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## ORIGINAL RESEARCH

### Comparison of Fluconazole and Nystatin in tissue conditioner for treatment of denture stomatitis

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#### ABSTRACT:

**Background:** Tissue conditioning materials were introduced about 50 years ago. These are soft, resilient, temporary relining materials which by reducing and evenly distributing stresses on the mucosa of the basal seat, have a rehabilitating effect on unhealthy tissue and allow the condition to return to normal health. Denture stomatitis affects 65% of healthy adult mouths. **Aim of the study:** To compare Fluconazole and Nystatin in tissue conditioner for treatment of denture stomatitis. **Materials and methods:** The present study was conducted in the Department of Prosthodontics of the Dental institutions. Clinical isolates of *Candida albicans* were obtained from the Department of Molecular Microbiology of the institute, to use as test organisms for the current study. Antifungal agents, Nystatin and Fluconazole were mixed into tissue conditioner powder at concentrations of 5% wt/wt in a sterile plate. All disks were contaminated with 100 µl of  $1 \times 10^6$  CFU/ml *C. Albicans* cell suspension and the cell culture plate were incubated at 35° C on a rotary shaker for 48 hours. The plates were incubated at 37° C for 48 hours and the colonies were enumerated. **Results:** We observed that that Nystatin 5% solution was the most efficient for inhibiting attachment and colonization of *C. albicans* (0.22). Fluconazole 5% solution is partially effective efficient for inhibiting attachment and colonization of *C. albicans*. **Conclusion:** The incorporation of Nystatin and Fluconazole into tissue conditioners is effective treatment for chronic atrophic candidiasis in denture users. Nystatin is more efficacious as compared to Fluconazole.

**Key words:** Candidiasis, Fluconazole, Nystatin.

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#### Introduction:

Tissue conditioning materials were introduced about 50 years ago.<sup>1</sup> These are soft, resilient, temporary relining materials which by reducing and evenly distributing stresses on the mucosa of the basal seat, have a rehabilitating effect on unhealthy tissue and allow the condition to return to normal health.<sup>2</sup> Denture stomatitis affects 65% of healthy adult mouths.<sup>3</sup> Denture stomatitis is more commonly seen in the maxillary mucosa. The prevalence of denture stomatitis is varied from 15 to 65%<sup>1</sup> and even more significant in the institutionalized denture wearing population at up to 72%.<sup>4,5</sup> This

condition is usually asymptomatic, although it can be associated with burning, bleeding, an unpleasant taste. Denture induced stomatitis can be managed by either denture repair or replacement, prophylactic measures adopted by the patients and prescribing antifungal drugs. The use of antifungal agents for prevention and treatment of denture stomatitis causes undesirable taste and also needs frequently doses; thus, it is frequently associated with poor patient compliance.<sup>6</sup> Hence, the present study was conducted to compare Fluconazole and Nystatin in tissue conditioner for treatment of denture stomatitis.

**Materials and methods:**

The present study was conducted in the Department of Prosthodontics of the Dental institutions. The ethical clearance for the study was approved from the ethical committee of the hospital. Clinical isolates of *Candida albicans* were obtained from the Department of Molecular Microbiology of the institute, to use as test organisms for the current study. *Candida albicans* was cultured onto Sabouraud dextrose agar plate and incubated at 37° C for 3 days. A colony from the stock culture was then diluted in 2 ml sterile saline and a suspension of 1×10<sup>6</sup> CFU/ml was prepared. Tissue conditioner was mixed and prepared according to manufacturer's instruction. Antifungal agents, Nystatin and Fluconazole were mixed into tissue conditioner powder at concentrations of 5% wt/wt in a sterile plate. A sterile glass rod was used to prepare a thin film of tissue conditioner with 1 mm thickness and punched as 5 mm diameter disks. One specimens of pure tissue conditioner was also prepared as negative control. All disks were contaminated with 100 µl of 1 × 10<sup>6</sup> CFU/ml *C. Albicans* cell suspension and the cell culture plate

were incubated at 35° C on a rotary shaker for 48 hours. The plates were incubated at 37° C for 48 hours and the colonies were enumerated. The data was tabulated for further evaluation.

The statistical analysis of the data was done using SPSS version 11.0 for windows. Chi-square and Student's t-test were used for checking the significance of the data. A p-value of 0.05 and lesser was defined to be statistical significant.

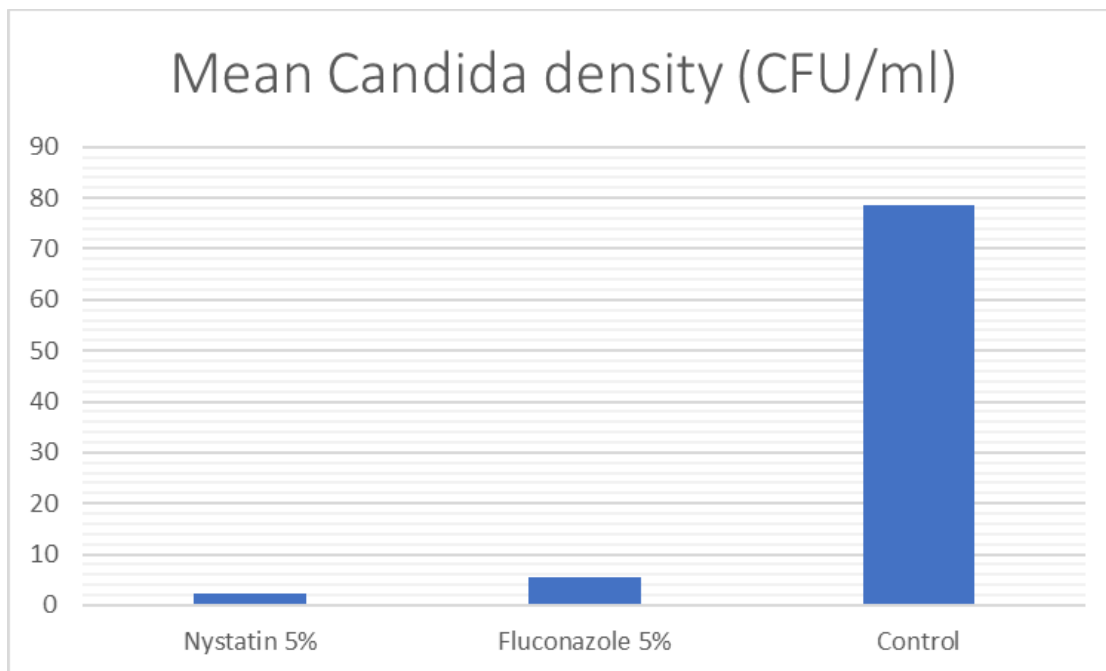
**Results:**

**Table 1** shows the mean *Candida* density (CFU/ml) for Nystatin 5%, Fluconazole 5% and control solution. We observed that that Nystatin 5% solution was the most efficient for inhibiting attachment and colonization of *C. albicans* (0.22). Fluconazole 5% solution is partially effective efficient for inhibiting attachment and colonization of *C. albicans*. The control solution was least effective with highest *Candida* density seen in control solution. The results were statistically significant (p<0.05) [Fig 1].

**Table 1: Mean Candida density (CFU/ml) for Nystatin 5%, Fluconazole 5% and control solution**

Specimens	Mean Candida density (CFU/ml)	p-value
Nystatin 5%	2.24	0.02
Fluconazole 5%	5.65	
Control	78.65	

**Figure 1:** Bar diagram showing mean candida density



**Discussion:**

In the present study nystatin showed higher inhibitory effects than fluconazole as it inhibited the production of *C. albicans* in tissue conditioner disks with high efficacy, however fluconazole could prevent the growth and adhesion of *Candida* only partially. The results were compared with studies from literature. Falah-Tafti A et al evaluated the efficacy of the two common antifungal agents mixed with tissue conditioner against *Candida albicans*. Tissue conditioner disks (Acrosoft) with 5mm diameter and 1mm thickness containing different concentrations of nystatin and fluconazole (1%, 3%, 5%, 10% wt/wt) as well as disks with no antifungal agents (8 disks for each group) were prepared for experimental biofilm formation by inoculation with *Candida albicans* cell suspensions. The specimens were incubated in cell culture microtiter plate wells containing Sabouraud's broth in a rotator shaker at 30°C for 48 hours. Then, the specimens were rinsed and sonicated in sterile water to remove surface organisms. The attached yeasts were enumerated by inoculation of the yeast suspension on Sabouraud's agar. The 1% to 10% mixture of nystatin and tissue conditioner completely inhibited the attachment and colonization of *Candida albicans*, although for fluconazole only a 10% concentration caused complete inhibition. Nystatin showed a potentially higher effect in inhibition of *Candida* attachment and colonization ( $P = 0.0001$ ) compared to that of fluconazole and a statistically significant difference was seen between 5% and 1% fluconazole. They concluded that tissue conditioner with 1% to 10% nystatin or 10% fluconazole can completely inhibit the adhesion and colonization of *Candida albicans*. Chow CK et al conducted study in which incorporated antifungal agents into tissue conditioners to investigate the effectiveness of this method of drug delivery. Combinations of nystatin, fluconazole, itraconazole and Coe Soft, Viscogel, Fitt were tested at 1, 3, 5, 7, 9 and 11 wt/wt%, with and without sterilized saliva. 6 mm diameter cores were punched in Sabouraud plates pre-grown with standardized *C. albicans*. Antifungal agents plus tissue conditioner mixtures were injected into each core. Inhibition diameters were measured for 14 days. Cores with only tissue conditioners acted as negative control and showed no significant inhibition activity. Peak activity was between 65 to 89 hours; followed by a plateau. Itraconazole had greater fungicidal activity than fluconazole; while nystatin was found to have the least fungicidal activity. The most effective concentration for nearly all combinations was 5% wt/wt. Specimens with saliva showed greater antifungal activity than those without. Itraconazole altered the physical properties of Viscogel hence this combination is not recommended for clinical use. It was concluded that the treatment of chronic atrophic candidiasis by

incorporation of antifungal drugs into tissue conditioners is efficacious. 5% wt/wt itraconazole mixed with Coe Soft or Fitt is recommended for clinical study where compliance of patient or care giver cannot be relied upon. Peak antifungal activity at 3 days suggests that mixtures prepared for clinical study may be replaced soon after this time for maximum effectiveness.<sup>7,8</sup>

Pachava KR et al investigated whether the incorporation of tea tree oil into denture soft liners would inhibit the growth of *Candida albicans*. Each 10 specimen disks incorporated with tea tree oil into soft liners (St) and without tea tree oil (S) were prepared. Both the tea tree oil daily. These disks were inoculated with *Candida albicans* suspension for assessment of fungal growth and were rinsed with sterile water to remove loosely attached surface organisms. The attached yeasts were measured by inoculating them on Sabouraud's agar. Treated and control disks were stored in distilled water for 1, 30, 60 days and washed daily with wet cotton. Data between treated and control disks were compared by applying t-test. The mean colony forming units (CFU) per mm<sup>2</sup> for specimens without tea tree oil after water storage and wash with wet cotton for 1, 30 and 60 days was  $7.1 \times 10^6$ ,  $6.5 \times 10^6$ ,  $6.8 \times 10^6$ , respectively and for specimens with tea tree oil CFU decreased significantly to  $2.1 \times 10^6$ ,  $2.8 \times 10^6$ ,  $3.1 \times 10^6$  after 1, 30 and 60 days. Treated disks were effective in controlling the growth of *C. albicans* for two months following water storage. They concluded that addition of tea tree oil to denture soft liner significantly reduced growth of *C. albicans* suggesting a new form of intra oral effective antifungal management for denture stomatitis. Jadhav V et al compared the hardness of two different tissue conditioners before and after the addition of three types of antifungal agents. Two types of tissue conditioners: viscogel and coe-soft were manipulated with three types of antifungal agents (fluconazole, clotrimazole, neem) and placed in the metal mold as per the manufacturer's instructions. A total of 240 samples were fabricated and divided into two groups of 120 samples each and were grouped as, T1 - Viscogel., T2 - Coe-soft. These were subjected to hardness test on 1 st , 7 th 14 th days with the Shore-A-Durometer equipment. Tissue conditioners showed an increase in hardness values with the use of antifungal agents in all the groups. The study can be concluded that hardness of Viscogel was statistically significant when mixed with fluconazole and compared with coe-soft as on 1 st day, 7 th day and 14<sup>th</sup> day.<sup>9,10</sup>

**Conclusion:**

Within the limitations of present study, we conclude that the incorporation of Nystatin and Fluconazole into tissue conditioners is effective treatment for chronic atrophic candidiasis in denture users. Nystatin is more efficacious as compared to Fluconazole.

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