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Peri-implant clinical profile and subgingival yeasts carriage among cigarette-smokers with peri-implant mucositis

Asmaa Saleh Almeslet[1] and Suha Mohammed Aljudaibi[2*]

Abstract

Background The present observational clinical investigation is based on the hypothesis that subgingival yeast carriage (SYC) is higher in cigarette-smokers with peri-implant mucositis (PM) than non-smokers with and without PM.

Objective The aim was to assess peri-implant clinical profile and SYC among cigarette-smokers with PM.

Methodology Participants were divided into four groups: Group-1—Cigarette-smokers with PM; Group-2—Cigarette-smokers without PM; Group-3—Non-smokers with PM; and Group-4—Non-smokers without PM. Information on duration and daily frequency of cigarette smoking (pack years), age, gender, familial history of smoking and most recent visit to a dentist and/or dental hygienist was collected. The following information was retrieved from healthcare records: implant dimensions, implant insertion torque, depth of insertion (credidastal or subcrestal), implant abutment connection, jaw location, implant surface characteristic, and mode of implant prosthesis retention. Peri-implant modified plaque and gingival indices (mPI and mGI), probing depth (PD) and crestal bone loss were recorded. Subgingival biofilm samples were collected, and SYC was recorded in colony forming units per milliliter (CFU/ml). $P < 0.05$ were considered statistically significant.

Results Eighty male individuals (20, 19, 21 and 20 individuals were included in groups 1, 2, 3 and 4, respectively) were included. The mPI was higher in Group-1 than groups 2 ($P < 0.05$) and 4 ($P < 0.05$). The mPI was higher in Group-3 than groups 2 ($P < 0.05$) and 4 ($P < 0.05$). The mGI was higher in Group-3 than groups 1 ($P < 0.05$), 2 ($P < 0.05$) and 4 ($P < 0.05$). The PD was higher in Group-1 than groups 2 ($P < 0.05$) and 4 ($P < 0.05$). The PD was higher in Group-3 than Groups 2 ($P < 0.05$) and 4 ($P < 0.05$). The CFU/ml were higher in Group-1 than groups 2 ($P < 0.05$) and 4 ($P < 0.05$). The CFU/ml were higher in Group-3 than groups 2 ($P < 0.05$) and 4 ($P < 0.05$).

Conclusion Peri-implant soft-tissue inflammatory parameters are worse and SYC is higher in moderate smokers than light smokers with PM and non-smokers without PM.

Keywords Dental implant, Subgingival, Yeast, Peri-implant mucositis, Smoking

*Correspondence:
Suha Mohammed Aljudaibi
smaljudaibi@pnu.edu.sa
[1]Department of Oral Maxillofacial Surgery and Diagnostic Sciences, College of Medicine and Dentistry, Riyadh Elm University, Riyadh, Saudi Arabia
[2]Department of Preventive Dental Sciences, College of Dentistry, Princess Nourah bint Abdulrahman University, P.O. Box 84428, Riyadh 11671, Saudi Arabia

Introduction

The oral microbiota is a complex ecosystem that includes a diverse array of microorganisms, and yeasts are among the normal inhabitants. Yeasts exist as symbiotic inhabitants in the oral flora and oral yeasts carriage (OYC) refers to the presence of various yeast species in the oral cavity with *Candida albicans* (*C. albicans*) being the most prevalent genus [1]. The OYC is usually assessed using traditional methods such as the concentrated-oral-rinse-culture technique [2]; however, these microbes have also been identified in subgingival biofilm (SB) [3]. Habitual tobacco smoking is a potential risk factor for alterations in the oral microbiota, including an increased prevalence of *Candida* species [2, 4]. Under such circumstances, these commensal microbes can transform into opportunistic pathithogens. Elevated yeast carriage is frequently implicated in the onset and advancement of oral mucosal conditions such as candidiasis [5]; nevertheless, scientific evidence indicates that oral yeasts (OY) might be a factor in the development of peri-implant diseases, specifically peri-implant mucositis (PM) and peri-implantitis [6, 7].

During initial phases, peri-implant diseases are restricted to soft tissues and this condition is identified as PM. The PM is characterized by the presence of gingival erythema, gingival bleeding (GB), increased probing depth (PD), and the absence of radiographic crestal bone loss (CBL) [8, 9]. However, when not promptly diagnosed and treated, the inflammatory condition of the soft tissues intensifies, ultimately posing a threat to the osseous tissues surrounding the implant, leading to peri-implantitis. Souza et al. [7] demonstrated that implant surfaces can be colonized by OY, particularly *Candida* species. Similarly, Aldosari et al. [10] assessed the subgingival yeasts colonization (SYC) among patients with PM. This study [10] confirmed the presence of yeasts in SB among all PM patients. Furthermore, it has been suggested that SYC fosters the growth of pathogenic bacteria such as *Porphyromonas gingivalis* (*P. gingivalis*), *Treponema denticola*, *Prevotella intermedia* and *Aggregatibacter actinomycetemcomitans*, resulting in heightened virulence of yeasts and subsequent damage to the soft tissues [7]. It is noteworthy that cigarette-smoking is a classical risk-factor of periodontal and peri-implant diseases including PM [11, 12]; and aside from its detrimental impact on oral mucosal and periodontal/peri-implant tissues, nicotine (a major component in tobacco) significantly influences the oral microbiome by modifying the growth, attachment, and biofilm formation of pathogenic microorganisms, including yeasts [13]. Experimental results by Haghighi et al. [14] showed that nicotine possibly influences the pathogenic traits of yeast, which include aspects such as hyphal growth, biofilm formation, and expression of genes associated with virulence. The present observational clinical investigation is based on the hypothesis that SYC is higher in cigarette-smokers with PM in contrast to non-smokers with and without PM.

The purpose of this investigation was to assess the peri-implant clinical profile and SYC among cigarette-smokers with PM.

Materials and methods

Ethical guidelines

The research protocol underwent rigorous review and received approval from an independent ethical committee, confirming its compliance with ethical standards. Participation in the study was entirely voluntary, and all individuals provided informed consent before inclusion. Throughout the study, participants were accorded the freedom to ask questions and seek clarifications regarding any aspect of the research. Importantly, all participants retained the autonomy to withdraw from the study at any stage without incurring any penalties or negative consequences. This commitment to voluntary participation, informed consent, and participant autonomy underscores our dedication to upholding the highest ethical standards in clinical research. The ethical approval was granted by the Institutional Review Board at the Riyadh Elm University, Riyadh, Saudi Arabia (Registration No. FRP/2024/543).

Protocol for patient eligibility for inclusion

For inclusion, the following criteria were implemented: (a) individuals aged at least 18 years; (b) self-reported cigarette-smokers (Individuals that reported to be smoking at least one cigarette daily for the past 12 months) [12]; (c) Self-reported non-smokers (individuals that reported to have never used any form of combustible and/or non-combustible nicotinic product) [12]; (d) Individuals with at least one dental implant in function for the past 180 days; (e) individuals diagnosed with peri-implant mucositis [15, 16]. Exclusion from the present study was based on the following: (a) dual smokers (individuals smoking cigarettes and using other forms of combustible nicotinic products such as cigars, pipe, waterpipe etc.); (b) Individuals who self-report systemic conditions, including cardiovascular diseases, metabolic disorders like obesity and diabetes mellitus (DM), renal and/or hepatic diseases, respiratory conditions such as chronic obstructive pulmonary disease, as well as those self-reporting viral infections like coronavirus disease-19 and acquired immune deficiency syndrome/HIV infections, along with individuals having oral/systemic malignancy; (c) pregnant or/and nursing females; (d) individuals using smokeless tobacco products; (e) individuals that reported to be currently using or had used antibiotics, antifungal medications, cancer therapy, and steroids and/or non-steroidal anti-inflammatory drugs within the past three-months

and (f) individuals diagnosed with peri-implantitis [15, 16].

Definition of peri-implant mucositis and peri-implant health

The presence of the following characteristics was used to define PM: peri-implant GB, coupled with signs such as redness, swelling, or suppuration, and without any associated CBL [15, 16]. A healthy peri-implant status was defined as the absence of peri-implant gingival, redness, swelling, GB and/or pus discharge, along with the absence of any indicators of inflammation [16].

Groups

Study participants were categorized into four groups: Group-1—Cigarette-smokers with PM; Group-2—Cigarette-smokers without PM; Group-3—Non-smokers with PM; and Group-4—Non-smokers without PM.

Questionnaire and evaluation of dental records

The principal investigator administered a questionnaire to all participants, collecting relevant information on duration and daily frequency of cigarette smoking (pack years [PY]), age, gender, any familial history of smoking and most recent visit to a dentist and/or dental hygienist. Participants were also asked about their daily frequencies of toothbrushing and flossing. Among cigarette smokers, sub-classification was performed into three subgroups: light-smokers (up to 20 PY), moderate-smokers (20.1–40.0 PY), and heavy-smokers (more than 40 PY) [17]. The following information was collected by the principal investigator from the individuals' digital dental healthcare records: (a) implant length; (b) implant diameter; (c) implant insertion torque; (d) depth of insertion (crestal or subcrestal); (e) implant abutment connection (platform switching); (f) jaw location (maxilla and/or mandible); (g) implant surface characteristic (moderately rough or smooth); (h) mode of implant prosthesis retention (screw or cement); and (i) segment of jaw in which, the implant was placed— implants replacing missing central incisors, lateral incisors, and/or canines were categorized as being positioned within the "anterior" region of the jaw and implants replacing missing premolars and/or molars, were categorized as being positioned within the "posterior" region of the jaw (Supplementary file attached).

Clinical and radiologic investigations

All clinical and radiologic investigations were performed before microbial investigations. Peri-implant indices, namely modified plaque index (mPI) [18], modified gingival index (mGI) [19], and PD [20] were meticulously gauged by a calibrated and blinded investigator (Kappa score 0.84) on four surfaces using a graded probe (UNC, HuFriedy, Chicago, IL, United States). The CBL on both mesial and distal surfaces of the implants was quantified through digital radiographs (Planmeca Romexis Intraoral X-Ray, Planmeca OY, Helsinki, Finland). This measurement involved determining the linear distance from the implant abutment interface to the alveolar crest [21]. The CBL assessments were conducted by a calibrated and blinded investigator (Kappa score 0.88) and documented in millimeters.

Collection of subgingival oral biofilm samples

The SB samples were obtained in accordance with a previously outlined protocol [10, 22]. Briefly, patients were comfortably seated in a dental chair, and the SB collection procedure was explained in a clear and accessible manner. Participants were encouraged to seek clarification or pose questions before the commencement of SB sample collection. To ensure isolation of peri-implant tissues, cotton rolls were utilized, and any supragingival plaque was delicately removed using sterile plastic hand curettes (Implan Prophy® Plastic Dental-Instrument-System-Kit, Tess Corporation, WI, USA). Subsequently, SB samples were gathered employing sterile plastic curettes (Implan Prophy® Plastic Dental-Instrument-System-Kit, Tess Corporation, WI, USA). The curette was gently inserted into the buccal and lingual peri-implant pockets, ensuring thorough contact with the subgingival area. Careful attention was given to minimize trauma to the surrounding tissues and prevent bleeding during sample retrieval. The collected SB samples were then carefully placed into a sterile plastic container equipped with a lid and containing phosphate-buffered-saline (PBS). All samples were subjected to further analysis within 30 min of collection.

Assessment of subgingival yeasts colony forming units

The determination of subgingival yeasts colony forming units followed the procedure outlined in previous studies [23, 24]. In summary, samples underwent vortexing at 1,000 rpm for 10 min, and the resulting pellet was resuspended in 1 ml PBS. Subsequently, a sample volume of 20 µl was extracted and evenly streaked using a sterile glass spreader across duplicates of Sabouraud's Dextrose Agar plates for culture. The plates were then incubated at 37 °C, and after 48 h, the *Candida* colonies were enumerated to calculate the colony-forming units per ml (CFU/ml) of SB. These investigations were performed by a trained, calibrated (kappa score 0.88) and blinded investigator.

Sample-size estimation (power analysis) and statistical analyses

Power analysis was conducted utilizing G*power version 3.0.10 (Franz Faul, Universität Kiel, Germany). The determination of sample size indicated that 18 individuals per

group would yield an 88% power to detect a genuine difference of 2 mm in probing depth (PD), which served as the primary outcome variable, between cigarette-smokers and non-smokers. This calculation was based on a two-tailed comparison with an alpha value of 0.05. Group comparisons were executed using one-way analysis of variance, and Bonferroni Post hoc adjustment tests were applied. Logistic regression analysis was employed to assess the correlation between SYC measured in colony-forming units per milliliter (CFU/ml) and variables such as age, gender, pack-years, clinicoradiographic parameters, and the duration of implants in function. The threshold for statistical significance was established at $P<5\%$.

Results

Study participants

During the patient screening phase, invitations were extended to 127 individuals for participation in the current study, comprising 98 males and 29 females. Eleven males with self-reported DM and seven dual-smokers were subsequently excluded. None of the invited females ($n=29$) opted to participate, and reasons for their non-participation were not disclosed. Consequently, a total of 80 male individuals proceeded to sign the informed consent form. These individuals were divided into four groups as follows: Group-1—Cigarette-smokers with PM ($n=20$); Group-2—Cigarette-smokers without PM ($n=19$); Group-3—Non-smokers with PM ($n=21$); and Group-4—Non-smokers without PM ($n=20$). These results are illustrated in Fig. 1.

Characteristics of patient cohort

A total of 80 male individuals agreed to participate in the present investigation and signed the informed consent form. Twenty, 19, 21 and 20 individuals were included in groups 1, 2, 3 and 4, respectively. There was no difference in the mean age of individuals in all groups. In groups 1 and 2, cigarette-smokers had a smoking history of 25.9±12.1 and 11.5±2.5 pack years, respectively. In groups 1 and 2, 60% and 15.8% of the individuals were moderate smokers, respectively. The family history of smoking was more often reported by individuals in groups 1 and 2 (75% and 68.4%, respectively) compared with individuals in groups 3 and 4 (19.04% and 15%, respectively). Toothbrushing twice daily was more often performed by individuals in groups 2 and 4 (84.2% and 85%, respectively) compared with individuals in Group-1 (15%). In Group-3, all individuals reported that they brushed teeth once daily. None of the individuals in groups 1 and 3 were performing flossing of interproximal spaces; and 78.9% and 80% of the individuals in groups 2 and 4, respectively were performing interproximal flossing once daily. In groups 1, 2, 3 and 4, participants visited a dentist/dental hygienist 3.1±1.6, 0.8±0.2, 3.5±1.5 and 0.7±0.2 years ago, respectively. These results are shown in Table 1.

Implant-related characteristics

All implants were platform-switched, were placed at bone-level and had moderately rough surfaces. All implants were loaded with cement-retained restorations and diameters and lengths ranging between 4.1
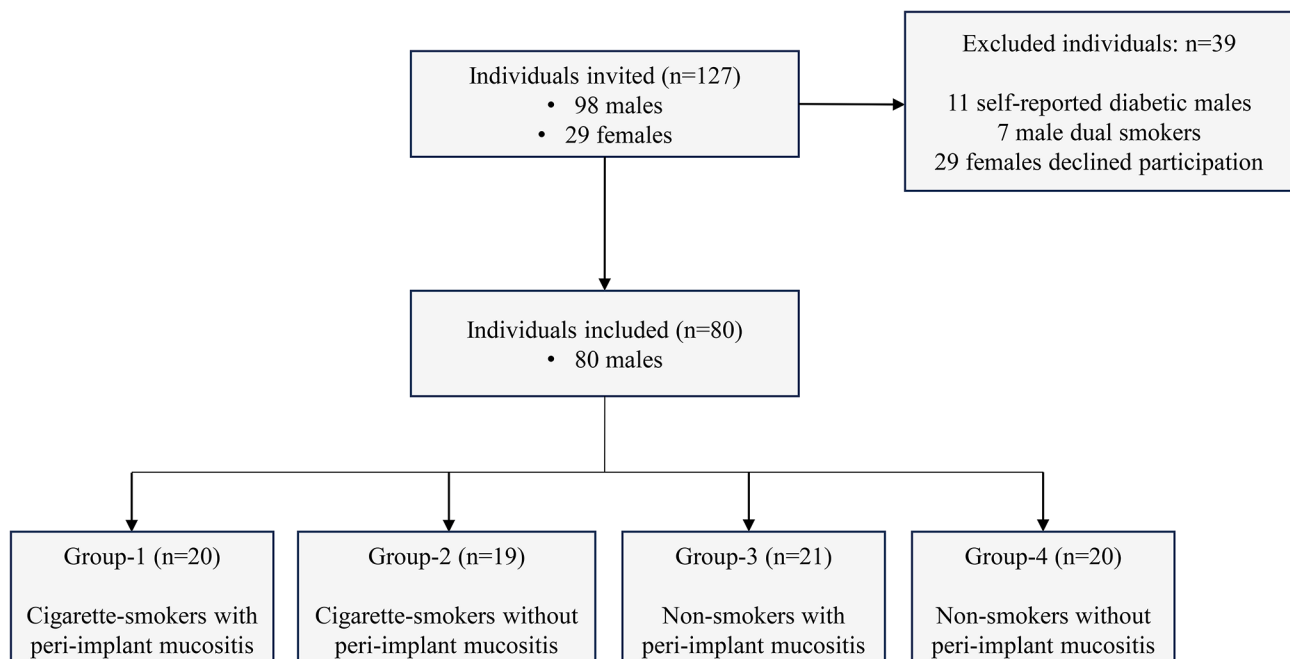
Fig. 1 Recruitment of study participants

Table 1 Characteristics of patient cohort

Parameters	Group-1	Group-2	Group-3	Group-4
Participants (n)	20	19	21	20
Gender (male)	20	19	21	20
Age in years (mean ± SD)	50.1 ± 4.5 years	52.6 ± 5.2 years	55.3 ± 5.8 years	51.8 ± 5.1 years
Pack years			NA	NA
Light smokers (n)	11.8 ± 3.17 pack years ($n=6$)	11.5 ± 2.5 pack years ($n=19$)	NA	NA
Moderate smokers (n)	29.2 ± 5.5 pack years ($n=12$)	None	NA	NA
Heavy smokers (n)	49 ± 4.9 pack years ($n=2$)	NA	NA	NA
All smokers (n)	25.9 ± 12.1 pack years ($n=20$)	11.5 ± 2.5 pack years ($n=19$)	NA	NA
Family history of smoking (%) (n)	75% ($n=15$)	68.4% ($n=13$)	19.04% ($n=4$)	15% ($n=3$)
Toothbrushing				
Once daily	85% ($n=17$)	15.8% ($n=3$)	100% ($n=21$)	15% ($n=3$)
Twice daily	15% ($n=3$)	84.2% ($n=16$)	NA	85% ($n=17$)
Not at all	NA	NA	NA	NA
Interproximal flossing				
Once daily	None	78.9% ($n=15$)	None	80% ($n=16$)
Twice daily	None	NA	None	None
Not at all	100% ($n=20$)	21.1% ($n=3$)	100% ($n=21$)	20% ($n=4$)
Last visit to dentist/hygienist	3.1 ± 1.6 years ago	0.8 ± 0.2 years ago	3.5 ± 1.5 years ago	0.7 ± 0.2 years ago

Group-1—Cigarette-smokers with PM; Group-2—Cigarette-smokers without PM; Group-3—Non-smokers with PM; and Group-4—Non-smokers without PM; light-smokers (up to 20 PY); moderate-smokers (20.1–40.0 PY), and heavy-smokers > 40 PY

Table 2 Characteristics of implants in the study groups

Parameters	Group-1	Group-2	Group-3	Group-4
Number of implants	20	19	21	20
Jaw location				
Maxilla : Mandible	12 : 8	10 : 9	12 : 9	10 : 10
Anterior maxilla	None	1 implant	4 implants	2 implants
Anterior mandible	None	None	None	None
Posterior maxilla	12 implants	10 implants	4 implants	8 implants
Posterior mandible	8 implants	8 implants	13 implants	10 implants
Implant surface	Moderately rough	Moderately rough	Moderately rough	Moderately rough
Implant abutment connection	Platform switched	Platform switched	Platform switched	Platform switched
Implant dimensions (diameter x length)	4.1 to 4.8 / 10 to 14 mm	4.1 to 4.8 / 10 to 14 mm	4.1 to 4.8 / 10 to 14 mm	4.1 to 5 / 10 to 14 mm
Insertion torque	30 to 35 Ncm	30 to 35 Ncm	30 to 35 Ncm	30 to 35 Ncm
Depth of implant insertion	Crestal	Crestal	Crestal	Crestal
Implant prosthesis retention	Cement-retained	Cement-retained	Cement-retained	Cement-retained
Duration in function (in years)	3.15 ± 0.9 years[*]	0.7 ± 0.5 years	1.7 ± 0.8 years	5.2 ± 1.4 years[†]

Group-1—Cigarette-smokers with PM; Group-2—Cigarette-smokers without PM; Group-3—Non-smokers with PM; and Group-4—Non-smokers without PM. Posterior: Implants located in areas of missing premolars or molars; Anterior: Implants located in areas of missing incisors or canines [*]Compared with group 2 ($P<0.05$) and 3 ($P<0.05$), [†]Compared with group 2 ($P<0.05$) and 3 ($P<0.05$)

and 4.8 and 10 and 14 mm, respectively. The total number of implants in groups 1, 2, 3 and 4 were 20, 19, 21 and 20, respectively. In groups 1, 2, 3 and 4, 12, 10, 12 and 10 implants were in the maxilla, respectively; and the remaining were located in the mandible. In all groups, most of the implants were present in the posterior jaws in the regions of missing premolars or molars. In all groups, the implants had been inserted at insertion torques ranging from 30 to 35 Ncm and were in function for 3.15 ± 0.9, 0.7 ± 0.5, 1.7 ± 0.8 and 5.2 ± 1.4 years in groups 1, 2, 3 and 4, respectively as shown in Table 2.

Clinical and radiographic peri-implant parameters

The mPI was significantly higher in Group-1 compared with Group-2 ($P<0.05$) and Group-4 ($P<0.05$). The mPI was significantly higher in Group-3 comparted with Groups 2 ($P<0.05$) and 4 ($P<0.05$). The mGI was significantly higher in Group-3 compared with groups 1 ($P<0.05$), 2 ($P<0.05$) and 4 ($P<0.05$). The PD was significantly higher in Group-1 compared with Group-2 ($P<0.05$) and Group-4 ($P<0.05$). The PD was significantly higher in Group-3 comparted with Groups 2 ($P<0.05$) and 4 ($P<0.05$). There was no statistically significant difference in medial and distal CBL in all groups (Table 3).

Table 3 Peri-implant clinical and radiological parameters

Peri-implant parameters	Group-1	Group-2	Group-3	Group-4
Modified plaque index	0.88 ± 0.05[*†]	0.2 ± 0.005	0.54 ± 0.04[§]	0.22 ± 0.06
Modified gingival index	0.23 ± 0.03	0.23 ± 0.08	0.84 ± 0.12[‡]	0.21 ± 0.08
Probing depth (in mm)	5.8 ± 1.05 mm[*†]	1.2 ± 0.16 mm	4.1 ± 0.08 mm[§]	1.1 ± 0.2 mm
Crestal bone loss (mesial)	0.54 ± 0.15 mm	0.35 ± 0.12 mm	0.45 ± 0.16 mm	0.26 ± 0.05 mm
Crestal bone loss (distal)	0.56 ± 0.17 mm	0.38 ± 0.16 mm	0.48 ± 0.15 mm	0.28 ± 0.07 mm

Group-1—Cigarette-smokers with PM; Group-2—Cigarette-smokers without PM; Group-3—Non-smokers with PM; and Group-4—Non-smokers without PM

[*]Compared with Group-2 ($P < 0.05$) [†]Compared with Group-4 [‡]Compared with Group-1 ($P < 0.05$), Group-2 ($P < 0.05$) and Group-4 ($P < 0.05$)

Table 4 Isolation and colony for units of yeasts in the subgingival biofilm

Parameters	Group-1	Group-2	Group-3	Group-4
Isolation of yeasts (n)	20 (100%)	7 (36.8%)	14 (66.7%)	4 (20%)
Colony forming units	1487.4 ± 205.3 CFU/ml[*]	787.6 ± 102.3 CFU/ml	1026.4 ± 114.7 CFU/ml[†]	236.2 ± 71.5 CFU/ml

[*]Compared with group-2 ($P < 0.05$) and Group-4 ($P < 0.05$)

[†]Compared with group-2 ($P < 0.05$) and Group-4 ($P < 0.05$)

### Isolation and colony for units of yeasts in the subgingival biofilm

Yeasts were isolated from SB in 100%, 36.8%, 66.7% and 20% individuals in groups 1, 2, 3 and 4, respectively. The CFU/ml were significantly higher in Group-1 compared with individuals in groups 2 ($P < 0.05$) and 4 ($P < 0.05$). The CFU/ml were significantly higher in Group-3 compared with individuals in groups 2 ($P < 0.05$) and 4 ($P < 0.05$) (Table 4).

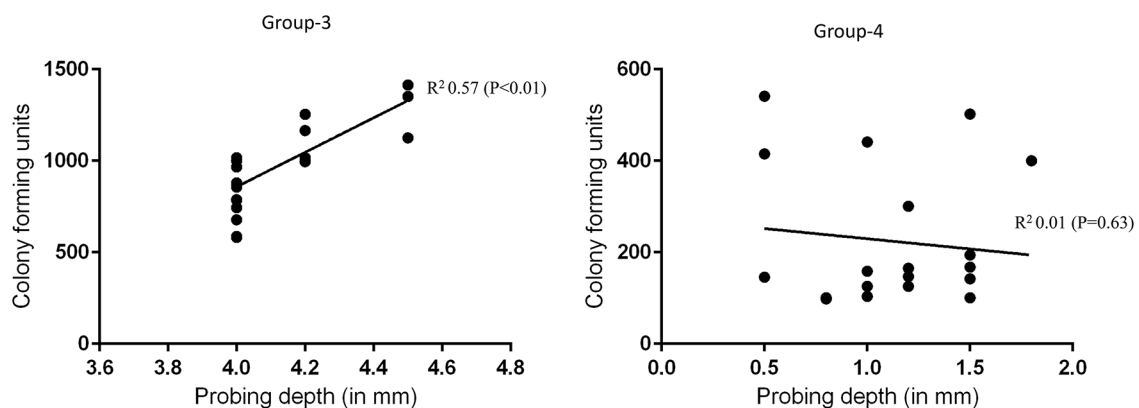### Correlation between smoking pack years, probing depth, interproximal flossing and oral yeasts colony forming units

In Group-1, the yeasts CFU/ml in the SB were statistically significantly correlated with smoking pack-years ($P < 0.001$) and peri-implant PD ($P < 0.01$). In Group-2, there was no statistically significant correlation between smoking pack-years and peri-implant PD and yeasts CFU/ml in the SB (Fig. 2). In Group-3, there was a statistically significant correlation between peri-implant PD and yeasts CFU/ml in the SB ($P < 0.01$); whereas in Group-4, there was no statistically significant correlation between peri-implant PD and yeasts CFU/ml in the SB (Fig. 3). In Group 2, there was a statistically significant correlation between daily flossing of interproximal spaces and yeasts CFU/ml in the SB (Fig. 4). There was no correlation between daily flossing of interproximal spaces and yeasts CFU/ml in the SB in groups 1, 3 and 4. There was no correlation between age, family history of smoking, implant dimensions (diameter and length), duration for which implants were in function, mPI, mGI and CBL and yeasts CFU/ml in the SB in all groups.

Discussion

The authors applaud results from a clinical investigation by Canabarro et al. [25], which investigated the potential connection between the SYC and severity of periodontal disease. The results showed that the CFU/ml of OY were significantly higher in the SB samples collected from patients with periodontitis in contrast to those collected from individuals with a healthy periodontal status. Authors of the present investigation applaud the results by Canabarro et al. [25] as CFU/ml of yeasts in SB were significantly higher among cigarette-smokers and non-smokers with PM (Group-1 and Group-3) compared with non-smokers without PM (Group-4). It is suggested that a variety of mechanisms contribute to this context. Nicotine is a major component of tobacco, which has been shown to have immunomodulatory effects [26]. Studies [6, 27, 28] have proposed that chronic nicotine exposure to tissues suppresses the immune response creating an immunocompromised state, which allows opportunistic microbes including yeasts to thrive and colonize the oral mucosa. Moreover, nicotine induced changes in the oral epithelium may create microlesions and/or disruptions providing entry-points for yeasts species to adhere and establish infections [29]. In a laboratory-based investigation focusing on titanium surfaces [30], the virulence of OY (predominantly *C. albicans*) within mixed-species biofilms, which included *P. gingivalis* and *Streptococcus sanguis* was assessed. The findings revealed that when coexisting with pathogenic bacteria, including the aforementioned species, *C. albicans* exhibited an elevated proportion of hyphae and an upregulation of hydrolytic enzymes [30]. The study [30] concluded that in conjunction with pathogenic bacteria found in oral biofilms, *C. albicans* expresses virulence factors that could potentially contribute to the development of peri-implant diseases. Furthermore, according to Nagler RM [31] nicotine jeopardizes salary flow rates and composition thereby compromising the ability of saliva to inhibit microbial growth; thus contributing to an environment favorable for yeasts colonization. Nevertheless, it is important to note that the relationship between nicotine and OYC is complex and further research is needed to elucidate the underlying mechanisms. Additionally, other components of tobacco smoke and their interactions with nicotine
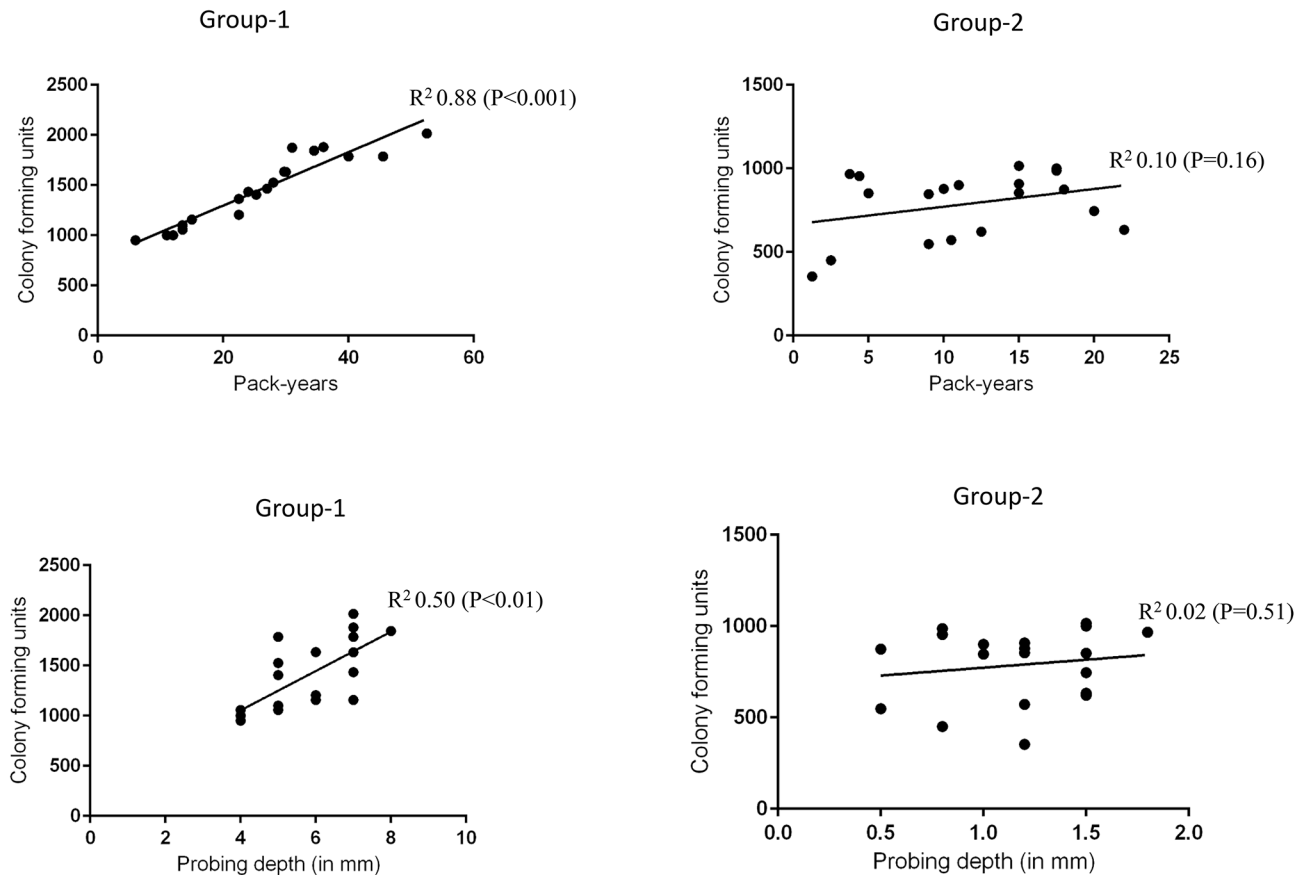
Fig. 2 Correlation between smoking pack-years and peri-implant probing depth and yeasts colony forming units per milliliter in the subgingival biofilm in groups 1 and 2


Fig. 3 Correlation between smoking pack-years and peri-implant probing depth and yeasts colony forming units permilliliter in the subgingival biofilm in groups 3 and 4

may also contribute to the observed effects on oral and peri-implant tissues.

An intriguing observation was noted in Group-2, where individuals, despite being cigarette smokers, exhibited healthy peri-implant tissues with significantly lower colony-forming units per milliliter (CFU/ml) in the SB compared to Group-1, as outlined in Table 4. One plausible explanation for this discrepancy may be linked to the

duration of the cigarette smoking habit within these two groups. Notably, individuals in Group-1 had an approximate smoking history of 26 pack years, while those in Group-2 had a significantly shorter smoking duration, totaling nearly 12 pack years. The diminished duration of smoking among individuals in Group-2 could potentially account for the lower yeast CFU in the SB compared to Group-1. Another factor worth considering is the
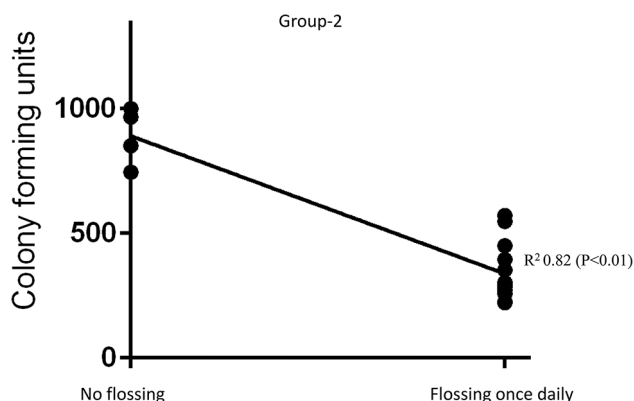
Fig. 4 Correlation between flossing of interproximal spaces and yeasts colony forming units per milliliter in thesubgingival biofilm in Group-2

duration for which implants were functional. In Group-1, the implants had been in function for approximately 3 years, whereas in Group-2, the duration was markedly shorter at around 8 months. This discrepancy in implant duration might explain the absence of peri-implant diseases and the lower yeast CFU/ml observed in Group-2. It is noteworthy, however, that a substantial majority (at least 80%) of individuals in Group-2 adhered to a diligent oral hygiene routine, including brushing twice daily and daily flossing of interproximal spaces in nearly 79% of the population. Furthermore, individuals in Group-2 seemed to be visiting oral healthcare providers and attaining routine dental prophylaxis compared with individuals in groups 1 and 3. The possible contribution of these factors towards maintaining peri-implant health in these patients cannot be overlooked. The results of logistic regression analysis indicated a significant increase in CFU/ml for specific SYC among individuals in Group-2 who did not engage in daily flossing of interproximal spaces compared to those who practiced daily dental flossing within the same group. Despite these positive oral hygiene practices, it is essential to emphasize that the maintenance of routine oral hygiene should not be construed as justification for the continued use of nicotinic products in general. In a prior investigation, Krishnan et al. [22] reported an elevated presence of SYC in the SB among individuals with periodontitis, regardless of their smoking status. Building upon this research, the current study establishes a statistically significant correlation between SYC, smoking duration (pack-years), and periodontal PD. The authors of this study commend these findings, noting a significant increase in SYC within the SB among both smokers and non-smokers with peri-implant mucositis (groups 1 and 3, respectively). In both groups, a robust statistical correlation was identified between pack-years of smoking and PD. This suggests that individuals classified as moderate and heavy cigarette smokers face an elevated risk of developing peri-implant diseases and hosting elevated

colonies of pathogenic microbes, including yeast species, within the SB, as compared to their counterparts who are light smokers or non-smokers. It is strongly recommended that community-based initiatives, focusing on anti-tobacco measures and oral health promotion, be regularly conducted to educate the public about the adverse impacts of smoking on oral, periodontal, peri-implant, and overall health. Emphasizing the advantages of consistent oral hygiene practices is crucial for fostering a superior oral health-related quality of life.

One limitation of the present study is that all participants were male. It has been reported that OYC varies among males and females with periodontal inflammation [21]. The postmenopausal phase has been associated with an increased OYC in females compared with males [21]. Therefore, it is hypothesized that the SYC in in the peri-implant sulci of females than males with peri-implant diseases. Moreover, identification of yeasts species was not performed in the present study. The primary reasoning for this was the limitation of resources that hindered further analyses such as polymerase chain reaction and DNA sequencing for yeast species identification. It is however, anticipated that most of the yeasts species in the SB were *C. albicans* as it is the most common yeasts species isolated from the oral cavity according to previous oral investigations [2, 3, 32]. Further power adjusted and well-designed studies are needed to test these hypotheses.

Limitations

One limitation of the current study lies in the exclusive inclusion of male participants. Previous research has indicated variations in oral yeast carriage between males and females with periodontal inflammation, with postmenopausal females demonstrating increased oral yeast colonization compared to males [21]. Consequently, it is postulated that females may exhibit higher SYC counts in peri-implant sulci than males in the context of peri-implant diseases. Additionally, the identification of yeast species was not undertaken in the present study. This decision was influenced by resource limitations that impeded more extensive analyses, such as polymerase chain reaction and DNA sequencing for yeast species identification. However, it is anticipated that most yeast species in the SB were *C. albicans*, given its prevalence as the most isolated yeast species from the oral cavity in previous investigations. Furthermore, immunocompromised individuals such as diabetic patients were excluded from the current investigation. It has been reported that peri-implant inflammatory conditions are worse and SYC is higher in diabetic compared with systemically healthy individuals [8, 33]. Therefore, it is likely that the CFU/ml of SYC in SB of diabetic smokers is higher than non-diabetic smokers and non-smokers. Additional robust, and

adequately powered studies are warranted to empirically test these hypotheses.

Conclusion

Peri-implant soft-tissue inflammatory parameters are worse and SYC is higher in moderate smokers than light smokers with PM and non-smokers without PM.

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s12903-024-04868-5.

Supplementary Material 1

Data availability
Data is available upon reasonable request.

Declarations

Human ethics and consent to participate
The research protocol underwent rigorous review and received approval from an independent ethical committee, confirming its compliance with ethical standards. This clinical study adhered to the principles of research ethics, ensuring the protection and well-being of participants. Participation in the study was entirely voluntary, and all individuals provided informed consent before inclusion. Throughout the study, participants were accorded the freedom to ask questions and seek clarifications regarding any aspect of the research. Importantly, all participants retained the autonomy to withdraw from the study at any stage without incurring any penalties or negative consequences. This commitment to voluntary participation, informed consent, and participant autonomy underscores our dedication to upholding the highest ethical standards in clinical research. The ethical approval was granted by the Institutional Review Board at the Riyadh Elm University, Riyadh, Saudi Arabia (Registration No. FRP/2024/543).

Clinical trial number
Not applicable.

Consent for publication
Not applicable.

Competing interests
The authors declare no competing interests.

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