

# Frequency of Oral Candida Colonization in Patients with Ankylosing Spondylitis

## Ankilozan Spondilit Hastalarında Oral Kandida Kolonizasyonu Sıklığı

Nilgül Üstün<sup>1</sup>, Melek İnci<sup>2</sup>, Hayal Güler<sup>1</sup>, Burçin Özer<sup>2</sup>, Çetin Kılınç<sup>2</sup>, Ayşe Dicle Turhanoglu<sup>1</sup>

<sup>1</sup> Mustafa Kemal University, Faculty of Medicine, Department of Physical Medicine and Rehabilitation, Hatay, Turkey

<sup>2</sup> Mustafa Kemal University, Faculty of Medicine, Department of Medical Microbiology, Hatay, Turkey

### ABSTRACT

Ankylosing spondylitis (AS) is a chronic inflammatory disease affecting the axial skeleton and peripheral joints. Anti-tumor necrosis factor (anti-TNF) agents improve signs and symptoms, spinal mobility, and physical function in these patients. A wide range of fungi infections associated with the use of anti-TNF agents have been described. The aims of this study were to compare the frequency of oral candida colonization in patients with AS as opposed to those of healthy subjects and, to compare the oral frequency of candida colonization from AS patients receiving conventional and anti-TNF agent therapy. Subject population consisted of 52 patients with AS and 51 age-sex-matched healthy individuals. Mycological examinations included frequency of Candida colonization in oral rinse samples. Number of the patients on anti-TNF agent and disease modifying anti-rheumatic drug therapy were 7 and 45, respectively. Candida colonization were in 18(34,6%) of the patients and 13(25,5%) of the healthy controls ( $p=0,313$ ). Candida albicans was the commonest species isolated from both patients and controls (94,4% vs. 69,2%,  $p=0,06$ ). Candida colonization were in 3(43%) and 15(33%) of the patients with and without using an anti-TNF agent, respectively ( $p=0,622$ ). In conclusion, frequency of oral candida colonization in patients with ankylosing spondylitis is not higher than that in healthy controls, and anti-TNF agent therapy is not correlated with increased frequency of oral candida colonization.

**Keywords:** Ankylosing spondylitis, anti-tumor necrosis factor agents, oral candida colonization

### ÖZET

Ankilozan spondilit aksiyel iskeleti ve periferik eklemleri tutan kronik inflamatuvar bir hastalıktır. Anti-tümör nekroz faktör ilaçlar bulgu ve semptomları, spinal mobilitayı ve fiziksel fonksiyonları düzeltir. Anti-tümör nekroz faktör ilaç kullanımı ile çeşitli mantar enfeksiyonları rapor edilmiştir. Bu çalışmadaki amaç ankilozan spondilit hastaları ve sağlıklı bireylerdeki ve anti-tümör nekroz faktör ilaç kullanan ve kullanmayan hastalardaki oral kandida kolonizasyonu sıklığını karşılaştırmaktır. Çalışmaya 52 ankilozan spondilit hastası ve 51 yaş/cinsiyet uyumlu sağlıklı birey alındı. Hastaların ve kontrollerin oral çalkalama sularında oral kandida kolonizasyonuna bakıldı. Ankilozan spondilit hastaları ve sağlıklı bireylerdeki ve anti-tümör nekroz faktör ilaç kullanan ve kullanmayan ankilozan spondilit hastalarındaki oral kandida kolonizasyon sıklığı arasında anlamlı fark yoktu. Sonuç olarak ankilozan spondilit hastalarındaki oral kandida kolonizasyonu sağlıklı bireylerden farklı değildir ve anti-tümör nekroz faktör ilaç kullanımı artmış oral kandida sıklığı ile ilişkili değildir.

**Anahtar sözcükler:** Ankilozan spondilit, anti-tümör nekroz faktör ilaçlar, oral kandida kolonizasyonu

Corresponding Author  
Yazışma Adresi

Nilgül Üstün

Mustafa Kemal Üniversitesi,  
Tıp Fakültesi, Fiziksel Tıp ve  
Rehabilitasyon AD, Hatay, Turkey

Phone: +90 326 227 63 67

E-mail: drnustun@yahoo.com.tr

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## Introduction

Ankylosing spondylitis (AS) is a chronic inflammatory disease affecting the axial skeleton and peripheral joints. Disease progression may result in loss of mobility and function. Short- and long-term controlled studies have shown that anti-tumor necrosis factor (anti-TNF) agents improved signs and symptoms, spinal mobility, and physical function in patients with AS (1-2). Despite the clinical benefits of anti-TNF agents, some concerns exist regarding the occurrence of infections in patients treated with these agents, especially in those with other comorbidities such as diabetes, heart disease, and in those receiving concurrent immunosuppressive medications (3). The most common infectious complication associated with the use of anti-TNF agents is tuberculosis (4). A wide range of fungi infections associated with the use of anti-TNF agents have also been described (5-9).

Most reports indicate that *Candida albicans* is the predominant yeast isolated in patients as well as in healthy subjects (10). The most common non-*albicans* *Candida* species are *Candida glabrata*, *Candida parapsilosis*, *Candida tropicalis* and *Candida krusei* which have been frequently isolated from oral candidiasis (11-12). In immunocompromised patients *Candida* species can cause a multitude of disease manifestations ranging from mild oral disease to disseminated candidiasis. *Candida albicans* is the predominant species associated with mucosal fungal infections from yeast (13).

The aims of this study are to compare the frequency of oral candida colonization in patients with AS and healthy subjects and, in patients with and without using anti-TNF agent.

## Materials and Methods

Subject population consisted of 52 patients with AS and 51 age-sex-matched healthy individuals. All AS patients fulfilled the modified New York criteria (14). Exclusion criteria were diabetes mellitus and presence of prosthetic appliances for AS patients, and presence of prosthetic appliances for healthy controls. The study protocol was approved by the local ethics committee.

Demographic and disease-specific data of the patients were recorded. Disease activity was evaluated by Bath Ankylosing Spondylitis Disease Activity Index (BASDAI: on a scale of 0-10) (15), erythrocyte sedimentation rate (ESR) and C-reactive protein (CRP).

Mycological examinations included frequency of *Candida* colonization in oral rinse samples. Participants were instructed to rinse their mouth with 10 mL of phosphate buffered saline (PBS) solution for 1 minute.

Samples from oral cavity were transferred into a vessel and were centrifuged at 1700 g for 10 min. Upper portion of the centrifuged solution was poured and then sediment at the bottom was resuspended in 2 mL PBS and mixed 20 seconds for homogenization by a Vortex mixer. Mouth rinsing solutions were cultured on Sabouraud dextrose agar (Merck, Darmstadt, Germany) and incubated at 37°C for 48 h. *Candida* species were identified by germ tube formation, colonial morphological features and carbohydrate absorption using the API 20C-AUX kit (bioMe´rieux, Marcy l’Etoile, France).

## Statistical Analysis

Statistical analysis were carried out using a computer program (SPSS version 13.0). Descriptive statistics, Chi-square test were used for statistical analysis. Differences were considered significant at  $p < 0,05$ .

## Results

Fifty-two AS patients (mean age  $41,21 \pm 11,20$  years; 81% male, 19% female) and 51 healthy controls (mean age  $41,10 \pm 16,85$  years; 78% male, 22% female) were included the study. There was no statistical significant difference between the groups in terms of age and gender ( $p > 0,05$ ). Demographic and disease-specific characteristics of patients are seen in table 1. Number of the patients on anti-TNF agent and disease modifying anti-rheumatic drug (DMARD) therapy were 7 and 45, respectively. Only 32,7% of the patients had active disease ( $BASDAI \geq 4$ ).

*Candida* colonization were in 18(34,6%) of the patients and 13(25,5%) of the healthy controls ( $p = 0,313$ ) (Table 2). *Candida albicans* was the commonest species isolated from both patients and controls (94,4% vs. 69,2%,  $p = 0,06$ ). Non-*albicans* *Candida* species were *C. glabrata* in the patients, and *C. glabrata*, *C. tropicalis* and *C. kefyr* in the controls. *Candida* colonization were in 3(43%) and 15(33%) of the patients with and without using an anti-TNF agent, respectively ( $p = 0,622$ ) (Table 2).

**Table 1. Demographic and disease-specific characteristics of the patients, mean  $\pm$  SD (min-max).**

Age, years	41,21 $\pm$ 11,20 (20-70)
Disease duration, years	10,54 $\pm$ 9,35 (1-50)
Male, n(%)	42(81%)
Anti-TNF agent users, n(%)	7(14)
BASDAI (range 0-10)	3,30 $\pm$ 1,74 (0-9,2)
ESR (normal $\leq$ 20 mm/h)	19,72 $\pm$ 20,03 (2-109)
CRP (normal $\leq$ 0,8 mg/L)	1,10 $\pm$ 1,25 (0,15-8,22)

**BASDAI:** Bath Ankylosing Spondylitis Disease Activity Index, **ESR:** Erythrocyte sedimentation rate, **CRP:** C-reactive protein.

Table 2. Findings associated Candida colonized participants.

	Variables	Candida Colonized participants, n(%)	p-value
Groups	Patients (n=52)	18 (34,6%)	0,313
	Controls (n=51)	13 (25,5%)	
Patients	Anti-TNF $\alpha$ therapy (n=7)	3 (42,8%)	0,622
	DMARD (n=45)	15(33,3%)	

## Discussion

We found that there was no significant difference in frequency of oral candida colonization between patients and healthy controls and, the patients with and without anti-TNF agent.

*Candida albicans* is the predominant yeast isolated in patients as well as in healthy subjects (16). In present study, *Candida albicans* was also found the predominant yeast in the patients and healthy controls.

Tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ) is crucial for the production of interferon  $\gamma$  which plays a pivotal role in the host defense against disseminated fungal infections (17). Platelet-activating factor has a protective role in systemic murine candida infection and that the effect of platelet-activating factor appears to be mediated by TNF- $\alpha$ . Platelet-activating factor is released immediately in response to an inflammatory stimulus and induces TNF- $\alpha$  expression through the activation of the inducible transcription nuclear factor-kB. Nuclear factor-kB plays a central role in the induction of genes encoding TNF- $\alpha$  which can confer protective activity against systemic *Candida albicans* infection (18-20). Fungi infections have been described with the use of an anti-TNF agent, especially in those with other comorbidities such as diabetes, heart disease, and in those receiving concurrent immunosuppressive medications (3). In our study, there was no significant difference in patients with and without using an anti-TNF agent in terms of prevalence of oral candida colonization. No patient in this study had comorbidities and using of concurrent immunosuppressive medications.

The main limitation in this study was the small number of patients on anti-TNF $\alpha$  therapy. *Candida* infections have been described in the patients using anti-TNF agents. In present study, frequency of oral *Candida* colonization was slightly higher although very small number of patients using anti-TNF agents.

In conclusion, in the patients with ankylosing spondylitis, frequency of oral candida colonization is not higher than healthy controls, and anti-TNF agent therapy is not correlated with increased frequency of oral candida colonization.

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