Assessment of outcome of single implants in the anterior maxilla following Standard Implant Treatment, Immediate Implant Treatment, and Implant Treatment in conjugation with Guided Bone Regeneration

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Abstract
The present study was aimed at documenting the outcome of single implants in the anterior maxilla. following three treatment modalities- Standard Implant Treatment (SIT), Immediate Implant Treatment (IIT), and Implant Treatment in conjugation with Guided Bone Regeneration (IGBR), all using the same implant system and biomaterials. In this study 30 subjects who required single implants in anterior maxilla were selected. Implant sites were examined and patients were divided into three groups; GROUP A- Standard implant treatment (SIT) was performed in 10 patients, GROUP B- Immediate implant treatment (IIT) was performed in 10 patients, and GROUP C- Implant treatment in conjugation with guided bone regeneration (IGBR) was performed in 10 patients. Clinical parameters included Probing depth, crestal bone levels, bleeding on probing, plaque score, implant stability and aesthetic evaluation. The aesthetics was in favour of Standard Implant Treatment cases, followed by Immediate Implant Treatment cases. They were least favourable in cases with GBR. Reduced papillae were a frequent finding in GBR cases. On the other hand, it is of the utmost importance from a clinical point of view to know whether comparable outcomes are realistic following complex (GBR/BGR) and innovative (IIT) treatment concepts in reference to a standard approach (SIT). After all, the primary expectation will be a – close to – perfect restoration and any pre-existing limitation to accomplish that goal may be of secondary concern for the patient. All the three treatment modalities used in our study were predictable from a clinical and radiographic point of view with Standard Implant Treatment cases giving best results followed by cases with Immediate Implant Treatment. However, GBR increased the risk for complications and compromised aesthetics.

Key words: Implant, Guided bone regeneration, Immediate implant

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Introduction
A number of techniques are available for the rehabilitation of the single – tooth space. The common techniques involved are conventional fixed prosthetics, removable partial dentures and, in some patients, orthodontic treatment. These methods, however, come with a set of disadvantages, such as loss of tooth substance, and a potential loss of tooth vitality, especially in young patients. In addition, the prognosis for reconstruction can be complicated by carious lesions progression, periodontal disease of abutment teeth and / or technical failures such as loss of retention and fracture of bridge components or abutment teeth etc.[1] The application of dental implants for single-tooth replacements has evolved into a viable alternative to conventional fixed bridgework, resin bonded restorations or removable partial dentures.[2,3] Ever since dental implant therapy evolved as a treatment option for replacement of missing teeth, the success of this treatment modality was defined as Osseointegration.[4]

Original protocols in implant dentistry advocated a healing period for implant of 4 to 6 months as a prerequisite for Osseointegration. In recent years, treatment protocols have been modified to shorten treatment time and to improve patient comfort. As a result, immediate implantation, which is the placement of implants directly into the socket immediately after extraction, has become widespread because it eliminates the delay required to allow for socket healing prior to fixture placement. Another predictable therapeutic modality appears to be immediate implant insertion in extraction sockets in conjunction with barrier membranes.[5]

With the predictability of Osseointegration of the current implant surfaces, clinicians find themselves focusing their efforts on the aesthetics of the restoration in order to have successful and acceptable outcomes. Single-tooth replacement by means of an implant-supported restoration has become a viable treatment option.[6] Still, achieving an aesthetically optimal result is not self-evident as this is determined by tooth-related factors such as tooth dimensions, form and colour, that contribute to aesthetics, as well as soft tissue-related factors including inter-dental and midfacial soft tissue dimensions, texture and colour. Recent findings suggest that extensive resorption of intact buccal plates is a common phenomenon following tooth removal. [7] In addition immediate implant placement has no impact on this remodelling process, making it a potentially risky procedure for aesthetic failure if patients are not well selected. Clearly, the vast majority in clinical practice do not qualify for such an approach. In these patients, early implant placement, usually combined with guided bone regeneration (GBR) in the aesthetic zone, can be pursued.[8]

Albeit successful results have also been described for various cases,[9,10] it is difficult to compare the outcome of aforementioned treatment concepts based on the available literature. Any conclusion in this regard may be significantly skewed due to heterogeneity in the healthcare providers, implant systems, biomaterials, and follow-up. In addition, aesthetic aspects of treatment outcome have been underexposed to research.[11]

Hence, the present study was conducted to document the outcome of single implants in the anterior maxilla following above mentioned treatment modalities when performed by using the same implant system and biomaterials. The proof for aesthetic outcome was studied from the pictures of maxilla and was based upon a 2-stage surgical protocol with a healing period of about 4 months. The study supported the traditional concept that, since the initial wound healing period is critical, loads applied prematurely to implants may jeopardize initial stabilization. As a result, a minimum waiting period of 4 months in the maxilla was advocated prior to applying any load to an implant.[12]

Aim of the study was to document the outcome of single implants in the anterior maxilla following three treatment modalities,

1. Standard Implant Treatment (SIT),
2. Immediate Implant Treatment (IIT),
3. Implant Treatment in conjugation with Guided Bone Regeneration (IGBR),

using the same implant system and biomaterials.

Materials & Methods

For this proposed study, a total of 30 patients were selected from the Out Patient Department of Periodontics and Implantology, Swami Devi Dyal Hospital and Dental College, Barwala (Panchkula) Haryana, India, who got implants placed in the department. An ethical approval for the study was obtained from the ethical committee of the institution. Each patient was given a detailed verbal and written description of the study and all the selected patients were required to sign an informed consent form prior to commencement of the study. In this study 30 subjects in which the implants were to be placed were selected. Implant sites were examined and patients were divided into three groups

GROUP A- In this group, standard implant treatment (SIT) was performed in 10 patients.

GROUP B- In this group, immediate implant treatment (IIT) was performed in 10 patients.

GROUP C- In this group, implant treatment in conjugation with guided bone regeneration (IGBR) was performed in 10 patients.

Group A: Under local anaesthesia with 2% lidocaine (1:200,000 epinephrine) in group A subjects, a standard mucoperiosteal flap was elevated following sulcular incision at both teeth facing the single-tooth gap and a palatally oriented crestal incision. Vertical releasing incisions were not made. Thereupon, all patients received a commercially available implant that was positioned according to surgical module. Sites were underprepared to ensure primary implant stability and non-submerged healing.

Group B: Under local anaesthesia with 2% lidocaine (1:200,000 epinephrine) in group B subjects, mucoperiosteal flap was elevated fully reflecting the papillae, yet without vertical releasing incisions. Following tooth removal using periotome, patients received commercially available implant hereby mainly engaging the palatal wall. A correct three-dimensional positioning of the implant was done. Sites were underprepared to ensure primary implant stability of at least 35 Ncm.

Group C: Under local anaesthesia with 2% lidocaine (1:200,000 epinephrine) in group C subjects, a wide mucoperiosteal flap was elevated following sulcular incision at both teeth facing the single-tooth gap and a palatally oriented crestal incision. Two vertical releasing incisions were made at the buccal paramedian aspect. Thereupon, all patients received a commercially available implant. Multiple bone perforations were performed in the buccal bone wall and the periosteum was released. In all cases, a synthetic bone graft (DM Bone) covered the buccal aspect of the ridge. Two layers of a collagen membrane were given to stabilize the grafting material. The GBR technique makes a two-stage procedure inevitable. Thus, tension-free primary wound closure was pursued in all patients by means of single sutures.

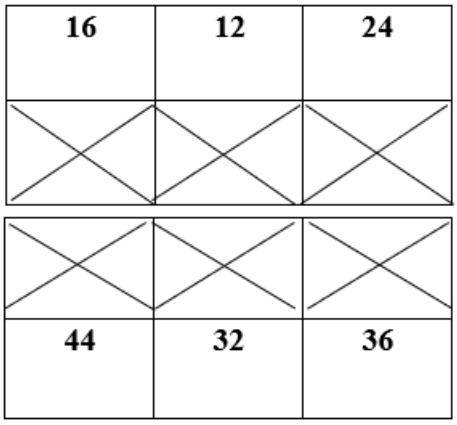
In all the groups, sutures were removed after 1 week and the second stage procedure was initiated after 4 months in the maxilla, after implant installation.

CLINICO-RADIOGRAPHIC EVALUATION

PARAMETERS

A) **Plaque Score-** Plaque Index (Silness & Loe, 1964)[13] was calculated at four sites per tooth and implant (mesial, midfacial, distal, and palatal).

PLAQUE INDEX (PI) (Silness & Loe, 1964)



B) **Probing Depth-** at Loading Stage, 3rd and 6th month following Prosthesis. Probing Depth was recorded at buccal, palatal, mesial and distal aspects of the placed implant with the help of graduated plastic periodontal probe and mean Probing Depth (PD) was calculated.

C) **Bleeding on Probing-** Modified Sulcular Bleeding Index (mSBI) (Mombelli et al, 1987)[14] at 3rd and 6th month after Loading Stage

MODIFIED SULCULAR BLEEDING INDEX (mSBI)
(Mombelli et al, 1987)

SCORE	INTERPRETATION
0	No bleeding when a periodontal probe is passed along the gingival margin
1	Isolated bleeding spots visible
2	Blood forms a confluent red line on margin
3	Heavy or profuse bleeding

D) **Aesthetic Evaluation-** The Pink Aesthetic Score (Fürhauser et al, 2005)[15] was used to evaluate the aesthetic outcome of the peri-implant soft tissues. Seven variables namely mesial papilla, distal papilla, midfacial level, midfacial contour, alveolar process deficiency, soft tissue colour, and soft tissue texture, are included in this index. The prostheses were photographed and the seven variables were evaluated versus a natural reference tooth at 1st, 3rd, and 6th month after Loading Stage.

Pink Aesthetic Score (PES)
(Furhauser et al 2005)

Variables	**0**	**1**	**2**	**Score Assigned**
Mesial papilla	Absent	Incomplete	Complete	
Distal papilla	Absent	Incomplete	Complete	
Level of soft-tissue margin	Major discrepancy > 2mm	Minor discrepancy 1–2mm	No discrepancy < 1mm	
Soft-tissue contour	Unnatural	Fairly natural	Natural	
Alveolar process	Obvious	Slight	None	
Soft-tissue color	Obvious difference	Moderate difference	No difference	
Soft-tissue texture	Obvious difference	Moderate difference	No difference	

E) **Height of Crestal Bone**- was recorded at baseline (at the time of Implant placement), 4th month, and 6th month using parallel cone technique for standardisation. Radiographic evaluation was done using X-ray grid mesh. Radiographic images were standardized and distance between the fixture shoulder and apical level of the marginal bone that is in contact with the implant was measured using implant height for calibration.

F) **Implant Mobility**-was recorded at Loading Stage, 3rd and 6th month after Loading Stage. To measure the implant mobility following Loading Stage, the Mobility Index of the endosseous implants developed by Wasserman[16] was used, which is a modification of the Miller Index[17] of horizontal tooth mobility. Quantification of the numerical index was assigned according to the subjective perception of the examiner considering the degree of mobility. The implant examined was lightly tapped with the handles of two dental instruments in a buccal-lingual direction and the degree of movement relative to adjacent teeth was observed. The implant was also tested for intrudability by applying pressure in an apical direction.

Mobility was scored as follows:

1. **Normal.**
2. **Slight mobility**—less than approximately ¾ mm of movement bucco-lingually.
3. **Moderate mobility**—up to approximately 2 mm of movement bucco-lingually.
4. **Severe mobility**—more than 2 mm of movement.

A tooth which was intrudable was given a score for its measured mobility plus one. All examinations were done by one examiner.

All measurements were made at the mesial and distal aspects of each fixture and the mean for each case was calculated.

DATA COLLECTION: All the above mentioned parameters were collected and subjected to statistical analysis using SPSS 21.0 (Armonk, NY: IBM Corp)

RESULTS

A study was undertaken between stage I implant surgery and stage II implant surgery in 30 subjects, and up to six months after final implant prostheses placement for single tooth implants.

Subjects were divided into 3 groups:

GROUP A- In this group, standard implant treatment (SIT) was performed in 10 patients.

GROUP B- In this group, immediate implant treatment (IIT) was performed in 10 patients.

GROUP C- In this group, implant treatment in conjugation with guided bone regeneration (IGBR) was performed in 10 patients

In all the groups, clinical and radiographic parameters including Plaque Score, Bleeding on Probing, Height of Crestal Bone, Implant Mobility, Probing Depth and Pink Aesthetic Score were recorded at different time intervals as per the preliminary time table determined for each subject. The results were compiled and statistically analysed using SPSS Version 21 Version. The data was found to be non-parametric for the all the six parameters – Plaque Score, Bleeding on Probing, Height of Crestal Bone, Implant Mobility, Pink Aesthetic Score and Probing Depth. The level of significance was fixed at $p \leq 0.05$.

PROBING DEPTH SCORE (Table 2 & 3) (Graph I & II)

Standard Implant Treatment (Group A):

Group A had mean Probing Depth scores of 1.60±0.52 mm at the baseline. Baseline for Probing Depth index was at prosthesis placement. The score increased at 3rd month (1.8±0.63mm) and 6th month interval (1.9±0.62 mm). There was absolute change of -0.2±0.57 mm at 3rd month and -0.3±0.53 mm at 6th month interval. The difference between baseline and 3rd month was statistically significant ($p<0.05$) whereas at the 6th month the difference from the baseline was also statistically significant ($p<0.05$).

Immediate Implant Treatment (Group B):

The mean Probing Depth scores was 01.66±0.49 mm at baseline, 1.82±.53mm at 3rd month and 2.3±0.49 mm at 6th month interval. The absolute change at the 3rd month interval and 6th month interval was -0.16±0.48 mm and -0.64±0.67 mm respectively. The difference between baseline and 3rd month was statistically significant ($p<0.05$) whereas at the 6th month the difference from the baseline was also statistically significant ($p=0.001$).

Implant Treatment in conjugation with Guided Bone Regeneration (Group C):

In Group C, mean Probing Depth score was 1.70±0.22 mm at the baseline. The score increased at 3rd month (1.83±0.32mm) and 6th month interval (2.86±0.55 mm). There was absolute change of -0.13±0.63 mm at 3rd month and -1.16±0.85 mm at 6th month interval. The difference between baseline and 3rd month was statistically significant ($p<0.05$) whereas at the 6th month the difference from the baseline was also statistically significant ($p<0.001$).

Comparison between Group A, Group B, and Group C:

The probing depth score for the Group A was 1.60±0.52 mm at the baseline, 1.80±0.63 mm at 3rd month and 1.90±0.62mm at 6th month interval. For Group B the respective values were 1.66±0.49 mm (at baseline), 1.82±0.53 mm (at 3rd month), and 2.30±0.49 mm (at 6th month). In Group C, values were 1.70±0.22 mm (at baseline), 1.83±0.32 mm (at 3rd month), and 2.86±0.55 mm (at 6th month). When the inter group comparison was made, there was statistically non-significant difference in the Probing depth scores between the Group A, Group B, and Group C, at all the intervals ($p>0.05$), analysed using ANOVA.

HEIGHT OF CRESTAL BONE (Table 4 & 5) (Graph III & IV)

Standard Implant Treatment (Group A):

Group A had a mean height of crestal bone of 0.57±0.21 mm at the baseline. The score increased at loading stage (1.30±0.00 mm) and 6th month (1.40±0.31 mm). There was absolute change of -0.73±0.21 mm from baseline to loading stage and -0.83±0.19 mm from baseline to 6th month interval. The difference between baseline and loading stage was statistically significant ($p=0.001$) whereas at the 6 months interval also, the difference from the baseline was statistically significant ($p<0.001$).

Immediate Implant Treatment (Group B):

Group B had a mean height of crestal bone of 0.49±0.28 mm at the baseline. The score increased at loading stage (1.20±0.24 mm) and at 6th month (1.45±0.28 mm). There was absolute change of -0.71±0.31 mm from baseline to loading stage and -0.96±0.39 mm from baseline to 6th month. The difference between the baseline and loading stage was statistically significant ($p<0.05$) whereas at the 6 months interval also, the difference from the baseline was statistically significant ($p<0.001$).

Implant Treatment in conjugation with Guided Bone Regeneration (Group C):

Group C had a mean height of crestal bone of 0.55±0.50 mm at the baseline. The score increased at loading stage (1.15±0.48 mm) and 6th month (2.80±0.88 mm). There was absolute change of -0.60±0.49 mm from baseline to loading stage and -2.25±0.59 mm from baseline to 6th month interval. The difference between baseline and loading stage was statistically significant ($p<0.05$) whereas at the 6 months interval also, the difference from the baseline was statistically significant ($p<0.001$).

Comparison between Group A, Group B, and Group C:

The mean height of crestal bone for the Group A was 0.57±0.21 mm at the baseline, 1.30±0.00 mm at loading stage and 1.40±0.31 mm at 6th month. For the Group B, the respective values were 0.49±0.28 mm (at baseline), 1.20±0.24 mm (at loading stage) and 1.45±0.28 mm (at 6th month). In Group C, the respective values were 0.55±0.50 mm (at baseline), 1.15±0.48 mm (at loading stage) and 2.80±0.88 mm (at 6th month). When the inter group comparison was made, there was statistically non-significant difference in the mean height of crestal bone scores between the Group A, Group B, and Group C at baseline and 6th month ($p>0.05$) and non-significant when

difference in the mean height of crestal bone scores between the Group A, Group B, and Group C at loading stage, analysed using the ANOVA as p value was 0.353 (p>0.05).

BLEEDING ON PROBING: (Table 6 & 7) (Graph V & VI)

Standard Implant Treatment (Group A):

The mean bleeding on probing score for the Group A at the 3 months interval was 0.9±0.57. The bleeding on probing score decreased to 1.10±0.74 at 6 months interval. The mean change in bleeding on probing scores recorded from the 3rd month to the 6th month interval was -0.2±0.42. The t value was -1.5. There was statistically non-significant decrease in the bleeding on probing scores from the baseline at each interval as the p value was 0.168 (p>0.05).

Immediate Implant Treatment (Group B):

The mean bleeding on probing score for the Group B at the 3 months interval was 1.1±0.74. The bleeding on probing score decreased to 1.40±0.70 at 6 months interval. The mean change in bleeding on probing scores recorded from the 3 months to 6 months interval was -0.3±0.67. The t value was -1.406. There was statistically non-significant decrease in the bleeding on probing scores from the baseline at each interval as the p value was 0.193 (p>0.05).

Implant Treatment in conjugation with Guided Bone Regeneration (Group C):

The mean bleeding on probing score for the Group C at the 3 months interval was 1.60±0.52. The bleeding on probing score decreased to 2.20±0.63 at 6 months interval. The mean change in bleeding on probing scores recorded from the 3 months to 6 months interval was -0.60±0.70. The t value was -2.714. There was statistically significant decrease in the bleeding on probing scores from the baseline at each interval as the p value was 0.024 (p>0.05).

Comparison between Group A, Group B, and Group C:

The mean bleeding on probing at the 3 months interval for the Group A, Group B, and Group C was 0.90±0.57, 1.10±0.74, and 1.60±0.52. The difference between the groups (at 3rd month) analysed using ANOVA (F value=3.441) was statistically significant as p <0.05 (p=0.047). At the 6 months interval Group A had bleeding on probing score of 1.10±0.74, for Group B 1.40±0.70 and for Group C it was 2.20±0.63. The difference between the groups at the 6 months was also analysed using ANOVA (F value = 6.767) and it was also, statistically significant at p=0.004 (p<0.05).

PLAQUE SCORE: (Table 8 & 9) (Graph VII & VIII)

Standard Implant Treatment (Group A):

The mean plaque score for the Group A 3 months after the loading stage was 1.41±0.69. The plaque score decreased to 1.01±0.65 at 6 months interval. The mean change in plaque scores recorded from the 3rd month to the 6th month after loading was 0.4±0.10. The t value was 12.000. There was statistically significant decrease in the plaque scores from the baseline at each interval as the p<0.001.

Immediate Implant Treatment (Group B):

The mean plaque score for the Group B 3 months after the loading stage was 1.43±0.65. The plaque score decreased to 0.99±0.62 at 6 months interval. The mean change in plaque scores recorded from the 3rd month to the 6th month after loading was 0.44±0.15. The t value was 9.242. There was statistically significant decrease in the plaque scores from the baseline at each interval as the p<0.001.

Implant Treatment in conjugation with Guided Bone Regeneration (Group C):

The mean plaque score for the Group C 3 months after the loading stage was 1.75±0.80. The plaque score decreased to 1.31±0.77 at 6 months interval. The mean change in plaque scores recorded from the 3rd month to the 6th month after loading was 0.44±0.19. The t value was 7.333. There was statistically significant decrease in the plaque scores from the baseline at each interval as the p<0.001.

Comparison between Group A, Group B, and Group C:

The mean plaque scores at the 3 months interval for the Group A, Group B, and Group C was 1.41±0.69, 1.43±0.65, and 1.75±0.80. The difference between the groups (at 3rd month) analysed using ANOVA (F value=0.705) was statistically non-significant as p>0.05 (p=0.503). At the 6 months interval Group A had plaque score of 1.01±0.65, for Group B 0.99±0.62 and for Group C it was 1.31±0.77. The difference between the groups at the 6 months was also analysed using ANOVA (F value = 0.682) and it was also, statistically non-significant at p=0.514 (p>0.05).

IMPLANT STABILTY (Table 10) (Graph IX):

Standard Implant Treatment (Group A):

The mean score for the implant mobility was 1.0±0.00 for the delayed implant surgery group assessed using the mobility index of the endosseous implants developed by Wasserman.

Immediate Implant Treatment (Group B):

The mean score of the implant mobility for the immediate implant placement surgery group system was 1.00±0.00 assessed using the mobility index of the endosseous implants developed by Wasserman.

Implant Treatment in conjugation with Guided Bone Regeneration (Group C):

The mean score of the implant mobility for this group was 1.00±0.00 assessed using the mobility index of the endosseous implants developed by Wasserman.

Comparison between Group A, Group B, and Group C:

The mean implant mobility scores for all the groups i.e. Group A, Group B, and Group C were 1.00±0.00. The difference between the groups for the implant mobility when analysed using Independent t Test was statistically non-significant at p= 1.000.

PINK ESTHETIC SCORE (Table 11) (Graph X)

Standard Implant Treatment (Group A):

The mean aesthetic score 1 month after prosthesis placement was 21.6, assessed using the Pink Aesthetic Score Index developed by Fürhauser ($p<0.001$).

Immediate Implant Treatment (Group B):

The mean aesthetic score 3 months after prosthesis placement was 5.948, assessed using the Pink Aesthetic Score Index developed by Fürhauser, with $p=0.203$ ($p>0.05$).

Implant Treatment in conjugation with Guided Bone Regeneration (Group C):

The mean aesthetic score 6 months after prosthesis placement was 5.963, assessed using the Pink Aesthetic Score Index developed by Fürhauser, with $p=0.051$ ($p=0.05$).

Comparison between Group A, Group B, and Group C:

The table reveals intergroup comparison of aesthetic score between group A, group B, and group C. A statistically significant difference in aesthetic scores was observed among different groups at 1st month after loading and 6th month after loading, although no significant difference in aesthetic score was observed among the three groups at 1st month and 3rd month, and 3rd month and 6th month after prosthesis placement.

TABLE 2-INTRAGROUP COMPARISION OF PROBING DEPTH

		Values of designated intervals	Changes from Loading Stage	Significance of difference from baseline using Paired sample t-test		
		Mean±SD	Mean±SD	t-value	P-value	Significance
Group A	At loading	1.60±0.52	-	-	-	-
	3 months after loading	1.80±0.63	-0.20±0.57	-6.128	0.041	Sig
	6 months after loading	1.90±0.62	-0.30±0.53	-9.000	0.022	Sig
Group B	At loading	1.66±0.49	-	-	-	-
	3 months after loading	1.82±0.53	-0.16±0.48	-4.583	0.032	Sig.
	6 months after loading	2.30±0.49	-0.64±0.67	-6.091	0.001	Sig
Group C	At loading	1.70±0.22	-	-	-	-
	3 months after loading	1.83±.032	-0.13±0.63	-4.000	0.006	Sig.
	6 months after loading	2.86±0.55	-1.16±0.85	-5.582	<0.001	Sig

TABLE 3- INTERGROUP COMPARISION OF PROBING DEPTH

	Group A	Group B	Group C	Significance of difference using ANOVA		
	Mean±SD	Mean±SD	Mean±SD	F-value	P-value	Significance
At loading	1.60±0.52	1.66±0.49	1.70±0.22	0.900	0.418	Non-sig.
3 months after loading	1.80±0.63	1.82±0.53	1.83±.032	0.618	0.547	Non-sig.
6 months after loading	1.90±0.62	2.30±0.49	2.86±0.55	1.735	0.196	Non-sig.

TABLE 4- INTRAGROUP COMPARISION OF CRESTAL BONE

		Values of designated intervals	Changes from Baseline	Significance of difference from baseline using Paired sample t-test		
		Mean±SD	Mean±SD	t-value	P-value	Significance
Group A	At Baseline*	0.57±0.21	-	-	-	-
	4 months	1.30±0.00	-0.73±0.21	-14.686	0.001	Sig.
	6 months	1.40±0.31	-0.83±0.19	-18.028	<0.001	Sig
Group B	At Baseline*	0.49±0.28	-	-	-	-
	4 months	1.20±0.24	-0.71±0.31	-9.000	0.003	Sig.
	6 months	1.45±0.28	-0.96±0.39	-8.820	<0.001	Sig
Group C	At Baseline*	0.55±0.50	-	-	-	-
	4 months	1.15±0.48	-0.60±0.49	-4.881	0.04	Sig.
	6 months	2.80±0.88	-2.25±0.59	-7.005	<0.001	Sig

TABLE 5- INTERGROUP COMPARISION OF CRESTAL BONE

	Group A	Group B	Group C	Significance of difference using ANOVA		
	Mean±SD	Mean±SD	Mean±SD	F-value	P-value	Significance
At Baseline*	0.57±0.21	0.49±0.28	0.55±0.50	0.207	0.827	Non-sig

4 months	1.30±0.00	1.20±0.24	1.15±0.48	1.134	0.353	Non-sig
6 months	1.40±0.31	1.45±0.28	2.80±0.88	0.816	0.457	Non-sig.

TABLE 6- INTRAGROUP COMPARISION OF BLEEDING INDEX

		Values of designated intervals	Changes from 3 months after loading	Significance of difference from baseline using Paired sample t-test		
		Mean±SD	Mean±SD	t-value	P-value	Significance
Group A	3 months after loading	0.90±0.57	-	-	-	-
	6 months after loading	1.10±0.74	-0.20±0.42	-1.50	0.168	Non-sig.
Group B	3 months after loading	1.10±0.74	-	-	-	-
	6 months after loading	1.40±0.70	-0.30±0.67	-1.406	0.193	Non-sig.
Group C	3 months after loading	1.60±0.52	-	-	-	-
	6 months after loading	2.20±0.63	-.060±0.70	-2.714	0.024	Sig.

TABLE 7- INTERGROUP COMPARISION OF BLEEDING INDEX

	Group A	Group B	Group C	Significance of difference using ANOVA		
	Mean±SD	Mean±SD	Mean±SD	F-value	P-value	Significance
3 months after loading	0.90±0.57	1.10±0.74	1.60±0.52	3.441	0.047	Sig.
6 months after loading	1.10±0.74	1.40±0.70	2.20±0.63	6.767	0.004	Sig.

TABLE 8- INTRAGROUP COMPARISION OF PLAQUE SCORE

		Values of designated intervals	Changes from 3 months after loading	Significance of difference from baseline using Paired sample t-test		
		Mean±SD	Mean±SD	t-value	P-value	Significance
Group A	3 months after loading	1.41±0.69	-	-	-	-
	6 months after loading	1.01±0.65	0.40±0.10	12.000	<0.001	Sig.
Group B	3 months after loading	1.43±0.65	-	-	-	-
	6 months after loading	0.99±.62	0.44±0.15	9.242	<0.001	Sig.
Group C	3 months after loading	1.75±0.80	-	-	-	-
	6 months after loading	1.31±0.77	0.44±0.19	7.333	<0.001	Sig.

TABLE 9- INTERGROUP COMPARISION OF PLAQUE SCORE

	Group A	Group B	Group C	Significance of difference using ANOVA		
	Mean±SD	Mean±SD	Mean±SD	F-value	P-value	Significance
3 months after loading	1.41±0.69	1.43±0.65	1.75±0.80	0.705	0.503	Non-sig.
6 months after loading	1.01±0.65	0.99±.62	1.31±0.77	0.682	0.514	Non-sig.

TABLE 10- INTERGROUP COMPARISON OF NORMAL IMPLANT STABILITY

Normal Implant Stability	Group A	Group B	Group C	Significance of difference using Chi-square		
	N (%)	N (%)	N (%)	Chi-square	P-value	Significance
At loading	10 (100)	10 (100)	10 (100)	-	-	-
3 months after loading	10 (100)	10 (100)	10 (100)	-	-	-
6 months after loading	10 (100)	10 (100)	10 (100)	-	-	-

TABLE 11- INTERGROUP COMPARISON OF ESTHETIC EVALUATION

	Group A	Group B	Group C	Significance of difference using Chi-square		
	N (%)	N (%)	N (%)	Chi-square	P-value	Significance
1 month after loading						
0	0 (0)	0 (0)	3 (30)	21.6	<0.001	Sig.
1	2 (20)	10 (100)	3 (30)			
2	8 (80)	0 (0)	4 (40)			
3 months after loading						
0	0 (0)	0 (0)	2(20)	5.948	0.203	Non-sig.
1	1 (10)	3 (30)	1 (10)			
2	9 (90)	7 (70)	7 (70)			
6 months after loading						
0	0 (0)	0 (0)	0 (0)	5.963	0.051	Sig
1	1 (10)	1 (10)	5 (50)			
2	9 (90)	9 (90)	5 (50)			

DISCUSSION

The present study was carried out to assess and compare crestal bone changes, aesthetics, and clinical outcomes after immediate and delayed implant placement, and use of bone grafts along with delayed implant placement. Partially edentulous patients with one or more missing teeth with good oral hygiene and systemic health were selected for the study. Patients with oral or systemic factors that would inhibit wound healing process for osseointegration were excluded.

PROBING DEPTH INDEX

Intra Group Analysis (Group A)

In the group A subjects, the change in the Probing Depth scores at the baseline was 1.60±0.52 mm, it was 1.80±0.63 mm at 3 months from the baseline and it was 1.9±0.62 mm at 6 months from the baseline. The difference at 3 months interval was statistically significant ($p<0.05$) and was also statistically significant at 6 month interval ($p<0.05$). The results obtained were in accordance with a study conducted by **Marwa M (2013),[18] and Shiva R et al (2014),[19]** and in which statistically significant increase in pocket probing depth was obtained.

Intra Group Analysis (Group B)

In the group B subjects, the change in the Probing Depth scores at the baseline was 1.66±0.49 mm, it was 1.82±0.53 mm at 3 months from the baseline and it was 2.3±0.49 mm at 6 months from the baseline. The difference at 3 months interval was statistically significant ($p<0.05$) and 6 month interval was also statistically significant ($p=0.001$). The results obtained were in accordance with a study conducted by **Anand S (2013),[20] and Gerber JA et al,[21]** in which statistically significant results were obtained.

Intra Group Analysis (Group C)

In the group C subjects, the change in the Probing Depth scores at the baseline was 1.70±0.22 mm, it was 1.83±0.32 mm at 3 months from the baseline and it was 2.86±0.55 mm at 6 months from the baseline. The difference at 3 months interval was statistically significant ($p<0.05$) and 6 month interval was also statistically significant ($p<0.001$). The results obtained were in accordance with a study conducted by **Buser D et al (2009)[22]** in which statistically significant results were obtained.

Inter Group Analysis

Statistically non-significant difference in the Probing Depth scores was observed when the inter group comparison was made, between the group A, group B, and group C subjects for the baseline ($p=0.418$), 3 months interval ($p=0.547$) and 6 months intervals ($p=0.196$) after the prosthesis placement. These results were in accordance to the study conducted by **Zafiropoulos GG (2010).[23] Schou et al (2002)[24]** compared probing depths around teeth and implants, reporting that probe penetration was deeper in implants if mild marginal inflammation was present. However, it is reasonable to assume that probing depth not exceeding 4.0 mm is preferable to facilitate the patient's ability for self-performed plaque control as well as accessibility for proper professional peri-implant cleaning in which similar results were observed at various intervals.

HEIGHT OF CRESTAL BONE

Intra Group Analysis (Group A)

Height of crestal bone was measured initially immediately after stage I surgery then after 4 months and 6 months. The mean height of the crestal bone at the Stage I was 0.57±0.21 mm, it was 1.30±0.00 mm at the Stage II and it was 1.40±0.31 mm at 6 months after implant placement and change was -0.73±0.21 at 4 months and -0.83±0.19 mm at 6 months after implant placement. Statistically the change from the Stage I to Stage II was statistically significant at $p=0.001$.

The present study is in contrast to the study conducted by **Schropp et al (2008**),[25] who concluded that there was no significant difference between immediate and delayed implants in approximal bone level changes during first year.

Intra Group Analysis (Group B)

In group B subjects, the mean height of the crestal bone at the Stage I and Stage II was 0.49±0.28 mm and 1.20±0.24 mm respectively and it was 1.45±0.28 mm at 6 months after implant placement and change was -0.71±0.31 mm at 4 months and -0.96±0.39 mm at 6 months after implant placement. Statistically the change from the Stage I to Stage II was statistically significant at $p<0.05$. These results were in accordance with study done by **Vigolo and Givani (2009),[26]** also reported significantly in which similar results were obtained.

Intra Group Analysis (Group C)

In group C subjects, the mean height of the crestal bone at the Stage I and Stage II was 0.55±0.50 mm and 1.15±0.48 mm respectively and it was 2.80±0.88 mm at 6 months after implant placement and change was -0.60±0.49 mm at 3 months and -2.25±0.59 mm at 6 months after implant placement. Statistically the change from the Stage I to Stage II was statistically significant at $p<0.05$. At the 6 months interval also, the difference from the baseline was statistically significant ($p<0.001$). These results were in accordance with study done by **Huang HY et al,**[27] in which similar results were obtained.

Inter Group Analysis

The difference of crestal bone height scores between the Group A, Group B, and Group C was statistically non-significant at Stage I ($p= 0.827$) and non-significant at Stage II ($p= 0.353$). They were also statistically non-significant at 6 months after loading ($p=0.457$). These results were in accordance to study done by **Canullo (2010),**[28] **Cappiello (2008),**[29] in which it was concluded that there is no difference in immediate implant placement and delayed implant placement.

BLEEDING ON PROBING

Intra Group Analysis (Group A)

In the group A subjects, the change in the Bleeding on Probing scores at the 3rd month was 0.90±0.57, it was 1.10±0.74 at 6 months after prosthetic placement. The difference at 6 months interval was statistically non-significant ($p=0.168$). The results obtained were in accordance with a study conducted by **Casado PL (2013),**[30] in which statistically non-significant results were obtained.

Intra Group Analysis (Group B)

In the group B subjects, the change in the Bleeding on probing scores at the 3rd month was 1.10±0.74, it was 1.40±0.70 at 6 months after prosthetic placement. The difference between baseline and 6 months interval was statistically non-significant ($p=0.193$). The results obtained were in accordance with a study conducted by **Tim De Rouck (2009),**[31] in which statistically non-significant results were obtained.

Intra Group Analysis (Group C)

In the group C subjects, the change in the Bleeding on probing scores at the 3rd month was 1.60±0.52, it was 2.20±0.63 at 6 months after prosthetic placement. The difference between baseline and 6 months interval was statistically significant ($p=0.024$). The results obtained were in accordance with a study conducted by **Talreja PS et al (2013),**[32] in which statistically significant results were obtained.

Inter Group Analysis

Statistically significant difference in the Bleeding on Probing scores was observed when the inter group comparison was made, between the group A, group B, and group C subjects for the baseline, 3 months' interval ($p=0.047$) and 6 months' intervals ($p=0.004$) after the prosthesis placement. These results were in accordance to the study conducted by **Alvira-Gonzalez J et al (2015),**[33] in which similar results were observed at various intervals while comparing delayed and immediate implants.

PLAQUE SCORE

Intra Group Analysis (Group A)

The mean plaque score for the Group A, 3 months after the loading stage was 1.41±0.69. The plaque score decreased to 1.01±0.65 at 6 months interval. There was statistically significant decrease in the plaque scores from the baseline at each interval as the $p<0.001$. This study is in accordance with the results obtained in a study by **Kumar PKS et al (2013),**[34] which showed decrease in plaque score during 6 months follow up.

Intra Group Analysis (Group B)

The mean plaque score for the Group B 3 months after the loading stage was 1.43±0.65. The plaque score decreased to 0.99±0.62 at 6 months interval. The mean change in plaque scores recorded from the 3rd month to the 6th month after loading was 0.44±0.15. There was statistically significant decrease in the plaque scores from the baseline at each interval as the $p<0.001$. The results of this study were similar to study conducted by **Anitha K et al (2014),**[35] in which satisfactory oral hygiene was observed in subjects who received immediate implants.

Intra Group Analysis (Group C)

The mean plaque score for the Group C 3 months after the loading stage was 1.75±0.80. The plaque score decreased to 1.31±0.77 at 6 months interval. There was statistically significant decrease in the plaque scores from the baseline at each interval with $p<0.001$. Our results agreed with the results of the study conducted by **Aly LA et al (2016).**[36]

Inter Group Analysis

Statistically non-significant difference was seen in mean plaque score in all the subjects of group A, group B, and group C. There was an overall decrease in plaque score of all the subjects. These results of our study were similar to studies conducted by **Bazrafshan N et al (2014),[37] Visser A et al (2005),[38] and Cosyn J et al (2012).[11]** Low plaque score in the subjects could be attributed to good oral hygiene maintenance during the follow-up period.

IMPLANT MOBILTY

Intra Group Analysis (Group A)

The mean score for the implant mobility was 1.00±0.00 assessed using the mobility index of the endosseous implants developed by Wasserman. Lack of mobility was observed in all group A subjects.

Intra Group Analysis (Group B)

The mean score for the implant mobility was 1.00±0.00 assessed using the mobility index of the endosseous implants developed by Wasserman. Lack of mobility was observed in all group B subjects.

Intra Group Analysis (Group C)

The mean score for the implant mobility was 1.00±0.00 assessed using the mobility index of the endosseous implants developed by Wasserman. Lack of mobility was observed in all group C subjects.

Inter Group Analysis

All subjects of group A, group B, and group C demonstrated lack of mobility at abutment placement during stage II. Statistically non-significant differences in values were obtained when implant mobility index was compared between group A, group B, and group C subjects. Similar results were obtained in the study by **Ueli Grunder (1999),[39]** in which he compared traditional and immediate implant surgeries.

AESTHETIC EVALUATION

Intra Group Analysis (Group A)

The mean aesthetic score 1 month after prosthesis placement was 21.6, assessed using the Pink Aesthetic Score Index developed by Fürhauser ($p<0.001$).

Intra Group Analysis (Group B)

The mean aesthetic score 3 months after prosthesis placement was 5.948, assessed using the Pink Aesthetic Score Index developed by Fürhauser, with $p=0.203$ ($p>0.05$).

Intra Group Analysis (Group C)

The mean aesthetic score 6 months after prosthesis placement was 5.963, assessed using the Pink Aesthetic Score Index developed by Fürhauser, with $p=0.051$ ($p=0.05$).

Inter Group Analysis

A statistically significant difference in aesthetic scores was observed among different groups at 1st month after loading and 6th month after loading, although no significant difference in aesthetic score was observed among the three groups at 1st month and 3rd month, and 3rd month and 6th month after prosthesis placement. Acceptable aesthetic outcomes were obtained in group A patients, where adequate healing time was given to periimplant soft tissue. These results were in accordance to the study conducted by **Cooper LF et al (2010),[40]** who concluded that the responses reflect the condition of the tissues prior to implant placement and that interproximal tissue formation should be part of the response to careful implant placement and restoration.

Conclusion

All the three treatment modalities used in our study were predictable from a clinical and radiographic point of view, with Standard Implant Treatment cases giving best results, followed by cases with Immediate Implant Treatment. However, GBR increased the risk for complications and compromised aesthetics. More research with a much larger sample size would be better in terms of more conclusive data.

References

1. Schropp L, Kostopoulos L, Wenzel A. Bone Healing Following Immediate Versus Delayed Placement of Titanium implants into Extraction Sockets: A Prospective Clinical Study. Int J Oral Maxillofac Implants 2003; 18:189–199.
2. Romeo E, Chiapasco M, Ghisolfi M and Vogel G. Long-term clinical effectiveness of oral implants in the treatment of partial edentulism. Sevenyear life table analysis of a prospective study with ITI dental implants system used for single-tooth restorations. Clin Oral Implants Res 2002; 13: 133–43.

3. den Hartog L, Slater JJ, Vissink A, Meijer HJ, Raghoebar GM. Treatment outcome of immediate, early and conventional single-tooth implants in the aesthetic zone: A systematic review to survival, bone level, soft-tissue, aesthetics and patient satisfaction. J Clin Periodontol. 2008; 35: 1073-86.
4. Belser U, Buser D, and Higginbottom F. Consensus statements and recommended clinical procedures regarding esthetics in implant dentistry. Int J Oral Maxillofac Implants 2004; 19: 73–4.
5. Kahnberg KE. Immediate Implant Placement in Fresh Extraction Sockets: A Clinical Report. Int Oral Maxillofac Implants 2009; 24:282–288.
6. Creugers NHJ, Kreulen CM, Snoek PA & de Kanter RJ. A systematic review of single tooth restorations supported by implants. Journal of Dentistry 2000; 28: 209-17.
7. Schropp L, Wenzel A, Kostopoulos L & Karring T. Bone healing and soft tissue contour changes following single-tooth extraction: a clinical and radiographic 12-month prospective study. International Journal of Periodontics and Restorative Dentistry 2003; 23: 313–23.
8. Cosyn J and De Rouck T. Aesthetic outcome of single-tooth implant restorations following early implant placement and guided bone regeneration: crown and soft tissue dimensions compared with contralateral teeth. Clin Oral Implants Res 2009; 20: 1063-9.
9. Raghoebar GM, Batenburg RH, Vissink A, Reintsema H. Augmentation of localized defects of the anterior maxillary ridge with autogenous bone before insertion of implants. Int J Oral Maxillofac Surg 1996; 54:1180–5.
10. Von Arx T, Wallkamm B, Hardt N. Localized ridge augmentation using a micro titanium mesh: a report on 27 implants followed from 1 to 3 years after functional loading. Clin Oral Implants Res 1998; 9:123–130.
11. Cosyn J, Eghbali A, Hanselaer L, De Rouck T, Wyn I, Sabzevar MM, Cleymaet R and De Bruyn H. Four Modalities of Single Implant Treatment in the Anterior Maxilla: A Clinical, Radiographic, and Aesthetic Evaluation. Clin Implant Dent Relat Res 2013; 15: 517-30.
12. Chen ST, Wilson Jr. TG, Hammerle CHF. Immediate or early placement of implants following tooth extraction: review of biological basis, clinical procedures, and outcomes Int J Oral Maxillofac Implants 2004; 19:12-25.
13. Silness J and Loe H. Periodontal Diseases in Pregnancy. II. Correlation between Oral Hygiene and Periodontal Condition. Acta Odontol Scand 1964; 22: 121-35.
14. Mombelli A, van Oosten MA, Schurch E Jr and Land NP. The microbiota associated with successful or failing osseointegrated titanium implants. Oral Microbiol Immunol 1987; 2: 145-51.
15. Furhauser R, Florescu D, Benesch T, Haas R, Mailath G and Watzek G. Evaluation of soft tissue around single-tooth implant crowns: the pink esthetic score. Clin Oral Implants Res 2005; 16: 639-44.
16. Wasserman, B., A. Geiger, and L. Turgeon. Relationship of occlusion and periodontal disease: VII. Mobility. J Periodontol 1973; 44:57–78.
17. Glickman, I. Clinical Periodontology. 2nd ed. Philadelphia, Pa:WB Saunders; 1958:694-96.
18. Marwa M, Osama Z, Kanako N, Masaki F, Shohei K. Peri-implantitis Progression Around Thin Sputtered Hydroxyapatite-Coated Implants: Clinical and Radiographic Evaluation in Dogs. Int J Oral Maxillofac Implants 2013; 28: 701–09.
19. Shiva R, Priya P, Taneja A, Ipsitha. Evaluation of osseointegration following placement of mini implants. I J Pre Clin Dent Res 2014; 1: 1-6.
20. Anand S, Gambhir RS, Aggarwal J, Kaushik A,Nanda T. Bone platform switching: Perpetuating crestal bone preservation.A pilot study. J Dent Implant 2013; 3: 9-15.
21. Gerber JA, Tan WC, Balmer TE, Salvi GE, Lang NP. Bleeding on probing and pocket depth in relation to probing pressure and mucosal health around oral implants. Clin Oral Implant Res 2009; 20: 75-8.
22. Buser D, Halbritter S, Hart C, Bornstein MM, Grutter L, Chappuis V, Belser UC. Early implant placement with simultaneous guided bone regeneration following single-tooth extraction in the esthetic zone: 12-month results of a prospective study with 20 consecutive patients. J Periodontol 2009; 80: 152-62.
23. Zafiropoulos GG, Kasaj A, Hoffmann O. Immediate implant placement in fresh mandibular molar extraction sockets: 8 years result. A case report. J oral implantol 2010; 36:145-51.

24. Schou S, Holmstrup P, Stoltze K, Hjørting-Hansen E, Fiehn NE, Skovgaard LT. Probing around implants and teeth with healthy or inflamed peri-implant mucosa. A histological comparision in cynomolgus monkeys. Clin Oral Impl Res 2002: 13; 113-26.
25. Schropp L, Isidor F. timing of implant placement relative to tooth extraction. J oral rehabil 2008; 35:33-43.
26. Vigolo P, Givani A, Platform-switched restorations on wide-diameter implants: a 5 years clinical prospective study. int J Oral Maxillofac Implants 2009; 24:103-09.
27. Huang HY, Ogata Y, Hanley J, Finkelman M, Hur Y.Crestal bone resorption in augmented bone using mineralized freeze-dried bone allograft or pristine bone during submerged implant healing; a prospective study in humans. Clin Oral Implants Res 2016; 27: e25-30.
28. Canullo L, Fedele GR, Iannello G, Jepsen S. Platform switching and marginal bone-level alterations: the results of a randomized controlled trial. Clin Oral Implants Res 2010; 21: 115-21.
29. Cappiello M, Luongo R, Di Iorio D, Bugea C, Cocchetto R, Celletti R. Evaluation of peri-implant bone loss around platform switched implants. Int J Periodontics Restorative Dent 2008; 28: 347-55.
30. Casado PL, Villas-Boas R, Leao da Silva CL, Andrade FC, Bonato LL, Granjeiro JM. Is bleeding on probing a differential diagnosis between periimplant health and disease? Braz. J. Oral Sci. 2013; 12: 165-88.
31. De Rouck T, Collys K, Wyn I, Cosyn J. Instant provisionalization of immediate single-tooth implants is essential to optimize esthetic treatment outcome. Clin Oral Implants Res 2009; 20: 566-70.
32. Talreja PS, Gayathri GV, Mehta DS. Treatment of an early failing implant by guided bone regeneration using resorbable collagen membrane and bioactive glass. J Indian Soc Periodontol 2013; 17: 131-6.
33. Alvira-Gonzalez J, Diaz-Campos E, Sanchez-Garces MA, Gay-Escoda C. Survival of immediately versus delayed loaded short implants: A prospective case series study. Med Oral Patol Oral Cir Bucal 2015; 20: e480-8.
34. Kumar PKS, Ravikumar A, Elavarasu S, Kumaran T, Murugan B. Clinical and radiographic evealuation of immediate and delayed single-tooth implant placement: An 18-month follow-up study.
35. Anitha K, Senthil SK, Babu MRR, Candamourty R, Thirumurugan. Immediate implants in anterior maxillary arch. J Nat Sci Biol Med 2014; 5: 82-89.
36. Aly LA, Nassar H. Outcome of immediate implant with bone augmentation in the anterior maxillary supporting partial denture. J Dent Implant 2016; 6: 13-21.
37. Bazrafshan N, Darby I. Retrospective success and survival rates of dental implants placed with simultaneous bone augmentation in partially edentulous patients. Clin Oral Implants Res 2014; 25: 768-73.
38. Visser A, Raghoebar GM, Meijer HJA, Batenburg RHK, Vissink A. Mandibular overdentures supported by two or four endosseous implants. A 5-year prospective study. Clin Oral Impl. Res 2005; 16: 19–25.
39. Grunder U, Pollizi G, Goene R, Hatano N, Henry P, Jackson WJ, Kawamra K, Kohler S, Renouard F, Rosenberg R, TriplettG, Werbitt M and Lithner B A 3-Year Prospective Multicenter Follow-up Report on the Immediate and Delayed-Immediate Placement of Implants. Int J Oral Maxillofac Implants 1999; 14:210–16.
40. Cooper LF, Raes F, Reside GJ, Garriga JS, Tarrida LG, Wiltfang J, Kern M, De Bruyn H. Comparison of radiographic and clinical outcomes following immediate provisionalization of single-tooth dental implants placed in healed alveolar ridges and extraction sockets. Int J Oral Maxillofac Implants 2010; 25: 1222-32.