# THE USE OF DEMINERALIZED FREEZE-DRIED BOVINE BONE XENOGRAFT IN REDUCING POST-SURGICAL PERIODONTAL <u>POCKET DEPTH</u>

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# ABSTRACT

This study evaluated the effectiveness of demineralized freeze-dried bone xenograft in reducing post-surgical pocket depth in moderate to advanced adult periodontitis in patients. Nine patients with a total of eighteen intrabony defects were selected for this study. The bony defects were matched for tooth type, location and pocket depth. Following an initial non-surgical treatment, only pockets of 5 to 7 mm deep were indicated for surgery. Periodontal pockets were measured pre-operatively and at 3, 6 and 9 months post-surgically. The study protocol included a split mouth design, where surgical treatment was carried out at both test and control sites. The test sites were assigned demineralized freeze-dried bone xenograft and the control sites were subjected to debridement alone without the use of demineralized freeze-dried bone xenograft. The results from this study showed a statistically significant difference in the mean pocket depth at 6 and 9 months post-operatively for both test and control groups, but there was no statistically significant difference at 3 months. In conclusion, demineralized freeze-dried bone xenograft was ineffective in reducing periodontal pocket depth in patients with moderate to severe periodontitis, as compared to surgical debridement alone.

Key words: periodontal pocket, demineralized freezedried xenograft.

### INTRODUCTION

The ultimate goals of periodontal therapy are to preserve the natural dentition by maintaining and improving periodontal health, comfort, aesthetic and function and to provide replacements of the periodontal tissues where necessary (1). The potential for regeneration of hard and soft tissues lost to periodontal diseases should be considered in managing periodontal disease.

The use of bone grafts for reconstructing osseous defects produced by periodontal diseases dated back to 1923 and was popularized by Nabers and O'Leary in 1965 (2). Of all the bone grafting materials being developed, bovine derived xenograft (BDX) has

#### **Origina.l Article**

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recently been shown to have the potential for periodontal regeneration (3). BDX possesses properties that are similar to human bone in terms of inner surface area, porosity, crystalline size and calcium-tophosphorous ratio (4). Thus, it provides an alternative to hard tissue transfer which however some patients may find unacceptable because of concerns of disease transmission.

All bone grafts possess osteoconductive potential i.e. by providing scaffolding for ossification. However, only some bone grafts have osteoinductive potential i.e. the ability to induce new bone formation by stimulating pleuri-potential stem cells to differentiate into osteoblasts to lay down bone (5). By demineralizing the bone in O.6M hydrochloric acid prior to freeze drying, the osteoinductive potential is enhanced as the bone morphogenic protein (BMP) is exposed. Demin~ralized freeze-dried bovine bone xenograft (DFDBBX) was shown to be highly osteoconductive when compared to osseous coagulum, bone blend and freeze-dried bone (6). Clinical and histological studies in human indicated that DFDBBX significantly enhanced new attachment formation in submerged and non-submerged intrabony defects (7,8). Animal investigations had also shown evidence of new host bone deposition at 6 months following the placement of the bone xenograft (8). The objective of this investigation was to determine the effectiveness of DFDBBX in reducing periodontal pocket depth in patients with moderate to advanced chronic periodontitis.

## METHODS

Participants in this study were patients of the Dental Faculty, University of Malaya who were referred for periodontal treatment. Nine patients with a total of nine pairs of intrabony defects were selected for this study. The defects were matched for tooth type, location and pocket depth (5 to 7 mm). Explanations were given to each patient regarding the procedures involved and each patient was required to sign a written consent form prior to commencement of treatment. Following initial non-surgical treatment that included oral hygiene instructions, scaling and root planing, all pockets at sites for surgery were measured post-operatively.

The design of the study was the split mouth design where the test sites were assigned DFDBBX and the control sites were subjected to surgical debridement alone. Following local anaesthesia (inferior dental nerve block or local infiltration) using xylocaine with adrenaline (1:80,000) scalloped internal bevel su1cular incisions were made along the gingival margin down to alveolar bone crest using a scalpel blade no. 12. Full thickness mucoperiosteal flaps were reflected exposing the crestal bone using a periosteal elevator on the facial/buccal and lingual/palatal surfaces of each tooth. During the flap surgery, the interproximal papilla were preserved to ensure maximum closure and graft coverage post-surgically. The exposed osseous intrabony defects were debrided of granulation tissues using hand instruments and the adjacent root surfaces planed to a hard, smooth surface with ultrasonic and hand instruments. The defect areas were irrigated with sterile saline to remove the debris. Following that, a rose bur with copious irrigation was used to create bleeding points on the alveolar bone to obtain fresh blood to wet the granulated DFDBBX which was then carefully placed in the intrabony defect area using a spoon excavator. Primary wound closure was achieved by means of simple interrupted or horizontal mattress sutures (4/0 silk). Every effort was made to ensure the repositioning of the flaps at the enamocemental junctions. Similar procedures were carried out for the control sites except there was no decortication of the alveolar bone and no DFDBBX placement in the bone defects. The flaps were replaced and sutured with interrupted sutures.

The operator verbally gave post-operative instructions to each patient. The patients were advised not to brush the surgical area for 2 weeks until the sutures were removed; instead, the patients were recommended to rinse with or to place wet gauze soaked in 0.12 % chlorohexidine gluconate solution twice daily. Each patient was given medications that consisted of an antibiotic, anti swelling agent and analgesic. The patients were recalled for review after 2 weeks, 3 months, 6 months and 9 months respectively.

Statistical analysis was carried out using the SPSS (version 10.0) to determine mean values and standard deviations. In order to explore the difference between the 2 sets of groups, Repeated Measure Analysis was used. In SPSS, the effect size is measured in terms

of Eta squared. The value of Eta ranges from 0 to 1. An Eta value more than 0.14 (Eta>0.14) is accepted as a strong evidence of significance difference. In small sample studies, Eta is chosen as a yardstick to measure the magnitude of the effect.

### RESULTS

# Table 1. Mean $(\pm$ SO) and Percentage Reduction in Pocket Depth at Various Time Intervals

Time	Groups	Mean ± SO (mm)	% reduction in pocket depth from baseline
0 months	Test (n=9) Control (n=9)	5.56 ± 1.01 5.56 ± 0.88	
3 months	Test (n=9)	4.33 ± 0.89	22.12
	Control (n=9)	4.89 ± 0.78	12.05
6 months	Test (n=9)	4.00 ± 0.89	28.06
	Control (n=9)	3.33 ± 1.39	40.11
9 months	Test (n=9)	3.44 ± 1.51	38.13
	Control (n=9)	3.43 ± 1.13	38.31

The result showed a gradual reduction in mean pocket depth for both test and control groups at 3, 6 and 9 months post-surgically compared to baseline.

For the test group, the percentage reduction in pocket depth was 22.12 at 3 months, 28.06 at 6 months and 38.13 at 9 months post-surgically. For the control groups, the percentage reduction was 12.05 at 3 months, 40.11 at 6 months and 38.31 at 9 months post-surgically.

Test of equality of error variance showed that there was no significant deviation from homogeneity between the test and control groups (p > 0.05). The test between subject effects was carried out to determine the effect of treatment to both groups. The test did not show any statistical significant difference in mean pocket depth between the test and control groups at baseline, 3, 6 and 9 months (p>0.05; Eta<0.14). Thus, there was no difference in the mean pocket depth between the two groups over time. In other words, the treatment intervention (DFDBBX) did not bring any advantage to the test group.

The test within subject effects was carried out to determine the effect of time and time by group interaction. As for time, the test gave p < 0.05 and Eta value of 0.46. Thus, there was strong evidence that the mean of at least one time period was significantly different. However, there was no difference in time by group interaction (p> 0.05; Eta < 0.14).

Further test using pair-wise comparison post-Hoc tests showed no significant difference between baseline and 3 months but there were significant improvements



Figure I: Graph showing probing pocket depth for both the Test (N = 9) and Control (N = 9) groups at various time intervals.

at 6 and 9 months compared to baseline mean values for both test and control groups. Therefore, it can be concluded that the significant improvement in test and control groups at 6 and 9 months post-surgically compared to baseline was mainly due to time factor and not due to the DFDBBX used.

### DISCUSSION

# 1. THE EFFECT OF DFDBBX ON POCKET DEPTH

At 3 months after surgery, the pocket depth reduction for the test group was 22.12 % and for the control group was 12.05 %. Although, it was not statistically significant, the difference in the mean and the percentage reduction in pocket depth in the test group relative to the control group could be attributed to the placement of DFDBBX in-situ which acted as scaffolds and occupied the space of the alveolar bone that was resorbed at the defect sites. The control group showed a lesser reduction since there was no xenograft placed in the defects to fill up the space, therefore the empty spaces that was previously occupied by granulation tissues just collapsed and caused recession.

At 6 months post-surgically, the percentage reduction in pocket depth for test group was 28.06% and for the control group was 40.11 %. These were statistically significant compared to baseline for both groups. In the test group, the significant reduction can be explained by the deposition and maturation of bone and incorporation of DFDBBX used. However, the residual pocket ( $\pm$  1 mm) observed in the test group could be due to simultaneous DFDBBX resorption that took place (9). In the control group, the significant

pocket depth reduction (40 %) at 6 months was due to further recession that took place.

During healing, gingival CT organization is associated with a progressive increase in collagen density having the net clinical effect of tissue shrinkage and tight "cuff-like" adaptation about the roots. Shrinkage may take place both concentrically and longitudinally about the tooth. This will result in reduced pocket depths by a closer adaptation of the gingiva about the tooth (pocket closure) and a reduction in the amount of detached tissue present (gingival recession).

At 9 months after surgery, the percentage reduction in pocket depth was 38.13 % for the test group and 38.31 % for the control group. There were statistically significant differences in the mean pocket depth at 9 months compared to baseline for both groups. Further reduction in the pocket depth for the test group was due to continued deposition and maturation of the host bone whereas in the control group, the reduction in pocket depth has almost stabilized since 6 months. At this juncture, the values of pocket depth for both test and control groups were approaching normal values.

Camelo et al (10) and Mellonig (3) have demonstrated that there was an increasing amount of bone formation from 4 months to 9 months postsurgically with the use of Bio-Oss® (Osteohealth Co., This was Shirley, NY). attributed to the osteoconductive and partly osteoinductive potential of Bio-Oss<sup>®</sup> (Osteohealth Co., Shirley, NY) that was The new bone placed in the periodontal defects. formation would gradually fill in the defect space causing reduction in the periodontal pocket depth. Similar process would be expected to happen to the DFDBBX used in this study as both materials

originated from similar source i.e. bovine bone and the DFDBBX used should at least possess osteoconductive potential. If this assumption of new bone deposition was correct, the author would expect further pocket depth reduction at 12 months postsurgically.

On the other hand, in the control group there was no difference between 6 and 9 months post-surgically. This was because there was no DFDBBX placed in situ, therefore the intrabony healing had maximized at 6 months and was quite stable and consequently the pocket depth did not change much after initial reduction.

The tendencies for patients to improve irrespective of any interventions are referred to as Hawthorne effects. In this study, the test and control groups received similar treatment i.e. both surgical and nonsurgical treatment but only the test group received DFDBBX in the bony defects. In addition the protocol included a split mouth design whereby non intervention factors such as patient oral hygiene can potentially influence the disease course for both test and control groups. Therefore, any observed change in a patient's response criteria may be due partly to Hawthorne effects and partly to the intervention itself. Thus, accurate assessment necessitates some adjustment for overseeing of the part of the Hawthorne effect.

### 2. BONE GRAFf

The selection was based on osteoconductive and osteoinductive (osteogenesis and cementogenesis) potential. The graft should also be biocompatible, noncariogenic, easily obtainable and relatively inexpensive to use.

Bio-Oss®(Osteohealth Co., Shirley, NY), a bovine derived xenograft (BDX), has been shown to be highly osteoconductive, with many BDX particles incorporated into newly formed bone (10). The same study showed significant clinical improvement on probing depth and attachment level. Similarly, the study also showed cementogenesis had taken place adjacent to the BDX particles. The degree of cementum formation was greater compared to using a synthesis membrane with DFDBA graft (6).

A similar BDX is available locally. The Tissue Bank in the Hospital Universiti Sains Malaysia Kubang Kerian has been producing DFDBBX for research purposes, as well as for oral, orthopaedics and ophthalmology surgeries. All the processes, techniques and stages were in accordance with the National Regulations and the Standards and Regulations of the European Association Tissue Banking. In this study, the author chose to use DFDBBX for a few reasons:

- •. It has closest the resemblance to human cancellous bone as compared to allograft and synthetic graft.
- .:. DFDBBX has been shown histologically in animal studies to have regenerative potential (9); and

.:. The DFDBBX possesses particle size of 250 to 1000", i.e. the size recommended for high osteogenic potential.

# CONCLUSION

The conclusion that can be drawn from this study was that DFDBBX was ineffective in minimizing periodontal pocket depth in moderate to advanced CIPD, as compared to surgical debridement.

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