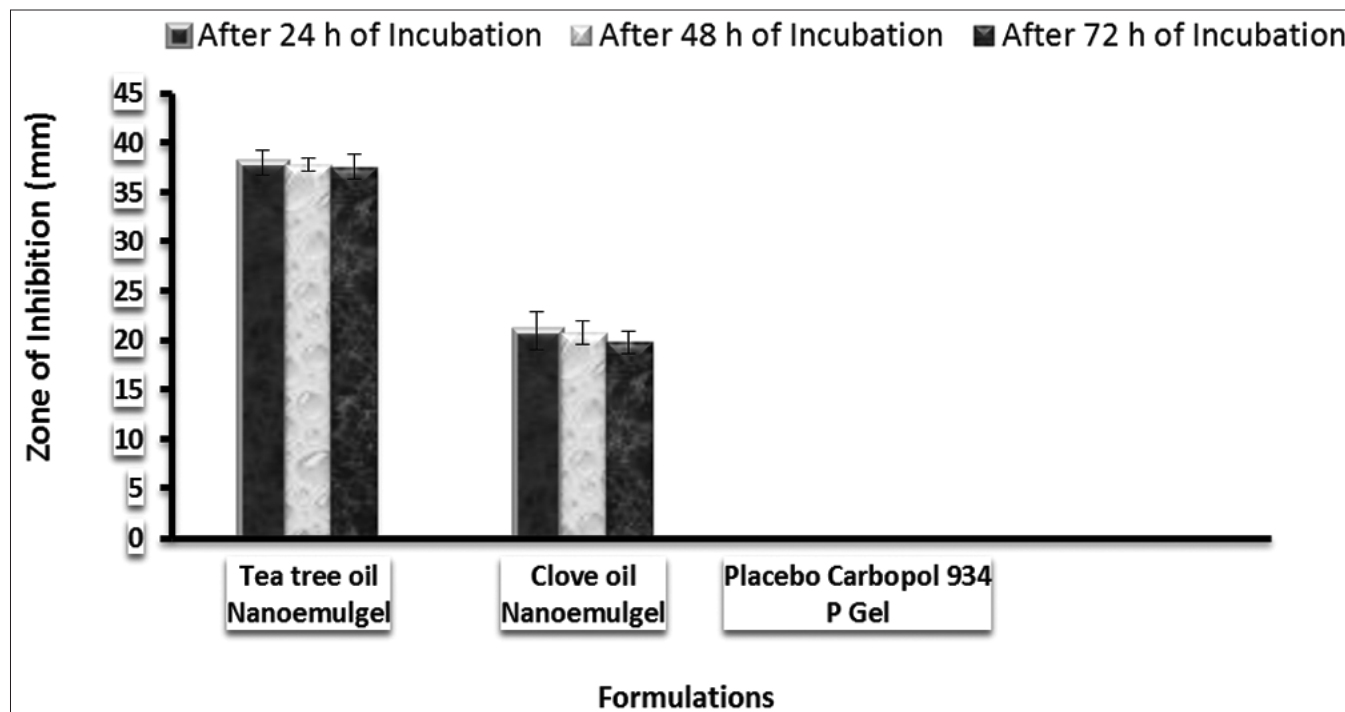


Figure 2: Comparison of zones of inhibition for different formulations during *in vitro* antifungal activity against *Candida albicans* (MTCC No. 227)

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