Asian Journal of Research in Animal and Veterinary Sciences

4(3): 1-7, 2019; Article no.AJRAVS.51580

Bovine Skeleton Preparation Using Hot Water Technique for Anatomical Studies

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Authors' contributions

This work was carried out in collaboration among all authors. Author SMA designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors SAH and AZJ managed the analyses of the study. Author AHB managed the literature searches. All authors read and approved the final manuscript.

Article Information

Editor(s):
(1) Dr. Fabio da Costa Henry, Associate Professor, Technology of Animal Products, State University of Northern of Rio de Janeiro, UENF, Brasil.
Reviewers:
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(3) Weena Koeypudsa, Chulalongkorn University, Thailand.
Complete Peer review History: https://sdiarticle4.com/review-history/51580

Original Research Article

Received 01 August 2019
Accepted 05 October 2019
Published 17 October 2019

ABSTRACT

Aims: The aim of this study was to prepare and document the processing of skeleton of a bovine species and to determine the time involved in the preparation.
Place and Duration of Study: Department of Animal Health and Production, College of Agriculture and Animal Science Bakura, Zamfara State, Nigeria, between October 2018 and November 2018.
Methodology: The skeleton of a Red Bororo bull was prepared and mounted using a hot water technique comprising skinning, evisceration and de-fleshing of carcass, carcass maceration, bone drying, frame construction, drilling and articulation of bones and bone varnishing. The time taken for each of the steps was determined using a digital stop watch.
Results: The entire skeleton preparation process lasted for five (5) days and 11 hours. The steps

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and time recorded in the skeleton preparation of the bull were: Skinning, evisceration and de-fleshing of carcass (6 hours), carcass maceration (16 hours), bone drying (4 days), frame construction (30 minutes), bone drilling and articulation (12 hours) and varnishing (30 minutes) respectively.

Conclusion: The method has shown to be faster (less than a week) than burying method, and safer, easier and cheaper compared to previous researches which requires the use of dangerous chemicals (chlorine bleach and fumes) and expensive items (skeleton preparator's Standard Toolkit). It is concluded that drying was the principal time-consuming component in the skeleton preparation. It utilized 73% (4 days) out of the total time used in preparing the skeleton (5 days and 11 hours), while frame construction and varnishing had the lowest duration (30) minutes each.

Keywords: Bovine anatomy; skeleton; appendicular and axial bones.

1. INTRODUCTION

Bones and skeletons serve as teaching tool in academic institutions and as exhibitions in museums [1]. For several years skeletons have been prepared for use in identification of animal remains in archaeological and forensic studies. For academic purposes different skeletons have also been prepared for comparative and anatomical studies. Certain techniques for processing such skeletons have been described such as burial maceration, cold water maceration, hot water maceration, insect maceration, chemical maceration, and enzymatic maceration [2,3]. Each of these techniques comprise several steps and their major limitation is time consumption, laborious and meticulous processes of which maceration is usually the lengthy and critical stage [4]. The preparation and actual time utilized in the preparation skeleton of Pigeon, Squirrel, Deer and Crocodile has been reported [5]. However there are little or no information on the procedure and duration of skeleton preparation in a Bovine species.

The aim of this research was to extract, process and assemble the appendicular and axial skeleton of a Bovine, to bridge the gap of information on the skeletal preparation of Bovine species and to determine the percentage time utilized in all the steps involved in the skeleton preparation. The findings of this research will serve as a guide in the skeleton preparation of a Bovine and contribute to the knowledge and museum of anatomy. Through anatomy (osteology), one learns about the bones in both an anatomical and physiological manner. In doing so, we can better understand related aspects about the body, we can understand better ways to treat the animals, find certain ways to mend and aid fractured or broken bones [6].

2. MATERIALS AND METHODS

A Red Bororo bull was purchased from the Talata Mafara livestock market of Zamfara State and transported to the Department of Animal Health and Production, College of Agriculture and Animal Science Bakura, Zamfara State (Plate 1). The bull was slaughtered using halal method [7].

Fig. 1. A photograph showing the Red Bororo bull

The methods employed in skeleton preparation were: Skinning, evisceration and de-fleshing of carcass, carcass maceration, bone drying, frame construction drilling and articulation of bones and bone varnishing

(1) Skinning, Evisceration and De-fleshing of carcass: The bull was skinned, eviscerated and the flesh were detached from the bones manually using knives (Fig. 2) according to [8].

(2) Carcass maceration: The bones were heated to over 80ºC for 4 hours in solution of crystallized hydrous carbonate of sodium (crystal soda) and anionic surfactant

(detergent) in a metal drum. The remnants on the boiled bones were separated with knives. The boiled bones were left to stand in a mixture of surfactant and water for 12 hours after which the separation of the remaining flesh, ligaments and the bones were done using knives and sponge. The limb bones were then rinsed in clean water.

(3) Bone drying: The moist bones were dried under Sun for a period of four (4) days (Fig. 3).

(4) Frame construction: The skeletal frame was constructed using a wooden floor of 48 inch and 24 inch length and breadth respectively, metal pipes, screws and nuts as shown in Fig. 4.

Fig. 2. A photograph showing the axial skeleton harvested from the Red Bororo bull

2.1 Drilling and Articulation of the Appendicular Bones

A drilling machine (Black & decker® KR532 England) with 230 V was used to drill the bones (including the axial and appendicular skeletons) and two wires of 2 mm and 1.34 mm thicknesses were passed through the drilled holes to join and articulate the corresponding bones *in situ*. A general purpose white glue (TOP BOND®) of 250g was used to attach the carpals and tarsal bones together.

2.2 Drilling and Articulation the Axial Bones

A metal pipe of 60inch and 0.5inch in length and diameter respectively was inserted into the vertebral canal of the sacral, lumbar, thoracic and cervical vertebrae into the foramen magnum of the skull. The occipital condyles of the skull and cranial part of the atlas wing were drilled on both sides and 2mm wire passed through corresponding holes and knots were made to articulate both bones *in situ* and a loop of 1.35mm wire was made and tied round the incisive and maxilla bones to keep the jaws closed.

The caudo-lateral and cranio-lateral bodies of the atlas and axis were drilled on both sides respectively, and 2mm wire passed through corresponding holes and knots were made to articulate both bones *in situ* and a 2 mm wire was passed through the transverse foramen of C2, C3 and C4 and looped to articulate corresponding bones *in situ*, while the ventral tubercle of C4 and C5 were drilled and 2mm wire passed through to articulate them *in situ*. Holes were drilled on the left and right transverse processes of all the thoracic vertebrae and 2mm wire passed through and tied to the neck of all the corresponding ribs to articulate them *in situ*. The distal parts of all the ribs and corresponding costal cartilages were drilled and 1.35mm wire passed through to articulate them *in situ*. The cartilaginous joints between the costal cartilages and sternebrae were left intact and *in situ*. The coccygeal vertebrae were drilled medially and a 2mm wire passed through them and into the vertebral canal of the sacrum and fixed to its arch to articulate them *in situ* and the entire vertebral column were mounted on a pipe of 50inch length (Fig. 4).

2.3 Assembling and Mounting of the Hindlimb

The skeleton of the hindlimb and forelimb were mounted respectively to the mounted axial skeleton (Fig. 4). The iliac and sacral wings were drilled on both sides and 2mm wire passed through the holes to articulate the sacro-iliac joint *in situ*. The left and right iliac shaft were drilled into the lunar surfaces of their acetabulum and the head of the femur into the neck on both sides and 2mm wire passed through to articulate the hip joint *in situ*. The left and right medial epicondyles and condyles of both limbs were drilled and 2mm wires passed through into the drilled holes on the left and right lateral and medial condyles of tibia (articular surface of the tibia) to articulate knee joint *in situ*, Holes were drilled from the center of the patella into the femoral trochlea and a 2mm wire passed through to articulate them *in situ*.

The tarsal bones of the limbs were articulated *in situ* using a top bond, there medial and lateral malleolus together with the tallus were drilled and a 1.34 mm wire passed through to articulate them *in situ*. The base of the medial and lateral metatarsal (proximal part) of both limbs were drilled 1.34 mm wires passed through into the holes drilled on the 4th tarsal to articulate them *in situ*. A hole was drilled into the centers of the proximal, mid and distal phalanx of the two digits of both limbs and 2 mm wires passed through into the holes drilled into the head of metatarsal bones (at the distal articular surface) to articulate them *in situ*. Holes were drilled on the coffin bones (distal phalanx) of both limbs and 1.34 mm wires passed through to articulate them (Fig. 5).

2.4 Assembling of the Forelimb

The infraspinatus and supraspinatus fossae of both limbs were drilled about 2 inches away from the scapular spines and a 2 mm wire passed through them, and across the thoracic vertebrae and a loop was made to articulate them *in situ*. The ventral angle of the scapular of both limbs were drilled caudally together with the head of the humerus and a 2 mm wire passed through to articulate them, forming the shoulder joint *in situ*. The lateral and medial condyles of the humerus of both limbs were drilled and a 2 mm wire passed through into the drilled holes of the lateral and medial neck of the radius to form and articulate the elbow joints *in situ*. The distal part of the radius of both limbs were drilled medially and laterally and a 2 mm wire inserted and passed through the corresponding holes drilled on the lateral and medial parts of the base of the metacarpals with the glued carpal bones placed in between the radio-ulna and metacarpals of both limbs. The proximal, mid and distal phalanx were articulated in a similar manner as in the hindlimbs (Fig. 5).

2.5 Varnish

Thinner 500 ml and varnish 250 ml were mixed and sprayed on the skeleton before mounting on the frame to further preserve and give it a shiny appearance.

3. RESULTS

The bones extracted comprised those of the axial and appendicular skeleton. The bones appeared whitish and intact. The axial skeleton comprised the skull, cervical, thoracic, sacral and coccygeal vertebrae, ossa-coxarum, ribs, costal cartilages and sternebrae (Fig. 4) and the appendicular skeleton comprised of the for elimb (scapular, humerus, radio-ulna, carpals, metacarpals and the digits) and hindlimb (femur, tibio-fibula, tarsals, metatarsal and digits) (Fig. 5). The time utilized by each of the steps of the skeleton preparation technique is as shown on Table 1.

Fig. 3. A photograph showing the drying process of the rib bones

Table 1. The duration of red bororo bull skeleton preparation

S/N	Steps	Time (Days/Hours/Minutes)	Time (%)
1	Skinning, Evisceration and De-fleshing	6 hours	5%
2	Carcass maceration	16 hours	12.21%
3	Bone drying	4 days	73.28%
4	Frame Construction	30 minutes	0.1%
5	Drilling and articulation of bones	12 hours	9.16%
6	Bone varnishing	30 minutes	0.1%
	Total time	**5 days and 11 hours**	**100%**

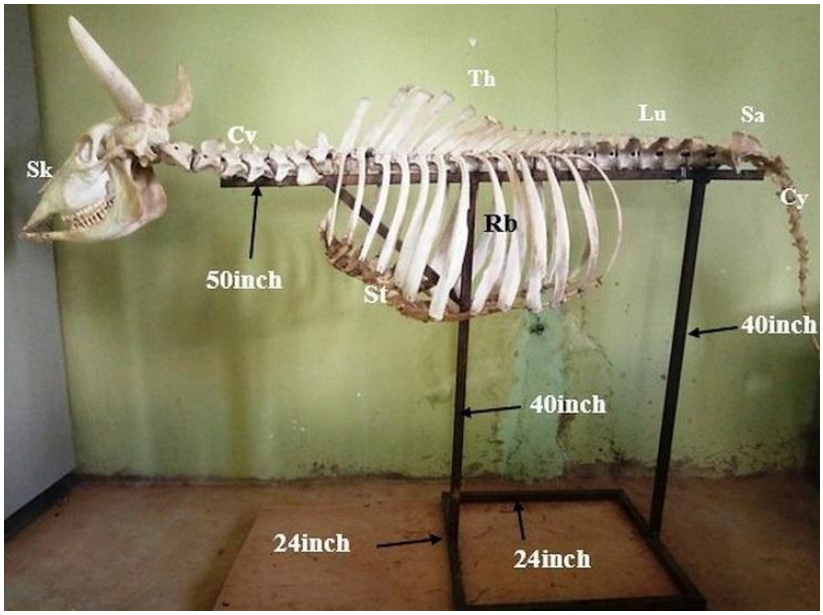

Fig. 4. A photograph of the axial skeleton of a Red Bororo bull, showing the skull (Sk), cervical vertebrae (Cr), thoracic vertebrae (Th), lumbar vertebrae (Lu), sacral vertebrae (Sa), coccygeal vertebrae (Cy), ribs (Rb) and sternum (St)

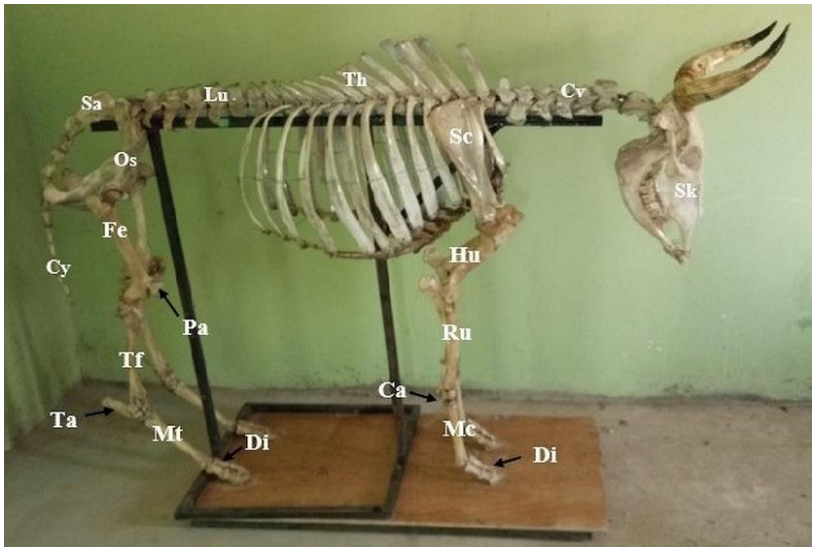

Fig. 5. A photograph of the skeletal system of a Red Bororo bull, showing the assembled and mounted axial skeleton, hindlimb femur (Fe), patella (Pa), tibio-fibula (Tf), tarsus (Ta), metatarsus (Mt) and digits (Di) and forelimb scapular (Sc), humerus (Hu), radio-ulna (Ru), carpus (Ca), metacarpus (Mc) and digital (Di), and the axial skeleton skull (Sk), cervical vertebrae (Cv), thoracic vertebrae (Th), lumbar vertebrae (Lu), sacral vertebrae (Sa), coccygeal vertebrae (Cy), ribs (Rb) and sternum (St)

4. DISCUSSION

The duration of skeleton preparation for Red Bororo bull in this study indicated that the entire procedure took 5 days-11 hours, which is shorter compared to the 18 days-2 hours both in pigeon and squirrel, 5 months-6 days-6 hours in deer, and 10 months-11 days-12 hours-30

minutes in crocodile [5], the difference in the methods of skeleton preparation could be responsible for the differences in the preparation duration.

The total number of bones extracted was 207; the bones of the appendicular and axial skeleton which helps in locomotion and protection of the brain, spinal cord and internal organs of the body respectively [9].

All the bones of the skeletal system in this research appeared white and intact as reported by Kyle [10] and Jesse [11], however, the method used in this present study is much faster and safer, compared to the report of Kyle [10] and Will [12], whose method involved burying of the dead animal for 60 days and the handling of decomposed carcass and dangerous chemicals such as chlorine bleach and fumes.

Furthermore, the maceration time in Pigeon, squirrel, deer and crocodile were 15 days 15 days 5 months and 10 months respectively [5] which were all longer compared to 16 hours reported in this study, as the boiling maceration method adopted in this research is faster compared to the burial maceration methods reported by Mahapatra et al. [5]. The method in this present study is also easier and cheaper compared to the report of Jesse [11], Will [12] and Kyle [10] whose methods requires a skeleton preparator's Standard Toolkit.

5. CONCLUSION

The method of preparing animal skeleton described in this present study lasted for 5 days and 11 hours, thus, the method has shown to be faster (less than a week), safer, easier and cheaper compared to previous researches. It is concluded that drying was the principal time-consuming component in the skeleton preparation. It consumed 73% (4 days) out of the total time used in preparing the skeleton (5 days and 11 hours).

CONSENT

It is not applicable.

ETHICAL APPROVAL

This study was approved by the research ethics committee of the College of Agriculture and Animal Science Bakura, Nigeria (CAAS/BKA/CREC/024).

ACKNOWLEDGEMENT

The technical contributions of all 2018 HNDII project students of A.H.T. Department CAAS Bakura, Zamfara State is acknowledged.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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Availble:www.skullsite.co.uk

Peer-review history:
The peer review history for this paper can be accessed here:
https://sdiarticle4.com/review-history/51580