Candida-Related Denture Stomatitis: A Pilot Study of the Efficacy of an Amorolfine Antifungal Varnish

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Purpose: *Candida albicans* is a component of the normal oral microflora, but local and systemic factors can transform this commensal *C albicans* to a pathogen. The most frequent cause of *Candida* opportunistic infections (candidiasis) is dentures, especially if poorly fitting or poorly cleaned. Management of oral candidiasis depends on an accurate diagnosis, identification and elimination of predisposing factors, and, often, use of antifungal agents. The aim of this study was to examine fingernail varnish, currently used for onychomycosis therapy, to reduce the fungal colonization in prosthetic biofilms. ***Materials and Methods:*** A varnish containing 5% amorolfine was applied once or twice a week for 6 months in six patients affected by nystatin-resistant denture-related stomatitis. In all six patients, the prostheses had previously been removed at night, and daily antimycotic topical therapy with nystatin had failed to resolve the stomatitis; after 30 days, these patients all showed persistence of candidal stomatitis. ***Results:*** After 1 month, five of the six patients were negative for *Candida;* this situation was unchanged in the following monthly controls. Only in the patient with suspected Sjögren syndrome was oral *Candida* found 15 days after the last varnish application. None of the patients had any complaints about the medication. ***Conclusion:*** This varnish containing 5% amorolfine, applied once or twice a week for 6 months, was able to suppress the nystatin-resistant denture-related stomatitis. *Int J Prosthodont 2005;18:55–59.*

Candida albicans is a component of the normal oral microflora, and up to 67% of people carry this microrganism without clinical evidence of infection.[1] Local and systemic factors can determine the transformation of *C albicans* from a commensal to pathogenic organism. The most frequent cause of *Candida* opportunistic infections is dentures, especially if poorly fitting or poorly cleaned.[2–5]

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Risk factors that may predispose the host to candidiasis are:

- Nighttime wearing of a prosthesis, with consequent acidity and proliferation of bacteria and yeasts
- Reduction of tissue resistance and increase in permeability of the epithelium to antigens and toxins, caused by trauma from a prosthesis
- Poor oral hygiene
- Allergy and irritating primary reaction to the constituents of the prosthesis
- Systemic factors, including general (diabetes, endocrine dysfunctions, anemia, malnutrition, neoplasia), drug therapy (antibiotics, oral contraceptives, corticosteroids), xerostomia, irradiation, immunosuppression (chemotherapy, corticosteroids), and HIV/AIDS[6–8]

Denture-related stomatitis presents different degrees of severity. According to a modified version of

Newton's classification, palatal inflammation can be divided into three grades: type I, with dispersed petechiae throughout all or any part of the palatal mucosa in contact with the denture; type II, with erythematic macules without hyperplasia; and type III, with diffuse or generalized erythema and papillary hyperplasia.[9]

Clinical examination cannot confirm if the stomatitis is due to *Candida,* a bacterial infection, or even an allergic reaction to the materials in the prosthesis. Only microscopic examination or culture testing can confirm the presence of *Candida* in the lesion.[10–12]

Management of oral candidiasis depends on an accurate diagnosis, identification and elimination of predisposing factors, and prescription of antifungal agents. When oral hygiene is not satisfactory, topical agents are sufficient. Nystatin and amphotericin are widely used, particularly for their ability to decrease the adhesion of the *Candida* to the prosthesis. Systemically administered antifungals are recommended for chronic, deep-seated infections and superficial cases refractory to topical agents. Ketoconazole, fluconazole, or amphotericin are the drugs most commonly used. These medications must be administered with particular caution because they can interact with anticoagulants and phenytoin and can be toxic.[10,13]

In denture-related stomatitis, biofilms are highly resistant to antifungal agents,[14–17] and *Candida* infection has often reestablished after antifungal therapy. Several clinical observations emphasize the importance of biofilm formation in candidiasis and the inability of antifungal drugs to eliminate the infection in a biofilm. A reproducible model of the *C albicans* biofilm has been established: Scanning microscopy, electronics, and fluorescence microscopy have demonstrated the presence of yeasts, hyphae, and similar hyphal structures on catheters.[18] Also, the observation of the *Candida* infection on prosthetic materials, such as methacrylate,[19] has shown the growth of the biofilm to be highly resistant to topical antifungal agents such as nystatin, amphotericin, fluconazole, and chlorhexidine.[20]

Amorolfine is a morpholinic derivative with both fungistatic and fungicidal activity. This agent inhibits the synthesis of ergosterol essential for the cellular membranes of *Candida,* in a manner similar to the azoles. In vitro, the antifungal activity is stronger against dermatophytes and yeasts other than *Candida.*[20] Amorolfine diffuses well into the keratinized tissues: In vitro after one application, *Candida* growth is inhibited for at least 10 days. After topical application on fingernails, the systemic absorption of amorolfine is negligible or nonexistent.[21,22]

The aim of this pilot study was to examine a new topical antifungal agent to reduce the fungal colonization in prosthetic biofilms. A type of varnish for fingernails, rich in amorolfine and currently suitable for onychomycosis therapy caused by dermatophytes, was used.

Materials and Methods

An open pilot study on the efficacy of a commercially available 5% amorolfine varnish in the control of nystatin-resistant denture-related stomatitis, approved by the local research ethics committee (amorolfine varnish is not currently licensed for intraoral use) was carried out. Because a possibly carcinogenic methacrylate copolymer is present in the preparation, the patients affected by hyperplastic candidiasis were excluded.

Six patients, aged 45 to 81 years, all with Newton type II denture-related stomatitis, were studied. Four patients used complete dentures, and two used removable partial prostheses. In all six patients, the prostheses had previously been removed at night, and daily antimycotic topical therapy with nystatin (Mycostatin, Bristol-Meyers Squibb) had failed to resolve the stomatitis; after 30 days, these patients all showed persistence of candidal stomatitis.

All patients but one were in apparent good health. One woman had xerostomia, lumbar spondylosis, and shoulder-blade periarthritis. Initially, Sjögren syndrome had been hypothesized, but sialography and anti-SSA and anti-SSB were negative.

Locetar 5% varnish (amorolfine chloride, copolymer of methacrylic acid, triacetin, butyl acetate, ethyl acetate, and ethyl alcohol) was applied to the denture once or twice a week for 6 months. Varnish was applied to the fitting surface of the prosthesis. Before varnish was applied, the surface of the prosthesis was carefully cleaned with gauze soaked with ethyl acetate solvent to remove every trace of varnish from the preceding application. All patients were examined before starting the treatment (day 0), during treatment (every 7 days for 1 month, days 90 and 180 from the first application), and after treatment (day 195 from the first application and day 60 from the last varnish application). All patients were subjected to examination of the palatal mucosa and quantitative cultures of *Candida* from the palatal mucosa and denture-fitting surface.

Triacetin, or glyceryl triacetate, is a cosmetic biocide, plasticizer, and solvent used in cosmetic formulations. It is a commonly used carrier for flavors and fragrances. Triacetin is a safe human food ingredient according to the Food and Drug Administration (FDA) and appears not to have any potentially dangerous effects.[23] Butyl acetate is a colorless and volatile liquid used as an industrial solvent in the production of essences, lacquers, photographic films, and fingernail varnish. It is characterized by low systemic toxicity, but possible symptoms include mucosal irritation, headache, mental confusion, nausea, vomiting, and cough. Ethyl acetate is an

Table 1 Evolution of *Candida* Infection According to Time and Varnish Application

Patient	Time (d)*								
	0	7	14	21	28	90	180	195	240
1	+	+	+	–	–	–	–	–	–
2	+	+	+	+	–	–	–	–	–
3	+	+	+	+	–	–	–	–	–
4	+	+	+	+	–	–	–	–	–
5	+	+	+	–	–	–	–	–	–
6	+	+	+	+	+	+	+	+	+

*Varnish was applied on days 0, 7, 14, 21, 28, 90, and 180.
+ = presence of *Candida* infection; – = absence of *Candida* infection.

inflammable solvent (soluble in water and oil), irritating to the eyes, respiratory streets, and skin. It has been associated with an increased risk of lymphatic leukemia in rubber workers who are heavily exposed.

To verify the presence of *Candida* on the mucosa and from the prosthesis, an Oricult-N test (Orion Diagnostica) was used at 3 days and then weekly. The Oricult-N semiquantitative dipslide method was developed for the enumeration of yeasts in clinical samples. In particular, it has been applied to the quantification of *Candida* species in the oral cavity.[24] The Oricult-N slide consists of Nickerson's medium containing chloramphenicol and gentamicin to control bacterial overgrowth. *Candida* species grow on it as characteristic brown-pigmented, smooth colonies.[24] The slides were incubated at 37°C for 2 days before examination, as recommended by the manufacturer. The numbers of colonies per square centimeter of agar surface were then counted. The number of yeasts per milliliter (range $< 10^3$ to 10^6 CFU/mL) was estimated by comparing the colony densities to color charts provided by the manufacturer.

Results

Twenty-eight days after the first varnish application, both examination of the palatal mucosa and quantitative cultures of *Candida* from the palatal mucosa and denture-fitting surface showed the complete disappearance of candidal infection in five of the six patients. Locetar varnish application was effective after 21 days from the first varnish application in two patients and after 28 days in three patients (Table 1). This situation was unchanged 195 days after the first application and 60 days after the last varnish application. Only in the patient with suspected Sjögren syndrome was oral *Candida* found after the last varnish application (Table 1). None of the patients had any complaints about the use of the varnish.

Discussion

Several antifungal medications have been developed for managing denture stomatitis associated with *Candida* infection. Generally, a topical and/or systemic approach can be used. Polyene agents (eg, nystatin, amphotericin B) and azole antifungals (eg, miconazole, ketoconazole, fluconazole, itraconazole) are the drugs most commonly used.[25,26] In the 1950s, nystatin was the first effective treatment for oral candidiasis. Nystatin is formulated for oral use as a suspension or pastille. The nystatin has a bitter taste, which may reduce patient compliance; therefore, the taste has to be disguised with sucrose and flavoring agents. If the candidiasis is due to xerostomia, however, the sucrose content of the nystatin preparation may contribute to xerostomia-related caries.[27–29] Amphotericin B has been restricted for many years to intravenous treatment of life-threatening systemic fungal infections. Subsequently, this medication has become available as an oral suspension for the management of oral candidiasis. Unfortunately, because the gastrointestinal tract poorly absorbs the polyene agents nystatin and amphotericin, multiple daily doses are necessary to adequately expose the yeasts to the drug.[25,30]

During the 1970s, the azole antifungal agents were developed. These medications represent a major step forward in the management of candidiasis.[31,32] A miconazole varnish and miconazole gel can be topically administered in denture-related stomatitis. To obtain a reduction of the yeasts and palatal erythema, a once-daily application of the miconazole varnish or a thrice-daily application of the miconazole gel for 15 days is sufficient.[33]

Because the biofilm formation in candidiasis is highly resistant to antifungal agents, systemic ketoconazole, fluconazole, or itraconazole can be used.[25,26] Ketoconazole can be absorbed across the gastrointestinal tract, thereby providing systemic therapy by an oral

route of administration. The single daily dose is much easier for the patient to use; however, several disadvantages have been noted. Patients must not take antacids or H_2-blocking agents because an acidic environment is required for proper absorption. Ketoconazole administration for more than 2 weeks could be responsible for idiosyncratic liver toxicity. For this reason, ketoconazole should not be used as an initial therapy for routine oral candidiasis. Furthermore, ketoconazole has been implicated in drug interactions with the macrolide antibiotics (eg, erythromycin) and the antihistamine astemizole, all of which may produce potentially life-threatening cardiac arrhythmias.[34]

Fluconazole is well-absorbed systemically, and an acidic environment is not required for absorption. A relatively long half-life allows for once-daily dosing, and liver toxicity is rare at the doses used to treat oral candidiasis. However, fluconazole may not be appropriate for long-term preventive therapy because resistance to the drug seems to develop in some instances. Drug interactions include a potentiation of the effects of phenytoin, warfarin compounds (anticoagulants), and oral hypoglycemic agents.

Itraconazole has proven efficacy against a variety of fungal diseases, including histoplasmosis, blastomycosis, and oropharyngeal candidiasis. This drug seems to have an efficacy equivalent to that of fluconazole, but significant drug interactions are possible. Itraconazole is contraindicated for patients taking astemizole, triazolam, midazolam, and cisapride.[34,35]

In this study, to avoid the use of systemic antifungal agents, and because antimycotic topical therapy with nystatin (Mycostatin) had failed, the 5% amorolfine varnish was used. This drug is useful in the treatment of onychomycosis.[36–40] In the literature, side-effects or complications caused by amorolfine varnish have been described. Administration of amorolfine varnish can be responsible for side-effects such as burning sensation, dryness of skin, and itching, and complications such as allergic contact dermatitis.[41,42]

An open, randomized study on 456 patients with onychomycosis treated once or twice weekly for up to 6 months with amorolfine 5% nail lacquer reported mild local irritation in only 4 of 456 patients.[39,40] Generally, no systemic side-effects occurred, and no patients suspended treatment because of an adverse event.[39,40] In a double-blind randomized study on 157 patients with onychomycosis treated once weekly for up to 6 months with amorolfine nail lacquer (2% or 5%), only 3 patients (2%) experienced mild local adverse events.[36] No systemic side-effects occurred, and no patients discontinued treatment because of an adverse event.[36]

Mild local adverse events are rare. Because amorolfine varnish is topically applied, there are no particular side-effects to its use.[36] However, the amorolfine varnish seems to be contraindicated for patients affected by hyperplastic candidiasis because, in the preparation of the varnish, a possibly carcinogenic methacrylate copolymer is present.

The current preliminary study showed the efficacy of a varnish of 5% amorolfine in the treatment of prosthetic stomatitis associated with *Candida* infection. However, further studies are required to confirm these encouraging results.

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