Original Article - Comparative Study

Healing Potential of Platelet Rich Fibrin in Impacted Mandibular Third Molar Extraction Sockets

Manzoor Mohammad Dar, Ajaz Ahmad Shah, A. Latief Najar, Mubashir Younis, Muneet Kapoor, Jahangir Irfan Dar

Department of Oral and Maxillofacial Surgery, Government Dental College and Hospital, Srinagar, Jammu and Kashmir, India

Abstract

Background: Healing of both hard and soft tissues have become one of the great challenges, faced in clinical research in development of bioactive surgical additives responsible for regulating inflammation and increasing healing. Platelet-rich fibrin (PRF) represents a new step in the platelet gel therapeutic concept with simplified processing minus artificial biochemical modification. **Aims and Objectives:** The aim of this study is to evaluate the effectiveness of PRF on soft-tissue healing and bone tissue healing in terms of postoperative pain, postoperative swelling, soft tissue healing, and the quality of bone healing at the mandibular third molar socket. **Materials and Methods:** A clinical study will be done on patients reporting to the Department of Oral and Maxillofacial Surgery in Government Dental College, Srinagar, requiring disimpaction of bilateral mesioangular impacted mandibular third molars in 60 patients. **Results:** The present prospective clinical study evaluates the effect of PRF in healing of mandibular third molar extraction sockets. There was no difference in the age gender and type of impaction between the two groups as the mean postoperative pain score (visual analog scale) was lower for the PRF group (Group A) at all points of time when compared with the control (Group B), and this was statistically significant ($P < 0.05$). The mean percentage swelling was lower for the PRF group (Group A) at all points of time when compared with the control (Group B). Evaluating the effect of treatments (with or without PRF) on lamina dura score shows that in both the groups at different time periods, significant ($P < 0.001$) difference was observed on lamina dura score. **Conclusion:** The results of the present study suggest that application of autologous PRF gel has a beneficial effect on the healing of extraction sockets after third molar surgery.

Keywords: Disimpaction, platelet-rich fibrin, third molar

Introduction

Healing of both hard and soft tissues have become one of the great challenges, faced in clinical research in development of bioactive surgical additives responsible for regulating inflammation and increasing healing. Understanding of this process at microcellular level is still not complete, but it is a proven fact that platelets do play an important role in wound healing.[1] Patients who undergo impacted third molar extraction experience intense inflammatory pain, swelling, and delayed bone healing. Socket healing is a highly coordinated sequence of biochemical, physiologic, cellular, and molecular responses involving numerous cell types, growth factors, hormones, cytokines, and other proteins, which is directed toward restoring tissue integrity and functional capacity after injury.[2] To accelerate the healing of an extracted socket autograft (mandibular symphyseal graft) or allograft demineralized freeze-dried bone graft, freeze-dried bone allograft can be used. Autograft is associated with high degree of donor-site morbidity and allograft is associated with risk of disease transmission which pushed the clinicians toward opting for more promising autologous material such as platelet-rich plasma (PRP) and platelet-rich fibrin (PRF) for more predictive results.[3] The use of platelet concentrates, based on the concept of cell therapy by growth factors, reopened technologic research on the autologous fibrin adhesives, especially the PRP.[4-6]

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Address for correspondence: Dr. Manzoor Dar, Room No 202, Department of Oral and Maxillofacial Surgery, Government Dental College, Srinagar, Jammu and Kashmir, India.
E-mail: itsdrmann@yahoo.com

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PRF represents a new step in the platelet gel therapeutic concept with simplified processing minus artificial biochemical modification. PRF is an immune and platelet concentrate collected on a single fibrin membrane containing all the constituents of a blood favorable for healing and immunity. The fibrin matrix supports them and is responsible for angiogenesis and immunity control.

Although the overall complication rate after the third molar (M3) extraction is low and most complications are minor, M3 removal is so common that the population morbidity of complications may be significant. As such, efforts to limit intraoperative or postoperative complications may have a great impact in terms of enhancing patient outcome.[7]

Materials and Methods

A clinical study will be done on patients reporting to the Department of Oral and Maxillofacial Surgery in Government Dental College, Srinagar, requiring disimpaction of bilateral mesioangular impacted mandibular third molars in 60 patients.

Inclusion criteria

- Patients aged 18–50
- Patients either male or female
- Bilateral mesioagular impacted mandibular third molars planned for extraction
- Surgical site free of active infection
- Patients free of significant systemic diseases.

Exclusion criteria

- Medically compromised patients
- Known mentally challenged patients and patients unable to communicate
- Pregnant and lactating women
- Patients with a history of smoking.

Study design

This is a split-mouth study where mesioangular impacted lower third molars on either side will be divided into two groups:

Group A – Test group: Those in which PRF will be placed into the extraction socket.

Group B – Control group: Those in which PRF will not be placed in the extraction socket.

In every patient, one side will belong to Group A and the other will be Group B.

In every odd patient, PRF is placed in the left socket.

In every even patient, PRF is placed in the right socket.

Surgical technique

Written informed consent was obtained from all patients. The area of the antecubital region was prepared with the cotton and spirit. Ten milliliters of intravenous blood was drawn from the antecubital region using 10 ml sterile syringe. This was transferred to centrifugal vials for the preparation of PRF (only for Group A cases). Patients were asked to gargle with chlorhexidine gluconate mouthwash 10 min before starting the procedure. As the PRF preparation requires 15–20 min, in the mean time, the extraction procedure was started. Surgical procedure was done after draping and painting the surgical area with povidone-iodine solution. All the patients were treated using 2% lignocaine hydrochloride with adrenaline in 1:80000 concentrations. Both right and left impacted molars were treated by the same surgeon with a time interval of minimum 30 days. Standard Ward's incision was given with B.P. blade No. 15 and mucoperiosteal flap was raised. After the mucoperiosteal flap reflection, bone removal was done with straight fissure bur (No. 702 carbide). Once the tooth was removed smoothening of bone margins, irrigation of socket with normal saline was done. PRF was placed in the Group A sockets with the help of tweezer and sutured. While in case of Group B, the extraction socket was sutured with 3–0 silk suture without placement of PRF. Primary closure was achieved with 3/4th circle reverse cutting needle 3–0 black silk after surgery. Immediate postoperative X-ray (intraoral periapical [IOPA]) of the extraction socket was taken. Postextraction instructions were given and patients were recalled for follow-up on 1st, 3rd, 7th, and 14th postoperative day and also after 1 month and 3 months. All the patients were prescribed analgesic and antibiotics as under:

- Tablet amoxicillin potassium clavulanate 625 mg BD for 5 days
- Tablet acefenac 100 BD for 5 days
- Tablet pantoprazole 40 mg OD for 5 days.

Method of preparation of platelet-rich fibrin

The 10 ml blood is drawn into test tubes without an anticoagulant and centrifuged immediately. Blood is centrifuged using a table top centrifuge for 12 min at 3000 rpm. The resultant product consists of the following three layers:

- Top most layer consisting of acellular plasma
- PRF clot in the middle
- Red blood cells at the bottom.

Due to the absence of an anticoagulant, the blood begins to coagulate as soon as it comes in contact with the glass surface. Therefore, for successful preparation of PRF, speedy blood collection, and immediate centrifugation, before the clotting cascade is initiated is absolutely essential [Figures 1-5].

Clinical parameters

Various preoperative, intraoperative, and postoperative parameters were used to evaluate the study subjects.

- Orthopantomograph (OPG)/IOPA radiograph (IOPAR)
  - Postoperative assessment
    - Postoperatively, patients were evaluated bilaterally for-Pain – 1st, 3rd, 7th, and 14th postoperative day
      - Swelling – 1st, 3rd, 7th, and 14th postoperative day
      - Periodontal health – immediate postoperatively, 4th week, and 12th week
      - Bone healing – on 4th week and 12th week

- PAIN
  - It will be evaluated using 10-point visual analog scale (VAS), with a score of "0" equals "no pain" and "10" equals "very severe pain." Pain will be evaluated on 1st, 3rd, 7th, and 14th postoperative day.

Swelling

Swelling will be calculated using a modification of the method of Schultze *et al.*[8] and this requires measuring the distances from the tragus to the soft tissue pogonion.

Swelling will be evaluated on 1st, 3rd, 7th, and 14th postoperative day.

Bone healing

The bone healing of the third molar socket is assessed using IOPAR/OPG. The criteria for IOPAR of bone healing and scoring system are based on modification of method used by Kelly *et al*. or computer-guided software for bone density in case of OPG. Three parameters, namely, lamina dura score, density score, and trabeculae pattern score will be assessed. Radiographs will be taken immediately after the procedure and on 4th and 12th weeks postoperatively. Bone healing will be evaluated after 4th and 12th week.

Lamina dura score

- +2 = Lamina dura essentially absent may be present in isolated areas
- +1 = Lamina dura substantially thinned, missing in some areas
- 0 = Within normal limits
- −1 = Portions of lamina dura thickened, milder degrees
- −2 = Entire lamina dura substantially thickened.

Overall density score

- +2 = Severe increase in radiographic density
- +1 = Mild-to-moderate increase in radiographic density
- 0 = Within normal limits
- −1 = Mild-to-moderate decrease in radiographic density
- −2 = Severe decrease in radiographic density.

Trabecular pattern score

- +2 = All trabeculae substantially coarser
- +1 = Some coarser trabeculae; milder degrees
- 0 = Within normal limits
- −1 = Delicate finely meshed trabeculations
- −2 = Granular, nearly homogenous patterns; individual trabeculations essentially absent.

Soft-tissue healing will be assessed using healing index of Landry, Turnbull, and Howley.

Results

The present prospective clinical study evaluates the effect of PRF in healing of mandibular third molar extraction sockets. The study was conducted from August 2013 to April 2015. A total of 30 patients, age between 18 and 50 years (mean ± standard [SE] error: 23.6 ± 4.385 years), either sex (Male = 13 [43.3%], Female = 17 [56.7%]), with bilaterally symmetrical impacted mandibular third molars, requiring surgical method for extraction were recruited and evaluated. One side of the participants was treated with PRF (Group A) and other side without PRF (Group B). The outcome measures of the study were postoperative pain, swelling, and healing. The comparative outcome measures (pain, swelling, and healing) of the two groups are summarized below in section A, B, and C, respectively.

There was no difference in the age [Table 1 and Graph 1], gender [Table 2 and Graph 2], and type of impaction [Table 3 and Graph 3] between the two groups as the mean postoperative pain score (VAS) was lower for the PRF group (Group A) at all points of time when compared with the control (Group B) and this was statistically significant ($P < 0.05$).

Table 1: Age distribution of patients

Age group	No.	%age
18-28	25	83.3
29-39	5	16.7
40-50	0	0.0

Mean±SD=23.6±4.385

Table 2: Gender distribution of patients

Gender	No.	%age
Male	13	43.3
Female	17	56.7
Total	30	100

Table 3: Comparison of pain evaluation between two groups

Pain at	Group A		Group B		*P*
	Mean	SD	Mean	SD	
Day 1	0.83	1.020	2.83	1.895	<0.001*
Day 3	0.50	0.938	2.20	1.864	<0.001*
Day 7	0.23	0.679	1.03	1.542	0.012*
Day 14	0.00	0.000	0.10	0.403	0.179

*Statistically significant difference

Table 4: Comparison of soft tissue healing between two groups

Soft tissue healing at	Group A		Group B		*P*
	Mean	SD	Mean	SD	
Day 1	2.03	0.320	1.80	0.407	0.016*
Day 3	2.83	0.648	2.50	0.572	0.039*
Day 7	3.87	0.571	3.27	0.640	<0.001*
Day 14	4.70	0.651	4.53	0.629	0.317

*Statistically significant difference

The mean percentage swelling was lower for the PRF group (Group A) at all points of time when compared with the control (Group B). Further, for each group, comparing the postoperative mean swelling score within the group (i.e., between different time periods) [Table 4 and Graph 4], there is a statistically significant ($P < 0.001$) decrease in swelling score over the time periods. Similarly, for each period, comparing the postoperative mean swelling score between the groups (i.e., Group A vs. Group B) [Table 4 and Graph 4], test revealed significantly ($P < 0.001$) different and higher swelling score of Group B as compared to Group A at all time periods.

The postoperative total lamina dura score in both groups increased with time and the increase was higher in Group A than Group B. Evaluating the effect of treatments (with or without PRF) on lamina dura score shows that in both the groups at different time periods [Table 5], significant ($P < 0.001$) difference was observed on lamina dura score. Further, comparing the postoperative total lamina dura score within the groups (i.e., between different time periods) [Table 5 and Graph 5], test (Group A) revealed significant ($P < 0.001$) increase in lamina dura score at 4th week as compared to 12th week.

Evaluating the effect of treatments (with or without PRF) on bone density score in both groups at different time periods [Table 6], significant difference between groups on bone density score was seen. Further, for each group, comparing the postoperative total bone density score within the groups (i.e., between different time periods) [Table 6 and Graph 6], significant difference ($P < 0.001$) in bone density score of both groups at 4th week as compared to 12th week was seen.

The postoperative total trabecular pattern score in both groups increased over the time periods, which was higher in Group A than Group B. Evaluating the effect of treatments (with or without PRF) on trabecular pattern score in both groups at different time periods [Table 7], significant increase on trabecular pattern score was seen in the PRF group. Further, for each group, comparing the postoperative mean trabeculae pattern score within the groups (i.e., between different time periods) [Table 7 and Graph 7], test (Group A) revealed significant ($P < 0.001$) increase in trabecular pattern score at 4th week while insignificant ($P > 0.05$) increase in Group A at 12th week as compared to 4th week.

Similarly, for each time period, comparing the postoperative mean trabecular pattern score between the groups (i.e., Group A vs. Group B) [Table 7 and Graph 7] revealed significantly ($P < 0.001$) different and lower trabeculae pattern score of Group B as compared to Group A at both time periods (4th week and 12th week).

The postoperative (4th week and 12th week) total (lamina dura + density + trabeculae pattern) bone healing score of two groups (Group A and Group B) are summarized in Table 8 and also depicted in Graph 8. Table 8 and Graph 8 both showed that the postoperative mean total bone healing score in both groups increased over the time periods, which was evident higher in Group A than Group B.

Comparing the soft-tissue healing between the two groups at different points of time, we observed a statistically significant difference between the groups at day 1, day 3, and day 7, but at day 14, the difference was statistically insignificant.

Hence, the soft-tissue healing difference with time was statistically significant for the test group.

Table 5: Swelling evaluation in two groups

Swelling at	Group A		Group B		*P*
	Mean	SD	Mean	SD	
Day 1	4.00	2.626	5.83	2.793	0.011*
Day 3	1.87	1.943	4.03	2.157	<0.001*
Day 7	0.23	0.626	0.97	0.850	<0.001*
Day 14	0.00	0.000	0.00	0.000	-

*Statistically significant difference

Table 6: Bone healing evaluation in two groups

	Group A		Group B		*P*
	Mean	SD	Mean	SD	
4th Week	2.43	0.935	0.53	0.900	<0.001*
12th Week	5.40	1.070	3.37	1.273	<0.001*

*Statistically significant difference

Table 7: Comparison of laminadura formation in two groups

Laminadura formation at	Group A		Group B		*P*
	No.	%age	No.	%age	
4th Week	20	66.7	2	6.7	<0.001*
12th Week	30	100.0	28	93.3	0.492

*Statistically significant difference

Table 8: Comparison of bone density in two groups

Bone Density	Group A		Group B		*P*
	No.	%age	No.	%age	
4th Week	25	83.3	4	13.3	<0.001*
12th Week	30	100.0	27	90.0	0.237

*Statistically significant difference

Discussion

One of the latest achievements in dentistry is the use of PRF/PRF/for the improvement of reparation and regeneration of the soft and hard tissue after different surgical procedures. PRF is concentrated platelets in a small volume of plasma. During platelet degranulation, many

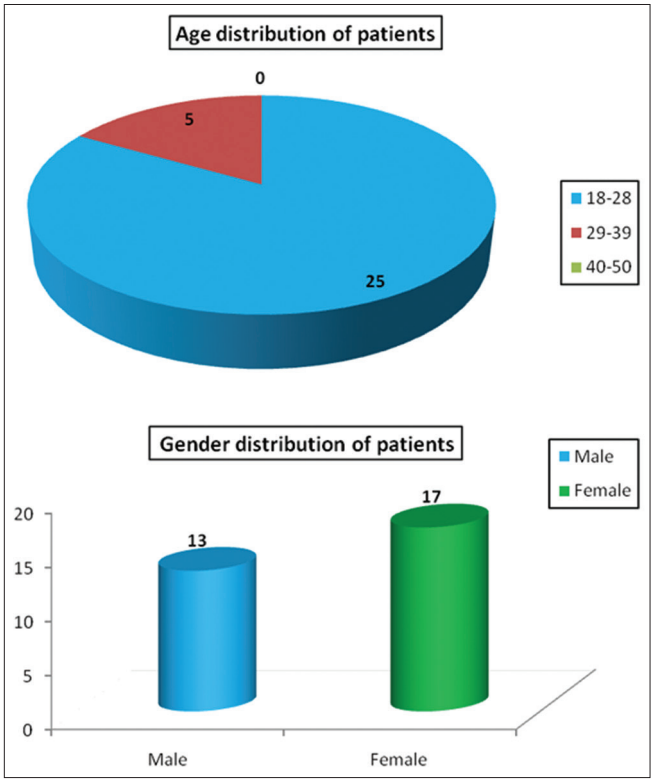
biologic active substances are released which participate in the primary hemostasis and help the following reparation and regeneration of the soft and hard tissue.[9] Today, PRF is an accepted and most extensively worked upon current biological material with immense regenerative potential by other disciplines of clinical dentistry.

Choukroun *et al*. (2001) developed a method of collection of platelets. The protocol aimed at collection of platelet and release of cytokines in a fibrin clot. Here, the fibrin matrix is the key for this product as it supports cell during the initial healing phase.[10]

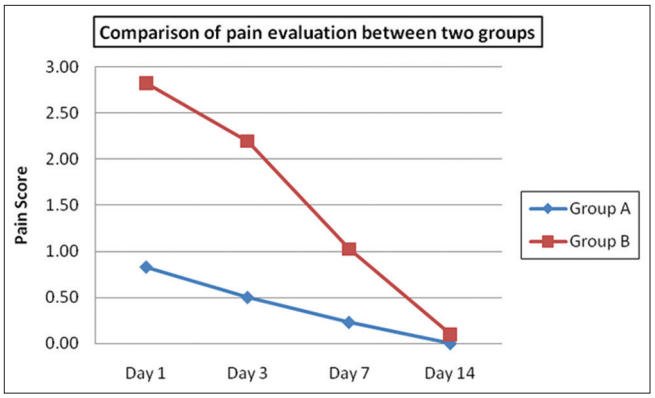
In our clinical study, we evaluated the usefulness of PRF in wound healing in 30 patients of extraction of bilaterally impacted mandibular third molars. Parameters such as age, gender, pain, swelling, soft-tissue healing, and bone healing were recorded.

Our study included 17 females and 13 males patients in the age group of 18–40 years. All of them were radiological diagnosed to have bilaterally impacted mandibular third molar. Smita Singh *et al*. (2013)[11] took a sample size of 15 for their study on application of PRF in surgical management of periapical lesions. Mozzati *et al*. (2007)[12] in their study took a sample size of 5. However, in a study done by Joy Das *et al*. (2014),[13] a sample size of 12 was taken. The present study includes a considerably larger sample size, one of the strengths of the study.

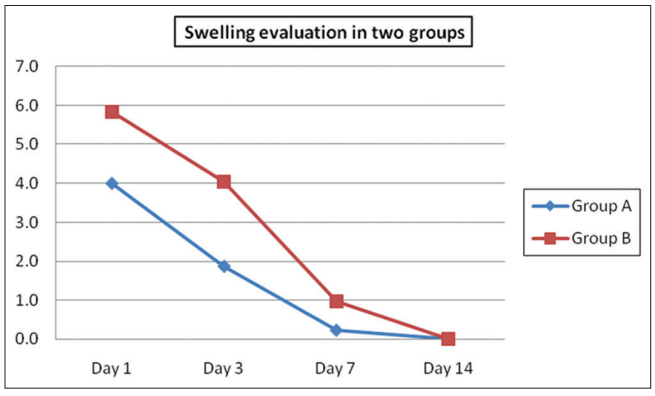
In the present study, 25 patients were in the age group 18–28 years, while 5 were in the age group of 29–39 years. The mean age of males (13) in the present study was 25.76 43.3%, while that of females (17) was 21.94 56.7% and the total mean age (30 patients) was ± SE: 23.6 ± 4.385 years. Kedarnath *et al*. (2011)[14] in their study had a sample size of 10 patients in age range of 17–35 years but they did not mention about mean age in the sample. Bello *et al*.(2011)[15] in their research demonstrated that the surrounding bone in young patients is relatively soft and more resilient compared to older patients where the bone is harder, necessitating more bone removal, with more difficulty in separating tooth from bone, resulting in more postoperative pain, swelling, and trismus.



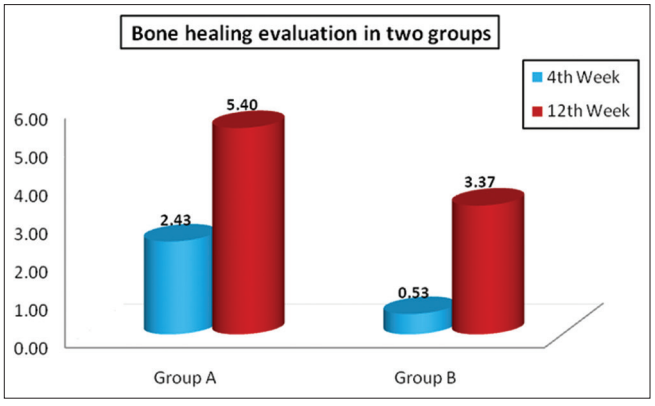
Graph 1: Age and gender distribution



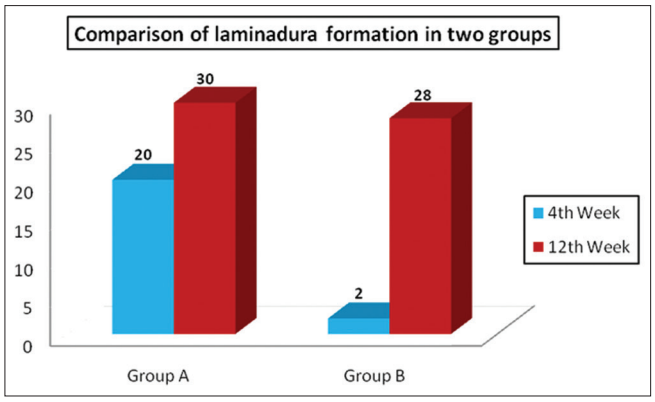
Graph 2: Comparison of pain evaluation between two groups



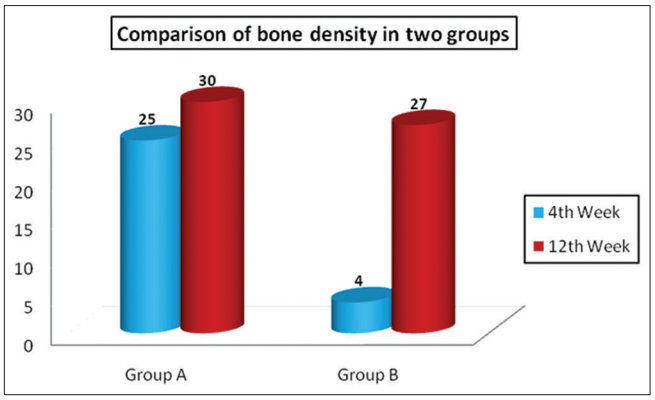
Graph 3: Swelling evaluation in two groups



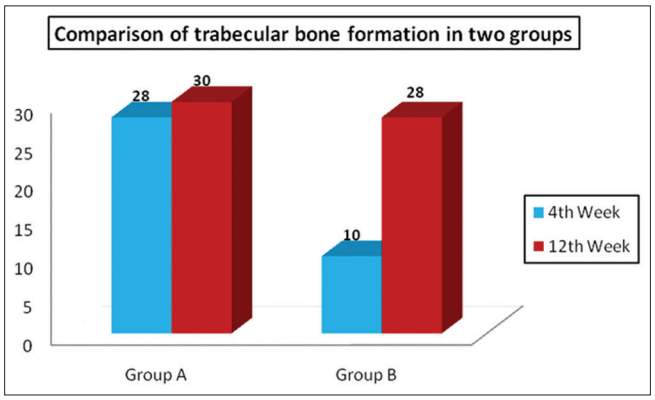
Graph 4: Bone healing evaluation in two groups



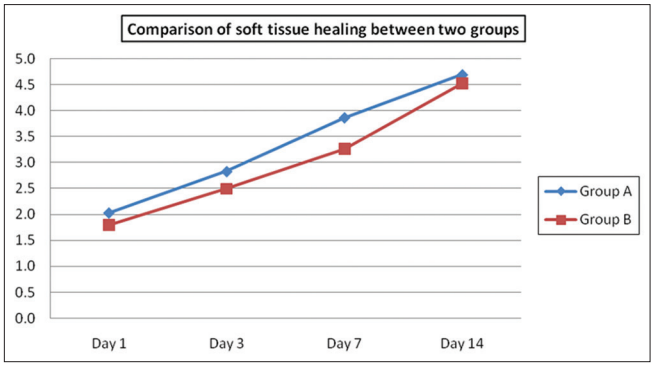
Graph 5: Comparison of lamina dura formation



Graph 6: Comparison of bone density



Graph 7: Comparison of trabecular bone formation



Graph 8: Comparison of soft tissue healing

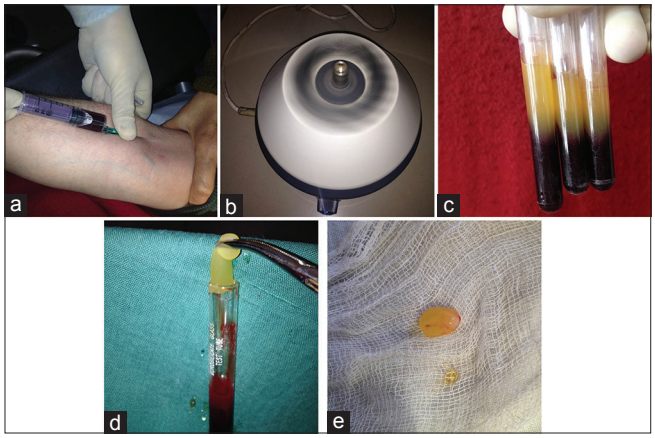


Figure 1: (a) Collection of blood. (b) Centrifugation. (c) Platelet-rich fibrin formed. (d) Separation of platelet-rich fibrin from blood. (e) Platelet-rich fibrin

Figure 2: (a) Preoperative (platelet-rich fibrin group). (b) Platelet-rich fibrin placed after disimpaction. (c) Suturing done. (d) Postoperative day 1 (platelet-rich fibrin). (e) Day 3. (f) Day 7

Pain was assessed with the help of VAS. In our study, we found that as compared to the control side postoperative pain was reduced on the study side on 1st, 3rd, and 7th day ($P \leq 0.001$ and $P < 0.001$, 0.012), statistically it was highly significant. However, difference was not significant on 14th day. Fiero-Serna *et al*. 2011[16] in their study also found that patients reported less pain on the side which received plasma rich in growth factors. Our findings were also supported by Pushkar and Rajshekhar H (2009).[17] The reason for this statistical difference in pain seems to be because of the accelerated growth factor release from the PRF which causes enhanced repair at the surgical site.

Overall in our study, PRF did make difference to the swelling. Although there are not many studies comparing swelling between PRF and non-PRF group, yet our findings were supported by Abhishek Singh *et al*. (2012)[18] who found that swelling was less on the PRF sides. The most important specific activities of platelet-derived growth factor (PDGF) in the PRF include

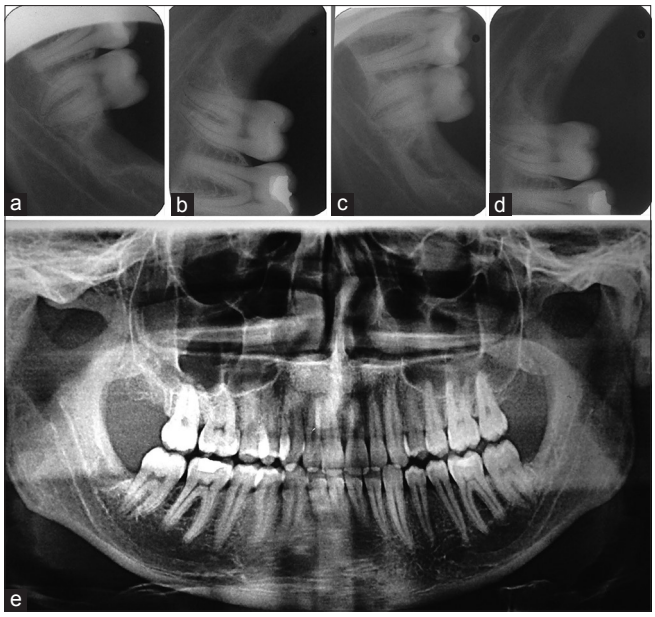


Figure 3: (a) Intraoral periapical radiograph 1 month (left). (b) Intraoral periapical radiograph 1 month (right). (c) Intraoral periapical radiograph 3 months (left). (d) Intraoral periapical radiograph 3 months (right). (e) Three-month orthopantomograph

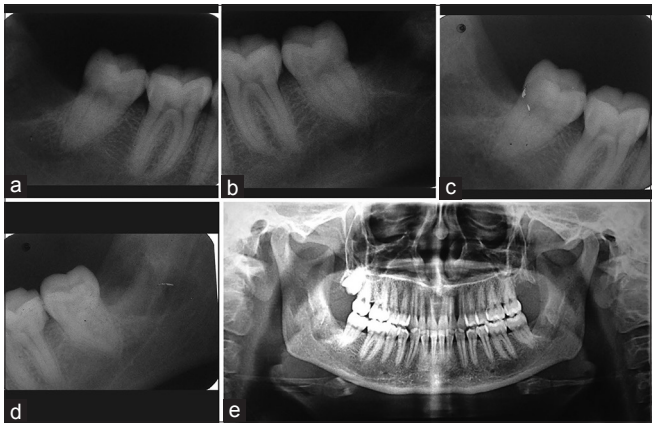


Figure 5: (a) Intraoral periapical radiograph 1 month (left). (b) Intraoral periapical radiograph 1 month (right). (c) Intraoral periapical radiograph 3 months (left). (d) Intraoral periapical radiograph 3 months (right). (e) Three-month orthopantomograph

mitogenesis (increase in the cell population on healing cells), angiogenesis (endothelial mitosis into functioning capillaries), and macrophage activation (debridement of the wound site and a second phase source of growth factors for continued repair and bone regeneration). Therefore, a threefold or greater concentration of platelets, as was measured in PRF, can be expected to have a profound effect on swelling reduction by virtue of it swashing away the exudates due to the above-mentioned activities. In our study, none of the patients (out of 30) had gaping and mucosal dehiscence in control side, PRF has been shown to have good effects on wound healing.

Comparing the soft-tissue healing between the two groups at different points of time, we observed a statistically significant

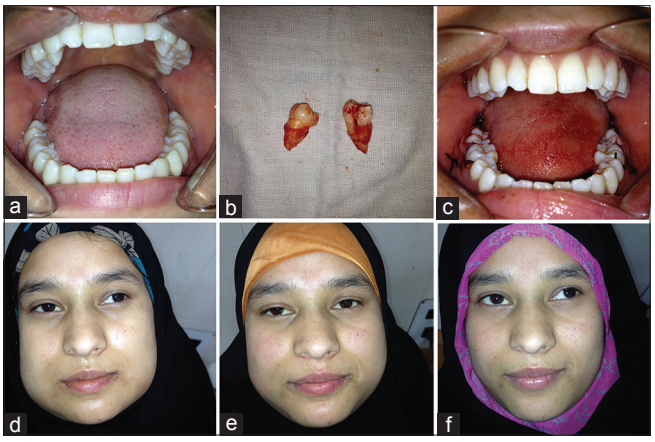


Figure 4: (a) Preoperative (nonplatelet-rich fibrin). (b) Bilateral disimpaction. (c) Suturing. (d) Day 1. (e) Day 3. (f) Day 7

difference between the groups during the early days of repair. Abhishek Singh *et al*. (2012)[18] showed that autologous PRF is biocompatible and has significantly improved soft-tissue healing. Gurbuzer *et al*. 2010[19] reported that PRF promotes wound healing and serve as an immune node that regulates inflammation and provide wound protection due to the presence of growth factors.

The postoperative total (lamina dura + density + trabeculae pattern) bone healing score showed a statistically significant difference between groups at all the time periods. In the 1st month postoperatively, the bone density on the PRF side was found to be significantly increased as compared to the non-PRF side. Three months postoperatively, a very highly significant difference was seen in the bone density between the PRF group and non-PRF group, PRF group showing better bone density. Similar increase in the bone density was observed in the 6th month postoperative follow-up. Girish Rao *et al*. (2013)[20] in their study found a definite improvement in the regeneration of bone after third molar surgery in cases treated with PRF as compared to the control group postoperatively. The enhanced bone density increase is because of the three most important growth factors from the PRF are PDGF, insulin-like growth factor-I, and transforming growth factor-β (TGF–β). Numerous studies, including some dental research have shown that these factors cause chemotaxis and mitogenesis of osteoblast precursors, and they also have the ability to stimulate osteoblast deposition of the collagen matrix of wound healing and of bone. In addition, TGF-β inhibits osteoclast formation and bone resorption, thus favoring bone formation over resorption.[21]

Conclusion

Uneventful and enhanced wound healing is desirable and critical in ascertaining the quality of life after third molar surgery. This will continue to attract the attention and priority of many clinicians and researchers. This study examined the effect of PRF gel on postoperative pain, swelling, and healing and bone regeneration potential on third molar extraction sockets. The results of the present study suggest that the

application of autologous PRF gel has a beneficial effect on the healing of extraction sockets after third molar surgery.

Declaration of patient consent

The authors certify that they have obtained all appropriate patient consent forms. In the form the patient(s) has/have given his/her/their consent for his/her/their images and other clinical information to be reported in the journal. The patients understand that their names and initials will not be published and due efforts will be made to conceal their identity, but anonymity cannot be guaranteed.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

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