Epiphyseal plate closure of radio-ulna bone in red Sokoto goat ecotype

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Abstract
This study was conducted with 57 Red Sokoto goats, which were randomly obtained from three different small ruminant farms with birth record within Sokoto metropolis, Nigeria. They were classified into different age groups and subgroups, from 1-144 weeks. The radiographs of their forearms were taken and the proximal and distal epiphyseal plate lengths of both radius and ulna bones were measured. The radiographic images of the bones showed that the proximal and distal epiphyseal plates of the radius were opened at week 1 with mean lengths of 0.50±0.05mm and 1.10±0.01mm respectively but fuses at week 8 and 36 respectively. The proximal ulna epiphyseal plate was opened at week 1, with mean length of 1.67±0.02mm and fused at week 144. However, the distal ulna epiphyses appeared radiolucent at week 1 and 2 and became radiopaque at week 3 with an epiphyseal plate mean length of 3.67±0.26mm which reduced chronologically and fused at 96 week. It was therefore concluded that in Red Sokoto goat, epiphyseal plate lengths decreases with increase in age and fuses at different age even within the same bone, and the epiphyseal plates of radius bones fuse earlier than the ulna bones.

Keywords: Epiphyseal plate, Red Sokoto goat, Radiography, Radius, Ulna

Introduction
The domestic goat (Capra aegagrus hircus) is a subspecies of goat domesticated from the wild goat (Capra aegagrus) (Belanger & Bredesen, 2010). Aina & Oppong (2011) stated that Red Sokoto goat is an indigenous breed of goat in Nigeria and is common to the Northern region of the country (Fabusoro, 2006).

According to Umar et al. (2013), five distinct colour coats were identified as Red Sokoto Goat (RSG) ecotypes; dark red, brown, light brown, black and variegated. Umar et al. (2013) further stated that these colours currently represent the population of RSG with only about 19% of the total population found within Sokoto and its environs and are classified as dark Red Sokoto goat.

The forearm consists of radius and ulna bones (Okpe & Adamu, 2002). The bones are rod-like, with a shaft and two extremities. They articulate with the humerus proximally and carpus distally (Larsen et al., 1999).

In recent years, there has been an increasing interest for a more accurate means of determining the age of animals, one of which is the use of epiphyseal plate closure (Choi et al., 2006). The epiphyseal plate is the area of longitudinal growth in a long bone (Boundless, 2016). It is a thin layer of hyaline
cartilage in the metaphysis at each end of an immature long bone where ossification occurs and is interpreted by a radiographer as a radiolucent line bordered by slightly higher radiopacities (Dyce et al., 2010).

Youssef et al. (2016a) stated that age determination with the aid of epiphyseal plate closure is of great importance in medico-legal cases, forensic pathology and anthropology. According to Gizada (1995), knowing the age of a goat enables one to decide when to cull, to decide when to mate,-know contemporaries for selection among them, and to have a good estimate of reproductive performance.

Understanding the anatomy of epiphyseal plate growth enables practicing veterinarians to provide a prognosis and assess indications for surgery (Summerlee, 2002). Furthermore, injured animals could be closely observed during the period of rapid growth as shown by epiphyseal growth plate pattern (Boskey, 2002).

Many researches were conducted on the epiphyseal plate of different animal species; (Choi et al., 2006, Chang et al., 2007, Genccelep et al., 2012, Alpdogan & Genccelep, 2012, Youssef et al., 2016a & Youssef et al., 2016b), however, there is dearth of information on age estimation using radiographic anatomy of epiphyseal plate closure of Red Sokoto goat.

Due to variations and complexities of epiphyseal plate closure pattern in different breeds and species of animals as stated by Evans & De Lahunta (2012) Choi et al. (2006), Chang et al. (2007), Alpdogan & Genccelep, 2012 and Genccelep et al. (2012), there is need to establish a pattern peculiar to Red Sokoto goat.

The aim of this study was to evaluate the epiphyseal plate closure pattern of radius and ulna bone across age groups in Red Sokoto goat using radiographic approach.

This study will provide information on the epiphyseal plate closure of radius and ulna bones and secondary ossification centers of the Red Sokoto goat whose birth records were known. Similarly, it will provide basic information for the estimation of age by radiography. Furthermore, it will reduce the problem of inadequate age record keeping among farmers.

**Materials and Methods**

This study was conducted with 57 Red Sokoto goats. The goats were randomly obtained from three different small ruminant farms with birth records within Sokoto metropolis and environs in Nigeria. Their ages ranged from week 1 to 144, and based on a single factor design (age), they were classified into different age groups and subgroups, a modified classification of Choi et al. (2006) as shown on table 1.

The goats were apparently healthy, as physical and clinical investigations were performed to exclude those with skeletal deformities and injuries within the limbs. They were then transported to Radiology unit, Department of Veterinary Surgery and Radiology, Usmanu Danfodiyo University, Sokoto, Nigeria.

The forearm region was groomed using a brush to remove any dirt that may be radiopaque. With the aid of physical restrain, each of the goats was placed on a right lateral recumbency, with the right limb down and the upper left limb retracted backwards, the right forearm was slightly extended and allowed to rest on the radiographic film cassette and the digital dry medical X-ray films were used. The x-ray beam was directed vertically from the focal spot to the center of the forearm as described by Douglas et al. (1987).

Two radiographic shots per goat and an exposure of 60kV and 10mAs for the kid goats and 65kv and 12mAs for adult goats and film-to-focus distance of 97cm were used (total of 114 shots) as described by Sirois and Anthony (2009) and the dates of exposure were documented (Table 1).

The proximal and distal epiphyseal plate lengths of the radius and ulna bones were measured using an illuminator, A4 paper, and electronic digital caliper (Raider® RDDC 706 model to the nearest 0.01mm. The epiphyseal plate length (the radiolucent area at each extremity of radius and ulna bones) was measured from the proximal end to the distal end, was marked on the A4 paper and the measurement was obtained from the point marked on the A4 paper (Plate II). The landmarks of the epiphyseal plate length as shown on plate I is further described below:

- **a.** Proximal Radial Epiphyseal Plate Length (PREPL): measured from the upper end to the lower end of the proximal epiphyseal plate of the proximal extremity of the radius bone.
- **b.** Distal Radial Epiphyseal Plate Length (DREPL): measured from the upper end to the lower end of the distal epiphyseal plate of the distal extremity of the radius bone.
- **c.** Proximal Ulna Epiphyseal Plate Length (PUEPL): measured from the upper end to the lower end of the proximal epiphyseal plate of the ulna at the proximal extremity of the ulna bone.
Table 1: Number and age of goats examined based on groups and subgroups

| Group (Age in weeks) | Subgroup (weeks) | Number examined (n=57) | Date of post-exposure |
|----------------------|------------------|------------------------|-----------------------|
| A (1-4)              | 1                | 3                      | 0                     |
|                      | 2                | 3                      |                       |
|                      | 3                | 3                      |                       |
|                      | 4                | 3                      |                       |
| B (8-16)             | 8                | 3                      | 7                     |
|                      | 12               | 3                      |                       |
|                      | 16               | 3                      |                       |
| C (20-28)            | 20               | 3                      | 14                    |
|                      | 24               | 3                      |                       |
|                      | 28               | 3                      |                       |
| D (32-44)            | 32               | 3                      | 21                    |
|                      | 36               | 3                      |                       |
|                      | 40               | 3                      |                       |
|                      | 44               | 3                      |                       |
| E (48-144)           | 48               | 3                      | 28                    |
|                      | 72               | 3                      |                       |
|                      | 96               | 3                      |                       |
|                      | 120              | 3                      |                       |
|                      | 144              | 3                      |                       |

Table 2: Mean ± SD length of radio-ulna epiphyseal plates radiographs across different age groups

| Age group (weeks) (n=57) | Subgroups (weeks) | Length of Epiphyseal Plate (mm) | Radial bone | Ulna bone |
|--------------------------|-------------------|---------------------------------|-------------|-----------|
|                          |                   | Proximal Distal                 | Proximal    | Distal    |
| A (1-4)                  | 1                 | 0.50±0.10<sup>a</sup> 1.10±0.02<sup>a</sup> 1.67±0.05<sup>a</sup> | N.O.C       |
|                          | 2                 | 0.44±0.03 0.84±0.09<sup>a</sup> 1.38±0.03<sup>a</sup> | N.O.C       |
|                          | 3                 | 0.36±0.02 0.68±0.05<sup>a</sup> 1.24±0.05<sup>a</sup> 3.67±0.45<sup>a</sup> |
|                          | 4                 | 0.30±0.02<sup>a</sup> 0.41±0.02<sup>a</sup> 0.92±0.03<sup>a</sup> 2.66±0.07<sup>a</sup> |
| B (8-16)                 | 8                 | 0.40±0.02<sup>b</sup> 0.90±0.03<sup>b</sup> 2.63±0.03<sup>b</sup> |
|                          | 12                | Fused 0.29±0.02<sup>b</sup> 0.70±0.03<sup>b</sup> 2.35±0.03<sup>b</sup> |
|                          | 16                | Fused 0.20±0.02<sup>b</sup> 0.57±0.03<sup>b</sup> 2.25±0.02<sup>b</sup> |
| C (20-28)                | 20                | Fused 0.19±0.02<sup>c</sup> 0.50±0.03 2.14±0.04<sup>a</sup> |
|                          | 24                | Fused 0.18±0.03<sup>c</sup> 0.51±0.02 2.03±0.03<sup>a</sup> |
|                          | 28                | Fused 0.17±0.02 0.50±0.02 2.01±0.02 |
| D (32-44)                | 32                | Fused 0.14±0.02 0.47±0.02 1.71±0.02<sup>ab</sup> |
|                          | 36                | Fused Fused 0.45±0.03 1.60±0.04<sup>ae</sup> |
|                          | 40                | Fused Fused 0.42±0.02 1.52±0.03<sup>ef</sup> |
|                          | 44                | Fused Fused 0.42±0.02 1.42±0.02<sup>ef</sup> |
| E (48-144)               | 48                | Fused Fused 0.41±0.02<sup>ab</sup> 1.41±0.02 |
|                          | 72                | Fused Fused 0.39±0.02<sup>b</sup> 0.97±0.02 |
|                          | 96                | Fused Fused 0.35±0.02<sup>d</sup> Fused |
|                          | 120               | Fused Fused 0.14±0.02<sup>abcd</sup> Fused |
|                          | 144               | Fused Fused Fused Fused |

Means with the same superscript across the subgroups in the columns differ significantly at (P<0.05) KEY: N.O.C= No Ossification Centre (n=57)
d. Distal Ulna Epiphyseal Plate Length (DUEPL): measured from the upper end to the lower end of the distal epiphyseal plate of the distal extremity of the ulna bone. The data obtained were sorted out and presented as mean ± Standard Deviation (SD). The differences were compared for statistical significance by analysis of variance (ANOVA). Differences were considered significant at P<0.05. The statistical analysis was performed using GraphPad InStat statistical software version 3.0.

Results
The radiographic images of the forearm of the Red Sokoto goats showed two separate long bones (radius and ulna bones), with each having a shaft and two extremities (proximal and distal) situated between the humerus dorsally and carpus ventrally, where they form the elbow and knee joints respectively. The ulna was caudal to the radius in the upper part of the forearm but lateral in the lower part with an interosseous space in between them. The proximal and distal extremities of the ulna bones were unattached to the radius bones at week 1 (Plate III) which gradually became attached as the age increased.

Across all the age groups, the ulna bone appeared wider proximally than distally (in contrast to the radius), strongly concaved cranio-caudally, the length and width of the bone increased progressively whereas the epiphyseal plate length decreased. The proximal extremity extends beyond the elbow joint to form the olecranon. The radius was slightly concaved cranio-caudally and wider distally than proximally.

The radiographic images of the radius showed radiolucent appearance in the proximal and distal epiphyseal plate at which no ossification occurred. This occurred between the age groups of 1 to 4 weeks, the proximal and distal radial epiphyseal plates fused at week 8 and 36 respectively (Plates V and VI and Table 2).

From week 1 to 2 the distal ulna epiphyses (distal secondary ossification center) were absent and appeared at week 3 (Plate III), though the proximal ulna epiphyses were seen at week 1 of life (Plate II) (Table 2). The proximal and distal ulna epiphyseal plates fused at week 144 and 96 respectively (Plates VIII, VII and Table 2).

The radiographic measurement of radius and ulna epiphyseal plates had the lowest mean lengths of 0.30±0.02mm for proximal radial epiphyseal plate, 0.14±0.02mm for distal radial epiphyseal plate, 0.14±0.02mm for proximal ulna epiphyseal plate and 0.97±0.02mm for distal ulna epiphyseal plates at week 4, 32, 120, and 72 respectively.

No significant difference (P<0.05) was seen for proximal ulna epiphyseal plate lengths (PUEPL) within the subgroups of 20 – 28 of group C and 32 – 44 of group D. In group A, the proximal radius epiphyseal plate lengths showed no significant difference (P<0.05) except for the subgroups of 1 and 4. Furthermore, distal ulna epiphyseal plates...
Plate III: A right medial view radiograph of radio-ulna bone at week 1, of Red Sokoto goat showing the proximal and distal epiphyseal plates (O), and proximal and distal end of the radius bone (RPM). The distal ulna epiphyseal plate was radiolucent (N.O.C), and the unattached rudimentary coronoid process (RC) and interosseous space (white arrow).

Plate IV: A left medial view radiograph of radio-ulna bone of a Red Sokoto goat at week 3, showing proximal and distal epiphyseal plates (O), slight appearance of distal ulna epiphysis (OC), proximal and distal end of the radius bone (RPM) and wide interosseous space (white arrow).

Plate V: A radiograph of radio-ulna bone of Red Sokoto goat at week 8 showing proximal radial bone plate is closed (C), proximal ulna and radio-ulna distal epiphyseal plates are radiolucent (O), proximal and distal end of the radius bone (RPM), Interosseous space (white arrow).

Plate VI: A right medial view radiograph of radio-ulna bone of Red Sokoto goat at week 36, showing the closed distal radial epiphyseal plate (C), proximal and distal ulna epiphyseal plates were radiolucent (O), proximal and distal end of the radius bone (RPM) and interosseous space (white arrow).
Plate VII: A left medial view radiograph of radio-ulna bone of Red Sokoto goat at week 96, closed distal ulna epiphyseal plate (C), radiolucent proximal ulna epiphyseal plate (O), and distal end of the radius bone (RPM) and interosseous space (white arrow)

Plate VIII: A left medial view radiograph of radio-ulna bone of Red Sokoto goat at week 144, showing closed epiphyseal plates (C), distal end of the radius bone (RLM) and interosseous space (white arrow)

(DUEPL) showed no significant difference (P<0.05) for groups A and B.

Discussion
The chronological decrease in the epiphyseal plate lengths of both radius and ulna bones observed in this study agrees with the findings of Choi et al. (2006) and Youssef et al. (2016a) who worked on the relationship between growth plate closure and age in sheep and goat. In this study, the radius and ulna bones appeared as two individual long bones, with a shaft and two extremities each (proximal and distal) situated between the humerus dorsally and carpus ventrally, where they form the elbow and knee joints respectively, this agrees with the findings of Colville & Bassert, (2002), where they stated that long bones are characterized by an elongated shaft and enlarged extremities (proximal and distal) that bear articular surfaces.

In the present study, the secondary ossification centers of proximal radius and ulna and distal radius were all present across all the age groups. While the distal secondary ossification center of ulna appeared at week 3, this observation corresponds with the findings of Choi et al. (2006).

Genccelep et al. (2002) reported that the proximal epiphyseal plate of radius occurs at week 36 in Morkaraman lambs in Turkey and the process last for about 3-5 months. In our study, the closure occurs at week 8 and the process lasted for 7 weeks.

The proximal radial epiphyseal plates also fuses earlier compared to the findings of Choi et al. (2006) in Korean native breeds of goats, Genccelep et al. (2012) in mohair goat kids and Asimus et al. (1995) Genccelep et al. (2012) and Peltonen (1989) reported that closure of distal plate of radius occurs at week 88 and 24 in kid goat and lambs respectively. In our present study, the closure occurred at week 36

The proximal ulna epiphyseal plates fuses later compared to the findings of Genccelep et al. (2012) in coloured Mohair Goat kids. But the proximal ulna epiphyseal plates fuses at same age (week 144) compared to the findings of Choi et al. (2006). Saber et al. (1989) and Genccelep et al. (2012) documented that union of distal epiphyseal plate of ulna was observed in the week 52 and 48-56 in lambs and kid goat respectively. However, in this present study the closure was observed at week 96. The chronological order of closure of radius and ulna epiphyseal plates in this study was proximal radius, distal radius, distal ulna and proximal ulna respectively. This is contrary to the order of closure reported by Genccelep et al. (2012) in mohair goat kids. Closure time of epiphyseal plate has been reported to vary based on several factors such as breed, sex, species and bones of the animal (Genccelep et al., 2002; Smith et al., 1991).

The proximal and distal radial epiphyseal plates fuses at week 8 and 36 respectively, while the proximal
and distal ulna epiphyseal plates fuses at week 144 and 96 respectively. This agrees with the findings of Chang et al. (2007). According to Dyce et al. (2010), the fusion of the epiphyseal plate of long bones is due to the proliferation, maturation and hypertrophy, calcification and ossification activities occurring in the three of the four different zones (reserve zone, proliferative zone, maturation and hypertrophy, zone of calcified matrix and zone of ossification) of cells within the epiphyseal plate.

In conclusion epiphyseal plates length decreases with age, due to fusion even within the same bone. The epiphyseal plates of radius bones fuses earlier than the ulna bones. Distal ulna epiphyses do not contribute to the growth of the bones at week 1 and 2 of life since it is absent at this age. The radio-ulna bone stops growing at week 144, since proximal and distal radius and ulna epiphyseal plates fuses at 8, 144, 36 and 96 weeks respectively.

The result of the present study showed that radiographic imaging can serve as an effective tool for determination of age in the Red Sokoto goat, through evaluation of the radius and ulna bones epiphyseal plates.

The findings of this work will contribute to the studies that will be conducted on bone developmental standards of Red Sokoto goat. It is recommended that studies on evaluation of epiphyseal plate closure of other long bones in goats and other species and breeds of animals should be conducted to determine their closure time. Finally, advanced machines such as micro X-ray CT scanner should be used in evaluating the radiographs.

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