Association of Self-Rated Pain with Clinical Peri-Implant Parameters and Cytokine Profile in Smokers and Never Smokers with and without Peri-Implantitis

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Abstract: To examine the association between self-perceived pain (SPP), clinical and radiographic peri-implant parameters, and biomarker levels among smokers and never smokers with and without peri-implantitis. Sixty individuals (20 smokers with peri-implantitis [group-1], 20 never smokers with peri-implantitis [group-2] and 20 never smokers without peri-implantitis [control-group]) were included. SPP was evaluated using a numeric pain rating scale (NPRS). Peri-implant plaque index (PI), probing depth (PD), and bleeding on probing (BOP) were recorded. After obtaining the samples, the levels of TNF-α, MMP-1, and IL-8 were measured. The mean SPP score in group-1, group-2, and the control group was 1.3 ± 0.05, 3.4 ± 0.5, and zero, respectively. The peri-implant mean PD (p < 0.05), BOP (p < 0.05), PI (p < 0.05), and crestal bone loss (CBL) (p < 0.05) were significantly higher among test groups than the control group. The levels of TNF-α, MMP-1, and IL-8 were significantly raised among group-1 and group-2 than the control group. A significant correlation between increasing SPP and PICF TNF-α, MMP-1, and IL-8 levels was observed based on regression analysis. Proinflammatory biomarkers were higher in smokers with peri-implantitis than never smokers with and without peri-implantitis, with a significant association between the proinflammatory cytokines and SPP.

Keywords: cigarette smoking; cytokines; pain; peri-implantitis; peri-implant crevicular fluid; inflammation

1. Introduction

Pain is an unpleasant sensory and emotional experience associated with actual or potential tissue damage or described in terms of such damage [1]. It can be associated with inflammation of soft tissue surrounding the dental implant and crestal bone loss (CBL); also called peri-implantitis [2]. A systematic review reported that the evaluation of self-perceived pain (SPP) is also a diagnostic parameter of peri-implantitis besides other diagnostic criteria such as CBL, bleeding on probing (BOP), probing depth (PD), and suppuration [3]. Furthermore, the International Congress of Implantologists (ICOI) also suggested SPP to be utilized as a diagnostic parameter for implant failure and peri-implantitis [4] The numeric pain rating scale (NPRS), developed by Downie et al. [5], is a valid and reliable scale often utilized to examine the SPP in subjects with musculoskeletal diseases, for instance, osteoarthritis [6]. The NPRS has also been utilized to evaluate links with dental implant surgeries [7].
The abutment screw loosening or failure is a rare, but very unpleasant failure. Several studies have reported that, after dental implant osseointegration, abutment screw loosening seems to be the most frequent issue linked with implants [8–11]. Many factors contribute to the etiology of these technical failures, such as fabrication failures, non-passive fit of the suprastructures, improper placement technique, fatigue, excessive occlusal load, and the utilization of unfavorable components including copy products [12]. A few reports suggest that most of the failures are related to the suprastructure rather than to the dental implants themselves [13].

Cigarette smoking is a well-established predisposing factor for both peri-implant and periodontal soft tissue inflammation; [14] and CBL surrounding dental implants and natural teeth [15]. One justification in this aspect might be that the generation and deposition of advanced glycation end products (AGEs) in the periodontal tissues is increased by cigarette smoking [16]. The linkage of AGEs with their receptors (RAGE), inflammatory cytokines including matrix metalloproteinase (MMP)-1, interleukin (IL)-8, and tumor necrosis factor-alpha (TNF-α) are generated by human gingival fibroblasts that stimulate inflammation [17]. Furthermore, collagen degradation is increased by cigarette smoking via influencing tissue inhibitors of metalloproteinases [18]. Moreover, the RAGE’s expression in the gingival tissues is upregulated; the production of reactive oxygen species (ROS) is stimulated by metabolites of nicotine, i.e., nornicotine which damages the periodontal tissues. Besides contributing to delayed periodontal wound healing, these variables might also compromise peri-implant tissue healing. The destructive inflammatory cytokines cause deteriorating clinical peri-implant parameters including plaque index (PI), PD, gingival index (GI), elevated CBL on the distal and mesial aspects of the dental implant. If untreated, implant failure may be caused [19]. Alqahtani et al. [20] evaluated the clinical peri-implant parameters and levels of cotinine in peri-implant crevicular fluid (PICF) of thirty-five never-smokers and thirty-five tobacco smokers. The findings reported that cotinine levels and peri-implantitis in the PICF were considerably lower in never smokers as compared to tobacco smokers. Furthermore, studies have suggested that nicotine harms human periodontal ligament cells and gingival fibroblasts by increasing the generation of inflammatory mediators and compromising cellular proliferation [21,22]. Regardless of its harmful effects on peri-implant and periodontal health, cigarette smoking is not an absolute contraindication to dental implant treatment [23].

To date, no study has reported the association between PICF levels of TNF-α, MMP-1, and IL-8 with clinic-radiographic peri-implant parameters (PI, PD, BOP, and CBL) along with SPP among patients with peri-implantitis. In the current study, it is hypothesized that clinical and radiographic peri-implant parameters along with pain scores are worse and the levels of TNF-α, MMP-1, and IL-8 in the PICF are higher in cigarette smokers with peri-implantitis in comparison with never-smokers with peri-implantitis. The present study aimed to assess the association between SPP, clinical and radiographic peri-implant parameters, and levels of PICF TNF-α, MMP-1, and IL-8 among cigarette smokers and never smokers with and without peri-implantitis.

2. Materials and Methods

2.1. Ethical Considerations

This study was submitted, reviewed, and approved by King Saud University, Riyadh, Saudi Arabia (UDRC/019-12). After reading the informed consent document, written in simple Arabic and English, all the volunteering participants provided written informed consent. The ethical standards of the 1964 Helsinki declaration and national and/or institutional research committee were strictly followed while performing all the procedures. All the participants were aware that they had the right to withdraw at any time throughout the study course without any indirect or direct consequences.
2.2. Inclusion and Exclusion Criteria

The inclusion criteria comprised of patients with the following characteristics: (1) reading and signing the written informed consent document; (2) patients aged ≥ 25 years who underwent dental implant treatment; (3) individuals suffering from peri-implantitis, i.e., peri-implant BOP at ≥30% sites, PD ≥ 4 mm, and CBL ≥ 3 mm [24]; (4) individuals without peri-implantitis, i.e., BOP of <30% sites, PD of <4 mm, and CBL of <3 mm [24]; (5) self-reported tobacco consumers: subjects who have been smoking a minimum of one cigarette/day over the past 2 years [25]; (6) never smokers: persons that stated to have never utilized tobacco in any form [26].

The exclusion criteria consisted of individuals with the following characteristics: (1) individuals who refused to participate and/or sign the written consent document; (2) individuals consuming smokeless tobacco products and/or areca nut; (3) dual smoker, i.e., persons smoking tobacco along with other nicotinic products including waterpipe, bidi, and electronic cigarettes; (4) individuals who consumed alcohol habitually; (5) completely edentulous individuals; (6) individuals with maligned dentition; (7) patients suffering from medical conditions such as diabetes mellitus, renal and hepatic disorders, cardiovascular diseases, and patients suffering from the HIV infection; (8) gestation and/or nursing individuals; (9) individuals who reported to have utilized steroids, NSAIDs, probiotics, antibiotics and bisphosphonates over the past 3 months.

2.3. Study Design and Participants

This is a retrospective case-control study conducted between January 2019 and February 2020 at the College of Dentistry, King Saud University, Riyadh, Saudi Arabia. The participants were classified into three groups on the basis of peri-implant status and self-reported habits of cigarette smoking: (1) cigarette smokers with peri-implantitis (Group-1); (2) never smokers with peri-implantitis (Group-2); and (3) never smokers without peri-implantitis (control-group).

2.4. Questionnaire

Information related to age, sex, highest education level, family history of smoking, daily frequency and duration of cigarette smoking, daily flossing and tooth brushing, duration of functional dental implants, location, and the number of dental implants in either jaw were collected. The questionnaire also collected data relating to SPP utilizing the NPRS. It was suggested that participants choose a number ranging from 0 to 10 that most appropriately fit their perceived pain’s intensity with 10 and 0 depicting the worst pain possible and no pain, respectively [27]. A trained investigator (T.A.) distributed the questionnaire to all participants.

2.5. Parameters Related to Dental Implant

A calibrated investigator (T.A.) gathered the data from patient’s records relating to the implant dimensions (diameter x length), jaw location (mandible or maxilla), surface characteristics (smooth or moderately rough surfaces), insertion torque (in Newton centimeters [Ncm]), implant-abutment connection (platform-switching), prosthetic restoration type (screw- or cement-retained). “Implant success” was described on the basis of the following characteristics: (1) existence of a fully functional implant within the oral cavity; (2) self-evaluated satisfaction of the patient regarding implant and the associated prosthesis [28].

2.6. Evaluation of Peri-Implant Clinic-Radiographic Parameters

As explained elsewhere [20], a trained investigator (F.A.V.) calculated and reported the peri-implant PD, BOP, PI, and CBL (distal and mesial) around every dental implant, as recommended by the specific guidelines declared in the consensus report of the Eleventh European Workshop on Periodontology [29]. To summarize, the peri-implant PD, BOP, and PI were calculated at 6 sites per implant, i.e., disto-palatal/-lingual, mid-palatal/-lingual, mesio-palatal/-lingual, distobuccal, mid-buccal, mesiobuccal. Utilizing a graded probe
(Hu-Friedy, Chicago, IL, USA), the measurement of peri-implant PD was carried out to the nearest mm. The digital bitewing radiographs, which were standardized utilizing long cone parallel methods, were employed to record the CBL [30]. The recording of the linear distance was performed 0.2 cm below the abutment–implant junction to the alveolar crest. The CBL was defined as the bone loss median measured on mesial and distal aspects of the dental implant.

2.7. Collection of PICF

PICF collection was carried out as described elsewhere [31]. The mechanical cleaning of the supragingival plaque present on the implant-supported crown was carried out. After the careful isolation of the peri-implant sites using sterile cotton, an air syringe was used for drying purposes. By introducing 1–2 mm of paper points (Periopaper, Pro Flow, Amityville, NY, USA) into the peri-implant sulcus for 30 s, the collection of PICF samples was performed. After discarding the samples contaminated with bacteria, blood, or saliva, the collection of the new specimens was carried out from the same site. An electronic calibrated gingival fluid device (Periotron 6000, Amityville, NY, USA) was utilized to measure the fluid sample volume. The elution of the collected PICF was carried out with 1 mL of phosphate-buffered saline (PBS) prior to freezing at −80 °C.

2.8. Evaluation of PICF Levels of TNF-α, MMP-1, and IL-8

Centrifugation of PICF samples was carried out at 15,000 × g for 15 min. Quantification of the levels of biomarkers was performed using enzyme-linked immunosorbent assay (ELISA) according to the manufacturer’s guidelines (Quintikine, R&D Systems, Minneapolis, MN, USA) and calculated as pg/mL. A trained investigator (P.A.) carried out all laboratory-based analysis, (i.e., kappa = 0.78). In brief, the addition of 100 mL samples and standards were incorporated into the corresponding wells. Then, their incubation was performed overnight at 4 °C after covering them properly. Later, to discard the solution, a multi-channel pipette was utilized and cleaned four times using a 0.3 mL 1 × wash solution. Post final wash, the removal of the residual liquid was carried out via aspiration. Then, the inversion of the plate was performed and blotted with sterile paper towels. Inclusion of 0.1 mL of 1 × Biotinylated anti-Human CRP Detector Antibody to individual wells was carried out. Later, this was incubated at room temperature, (i.e., 25 °C) for 60 min. The rewashing of the solution was carried out after disposing it off. Before incubating it at 25 °C for 45 min, 0.1 mL of 1 × HRP-Streptavidin solution was incorporated into each individual well. The solution was re-discarded and washed again. To each well, 0.1 mL of TMB One-Step Substrate Reagent was incorporated and incubation was carried out at 25 °C for half an hour in a dark room. The addition of 50 μL of stop solution to each well was performed before instantly taking the reading at 450 nm. Prior to each individual assay, standard curves were produced and employed for plotting outcomes. For the levels of PICF of TNF-α, MMP-1, and IL-8, the ELISA’s sensitivity was 96%, 97.2%, and 96.8%, respectively.

2.9. Statistical and Power Analysis

The Statistical Package for Social Sciences (SPSS) version 21.0 was used to perform statistical analysis. The goodness of fit test was carried out to assess the normality of the data. Unless otherwise specified, the results were shown as mean ± SD of the mean. To assess between-group differences of non-normal data distribution, the Kruskal–Wallis nonparametric test was conducted. Bivariate multiple regression analysis was carried out to evaluate the association between PICF cytokine levels and SPP. A total of 15 patients were required with 25 dental implants per group to attain a power of 90% with a probing depth of 1 mm. To allow a 20% attrition for compensation, a total of 20 patients were selected. A p-value of <0.05 was considered statistically significant.
3. Results

3.1. General Description of Study Participants

A total of 60 patients (29 males and 31 females) participated in this study and signed the written informed consent and completed the clinical trial. Group-1 and group-2 participants had a mean age of 41.7 ± 6.6 and 39.4 ± 5.8 years, respectively, while the control group had a mean age of 38.2 ± 7.3 years. Among the smokers and never smokers with peri-implantitis, 42 and 36 dental implants were examined, respectively, while 28 dental implants were evaluated among never smokers without peri-implantitis. Among group-1 and group-2, an average of dental implants in service was 37.4 ± 3.2 and 31.6 ± 2.1 months, respectively, while among the control group an average of dental implants in use was 29.9 ± 5.0 months (Table 1).

Table 1. Demographics of the study groups.

| Characteristics               | Group-I (n = 20) | Group-II (n = 20) | Group-III (n = 20) |
|-------------------------------|-----------------|-------------------|-------------------|
| Total number of samples ‘n’   | 20              | 20                | 20                |
| Mean age (±SD)                | 41.7 ± 6.6      | 39.4 ± 5.8        | 38.2 ± 7.3        |
| Gender (M/F)                  | 11/9            | 8/12              | 10/10             |
| Number of implants assessed   | 42              | 36                | 28                |
| Duration of implants in months (±SD) | 37.4 ± 3.2 | 31.6 ± 2.1        | 29.9 ± 5.0       |

3.2. Clinical and Radiographic Peri-Implant Parameters

The peri-implant mean PD (p < 0.05) and mesial (p < 0.05) and distal (p < 0.05) CBL were significantly higher among group-1 than group-2 and control group. However, the peri-implant mean BOP (p < 0.05) and PI (p < 0.05) were significantly higher in group-2 when compared with group-1 and the control group. Overall, the peri-implant mean PI (p < 0.05), BOP (p < 0.05), PD (p < 0.05), and CBL (p < 0.05) were significantly higher among test groups than the control group. The mean SPP score was highest in group-2 (3.4 ± 1) when compared with group-1 (1.3 ± 1) and the control group (0 ± 0), respectively (Table 2).

Table 2. Peri-implant clinical parameters of the research groups.

| Variables (Mean ± SD) | Group-I (n = 20) | Group-II (n = 20) | Group-III (n = 20) |
|-----------------------|-----------------|-------------------|-------------------|
| PI in % (±SD)         | 41.5 ± 8.3      | 46.6 ± 11.4       | 13.7 ± 5.1        |
| BOP in % (±SD)        | 21.6 ± 6.9      | 53.5 ± 7.7        | 15.8 ± 7.0        |
| PD in mm (±SD)        | 5.0 ± 0.5       | 4.8 ± 0.3         | 1.2 ± 0.8         |
| CBL in mm (±SD)       | 2.9 ± 0.6       | 2.7 ± 0.4         | 0.7 ± 0.02        |
| SPP score             | 1.3 ± 1         | 3.4 ± 1           | 0 ± 0             |

Dissimilar lowercase letters indicate a statistically significant difference (p < 0.05) between groups along rows. Abbreviations: BOP = bleeding on probing; CBL = crestal bone loss; PD = probing depth; PI = plaque index; SPP = self-perceived pain.

3.3. Peri-Implant Crevicular Fluid Cytokine Levels and Self-Perceived Pain

PICF levels of TNF-α, MMP-1, and IL-8 were raised significantly among group-1 and group-2 than the control group (Table 3).

Table 3. Peri-implant crevicular fluid cytokine levels of the study groups.

| Cytokines (pg/mL) | Group-I (n = 20) | Group-II (n = 20) | Group-III (n = 20) |
|-------------------|-----------------|-------------------|-------------------|
| TNF-α             | 648 ± 335       | 395 ± 182         | 87 ± 23           |
| MMP-1             | 4612 ± 2296     | 3179 ± 1498       | 174 ± 82          |
| IL-8              | 442 ± 202       | 387 ± 164         | 53 ± 22           |

Dissimilar lowercase letters indicate a statistically significant difference (p < 0.05) between groups along rows.
Statistically, a significant association was observed between SPP and TNF-α, MMP-1, and IL-8 expression in the PICF of test groups and control group as per bivariate multiple logistic regression analysis (Table 4).

Table 4. Statistical comparisons of the SPP score with PICF cytokines levels.

| SPP Score | OR   | 95% CI       | p-Value |
|-----------|------|--------------|---------|
| TNF-α     |      |              |         |
| 1         | 0.45 | 0.32–0.58    |         |
| 2         | 0.13 | 0.09–0.17    |         |
| 3         | 0.72 | 0.62–0.82    | 0.032   |
| 4         | 0.32 | 0.15–0.49    |         |
| 5         | 0.28 | 0.22–0.34    |         |
| 6         | 0.16 | 0.10–0.22    |         |
| MMP-1     |      |              |         |
| 1         | 0.15 | 0.05–0.25    |         |
| 2         | 0.36 | 0.31–0.41    |         |
| 3         | 0.52 | 0.43–0.61    |         |
| 4         | 0.23 | 0.15–0.46    | 0.044   |
| 5         | 0.19 | 0.11–0.27    |         |
| 6         | 0.08 | 0.05–0.11    |         |
| IL-8      |      |              |         |
| 1         | 0.16 | 0.12–0.21    |         |
| 2         | 0.47 | 0.40–0.54    |         |
| 3         | 0.39 | 0.21–0.57    |         |
| 4         | 0.61 | 0.56–0.66    | 0.0082  |
| 5         | 0.07 | 0.06–0.08    |         |
| 6         | 0.26 | 0.18–0.34    |         |

Abbreviations: CI = confidence interval; IL-8 = interleukin-8; MMP-1 = matrix metalloproteinase-1; OR = odds ratio; PICF = peri-implant crevicular fluid; SPP = self-perceived pain; TNF-α = tumor necrosis factor-alpha.

4. Discussion

It is postulated that SPP, peri-implant clinical, and radiographic parameters are worse among cigarette smokers with peri-implantitis than never smokers without peri-implantitis. In this study, the clinic-radiographic peri-implant parameters were poorer among cigarette smokers with peri-implantitis than never smokers without peri-implantitis; no statistically significant correlation was observed between SPP and expression of TNF-α, MMP-1, and IL-8. However, several variables might have altered the findings, therefore, these results should be interpreted with extreme caution.

A unique ecological niche is formed by the gingival sulcus for bacterial colonization. The bacterial biofilm in the gingival sulcus promotes inflammation in the surrounding connective tissues [32]. An inflammatory process is triggered by the imbalance between host response and microbial challenge at the implant–soft tissue interface [33]. Cytokines, including IL-6, IL-1β, TNF-α, are released from the cells of neutrophils, macrophages, connective tissue fibroblasts, dendritic cells, and gingival epithelium. Moreover, a number of enzymes, including aspartate aminotransferase, alkaline phosphatase, and MMPs, are formed by osteoclasts, fibroblasts, and neutrophils, resulting in the degradation of connective tissue collagen and alveolar bone [34,35]. Over 90 different molecular components in gingival crevicular fluid have been assessed for potential periodontal disease diagnosis linked with the natural dentition [36]. However, considerably fewer PICF components have been investigated around dental implants.

In the current study, certain disparities were also observed. For example, clinical peri-implant examination revealed that PI and BOP were significantly raised in never smokers with peri-implantitis than smokers with peri-implantitis and the control group. Moreover, in cigarette smokers, BOP is masked as nicotine causes the vasoconstriction of gingival blood vessels [37]. From a clinical perspective, the findings of this study conform
to the proposed hypothesis as scores of peri-implant PD are significantly elevated among cigarette smokers than never smokers. One explanation for this finding is that cell death and formation of matrix metalloproteinases are induced and enhanced by cigarette smoke extracts, respectively, leading to the deterioration of the extracellular matrix proteins such as collagen [38]. Furthermore, the expression for RAGE is increased four times by nicotine in human gingival cells [39]; an increased AGE/RAGE interaction aggravates periodontal tissue inflammation [39].

In the current study, smokers and never smokers with peri-implantitis did not show a significant difference in CBL. Among all three groups, the subjects had a mean age between 38 and 42 years. It is well-established that advancing or increasing age is a predisposing factor for the increased loss of alveolar bone around dental implants and natural dentition [14]. In one study, Javed et al. [25] reported that alveolar bone loss around teeth was significantly less among younger non-smokers (≤45 years of age) than old non-diabetic smokers (≥65 years of age). Furthermore, subjects in group-1 (smokers with peri-implantitis) had a cigarette smoking history of one cigarette per day over the past two years. It is possible that participants with such a brief history of tobacco smoking may exhibit better radiographic bone levels when compared with subjects with a prolonged history of cigarette smoking, (i.e., >15 packs per year) [14]. However, further research is required for testing these hypotheses.

The current study reports that TNF-α, MMP-1, and IL-8 levels were significantly raised among smokers and never smokers with peri-implantitis than never smokers without peri-implantitis. TNF-α, MMP-1, and IL-8 appear to have a crucial role in the destruction of the peri-implant tissue. TNF-α is a proinflammatory mediator and its level reflects the bacterial amount and the levels of inflammation [40]. MMPs are considered to be involved in wound repair, normal tissue turnover, and periodontal destruction; MMP-1 may initiate the deterioration of the extracellular matrix. Overproduction of MMP-1 may cause accelerated degradation of the matrix in pathologic conditions including peri-implantitis [41]. IL-8 increase causes the activation of polymorphonuclear cells and influences their selective migration from gingival blood vessels. This leads to an increased amount of cells in a limited time and produces inflammatory conditions. Hence, an excessive quantity of IL-8 might indicate the incipient stage of peri-implantitis [42]. Several studies have reported that the secretion of TNF-α, MMP-1, and IL-8 is upregulated by nicotine in tobacco; which appears to play a vital role in the destruction of alveolar bone around dental implants and natural dentition [17]. Elevated levels of these proinflammatory cytokines have been observed in the PICF of patients with peri-implantitis [43]. Moreover, a meta-analysis reported that cigarette smoking jeopardizes bone-to-implant contact by impairing new bone formation around the dental implant [44]. Another explanation in this regard is that cigarette smoking escalates the production and accumulation of AGEs in periodontal tissues [39]. Strong interfaces between AGEs and RAGEs have been linked with the production of ROS that promotes oxidative burst within gingival tissues, functional changes of phagocytosis and chemotaxis of polymorphonuclear cells (PMNs), alleviated formation of antibodies, increased attachment of bacterial adhesion, and raised local and systemic burden by escalating the expression of cytokines in the crevicular fluid and serum [45]. These mechanisms have been associated with the inflammation of oral connective tissues and destruction of alveolar bone around dental implants and natural teeth in cigarette smokers [16].

In literature, a dearth of reports exist that assessed SPP and its association with the inflammatory cytokines’ levels discharged saliva, serum, or PICF. In this aspect, the authors hypothesized that PICF levels of TNF-α, MMP-1, IL-8 are raised in smokers with peri-implantitis with high SPP scores than non-smokers with and without peri-implantitis. The findings of the present study are in opposition with this hypothesis as no statistically significant association between SPP and PICF levels of TNF-α, MMP-1, IL-8. One justification in this regard might be that an array of factors such as coping strategies, cognition, expectations, beliefs, and interpretation influence pain evaluation.
Furthermore, from these findings, it is not possible to calculate the minimum amount of proinflammatory cytokines required to evoke SPP in individuals with peri-implantitis. Despite a well-documented association between pain conditions and cytokines \[46,47\], the association between pain perception and cytokine expression in oral fluids remains unclear and poorly understood. Hence, further research is required in this regard.

Although questionnaires are not particularly reliable regarding clinical peri-implant parameters, including PI, PD, and BOP \[48\], they are valid and reliable tools for evaluation of SPP and other oral symptoms \[48\]. Moreover, validated and well-designed questionnaires could too yield valuable data in survey-based epidemiological large sample-size reports \[26\]. In the current study, NPRS was utilized to assess the SPP following the recommendations by Downie and co-workers \[5\] almost forty years ago, who reported that the NPRS (0–10) was a better method to assess SPP than other scales including the four-point descriptive and the visual analogue scales. Because NPRS was the only utilized scale for assessing the SPP, it is difficult to either contradict or support the Downie study \[5\]. Based on the present study’s findings, the NPRS appears to be an efficient scale for pain evaluation in periodontology and implant dentistry. However, future studies utilizing several pain evaluation scales are required to identify the most efficient and suitable pain scale that can be employed on individuals with peri-implant diseases. It is important to recognize the limitations of this study. First, the cross-sectional study and self-reported results that depend on the subject’s recall capacities might have influenced the outcomes of our findings. The assessment of pain biomarker(s) would have helped to evaluate the pain parameter on the molecular level. Second, strict criteria were adopted regarding patient selection. For example, patients with endocrine disorders and tobacco-product consumers were excluded. As habitually consuming tobacco products is a predisposing factor of peri-implant diseases \[49\], it was speculated that the levels of PICF proinflammatory cytokines and the severity of SPP are less in never smokers than smokers with peri-implantitis. The primary strength of this study is the assessment of local proinflammatory cytokines. In terms of accuracy, the use of bitewing radiographs in this study provided better-standardized measurements. Future research studies using several pain evaluation scales and biomarkers related to pain and dental implant therapy are required to identify the most efficient and suitable diagnostic criterion that could be employed on individuals with peri-implant diseases. In addition, longitudinal reports might help to evaluate the PICF biomarkers’ evolution over time for better understanding in terms of peri-implant infections among smokers and non-smokers.

5. Conclusions

Within the limitations of the present study, proinflammatory biomarkers were higher in smokers with peri-implantitis than never smokers with and without peri-implantitis, with a significant association between the proinflammatory cytokines and self-perceived pain.

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References

1. Nugraha, B.; Gutenbrunner, C.; Barke, A.; Karst, M.; Schiller, J.; Schäfer, P.; Falter, S.; Korwisi, B.; Rief, W.; Treede, R.D.; et al. The IASP classification of chronic pain for ICD-11: Functioning properties of chronic pain. Pain 2019, 160, 88–94. [CrossRef] [PubMed]
2. Coli, P.; Christaens, V.; Semnerby, L.; Bruyn, H.D. Reliability of periodontal diagnostic tools for monitoring peri-implant health and disease. Periodontology 2000 2017, 73, 203–217. [CrossRef]
3. Ramanaukeite, A.; Juodzbalys, G. Diagnostic principles of peri-implantitis: A systematic review and guidelines for peri-implantitis diagnosis proposal. J. Oral Maxillofac. Res. 2016, 7, e8. [CrossRef]
4. Misch, C.E.; Perel, M.L.; Wang, H.L.; Sammartino, G.; Galindo-Moreno, P.; Trisi, P.; Steigmann, M.; Rebaudi, A.; Palti, A.; Pikos, M.A.; et al. Implant success, survival, and failure: The International Congress of Oral Implantologists (ICOI) pisa consensus conference. Implant Dent. 2008, 17, 5–15. [CrossRef]
5. Downie, W.; Leatham, P.; Rhind, V.; Wright, V.; Branco, J.; Anderson, J. Studies with pain rating scales. Ann. Rheum. Dis. 1978, 37, 378–381. [CrossRef]
6. Saltychev, M.; Bärlund, E.; Laimi, K. Correlation between the pain numeric rating scale and the 12-item WHO Disability Assessment Schedule 2.0 in patients with musculoskeletal pain. Int. J. Rehabil. Res. 2018, 41, 87–91. [CrossRef]
7. Iero, P.T.; Mulherin, D.R.; Jensen, O.; Berry, T.; Danesi, H.; Razook, S.J. A prospective, randomized, open-Label study comparing Core-Vent dental implants. J. Oral Maxillofac. Implant. 2018, 33, 1155–1164. [CrossRef] [PubMed]
8. Scaranò, A.; Murmura, G.; Sinijari, B.; Sollazzo, V.; Spinelli, G.; Carinci, F. Analysis and structural examination of screw loosening in oral implants. Int. J. Immunopathol. Pharmacol. 2011, 24, 77–81. [CrossRef] [PubMed]
9. Assenza, B.; Scaranò, A.; Leghissa, G.; Carusi, G.; Thams, U.; Roman, F.S.; Piattelli, A. Screw-vs Cement-implant–retained Restorations: An Experimental Study in the Beagle. Part I. Screw and Abutment Loosening. J. Oral Implantol. 2005, 31, 242–246. [CrossRef]
10. Jent, T.; Laney, W.R.; Harris, D.; Henry, P.J.; Krogh, P.H., Jr.; Polizzi, G.; Zarb, G.A.; Herrmann, I. Osseointegrated implants for single tooth replacement: A 1-year report from a multicenter prospective study. Int. J. Oral Maxillofac. Implant. 1991, 6, 29–36.
11. Al-Aali, K.A.; ArRejaie, A.S.; Alrahlah, A.; Al-Fawaz, Y.F.; Abduljabbar, T.; Vohra, F. Clinical and radiographic peri-implant health status around narrow diameter implant-supported single and splinted crowns. Clin. Implant Dent. Relat. Res. 2019, 21, 386–390. [CrossRef]
12. Cranin, A.N.; Dbling, J.B.; Simons, A.; Klein, M.; Sirakian, A. Report of the incidence of implant insert fracture and repair of Core-Vent dental implants. J. Oral Implantol. 1990, 16, 184–188.
13. Dixon, D.L.; Breeding, L.C.; Sadler, J.P.; McKay, M.L. Comparison of screw loosening, rotation, and deflection among three implant designs. J. Prosthet. Dent. 1995, 74, 270–278. [CrossRef]
14. Alasqah, M.N.; Al-Fawaz, Y.F.; Al-Hamoudi, N.; Vohra, F.; Alotaibi, B.M.; Abduljabbar, T. Longitudinal assessment of clinical and radiographic peri-implant status around narrow and regular diameter implants placed in cigarette-smokers and nonsmokers. Clin. Implant Dent. Relat. Res. 2019, 21, 910–915. [CrossRef]
15. Veitz-Keenan, A. Marginal bone loss and dental implant failure may be increased in smokers. Evid. Based Dent. 2016, 17, 6–7. [CrossRef] [PubMed]
16. Akram, Z.; Alqahtani, F.; Alqahtani, M.; Al-Kheraif, A.A.; Javed, F. Levels of advanced glycation end products in gingival crevicular fluid of chronic periodontitis patients with and without type-2 diabetes mellitus. J. Periodontol. 2020, 91, 396–402. [CrossRef] [PubMed]
17. Yu, S.; Li, H.; Ma, Y.; Fu, Y. Matrix metalloproteinase-1 of gingival fibroblasts influenced by advanced glycation end products (AGEs) and their association with receptor for AGEs and nuclear factor-κB in gingival connective tissue. J. Periodontol. 2012, 83, 119–126. [CrossRef] [PubMed]
18. Zhang, W.; Fang, M.; Song, F.; Windsor, L.J. Effects of cigarette smoke condensate and nicotine on human gingival fibroblast-mediated collagen degradation. J. Periodontol. 2011, 82, 1071–1079. [CrossRef]
19. Al-Aali, K.A.; Al-Helal, A.; Alhamoudi, N.; Al-Henaki, A.M.; Javed, F.; Abduljabbar, T. Assessment of advanced glycation end products in the peri-implant sulcular fluid among moderate cigarette-smokers and nonsmokers with peri-implantitis. Clin. Implant Dent. Relat. Res. 2020, 22, 1–7. [CrossRef] [PubMed]
20. Alqahtani, F.; Alqahtani, M.; Albaqawi, A.H.; Al-Kheraif, A.A.; Javed, F. Comparison of cotinine levels in the peri-implant sulcular fluid among cigarette and waterpipe smokers, electronic-cigarette users, and nonsmokers. Clin. Implant Dent. Relat. Res. 2019, 21, 702–707. [CrossRef] [PubMed]
21. Holliday, R.S.; Campbell, J.; Preshaw, P.M. Effect of nicotine on human gingival, periodontal ligament and oral epithelial cells. A systematic review of the literature. J. Dent. 2019, 86, 81–88. [CrossRef]

22. Alamri, A.; Semlali, A.; Jacques, E.; Alanazi, M.; Zakrzewski, A.; Chmielewski, W.; Rouabhia, M. Long-term exposure of human gingival fibroblasts to cigarette smoke condensate reduces cell growth by modulating Bax, caspase-3 and p53 expression. J. Periodontal Res. 2015, 50, 423–433. [CrossRef] [PubMed]

23. DeLuca, S.; Habsa, E.; Zarb, G.A. The effect of smoking on osseointegrated dental implants. Part I: Implant survival. Int. J. Prosthodont. 2006, 19, 491–498.

24. Abduljabbar, T.; Vohra, F.; Ullah, A.; Alhamoudi, N.; Khan, J.; Javed, F. Relationship between self-rated pain and peri-implant clinical, radiographic and whole salivary inflammatory markers among patients with and without peri-implantitis. Clin. Implant Dent. Relat. Res. 2019, 21, 1218–1224. [CrossRef]

25. Alsahhaf, A.; Alshagroud, R.S.; Al-Aali, K.A.; Alofi, R.S.; Vohra, F.; Abduljabbar, T. Survival of titanium-zirconium and titanium dental implants in cigarette-smokers and never-smokers: A 5-year follow-up. Chin. J. Dent. Res. 2019, 22, 265–272. [PubMed]

26. Vohra, F.; Bukhari, I.A.; Sheikh, S.A.; Albaian, R.; Naseem, M. Comparison of self-rated oral symptoms and periodontal status among cigarette smokers and individuals using electronic nicotine delivery systems. J. Am. Coll. Health 2020, 68, 788–793. [CrossRef]

27. Jensen, M.P.; McFarland, C.A. Increasing the reliability and validity of pain intensity measurement in chronic pain patients. Pain 1993, 55, 195–203. [CrossRef]

28. Clark, D.; Levin, L. Dental implant management and maintenance: How to improve long-term implant success. Quintessence Int. 2016, 47, 417–423.

29. Lang, N.P.; Berglundh, T.; Working Group 4 of Seventh European Workshop on Periodontology. Periimplant diseases: Where are we now?—Consensus of the Seventh European Workshop on Periodontology. J. Clin. Periodontol. 2011, 38, 78–811. [CrossRef]

30. Khocht, A.; Janal, M.; Harasty, L.; Chang, K.M. Comparison of direct digital and conventional intraoral radiographs in detecting alveolar bone loss. J. Am. Dent. Assoc. 2003, 134, 1468–1475. [CrossRef]

31. Alasqah, M.N.; Al-Shibani, N.; Al-Aali, K.A.; Qutub, O.A.; Abduljabbar, T.; Akram, Z. Clinical indices and local levels of inflammatory biomarkers in peri-implant health of obese and nonobese individuals. Clin. Implant Dent. Relat. Res. 2019, 21, 80–84. [CrossRef]

32. Costalonga, M.; Herzberg, M.C. The oral microbiome and the immunobiology of periodontal disease and caries. Immunol. Lett. 2014, 162, 22–38. [CrossRef] [PubMed]

33. Wang, H.L.; Garaioco-Pazmino, C.; Collins, A.; Ong, H.S.; Chudri, R.; Giannobile, W.V. Protein biomarkers and microbial profiles in peri-implantitis. Clin. Oral Implant. Res. 2016, 27, 1129–1136. [CrossRef]

34. Barros, S.P.; Williams, R.; Offenbacher, S.; Morelli, T. Gingival crevicular fluid as a source of biomarkers for periodontitis. Periodontology 2000 2016, 70, 53–64. [CrossRef] [PubMed]

35. Paknejad, M.; Emtiaz, S.; Khoobyari, M.M.; Gharb, M.T.; Yazdi, M.T. Analysis of aspartate aminotransferase and alkaline phosphatase in crevicular fluid from implants with and without peri-implantitis. J. Periodontal Res. 2015, 50, 1383–1389. [CrossRef] [PubMed]

36. Lang, N.P.; Berglundh, T.; Working Group 4 of Seventh European Workshop on Periodontology. Periimplant diseases: Where are we now?—Consensus of the Seventh European Workshop on Periodontology. J. Clin. Periodontol. 2011, 38, 78–811. [CrossRef]

37. Khocht, A.; Janal, M.; Harasty, L.; Chang, K.M. Comparison of direct digital and conventional intraoral radiographs in detecting alveolar bone loss. J. Am. Dent. Assoc. 2003, 134, 1468–1475. [CrossRef]

38. Alasqah, M.N.; Al-Shibani, N.; Al-Aali, K.A.; Qutub, O.A.; Abduljabbar, T.; Akram, Z. Clinical indices and local levels of inflammatory biomarkers in peri-implant health of obese and nonobese individuals. Clin. Implant Dent. Relat. Res. 2019, 21, 80–84. [CrossRef]

39. Costalonga, M.; Herzberg, M.C. The oral microbiome and the immunobiology of periodontal disease and caries. Immunol. Lett. 2014, 162, 22–38. [CrossRef] [PubMed]

40. Barros, S.P.; Williams, R.; Offenbacher, S.; Morelli, T. Gingival crevicular fluid as a source of biomarkers for periodontitis. Periodontology 2000 2016, 70, 53–64. [CrossRef] [PubMed]

41. Paknejad, M.; Emtiaz, S.; Khoobyari, M.M.; Gharb, M.T.; Yazdi, M.T. Analysis of aspartate aminotransferase and alkaline phosphatase in crevicular fluid from implants with and without peri-implantitis. J. Periodontal Res. 2015, 50, 1383–1389. [CrossRef] [PubMed]

42. Lang, N.P.; Berglundh, T.; Working Group 4 of Seventh European Workshop on Periodontology. Periimplant diseases: Where are we now?—Consensus of the Seventh European Workshop on Periodontology. J. Clin. Periodontol. 2011, 38, 78–811. [CrossRef]

43. Khocht, A.; Janal, M.; Harasty, L.; Chang, K.M. Comparison of direct digital and conventional intraoral radiographs in detecting alveolar bone loss. J. Am. Dent. Assoc. 2003, 134, 1468–1475. [CrossRef]

44. Alasqah, M.N.; Al-Shibani, N.; Al-Aali, K.A.; Qutub, O.A.; Abduljabbar, T.; Akram, Z. Clinical indices and local levels of inflammatory biomarkers in peri-implant health of obese and nonobese individuals. Clin. Implant Dent. Relat. Res. 2019, 21, 80–84. [CrossRef]

45. Costalonga, M.; Herzberg, M.C. The oral microbiome and the immunobiology of periodontal disease and caries. Immunol. Lett. 2014, 162, 22–38. [CrossRef] [PubMed]
46. Khan, J.; Hassun, H.; Zusman, T.; Korczeniewska, O.; Eliav, E. Interleukin-8 levels in rat models of nerve damage and neuropathic pain. *Neurosci. Lett.* 2017, 657, 106–112. [CrossRef] Clin. Implant Dent. Relat. Res.

47. Khan, J.; Ramadan, K.; Korczeniewska, O.; Anwer, M.M.; Benoliel, R.; Eliav, E. Interleukin-10 levels in rat models of nerve damage and neuropathic pain. *Neurosci. Lett.* 2015, 59, 99–106. [CrossRef]

48. Buhlin, K.; Gustafsson, A.; Andersson, K.; Håkansson, J.; Klinge, B. Validity and limitations of self-reported periodontal health. *Community Dent. Oral Epidemiol.* 2002, 30, 431–437. [CrossRef]

49. Monje, A.; Pons, R.; Insua, A.; Nart, J.; Wang, H.L.; Schwarz, F. Morphology and severity of peri-implantitis bone defects. *Clin. Implant Dent. Relat. Res.* 2019, 21, 635–643. [CrossRef] [PubMed]