

## Original Article

### Clinical evaluation of the essential oil of “*Satureja Hortensis*” for the treatment of denture stomatitis

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#### ABSTRACT

**Background:** The prevalence of denture stomatitis has been shown to vary from 15 to 65% in complete denture wearers. *Satureja hortensis* L. has been considered to have antinociceptive, anti-inflammatory, antifungal and antimicrobial activities *in vitro* and exhibits strong inhibitory effect on the growth of periodontal bacteria. The aim of this study was to evaluate the efficacy of a 1% gel formulation of *S. hortensis* essential oil for the treatment of denture stomatitis.

**Materials and Methods:** A randomized, controlled clinical trial study was conducted on 80 patients (mean age 62.91±7.34) in two parallel groups treated either with *S. hortensis* essential oil 1% gel or placebo applied two times daily for two weeks. Denture stomatitis was diagnosed by clinical examination and paraclinical confirmation with sampling the palatal mucosa for *Candida albicans*. Data were analyzed using Chi-squared or Student's *t* tests.

**Results:** The erythematous lesions of palatal area were significantly reduced ( $P<0.0001$ ) in the treatment group who applied 1% topical gel of *S. hortensis* essential oil and *Candida* colonies count were reduced significantly ( $P=0.001$ ).

**Conclusion:** Topical application of the essential oil of *S. hortensis* could be considered as an effective agent for the treatment of denture stomatitis.

**Key Words:** *Candida albicans*, denture stomatitis, *Satureja hortensis*

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## INTRODUCTION

Denture stomatitis is an oral pathology in denture-bearing patients, especially under the maxillary prosthesis.<sup>[1,2]</sup> The prevalence of it has been shown to vary from 15 to 65%.<sup>[3]</sup> It usually manifests itself as an erythematous, edematous mucosa underlying and confined to the area covered by a complete upper denture.<sup>[4]</sup> *Candida* species are considered the primary causative agents of denture stomatitis.<sup>[2,4-6]</sup> The formation of *Candida* biofilms reduces the susceptibility of yeasts to antifungal agents.<sup>[1,7]</sup>

It seems that resistance of pathogenic fungi to antifungal agents has increased. Because of this side effects to common antifungal drugs,<sup>[1,4]</sup> novel fungal therapies with fewer side effects on humans are urgently required for effective management of candidiasis infections, so there are many studies on the use of herbal antifungal essential oils and extract.<sup>[1,4,8-13]</sup>

In several studies, about the antifungal effect of essential oils, specific *anti-Candida* activity of many aromatic plants has been described. The phenolic major components of these plants have been suggested to have a potent antifungal activity.<sup>[14-16]</sup>

Different species of the *Satureja* spp. are used in traditional medicine, food and pharmaceutical industries.<sup>[17]</sup> Some investigations suggest that the essential oil of the *S. hortensis* L. has antinociceptive,<sup>[18,19]</sup> anti-inflammatory,<sup>[18,19]</sup> antifungal<sup>[20,21]</sup> and antimicrobial activities<sup>[21-25]</sup> and a recent study has also demonstrated

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that growth of periodontal bacterial inhibition effect.<sup>[18,26]</sup> The main compounds in the essential oil of *Satureja* spp. are carvacrol,  $\gamma$ -terpinene and p-cymene.<sup>[21,24,25,27]</sup> To date, many diverse activities of carvacrol such as antimicrobial, antifungal, antitumor, analgesic, antispasmodic, anti-inflammatory are shown and discussed elsewhere.<sup>[15,18,25,28,29]</sup>

The aim of the present study was to evaluate the efficacy of *S. hortensis* essential oil gel for the treatment of denture stomatitis.

## MATERIALS AND METHODS

This double-blind clinical trial was conducted in dental clinics affiliated to the Isfahan University of Medical Sciences Isfahan, Iran, from July 2010 until May 2011. Eighty complete denture wearers with denture stomatitis were enrolled. Forty outpatients (19 males and 21 females) were included in the study as case group, and 40 (12 males and 28 females) were included in the control group.

All patients who were wearing complete dentures for more than one year and suffered from type II or III palatal lesions according to Newton's classification<sup>[30]</sup> were included in this study. Accordingly moderate palatal lesions (Type II) were defined as a generalized inflammation of the denture-bearing area, and the severe one (Type III) was defined as a hyperplastic palatal surface of the palatal area. However patients who had diabetes mellitus, immunity suppression or systemic diseases, used antibiotic, antifungal drugs or antiseptic mouth wash within one month before the study as well as during the treatment course, had not followed the medical procedure during treatment and suffered of changing within the structure in denture, were excluded from the study.

All patients were examined by a specialized dentist and all demographic data, full medical and dental history for each patient were recorded.

A gel formulation of the essential oil of *S. hortensis* was prepared at the school of Pharmacy, Isfahan University of Medical Sciences. We formulated a carboxy methyl cellulose, Carbomer 934 (Merck®, Germany) and Plastibase (Jaber ebne Hayyan®, Tehran, Iran) gel base as placebo and the same gel base containing 1% of essential oil of the *S. hortensis* L. (Barij® Essence Company, Kashan, Iran) for the use of patients in this study.

At the first visit, erythema of the palatal mucosa

was assessed visually, and the palatal mucosa were examined for the presence of yeasts by cultivating the samples on sabouraud dextrose agar plates with sterile swabs and were inoculated over the culture media, then incubated at 37°C and after 48 h the colonies were counted. The presence of one colony or more were considered as a positive result.<sup>[4]</sup>

Patients were randomly received either 1% gel of *S. hortensis* essential oil (cases) or placebo (controls) according to their profile numbers and the random figure tables. They were instructed to apply thin layer gel equals 1 cm on the base of the denture and the palatal mucosa twice daily (morning and night) for a period of two weeks. All patients were asked to keep the recommended hygienic instructions during treatment course. After 14 days, clinical and mycological evaluation was repeated to determine the efficacy of *S. hortensis* essential oil 1% gel.<sup>[1,4]</sup> If clinical alterations in the severity and extension of lesions were not observed, as well as any reduction in colony counts were seen, we reported it as no improvement. When the erythema surface of the palatal surface and colony counts were reduced, we considered it as partial improvement and in case of total elimination of erythematous lesions and the absence of any colonies, it was reported as complete improvement.

On days 14 and 21 after the first visit, all patients were assessed for treatment, local adverse reactions and asked as to whether they have experienced any adverse reactions.

The study protocol was approved by the board of ethics for human research at Isfahan University of Medical Sciences. All patients have completed informed consent form prior to the study, and no patient was deprived from the normal ordinary treatment if he/she refused for participation in the study.

All data were analyzed using SPSS® for windows (v.18). We have used  $\chi^2$  test for analyzing qualitative data and Student's *t* test for quantitative data, considering  $P < 0.05$  as significant.

## RESULTS

Eighty patients with the mean ( $\pm$ SD) age of 62.91 $\pm$ 7.34 completely fulfilled the study protocol and 49 out of them were females. Participants' demographic characteristics are summarized in Table 1. All patients of the two study groups were matched in terms of age, gender, colony count of *C. albicans* on palatal sample,

duration of denture wearing, removal of denture during at night and Newton's type of denture stomatitis [Table 1]. And 77.5% of control group and 67.5% of the case group did not remove their denture during the night. According to this data, no significant differences was seen between the severity of lesions in both groups ( $P=0.317$ ).

In this study, the most common form of observed denture stomatitis lesions in both groups (cases and controls) was in Newton's Type II classification.<sup>[30]</sup>

Figure 1 shows the change in the inflammatory state of the palatal mucosa before and after treatment with the topical drug and placebo in cases and control groups respectively.

Patients in the intervention arm of the study (cases) who applied 1% gel of *S. hortensis* for two consecutive weeks had a statistically significant improvement in colony count of *Candida* ( $P=0.001$ ) and the erythema of palatal mucosa ( $P=0.000$ ) [Figure 1].

## DISCUSSION

Denture stomatitis is an inflammatory process of the oral mucosa associated with the presence of *Candida* or other microorganisms and factors, such as denture wearing and long term antibiotic therapy. *Candida albicans* represents the most common causative agent of the oral candidiasis.<sup>[13,31]</sup>

*Candida albicans* is a part of the normal flora of mouth in human, and the density of growths of *Candida* may have a role in candidial infections. In this way, the rational use of antifungal agents in the treatment of these clinical conditions seems to be justifiable.<sup>[4,13]</sup>

In this study, all patients of the two study groups were matched in terms of age, gender, colony count of *C. albicans* on palatal sample, duration of denture wearing, removal of denture during at night and Newton's type of denture stomatitis [Table 1]. In agreement with many other studies, predisposing local factors include poor denture hygiene, continual and night time wearing of removable dentures.<sup>[32-34]</sup>

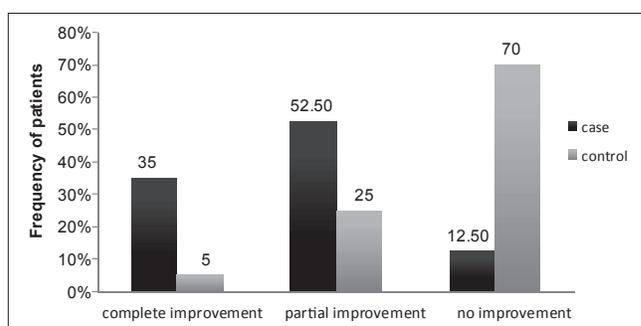
Patients in the intervention arm of the study (cases) who applied 1% gel of *S. hortensis* for two consecutive weeks had a statistically significant improvement in colony count of *Candida* ( $P=0.001$ ) [Table 1] and the erythema of palatal mucosa ( $P=0.000$ ) [Figure 1].

As mentioned in other study,<sup>[1]</sup> For oral topical drug

**Table 1: Baseline characteristics and the outcome of treatment with Savory essential oil in the study patients**

Variable	Cases (40)	Controls (40)	P value
Age (years)	61.9 (7.4)*	63.92 (7.216)	0.219
Gender (female)	52.5%**	70%	0.108
Denture usage (years)	11.32 (5.95)	10.65 (5.838)	0.610
Newton's type of denture stomatitis (II)	90%	87.5%	1.000
Removal of dentureduring at night	32.5%	22.5%	0.317
Colony counts (before study) CFU/each sample	23.55 (24.075)	30.40 (26.920)	0.543
Colony counts (after study) CFU/each sample	10.00 (16.148)	26.20 (27.886)	0.001
Outcome (recovery)	87.5%	30%	0.000

\*Independent sample t-test: Mean (SD) of these variations. \*\*Chi-square tests: (%) Percentage of these variations



**Figure 1: Inflammation changes of palatal mucosa in case and control groups**

administrations, it is necessary to note that the constant flow of saliva, and the mobility of the involved tissues may cause a rapid decline in therapeutic concentration of drug and consequently result in treatment failure. In this case, selection of buccal mucoadhesive formulations, such as these orabase formulations, can help to overcome these problems.

In our study, spicy odor of the essential oil was the only minor complaint of patients which is comparable to similar oral topical gels.<sup>[35]</sup> However the topical formulation which we used could be corrected and developed in further pharmaceutical studies. We have not observed any other reportable side effects in patients receiving the herbal gel.

Due to the increasing resistance of micro-organisms against antifungal agents and their side effects, formation of *Candida* biofilm and generally positive attitude of patients for natural treatment, novel herbal antifungal therapies with fewer side effects on humans are rational for effective management of *candidiasis* infections. There are studies on the use of herbal

antifungal and especially anti *candidial* characteristics of some essential oils.<sup>[8-13]</sup>

*Satureja* is one of the aromatic plants with a wide spectrum of pharmacologic properties.<sup>[18,20]</sup>

Another species of this plant, *S. khuzestanica*s has been shown to have analgesic, antiseptic, antioxidant, antimicrobial, antiviral, antifungal, anti-inflammatory, anti-nociceptive, antidiabetic and anti-hyperlipidemic effects.<sup>[19,36,37]</sup> *S. hortensis* is used as a drug in traditional medicine and as a food additive in food industries.<sup>[17]</sup> Evidences for antinociceptive, anti-inflammatory, antifungal and antimicrobial activities of the essential oil *S. hortensis* are reported previously elsewhere.<sup>[18,20,26,38]</sup> Essential oil of *S. hortensis* L. exhibits a strong inhibitory effect on the growth of periodontal bacteria.<sup>[18,26]</sup> Hence, researchers have studied the chemical composition of extracts and essential oils isolated from this aromatic plant.<sup>[21,25]</sup> The phenolic major components of them have been suggested to have a potent antifungal activity.<sup>[14-16]</sup> Carvacrol,  $\gamma$ -terpinene and p-cymene are major components of *Satureja* spp.<sup>[19,21]</sup> Many pharmacologic activities of the essential oil of *Satureja* such as antimicrobial, antifungal, antitumor, analgesic, antispasmodic, anti-inflammatory effects are attributed to the chemical structure of these phenolic compounds.<sup>[15,28,29]</sup>

In this study, reduction in the number of *Candida* and erythematous surface in both of two arms may be justifiable because of the effect of the standard hygienic recommendations (e.g., cleaning the dentures and denture brushing, removal of denture overnight) during the study which is consistent when compared with previous one.<sup>[13]</sup> On the other hand, because of coverage of the palatal mucosa with the denture, especially during the night, antimicrobial effect of saliva which has an important role in the maintenance of oral health is attenuated.<sup>[11]</sup>

In our study, reduction of the erythema surface was more significant in the treatment group which supports previous reports on above mentioned pharmacological effects.

In conclusion, this study showed that the gel formulation of the essential oil of *S. hortensis* could be an effective agent for the treatment of denture stomatitis and may be considered safe regarding its nutraceutical and folklore usage. However, further studies are needed to investigate other therapeutic aspects of this essential oil for maintaining oral health.

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