

Radiographic Anatomical Studies on the Developmental pattern of Ossification Centres in the Forelimb Bones of Local sheep of Kashmir valley

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ABSTRACT

The present investigation was undertaken to document the chronological appearance and fusion of ossification centres in the developing forelimb bones of prenatal sheep of Kashmir region using radiographic techniques. A total of eighteen sheep fetuses of varying gestational ages and one 15 days old postnatal dead sheep were utilized and categorized into three groups: Group I (0–50 days), Group II (51–100 days) and Group III (101 days to full term). Radiographic evaluation was performed to study the ossification pattern of the scapula, humerus, radius, ulna, carpal bones, metacarpals and phalanges. The skeletal system remained cartilaginous up to the 7th week of gestation. Primary ossification centres of most long bones appeared around 60 days of gestation. In general, distal epiphyses ossified earlier than proximal epiphyses. Sesamoid bones were the last to ossify. The developmental progression of forelimb bones in local sheep of the Kashmir Valley appeared relatively delayed compared to reports from other Indian sheep breeds. The findings provide baseline data useful for fetal age estimation, assessment of skeletal maturity and veterinary forensic investigations.

Key words: Forelimb, Ossification centres, Prenatal sheep, Radiography

INTRODUCTION

The skeletal system is a fundamental structural framework of the body and plays a vital role in species characterization and functional adaptation. The developmental morphology of bones is of considerable importance for diagnosing skeletal abnormalities in young and prenatal animals. Variations in skeletal development may occur due to breed, age, sex, nutrition and environmental factors (Outram and Conwy, 1998). The appendicular skeleton, particularly the limb bones, is commonly used for estimating fetal age and assessing developmental maturity. Radiographic examination is considered one of the most reliable techniques for studying ossification, as it allows non-destructive visualization of developing skeletal elements while preserving anatomical integrity. The appearance of ossification centres provides valuable information for determining prenatal age, fetal bone maturation and early detection of congenital anomalies (Oishi *et al.*, 1996).

Although several studies have described prenatal ossification in sheep and goats, detailed radiographic documentation of ossification chronology in local sheep of the Kashmir Valley is limited. Therefore, the present study was designed to establish a radiographic chronology of ossification in the forelimb bones of sheep fetuses, which may contribute to clinical anatomy, developmental biology and veterinary forensic science.

MATERIALS AND METHODS

Experimental Material

For the present study, sheep fetuses (n=18) and 15 days old sheep (n=01) of either sex without any external anomaly were used. Specimens were collected from the local slaughter houses present in and around Srinagar city and from sheep research station of SKUAST-K.

Estimation of Gestational Age

The approximate gestational age of each fetus was determined using Richardson's formula (Richardson, 1980): $X = 2.1(Y + 17)$ where X represents the age in days and Y denotes the crown–

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anus length in centimetres. Based on gestational age, fetuses were divided into three groups: 1. Group I: 0–50 days 2. Group II: 51–100 days 3. Group III: 101 days to full term.

Radiographic Procedure

Each fetus was subjected to radiographic examination to study the appearance and fusion of ossification centres in the forelimb bones. Exposure parameters were adjusted according to fetal size and tissue thickness. Early-stage fetuses were radiographed using 50 KVP (Kilo Volt Potential), 6.4 mA and an exposure time of 0.2–0.25 seconds, while later-stage fetuses were radiographed at 60–64 KVP, 12.4 mA and 0.3–0.45 seconds exposure time. Computed radiography was employed for image acquisition. The first appearance of a radio-opaque area at the appropriate anatomical location was considered as the onset of ossification.

RESULTS AND DISCUSSION

General Observations

Up to 54 days of gestation, the entire skeletal system of the sheep fetus was cartilaginous in appearance (Fig. 1). Primary ossification centres of most long bones began to appear around 60 days of gestation. A consistent finding throughout the study was that distal epiphyseal centres appeared earlier than proximal epiphyseal centres, a pattern also reported in earlier studies. Sesamoid bones were the last skeletal elements to ossify.

Scapula

The scapula developed from two primary ossification centres that is Corpus scapulae and Tuber scapulae. The centre for the corpus scapulae appeared at approximately 60 days of gestation, while the ossification centre for the tuber scapulae was observed near 149 days (Fig. 2). Ossification progressed from the distal to proximal direction, and bone density was relatively lower in the central region of the scapula. These findings are in agreement with earlier (Ahmed, 2008), although minor variations in timing were noted. Compared to our observations, the centre for ossification of the corpus scapulae and tuber scapulae in sheep has been reported at 45 days and 107 days of

gestational age, respectively (Supriya *et al.*, 2016). Similarly, Wenham (1977) reported that using radiographic method, the ossification centre for tuber scapulae appeared at 107 days in sheep, indicating slight regional or breed variation. Compared to those findings, ossification in the present study appeared relatively delayed.

Humerus

The humerus exhibited six ossification centres: one diaphyseal and five secondary centres (distal epiphysis, caput humeri, tuberculum majus, medial and lateral epicondyles). The diaphyseal centre appeared at about 60 days of gestation, unlike the earlier reports, where the ossification centre for the shaft was observed at 45 days of the gestational age in sheep foetuses (Supriya *et al.*, 2016). The distal epiphysis was observed at approximately 113 days (Fig. 3), followed by the appearance of centres for the caput humeri, tuberculum majus and the medial and lateral epicondyles between 97–128 days. The secondary centres remained separated from the diaphysis until late gestation, corroborating findings reported by Wenham (1981). However, the ossification, in general appeared slightly delayed in the present study when compared with the reports of Supriya *et al.*, (2016) in sheep.

Radius and Ulna

The diaphyseal ossification centres of the radius and ulna appeared between 60–65 days of gestation. The distal epiphysis of the radius was observed at around 113 days, while the proximal epiphysis appeared at approximately 119 days. Ossification of the olecranon process was evident near 149 days (Fig. 4). These results align with the observations of Chaudhary *et al.* (2018), though slight variations in gestational timing were evident. Three centres of ossification for radius and ulna have also been observed in different dog breeds (Charjan *et al.*, 2014). Compared to our findings, the appearance of ossification centres in the radius and ulna of sheep has been reported much earlier, with an overall appearance of all the three centres of ossification as early as 128 days of fetal gestational age.

Carpals, Metacarpals and Phalanges

Ossification centres for the carpal bones appeared at approximately 104 days of gestation. The accessory carpal bone was the last to ossify, with its centre appearing near 149 days (Fig. 5). Similar observations have been reported in previous radiographic studies of ovine fetuses. The diaphysis of the large metacarpal bone was visible at around 60 days, while the distal epiphyseal centre appeared at approximately 125 days (Fig. 6). Ossification centres for the phalanges were observed at around 65 days, dewclaws at around 67 days and proximal sesamoid bones appeared near the terminal stages of gestation (Fig. 7).

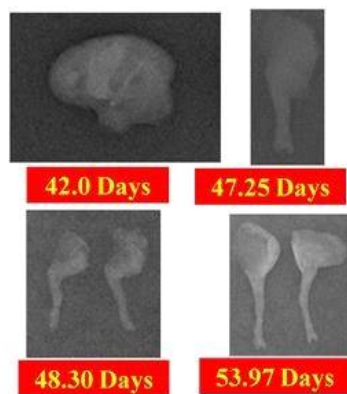


Fig. 1. Radiograph of a 42-day-old sheep fetus and forelimbs of sheep fetuses at 47, 48, and 54 days of gestation, demonstrating that the skeletal system was cartilaginous at these stages.

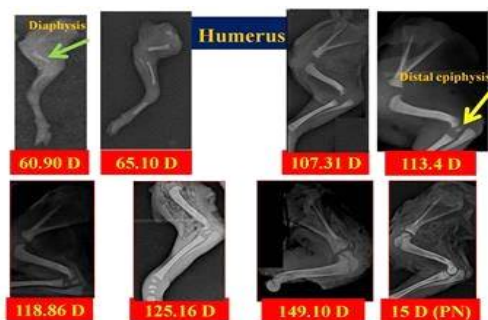


Fig. 3. Radiographs of the forelimbs of sheep fetuses at 60, 65, 107, 113, 119, 125, and 149 days of gestation, along with a 15-day-old postnatal sheep, demonstrating that the primary ossification centre of the humeral diaphysis appeared at 60 days, whereas the distal epiphyseal centre became evident at 113 days of gestation.

CONCLUSION

The present radiographic study establishes a detailed chronological sequence of ossification in the forelimb bones of prenatal sheep of the Kashmir region. The skeletal system remains entirely cartilaginous up to approximately 54 days of gestation, with primary ossification centres appearing around 60 days. The sesamoid bones were the last to show ossification. These findings provide valuable baseline data for fetal age estimation, assessment of skeletal maturity and identification of developmental anomalies in sheep. Our findings suggest that the development of the bones of fore limb in the local sheep of Kashmir region was slow in progress when compared with other sheep breeds of India.

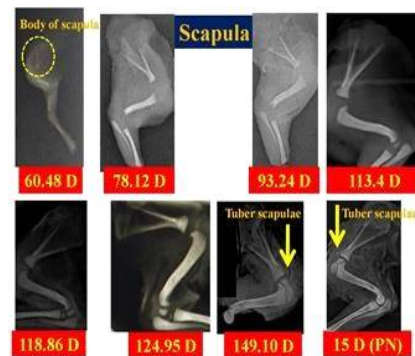


Fig. 2. Radiographs of the forelimbs of sheep fetuses at 60, 78, 93, 113, 119, 125, and 149 days of gestation, along with a 15-day-old postnatal sheep, demonstrating that the primary ossification centre of the scapula appeared at approximately 60 days, whereas the tuber scapulae became evident at 149 days of gestation.

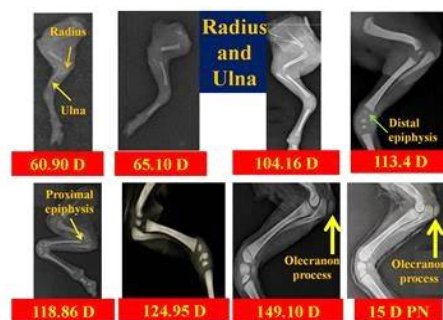


Fig. 4. Radiographs of the forelimbs of sheep fetuses at 61, 65, 104, 113, 119, 125, and 149 days of gestation, along with a 15-day-old postnatal sheep, demonstrating that the primary ossification centres of the radius and ulna appeared at 61 days. The distal and proximal epiphyseal centres of the radius became evident at 113 and 119 days, respectively, while the olecranon process ossified at 149 days of gestation.

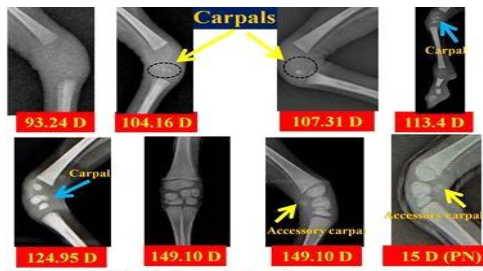


Fig. 5. Radiographs of the forelimbs of sheep fetuses at 93, 104, 107, 113, 125, and 149 days of gestation, along with a 15-day-old postnatal sheep, demonstrating that the primary ossification centres of the carpal bones appeared at 104 days of gestation, whereas the accessory carpal bone ossified at 149 days of gestation.

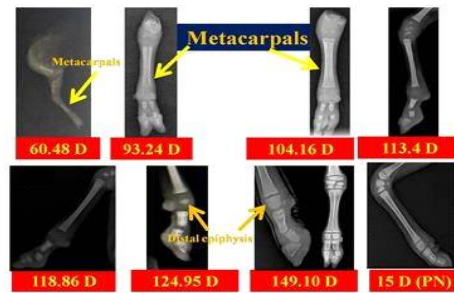


Fig. 6. Radiographs of the forelimbs of sheep fetuses at 60, 93, 104, 113, 119, 125, and 149 days of gestation, along with a 15-day-old postnatal sheep, demonstrating that the primary ossification centre of the large metacarpal bone appeared at 60 days of gestation, whereas the distal epiphyseal centre became evident at 125 days of gestation.



Fig. 7. Radiographs of the forelimbs of sheep fetuses at 65, 67, 78, 113, 118, 125, and 149 days of gestation, along with a 15-day-old postnatal sheep, demonstrating that the primary ossification centre of the first phalanx appeared at 65 days of gestation, those of the second phalanx and dewclaws at 67 days, and the proximal sesamoid bone of the digit at 149 days of gestation.

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