



## A Comparative Evaluation Of Decalcified Freeze Dried Bone Allograft And Its Combination With Hydroxyapatite In Osseous Defects Of Maxillofacial Surgery

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### Abstract:

**Background:** The development of bone graft material to replace bone remains a formidable challenge in maxillofacial surgery. Although autogenous bone is the best material, however, the advantage of an autograft is offset by the limited supply of such bone and morbidity associated with surgery to harvest the graft. The thesis aims to study and compare the efficacy of two such bone substitutes in the healing of osseous defects of the maxillofacial surgery.

**Method:** 20 patients reporting to DEPARTMENT OF ORAL AND MAXILLOFACIAL SURGERY, AL-AMEEN DENTAL COLLEGE AND HOSPITAL, BIJAPUR, requiring

Treatment for osseous defects were selected for the study. They were categorized in two groups of 10 each. GROUP A were treated by DFDBA and GROUP B were using DFDBA and HYDROXYAPATITE in combination. Treatment outcome was evaluated using many parameters like pain, swelling clinically and bone density radiographically pre operative, 1<sup>st</sup> day, 1<sup>st</sup> week, 4<sup>th</sup> week and 12<sup>th</sup> week post operative. Results: Analysis of the two groups did not reveal high statistically significant differences at any of the follow up periods except for radiological evidence at four weeks.

All the patients had mild to moderate pain on GROUP A on first post-operative day. Pain was maximum on 2<sup>nd</sup> post-operative day and decreased gradually and almost nil by the end of 4<sup>th</sup> post-operative day. In case of GROUP B, all patients had moderate to severe pain on first post-operative day. Pain was maximum from 2<sup>nd</sup> to 4<sup>th</sup> post operative day and then gradually decreased but slight pain

<p><b>CC License</b> CC-BY-NC-SA 4.0</p>	<p>was there at the end of first post-operative week All the patients had mild to moderate swelling in GROUP A on first post-operative day. Swelling decreased gradually towards normal. By the end of first week post-operative, swelling was nil.</p> <p>In case of GROUP B, all patients had moderate to severe swelling on first post-operative day. Swelling was maximum from 1<sup>st</sup> post-operative week and then started decreasing towards normal. By the end of first week post-operative, slight swelling was still present. GROUP A No signs of infection (persistent post operative swelling, pain or pus discharge) were seen in any patient of GROUP A. No signs of implant (graft material) rejection (discharge, extrusion, tissue dehiscence) were seen in any patient of GROUP A. In GROUP B, however, all these three signs of implant (graft material) rejection were seen in one patient at fourth week. The graft material was surgically removed at fourth week. Radiological evidence of calcification, bone formation and bridging of the gap with new bone as evident by formation of irregular trabeculae of bone and appearance of radio-opaque areas in the defect was evident from fourth week onwards. It was seen 70 % cases of GROUP A, in 30% cases of GROUP B. The difference between GROUP A and GROUP B was statistically significant (P&lt;0.01) at this point of time. Bone density was assessed by grey scale histogram.</p> <p><b>Conclusion:</b> DFDBA when used for the obliteration of osseous defects bypasses the phase of obligatory resorption and shows early evidence of new bone formation. HA when used for Obliteration of osseous defects shows delayed first evidence of bone formation as compared to the decalcified freeze-dried bone matrix allograft and it undergoes resorption but takes a long period to resorbed completely and be replaced by bone. The radiograph assessment score over grey scale histogram indicates early bone formation with DFDB and the combination of DFDBA and HA.</p>
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### Introduction:

Bone is a specialized connective tissue that provides support and protection for the delicate and vital organs of the body and also allows for locomotion. Bone is a dynamic living tissue that shows marked structural alteration in response to injury, changes of stress and Vascular, endocrine, genetic and nutritional influences. It is one of the few human organs that can undergo regeneration rather than repair with formation of scar tissue. The development of bone graft material to replace bone remains a formidable challenge in maxillofacial surgery.<sup>15</sup>

The use of osseous and cartilaginous allogenic grafts for surgical success is of great importance. The methods of processing grafts to achieve optimal results; the biology of behavior of allografts and new bone substitutes, their corporation with the living tissues are seriously investigated Moreover, numerous researches mentioned for the biomechanical characteristics and behaviors of allografts corresponding with the vital bone. Bone possesses the intrinsic capacity for regeneration as part of the repair process in response to injury, as well as during skeletal development or continuous remodeling throughout adult life.

Although autogenous bone is the best material, however the advantage of an autograft is offset by the limited supply of such bone and morbidity associated with surgery to harvest the graft. The use of allogenic cartilaginous and osseous grafts of cadaveric origin in the reconstruction of skeletal defects has a long history in orthopedic surgery. Even there exists a legend from the 3<sup>rd</sup> century that stems from ancient times, concerning this history. Job Van Meekeren has performed the earliest recorded bone transplantation operation. In this operation, bone obtained from the hip of a dog, has been implanted to the skull defect of a wounded soldier. Later, in 1881, human fibula was used as an allogenic graft material for a child suffering from osteomyelitis and the operation turned out to be successful.

Autogenous cancellous bone is the best grafting material and has been used with clinical success for treatment of such lesions for many years. However, an additional surgical procedure, with increased morbidity, is required to obtain the graft, and there may be insufficient quantities of autogenous bone for grafting large or multiple defects. As an alternative, allogeneic bone, xenogeneic bone, or alloplastic bone substitute have been used. One type of allogeneic bone is decalcified freeze-dried bone allograft (DFDBA). Clinical studies using DFDBA in the treatment of human intraosseous periodontal lesions report significant

new bone formation. DFDBA has also been used as augmentation, construction, and interpositional grafts in maxillocraniofacial deformities with variable degrees of success.

The aim of thesis to study and compare the efficacy of two such bone substitutes in the healing of osseous defects of the jaw. The materials are DFDBA and HYDROXYAPATITE are used in 21 patients reporting to dept of oral and maxillofacial surgery al ameen dental college and hospital, Bijapur with osseous defects, will be selected. They are divided into two group in one group only DFDBA is used and in other group DFDBA is mixed with HYDROXYAPATITE. Post operative healing, bone density, and complications are assessed after regular intervals.<sup>3</sup>

### **Materials and Methods:**

This prospective study was conducted on 20 patients, reporting to department of oral and maxillofacial surgery, Al-Ameen Dental College and Hospital, Bijapur, requiring treatment related with osseous defects like cystic defects.

#### **Inclusion criteria:**

1. Age :18 to 55 years.
2. Both odontogenic and non-odontogenic cystic defect.
3. Any osseous defects in maxillofacial surgery in which allograft of proper size and shape can be placed.

#### **Exclusion criteria:**

1. Patient not willing to come under study.
2. Systemically compromised patients.
3. Communited fractures.
4. Infected cystic defect or bony cavities.

In this study 20 patients presenting with osseous defects were selected and were divided into two groups of 10 each.

For both groups of patients, pre-operative evaluation consisting of a complete case history, general physical examination, routine blood and urine examination, tests for HIV & HbsAg and orthopantomography, iopa and occlusal (if needed) radiographs were done. After thorough pre-operative evaluation, physical fitness was obtained for all the patients included in the study. All patients were covered under proper antibiotic coverage 12 hours preoperatively. Patients were taken up for surgery after informed written consent for using graft material in defect under local anesthesia or general anesthesia depending upon situation.

Source of bone graft material for study

- Material one: Decalcified freeze-dried bone allograft (DFDBA) is supplied by TISSUE BANK TATA MEMORIAL HOSPITAL, MUMBAI both in granular and block forms depending upon defect.
- Material two: Hydroxyapatite. From BIOGRAFT in both granules and block forms.

Patients with osseous defects were randomly divided into two groups.

- GROUP A osseous defects to be treated with DFDBA.
- GROUP B osseous defects treated with DFDBA and HYDROXYAPATITE in equal volume.

The size and shape of graft materials depends upon type and site of defect. Both materials are in granular and blocks form. For both group initial treatment is same as above mentioned.

The selection of type of radiograph depends upon type and site of osseous defect. Surgery was carried out under local anaesthesia or general anaesthesia depending upon patient's condition.

The patients in the group A are treated with DFDBA. Surgical process involves routine reflection of mucoperiosteal flap, exposing the bone site like in cystic defect cases. In case of cystic defect enucleation of cyst was done. In case of other osseous defects, the defect site itself act as anatomical cavity. The bony defect was packed with DFDBA. The materials can be used either by making with normal saline paste or in mixing with patients own blood. The wound was closed by suture vicryl 3-0 is used in all if cases.

The patients In the group B are treated with combination of DFDBA and HYDROXYAPATITE in equal volumes.

**Evaluation:**

- a) 1<sup>st</sup> postoperative day: Pain and swelling evaluated clinically.
- b) 1<sup>st</sup> postoperative week: Pain, swelling and wound dehiscence evaluated clinically and bone density radiographically with grey scale histogram.
- c) 4<sup>th</sup> post operative week: Pain swelling, wound dehiscence evaluated clinically and bone density radiographically with grey scale histogram.
- d) 12<sup>th</sup> postoperative week: Pain, swelling evaluated clinically and bone density radiographically with grey scale histogram.

**Measurement of study variables:**

- 1) VISUAL ANALOG SCALE of 0 to 10 was used to estimate pain by subjectively asking the patient to rate the nociceptive experience.
- 2) SWELLING was assessed by measuring the distance between the various anatomical landmarks, depending upon location of swelling e.g., mid pupillary line in upper half face, angle of mouth, ala tragus line etc. The mean difference between the preoperative and postoperative measurements was calculated.
- 3) BONE DENSITY The dimensions of the defects were evaluated on the preoperative radiographs mesiodistally and follow up was based on clinical and radiographic examinations at 1<sup>st</sup> day ,1<sup>st</sup> week, 4<sup>th</sup> week and 12<sup>th</sup> week after surgical treatment to evaluate the reduction in size of the residual cavity. Also change in bone density in the 1<sup>st</sup> day ,1<sup>st</sup> week, 4<sup>th</sup> week and 12<sup>th</sup> week were compared with the immediate postoperative radiographs. The radiographic findings were analyzed both subjectively and by using a digital technique to reduce the bias derived from the subjective evaluations. The computer analysis was performed using a PC (intel core I5, 2.3.0 ghz, Intel Corporation) with Adobe photoshop software to transfer the areas on the radiograph into pixels. A Digital camera (sony hx 10v) was used to photograph the radiographs on x ray viewer. Following the measurements of density of bony defects according to radiographs, they were recorded for statistical evaluation.

**Results:**

20 patients reporting to Department of Oral and Maxillofacial Surgery, Al-Ameen Dental College and Hospital, Bijapur, with defects were selected for the study. Patients with osseous defects were randomly divided into two groups. GROUP -A osseous defects to be treated with DFDBA. GROUP-B osseous defects treated with DFDBA and HYDROXYAPATITE in equal volume.

Assessment of postoperative swelling was done at 1<sup>st</sup> day ,1<sup>st</sup> week, 4<sup>th</sup> week and 12<sup>th</sup> week. Pain was assessed using pain analogue scale by asking the patient questionnaires at 1<sup>st</sup> day ,1<sup>st</sup> week, 4<sup>th</sup> week and 12<sup>th</sup> week. Radiological assessment was done using proper radiographs to document the osseous fill at 1<sup>st</sup> day ,1<sup>st</sup> week, 4<sup>th</sup> week and 12<sup>th</sup> week. greyscale evaluation of radiographs was done and bone formations at both .

**Results of clinical assessment Assessment of swelling:**

All the patients had mild to moderate swelling in GROUP A on first post-operative day. Swelling decreased gradually towards normal. By the end of first week post-operative, swelling was minimal.

In case of GROUP B, all patients had moderate to severe swelling on first post-operative day. Swelling was maximum from 1<sup>st</sup> post-operative week and then started decreasing towards normal. By the end of first week post-operative, slight swelling was still present.

**Assessment of pain**

All the patients had mild to moderate pain on GROUP A on first post-operative day. Pain was maximum on 2<sup>nd</sup> post-operative day and decreased gradually and almost nil by the end of 4<sup>th</sup> post-operative day.

In case of GROUP B , all patients had moderate to severe pain on first post-operative day. Pain was maximum from 2<sup>nd</sup> to 4<sup>th</sup> post-operative day and then gradually decreased but slight pain was there at the end of first post-operative week.

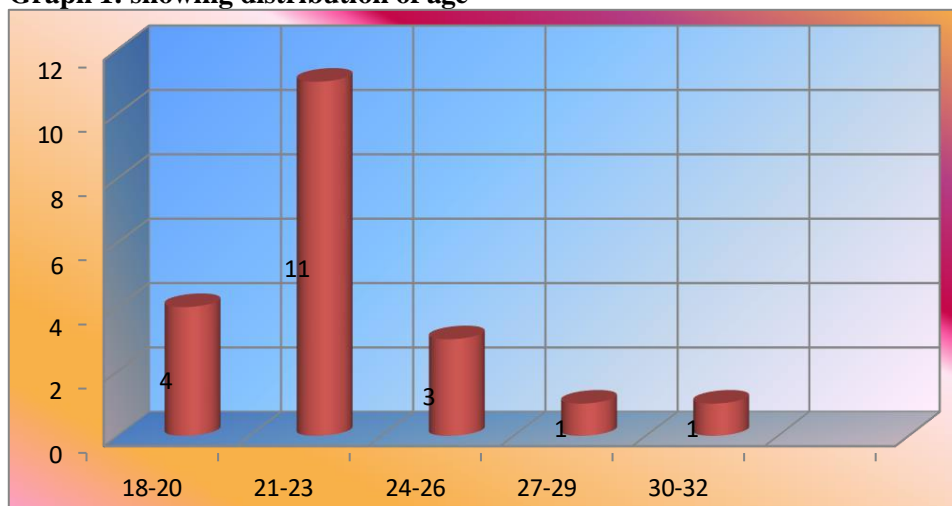
**Assessment of bone density**

Bone density was assessed by comparing bone formation on both groups using suitable radiographs at 1<sup>st</sup> day ,1<sup>st</sup> week, 4<sup>th</sup> week and 12<sup>th</sup> week by grey scale histogram. Mean difference of bone formation was moderately significant in graft site at 4<sup>th</sup> week post operative as compared to non-graft site. At 12<sup>th</sup> month post operative there was no significant difference between both group densities.

**Table 1: showing distribution of age**

Age	Group A and Group B
18-20	4
21-23	11
24-26	3
27-29	1
30-32	1
<b>Mean±SD</b>	<b>22.5 ± 2.554</b>

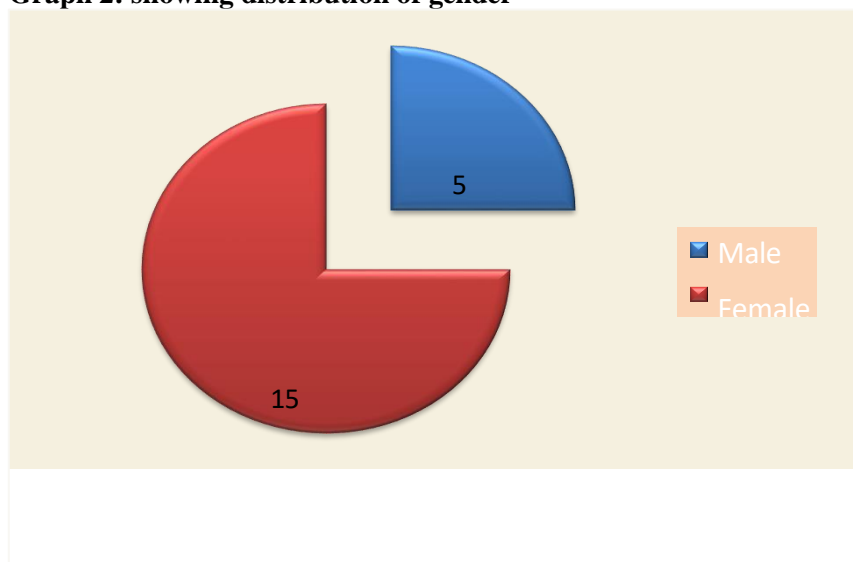
**Graph 1: showing distribution of age**



**Table 2: showing distribution of gender**

Sex	GROUP A AND GROUP B
Male	5
Female	15

**Graph 2: showing distribution of gender**

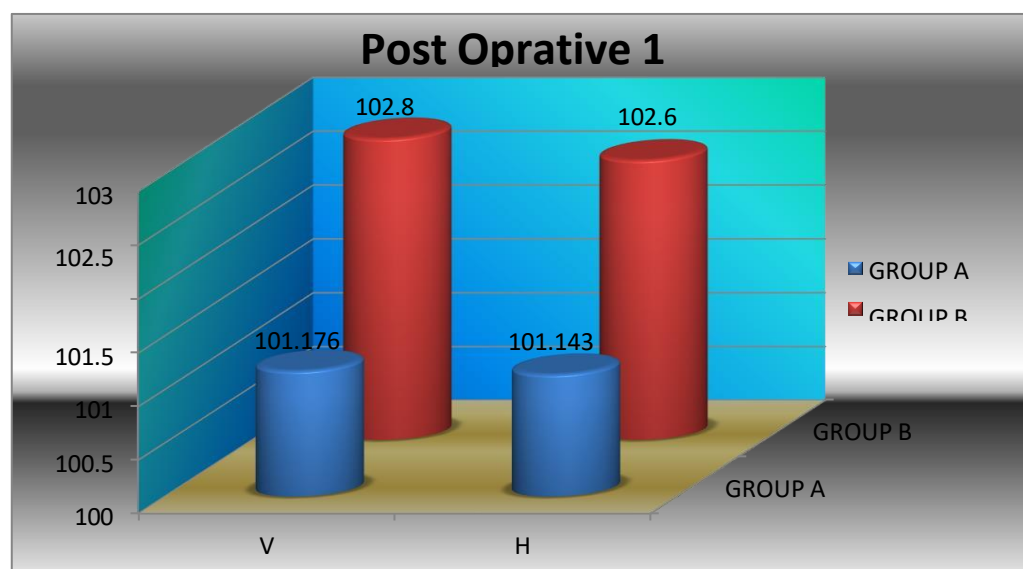
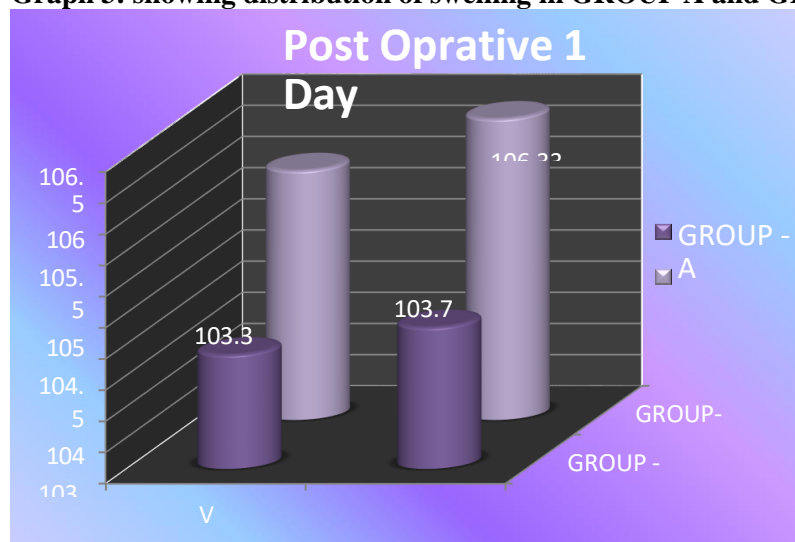


**Table 3: showing mean of swelling**

Swelling	GROUP A	GROUP B	Mann Whitney U test
Pre Operative	101.77±3.098 101.0±3.485	101.77±3.098 101.0±3.485	
Post Operative 1 Day V	103.33±.0133	105.5±2.945	

H	103.77±3.544 (103.49±3.312)	106.33±3.104 (105.46±3.024)	<b>U=1131.4</b> <b>P=0.0005 HS</b>
Post Operative 1 week	101.176±3.088	102.8±2.797	
V	101.143±3.472	102.6±3.15	<b>U=1383.0</b> <b>P=0.0282 S</b>
H	(101.4±3.269)	(102.7±2.942)	

**Graph 3: showing distribution of swelling in GROUP A and GROUP B**

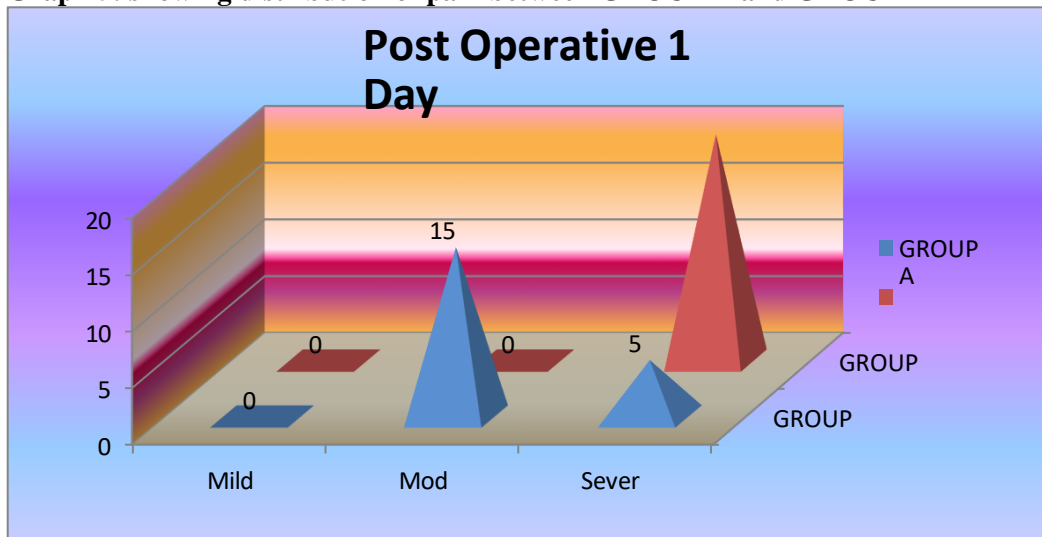


**Graph 4: showing distribution of swelling of GROUP A and GROUP B**

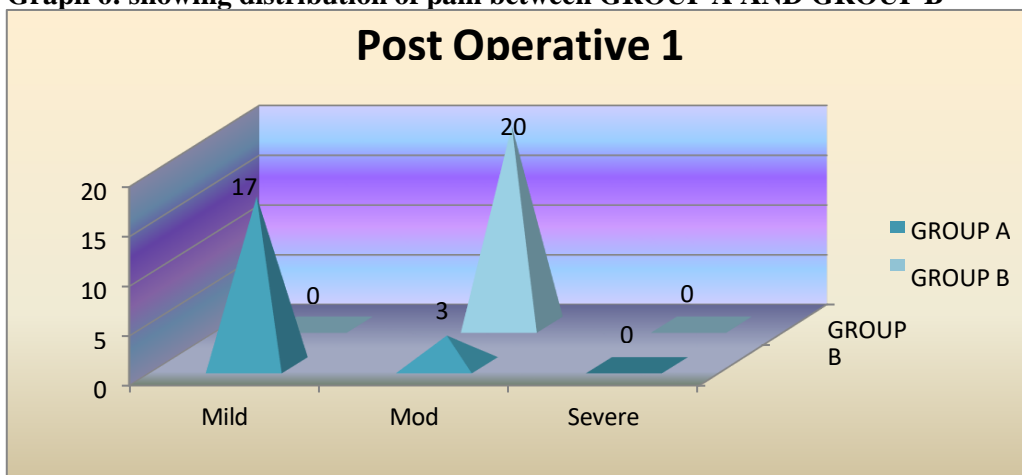
**Table 4: showing distribution of pain**

Pain	Group A	Group B	Fisher's exact test
Pre Operative Nil	20	20	
Post Operative 1 day Mild			<b>P&lt;0.0001 HS</b>
Moderate	0	0	
Severe	15	0	
Post operative 1 <sup>st</sup> week Mild			<b>P&lt;0.001 HS</b>
Moderate	17	20	
Severe	3	0	
	0		

**Graph 5: showing distribution of pain between GROUP A and GROUP B**



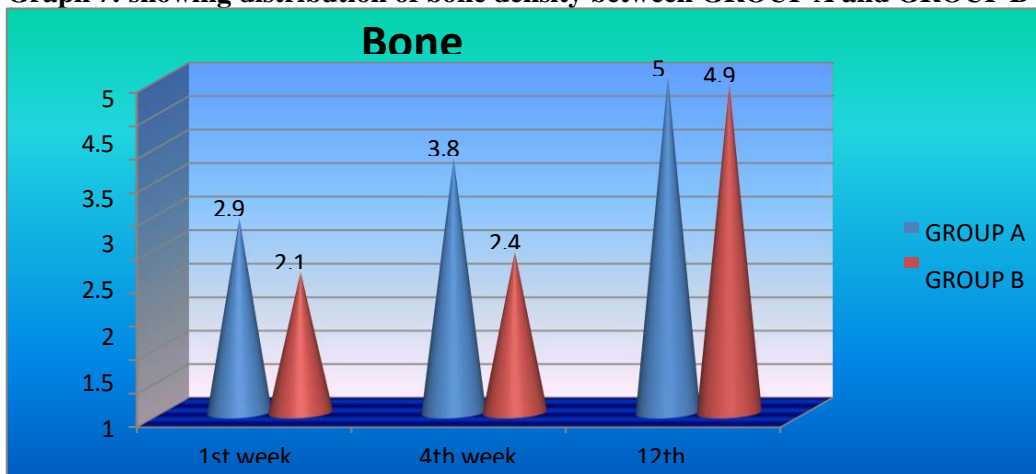
**Graph 6: showing distribution of pain between GROUP A AND GROUP B**



**Table 7: showing mean of bone density**

Bone density	Mean ± SD		Mann Whitney U test
	GROUP A	GROUP B	
1 <sup>ST</sup> WEEK	2.9 ± 0.3862	2.165 ± 0.406	p<0.0001 HS
4 <sup>TH</sup> WEEK	3.8 ± 0.4068	2.466 ± 0.507	p<0.001 HS
12 <sup>TH</sup> WEEK	5 ± 0.00	4.9.0.305	

**Graph 7: showing distribution of bone density between GROUP A and GROUP B**



## Discussion:

The allograft, DFDBA and the alloplastic material, HA were tried in this study due to their osteoinductive and osteoconductive properties respectively and their availability in sufficient quantities as and when needed. There have been numerous encouraging results with both these bone substitutes. Since both these materials were found to be useful, it was thought to combine them together and to assess their collective effectivity in bone formation and to see whether these collectively offered any additional advantage as opposed to their singular use.

The DFDBA was used because the very processes involved in its preparation i.e., decalcification exposes on its surface, the bone morphogenetic proteins (BMPs) which are osteoinductive that is, they induce differentiation of mesenchymal cells into cartilage and bone.<sup>15</sup>

BMPs are natural proteins which play important roles during embryogenesis and mediate in specific aspects of skeletal growth and development during later adult life. BMPs have been extracted from bone, dentin, and osteosarcoma tissue. Thus, in group A, the bone formation ensued immediately after filling DFDBA because of the BMPs which were exposed during the decalcification process. Also, the freeze drying at -196°C destroys the antigenicity<sup>15</sup> of the DFDBA. Porous HA is an abundantly available nontoxic material that is bioactive and allows new bone to be formed directly on its surface without any intervening layer of fibrous tissue. Synthetic HA possesses a similar composition to HA crystals present in bone, enamel and dentin and exhibits osteoconduction by acting as a scaffold for new bone to grow through the implant material as long as there is enough vital host bone surrounding it.<sup>15</sup>

HA, when implanted does not evoke an inflammatory or foreign body response and has a good tissue tolerance. Thus, in group B, the initiation of bone formation is different i.e., by osteoconduction alone as the HA granules used merely act as a trellis for blood vessels.<sup>15</sup> Migration of osteoblasts from surrounding healthy bone, a process called “creeping substitution”, whereby the HA is slowly resorbed and replaced by bone.

HA when used with DFDBA as opposed to their single use also showed good results as a synergistic effect of osteoconduction and osteoinduction was observed. Osteogenic induction is the vital process that a tissue or products derived from it causes a second undifferentiated tissue to differentiate into bone. The need for allogenic grafts in surgery has promoted the developing of the most suitable, the fastest, the easiest, the least risky, and the healthiest method for obtaining grafts, from the host, donor, and surgeon points of view.

Unlike in other tissues, the majority of bony injuries (fractures) heal without the formation of scar tissue, and bone is regenerated with its pre-existing properties largely restored, and with the newly formed bone being eventually indistinguishable from the adjacent uninjured bone.

However, there are cases of fracture healing in which bone regeneration is impaired, with, for example, up to 13% of fractures occurring in the tibia being associated with delayed union or fracture non-union. In addition, there are other conditions in orthopedic surgery and in oral and maxillofacial surgery in which bone regeneration is required in large quantity (beyond the normal potential for self-healing), such as for skeletal reconstruction of large bone defects created by trauma, infection, tumor resection and skeletal abnormalities, or cases in which the regenerative process is compromised, including avascular necrosis and osteoporosis.<sup>3</sup>

Bone grafting is a commonly performed surgical procedure to augment bone regeneration in a variety of orthopedic and maxillofacial procedures, with autologous bone being considered as the ‘gold standard’ bone-grafting material, as it combines all properties required in a bone graft material: osteoinduction (bone morphogenetic proteins (BMPs) and other growth factors), osteogenesis (osteoprogenitor cells) and osteoconduction (scaffold). It can also be harvested as a tricortical graft for structural support or as a vascularized bone graft for restoration of large bone defects or avascular necrosis. A variety of sites can be used for bone-graft harvesting, with the anterior and posterior iliac crests of the pelvis being the commonly used donor sites.<sup>3</sup>

The extent of bone induction by decalcified bone is a function of the surface area of the implanted bone; therefore, powder provides the maximum surface area necessary for interaction with recipient target cells.’ The decalcified bone graft appears to heal by osteoinduction. The BMP that the decalcified bone contains stimulates conversion of host fibroblast to chondroblasts on contact; osteogenesis follows chondrogenesis, which leads to new bone formation.

As the grafted material in the current study is decalcified, it appears radiolucent on the radiograph taken immediately postoperatively. This allows the use of careful radiographic follow-up of density increase as bone formation occurs. This is in contrast to studies in which non decalcified freeze-dried allogeneic bone or xenogeneic bone were grafted to bone defects; the immediate postoperative radiograph showed a radiopacity, almost like normal bone, and during the month after the operation the radiodensity diminished, associated



with resorption

And remodeling of the graft. Resorption of a standard mineral-containing bone graft has been estimated to be in the range of 30% to 70% of graft bulk.

An advantage of the decalcified bone graft is that it bypasses this process. Resorption does not occur in the decalcified bone graft, and bone healing starts immediately. This is probably the reason for the faster bone healing observed in their study, which is in contrast to the longer healing period with mineral-containing grafts. A concern about grafting bone obtained from cadaveric sources has been the potential transmission of infectious diseases, notably human immunodeficiency virus. A recent work, however, has shown that the demineralization process effectively destroys the viruses. It can be concluded from the current study that grafting of allogeneic decalcified bone to large jaw defects enhances bone formation and should be considered as an alternative to autogenous bone grafting.<sup>4</sup>

**The mechanisms for bone formation Induced by DFDBA could be:**

a) The demineralized particles undergo a biochemical change that brings about the remineralization of the particles.

b) The demineralized particles are colonized by mononuclear osteoclasts from the neighbouring bone, that are able to attract osteoblasts on their surface, thus allowing successive bone layering.

c) In the FDDBA particles we found, on the contrary, that many particles were completely surrounded by newly formed bone. Even the particles that were farthest from the host bone were lined by osteoblasts actively secreting osteoid and newly formed bone.

In conclusion, according to our histological results, the main differences between these two allografts seem to be as follows.

1) In FDDBA the resorption processes are very scarce and it has not been possible to find cells positive for ACP, while, on the contrary, in the DFDBA particles the resorption processes were present and cells positive for ACP were found.

2) In FDDBA even the particles farthest from the host bone were lined by or embedded in newly formed bone, while the DFDBA particles located far from the host bone tended to be surrounded by a scarcely cellular connective tissue, composed mainly of collagen fibres.

3) In FDDBA all the osteolytic lacunae were filled by osteocytes and in some areas Haversian systems with a capillary at the centre were found, while in the DFDBA the osteolytic lacunae tended, for the most part, to remain empty.

4) FDDBA and DFDBA did not show any osteoinductive effects.<sup>6</sup>

The clinical demand for bone void fillers that obviate the need to harvest autograft has prompted the development of synthetic and biological autograft substitutes. Among the Clinically most successful filler is DBM. DBM is osteoconductive, osteoinductive, and relatively easy to use clinically, especially in carrier formulations of diverse offerings.

The following are the criteria for ideal graft materials: the ability to facilitate osteogenesis, Stability of the implant when placed with the graft, low risk of infection, ease of availability, low antigenicity, and high level of reliability. The chosen implant material must provide the proper viable bone to stabilize the implant and facilitate osseointegration. The viability of the implanted bone is important in the long-term maintenance of the implant.

The success rate of implantation can be increased by guiding the osseointegration of the bone defect with bone grafting on top of the implant. Materials such as autogenous, allogenic, and xenogeneic bones, as well as synthetic materials, can be used.

The volume of bone formed is related to the quantity of BMP present. Factors/proteins present in DBP stimulate

1. Migration and attachment of cells at the healing site,
2. Proliferation of cells,
3. Biosynthetic activity by cells, and
4. Chondroblastic and osteoblastic cell differentiation.

DBP has been extensively used, often with controversial results. Recent studies showed that osteoinductive proteins, such as BMPs, enhanced osteoblast differentiation but not cell proliferation. In contrast, other researchers reported that DBP has a lower osteogenic capacity and has produced a significantly diminished degree of osseointegration. Becker et al reported that DBP promoted the least amount of new bone within the osseous defects. Pansegrau et al reported diminished integration of implants grafted with DBP. In contrast, Landsberg et al reported that DBP is capable of promoting bone formation around dental implants if

complete flap coverage and membrane presence can be maintained throughout the healing phase. The goal of this study was not to quantify the amount of regeneration that occurred but rather to determine whether any regeneration ever occurred.<sup>7,8</sup>

Freeze-dried bone allograft and decalcified freeze-dried bone allograft have been compared with porous particulate hydroxyapatite. Some studies suggest only a moderate difference in favor of the allografts when post-treatment clinical parameters are compared. Another study suggests a slight difference in favor of the alloplast. Decalcified freeze-dried bone allograft has also been compared with polylactic acid granules treatment of periodontal bone defects. A statistically significant improvement was found in the fill of the osseous defects when using decalcified freeze-dried bone allograft compared with the polylactic acid granules. The major difference between allografts and synthetic grafts is in the histologic results. Allografts heal by regeneration of the periodontium, whereas grafts of synthetic bone heal by encapsulation of the graft particles by connective tissue.<sup>15</sup>

While both DFDBA and FDBA are osteoconductive, only DFDBA has been proven to be osteoinductive. In the 1960s, Urist et al. showed that demineralized bone has osteoinductive potential by stimulating bone formation in extra skeletal sites. The osteoinductive potential of DFDBA is related to the amount of bone morphogenetic proteins (BMP) that remain after commercial processing has been completed. Shigeyama et al. detected BMP and in a commercial lot of demineralized bone matrix. Schwartz et al tested commercial lots of DFDBA from 6 different bone banks and found that most of the lots were able to induce ectopic bone formation when placed in a nude mouse muscle, while other lots did not induce new bone at all. In a ridge preservation study by Becker et al. DFDBA failed to show any signs of osteoinduction while autologous bone grafts had significant new bone formation. The BMPs in DFDBA can either be active or inactive due to a number of factors; if inactive, the inductive properties are lost. The release of these self-contained BMPs stimulates the differentiation of mesenchymal cells to osteoblasts in a location such as muscle, even though bone does not normally form there. While the absence of BMPs from DFDBA will eliminate inductive capabilities in the nude mouse model, the addition of BMP-2 has been shown to restore the osteoinductive nature of DFDBA. In addition, an osteopromotor such as enamel matrix derivative (EMD) will increase the osteoinductive potential of active DFDBA.<sup>18</sup>

Augmentations in the vertical dimension have mainly been performed using autogenous bone grafts, either as intraorally harvested blocks or as particulate supported by a space-keeping device. In maxillary sinus floor elevations using the lateral window technique, the following grafting protocols may be considered well-documented: coagulum (in combination with immediate implant placements), autogenous particulate alone or in combination with DBM or DFDBA, DBBM alone or in combination with DFDBA, and an alloplastic HA alone. The best documented sinus grafting materials using the trans alveolar approach are coagulum, particulate autograft, and DBBM.<sup>19</sup> The current widespread use of decalcified freeze-dried bone allograft (DFDBA) is based on the purported osteoinductive ability of bone graft preparations. Demineralization of the graft exposes the bone inductive proteins located in the bone matrix and in fact, may activate them.<sup>20</sup>

Human allografts can be classified into cortical, cancellous, and cortico-cancellous allografts according to their source. They can also be classified into freeze-dried bone allografts (FDBA) and decalcified freeze-dried bone allografts (DFDBA), also known as demineralized bone matrix (DBM), according to their decalcification process. There have been many studies comparing the osteoinductive effects of FDBA and DFDBA. However, the differences in osteoinductive effects among cortical, cancellous, and cortico-cancellous human bone have not yet been reported.<sup>21</sup>

Although autogenous bone grafts seem to be preferable as a grafting material, a meta-analysis by tong et al reported comparable success rates of implants placed in sinuses grafted with different materials including HA, DFDBA, and autogenous bone. Limitations and side

Effects related to autogenous grafts should also be considered. A second surgical site, the increase in surgical time, patient morbidity, and the need for hospitalization and general anesthesia should be weighed against therapeutic alternatives that may be less invasive and expensive. Bone substitutes have the advantage of being readily available, with no limitations in their procurement. Furthermore, they can be considered safe in terms of disease transmission.<sup>24</sup> Bryan c Mendelson et al shows that porous hydroxyapatite granules maintain bony and overall projection at 2 years when used for augmentation of the facial skeleton in the aesthetic patient. The completeness of the volume maintenance, although only over a 2-year follow-up in our study, suggests that the volume enhancement may be permanent.<sup>26</sup>

David c Greenspan carried out a study which concludes that Bone substitutes are in great demand in the treatment of various disorders like periodontal diseases, dental periapical abscess, bone tumors, trauma and other bone defects. The use of autogenous bone has remained the gold standard in restoring bone defects, but it is not always possible to obtain enough bone or the amount of bone needed may

exceed than the available<sup>27</sup> Johannes Franz Honig, Hans Albert Merteen, Axel Nitsch and Raphaela Verhaegen carried out study on contouring of cranial vault with hydroxyapatite cement. Their study was performed on group of patients ranges from 23 to 57 average age was 38.5 years. All of the patients were male. Their study shows that Hydroxyapatite cement (Bone Source) will gradually be reabsorbed and replaced by bone, if not the internal table together with the external table calvaria bone. It permits osseointegration, which makes it relatively resistant to infection. The substrate is available in amounts (volumes) that are easy to apply and shape to suit individual needs<sup>28</sup>

In this study, 20 patients reporting to Department of Oral and Maxillofacial Surgery, Al-Ameen Dental College and Hospital, Bijapur, with defects were selected for the study. Patients with osseous defects were randomly divided into two groups. GROUP -A osseous defects to be treated with DFDBA. GROUP-B osseous defects treated with DFDBA and HYDROXYAPATITE in equal volume.

Assessment of postoperative swelling was done at 1<sup>st</sup> day ,1<sup>st</sup> week, 4<sup>th</sup> week and 12<sup>th</sup> week. Pain was assessed using pain analogue scale by asking the patient questionnaires at 1<sup>st</sup> day ,1<sup>st</sup> week, 4<sup>th</sup> week and 12<sup>th</sup> week. Radiological assessment was done using proper radiographs to document the osseous fill at 1<sup>st</sup> day ,1<sup>st</sup> week, 4<sup>th</sup> week and 12<sup>th</sup> week. Greyscale evaluation of radiographs was done and bone formations at both. This study was conducted to determine effectiveness of healing ability of DFDBA and HA and aimed to evaluate the clinical outcome of Demineralized Freeze-Dried Bone Allograft as an osteoinductive bone replacing material for treating osseous defects in humans which is more effective than hydroxyapatite. The following conclusions were drawn from the study. DFDBA has high osteoinductive potential that can be used as an efficient bone replacing agent, which this study proved with the radiographical evidence of new bone deposition within 4 weeks. The demineralization process of the allograft exposes the bone morphogenic protein (BMP) of the graft material, making it a possible reason for hastening the bone healing, which was revealed in this study proving demineralized bone grafts are better than mineralized bone grafts. Usually, the initial phase of bone healing is resorption but present study concludes that demineralized bone allografts find a way around this phase of resorption and induces new bone formation within 1 month period, making Demineralized Freeze-Dried Bone Allograft clinically efficient. As already known autograft is the ideal bone replacing option, but present study has proved that decalcified bone allografts are the next promising option for bone replacement.

Further long-term studies should be directed towards the use of Decalcified Freeze- Dried Bone Allograft in the treatment of large osseous defects in the field of oral and maxillofacial surgery. DFDBA when used for the obliteration of osseous defects bypasses the phase of obligatory resorption and shows early evidence of new bone formation. HA when used for obliteration of osseous defects shows delayed first evidence of bone formation as compared to the decalcified freeze-dried bone matrix allograft and it undergoes resorption but takes a long period to resorb completely and be replaced by bone. The radiograph assessment grey scale histogram indicates early bone formation with DFDBA alone than the combination of DFDBA and HA.

### **Conclusion:**

Our study was conducted on 20 patients who underwent treatment for osseous defects were selected for the study. They were categorized in two groups of 10 each. GROUP A were treated by DFDBA and GROUP B were treated by HA and DFDBA. Treatment outcome was evaluated using many parameters like pain, swelling, wound dehiscence clinically and bone density radiographically pre operative, 1<sup>st</sup> day ,1<sup>st</sup> week, 4<sup>th</sup> week and 12<sup>th</sup> week post operative.

In our study, considering our results and experience in a comparative evaluation of DFDBA and its combination with HA in osseous defects of maxillofacial surgery, we observed that there is no statistically significant difference between DFDBA alone versus DFDBA mixed with HYDROXYAPATITE in terms of pain, swelling, wound dehiscence clinically and bone density radiographically. It can be concluded that DFDBA is alone better than DFDBA mixed with HYDROXYAPATITE.

DFDBA when used for the obliteration of osseous defects bypasses the phase Of obligatory resorption and shows early evidence of new bone formation. HA when used for obliteration of osseous defects shows delayed first evidence of bone formation as compared to the decalcified freeze-dried bone matrix allograft and it undergoes resorption but takes a long period to resorb completely and be replaced by bone. The radiograph assessment grey scale histogram indicates early bone formation with DFDBA than the combination of DFDBA and HA.

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