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Comparative evaluation of Hydroxyapatite with and without platelet rich fibrin as a bone graft material in mandibular third molar extraction socket

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Abstract

Aim: In this prospective study, we evaluated the effects of hydroxyapatite alone and hydroxyapatite along with Platelet Rich Fibrin (PRF) when used as graft materials on the density of the bone formed and also its effect on post-operative complications.

Patients and Methods: In total, 80 patients were included in this study. The patients were randomly divided into 2 groups: Group 1 - Patients who were grafted with Hydroxyapatite, Group 2 - Patients who were grafted with PRF and Hydroxyapatite.

Results: The visual analogue scale was used to assess pain on the 1st, 2nd and the 3rd post-operative day. No significance between the pain between the two groups was noted. But there was significant difference in mean densitometry analysis readings at 3 months and 6 months. The bone density was significantly high in PRF with Hydroxyapatite group compared to Hydroxyapatite group.

Conclusions: Our results imply that PRF with Hydroxyapatite can help in the formation of bone with significantly higher density compared to hydroxyapatite alone when used as graft materials in surgically extracted third molar sockets.

Keywords: PRF, OPG, Molar Sockets

Introduction

Removal of impacted third molars has become a common procedure in dental practice and about 10 million impacted third molar teeth are removed every year all over the world by oral and maxillofacial surgeons. It is a well-known fact that the extraction of teeth especially impacted ones result in significant

dimensional changes in the jaw bone. The rate of the change and magnitude of these changes have been widely studied, both in various human and in animal studies.(1) (2) These dimensional changes of the bone mimic the bone defects and thus arises the need for appropriate material to replace the lost bone with like material. Bone is a dynamic organ that can regenerate and bone grafting is a dynamic phenomenon.(1) Bone regenerative techniques like graft materials, protein and barrier membrane are often used to improve the bone quality. Bone is the second most commonly transplanted tissue after blood. A bone graft is defined as an implanted material, which promotes bone healing either in combination or alone, through osteogenesis, osteoinduction, and osteoconduction.(3) Although autografts are the gold standard, allograft have been extensively used for the management of osseous defects with varying degree of success and are gaining new grounds.

Synthetic hydroxyapatite has been one of the most frequently used allograft material due to its chemical composition, which is similar to human bone(1). It has been proven that porous hydroxyapatite has excellent bone conductive property which allows the growth of osteogenic cells from existing bone surfaces into adjacent bone graft material.(4) It is non- toxic and has a high chemical stability along with less inflammatory and antigenic reaction. Another important property is that its microstructure can be controlled to promote the formation of pores that can allow the migration of blood vessels and bone tissues into the material. Synthetic hydroxyapatite has been used as a graft material with varying degrees of success. This material has the property of osteoconduction.

Autologous platelet concentrates such as platelet rich plasma (PRP) and platelet rich fibrin (PRF) are widely used for superior wound healing. Platelet Rich Fibrin (PRF) blood-derived and autogenous living biomaterial, has been and is increasingly being investigated and used by clinicians as an aid to promote both hard and soft tissue healing and regeneration.(5) PRF, a second generation platelet concentrate, is known to have a more sustained release of growth factors. Its simplified processing technique with minimal biochemical blood handling as compared to platelet rich plasma serves as an advantage over other blood products. PRF in the form of a platelet gel can be used in conjunction with bone grafts, which has several advantages, such as promoting wound healing, bone growth and maturation, wound sealing and haemostasis, and imparting better handling properties to graft materials(6). Evidence regarding the effect of PRF when used along with hydroxyapatite on the density of the bone formed and also its effect on post-operative complications has been sparse and hence this study has been taken up.

Material and Methods

The study was conducted in the Department of Oral and Maxillofacial Surgery, A J Institute of Dental Science, Mangalore from January 2017 to May 2018, 80 patients with mandibular 3rd molar impactions were selected for the study provided they met the inclusion critera.

Inclusion criteria

1. Impacted mandibular 3rd molar definitely diagnosed for removal
2. Subjects of both sexes
3. Age group 18 to 35 years

Exclusion criteria

1. Patients with localized infection.
2. Patients with any pathology in the desired area.
3. Known allergy to lignocaine
4. Systemic disease contraindicating tooth extraction.
5. Pregnant females or lactating mothers.

Materials

- Diagnostic instruments
- Dis-impaction kit
- 10 ml syringe
- Vacutainer – Red
- Centrifuge machine (REMI R-8)
- Synthetic hydroxyapatite (G- Bone)

Following institutional ethical clearance and after explaining the whole procedure, an informed consent was obtained from the patient participating in the study after all routine investigations. 80 patients were randomly divided equally into two groups of 40 each, group 1 and group 2.

Group 1: patients who were grafted with Hydroxyapatite.

Group 2: patients who were grafted with PRF and Hydroxyapatite.

Fig. 1a: Fractions of blood in vacutainer.

All the patients had a pre-operative OPG to evaluate the impacted tooth. Surgical Extractions was done by the same surgeon in both the groups. The extraction was carried out under standard aseptic conditions. Inferior alveolar nerve block was given, incision placed (according to the type of impaction and the surgeon's preference) and the flap was raised, bone removal and extraction was done based on the type of impaction. An OPG was taken after the removal of tooth and before placement of the graft. In group 1 following the extraction OPG was taken, the socket was then grafted with only hydroxyapatite. Wound closure was done using 3-0. In group 2 the preparation of the graft was done, cubital vein was used to draw 10ml of blood from the patient. The blood was then transferred to a vacutainer/test tube which was kept in the centrifuge machine. This centrifuge machine was maintained at a speed of 3000rmp for 10 minutes, this procedure separates the blood into three fractions the lower fraction containing red blood cells, the middle fraction containing the fibrin clot and the upper fraction is the straw-colored cellular plasma. (Figure 1a)

The middle fraction is the PRF which was used. (Figure 1b) This is separated from the blood using a scalpel; it is mixed with hydroxyapatite and grafted into the socket. Following extraction OPG was taken and then the prepared graft was placed in the socket. Regular post extraction instruction was given to the patient. Routine antibiotics and analgesics were prescribed. An assessment sheet for pain was given to the patient VAS for pain, which was assessed by the patient on day 1, day 2 and day 3 and 7th post-operative day. Intraoral sutures were removed, following a through clinical examination. At the 3rd month and 6th month (post – operatively), an OPG was taken bone density of the extraction sockets was measured at four random areas where graft was placed within the extraction socket through 'densitometric analysis' software in the OPG program (Kodak 8000C Digital Panoramic System, Eastman Kodak Company). From the four obtained values, an average value was recorded through the software.

Results

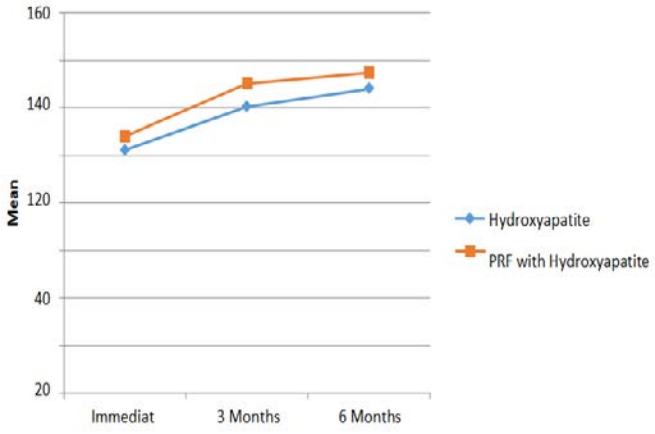
Our study which included 80 patients –

Group I – 40, socket grafted with hydroxyapatite (G-bone)

Group II – 40, socket grafted with PRF and hydroxyapatite (G-bone), yielded the following results.

In both Hydroxyapatite group and PRF with Hydroxyapatite group, majority of subjects were in the age group 21 to 30 years. There was no significant difference in age distribution between two groups. In both the groups, majority of subjects were males. There was no significant difference in sex distribution between two groups. The type of impaction as per radiographic analysis showed that most of the third molar 53.8% (43) were mesioangular impactions, followed by distoangular 25% (20), vertical 13.8% (11) and horizontal 7.5% (6). The visual analogue scale was used to assess pain on the 1 st, 2 nd and the 3 rd post-operative day. This was compared between the group I and group II using paired t test. The test revealed no significance between the pain between the two groups. But it was also noted that the pain in both the groups was significant when compared to the days within the group itself, which indicated the pain was reducing from the 1 st to the 3 rd post – operative groups irrespective of the group and yielded a highly significant p value. This was assessed using ANOVA test. Bone density was measured by digital orthopantomogram (OPG) (in pixels), orthopantomogram (OPG) images were taken intraoperatively, just after extraction, before the graft placement in Group I (hydroxyapatite) & group II (hydroxyapatite with PRF) and postoperatively at the end of third month and sixth month. The mean density immediately in group I was 102.4 and in group II was 108.2. At the end of the third post – operative month the mean density in group I was120.6 and in group II was130.3. And at the end of the 6 th post – operative month group I was 128.1 and in group II was 134.9. (Graph 1). In the study there was no significant difference in mean Densinometric analysis readings at baseline between two groups. But there was significant difference in mean Densinometric analysis readings at 3 months and 6 months b/w 2 groups. Mean Densinometric analysis readings was significantly high in PRF with Hydroxyapatite group compared to Hydroxyapatite group.

Fig. 1b: Platelet rich fibrin.



Graph 1: Densinometric analysis of OPG between groups.

Discussion

The success of bone formation in bone grafts is attributed to the following Osteogenetic cells to facilitate bone reconstruction, Osteoinductive factors to induce bone formation and Osteoconductive matrix to stimulate

bone deposition. Although osteoconductive materials have no potiential to induce bone formation, they do act as an interconnected scaffold that is biocompatible. Thus, this osseous tissue can be used for the regeneration of bone. On the other hand, osteoinductive materials promote new bone formation by allowing cells in the adjoining area to undergo transformation in order to form osteoprogenitor cell types which are capable of bone formation. Osteogenic material is a graft that has an inherent property of bone formation.(7) Various studies have been carried out in the past in order to evaluate the bone density of the grafted site. Standard radiography is the most common imaging method used to assess bone healing because its ease of availability and accessibility, it is economic and relatively safe.(8)(9) However, that assessment of bone healing by radiographic methods is subjective thus giving more scope for errors. Therefore, other imaging technologies and methods are being investigated that help quantify bone healing. But, these techniques are not economic, high maintaince and need extra manpower and also have radiation hazards.(10)

In this study we used the Densitometric Analysis software for the assessment of bone density by densitometric comparisons between digital orthopantogram (OPG) taken intra-operatively, just after extraction in) after extraction but before graft placement in the Group I (hydroxyapatite, G-bone) & Group II (PRF with Hrdroxyapetite) and postoperatively at the end of the third month and the sixth month. Bone density of the post- extraction sockets was measured at four random areas through ‘densitometric analysis’ software in the OPG program (Kodak 8000C Digital Panoramic System, Eastman Kodak Company) and an average value was recorded at each review.

The objective of this study was to radiologically assess and compare the regenerative potential of hydroxyapatite (G- bone) & PRF with hydroxyapatite from digital orthopantogram (OPG) using the Densitometric Analysis software and to evaluate the clinical usefulness of these materials to enhance soft tissue healing in third molar extraction sites. Both materials are indicated for a wide range of bone grafting procedures induvially as well as in combination.

A similar study was conducted by V. Panday et al in which they also used the same investigation procedure in order to determine the bone density, however in their study they had three groups which included 2 study groups comparing between the G-bone and G-graft and one control group.(1)

Another study which was conducted by Khalid et al who concluded that software analysis could be used to test the changes following bone augmentation procedures(10). Our results regarding the interpretation of bone density by using densitometric analysis were in agreement to this study.

There are a number of fabricated bone graft substitutes which based on hydroxyapatite or other calcium phosphate minerals that have been employed for these applications.

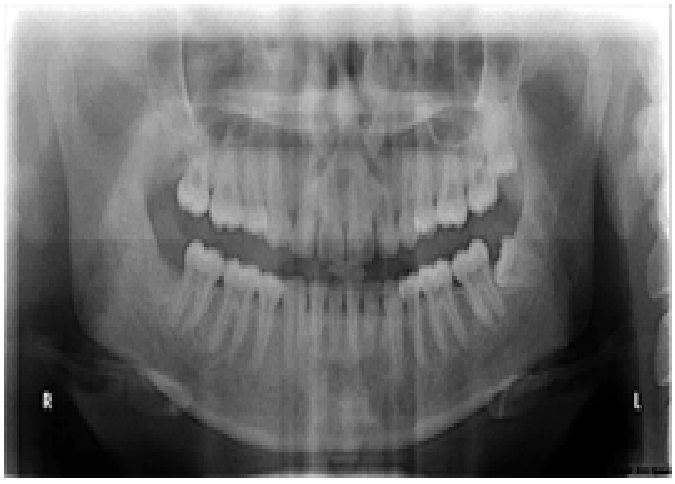


Fig. 2a: Immediate Post op OPG.

Fig. 2b: 6 months Post op OPG.

These materials are available either as porous or dense granules of various sizes and are similar to the natural mineral found in human bone in many ways. They are not only biocompatible but also have osteoconductive properties. They can be easily sterilized and are easy to be used in the clinic. The calcium phosphate materials exhibit a higher solubility whereas hydroxyapatite materials demonstrate a low solubility, and thus are thought of as resorbable bone graft materials.

The use of PRF along with an osteoconductive material such as hydroxyapatite or tricalcium phosphate or other calcium derivatives gives the graft material an added advantage of making the material osteoconductive in nature. This is what we have tried to analyze in our study.

PRF is the second generation of platelet concentrates. PRF contains various autologous cytokines, immune cells and various growth factors. In the oral and maxillofacial region, PRF has been widely used in sinus augmentation as a sole grafting material or in combination with other graft materials and bone substitutes.(11) Extraction socket preservation, intrabony defects, and periodontal problems are the other indications.(12) In our study we used PRF along with hydroxyapatite crystals to assess its role not only in soft tissue healing but also in hard tissue regeneration.

In our study we did not find any significant change in the degree of pain in the post – operative day 1,2 and 3 (which was assessed using VAS) between the two groups, group I (hydroxyapatite) and group II (PRF and hydroxyapatite). Although there was no difference in pain between the two groups there was a significant decrease in pain in both groups from the post – operative day 1 to day 3 in both the groups. A similar kind of study was conducted at the Ankara University by Uğur Gülşen et al on the effect of PRF on pain and edema following third molar extraction. In this study 30 patients with bilateral impactions were chosen and a split mouth randomized control study was carried out. This study concluded that there was no significant difference in edema or pain in the two groups(13) Pain although measured, is a subjective symptom. In this study we have not standerized the impactions or the indivuals and hence pain cannot be considered as the sole criteria to assess the role of PRF when used with the hydroxyapatite.

The bone density in our study as compared between the two groups, group I (G-bone) and group II (G-bone with PRF) showed that the density was more in group II as compared to group I. (Figure 2a,b) (Figure 3a,b) This indicated that using PRF, an autologous material along with an alloplastic material has an advantage over the latter. An osteoconductive material used alongside with an osteoinductive material.

The use of PRF during bone grafting provides the following four advantages.

✓ PRF plays an important mechanical role by maintaining and serving the grafted materials.

✓ the fibrin network at the regenerative site facilitates cellular migration, vascularization, and survival of the graft. (14)(15)

✓ The growth factors (PDGF, TGF-β, and IGF-1) are gradually released over a period of time as the fibrin matrix is resorbed, thus creating a continual process of healing.

✓ the presence of leukocytes and cytokines in the fibrin network play a significant role in the self-regulation of inflammatory and infectious phenomena within the grafted material

According to a study done by Nesligül Niyaz Kökdere, et al on PRF mixed with autogenous bone graft the defects with grafted with PRF presented far more osteoblasts and newly formed bone area values than the control group. In this study, PRF had a positive effect on bone formation after 8 weeks of grafting (16)

However in our study we saw positive bone formation by the end of 3 months, although both the groups showed positive results we observed that the density of bone formed was more in group II (G-bone with PRF) as compared to group I (G-bone). The mean density of group II was 130.36 at the end of the 3 months whereas it was 120.62 in group I with a highly significant p value <.001 this indicated that group II in which PRF was used showed faster bone formation as compared to the group I in which it was not used. In our study we followed up both the groups for 3 months and 6 months post – operatively, so the first incidence of bone formation was noted at the end of 3 months, but it could have been earlier than this. On the subsequent follow up on the 6 month the radiograph showed differences in the density of formed bone between the two groups with a high significant p value of <.001. In both the groups we noted areas or specks of radio-opaque regions indicating that some amount of graft material has still not completely formed bone. Literature shows that alloplastic materials such as hydroxyapatite when used alone takes nearly 6-8 months to completely dissolve and form bone. In our study there was no comprise of the alveolar height, although this was not quantified radiographic changes as such was not noted proving otherwise.

In oral and maxillofacial surgery applications, rehabilitation of the bone defects requires more than a 3-6-month interval after bone grafting. Thus, the growth factors that cause differences in osteogenic effects at early stages may have little or no effect in the long-term.

Fig.3a: immediate Post op OPG.

Fig.3b: 6 months Post op OPG.

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