ORAL DISEASES

Oral Diseases (2010) 16, 172–175. doi:10.1111/j.1601-0825.2009.01618.x

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ORIGINAL ARTICLE

Colonization of *Candida*: prevalence among tongue-pierced and non-pierced immunocompetent adults

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OBJECTIVE: To evaluate the colonization of *Candida* at the tongue-piercing site of immunocompetent individuals.
SUBJECTS AND METHODS: Swabs samples were obtained from the anterior lingual mucosa of healthy young adults with tongue piercing (*N* = 115); 86 subjects with (non-intra-oral) facial piercing served as a comparison group. *Candida* colonization was examined by light microscopy after 5-day incubation. Positive specimens were re-cultured on Chromagar™ *Candida* plates for species identifying.
RESULTS: *Candida* colonization was more prevalent among tongue-pierced (20.0%) than facial-pierced subjects (9.4%; *P* = 0.048). All colonies were of *Candida albicans*. No difference was found between current tongue ornament wearers (21.2%) and non-wearers (19.5%; *P* = 0.803). In multivariate analysis, the only significantly positive influencing factors on colonization were tongue piercing (*P* = 0.034) and daily smoking of more than 10 cigarettes (*P* = 0.024).
CONCLUSIONS: Piercing of the tongue was found to be a risk factor for colonization of *Candida albicans*, without an influence of whether or not an ornament is in place.
Oral Diseases (2010) **16**, 172–175

Keywords: oral piercing; fungal infection; candidiasis; candidosis; oral hygiene; compliance

Introduction

Candida, a yeast-like fungus, is a common colonizer in the oral and gastrointestinal flora (Epstein, 2003). Up to 66% of the general population was reported as harboring *Candida* in oral flora (Guida, 1988; Brawner and Cutler, 1989; Epstein, 1990; Ben-Aryeh *et al*, 1995) and was termed '*Candida* carriers' (Scully, 2008). The dorsum of the tongue is the most densely colonized region in the oral cavity, followed by palate and buccal mucosa (Muzyka and Glick, 1995).

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Received 25 June 2009; revised 24 July 2009; accepted 28 July 2009

Candidiasis (also termed candidosis, moniliasis) is the condition when a lesion is caused by *Candida* species (Scully, 2008). Oral *Candida* is mostly involved in superficial fungal oral infections, with symptoms ranging from none to a painful burning sensation causing dysphagia (difficulty in swallowing) and interfering with nutritional intake (Muzyka and Glick, 1995). Oral *Candida* can be of major significance in immunosuppressed patients. In patients suffering from malignancies, *Candida* can complicate oral mucositis and is associated with high rate of morbidity and mortality (Epstein *et al*, 1993; Wingard and Leather, 2001). In patients receiving head and neck radiotherapy, preexisting *Candida* colonization in oral flora before radiation therapy was found to be a risk factor for clinical candidiasis (Epstein *et al*, 1993).

Oral *Candida* colonization and infection are influenced by systemic as well as local factors. Systemic factors that increase the presence of *Candida* in the oral flora include diabetes mellitus, immunosuppression (e.g. HIV), nutritional factors (e.g. deficiencies of iron, folic acid, and vitamin B12), broad-spectrum antibiotics, and oral administration of steroids (Epstein, 2003). Invasion of *Candida* from the oral flora and presentation of clinical infection are most commonly related to neutropenia, immunosuppression, and mucosal injury (Epstein *et al*, 1993).

Local risk factors for *Candida* in the oral flora include the using of dentures, smoking, hyposalivation, and local glucocorticoids application in the oral cavity (Cardash *et al*, 1989; Epstein, 2003). It seems that denture enhances the risk by the adherence of *Candida* to the acrylic base (Segal *et al*, 1992), the interference with salivary rinsing of the tissue underneath the denture, and the associated poor hygiene (Moskona and Kaplan, 1992; Epstein, 2003).

There are no reports of the risk for tongue *Candida* colonization associated with tongue piercing (Figure 1). The aim of this study was to examine this possible association. Our hypothesis was that there is an elevation in the prevalence of colonization of *Candida* as tongue-piercing sites can serve as relatively protected, warmed, and moisten colonization niche, especially when acrylic ornament and poor hygiene are involved.

Figure 1 Metal tongue ornament. Tongue piercing is an irreversible operation; the piercing hole would not reattach even after permanent removal of the ornament

Subjects and methods

Study population

The participants in the study were healthy young adults who arrived at an Israeli military central dental clinic for periodic dental examination. Inclusion criteria included the presence of oral and/or facial piercing (not including conventional earlobe piercing). Exclusive criteria included having complaint of related symptoms (e.g. swelling, pain, or discomfort in the tongue); using intra-oral acrylic removal prosthodontic or orthodontic appliance; having diabetes mellitus; using hyposalivation-induced medications, antibiotics, glucocorticoids (by oral administration, inhalation, or oral topical agents), or other immunosuppressive agents within 6 months prior to examination; history of head and neck irradiation and/or anti-cancer cytotoxic chemotherapy; or self-reporting of HIV infection in the written medical history questionnaire. The examinees who fulfilled these criteria were briefed on the purpose of sample collection and were asked to participate in the study. The examinees could refuse to participate without prejudice or loss of benefits to which they were otherwise entitled. Informed consents were obtained from examinees who agreed to participate in the study.

Subjects who had their tongue pierced (with or without current wearing of ornament) were included in the study group. Subjects with non-intra-oral facial piercing (including subjects with lip piercing) were included in the comparison group. Subjects who had tongue as well as other facial piercing were included in the (tongue piercing) study group.

Data included age, gender, smoking and alcohol consumption, oral hygiene practice, piercing sites, time from piercing procedure, and ornament use were collected from each participant.

Laboratory methods

Samples were collected from the tongue mucosa of each subject using a swab with Amies gel (Copan, CA, USA); in tongue-pierced subjects (study group), from the tongue-piercing site by dorsal approach; and in non-tongue-pierced subjects (comparison group), from the estimated anterior third of the dorsal tongue. No samples were obtained from the ventral aspect of the tongue.

Samples were cultured using DTM (Dermatophytes Test medium)/SDA (Sabouraud glucose agar) divided dishes (HyLabs, Rehovot, Israel) and incubated at 28°C for 5 days.

Cultures were examined macroscopically and microscopically by two experienced microbiologists, who were not aware to the subjects' grouping at the time of examination. Disagreements were settled by discussion and convincing between the two microbiologists. Positive *Candida* species were re-cultured on Chromagar™ *Candida* plates (HyLabs, Rehovot, Israel) to identify the *Candida* species.

Statistical analysis

Prevalence of tongue *Candida* colonization in study group *vs* comparison group was tested using the chi-square test. Logistic Regression model for *Candida* colonization, with gender, smoking, and oral self-care practice as explanatory variables, was performed as well.

For assaying the hypothesis that the tongue ornament could have a cleansing effect, thus decreasing superficial colonization of *Candida*, the prevalence of positive colonization was compared between current lingual ornament wearers and non-wearers (all of the tongue-pierced study group).

Data were recorded into Microsoft® Access® database and analyzed by SPSS® 15.0 (SPSS, Inc., Chicago, IL, USA) statistical software. Two-sided $P < 0.05$ was considered statistically significant.

The study design was approved by the Committee for Ethics Research on Human Subjects of the Israeli Defense Force Medical Corps.

Results

A total of 201 examinees fulfilled the inclusion criteria and agreed to participate in the study; 115 subjects were tongue pierced and included in the study group, and 86 were (non-intra-oral) facial pierced and included in the comparison group (Table 1). Mean age (±s.d.) of the study and comparison groups was 20.4 (±1.6) and 20.7 (±1.7) years (range 18–25 years), respectively, and mean time (±s.d.) from piercing was 26.0 (±19.8) and 21.0 (±16.2) months, respectively. Thirty-three of the tongue-pierced subjects (28.7%) were with tongue ornament regularly at the time of examination and 82 (71.3%) were without tongue ornament. Among the 33 tongue ornament wearers, 19 ornaments were of metal and 14 of acrylic jewelry.

Positive tongue *Candida* colonizations were obtained in 23 (20.0%) of the tongue-pierced subjects and in 8 (9.4%) of the comparison group ($P = 0.048$). All colonizations were of *Candida albicans*. Within the tongue-pierced group, positive grows were found in 7 (21.2%) and 16 (19.5%) of the 33 current ornament wearers and 82 non-wearers, respectively (OR = 1.111, 95% CI = 0.410–3.011, p = 0.803). Of the 19 metal ornament wearers, 5 (26.3%) were with tongue colonizations, and among the 14 acrylic ornament wearers, 2 (14.3%) were colonized (no statistical analysis was made owing to relatively small numbers).

The univariate and logistic regression analyses of possible risk factors for tongue colonization are presented in Table 2. Tongue piercing and smoking of more than 10 cigarettes per day were the only statistically significant influencing factors in the multivariate analysis ($P = 0.034$, $P = 0.024$, respectively). Male gender, daily alcohol consumption of more than 15 g, or regular daily tongue brushing have a non-significant protecting effect against *Candida* colonization in the tongue. Statistical analysis regarding mouthwash practice was not carried out because of an inconsistent practice of mouth washing (frequency, amount, duration, and time interval until next drinking or eating) and the different brand names of the solutions with varying active components' concentrations.

Table 1 Prevalence *Candida albicans* colonization by groups

Group	*N*	*Colonizations (%)*
Study group –Tongue piercing	115	23 (20.0)
Current non-wearers of tongue ornament	82	16 (19.5)
Current tongue ornament wearers	33	7 (21.2)
Comparison group –Facial (non-intra-oral) piercing	86	8 (9.3)

Discussion

Body art (piercing, tattooing, tongue splitting, and tooth decoration) is a practice that is gaining acceptance as a sign of individuality, marginality, decoration, or group membership (Stirn, 2003; Rabinerson and Horowitz, 2005). Prevalence of oral and peri-oral piercing among studied young adult population ranges between 3.4% and 20.3% (Levin and Zadik, 2007). However, despite the banal appearance of tongue piercing, it is not without complications. These complications can be divided into immediate postoperative complications (e.g. bleeding, infection, and swelling) (Ram and Peretz, 2000; Shacham *et al*, 2003) and long-term complications (e.g. irreversible gingival recession and dental fracture) (Levin *et al*, 2005; Zadik and Sandler, 2007).

Regarding infection-related complications, there are reports of tongue-piercing-associated infective endocarditis (Armstrong *et al*, 2008) and Ludwig's angina (Perkins *et al*, 1997), which are life-threatening. However, no association between tongue piercing and fungal infection was previously reported.

In the current clinical study, the prevalence of tongue *Candida* colonization was assessed among young adults, who were not having local or systemic risk factors, such as oral acrylic appliance, diabetes mellitus, relevant medications, or irradiation, and reported themselves as non-HIV-carriers; *Candida* colonization was found to be significantly higher among tongue-pierced subjects than among subjects without tongue piercing. No difference was found for *Candida* colonization between current tongue ornament wearers and non-wearers; thus it seems that tongue ornament does not have a local cleansing effect.

As acrylic ornaments are flexible and thus less harmful to teeth and gingival tissues than the rigid metal ornaments, it was suggested that the dental practitioner has to recommend the former when the patient insists to have a tongue ornament (Zadik *et al*,

Table 2 Univariate and logistic regression analysis of possible risk factors for *Candida* colonization in the study population

Factor	*N*	*Colonization, N (%)*	*Univariate analysis*			*Logistic regression analysis*		
			OR	*95% CI*	P *value*	*OR*	*95% CI*	P *value*
Tongue piercing								
No	86	8 (9.3)	1.000			1.000		
Yes	115	23 (20.0)	2.438	1.032–5.756	0.048*	2.778	1.079–7.149	0.034*
Gender								
Male	66	9 (13.6)	1.000			1.000		
Female	135	22 (16.3)	1.233	0.533–2.851	0.683	1.677	0.674–4.174	0.266
Daily cigarettes smoking								
0	81	10 (12.3)	1.000			1.000		
1–10	82	11 (13.4)	1.100	0.440–2.753	1.000	1.333	0.519–3.423	0.550
>10	38	10 (26.3)	2.536	0.952–6.754	0.069	3.332	1.172–9.472	0.024*
Daily alcohol consumption								
0	79	14 (17.7)	1.000			1.000		
1–15 g	61	10 (16.4)	0.910	0.374–2.218	1.000	0.935	0.372–2.354	0.887
>15 g	61	7 (11.5)	0.602	0.227–1.598	0.348	0.406	0.137–1.205	0.104
Regular tongue brushing								
Yes	90	11 (12.2)	1.000			1.000		
No	111	20 (18.0)	1.578	0.713–3.496	0.327	1.500	0.663–3.394	0.330

OR, odds ratio; CI, confidence interval.
*Statistically significant.

2007). However, oral acrylic appliances were reported to have affinity to fungal colonization (Segal *et al*, 1992). Further studies are needed to investigate whether tongue acrylic ornament also has such affinity.

Using Chromagar media for determining the species, all of the colonizations in this study were identified as colonies of *Candida albicans*, which is also in agreement with the previous studies. For example, Ben-Aryeh *et al* (1995) studied oral candidal growth among healthy volunteers and reported that 76% of the colonizations were of *C. albicans*. However, the using Chromagar media for this purpose has a limitation as although the vast majority of green colonies are of *Candida albicans*, they could also be of *Candida dubliniensis* in the lesser extent and mostly in HIV carriers, or of other species.

The significant positive effect of smoking on colonization is in agreement with previous reports (Epstein *et al*, 1993). Male gender, daily alcohol consumers of more than 15 g, or regular daily tongue brushers had non-significantly lower prevalence of positive colonizations. In the general population, women have higher prevalence of *Candida* carriers than men (Scully, 2008), despite reported better oral hygiene measures held by women compared with men (Zadik *et al*, 2008). The other results regarding alcohol consumption and tongue brushing can be partly explained by the antiseptic effect of alcohol, and the mechanical cleansing effect of tongue brushing on tongue colonization. More research on the possible effect of mouthwash in the tongue-piercing population is suggested.

The relatively low percentage of colonizations (9.4% to 20.0%) found in this study, compared with the previous reports among general population (up to 66%), can be explained by the healthy (non-medicated) young adult population of this study (after exclusion of several high risk groups), whereas the general population includes the high risk groups of infants (Makhoul *et al*, 2002) and old age persons (Moskona and Kaplan, 1992) as well as immunodeficient patients or patients with diabetes mellitus, antibiotic-, steroid-, immunosuppressive-, or hyposalivation-induced-medicated persons. Moreover, in this study, we sampled only the anterior tongue region and not the whole oral cavity.

The results obtained in this study suggest that the piercing of the tongue is a risk factor for colonization of *Candida albicans*, without the influence of whether or not an ornament is in place. As body piercing is becoming popular mostly among young adults only during recent years, the possible clinical significance of these findings has to be further investigated in the next decades as these current young adults will get older and become medically compromised with additional local and systemic risk factors for Candidal infection.

Author contribution

Y. Zadik designed the study and wrote the article. S. Burnstein managed the laboratory work and performed it. E. Derazne analyzed the data. C. Ianculovici and V. Sandler recruited the subjects, and collected the data and samples. T. Halperin performed the laboratory work (with S. Burnstein), and co-wrote the article.

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