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A comparative study of the effects of advanced platelet-rich fibrin and resorbable collagen membrane in the treatment of gingival recession: a split-mouth, randomized clinical trial

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Abstract

Aim This study aimed to assess the effectiveness of advanced platelet-rich fibrin (A-PRF) combined with the pinhole surgical technique (PST) for enhancing root coverage (RC) in individuals with Miller class I or II gingival recessions (GR). Additionally, it compared the clinical effect of A-PRF and resorbable collagen membrane (RCM).

Materials and methods A total of 18 patients, encompassing 36 treatment sides of 18 Miller class I or II, were randomly assigned to the PST + A-PRF side (18 sides) and the PST + RCM side (18 sides). Clinical assessments of various parameters, including plaque index (PI), clinical attachment level (CAL), keratinized tissue width (KTW), recession depth (RD), recession width (RW), and gingival thickness (GT) were conducted at baseline and three months after the surgical procedure. A numeric rating scale (NRS) was also evaluated during the 1st, 2nd, 3rd and 4th days. This study was formally recorded under the TCTR identification number TCTR20230613005 in the Thai Clinical Trials Register-Medical Research Foundation of Thailand (MRF) on 13/06/2023. Furthermore, it was ethically approved by Sana'a University's Ethical Committee for Medical Research.

Results When comparing the values of 3 months follow-up with the baseline values, intra-side comparison of the PST + A-PRF group showed significant improvements in PI ($P = 0.02$), CAL ($P = 0.01$), and RD ($P = 0.04$), and GT values ($P < 0.01$). The improvements in the PST + A-PRF group were through the reduction of baseline values of PI, CAL, and RD; the mean reductions in PI, CAL, and RD were 0.44 ± 0.71, 0.33 ± 0.45, and 0.22 ± 0.43 respectively, and a significant increase in GT value (0.44 ± 0.24). While there was an insignificant increase in KTW value with no change in RW values (4.50 ± 0.71, $P = 1$). In contrast, intra- side comparison of PST + RCM side showed only a significant reduction in PI value (0.44 ± 0.71, $P = 0.02$) and a significant increase in GT value (0.42 ± 0.26, P = < 0.01). Meanwhile, there were insignificant

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improvements in CAL (2.89 ± 0.95), KTW (3.97 ± 0.74), and RD (1.94 ± 0.87) values. Regarding inter-side comparison, there were no statistically significant among all variables ($p > 0.05$). The pain scores of the numeric rating scale were significantly lower on the PST + A-PRF sides compared with the PST + RCM sides, especially on the 1st, 2nd, and 3rd days ($P < 0.001$).

Conclusion Both A-PRF and RCM showed not wholly satisfactory outcomes in gingival recession treatment. Interestingly, the combination of PST with A-PRF has proven more effective than combining PST with RCM. Additionally, the localized application of A-PRF has been shown to reduce post-operative pain following the pinhole surgical technique.

Keywords Gingival recession, Pinhole technique, Advanced platelet-rich fibrin, Resorbable collagen membrane

Introduction

The primary objectives of plastic periodontal surgical (PPS) techniques in addressing gingival recessions (GR) revolve around achieving two key outcomes: full coverage of the exposed tooth root (RC) and the attainment of an aesthetically pleasing result [1]. The pinhole surgical technique (PST) is one of these recent procedures invented by John Chao in 2012 and provides a conservative approach for managing Miller's class I and II types of GR using collagen membranes [2]. The rationale for using PST is that it is a minimally invasive approach; scalpels are limited to pinholes with no damage to intracellular tissues; sutures are not required; there are few postoperative complications such as bleeding or pain, and better healing occurs because soft tissue is managed with less force [3].

A biodegradable copolymer of glycoside and lactide is used to make collagen membranes [4]. It is a resorbable product that integrates with the flap connective tissue to prevent epithelial down growth [5]. Absorption within the body, accomplished through hydrolysis, is minimal for 4–6 weeks and complete by 8 months. [6]. Additional advantages of collagen include improved cell migration into wound areas, clot stabilization, fibroblast chemotaxis, and increased growth factors [7].

Platelet-rich fibrin (PRF) was first created by Choukroun et al. in 2001. It is a second-generation platelet concentrate that is autologous, resorbable, and contains a variety of cytokines, growth factors, and cells that are slowly released over time [8]. Choukroun et al. modified it to advanced platelet-rich fibrin (A-PRF) in 2014. It forms a more tensile blood clot rich in GF and a scaffold of fibrin matrix, platelets, leukocytes, and stem cells to interact together with a longer centrifugation time and slower centrifugation speed. The current PRF protocols are as follows [9]: the original PRF protocol (standard protocol): 3000 rpm for 10 min; A-PRF: 1500 rpm for 14 min [10].

When combined and prepared properly, PRF participates in key processes of tissue repair and regeneration, such as cell proliferation and differentiation, extracellular matrix formation, chemotaxis, angiogenesis (neo-vascularization), faster healing, and pain reduction [9].To the best of our knowledge, comparative clinical studies between the application of A-PRF and resorbable collagen membrane (RCM) for gingival recession treatment are limited to the available scientific evidence. Thus, this randomized controlled clinical study aimed to evaluate and compare the clinical effect of A-PRF and RCM application with PST for the treatment of Miller class I and II gingival recession considering root coverage. The study's null hypothesis is that there is no significant difference in the effectiveness of advanced platelet-rich fibrin combined with the pinhole surgical technique compared to resorbable collagen membrane in treating Miller class I or II gingival recessions.

Materials and methods

Selection and preparation of participants

This split-mouth, randomized, single-blind study was conducted on 18 patients, encompassing 36 treatment sides, who came to the Department of Periodontology, Faculty of Dentistry, Sana'a University.

The criteria for patient inclusion were as follows: the presence of bilateral Miller Class I or II gingival recessions on bilateral lower canines, vital anterior teeth or premolars, an identifiable cementoenamel junction (CEJ), gingival thickness of at least 1 mm for the recession area, and good periodontal health. Pregnant or lactating females, those with fully restored teeth or mobility, cervical abrasion, caries, or abfraction, a history of prolonged antibiotic use, smokers, and khat chewing are all excluded. This study was formally recorded under the TCTR identification number TCTR20230613005 in the Thai Clinical Trials Register-Medical Research Foundation of Thailand (MRF) on 13/06/2023. Furthermore, it was ethically approved by Sana'a University's ethical committee for medical research and has been conducted in accordance with the guidelines of the declaration of Helsinki. Before their participation, all study participants provided written informed consent.

Sample size calculation

The sample size of the study was calculated using G*Power software (version 3.1.9.2) employing t-test, tow-tailed, Power of (1 − β err) 0.80, α=0.05, and an allocation ratio of 1. The effect size (0.989) was determined based on the previous study for the difference in CAL between test and control groups [11]. Accordingly, the calculated sample size was 18 samples per side. In addition, it was conducted based on previous comparable studies [12, 13].

Randomization

The patients underwent assessment by blinded outcome assessors (M.S. & M.A.). To determine the placement of either A-PRF or RCM, a random selection process was employed. This selection process was carried out using sealed envelopes, each containing cards marked 'A-PRF' or 'RCM.' Subsequently, the periodontist (B.A.) opened these envelopes once the patients had made their choice. There were two distinct sides: one designated as the A-PRF side, where PST+A-PRF treatment was administered, and the other as the PST+RCM side, where RCM treatment was provided [14, 15].

The A-PRF preparation protocol

Intravenous blood was collected from the antecubital vein in 10-ml sterilized glass-coated plastic tubes without anticoagulant before surgery and centrifuged immediately using a lower-speed centrifugal machine at 1500 rpm for 14 min [16]. Once a fibrin clot had formed in the middle section of the tube, it was removed and gently compressed to form the A-PRF membrane. The fibrin clot formed in the tube's middle. The upper portion contained acellular plasma, while the lower portion contained red corpuscles. The fibrin clot was easily separated from the centrifuged blood and distributed on sterile gauze [17]. The period between PRF extraction and recipient site preparation was less than nine minutes.

Study variables

The primary outcome variables of the study were PI, CAL, KTW, RD, RW, and GT.

The secondary outcome variable was NRS.

Surgical protocol

Each patient had received initial periodontal treatment, which included oral hygiene instructions, plaque control, scaling, and root planning. They were carried out three weeks before the root-coverage surgical procedure.

According to Chao's 2012 technique [2], the same periodontist performed all surgical procedures for both sides in a single appointment; (a) local infiltration of 2% lidocaine with 1:100,000 epinephrine was used to anesthetize the surgical area, (b) root planning with ultrasonic instruments, followed by hand curettes, (c) a minimal horizontal incision, approximately 2 to 3 millimeters in size, was made near the bottom of the vestibule, just apical to the recipient site, on both sides. This incision was skillfully executed using a no. 12 scalpel (Bard-Parker) (Fig. 1), (d) a gingival elevator was inserted through the pinhole used for blunt dissection and raised a mucoperiosteal flap, (e) the flap was coronally and horizontally

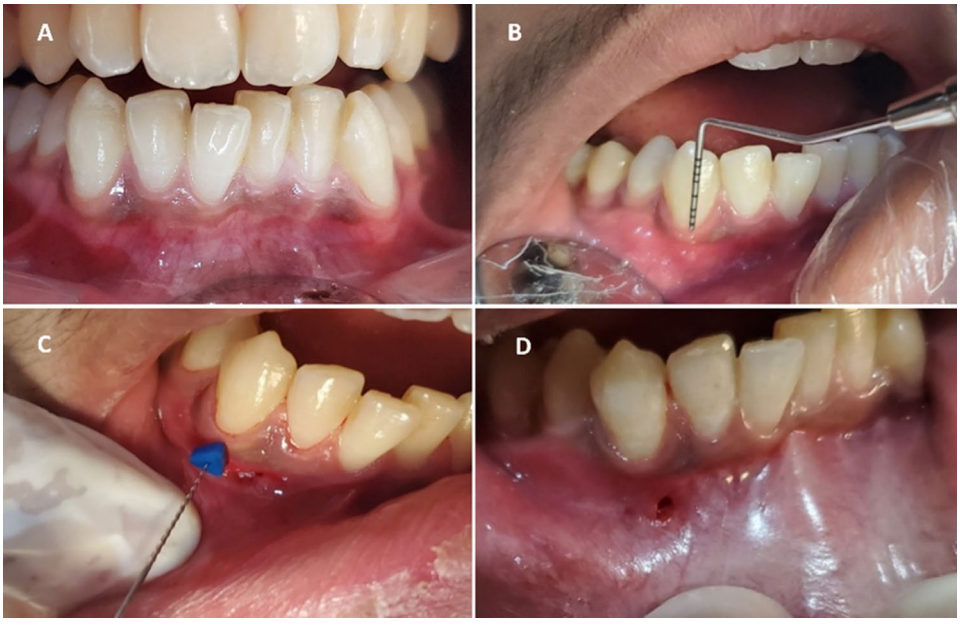

Fig. 1 The surgical procedures of the pinhole surgical technique: (**A**) Scaling and root planning. (**B** &**C**) Baseline measurements. (**D**) A minimal horizontal incision, approximately 2 to 3 millimetres in size

extended to allow the elevation of two adjacent papillae on each side of the denuded root, (f) The interdental elongation of the flap produced an advanced flap that was freely movable and free of tension, which was then used to cover the CEJ [18]. (g) A-PRF was carefully adapted through the pinhole and packed properly on one side until the advancement was stabilized and sufficient fullness in the papillary tissues was created for adhesion to the mucogingival tissue (Fig. 2). (h) The RCM (SYNERGY brand) was cut into small strips and moistened with normal saline before being inserted through the pinhole into another side (Fig. 3). The number of strips employed was established based on the amount of material needed to firmly secure the flap in its intended location, ensuring it remains in position without the need for sutures, surgical dressings, or tissue adhesive [19].

Postoperative care

200 mg of ibuprofen was prescribed as an analgesic to relieve the pain. The patients were instructed to brush untreated teeth gently (using the roll technique) with a soft toothbrush. Patients were also instructed to spray chlorhexidine solution (0.12%) on their teeth for two weeks instead of brushing the treated area to ensure the necessary plaque control measures were taken. Patients were clinically observed and evaluated for complications like pain, swelling, and healing. The patients were reevaluated.

Clinical assessments

The subsequent variables underwent assessment both at the initial baseline and again three months following the surgical procedure: plaque index (PI) [20], gingival index (GI) [21], RD measured from the cementoenamel junction (CEJ) to the lowest point of the free margin [22], RW is a horizontal measurement of the recession in the mesiodistal direction at the CEJ [23], CAL measured from the CEJ to the pocket's base, KTW; measured from the marginal gingiva to the mucogingival junction (MGJ) [24], GT measured from the mid-point location between the gingival margin and MGJ using an endodontic file with a rubber stopper [25]. RD, CAL, KTW, and RW measurements were taken at the mid-buccal point of the affected teeth. Patients were recalled after the first, second, and third weeks and at the end of the first, second, and third months. The RC percentage (%) was calculated using the following formula: [(pre-operative RD - post-operative RD) / pre-operative RD] × 100 [26].

The assessment of postoperative pain levels occurred on the first, second, third, and fourth days following the surgical procedure. This evaluation employed the numeric rating scale (NRS), which ranges from 0 to 10. Within this scale, pain intensity was categorized as follows: a score of 1 to 3 denoted mild pain, 4 to 6 signified moderate pain, and 7 to 10 represented severe pain. Typically, a score of 0 on this scale indicated the absence of any pain [27].

Fig. 2 The surgical procedures of the pinhole surgical technique with advanced platelet-rich fibrin (A-PRF): (**A**) A gingival elevator was inserted through the pinhole used for blunt dissection and raised a mucoperiosteal flap. (**B**) A-PRF after centrifuge. (**C** & **D**) A-PRF was carefully adapted through the pinhole and packed properly in one side without suturing

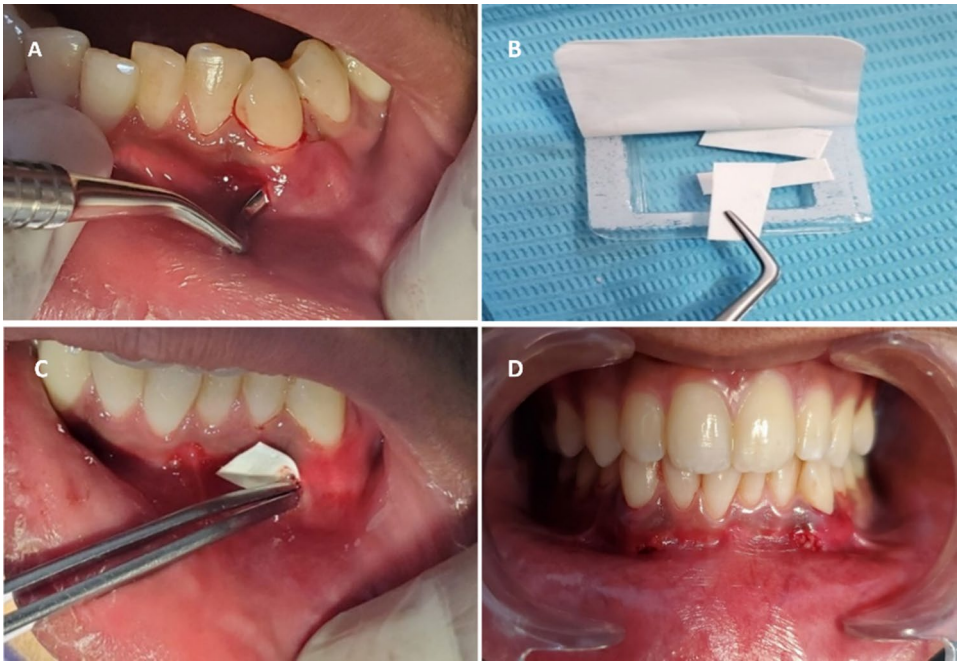

Fig. 3 The surgical procedures of the pinhole surgical technique with resorbable collagen membrane (RCM): (**A**) A gingival elevator was inserted through the pinhole used for blunt dissection and raised a mucoperiosteal flap. (**B**) The RCM was cut into small strips and moistened with normal saline before being inserted through the pinhole into another side. (**C** & **D**) RCM was carefully adapted through the pinhole and packed properly into another side without suturing

Fig. 4 A case with gingival recession Miller class I of bilateral lower canine; (**A**) Preoperative view showing bilateral lower canine with gingival recession Miller class I. (**B**) Immediately postoperative view showing the right pinhole received advanced platelet-rich fibrin (A-PRF), the left pinhole received resorbable collagen membrane (RCM). (**C**) Healing at 1st week following surgery. (**D**) Postoperative view at 1st month following surgery. (**E**) Postoperative view at 3rd month following surgery

Statistical analysis

The Social Sciences Program (SPSS) version 22 was used for all analyses. The descriptive data were expressed as mean (SD). The Chi-square test was performed to analyze the difference in NRS between the two sides. Mann–Whitney test was used to compare all variables measured between the sides. Additionally, the reliability of the measurements between assessors was evaluated using the intraclass correlation coefficient (ICC) test. All metrics showed ICC values exceeding 0.9, indicating a high level of agreement. At $P<0.05$, differences were considered statistically significant.

Results

A total of 18 patients (12 males and 6 females), encompassing 36 treatment sides with an average age of 25 years, were enrolled in this clinical trial study.

When comparing the values of 3 months follow-up with the baseline values (Fig. 4), intra-side comparison of the PST + A-PRF group showed significant improvements

Table 1 Intra-sides comparison of clinical parameters regarding mean with standard deviation at different time intervals in both sides

Variables	PST + A-PRF			PST + RCM		
	T1	T2	*P*	T1	T2	*P*
Plaque index	1.00 ± 0.59	0.56 ± 0.51	0.02	1.00 ± 0.59	0.56 ± 0.51	0.02
Clinical attachment level	3.11 ± 0.90	2.78 ± 0.86	0.01	3.11 ± 0.9	2.89 ± 0.95	0.15
Keratinized tissue width	3.94 ± 0.94	4.19 ± 0.71	0.07	3.83 ± 0.92	3.97 ± 0.74	0.33
Recession depth	2.06 ± 0.94	1.83 ± 0.78	0.04	2.11 ± 0.96	1.94 ± 0.87	0.27
Recession width	4.50 ± 0.71	4.50 ± 0.71	1	4.56 ± 0.71	4.61 ± 0.61	0.33
Gingival thickness	1.38 ± 0.27	1.83 ± 0.29	< 0.01	1.36 ± 0.29	1.77 ± 0.31	< 0.01

A-PRF; advanced platelet-rich fibrin, RCM; resorbable collagen membrane, PST; pinhole surgical technique, T1; baseline values, T2; three months post-surgery values, P; P-value

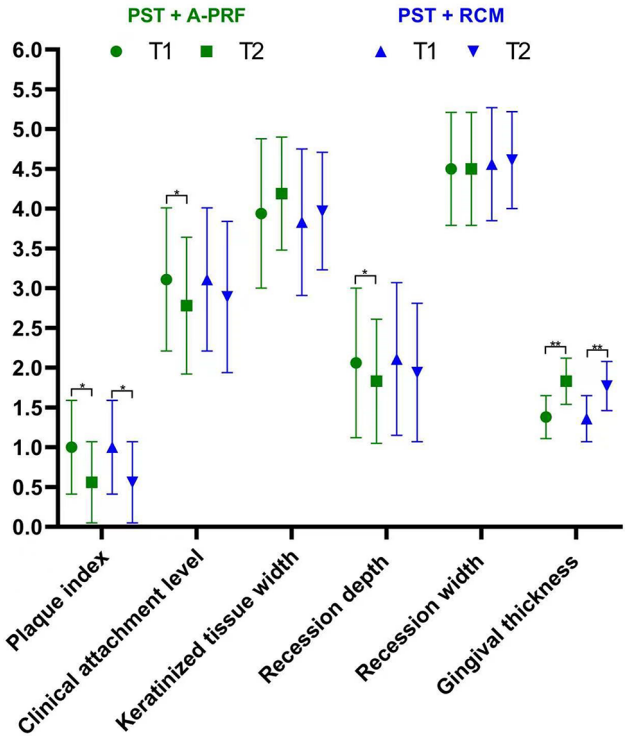

Fig. 5 Intra-sides comparison of the means of the improvement difference between baseline and 3 months post-surgery values

in PI (P=0.02), CAL (P=0.01), and RD (P=0.04), and GT values (P<0.01). The improvements in the PST+A-PRF group were through the reduction of baseline values of PI, CAL, and RD; the mean reductions in PI, CAL, and RD were 0.44±0.71, 0.33±0.45, and 0.22±0.43 respectively, and a significant increase in GT value (0.44±0.24). While there was an insignificant increase in KTW value with no change in RW values (4.50±0.71, P=1) (Table 1; Fig. 5).

In contrast, intra- side comparison of PST+RCM side showed only a significant reduction in PI value (0.44±0.71, P=0.02) and a significant increase in GT value (0.42±0.26, P=<0.01). Meanwhile, there were insignificant improvements in CAL (2.89±0.95), KTW (3.97±0.74), and RD (1.94±0.87) (Table 1; Fig. 5).

Table 2 Inter-sides comparison of the means of the improvement difference between baseline and 3 months post-surgery values are reported as mean with standard deviation

Variables	PST + A-PRF	PST + RCM	*P*.Value
Plaque index	¯0.44 ± 0.71	¯0.44 ± 0.71	1
Clinical attachment level	¯0.33 ± 0.45	¯0.14 ± 0.59	0.88
Keratinized tissue width	+ 0.25 ± 0.55	+ 0.14 ± 0.59	0.86
Recession depth	¯0.22 ± 0.43	¯0.17 ± 0.62	0.29
Recession width	0.00 ± 0.00	0.06 ± 0.24	0.04
Gingival thickness	+ 0.44 ± 0.24	+ 0.42 ± 0.26	0.53

A-PRF; advanced platelet-rich fibrin, RCM; resorbable collagen membrane, PST; pinhole surgical technique, ($^{+/-}$values); means the improvement value that occurred

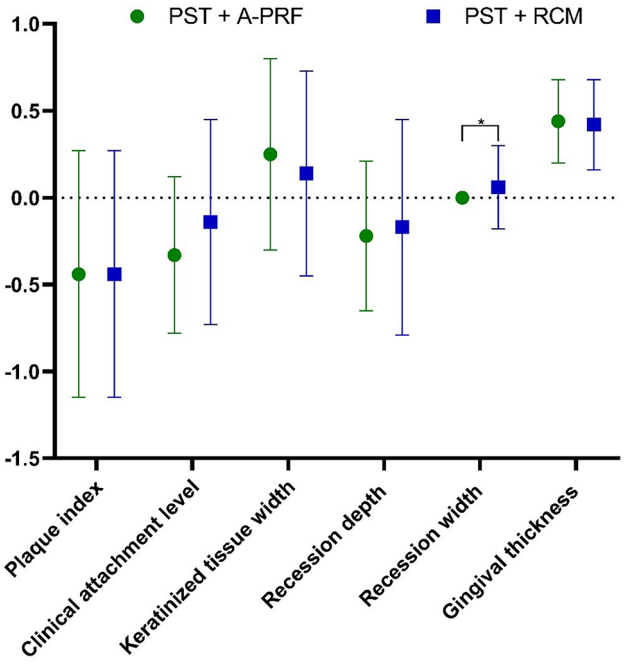

Fig. 6 Inter-sides comparison of the means of the improvement difference between baseline and 3 months post-surgery values

Regarding inter-side comparisons (Table 2; Fig. 6), among all variables, there was no statistical significance except in Recession width. PST+A-PRF sides showed the same value before and after the treatment, whilst PST+RCM sides showed increased Recession width value. The pain scores of the numeric rating scale were

Table 3 Comparison of pain scores on the numeric rating scale (NRS) between the A-PRF and RCM sides during the different time intervals using the Chi-square test

Numeric rating scale (NRS)	1st day		2nd day		3rd day		4th day	
	A-PRF side	RCM side	A-PRF side	RCM side	A-PRF side	RCM side	A-PRF side	RCM side
No pain	8	0	18	0	18	4	18	15
Mild pain	10	0	0	4	0	11	0	3
Moderate	0	5	0	11	0	3	0	0
Severe	0	13	0	3	0	0	0	0
P-value	< 0.01		< 0.01		< 0.01		0.35	

A-PRF; advanced platelet-rich fibrin, RCM; resorbable collagen membrane

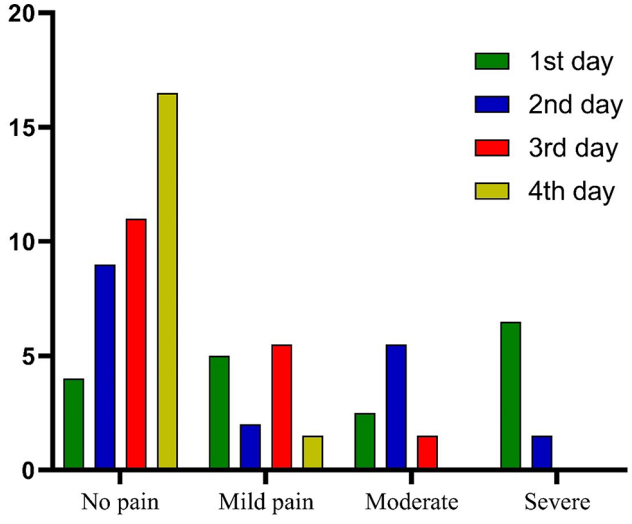

Fig. 7 Comparison of pain scores on the numeric rating scale (NRS) between the A-PRF and RCM sides during the different time intervals

significantly lower in the PST+A-PRF sides than the PST+RCM sides, especially on the 1st, 2[nd,] and 3rd days ($P<0.01$), with no statistical significance on the 4th day (Table 3; Fig. 7).

Discussion

The primary cause of dentin hypersensitivity is when the root surfaces become exposed due to gingival recession [28]. To address this issue, various periodontal plastic surgery techniques have been developed over the years to conceal these exposed root surfaces [29]. One such method is the pinhole surgical technique, which offers a more conservative approach to treating these recession-related defects [2]. The current study aimed to compare the efficacy of A-PRF and RCM for treating Miller Class I and II recessions with PST.

In the present study, both sides (PST+A-PRF and PST+RCM) showed a significant reduction in PI, CAL, and RD values with a greater gain in KTW and GT at the time point of 3 months. These results are consistent with a study conducted by Trivedi DS et al. [30]., in which they evaluated the efficacy of the pinhole surgical technique with and without the use of A-PRF for treating multiple adjacent recession defects. Their study's conclusions indicated that certain key parameters, such as PD, CAL, and GRD, showed improved outcomes in terms of greater gains in KTW and GT for both groups. Notably, these parameters exhibited a decrease in values at both the 6-month and 12-month follow-up points. Importantly, the A-PRF group demonstrated even more significant reductions in these values compared to the control group. Padma R et al. in 2013 found that a split-mouth study design increased the KTW when PRF was added to CAF [31]. Also, in individuals with thin periodontal phenotypes, using injectable-PRF (I-PRF) alone or in combination with microneedling (MN) can potentially enhance gingival thickness. The findings indicate that applying I-PRF and MN may serve as an initial non-surgical approach to augment gingival thickness [32]. Furthermore, based on the findings of Patra L's study, it can be considered that multiple or isolated gingival recessions of Miller's class-I and class-II defects can be successfully treated with the minimally invasive vestibular incision subperiosteal tunnel access technique, along with a collagen membrane that functions as a scaffold and chemoattractant. An additional benefit of this approach is the use of an injectable form of PRF that has the potential to release more growth factors and regenerative cells responsible for tissue regeneration [33].

Paolantonio M et al. [34] concluded that augmenting the thickness of gingival tissue yields a favorable outcome by reducing the likelihood of recurring gingival recession. This reduction is attributed to the fact that chronic trauma caused by insufficient toothbrushing or inflammatory responses within the delicate marginal tissue can lead to the development of gingival recession [35]. Agarwal SK et al. reported that PRF emerges as a superior choice for root coverage when contrasted with the utilization of fresh amniotic membrane (FAM) and connective tissue graft (CAF) alone. The study encompassed 45 sites affected by gum recession, and treatments were randomly allocated into three groups: CAF+PRF, CAF+FAM, and CAF, used independently. The observed discrepancies in the outcomes were attributed to the presence of thin gingival tissue measuring just 1 mm in all the groups at the study's outset [36].

As well as, Choudhury et al. [37] reported that the pinhole surgical technique has exhibited favorable outcomes when used in conjunction with both collagen membranes and PRF. Notably, there has been a significant improvement in recession height (RH), recession RW, and WKT with the application of both collagen membranes and PRF.

Our result contrasted with this study in recession width; PST+A-PRF, and PST+RCM sides showed no improvement in RW values. Also, Moraschini and Barboza et al. reported that the application of PRF in the management of GR did not yield better results in terms of RC, KTW, and CAL when compared to alternative treatment approaches. This lack of improvement can be attributed to the swift degradation of PRF at the surgical site, potentially impeding the early stabilization of periodontal tissues during the healing process [38]. Clinical and experimental study has demonstrated that the utilization of a collagen membrane plays a pivotal role in facilitating the formation of a new connective tissue attachment [39]. Other studies concluded that using a PRF membrane with CAF provided no significant advantage in terms of recession coverage on single and multiple recessions, except for an increase in GT [40].

In terms of postoperative pain, the pain scores of the numeric rating scale were significantly lower on the PST+A-PRF sides compared with the PST+RCM sides, especially on the 1st, 2nd and 3rd days ($P<0.001$). These findings were associated with the regenerative abilities of A-PRF and the heightened concentration of fibrin fibers, which stimulate the process of angiogenesis by releasing crucial growth factors. The use of this autogenous biological material in the test group completely eliminates the risk of adverse reactions to foreign (non-autogenous) materials in the control group. Several studies found similar results, using PRF to reduce pain scores was more effective than using CAF alone or CAF and CTG. As a result, the use of PRF reduces patient-reported morbidity at the donor site when compared to using CAF alone. This improvement also extends to the process of harvesting CTG at the recipient site [41].

The study was limited by a small sample size and short-term follow-up, which could provide a more detailed understanding of the specific type of attachment achieved. Additionally, the patients in the study had received surgical treatment on both sides during the same appointment, making it challenging for them to distinguish the level of pain on each side accurately.

Considering the growing interest in minimally invasive procedures, it is imperative to conduct a larger sample size further study considers the factors that may be included in the regression model (e.g. gender, age, oral hygiene, missing teeth, presence of any prosthesis, root coverage esthetic score, etc.) to gain deeper insights into this subject.

Conclusion

Both A-PRF and RCM showed not wholly satisfactory outcomes in gingival recession treatment. Interestingly, the combination of PST with A-PRF has proven more effective than combining PST with RCM. Additionally, the localized application of A-PRF has been shown to reduce post-operative pain following the pinhole surgical technique.

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Author contributions
M.S.A, B.A, and S.A.E contributed to data collection, interpretation of data, designing the study and writing the original manuscript. S.A.E and M.A have critically revised the manuscript. All authors have approved the final manuscript before its submission.

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Data availability
The datasets used and/or analysed during the study are available from the corresponding author upon reasonable request.

Declarations

Ethics approval and consent to participate
This study was registered with the TCTR identification number TCTR20230613005 at Thai Clinical Trials Register-Medical Research Foundation of Thailand (MRF) on 13/06/2023. Also, it was ethically approved from Sana'a University's ethical committee for medical research, and has been conducted in accordance to the guidelines of the declaration of Helsinki. Written informed consent was obtained from all patients in the study.

Consent for publication
Not applicable.

Competing interests
The authors declare no competing interests.

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