Abstract:
Objectives: The aim of this study will be to evaluate the clinical and radiographic efficacy of platelet rich fibrin (PRF) membrane used with immediately placed dental implants in the esthetic zone.

Methods: A clinical trial was carried out on fifteen fresh extraction sockets in 7 males and 8 female’s patients with age range from 18 to 43 years, sockets implanted with immediate endosseous implant and grafted with deproteinized bovine bone mineral (DBBM) and PRF. After implant placement all implants were evaluated clinically and radiographically at baseline, 3 months, 6 months, 9 months and 12 months. Clinical (modified sulcus bleeding index mSBI, probing pocket depth PPD and degree of mobility using Periotest device) and radiographic parameters (marginal bone loss MBL) were assessed.

Results: During the 1-year interval, no implant was lost and all patients showed favorable clinical and radiographic finding. Periotest evaluation showed that all implants were well osseointegrated and stable. There was a statistically significant increase in PD and MBL through the whole study period (P value <0.001).

Conclusions: Immediate placement of implants with a PRF membranes could be considered a valuable option to replace a missing teeth. Integrating this technique into common practice could provide important benefits for the patients regarding aesthetics, without any risk of infection or transmission of diseases.

Keywords: Immediate implant, PRF, DBBM, periotest.

Introduction

Teeth replacement using dental implants has proven to be a successful and predictable treatment procedure; different placement and loading protocols have evolved from the first protocols in order to achieve quicker and easier surgical treatment times [1]. In situations where a tooth requires extraction and replacement, original protocol (gold standard) suggested a 6-12 month waiting period before implant placement [2]. The original protocol has been challenged within the last decade and new protocols have been developed in which implants are placed at the time of extraction of the tooth. This protocol where implants have been placed at the time of tooth extraction is known as immediate implants [3].

The benefits of immediate implant insertion after tooth extraction are elimination of post-extraction healing period, reduction of the number of surgical sessions, preservation of alveolar width and height, lower risk of dehiscences or fenestrations around dental implant, better angulation leading to improved esthetics and axial occlusal loading, and improved surgical orientation relative to pertinent anatomical structures [4]. These benefits are usually accompanied by a minor drawback as lack of adequate available bone apical to socket may compromise primary stability, site morphology may complicate optimal placement and anchorage, tension-free closure may be difficult to achieve in case simultaneous use of biomaterials is needed. Even, if achieved, it can lead to alteration of mucogingival junction, thin tissue biotype may compromise optimal outcome, difficulty in preparing the osteotomy due to bur movement (chatter) on the walls of the extraction site, and the added cost of bone grafting [5,6].

When an implant is placed in a fresh or recent extraction alveolus a gap between the implant surface and the bone walls of the socket may occur. The presence or size of the gap is both influenced by the configuration of the alveolus and by the design and width of the implant [7]. Wilson et al. [8] and Paolantonio et al. [9] demonstrated that for implants with a horizontal defect of 2 mm or less, spontaneous bone healing and osseointegration take place if the implant has a rough surface [8,9]. Horizontal defects in excess of 2 mm have been shown to not heal predictably with bone [7]. So To compensate for these problems, guided bone regeneration (GBR) using autografts, allografts, or alloplasts; barrier membranes; or combination therapy has been accomplished with what appear to be successful clinical results [10].

Choukroun’s Platelet-rich fibrin (PRF) has been recently proposed as an aid for promoting hard and soft tissue regeneration in the field of oral surgery [11]. It is a simple, natural and inexpensive technique for the production of leukocyte and PRF concentrates [12]. PRF is a consistent fibrin biomaterial and not improved fibrin glue from the platelet-rich plasma (PRP) family [13]. PRF releases high amounts of growth factors such as transforming growth factor B1 (TGF-B1), platelet-derived growth factor AB (PDGF-AB), vascular endothelial growth factor (VEGF) and matrix glycoprotein such as (thrombospondin-1) during at least 7 days in vitro [14]. Thus this biomaterial presents a specific biology which offers several advantages including promoting wound healing, bone growth and maturation.
graft stabilization, wound sealing and hemostasis, and improving the handling properties of graft materials. PRF can also be used as a membrane. Clinical trials suggest that the combination of bone grafts and growth factors contained in PRP and PRF may be suitable to enhance bone density [15,16].

The aim of this study is to describe a technique using PRF membranes technology for preservation of hard and soft tissue architecture around a postextraction implant immediately placed in esthetic zone.

**Patients and methods**

**Subjects**

Fifteen patients, 7 male and 8 female with an average age 25 years (range from 18 to 43), were included in this study. They are selected from the Outpatient Clinic in the Oral and Maxillofacial Surgery Department, Faculty of Dentistry, Mansoura University for extraction of a non-restorable maxillary anterior and premolar teeth (esthetic zone) and placement of a dental implant into fresh extraction sockets.

**Inclusion Criteria**

1. Presence of non-restorable maxillary anterior and premolar teeth due to trauma, caries, root resorption, root fracture, endodontic or periodontal failure.
2. Patient’s cooperation, motivation and good oral hygiene.
3. No Acute Infection is present.

**Exclusion Criteria**

1. Poor oral hygiene, traumatic occlusion, smokers and presence of parafunctional habits.
2. Presence of acute and chronic systemic disorders such as uncontrolled diabetes, hemorrhagic disorders and other conditions that can affect wound healing responses.
3. Extreme bone atrophy.

**Preoperative therapy**

Each case was evaluated through examination of diagnostic casts for intra-arch relationship, panoramic and periapical radiographs to evaluate the anatomic conditions (Fig.1). All patients were subjected to proper oral hygiene instructions, scaling and root planning for all teeth and periodontal treatment if needed to provide an oral environment more favorable to wound healing

**Platelet Rich Fibrin Preparation**

A convenient blood sample was collected from a peripheral venous blood from the patient in sterile 10 ml tubes without the addition of an anticoagulant and centrifuged at 3000 revolutions (400g) per minute for 10 minutes. PRF settles down between the platelet poor plasma (PPP) at the top and the red blood cells (RBC) at the bottom of the tube (Fig.2).

**Surgical Procedure**

Each patient was asked to rinse her mouth with 0.2% chlorhexidine solution. All the surgical procedures were performed under local anesthesia and strict aseptic conditions. Intrasulcular incisions were made to raise full thickness mucoperiosteal flap and the teeth were carefully removed by a gentle extraction using forceps in order to protect and to preserve the alveolar bone.

Sockets were curetted and irrigated with saline to remove granulation tissue and residual periodontal ligament (Fig.3). Drilling was performed at 600-800 rpm at the accurate direction guided by the surgical drill guide. Sequential drilling with copious irrigation was carried out till the desired dimensions were achieved depending on the selected implant (Dentium® System, Superline, Seoul, Korea). Manual key and ratchet were used for implant insertion. The implants were placed 2-3 mm beyond the apex to achieve primary stability. The cover screw was placed on the top the implant (Fig.4).

The residual gap between the socket wall and implant threads was grafted with DBBM (Biogen®, Xenographic mix granules, Bioteck S.p.A, Italy) and PRF membrane was positioned and stabilized over the graft (Fig.5, 6). Prior to wound closure with simple interrupted suture, releasing incisions were performed to allow better flap adaptation around implant neck (Fig.7). Then, an immediate post-operative x-ray was done (Fig.8).

**Post-operative care**

Amoxicillin 500mg every 8 hours was continued for 5 to 7 days post-surgery. Instruction for good oral hygiene measures was also given. Further advices included adhering to a soft diet and avoiding trauma to the gingival tissue at the implant site especially in the first few weeks. Sutures were removed after 7 days and patients were examined every week during first 3 weeks following surgery then monthly until termination of study.

**Prosthetic procedure**

4-6 months later, under local anesthesia, the surgical covering screw was exposed and removed then healing cap was then placed for 2 weeks. After that, Impression was made with the aid of impression post and laboratory analogue using silicone rubber base material to fabricate working cast then the final restoration of porcelain fused to metal was fabricated and cemented on the abutment.

**Clinical evaluations**

The following parameters were evaluated at BL (after implant loading), 3, 6, 9 and 12 months of implant insertion.

A-Implant stability: was measured by Periotest instrument (Periotest M, Medizintechnik Gulden, Germany). The readings were correlated with a grading scale provided by manufactures of Periotest instrument

B-Modified sulcus bleeding index(17): at four aspects around the implants: score 0, no bleeding when a periodontal probe is passed along the gingival margin adjacent to the implant; score 1, isolated bleeding spot visible; score 2, blood forms a confluent red line on margin; and score 3, heavy or profuse bleeding.

C-Peri-implant pocket depth (PDP): The distance between the base of the pocket and the gingival margin was measured using a graduated probe. The probe was inserted at 6 sites around each implant in a line with the vertical axis of the implant until the blunt edge of the probe contact the base of the pocket.

D-Radiographic evaluation: Intra-oral periapical radiographs were taken using parallel long cone technique with Rinn XCP (DENTSPLY Friadent Schweiz, Nidau, Switzerland.) film holder and custom- fabricated bite blocks. A digital tracing for the implant body and alveolar crest was performed
by the aid of Scanora 5.2 software program for periapical radiograph. The analysis tools were used to measure the distance from the abutment-implant connection to the marginal bone level in millimeters using the implant length as a reference for judgment for the correctness of the measurements. The known length of the implant (measured from the implant-abutment connection to the implant apex) according to the manufacturer was used as reference point. The distance from implant-abutment connection to marginal bone level was measured on the magnified images. To account for variability, the true length of the implant and the length of the implant on the magnified radiograph were used as a correction factor for the magnification to calculate the length of marginal bone levels changes at the distal and mesial surfaces of all implants on the radiographs in millimeters.

**Statistical Methods**

Numerical data were explored for normality using Kolmogorov-Smirnov and Shapiro-Wilk tests. Data showed non-parametric distribution. Data were presented as mean, median, standard deviation (SD) and range values. Friedman’s test was used to study the changes by time within each variable. Wilcoxon signed-rank test was for pair-wise comparisons when Friedman’s test is significant. The significance level was set at $P \leq 0.05$. Statistical analysis was performed with IBM® (IBM Corporation, NY, USA) SPSS® Statistics Version 20 for Windows (SPSS, Inc., an IBM Company).

**Results**

**Clinical findings**

The study was conducted on 15 subjects; 7 males (46.7%) and 8 females (53.3%). The mean and standard deviation values for age were 26.3±6.2 years. The patients attended the follow-up recall till the end of the study period (12 months). All patients showed no postoperative inconveniences and all implants showed successful signs of osseointegration with no signs of failure (soft tissue dehiscence, infection, looseness of the implant).

The implant lengths used in this study were 12 mm (33.3%) and 14 mm (66.7%). Moreover, the most commonly used implant diameter used in this study was 3.6 mm (46.7%) and 4.0 mm (46.7%), followed by 4.5 mm (6.6%). Implant stability and the periodontal indexes are reported in Table 1.

Periotest scores showed that the mean value were decreasing with time of measurement where it was -2.7±1.3 at baseline and decrease to -2.4±1.0 at three months with statistically significant difference and became -2.3±0.9, -2.1±1.0 and -2.2±0.9 respectively with no statistically significant difference. (mSBI) values showed that the mean value were not statistically significant through the whole study period where it was 0.3±0.3 at baseline and became 0.4±0.4 at 12 months. (PPD) values showed that the mean value were increasing with time of measurement where it was 2.2±0.4mm at baseline and increase to 2.4±0.3mm at 3 months with statistically significant difference and became 2.5±0.4mm, 2.5±0.4mm, and 2.6±0.5mm at 6, 9, and 12 months respectively with no statistically significant difference.

**Radiographic finding**

The periapical radiographs taken for all implants revealed no signs of continuous peri-implant radiolucency, at the postoperative radiographic examination, the mean MBL was 0.8±0.5 mm at base line and increased to 1.0±0.5mm at 3 months with statistically significant difference and became 1.0±0.5mm, 1.1±0.5mm, 1.1±0.6mm at 6, 9, and 12 months respectively with no statistically significant difference. (Table 2).

**Discussion**

The biologic advantage of immediate implant is the contention that the implant will prevent postsurgical bone resorption seen following tooth extraction as a normal part of the socket healing [18,19]. It also eliminates the waiting period for socket consolidation, has fewer surgical sessions, a shorter total treatment time, reduced over-all costs [4,18].

Membranes either resorbable or non-resorbable with or without bone grafting have been used for many years in guided tissue regeneration (GTR) to heal periodontal defects and to regenerate peri-implant defects in GBR [20]. Because the membrane is generally expensive and has a possibility of disease transmission, a product from the patient’s own blood has gained much popularity as a supplement to procedures of tissue regeneration [21].

Choukroun PRF may be a good source. It is autogenous and is not associated with any issues related to immune reactions or infections. Besides, its gelatinous consistency enhances clot and graft stability [22]. Platelets release cytokines, such as platelet-derived growth factor (PDGF), insulin-like growth factor (IGF), and vascular endothelial growth factor (VEGF). PDGF and IGF are well known osteogenic cytokines [23]. Choukroun PRF is known to release growth factors for at least 7 days [14].

Apart from these advantages, PRF allows to avoid the use of membranes and barriers, thus reducing the risk of possible exposure to the oral cavity and of the consequences that the bacterial contamination may have on the regenerative process [24]. The present study was conducted to evaluate the clinical and radiographic efficacy of platelet rich fibrin (PRF) membrane used with immediately placed dental implants in the esthetic zone. All the implants were successfully osseointegrated over a one year follow-up period with a success rate of 100% with insignificant change in the crestal bone level.

The higher success rate which was noticed in the current study was probably attributed to the smaller sample size or the strict case selection. All implants were totally submerged and loaded after 6 months in this study as two stages implant type keep the implant out of function and reduce the effect of micro movement during the healing period to gain osseointegration, this is in agreement with Severson et al. [25]. The rational for reflecting full thickness mucoperiosteal flaps in this study was threefold; first, it facilitates tooth removal, which can be quite delicate, especially when the tooth is fractured or in case of root resorption. Second, a flap allows the clinician to inspect the buccal socket wall properly for fenestrations and dehiscencies. Third, flapless surgery increases the risk of perforation. This is in accordance with Misch [26] who reported that, the full thickness flap allows preservation of

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the delicate soft tissue from laceration and subsequent infection [26].

Regarding implant stability, our study declared that, the mean average periotest values (PTV) were -2.7±1.3, -2.4±1.0, -2.3±0.9, -2.1±1.0, and -2.2±0.9 at baseline, 3, 6, 9 and 12 months respectively. There was a significant difference between the mean (PTV) from baseline to 3 months but the difference was insignificant thereafter from 3 to 12 months. This was in agreement with Lorenzoni et al. [27] who showed an ankylosis healing and successful maintenance of osseointegration of all implants.

The diminution in (PTV) may be attributable to an increased incidence of loosening of superstructures and screws. This was seen so agreeably with the results of Gomez-Roman and his colleagues [28] who found similar finding and concluded that an increased Periotest value over time, therefore, is a reason to check the fastening screw loosening.

Within limitation of the study and short period of periotest evaluation, the records are closely to each other which is insensitive to detect the bone turnover or stability changes. Regarding to Modified sulcus bleeding index (mSBI) as a clinical indicator for absence or presence of inflammation, there was no suppuration detected during the follow up period. Decreased incidence of gingival inflammation can be attributed to the application PRF membranes as it behaves like a true fibrin tissue which considered as an optimized blood clot with platelet growth factors that can improve the vascularization of the surgical site, promoting neoangiogenesis and decreases the wound healing time [24].

During assessment of peri-implant probing depth (PPD), our study declared that, the mean (PPD) were 2.2±0.4mm, 2.4±0.3mm at baseline, 3 months respectively and 2.5±0.4mm for both 6 and 9 months and 2.6±0.5mm at 12 months. Through the whole study period (Baseline to 12 months); there was a statistically significant increase in PPD.

Increased incidence of (PPD) values can be attributed to reflecting a full thickness mucoperiosteal flaps in this study which leads to a more apically positioned junctional epithelium which is directly related to an increased probing depth around the implant [29]. Besides large open wounds heal slowly and with significant scarring due to impairment in amount of vascular structures in the peri-implant mucosa [30].

During assessment of marginal bone level (MBL), our study declared that, the mean (MBL) were 0.8±0.5mm, at baseline and 1.0±0.5mm at 3, 6, and 9 months respectively and 1.1±0.6mm at 12 months. Through the whole study period (Baseline to 12 months); there was a statistically significant increase in MBL.

In our study, the bone loss occurred may be a result of irritation from plaque accumulation, natural bone remodeling around the implant as a sequel for placement of final prosthesis that may be associated with increased load and in turn increased transferred stress on bone implant interface [31,32]. Similarly, it may be attributed to subcrestal placement of implant by 1mm [33].

The bone implant contact ratio was stable throughout the observation period. This may be attributed to the biologic properties of PRF which utilize the potential of several autologous platelet-derived growth factors (PDGF, TGF-β, VEGF, IGF-1, FGF) obtained with a simple centrifugation procedure, to specifically stimulate several biologic functions such as chemotaxis, angiogenesis, proliferation, differentiation, modulation, thereby representing an effective therapeutic device for more rapid and effective regeneration of hard and soft tissues [23,34].

To the best of our knowledge, there have been no studies reported to assess the combined application of PRF membranes in conjunction with immediate implant placement. We believe that, such combined technique could potentially improves implant bed preparation and motivates osseointegration and decrease overall healing time. This hypothesis needs to be confirmed by long-term studies using cone-beam computed tomography (CBCT) because this new radiologic technique is able to visualize and assess the facial bone wall at implants with excellent resolution and low radiation exposure.

**Conclusion**

From this study, we could conclude the following:

1. PRF membranes represents a new technology for stimulation and acceleration of bone regeneration.
2. Integrating the present technique into common practice could provide important benefits for patients in terms of esthetics without any risk of infection or transmission of diseases as PRF membranes are autologous source of growth factors, prepared by simple technique, minimal cost and minimal invasive.

Addition of PRF to deprotenized bovine bone may be a useful adjunct for alveolar ridge augmentation around immediate implant placement.

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<tr>
<th>Table 1: Implant stability and the periodontal indexes recording at different time intervals (BL, 3, 6, 9, &amp; 12 months).</th>
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<td><strong>Base line</strong></td>
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<td><strong>Periotest values</strong></td>
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<td><strong>mSBI values</strong></td>
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<td><strong>PPD values</strong></td>
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<sup>a</sup>: Significant at P ≤ 0.05, Different superscripts are statistically significantly different.
Figure 1: Preoperative view.

Figure 2: Drawn blood centrifuged.

Figure 3: Extraction socket after debridement.

Figure 4: Implant installation.

Figure 5: Bone substitute application.

Figure 6: PRF membranes application.

Figure 7: Flap sutured.

Figure 8: Immediate postoperative x-ray.

Figure 9: Final restoration at 12 months.
Table 2: Marginal Bone Level recording at different time intervals (BL, 3, 6, 9, & 12 months.

<table>
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<tr>
<th></th>
<th>Base line</th>
<th>3 months</th>
<th>6 months</th>
<th>9 months</th>
<th>12 months</th>
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<tr>
<td>Mean ± SD</td>
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<tr>
<td>Median</td>
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<td>1.0 ± 0.5</td>
<td>1.0 ± 0.5</td>
<td>1.1 ± 0.5</td>
<td>1.1 ± 0.6</td>
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<td>Range</td>
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<td>0.8</td>
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* Significant at P ≤ 0.05, Different superscripts are statistically significantly different

References


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