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Occurrence of Os Rostrale in a Buffalo (*Bubalus bubalis*) Skull

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

The Os rostrale, also known as the prenasal bone, is commonly found in members of the Suidae family, particularly in pigs. It completes the snout's rigid skeleton, aids in digging and provides structural integrity during excavation activities. However, its occurrence in bovines, including buffalo breeds, is rare and understudied. This study aimed to explore the morphological and histological features of the Os rostrale in an adult Pandharpuri buffalo. Additionally, it aimed to investigate the rarity of the occurrence of Os rostrale in buffalo skulls. After careful gross morphological observations, the Os rostrale of an adult Pandharpuri buffalo was subjected to radiographic analysis and histological examination. The Os rostrale appeared sword-shaped with a mediolateral

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curvature and was radio-opaque. Histological analysis revealed active hyaline cartilage transitioning into compact bone with Haversian system, indicating its endochondral ossification. Despite belonging to an adult animal, the structure retained cartilage elements, suggesting a slow replacement process requiring further investigation. This study provided essential baseline information regarding the Os rostrale in buffaloes, contributing to a better understanding of its structural significance. Additionally, it sheds light on the unusual occurrence of the Os rostrale in buffalo skulls, which is uncommon within the Bovidae family.

Keywords: Bubalus bubalis; pandharpuri; os rostrale; skull; radiography; ossification; histology.

1. INTRODUCTION

Visceral bones or splanchnic bones are non-weight-bearing bones found in a diverse range of mammals and birds. The baculum, which was found in various mammals including carnivores, bats, rodents, insectivores, flying lemurs, and certain primates [1,2], Os clitoridis of felines [3], Os diaphragmaticum of camel [4], Os cordis of bovines [5], Os rostrale of pigs and scleral ossicles in birds [6] are some of the noteworthy examples of visceral bones. These bones are observed to occur to aid in certain functions *viz*., scleral ossicles maintain the shape of the eyeball in variable environments in birds; Os penis provides support to the passage of penile urethra; Os phrenic presumably maintains the aperture size of afferent caudal vena cava in camels and Os rostrale furnished structural integrity to snout in pigs as fossorial adaptation. The occurrence and anatomical characteristics of the Os rostrale, or the prenasal bone, were predominantly documented within the Suidae family [7]. Os rostrale in pigs completed the rigid skeleton of the snout by replacing part of the nasal septum thus enhancing digging and helping the snout to move forcefully during excavation activities [8,9,10]. The occurrence of this bone is elusive and rare as far as bovines are considered. The presence of the Os rostrale in ox skull had been documented rarely [11]. Despite such previous observations in cattle, the existence and importance of the Os rostrale in water buffalo breeds had not been sufficiently addressed. In this study, the skull of an adult Pandharpuri, adorned with caudally oriented dagger-shaped horns characteristic of the breed was utilized [12]. This breed was recognized as one among the indigenous Indian buffalo breeds. The present work aims to ascertain the presence of the Os rostrale in the Pandharpuri breed and its morphological and histological features.

2. MATERIALS AND METHODS

The head of a healthy adult Pandharpuri buffalo was collected from the Department of Veterinary Pathology, College of Veterinary and Animal Sciences, Pookode, during post-mortem examination. Subsequently, maggot-induced sarco-digestion was performed, followed by thorough cleaning and air drying before subjecting the specimen to further morphological, radiographic, and histological observations at the Department of Veterinary Anatomy, College of Veterinary and Animal Sciences, Pookode. After cleaning, a non-articulating, hardened structure was observed in the rostral region. Morphometric observations were conducted using a digital Vernier Calliper. Radiography was performed with 70 kV and 26 mAs using lateral and dorsoventral views. A small tissue sample was incised, observed under stereo-zoom microscopy (model: CZM6, make: Labomed), and subjected to Nitric acid treatment for decalcification, enabling further tissue processing. Routine histological processing and Hematoxylin and Eosin staining [13] unveiled microscopic details of cellular structures and matrix. Additionally, Alcian blue staining [14] was performed to distinguish any cartilage present.

3. RESULTS

The morphological examination of an elongated sword-shaped structure within the rostral region of the Pandharpuri buffalo skull revealed its attachment to the nasal septum. The structure had a pronounced curvature along the median plane, becoming more evident as one progressed in a rostro-caudal direction (Fig. 1). This curvature was observed to be more concave from dorsal side.

Morphometry revealed that, the Os rostrale had a rostro-caudal length of 41mm with a variable thickness along its length. The maximum thickness was found at the rostral tip, measuring 3.35mm, whereas, the least thickness was observed at its caudal-most end extreme, with a corresponding value of 1.55mm. The thickness was found to be lesser than the rostral but higher than the caudal extremities at about its mid-length, with a corresponding value of 1.83mm.

Stereo-zoom microscopic observation of the cross-section of the Os rostrale revealed a spongy appearance at its core with numerous caverns separated by rounded mounds (Fig. 2).
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Radiographic observations, indicated a radiopaque structure with distinct borders (Fig. 3).

Inset view of sword-shaped Os rostrale showing ventral adherence to nasal septum.

Histological observations revealed that the os rostrale was characterized by the presence of unevenly distributed cartilaginous and osseous portions (Fig. 4). In the region of cartilage, the matrix exhibited homogeneity without significant fibrous elements, suggesting that the cartilage observed was of the hyaline type studded with numerous isogenous cell groups, cell nests (Fig. 5). Active cell division was evident within each cell nest that housed chondrocytes. This hyaline cartilage was also observed to be a crucialcomponent in providing structural support and flexibility to the nasal septum, from which Os rostrale appeared to spring. The predominant tissue of the Os rostrale however, appeared to be osseous. Moreover, the presence of compact bone was well established, featuring a concentric arrangement of the lamellae. Between the lamellae, lacunae filled with osteocytes were present and the canaliculi perpendicular to the lacunae highlighted the nutrient exchange. The centrally located lumen observed corresponded to the Haversian canal, indicative of compact bone formation (Fig. 6).

The transition from cartilage to compact bone, a critical aspect of skeletal development, was distinctly observed (Fig. 7). The presence of isogenous cell groups and the progression from lacunae-filled with chondrocytes to lacunae occupied by osteocytes highlighted the dynamic nature of this structure development.

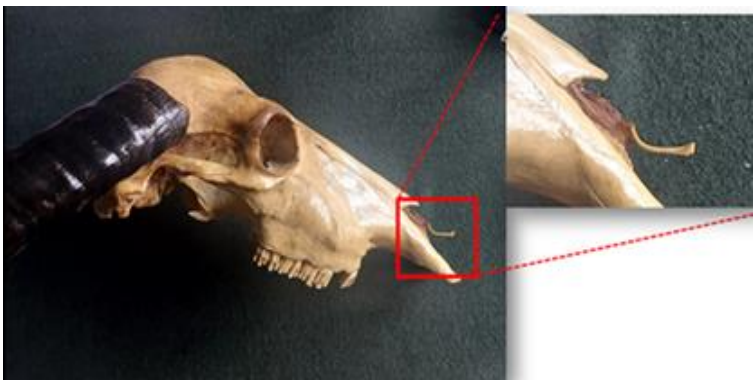

Fig. 1. Right lateral view of Pandharpuri buffalo skull showing the presence of Os rostrale

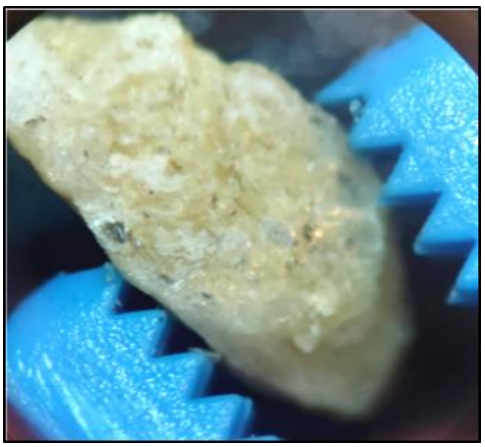

Fig. 2. Stereo-zoom microscopic view (X 12) of Os rostrale showing spongy appearance

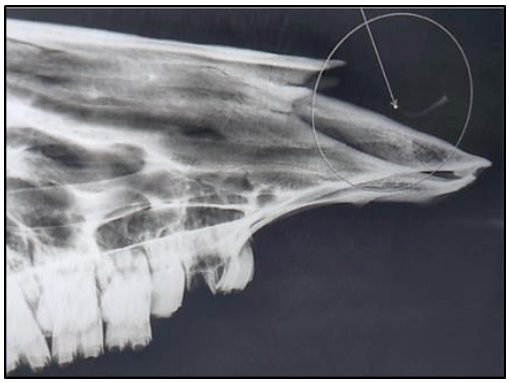

Fig. 3. Radiograph of Os rostrale (right lateral view) showing its radio-opaque nature

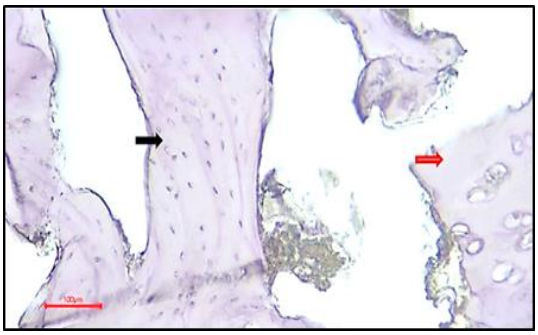

Fig. 4. Photomicrograph showing Os rostrale with uneven regions of cartilage (red arrow) and compact bone (black arrow), H & E (X 100)

Fig. 5. Photomicrograph showing isogenous cell group, H & E (X 400)

Fig. 6. Photomicrograph showing Haversion canal with concentric lamellae, H & E (X 400)

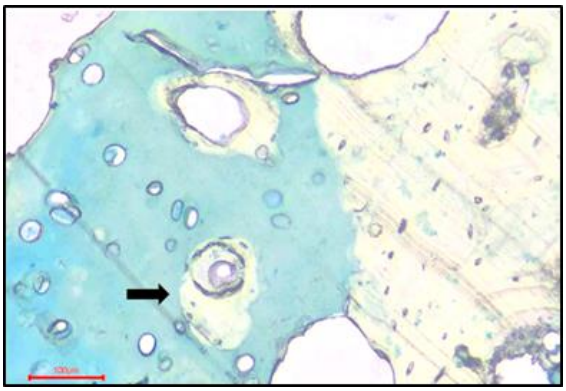

Fig. 7. Photomicrograph showing bone encroachment into cartilage (arrow) Alcian blue (X 100)

4. DISCUSSION

The presence of the Os rostrale in the Pandharpuri buffalo skull, although not entirely osseous, aligns with previous observations in other members of the Suidae family, such as pigs. In pigs, the Os rostrale has been documented as a normality, yet its attachment within the nasal cavity appears variable. The bone had been observed to either rest dorsally on the premaxilla [15], or, attach only to the cartilaginous part of the nasal septum [16]. Similarly, in the present study, it was observed that the ventral border of the Os rostrale was firmly adhered to the cartilaginous part of the nasal septum and had no direct contact with the premaxilla.

The histological examination revealed the endochondral process of ossification, with the transition from hyaline cartilage to compact bone being a critical aspect of skeletal development. The presence of numerous cell nests containing chondrocytes in the cartilaginous region suggested ongoing cartilage growth and remodelling. The identification of compact bone with features like lacunae, osteocytes, canaliculi, and a central Haversian canal further supported the bone growth at the cost of cartilage ensuring an ongoing process of endochondral ossification. The histology of Os rostrale has received poor attention throughout the available literature and the present work shows the microscopic peculiarities of Os rostrale *vis-à-vis* the mode of development on lines of endochondral ossification for the first time.

5. CONCLUSION

The Os rostrale found in Pandharpuri buffalo was not entirely osseous although the skull belonged to an adult animal. The presence of numerous cell nests containing chondrocytes and the presence of hyaline cartilage, a key components for structural support and flexibility in the nasal septum. Furthermore, the identification of compact bone with features like lacunae, osteocytes, canaliculi, and a central Haversian canal highlighted the ongoing process of ossification, the transition from cartilage to bone. However, the reasons for such slow replacement of cartilage need further investigation. This finding is significant as there are no existing records to compare this study with, indicating a novel aspect of visceral bone development in this species.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of manuscripts.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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