

Original Research Article

Efficacy of injectable platelet-rich fibrin in reconstruction of interdental papilla: a case series

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Abstract – Objective: To evaluate the effectiveness of I-PRF in the reconstruction of recessed interdental papillae after three injections spaced two weeks apart. **Materials and methods:** The study included 8 participants aged 20–35 years. Periapical radiographs were taken and entered into ImageJ software to measure the distance between the apex of the alveolar process and the contact area. Cases were included when the distance was 5 mm or less. The following indices and parameters were evaluated: plaque index (PI), gingival inflammation index (GI), gingival bleeding index (GB), height of papillary deficiency (HPD), measure of change in the height of papillary deficiency (MHPD), and percentage of change in the height of papillary deficiency (PHPD). Autologous PRF was obtained and immediately centrifuged. The I-PRF was injected at the interdental papilla base. Patients were followed up three months post-treatment. **Results:** The mean papillary dimensions after three months (5.43 ± 0.99) were lower than the baseline mean (5.55 ± 1.05), with no significant difference ($P = 0.171$). No significant differences were observed between genders in the parameters ($P > 0.05$): HPD, MHPD, and PHPD. Although the mean HPD in males after three months (5.88 ± 0.83) was lower than at baseline (6.00 ± 0.76), and a similar trend was observed in females after three months (5.13 ± 1.00) compared to baseline (5.25 ± 1.14), independent t-tests revealed no significant differences between genders. Improvements in PI and GI were noted after three months. **Conclusion:** Within the limits of this study, the I-PRF injection technique was ineffective in managing recessed interdental papillae three months post-treatment.

Introduction

The interdental papilla is an essential component of an attractive smile. Recession of the papilla leads to black triangles, which may compromise a perfect smile. A black triangle forms when the embrasure apical to the proximal contact point is unfilled by the interdental papilla. The presence of these embrasures affects phonetics and leads to food impaction, which has negative effects on periodontal health [1–3].

Black triangles more frequently appear in adult patients with bone loss (over 30% of adults) [4]. The prevalence of black triangles is 18% among individuals under the age of 20 and it was 67% among those above the age of 20 [5].

One of the etiological factors affecting papilla deficiency is the distance between the interproximal contact position and the bone crest. Therefore, when the distance was less than 5 mm, the interdental papillary filling was 100%

complete. However, when the distance was to 6 mm, the interdental papillary filling was 56%, and it was only 27% when the distance was more than 7 mm [6].

Black triangles can also result from traumatic brushing and improper use of interdental floss [7,8]. Older age, periodontal diseases, and interproximal space are also considered etiological factors of open gingival embrasures [8–10].

Previous studies suggested surgical and non-surgical techniques to reconstruct lost soft tissue and manage interdental space. Non-surgical methods include orthodontic, prosthodontic, and restorative procedures. Surgical methods aim to remodel, maintain, or reconstruct the interdental soft tissue [11].

Restorative or prosthetic approaches can correct open gingival embrasures by lengthening the contact point apically to the tip of the papilla to close the space. These approaches require the tooth to have an abnormal shape and have disadvantages like food impaction and increased bacterial growth [2].

Orthodontic approaches can also correct black triangles by changing root inclination to place the contact point more apically. This approach is applied in cases of divergent roots. Some cases of diastema need orthodontic approaches followed by papilla regeneration to close the gap [2].

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Repeated curettage every 15 days for 3 months can recreate the papilla that has been destroyed by necrotizing gingivitis. Some papillae present complete regeneration; however, not all cases respond to periodic curettage [12]. Therefore, the response to this treatment cannot be predicted.

Several surgical techniques have been introduced to reconstruct the interdental papilla, such as pedicle flap, envelope-type flap, semilunar coronally repositioned flap, and connective tissue graft. However, the majority of these surgical techniques have shown limited success, largely attributed to the minor blood supply in the interdental papilla [2,13].

Platelet-rich fibrin (PRF) is widely utilized in dentistry to expedite the healing process of soft and bone tissues. It is differentiated from the first generation (platelet-rich plasma) by its ease of preparation, application, and prolonged release of growth factors, in addition to its cost-effectiveness [14].

Platelets are the main cells responsible for the biological activity of PRF. They contain alpha, dense, and glycolytic granules. Alpha granules contribute to the wound healing process through the presence of the following growth factors: platelet-derived growth factor, transforming growth factor β , epidermal growth factor, endothelial vascular growth factor, and insulin-like growth factor [15].

Injectable platelet-rich fibrin (I-PRF) is the injectable form of platelet-rich fibrin, offering greater ease of use compared to the gel form [16]. It is noteworthy that while I-PRF has recently found applications in various fields beyond dentistry, however its effects on deficient interdental papillae have not widely been evaluated.

Therefore, this clinical study aimed to evaluate the effectiveness of the I-PRF technique in reconstructing recessed interdental papillae following three injections. The null hypothesis states that there are no statistically significant differences in the studied variables before and after applying the I-PRF technique. The alternative hypothesis states that there are statistically significant differences in the studied variables before and after applying the I-PRF technique.

Materials and methods

Study design

This study included 8 participants who had 20 recessed interdental papillae classified as Class II according to the *Papilla Presence Index* (PPI) [17]. PPI class 2 indicates the papilla is apically positioned to the contact point and does not completely fill the gingival embrasure, nor is it at the same level as the adjacent papillae; however, the cemento-enamel junction is still not visible. Recessed interdental papillae located in the frontal esthetic zone (between upper and lower canines).

Sample size determination

According to Singh *et al.*'s study the 1.3% effect size was adopted [18]. Using G*Power Version 3.1.9.2 (Franz Faul, Universität Kiel, Germany), and considering 5% margin of

error, 95% power, the minimum sample size required will be 12 sites.

Ethical approval

The study was performed in accordance with the Declaration of Helsinki. Ethical approval was obtained from the Ethics Committee and the Board of Scientific Research at the Faculty of Dentistry at Damascus University, and the Ministry of Higher Education and Research (No.3480, date: 4-9-2018). Additionally, official written permissions were obtained from all participants to participate in the study.

Each participant received a form in which the materials used, treatment plan, and complete procedures were explained.

The study procedures and follow-up occurred in the Periodontology Department and the Maxillofacial Surgery Hospital, and were carried out from September 2018 to August 2019.

Participants were selected from patients who attended the Periodontology Department, Faculty of Dentistry at Damascus University, Damascus, Syria.

Inclusion criteria

- Patients aged 20–35 years.
- Presence of at least one papilla meeting the following criteria; papilla deficiency classified as class II according to PPI [17], presence of contact area between two adjacent teeth, adjacent teeth free of caries, restorations, and prosthodontics, and the space between the apex of the alveolar process and the contact area was less than or equal to 5 mm.

Exclusion criteria

- Diagnosed chronic diseases such as diabetes, heart diseases, bleeding disorders, and blood pressure disorders.
- Vitamin C deficiency.
- Poor oral hygiene.
- Traumatic occlusion on adjacent teeth.
- Periodontal diseases.
- Orthodontic treatment.
- Smokers and alcoholics.
- Pregnancy and breastfeeding.
- Any condition affecting procedures and/or I-PRF preparation, and/or healing.

During the selecting stage, clinical and radiographic examinations were conducted, and data was recorded using the examination form. Each participant had two examination forms: one for pre-treatment examinations and another for post-treatment examinations.

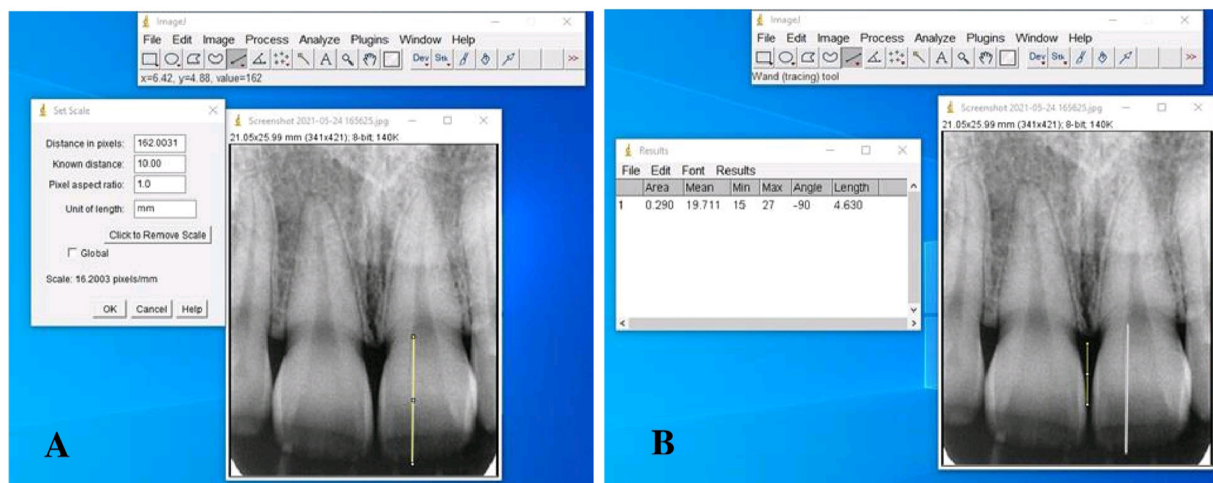


Fig. 1. Periapical digital radiographs. (A) Calibrate the radiograph dimensions by a 10 mm metal wire. (B) Measure the distance between the apex of the alveolar process and the contact area.

The following parameters were investigated

- **Height of Papillary Deficiency (HPD):** This refers to the distance between the tip of the interdental papilla and the base of the contact area (the vacuum's reference point). It was measured using UNC15 probe and estimated in millimeters (mm).
- **Measure of Change in the Height of Papillary Deficiency (MHPD):** This was estimated in mm and calculated using the formula:

$$\text{MHPD} = \text{Post - treatment HPD} - \text{Pre - treatment HPD.}$$

- **Percentage of Change in the Height of Papillary Deficiency (PHPD):** This represents the percentage change in distance and was calculated using the formula:

$$\text{PHPD} = \text{MHPD} \div \text{Pre - treatment HPD} \times 100.$$

Radiographic examination

The distance between the apex of the alveolar process and the contact area was measured using periapical digital radiographs taking with a parallel technique. Before capturing the radiograph, a 10 mm metal wire was temporarily affixed parallel to the buccal surface of the tooth adjacent to the target papilla. This metal wire served as a calibration tool for the radiograph dimensions and facilitated measurement of the required distance using ImageJ Software (Fig. 1).

Clinical examination

The following periodontal indices were also recorded to assess oral hygiene level and periodontal status:

Plaque Index (PI) and Gingival Inflammation Index (GI) according to (Silness and L oe) [19], Gingival Bleeding Index (GB) [20], and interdental papilla index. These indices were evaluated twice: first at baseline (pre-treatment), and then in a follow-up after 3 months.

Preparation stage

This stage involved manual scaling using U15 and CK6 tools, along with educating the patient about oral hygiene care. Subsequently, a vacuum plate was custom-manufactured for the target jaw using silicon impression materials (Fig. 2). The vacuum plate served to establish a reference point for subsequent measurements.

The space between the papilla tip and the vacuum plate's reference point was measured by UNC15 probe (Fig. 3).

Preparing I-PRF

An autologous PRF was obtained by drawing 10 ml of blood, then placing it in a dry, sterile, and anticoagulant-free tube. The sample was immediately centrifuged using centrifuge device (centrifugal) with speed of 700 rounds per minute for three minutes (Fig. 4). The material was applied directly to retain maximum regeneration potential [16].

Injection stage

Local anesthetic (2% lidocaine with 1:80,000 adrenaline) was applied by infiltration to the target area. Then I-PRF was injected using a 27-gauge needle's tip at the base of the interdental papilla with depth of 2 mm from the papilla's surface, (Fig. 5). Choukroun and Miron technique was adopted in terms of the intervals and frequency of injections [16].

The injection was repeated with the same steps mentioned previously after 2 weeks then after 4 weeks (Fig. 6).

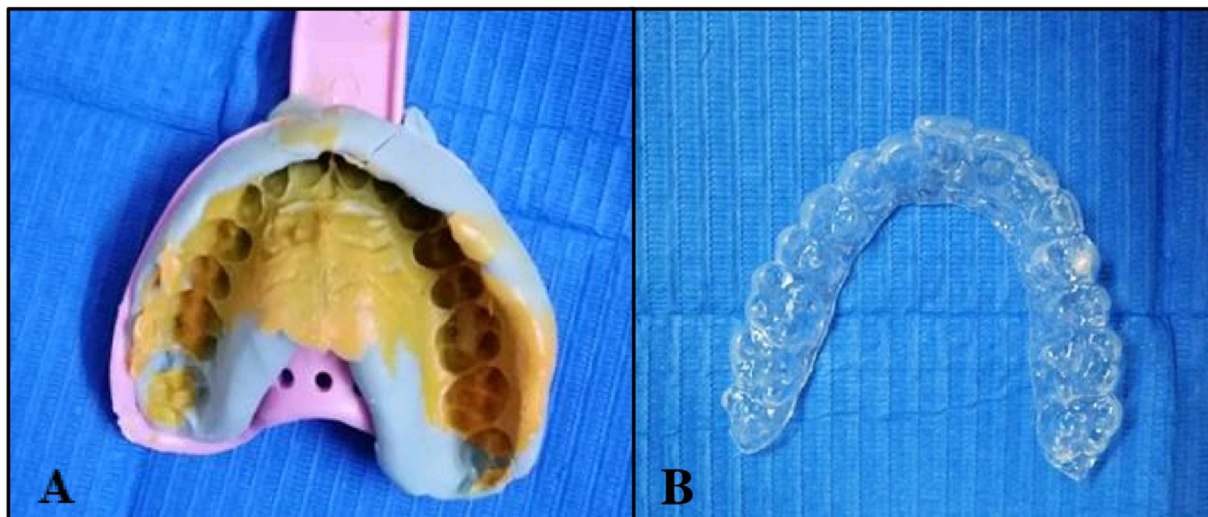


Fig. 2. Manufacturing a vacuum plate. (A) The silicon impression. (B) The vacuum plate.



Fig. 3. Using the vacuum plate to determine a reference point for measuring the distance between the papilla tip and the vacuum plate's reference point by UNC15 probe.

Post-procedures instructions

After performing the injection, the following instructions were given: avoid eating harsh food on the injection area. Refrain from mechanical actions on the day of the injection to control dental plaque, which means replacing brushing and flossing with rinsing using 0.12% Chlorhexidine twice per day. Use the toothbrush after the first 24 hours of injection, and brush using the Roll Technique.

Follow-up stage

Cases were followed up three months after the last injection. The measurements and examinations at all stages were conducted by a single examiner.

Statistical analysis

Data were collected and entered into Microsoft Office Excel 2016, then analyzed using IBM SPSS Statistics version 26. Independent *t*-test, Paired *t*-test, and Wilcoxon tests were implemented. A *p*-value of <0.05 was considered significant at a 95% confidence level.

Results

The study sample contained 20 recessed interdental papillae among 8 participants (3 males and 5 females) aged from 20 to 35 years, with a mean age of 25.6 ± 4.8 .

The mean of the measurement of papillary dimension after 3 months was lower than the pre-treatment mean. However, there were no significant differences between the two means (Tab. I).

When comparing between genders, there were no significant differences between the pre-treatment and after 3 months' means (Tab. II).

Descriptive results of the measure of change in the papillary dimensions, and the percentage change in the measure of papillary dimensions for the study sample represented in (Tab. III).

Results of the measure of change in the papillary dimensions, and the percentage change of the papillary dimensions according to gender showed no significant differences (Tab. IV).

Results of evaluated plaque index (PI), and gingival inflammation index (GI) pre- and post-3 months are represented in (Figs. 7 and 8), respectively.

PI and GI improved after 3 months compared to pre-treatment assessments with statistically significant differences (Tab. V).

The percentage change in the gingival bleeding index rate for the study sample showed no significant differences between pre-treatment and after 3 months (Tab. VI).

Discussion

Loss or deficiency of the interdental papilla is a concern for both the dentist and the patient due to the various resulting problems, including aesthetic issues.

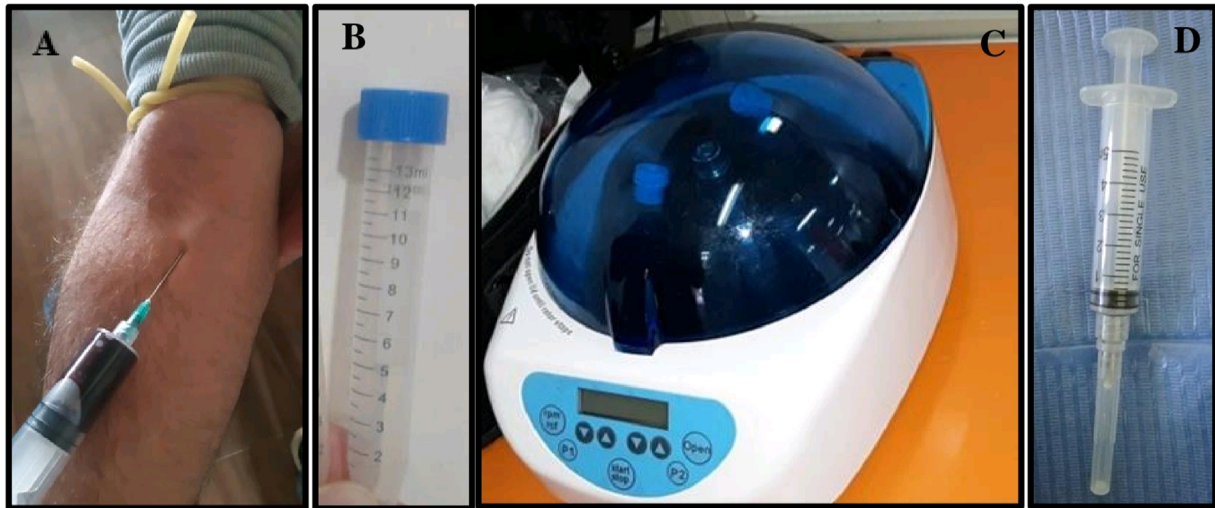


Fig. 4. Preparing I-PRF. (A) Blood drawing. (B) The dry, sterile, and anticoagulant-free tube; (C) Centrifugal (Hettich®, Germany, EBA20). (D) The 27-gauge needle's tip for extracting I-PRF and using it in injection stage.

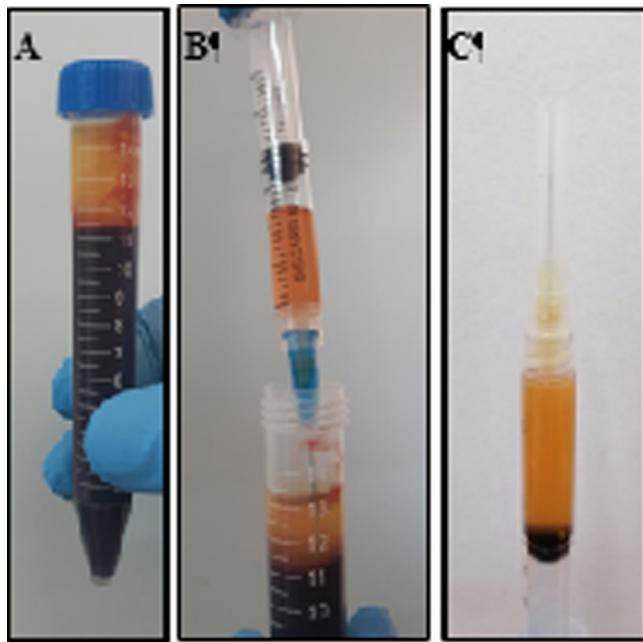


Fig. 5. Injection stage. (A) An image from the study sample showing sections of I-PRF. (B) Extracting the I-PRF from the tube. (C) The I-PRF is ready for injection.

The use of platelet-rich fibrin for papilla reconstruction has been reported in several studies [18,21]. However, most medical applications require platelet substrates in liquid form rather than coagulated form. Therefore, this study focused on PRF in its injectable form, which is rich in white blood cells in addition to platelets, enhancing healing properties while adhering to the principle of centrifugation.

PRF is an endogenous biomaterial derived from blood after brief centrifugation at low speed. With advancements in the concept of low-speed centrifugation, I-PRF has been developed, which has a higher concentration of leukocytes and

growth factors, thereby possessing a greater capacity for wound healing and regeneration [16].

Since I-PRF is autogenous, it reduces adverse immune reactions compared to other types of grafting materials [22]. Moreover, I-PRF surpasses avascular materials currently used in tissue engineering, such as hyaluronic acid and xenografts, which are incapable of providing the necessary vascular supply for fully successful regeneration of either soft or hard tissue [23].

In addition, growth factors are essential for promoting osteogenesis and neovascularization by recruiting mesenchymal stem cells and stimulating cell differentiation. Growth factors previously found in I-PRF are VEGF, PDGF, and TGF-β1, which can directly stimulate new bone formation or indirectly promote bone healing [24,25]. It should also be noted that obtaining I-PRF requires using tubes that are free of anticoagulants, and it turns into a fibrin clot several minutes after the injection.

This study evaluated the effectiveness of I-PRF on recessed interdental papillae (class II according to Cardaropoli *et al.* [17]) in the esthetic area. Previous studies have evaluated the effect of injecting other substances into the papilla, such as hyaluronic acid [26], or combining the use of mesenchymal stem cells with PRP and hyaluronic acid as a scaffold [27].

The ages of the participants ranged between 20 and 35 years in order to exclude the effects of aging in the recession of the papillae. Additionally, patients with chronic diseases were excluded due to their potential effects on gingival position. The study also focused on patients with good oral hygiene to exclude the negative effects of poor oral hygiene on tissue healing [28]. Moreover, poor oral hygiene may also lead to periodontitis, which in turn causes papillary deficiency.

A contact point between adjacent teeth was required to make it possible for interdental papilla to reconstruct, according to the study of Tarnow *et al.* [6]. Traumatic occlusion is a significant factor in periodontitis etiology, in

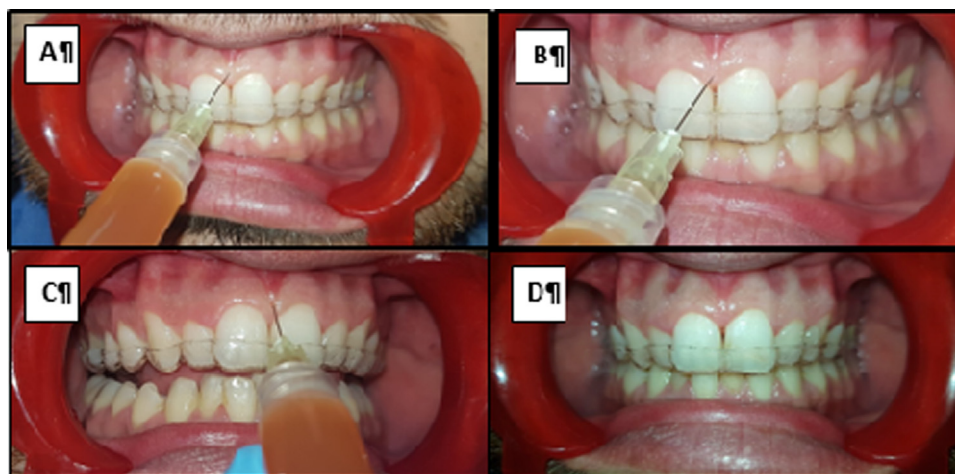


Fig. 6. Injection stage. (A) During the first injection. (B) During the second injection. (C) During the third injection. (D) After three months.

Table I. Means, standard deviation (SD), and dependent samples *t*-test for the measure of papillary dimensions in mm.

| Variable | | Pre- treatment | After 3 months | Paired <i>t</i> -test <i>P</i> value |
|----------|----------|----------------|----------------|--------------------------------------|
| HPD (mm) | <i>n</i> | 20 | 20 | 0.171 |
| | mean | 5.55 | 5.43 | |
| | SD | 1.05 | 0.99 | |

Table II. Means, standard deviation (SD), dependent samples *t*-test, and independent samples *t*-test for the measure of papillary dimensions in mm according to gender.

| Variable | Gender | | Pre-treatment | After 3 months | Paired <i>t</i> -test <i>P</i> value |
|---|--------|----------|---------------|----------------|--------------------------------------|
| HPD (mm) | Male | <i>n</i> | 8 | 8 | 0.351 |
| | | Mean | 6.00 | 5.88 | |
| | | SD | 0.76 | 0.83 | |
| | Female | <i>n</i> | 12 | 12 | 0.339 |
| | | mean | 5.25 | 5.13 | |
| | | SD | 1.14 | 1.00 | |
| Independent <i>t</i> -test <i>P</i> value | | | 0.120 | 0.098 | |

addition to its negative effects on gingiva and teeth. It can also increase plaque accumulation, which leads to gingivitis [29]. The effect of hormones during pregnancy and breastfeeding was excluded because of their effect on gingival tissue [30].

When studying PI and GI indices, statistically significant differences were found between scores before and after treatment. This is due to controlling oral hygiene and emphasizing care instructions during treatment sessions and follow-up.

In this study, the papillary dimensions remained unchanged three months after three sessions of I-PRF injection. This result is likely because I-PRF affects the thickness of the gingiva and not the height, as shown in Ozsagir *et al.*'s study [31]. Bissar *et al.*'s study found no statistical differences in volumetric changes at the interdental papillae between using I-PRF and subepithelial connective tissue graft; however, patients were obviously more satisfied with the I-PRF technique [32].

Table III. Descriptive statistics of measure of change in the height of papillary deficiency (MHDP) and percentage of change in the height of papillary deficiency (PHPD).

| | <i>n</i> | Mean | SD | Lowest rate | Highest rate |
|------|----------|-------|------|-------------|--------------|
| MHDP | 20 | -0.13 | 0.39 | -1.5 | 0 |
| PHPD | 20 | -1.90 | 5.91 | -21.43 | 0 |

Table IV. Differences in the measure of change in the height of papillary deficiency (MHDP) and the percentage of change in the height of papillary deficiency (PHPD) means according to gender.

| | Gender | <i>n</i> | Mean | SD | Independent <i>t</i> -test <i>P</i> value |
|------|--------|----------|-------|------|---|
| MHDP | Male | 8 | -0.13 | 0.35 | 1.000 |
| | Female | 12 | -0.13 | 0.43 | |
| PHPD | Male | 8 | -2.08 | 5.89 | 0.916 |
| | Female | 12 | -1.79 | 6.19 | |

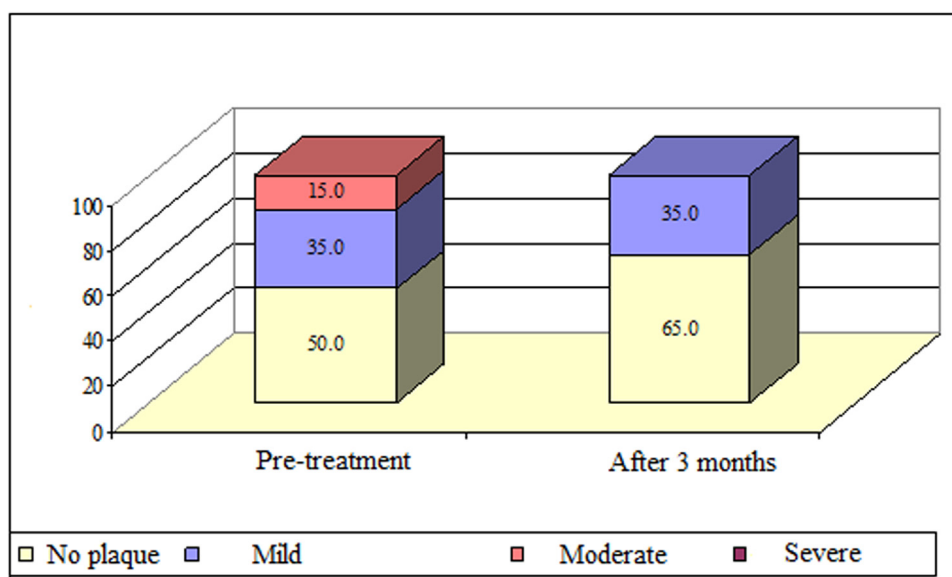


Fig. 7. The percentage of the plaque index (PI) in the study sample pre-treatment and after 3 months.

Likewise, the ability of reconstruct the papilla is limited due to poor blood supply at the apex of the papilla, as the blood supply to the papilla depends on its base, which is the main source of perfusion for the papilla [13]. A significant relationship was found between the base width of papilla and its vertical dimension (height) [33].

The results of this study differed from Awartani *et al.*'s study [26]. This is due to the overall difference between the injected materials. In the Awartani's study, hyaluronic acid was injected into the recessed papillae, and it gave positive results on the papillae height due to the filling nature of the hyaluronic acid.

This study also differed from the results of Choukroun and Miron's research [16], which injected I-PRF into the wrinkles of the forehead, face, and lips. This may be due to the difference

in the nature of the labial tissue and its richness in blood supply, unlike the gingival tissue, especially in the area of interdental papillae, which has poor blood supply. In addition, the interstitial spaces in the face and the grooves in which the injection was made differ in size compared to the very small papilla area.

No statistical differences were found between genders, these findings agreed with Chang's study which also found no statistical differences when comparing the papillary dimension between genders, regardless of the time period studied (pre-treatment and after three months) [34].

Abd El Raouf *et al.*'s study [35] evaluated the effectiveness of I-PRF compared to platelet-rich plasma (PRP) on transplanted chondrocytes and osteochondral regeneration in a large rabbit knee osteochondral defect. They found using I-PRF

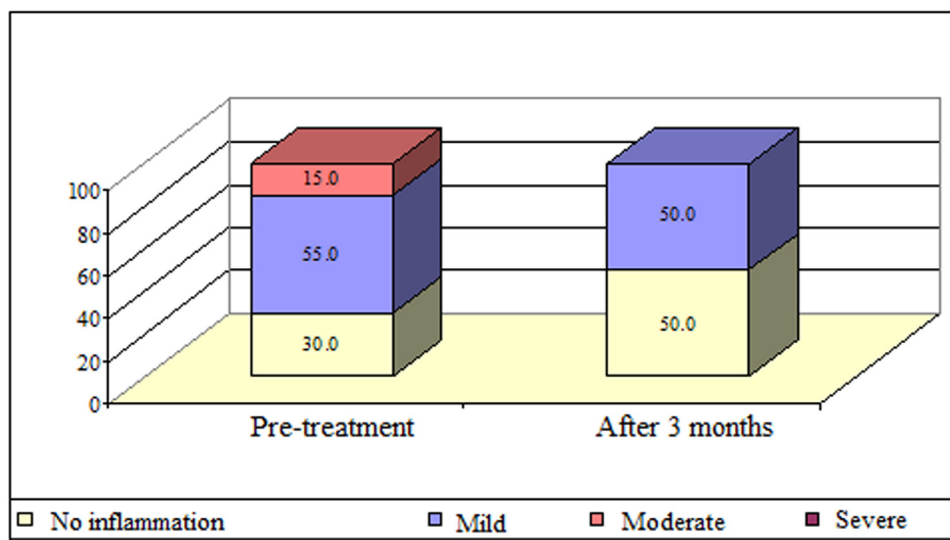


Fig. 8. The percentage of the gingival inflammation index (GI) in the study sample pre-treatment and after 3 months.

Table V. Frequency of plaque index (PI) and gingival inflammation index (GI) scores pre-treatment and after 3 months.

| Variable | | No plaque/ inflammation | Mild <i>n</i> | Moderate <i>n</i> | Severe <i>n</i> | Total | Wilcoxon <i>P</i> value |
|----------|----------------|----------------------------|------------------|----------------------|--------------------|-------|----------------------------|
| PI | Pre-treatment | 10 | 7 | 3 | 0 | 20 | *0.034 |
| | After 3 months | 13 | 7 | 0 | 0 | 20 | |
| GI | Pre-treatment | 6 | 11 | 3 | 0 | 20 | *0.020 |
| | After 3 months | 10 | 10 | 0 | 0 | 20 | |

**P* < 0.05.

Table VI. Mean and standard deviation (SD) of the change percentage in gingival bleeding index (GB) pre-treatment and after 3 months.

| Variable | | <i>n</i> | Mean | SD | Paired <i>t</i> -test |
|-------------------------|----------------|----------|-------|-------|-----------------------|
| Change percentage in GB | Pre-treatment | 20 | 31.35 | 6.78 | 0.672 |
| | After 3 months | 20 | 30.50 | 13.27 | |

significantly enhanced the activity of chondrocytes, which regenerated cartilage more than PRP, as the I-PRF enhanced chondrocyte proliferation, RNA levels, and collagen II compared to PRP and control groups.

This study did not take gingival biotype into account, so we recommend conducting further studies to compare the response of thin and thick gingival biotypes when applying the I-PRF technique.

Indeed, there are many approaches, other than PRF preparation, to manage papilla deficiency; however, each approach has its limitations and disadvantages. This study focused on a non-surgical technique to reconstruct the papilla. Although the injections' effectiveness has not been achieved sufficient results, it cannot be denied that it offers a practical, uncomplicated, and inexpensive approach. Additionally,

promising results may be obtained in future studies by increasing the frequency of injections and following-up procedures for longer periods.

Conclusion

Within the limits of this study, the I-PRF injection technique proved ineffective in managing recessed interdental papillae 3 months after treatment. Nevertheless, employing the I-PRF injection technique offers advantages such as practicality and cost-effectiveness. This research recommends conducting studies with larger sample sizes and increased injection frequencies to reconstruct interdental papillae. It also suggests studying the effect of combining PRF and I-PRF in reconstructing recessed interdental papillae.

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Conflicts of interest

The authors declare no competing interests.

Data availability statement

All data are available from the corresponding author upon reasonable request.

Ethics approval

Ethical approval was obtained from the Ethics Committee and the Board of Scientific Research at the Faculty of Dentistry at Damascus University, and the Ministry of Higher Education and Research (No. 3480, date: 4-9-2018). In addition, official written permissions were obtained from all participants to participate in the study.

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