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Original Article

The Clinical and Radiological Outcome of Bovine Hydroxyapatite (Bio Hydrox) as Bone Graft

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ABSTRACT

Background: Bone grafting is a surgical procedure that involves transplanting material to enhance bone healing through osteogenesis, osteoconduction, and osteoinduction. While autografting has long been considered the gold standard, it has limitations, including a restricted supply and donor site morbidity. These limitations, along with those associated with allografts, have led to the development of bovine hydroxyapatite (BHA) as a bone graft substitute. This study aimed to evaluate the clinical and radiological outcomes of BHA.
Methods: This descriptive study used a consecutive sampling design, including all trauma patients who received BHA at our hospital between 2016 and 2018. The BHA was obtained from the tissue bank at our institution. The clinical and radiological outcomes were evaluated, and the data was tabulated and analyzed descriptively.
Results: Of the 56 patients who underwent surgery with BHA, most (80.36%) had excellent outcomes, 12.5% had good outcomes, 3.57% had fair outcomes, and 3.57% had poor outcomes.
Conclusion: Bovine hydroxyapatite can be considered an alternative bone graft to support the bone healing process.

Keywords: Bone graft; Bone healing; Bovine hydroxyapatite; Human and medicine; Osteoconductive

INTRODUCTION

Bone grafting is the transplantation of material into a recipient site to enhance bone healing through osteogenesis, osteoconduction, and osteoinduction.[1,2] Osteogenesis refers to the graft's cellular component, enabling bone apposition using the replaced tissue. Osteogenic grafts contain living cells, such as osteoblasts, which can differentiate into mature bone cells and directly contribute to new bone formation. Osteoconduction occurs when the graft provides a porous scaffold that facilitates the migration and adhesion of osteoblasts and progenitor cells from the surrounding host tissue. The interconnected pores of an osteoconductive graft allow for the ingrowth of new blood vessels, which supply the oxygen and nutrients essential for bone healing. Osteoinduction is the graft's ability to stimulate the differentiation of undifferentiated stem cells or osteoprogenitor cells into osteoblasts. Osteoinduction is a complex process involving a cascade of growth factors and signaling molecules that regulate cell proliferation and differentiation. Osteoinductive grafts typically contain bone morphogenetic proteins (BMPs), which are potent signaling molecules that can induce bone formation.[3]

Bone grafting is a common surgical procedure with a high demand, particularly in the United States, where over 1 million cases are performed annually. The demand for bone grafts is steadily increasing, with a projected growth rate of 13% per year.[2] VVarious bone graft options, including synthetic and natural materials, are offered by tissue banks. A significant amount of bone graft material is from The Cell and Tissue Bank Regenerative Medicine at Dr. Soetomo General Academic Hospital in Surabaya, Indonesia. In 2017, the use of approximately 3,763 packs of bovine bone graft, 282 human allografts, and 362 packs of bovine hydroxyapatite was recorded.[3]

Organic bone grafts are categorized into three types: autografts, allografts, and xenografts. The autogenous bone graft is considered the gold standard. It has minimal immunological rejection, excellent histocompatibility, and provides the best osteoinductive, osteoconductive, and osteogenic properties.[4] Autografts, harvested from the patient's own body, offer several advantages. They are highly biocompatible, minimizing the risk of immune rejection and disease transmission. Autografts also provide optimal osteoinductive properties, stimulating the body's natural healing processes. However, autografts have limitations, including donor site morbidity, limited availability, and the need for additional surgical procedures.

Due to these limitations, bone allografts, derived from deceased human donors, are considered an alternative for filling bone defects when autograft availability is insufficient. Allografts offer advantages such as size adjustability and the possibility of mass production. However, potential drawbacks of allografts include disease transmission, rejection reactions, delayed union, and limited donor availability. To mitigate these risks, allografts undergo rigorous processing, including sterilization and tissue banking, to ensure safety and efficacy.[6–8]A proper and meticulous process is essential for the production of bone allografts.

To address the limitations of autografts and allografts, bone xenografts have been developed. Xenografts, derived from other species such as bovine sources, offer abundant availability, osteoconductive properties for mechanical support, and have a low production cost. The production process involves high-temperature incineration at 1000°C to eliminate protein components from the bovine bone graft.[9] Xenografts are obtained through freeze-drying, demineralization, and deproteinization. However, freeze-drying can cause significant degradation of bone strength. Methods adapted from Professor Frank Dexter of Tissue Bank Yorkshire and implemented by various tissue banks in the Asia Pacific aim to mitigate this issue, but some reduction in strength may still occur.[10,11]

Another bone graft material, Bovine hydroxyapatite is also obtained from bovine bones through a freeze-drying process. The material is harvested from bovine bones where all organic components have been extracted (deproteinized) by a furnacing process. The process consists of several steps: dissection, division, cleansing, freeze-drying, washing, and finally, oven-drying. The hydroxyapatite produced from bovine bone undergoes rigorous testing for biocompatibility, microstructure, and composition.[9]

Bovine hydroxyapatite has been developed in our hospital institution and has been widely used in orthopedic cases. This study aims to evaluate the clinical and radiological outcomes of bovine hydroxyapatite as a bone graft.

METHODS

Study Design

This is a descriptive study examining the clinical and radiological outcomes of patients who underwent orthopedic surgery with bovine hydroxyapatite bone grafts.

Sample Selection

The samples were obtained by consecutive sampling. All patients who underwent surgery with bovine hydroxyapatite from January 2016 to December 2018 and consented to attend a scheduled check-up in the Orthopedic Outpatient Clinic or to be home-visited were included in the study. Due to the limited number of subjects, all patients meeting the inclusion criteria during the study period were evaluated.

Inclusion and Exclusion Criteria

The inclusion criteria included: 1) Patient with a fracture in a single bone; and 2) Patient with a non-union fracture. The exclusion criteria included: 1) Patient with multiple fractures; 2) Patient with a pathological fracture due to a bone tumor; 3) Patient with osteomyelitis; and 4) Patient with an open fracture Grade 3B based on the Gustilo-Anderson classification.

Follow-up and Data Collection

Patients were followed up for a minimum of 6 months before clinical and radiological assessments were performed. Data was collected from their patient medical records, including patient identity, address, and the results of the clinical and radiological check-ups from the Orthopedic Outpatient Clinic or home visits.

Outcome Assessment

A single assessor evaluated the clinical and radiological outcomes to prevent observation bias. The outcome assessment categories were:[12]

- Excellent: Good radiologic union with complete graft incorporation and complete functional recovery;
- Good: Radiologic union with partial graft incorporation and no need for orthotic protection.
- Fair: Poor graft incorporation with the need for orthotic protection.
- Poor: Marked fragmentation of the graft with no radiologic incorporation and no functional improvement over the preoperative condition.

Ethical Considerations

This study was approved by the Institutional Ethics Committee of Dr. Soetomo General Academic Hospital, Surabaya, Indonesia (Ethical Clearance No. 1353/KEPK/VII/2019).

Data Analysis

The data was recorded and tabulated according to demographic characteristics. Descriptive analysis was performed to elaborate on the clinical and radiological outcomes without using comparative statistical analysis.

RESULTS

Eighty-six trauma patients underwent surgery using bovine hydroxyapatite in our hospital institution between January 2016 and December 2018. However, only 56 patients had complete medical records and were available for evaluation. The remaining patients could not be evaluated due to loss to follow-up.

Table 1 provides details on the age distribution of the patients in this study. The majority of patients were between 41 and 50 years old (39.28%), followed by the 31-40 years age group (25%). The 51-60 year

Table 1. Patient demographics.

Groups	N	Percentage (%)
Age		
< 20 years old	5	8.92
21-30 years old	9	16.07
31-40 years old	10	17.86
41-50 years old	22	39.28
51-60 years old	8	14.28
Bone		
Humerus	11	19.64
Radius	4	7.14
Femur	22	39.28
Tibia	17	30.36
Metacarpal	1	1.78
Phalanges	1	1.78
Location		
Metaphysis	31	55.36
Shaft	25	44.64
Outcome		
Excellent	45	80.36%
Good	7	12.5%
Fair	2	3.57%
Poor	2	3.57%

Table 2. Patient distribution based on cases.

Pathology	Bone region	Numbers
Tibial plateau fracture	Tibia	13
Lower end radius fracture	Radius	3
Upper-end humerus fracture	Humerus	5
Intercondylar fracture	Femur	9
	Humerus	4
Non-union	Femur	4
	Humerus	2
	Tibia	2
Comminuted fracture	Femur	9
	Tibia	2
	Radius	1
	Metacarpal	1
	Phalanx	1
Total		56

Table 3. Patient distribution based on the complication.

Complication	Numbers	Percentages
Infection	2	3%
Implant failure	2	3%
Nerve lesion	0	-

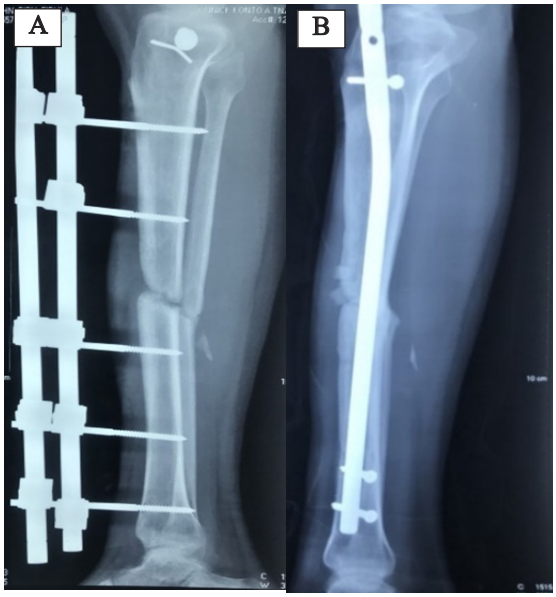

Figure 1. (A) Non-union case of tibial fracture in external fixator application and (B) The results 6 months post-application of the external fixation and addition of bone hydroxyapatite inside the fracture site.

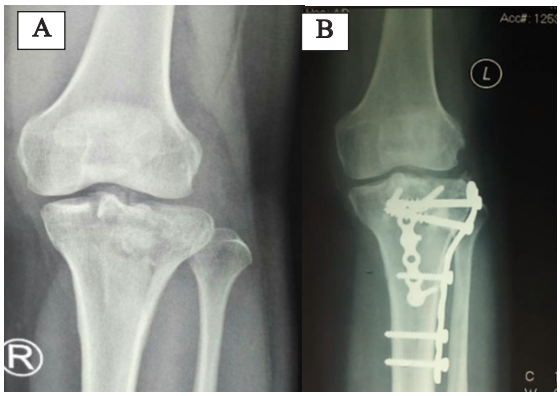

Figure 2. (A) Tibial plateau fracture ore-ORIF and (B) post-ORIF with a plate and screw added alongside the bone graft.

age group accounted for 21.43% of patients, while only two subjects were over 60 years old. The most frequent defect location was the metaphysis (55.36%). Most patients (80.36%) had excellent outcomes, 12.5% had good outcomes, 3.57% had fair outcomes, and 3.57% had poor outcomes.

The femur was the most common bone region requiring grafting (39.28% of patients). Table 2 shows that the most common fractures were tibial plateau fractures (13 cases), followed by intercondylar femur fractures (9 cases).

Post-surgical complications occurred in four patients, including two cases of infection and two cases of implant failure (Table 3).

Figure 1A illustrates a case of non-union in a patient treated with an external fixator. Despite prolonged fixation, the fracture site failed to heal. To address this, bone hydroxyapatite was added to the fracture site six months after the initial application of the external fixator. The results were excellent, as demonstrated by Figure 1B, which shows the healed fracture site with complete bone union and restoration of bone continuity.

Figure 2 illustrates a complex tibial plateau fracture with significant joint depression and comminution. The fracture fragments were displaced and unstable, making surgical repair challenging. A bone graft was implanted to fill the defect, restore the articular surface, and provide structural support for healing. The pre-operative (Figure 2A) and post-operative (Figure 2B) images demonstrate successful fracture union, the reduction of joint depression, and restoration of the articular surface. The bone graft integrated well with the host bone, facilitating healing and improving long-term functional outcomes.

DISCUSSION

Bone grafts play a crucial role in bone healing and reconstruction by fulfilling several essential functions. They provide a scaffold for new bone growth, promote the migration and differentiation of bone-forming cells, and contribute to the restoration of bone structure and function. Autografts and allografts have traditionally been used for bone grafting but they are limited by donor site morbidity, immunological risks, and limited availability.

Bovine hydroxyapatite has emerged as a promising alternative to autografts and allografts due to its numerous advantages. It offers abundant availability, excellent biocompatibility, and osteoconductive properties, making it a suitable choice for various bone grafting applications. Unlike autografts and allografts, bovine hydroxyapatite does not carry the risk of disease transmission or immune rejection, further enhancing its safety profile. Additionally, its ease of processing and storage makes it a convenient option for clinical use.[13]

The Cell and Tissue Bank of Regenerative Medicine at Dr. Soetomo General Academic Hospital in Surabaya, Indonesia, has developed a variety of bone grafts. Fresh-frozen bone allografts are used for larger bone defect reconstruction. However, their distribution presents challenges due to the requirement for a -80°C cold chain for storage.

In addition to other graft types, the Cell and Tissue Bank of Regenerative Medicine at Dr. Soetomo General Academic Hospital in Surabaya, Indonesia, produces freeze-dried bone allografts. These grafts have a low water content (less than 8%) due to the sublimation process, making them easier to store and distribute at room temperature.[9]

Freeze-dried bone allografts can be processed in several ways. Initially, both organic and inorganic components are present. Deproteinization removes the organic components, leaving only inorganic mineral hydroxyapatite. Alternatively, demineralization can be performed to produce grafts containing only organic compounds, known as Demineralized Bone Matrix (DBM).[9]

Due to the high demand for bone grafts, our tissue bank produces bovine hydroxyapatite (Bio-Hydrox) as a cost-effective alternative. Bovine hydroxyapatite has demonstrated a high success rate and lower expense in orthopedic surgeries.[14]

This study evaluated the efficacy of bovine hydroxyapatite as a bone graft in 56 patients. The patients' ages ranged from 11 to 68 years. Age and gender are important factors influencing pain perception, with children, the elderly, and women typically experiencing greater pain sensation than men. These factors were considered in the pain evaluation of patients who underwent surgeries using bovine hydroxyapatite.

The most common fracture types treated with bone grafts in this study were metaphyseal (56%), comminuted (31%), and tibial plateau (20%). These findings are consistent with the research by Shibuya et al., which showed the effectiveness of bovine hydroxyapatite in reconstructing, elevating, and restoring joint depression contours. This allows for good structural strength, particularly in leg and ankle surgeries. Successful rehabilitation relies on achieving new ranges of motion, consistent exercise, and appropriate weight-bearing.[15]

The success rate of bovine hydroxyapatite in this study was very high. Most patients achieved excellent (80.36%) or good (12.5%) outcomes, with only a small percentage experiencing fair (3.57%) or poor (3.57%) outcomes. Mahyudin et al. conducted a study using rabbits and found similar results, demonstrating comparable bone healing with bovine hydroxyapatite and allografts.[16] Hydroxyapatite, with or without bone marrow, is a compelling option for cancellous bone grafting, although its efficacy in cortical bone requires further investigation.[1,17]

Tsai et al.'s study demonstrated the successful use of HA bone substitutes in 27 out of 33 patients. After 6-12 months of follow-up, x-ray images showed proper bone healing and an 81.8% fusion rate. A notable finding of this study was the successful integration of sintered bovine HA, even when placed outside the cortex.[18]

Hydroxyapatite offers several advantages when it comes to bone regeneration. Chemically similar to natural bone, HA exhibits excellent biocompatibility, integrating seamlessly into the host bone without triggering an immune response. Its porous structure facilitates vascularization, promoting nutrient delivery and waste removal during the healing process. Additionally, HA's osteoconductive properties provide a scaffold for bone cell migration and proliferation, guiding new bone formation. This scaffold-like structure encourages the attachment and differentiation of osteoblasts, leading to the production of new bone matrix. A study by Kotobuki et al. explained that HA provides calcium and alkaline ions for osteoblasts, enabling them to mineralize extracellular mesenchymal tissue and secrete ATPase. It can also encourage cells to show an osteoblastic phenotype and form bone tissue.[19]

Four complications were observed in this study. Two patients with open fractures (a supracondylar femur fracture with bone loss and an intercondylar humerus fracture) who underwent open reduction and internal fixation with double plating and bone grafting had poor outcomes. Infection was suspected as the primary cause of non-union in these cases, as open fractures have a high risk of infection. This is consistent with the findings of Tsai et al., who also identified infection as a cause of bone graft failure.[18]

The use of bovine hydroxyapatite is relatively safe and has shown satisfactory results in fracture patients. This is attributed to its deproteinized nature and the fact that humans are routinely exposed to bovine proteins through the consumption of meat and dairy products. However, infection can occur, particularly in open fractures, and this can lead to non-union.[20]

This study has several limitations. The number and variety of cases were limited and may not represent the full spectrum of possible injuries. Comparative statistical analysis was not performed to evaluate outcomes based on injury type. Longer follow-up is needed to better understand the long-term outcomes of bovine hydroxyapatite in trauma patients.

Further research should include a larger and more diverse patient population with a longer follow-up to assess long-term outcomes. Comparative studies with other bone graft materials are needed to establish the relative effectiveness of bovine hydroxyapatite. Statistical analysis should be performed to evaluate the outcomes based on specific injury types and patient demographics. Investigation into the effects of different processing techniques and the use of combination therapies with other biologics may lead to improved graft performance. Finally, a cost-effectiveness analysis should be conducted to compare bovine hydroxyapatite to other bone graft materials.

CONCLUSION

This study suggests that bovine hydroxyapatite may be a suitable bone graft substitute as indicated by the descriptive clinical and radiological outcomes. Bovine hydroxyapatite appears to be a viable option for promoting bone healing, filling bone defects, and restoring bone contour and structure. However, further research with a larger sample size, longer follow-up, and comparative analysis is needed to confirm these findings.

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CONFLICTS OF INTEREST

The authors declare that there are no conflicts of interest.

REFERENCES

1. Elhalawany SK, Tarakji B, Azzeghaiby SN, Alzoghaibi I, Baroudi K, Nassani MZ. Clinical and radiographic evaluation of biodegradable bone plates in the treatment of mandibular body fractures. Niger Med J 2015;56(1):48.
2. Coleman BE. A clinical review of autogenous bone grafting for orthopaedics. The Avitus ® Bone Harvester; 2018.
3. Wang W, Yeung KW. Bone grafts and biomaterials substitutes for bone defect repair: A review. Bioact Mater 2017;2(4):224-47.
4. Samartzis D, Shen FH, Goldberg EJ, An HS. Is autograft the gold standard in achieving radiographic fusion in one-level anterior cervical discectomy and fusion with rigid anterior plate fixation? Spine (Phila Pa 1976) 2005;30(15):1756–61.
5. Brydone AS, Meek D, MacLaine S. Bone grafting, orthopaedic biomaterials, and the clinical need for bone engineering. Proc Inst Mech Eng H, J Eng Med 2010;224(12):1329–43.
6. Myeroff C, Archdeacon M. Autogenous bone graft: Donor sites and techniques. J Bone Jt Surg 2011;93(23):2227–36.
7. Hung NN. Basic knowledge of bone grafting. Zorzi A. Bone Grafting. Croatia: InTech, Chapters. 2012:11-38.
8. Mahyudin F. Bone grafts & bone substitute materials (characteristics and clinical application strategies) [Graf tulang & material pengganti tulang (Karakteristik dan strategi aplikasi klinis)]. In: Utomo DN, editor. Surabaya: Airlangga University Press; 2018. p. 57–102.
9. Mahyudin F and Rantam FA. Regeneration in massive bone defects with bovine hydroxyapatite as a scaffold for mesenchymal stem cells

[Regenerasi pada massive bone defect dengan bovine hydroxyapatite sebagai scaffold mesenchymal stem cell]. J. Biosains Pascasarjana 2011;13(3):179–95.
10. Cornu O. Influence of freeze-drying and irradiation on mechanical properties of human cancellous bone: Application to impaction bone grafting. Bone Grafting. 2012.
11. Hapuhinna K, Gunaratne R, Pitawala J. Comparison between differently synthesized hydroxyapatite composites for orthopedic applications. J Mater Sci and Chem Eng 2019;7(5):16-28.
12. Balakrishnan M, Agarwal D, Kumar S. A study of efficacy of heterogeneous bone grafts (Surgibone) in orthopaedic surgery. Med J Armed Forces India 2000;56(1):21–3.
13. Bano N, Jikan SS, Basri H, Adzila S, Nuhu AH. Natural Hydroxyapatite Extracted From Bovine Bone. J Sci Tech 2017;9(2):22–8.
14. Campana V, Milano G, Pagano E, Barba M, Cicione C, Salonna G, et al. Bone substitutes in orthopaedic surgery: from basic science to clinical practice. J Mater Sci Mater Med 2014;25(10):2445–61.
15. Shibuya I, Yoshimura K, Miyamoto Y, Yamada A, Takami M, Suzawa T, et al. Octacalcium phosphate suppresses chondrogenic differentiation of ATDC5 cells. Cell Tissue Res 2013;352(2):401-12.
16. Mahyudin F, Utomo DN, Suroto H, Martanto TW, Edward M, Gaol IL. Comparative effectiveness of bone grafting using xenograft freeze-dried cortical bovine, allograft freeze-dried cortical New Zealand white rabbit, xenograft hydroxyapatite bovine, and xenograft demineralized bone matrix bovine in bone defect of femoral diaphysis of white rabbit: Experimental study in vivo. Int J Biomater 2017;2017:7571523.
17. Fernandez de Grado G, Keller L, Idoux-Gillet Y, Wagner Q, Musset AM, Benkirane-Jessel N, et al. Bone substitutes: a review of their characteristics, clinical use, and perspectives for large bone defects management. J Tissue Eng 2018;9:1-18.
18. Tsai WC, Liao CJ, Wu CT, Liu CY, Lin SC, Young TH, et al. Clinical result of sintered bovine hydroxyapatite bone substitute: Analysis of the interface reaction between tissue and bone substitute. J Orthop Sci 2010;15(2):223–32.
19. Kotobuki N, Ioku K, Kawagoe D, Fujimori H, Goto S, Ohgushi H. Observation of osteogenic differentiation cascade of living mesenchymal stem cells on transparent hydroxyapatite ceramics. Biomaterials 2005;26(7):779–85.
20. Schnetzke M, Morbitzer C, Aytac S, Erhardt M, Frank C, Muenzberg M, et al. Additional bone graft accelerates healing of clavicle nonunions and improves long-term results after 8.9 years: A retrospective study. J Orthop Surg Res 2015;10(1):1–9.