Archives of Oral Biology 70 (2016) 55–61

Contents lists available at ScienceDirect

Archives of Oral Biology

journal homepage: www.elsevier.com/locate/aob

Impact of a chronic smoking habit on the osteo-immunoinflammatory mediators in the peri-implant fluid of clinically healthy dental implants

Brenno Marcondes Negri, Suzana Peres Pimentel*, Marcio Zaffalon Casati, Fabiano Ribeiro Cirano, Renato Correa Casarin, Fernanda Vieira Ribeiro

Dental Research Division, School of Dentistry, Paulista University, Av. Dr. Bacelar, 1212, 4° andar, Vila Clementino, São Paulo, SP 04026-002, Brazil

ARTICLE INFO

Article history:
Received 16 January 2015
Received in revised form 8 December 2015
Accepted 20 May 2016

Keywords:
Biological markers
Dental implants
Cytokines
Smoking

ABSTRACT

Objective: The aim of this study was to evaluate the influence of chronic cigarette smoking on the profile of osteo-immunoinflammatory markers in the peri-implant crevicular fluid (PICF) from clinically healthy implants

Designs: Twenty-five smokers and 23 non-smoker subjects with a unitary screwed implant-supported crown in the molar or pre-molar region were enrolled in this study. The implants should have been in functioning for at least 12 months, and the peri-implant tissue should be clinically healthy [probing depth (PD) < 4 mm with no bleeding on probing (BoP) and no evidence of radiographic bone loss beyond bone remodeling]. The levels of interferon (INF)-γ, interleukin (IL)-4, IL-17, IL-1β, IL-10, IL-6, IL-8, tumor necrosis factor (TNF)-α, matrix metalloproteinase (MMP)-2, MMP-9, osteoprotegerin (OPG), soluble receptor activator of nuclear factor-κβ ligand (RANKL), osteocalcin (OC), osteopontin (OPN), transforming growth factor (TGF)-β, and cross-linked telopeptide of type I collagen (ICTP) in the PICF were quantified by a multiplexed bead immunoassay.

Results: The smokers presented reduced levels of IL-4, IL-8, and TNF-α compared with the non-smoker individuals ($p < 0.05$). In addition, although lower OPG levels were detected in the PICF of the smokers, the RANKL/OPG ratio did not show a significant difference ($p > 0.05$). Moreover, higher ICTP concentrations and a higher TH1/TH2 ratio were observed in the PICF of the smoker patients ($p < 0.05$). No differences between the groups were observed for the other markers evaluated ($p > 0.05$).

Conclusions: Smoking habit modulate peri-implant cytokine profile, leading to reductions in IL-4, -8 TNF-α, and OPG levels and an increased ICTP and TH1/TH2 ratio in peri-implant crevicular fluid.

1. Introduction

Implant therapy has been described as a successful and predictable alternative to oral rehabilitation (Levin, Ofec, Grossmann, & Anner, 2011). However, some factors could negatively interfere with implant survival and the incidence of complications, such as peri-implant diseases. Although the prevalence of peri-implant lesions has been inconsistently described due to a lack of consensus in defining peri-implant disease from a clinical perspective, peri-implant mucositis and peri-implantitis have been reported as common clinical conditions with relatively high occurrences (Adell, Lekholm, Rockler, & Branemark, 1981; Atieh, Alsabeeha, Faggion, & Duncan, 2013; Fransson et al., 2005; Heitz-Mayfield, 2008; Marrone, Lasserre, Bercy, & Brecx, 2013; Zitzmann & Berglundh, 2008).

Smoking is associated with a higher prevalence and severity of peri-implant lesions and is considered a risk factor for bone loss around the implants (Atieh et al., 2013; Esposito, Hirsch, Lekholm, & Thomsen, 1998; Heitz-Mayfield, 2008; Mombelli, Müller, & Cionca, 2012; Roos-Jansåker, Lindahl, Renvert, & Renvert, 2006a; Roos-Jansåker, Lindahl, Renvert, & Renvert, 2006b; Roos-Jansåker, Renvert, Lindahl, & Renvert, 2006c; Saaby, Karring, Schou, & Isidor, 2014). It is well known that tobacco use induces alterations in the microbiota community in periodontal disease sites (Haffajee & Socransky, 2001; van Winkelhoff, Bosch-Tijhof, Winkel, & van der Reijden, 2001; Zambon, 1996). Recently, it was reported that smokers present a highly pathogen-rich anaerobic microbiome that is more closely aligned with a disease-associated community, even in periodontally healthy individuals, which creates an at-risk-for-harm environment that is at risk for future tissue breakdown (Mason et al., 2015).

* Corresponding author at: Depto de Odontologia, Universidade Paulista, UNIP, Av. Dr. Bacelar, 1212, 4° andar, Vila Clementino, São Paulo, SP 04026-002, Brazil.
E-mail address: suppimentel@yahoo.com (S.P. Pimentel).

http://dx.doi.org/10.1016/j.archoralbio.2016.05.014
0003-9969/

However, the presence of periodontopathogens *per se* is not sufficient for disease initiation, and studies clearly demonstrate that osteo-immunoinflammatory mediators induced by the host response play a critical role in peri-implant tissue breakdown (Arikan, Buduneli, & Lappin, 2011; Duarte et al., 2009; Javed, Al-Hezaimi, Salameh, Almas, & Romanos, 2011; Monov et al., 2006; Rakic et al., 2013; Strbac et al., 2006). It is well established that smoking alters the host response, including vascular function, neutrophil/monocyte activities, adhesion molecule expression, and antibody production, as well as immunoinflammatory mediator release (increasing pro-inflammatory cytokine release – i.e., IL-1β, IL-6, TNF-α – and reducing anti-inflammatory ones—IL-10, for example), contributing to periodontal lesions (Barbour et al., 1997; Kinane & Chestnutt, 2000; Palmer, Wilson, Hasan, & Scott, 2005; Ryder et al., 1998). Some evidence also suggests a modified pattern of important modulators of inflammation and of bone tissue metabolism in smoker individuals when peri-implant diseases are present (Ataoglu et al., 2002; Feloutzis et al., 2003; Konermann et al., 2014). However, no study is available demonstrating which disturbances in the profile of osteo-immunoinflammatory mediators are promoted by a smoking habit prior to disease initiation. This knowledge could explain the increased disease susceptibility, especially associated with dental implants, and, in the future, allow more predictable therapies and preventive approaches (Heitz-Mayfield & Mombelli, 2014).

Therefore, the aim of the present study was to evaluate whether smoking status could interfere with the levels of pro-inflammatory cytokines [interferon (INF)-γ, tumor necrosis factor (TNF)-α, interleukin (IL)-17, IL-1β, IL-6, IL-8], proteases [matrix metalloproteinase (MMP)-2, MMP-9], anti-inflammatory mediators [IL-4, IL-10, transforming growth factor (TGF)-β], and osteoblasto/clastogenesis-related factors [osteoprotegerin (OPG), receptor activator of NF-$_K$B ligand (RANKL), osteocalcin (OC), osteopontin (OPN), cross-linked telopeptide of type I collagen (ICTP)] in the peri-implant crevicular fluid (PICF) under clinically healthy conditions. These molecules are key markers of host immune responses and have been hypothesized to be critical mediators of the initiation and progression of peri-implant diseases. The hypothesis was that smoking modulates the local pattern of these osteo-immunoinflammatory mediators, even in the clinically healthy peri-implant condition.

2. Material and methods

2.1. Study design

This investigation was designed as a cross-sectional clinical study to evaluate the effects of chronic cigarette smoking in the profile of osteo-immunoinflammatory markers in the peri-implant fluid on the status of clinical health. This study was approved by the ethics committee of Paulista University (São Paulo-Brazil (Protocol 97.117)).

2.2. Population screening

Patient recruitment started in April 2013 and was completed by the end of February 2014. The clinical procedures and evaluations were performed between May 2013 and March 2014. All of the patients in the study were recruited from the patients referred to Paulista University by their regular dental visit program.

2.3. Inclusion and exclusion criteria

To be included in the study, all of the subjects had to be >30 years old and present at least a two-stage unitary screwed implant-supported crown in the molar or pre-molar region. The implant connection had to be external hexagonal, and the implants had to be functioning for at least 12 months, with a width of keratinized tissue $\geq$ 2 mm around the implants. The peri-implant tissue should be healthy [probing depth (PD) $\leq$ 4 mm with no bleeding on probing (BoP) and no evidence of radiographic bone loss beyond bone remodeling (American Academy of Periodontology, 2013). Individuals should be periodontally healthy (absence of periodontal pockets $\geq$ 4 mm and bleeding) and present full mouth plaque scores (FMPS (Ainamo & Bay, 1975)) and bleeding scores (FMBS (Mühlemann & Son, 1971)) $\leq$ 20%. Patients were classified as smokers if they smoked more than 10 cigarettes/day regularly, for at least two years. Non-smokers were subjects who had never smoked.

The exclusion criteria were pregnancy, lactation, systemic conditions reported during anamneses that could affect the progression of peri-implant diseases and bone metabolism (e.g., immunologic disorders), the long-term use of anti-inflammatory and immunosuppressive medications, antibiotic therapies in the previous six months, patients who required bone grafts before or alongside the implant surgery, and a history of previous regenerative procedures in the area treated with implant therapy.

All of the eligible patients were thoroughly informed of the nature, potential risks, and benefits of their participation in the study, and they each signed an informed consent document.

2.4. Experimental groups

The patients were allocated into one of the following groups: Smoker (n = 25) and Non-smoker (n = 23). Approximately 850 subjects referred to the Dental clinic for treatment were examined to obtain this final population. The number of patients included in the present study was based on previous investigations that found differences in the peri-implant and gingival crevicular fluid levels of various bone-related and immune-inflammatory markers in different clinical statuses (Barewal, Stanford, & Weesner, 2012; Gokhale et al., 2014; Prati et al., 2013; Ribeiro et al., 2011).

All patients were enrolled in a periodontal maintenance regimen, and before clinical and peri-implant fluid assessment patients were submitted to supragingival scaling and root planning, when necessary.

2.5. Clinical examination

The same examiner (BMN), who was blinded to the groups, performed all of the clinical evaluations. To perform the intra-examiner calibration, 15 non-study subjects presenting dental implants were selected. The examiner measured the PD of all of the patients twice within 24 h. The examiner was judged to be reproducible after fulfilling the pre-determined success criteria (the percentage of agreement within ±1 mm between repeated measurements had to be at least 90%). The intra-class correlation was calculated as a 92% reproducibility.

The following parameters were assessed at four sites of the experimental dental implants using a periodontal probe (North Carolina—Hu-Friedy, Chicago, IL, USA): 1) plaque index (PI) was scored using a dichotomous plaque index along the mucosal margin around the implants, 2) gingival index (GI) scored using a dichotomous index of mucosal marginal bleeding around the implants, 3) bleeding on probing (BoP) measured the presence or absence of bleeding up to 15 s after gentle peri-implant probing, and 4) peri-implant probing depth (PD/mm)—distance between the mucosal margin and the bottom of the sulcus. The full mouth plaque scores (FMPS) (Ainamo & Bay, 1975) and bleeding scores (FMBS) (Mühlemann & Son, 1971), as well as the probing depth—distance between the bottom of the pocket and the gingival margin and the clinical attachment level distance between bottom of the