THE FOETAL GROWTH OF THE SHEEP

By H. A. HARRIS
Anatomy School, Cambridge

It is known that the state of development of the newborn in different species varies over wide limits. On the one hand the huge Polar bear gives birth to one or two cubs weighing from 1 to 1½ lb. each. The bat, on the other hand, gives birth to one baby, the weight of which is one-third that of the mother. Among the more familiar animals we may compare various forms such as the rat or mouse with the guinea-pig, the rabbit with the hare, the cat or dog with the horse and sheep. The newborn rat weighs but 5 g.; it is blind, helpless, hairless, and entirely dependent on the mother's milk for 4 weeks. Its skeletal development is comparable with that of a human embryo of the 4th month. On the contrary, the guinea-pig at birth is a "going concern", able to see, to walk and to maintain its warmth with the aid of a well-developed coat; it sucks and eats.

In comparison with the human babe at birth the newborn mammals may range in development from extreme pre-maturity, as in the mouse and rat, to extreme post-maturity, as in the lamb. These differences are not due to the number of young in the litter, for post-maturity is a feature in both the twelve piglets of the sow and the single colt of the mare. Nor is the period of gestation a determining factor, for the sheep, goat and pig show a combination of a short gestation period with post-maturity of the newborn. Man, in common with the great apes, shows a long gestation period with relative helplessness of the newborn.

It may be of interest to consider the rate of post-natal growth in the various forms. This rate is not determined by the state of development at birth, for the human babe doubles its weight in 6 months, the calf in 47 days, while the post-mature pig and sheep double their weight in 14 days and the pre-mature rabbit, cat and dog in 1 week. The rate of post-natal growth is not determined by diet, although attempts have been made to correlate growth with the dietetic value of the milk of the species. Cow's milk is three to four times as rich as human milk in protein, calcium and phosphate. The newborn calf doubles its weight in one-quarter of the time taken by the newborn babe, but this arithmetical ratio does not apply too closely to the whole mammalian species. It was Bunge(1) who first called attention to the close similarity in percentage composition between the mineral ash of the newborn and that of the mother's milk—iron alone proving the exception.

It is difficult to ascribe variation in pre-natal growth to differences in the placenta both as regards placental type and transmissibility. In some un-

gulates and insectivores there is no erosion of the maternal uterus by the chorion of the embryo, whereas in carnivores the erosion has extended almost as far as in the primates where the chorionic villi have eroded the decidua, connective tissue and endothelium of the mother to such an extent that the foetal chorionic villi are bathed in maternal blood. The type of placenta cannot be regarded as the determining factor in the pattern of growth of the young, for the human haemochorial placenta is closely imitated by the rodents, both the pre-mature rat and the post-mature guinea-pig. The placenta of the rodents and of the primates, including man, displays senile degeneration, proliferative endarteritis, infarction and calcification of the tissues in the marginal areas of the vascular territories.

Neither comparative anatomy nor physiology can afford a satisfactory explanation of the stage of development at which the young is born, the rate of post-natal growth or the pattern of skeletal development and dentition. The recent advances in the study of nutrition and of the hormones of the pituitary, Graafian follicle and corpus luteum indicate the need for a return to the study of morphology with a view to ascertaining to what extent, if any, the behaviour of the newborn is related to the degree of anatomical development.

THE PRE-NATAL DEVELOPMENT OF THE SHEEP

Through the courtesy of Sir Joseph Barcroft and his co-workers in the School of Physiology, Cambridge, it has been possible to examine a series of sheep embryos and foetuses from the earliest stages of gestation to term at about 154 days. The methods employed have been radiographic, dissectional and staining in bulk with madder (alizarin), whereby the bony skeleton is clearly demarcated from the cartilaginous. The earlier embryos, under 40 days, have been stained for cartilage by Lundvall's toluidine blue method.

The earliest distinct centre of ossification to appear in the sheep is in the mandible. This centre appears at the 39th day of gestation. In man the mandible commences to ossify at the 42nd day, being preceded by the clavicle at the 39th day. In the adult sheep, in common with ruminants generally, the clavicle is absent, but in the embryo a minute bony clavicle, 3 mm. long, is present at the 42nd day of embryonic life, and is absorbed before the 45th day. The centre for the mandible in the sheep is rapidly followed by that for the pre-maxilla, maxilla, and the tectum posticum which is a bridge of cartilage in the position of the future supra-occipital bone. At 41 days eleven pairs of ribs are ossified, the rapidity of their appearance indicating the large amount of membrane bone therein as previously suggested by Harris(2). At 42 days centres of ossification are present in the humerus, radius, ulna and metacarpals (cannon bone) of the forelimb; in the femur, tibia and metatarsals (cannon bone) of the hindlimb. At 45 days the basiocciput and exocciput commence to ossify. The lateral masses of the vertebrae display centres in descending order of magnitude from the first cervical to the first sacral, including seven

cervical, thirteen dorsal and seven lumbar vertebrae. Centres are present in the centra of the vertebrae from the third cervical to the second sacral, commencing about the twelfth and thirteenth dorsal and extending cranially and caudally. Well-developed centres are present for the scapula, ilium and ischium. The basi-sphenoid, pre-sphenoid, frontal and parietal bones are partly ossified.

Thus far the pattern and sequence of ossification in the sheep embryo does not differ essentially from the human except in the suppression of the clavicle and the development of only two bones in the metacarpus and metatarsus. At 61 days it would appear that the growth processes in the sheep are accelerated. In the vertebral column centres of ossification are present for the centra from the first cervical vertebra to the fifth sacral and extend to the fourteenth caudal. Centres for the lateral masses extend from the first cervical to the fifth sacral. In the breast plate, which consists of seven segments, ossification has appeared in the fourth, fifth and sixth segments. In the forelimb centres are present for the first, second and third phalanges of both digits; in the hindlimb centres for the os calcis and the three phalanges of both digits have appeared.

At 68 days both the tympanic ring surrounding the drum of the ear and the squama of the temporal bone are clearly defined in membrane bone. The dental crypts are visible as excavations in the bony alveolus of the upper and lower jaws. Five segments of the sternum are ossified. At 73 days the superior semicircular canal is heavily ossified and in the sacrum costal elements have appeared in bone near the auricular surface of the ala in the first and second segments. At 80 days all three semicircular canals are ossified and virtually adult in size. All the milk teeth are heavily calcified. A small centre is present in the pubis and in the talus. In short, between the 8th and 11th weeks the sheep embryo has rushed through its skeletal development to such an extent that the skeleton is comparable with that of the human foetus of 22 weeks.

At 92 days a most precocious event occurs in the spine of the upper dorsal vertebrae. Separate bony centres appear in the elongated spines from the second to the eighth dorsal. These are the first secondary centres to appear in the vertebral column. In man they do not appear until puberty. It should be noted that the vertebrae presenting precocious ossification of the spinous processes are those giving attachment to the muscles of the scapula and upper limb. In the sternum six segments present an advanced stage of ossification. In the carpus two small centres are present; in the tarsus two centres have appeared in addition to the os calcis and talus. At this stage, in addition to the fused third and fourth metacarpals (the cannon bone), ossification occurs in the transient fifth metacarpal. This bone is 20 mm. long by 0·5 mm. in diameter. There is no trace of a supernumerary metatarsal.

At 95 days five bones in the carpus and four in the tarsus are ossified. As many as nineteen caudal vertebrae are represented by ossification in their centra. At 99 days the centre of ossification appears in the distal epiphyses of

the femur—the centre which is used so extensively as an index of maturity in the human newborn. The supernumerary metacarpal has now disappeared in much the same way as the clavicle of early embryonic life.

At 104 days the anterior arch of the atlas is heavily ossified (Pl. I). The distal epiphyses appear in the two fused metacarpals (cannon bone). These are the first secondary centres to appear in the manus or pes. At 110 days all seven segments of the sternum are well ossified. In the forelimb the distal epiphyses of the humerus and radius show considerable ossification and the centre for the proximal end of the latter is about to appear. In the hindlimb the proximal epiphysis of the tibia is present; two centres have appeared for the distal extremity of the metatarsals (cannon bone) and three bones are present in the tarsus in addition to the os calcis and talus. At 111 days a small centre appears for the head of the humerus and a minute centre is present for the small coracoid process. The bar of cartilage interposed between the rami of the ischium and pubis is almost obliterated and indicates approaching union. At 120 days a centre of ossification appears for the proximal end of the ulna and ossification commences in the patella.

At 124 days a marked advance occurs in the vertebral column, for plate-like bony epiphyses appear on the bodies of the vertebrae. These extend from the third cervical to the fourth lumbar vertebra and have their maximum development at about the third or fourth dorsal. The epiphyses for the spinous processes which appeared at 92 days have commenced to unite with the neural arches and union is complete from the second to the fifth dorsal. Thus, epiphysial phenomena associated with puberty in the development of the human vertebrae are present in the foetal sheep of 124 days. A large centre of ossification is present for the head of the humerus. In the hindlimb the distal epiphysis of the tibia is present. In both limbs the two digits have epiphyses at the proximal extremities of all three phalanges, those in the forelimb being the more advanced. These phalangeal epiphyses appear in man early in the third year.

At 126 days the conjoined ischio-pubic ramus is completely ossified. Two days later a second centre has appeared at the distal end of the humerus (internal condyle) and a secondary epiphysis is ossifying at the posterior extremity of the os calcis. All these occur in man between the 7th and 11th years.

At 131 days the centre appears for the distal epiphysis of the ulna, for the head of the femur and for the tibial tubercle (Schlatter's). Bony sesamoids are present under the distal extremities of the metacarpals and metatarsals. At 134–135 days a third centre appears at the lower extremity of the humerus for the external condyle. At 137 days the great trochanter of the femur ossifies and at 138 days the distal epiphysis of the ulna is distinct. The first costal cartilage commences to calcify so that the superior aperture of the thorax is relatively fixed. Plate-like epiphyses are present on all five sacral vertebrae. A centre appears for the lesser trochanter of the femur and in one

specimen ossification is present in the fabella (sesamoid of the lateral head of the gastrocnemius), notwithstanding the numerous statements that no sesamoids are found in the neighbourhood of the knee joint in sheep (Pearson & Davin)(4).

ORDER OF APPEARANCE OF OSSIFICATION CENTRES IN THE SHEEP (*OVIS OVIS*)

Copulation age in days	Serial no.	Crown-rump length	Ossification centre visible on radiograph
36	112	27 mm.	Mandible condensation present
38	60	31 ,,	Mandible, maxilla
39	104	39 ,,	Clavicle. Mandible, maxilla. Tectum posticum of supra-occipital
40	66	40 ,,	Ribs 1–8. Humerus, radius, ulna
41	103	40 ,,	Supra-occipital. Ribs 1–10
43	69	46 ,,	Frontal, zygomatic. Ribs 1–13. Femur
45	122	50 ,,	Premaxilla, parietal. Ilium
46	96	65 ,,	Squamosal and parietal. Lateral mass of cervical vertebrae 1–7. Centrum dorsal vertebrae 12 and 13. Third metacarpal. Tibia and third metatarsal
47	95	65 ,,	Exoccipital, basi-occipital, basi-sphenoid. Lateral mass dorsal vertebrae 1–13. Centrum dorsal 1—lumbar 4
48	100	80 ,,	Presphenoid, periotic. Lateral mass cervical 1—sacral 2. Centrum cervical 2—sacral 3. Scapula; metacarpals 3 and 4. Ischium, metatarsals 3 and 4
61	102	140 ,,	Manus phalanges 1, 2, 3. Pes phalanges 1, 2, 3. Os calcis
64	107	130 ,,	Tympanic ring (trace of). Sternum four segments
66	106	140 ,,	Nasal bone. Dens of axis (centrum of cervical 1)
68	149	130 ,,	Sternum six segments
73	6	180 ,,	Semicircular canals partly ossified. Fifth metacarpal 1 cm. long. Trace of costal element of sacral 1
80	9	200 ,,	Semicircular canals ossified. Os magnum, hamatum. Astragalus
92	14	210 ,,	Anterior arch of atlas. Spines of dorsal vertebrae 2–9. Pubis, cuboid
95	7	250 ,,	Auditory ossicles. Five carpal bones; fifth metacarpal 2 cm. long. Four tarsal bones
99	8	300 ,,	Costal element of sacral 1 and 2 distinct. Secondary centre at lower end of femur
104	10	28 cm.	Secondary centre distal end of humerus, distal end of radius, distal extremity of metacarpals 3 and 4
110	11	28 ,,	Secondary centre proximal end of radius. Secondary centre proximal end of tibia, distal end of metatarsals 3 and 4. Five tarsal bones
111	12	32 ,,	Secondary centre proximal end of humerus and coracoid. Secondary centre distal extremity of tibia
120	31	40 ,,	Secondary centre proximal extremity of ulna. Bony union of conjoined ischio-pubic ramus, ossification of patella
124	16	40 ,,	Secondary centres epiphysial plates of vertebrae C_3–L_4. Fusion of bony dorsal spines with laminae. Secondary centre greater tuberosity of humerus. Secondary centre head of femur. Secondary centre proximal end of phalanges 1 and 2 in manus, and trace thereof in pes
128	15	35 ,,	Body of hyoid. Secondary centre os calcis
130 (±1)	18	—	Secondary centre internal and external epicondyles of humerus. Secondary centre tibial tubercle
131	23	—	Secondary centres of epiphysial plates of vertebrae C_1 to S_3. Secondary centre great trochanter of femur
137	21	—	Secondary centre distal extremity of ulna. Secondary centre small trochanter of femur

BEHAVIOUR IN THE EMBRYO AND FOETUS

The number of problems suggested by the differences in skeletal development of the newborn lamb and the human babe is infinite. It has been shown by Barcroft *et al.*(3) that even as early as the 36th day (27–29 mm. crown-rump length) the sheep embryo will display mass movements on touching the nose with a fine glass rod. From the 38th to the 49th days rhythmic trunk movements, closely resembling those of normal respiration, are seen. After the 50th day these rhythmic movements disappear as a spontaneous phenomenon but can be revived by stoppage of the blood flow in the umbilical cord. These early embryonic movements focus attention on the precocious development of the nervous system, and throw considerable doubt on the prevailing view that myelinization of the tracts of the spinal cord determines the onset of function therein.

In man it may be permissible to associate certain steps in the pattern of ossification of the skeleton with certain physiological events. For instance, the first subjective symptoms of early pregnancy coincide with the time of ossification in the clavicle and mandible about the 40th day. The first subjective symptoms of quickening in the mother occur about the 20th week of pregnancy when the tympanic ring and semicircular canals of the foetus are ossifying rapidly and the internal ear has reached virtually adult size; the muscular system is growing rapidly and the vestibulo-spinal tract is myelinating. It is customary in post-natal life to associate the appearance of certain centres of ossification with the cutting of the milk teeth, walking, the cutting of the first permanent molar (6 years), puberty and adolescence. It is difficult to analyse the sequence of events in the lamb, which is born in so advanced a stage in accord with its ability to walk, gambol and graze within a few hours of birth. In comparison with the human the lamb at 130 days of gestation displays in the hindlimb a degree of ossification somewhat akin to that of the human of 7–8 years. The development of the vertebral column is still more advanced, since the epiphysial plates of the vertebrae are at the same stage as those of the human at puberty. Again, it is tempting to associate union of the ilium, ischium and pubis at the acetabulum with the dramatic onset of puberty at about 8 months after birth, but it is difficult to fit in with this the bony union of the conjoined ischio-pubic ramus which occurs about 30 days before birth. It is known in man that increase in length of the lower limb is maximal in the first springing-up period of the suckling, in the second springing-up period associated with eruption of the second dentition and again in the third springing-up period of puberty. In post-natal life the femur grows more than the tibia and the humerus grows more than the radius. In the sheep Hammond(5) has shown that the digits, metacarpals and metatarsals show relatively little post-natal growth as compared with the bones of the more proximal segments. The post-natal growth in the forelimb of the lamb is greater than in the hindlimb. The shorter forelimb in the newborn

may be an advantage from the point of view of suckling, but all four limbs need to be well developed at birth in order that the lamb may keep alongside the ewe in search of food. The advanced stage of the vertebral column may be associated with the great length of the neck and the need for controlling the dependent head in the position of grazing by means of powerful muscles.

The brain weight of the newborn lamb is about 80 g. and that of the adult sheep is about 95 g. so that relatively little post-natal growth occurs. This fits in with the notion derived from the skeleton of the limbs that the age of the lamb at birth is equivalent to 7 or 8 years on the human scale. It is proposed to analyse the body weight and brain weights of this series of sheep with a view to tracing the changes in the Index of Cerebral Value, as previously done in a series of pigs by Harris(6).

REDUCTION OF ELEMENTS IN THE LIMB

In a previous brief communication to *Nature* Harris(7) reported the presence of a rudimentary clavicle in the sheep embryo. Ossification occurs in this rudiment at 39 days. At 45 days no trace of the bony clavicle is seen either by radiographic or staining methods. The presence of a transient fifth metacarpal in the forelimb of the sheep was reported at the same time. The problem of these transient metacarpals has since been studied in some detail, both by bulk staining of cartilage, bulk staining of bone, and serial sections.

In a sheep embryo of 40 days a transverse section of the metatarsal bones illustrates the manner in which the second and fifth metatarsals are present on either side of the third and fourth (Fig. 1 *a*). Two sections taken more distally in the limb (Fig. 1 *b* and *c*) show the manner in which the second and fifth disappear by gradual incorporation in the cortex of the rapidly growing third and fourth, which are beginning to fuse along their contiguous surfaces.

In the hindlimb of a sheep embryo of 45 days the transverse sections (Fig. 2 *a* and *b*) illustrate certain stages in the burial of the second and fifth metatarsals, whilst the latter are in a cartilaginous stage. The absorption of the perichondrium of the smaller metatarsals is more advanced for the fifth than for the second.

In the case of the forelimb the process of burial is not only slower but there is a marked difference between the pre-axial and post-axial borders, inasmuch as the fifth metacarpal actually undergoes ossification so that its disappearance is a matter of some time. In sections of the forelimb of a sheep embryo of 45 days (Fig. 3 *a* and *b*) it is seen that the second metacarpal is absorbed into the third at a time when the fifth is still distinct from the fourth. The fifth metacarpal actually persists and ossifies, appearing as a thin thread of bone both in radiographs and in specimens cleared by the Spalteholz method (Fig. 4) in embryos ranging from 70 to 95 days. This bony fifth metacarpal is about 0·5 mm. in diameter and its length decreases from about 2 cm. at 65 days to 1 cm. at 90 days. It appears to atrophy at the distal end and at 104 days

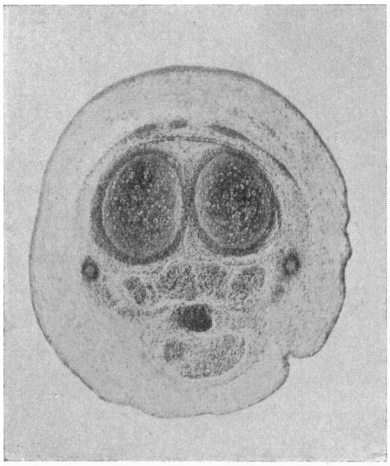


Fig. 1*a*.

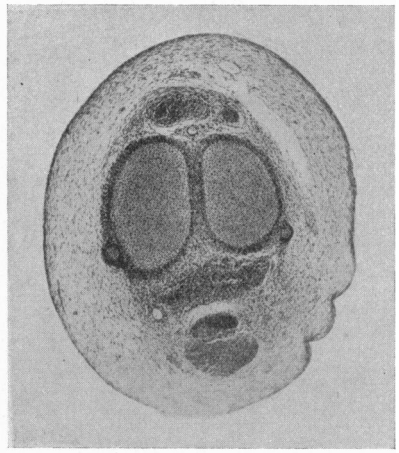


Fig. 1*b*.

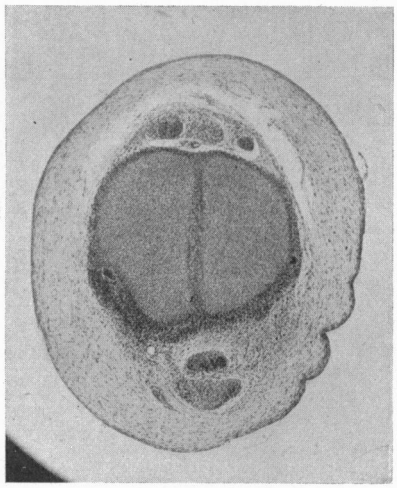


Fig. 1*c*.

Fig. 1 *a*, *b* and *c*. Three sections of the metatarsals of a sheep embryo of 40 days showing the gradual incorporation of the second and fifth in the cortex of the third and fourth metatarsals respectively.

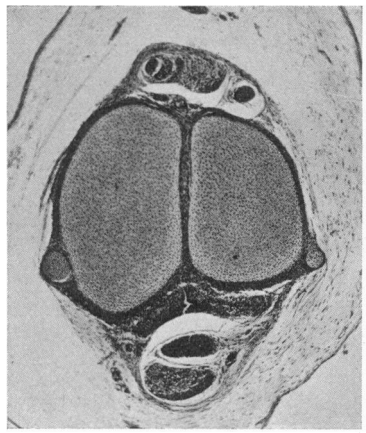


Fig. 2*a*.

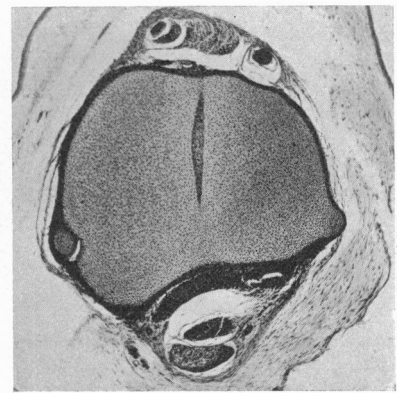


Fig. 2*b*.

Fig. 2 *a* and *b*. Two sections of the metatarsals of a sheep of 45 days showing the stages n the absorption of the second and fifth metatarsals in the cannon bone (third and fourth metatarsals).

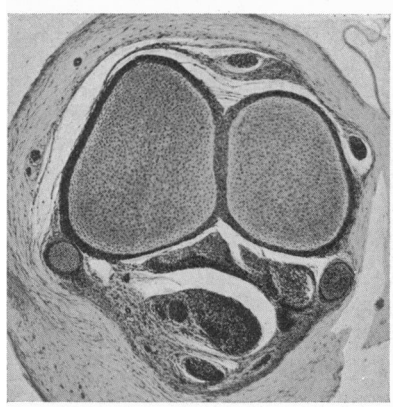


Fig. 3*a*.

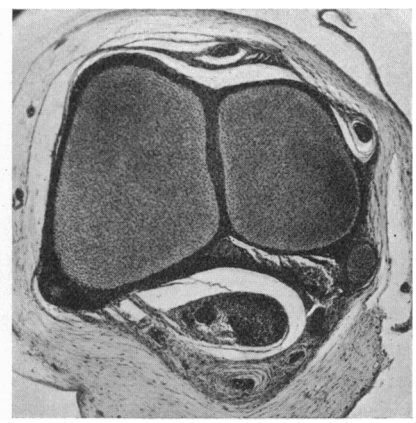


Fig. 3*b*.

Fig. 3 *a* and *b*. Two sections of the metacarpals of a sheep embryo of 45 days. The second metacarpal is in process of absorption but the fifth remains distinct from the fourth and later undergoes ossification.

the proximal end alone is visible as a small nodule of bone with a much attenuated shaft gradually decreasing in diameter distally. At birth the fifth metacarpal varies within wide limits and may consist merely of a proximal end or of a proximal end tapering away into a shaft even as long as 2·5 cm. Its general shape closely resembles that of the fibula of the domestic chick.

It would appear that in the hindlimb the precocious growth of the cannon bone in length and diameter is such in time and extent as to bury the cartilaginous second and fifth metatarsals. In the forelimb the growth of the cannon bone is such that the second metacarpal is buried by the third whilst in the cartilaginous stage, but the relatively slower growth in the diameters of the fourth metacarpal is not sufficient to incorporate the fifth. Thus the fifth metacarpal undergoes ossification *in situ* and is subsequently subjected to a slow process of atrophy.

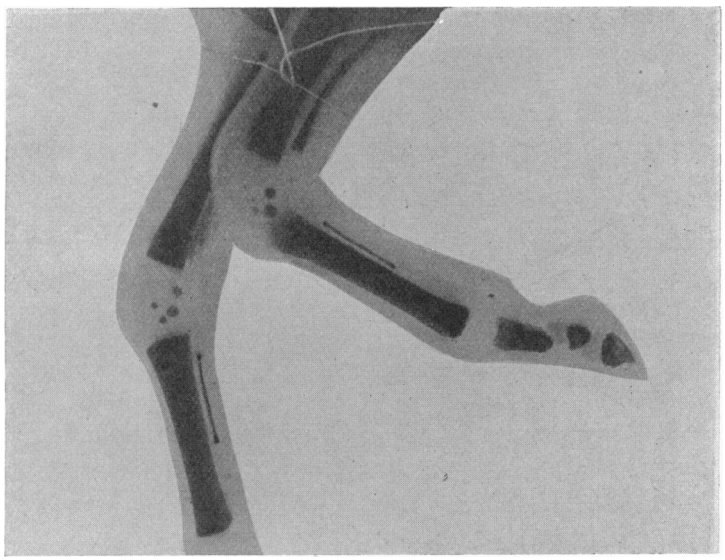


Fig. 4. The forelimb of a sheep embryo of 92 days cleared in oil of wintergreen to show the persistence of the diminutive fifth metacarpal bone at a time when the second has been completely absorbed.

It is well known that in the even-toed ungulates the second and fifth metacarpals display a wide range of variation. In some forms the proximal end persists in bone; in others the distal end. Sedgwick(8) maintains that in the artiodactyls "no traces of the skeletal parts of digits which are totally missing in the adult have so far been discovered in the embryo". It can be clearly stated that in the embryonic form of the sheep all four metacarpals and metatarsals and the corresponding digits are present in mesenchyme or cartilage. The various stages in the reduction of the digits can be traced in the

embryo until at the final stage nothing remains but two digits, plus a trace of the fifth metacarpal in bone and of the terminal phalanges of the second and fifth digits in bone.

Edgeworth (9) has recently indicated that the important problem in morphology is to ascertain whether a given structure undergoes atrophy during life or whether it has ceased to exist. It is well known that a separate bony costal element near the transverse process of the seventh cervical vertebra is present in almost all human foetuses. After birth this fuses with the transverse process. Occasionally it continues to grow and gives rise to the condition known as "cervical rib". The cervical rib sometimes gives rise to a palsy of the small muscles of the hand as a result of pressure on the brachial plexus. The patient is not to be regarded as a freak who has grown a new cervical rib, but as an unfortunate who has failed to keep the embryonic rib within normal bounds by fusion and suppression. Similarly, many of the abnormalities of the hands and feet in the quadrupeds are a failure to absorb and suppress various elements in the normal embryological sequence, rather than the acquirement of additional bony elements in the limbs.

The process of burial and absorption of the bony elements of ungulates may be significant in the study of the distribution of tumours and cysts in the limbs, for the sites of embryological happenings may often be the sites of pathological changes. Finally it is not without interest that the date of ossification in the clavicle of the sheep, the 39th day, is identical with that in the human embryo, as is the ossification in the transient fifth metacarpal in the 8th to 9th week of foetal life. The divergence in the pattern of ossification in the sheep and human forms is negligible in the first 6–8 weeks and becomes of rapidly increasing significance, inasmuch as the sheep at birth has reached a measure of skeletal ossification which is not reached by man until 7 years after birth. The embryological evidence here produced indicates that the ungulates have not departed from the common mammalian pattern to such an extent as is suggested by the study of fossil forms.

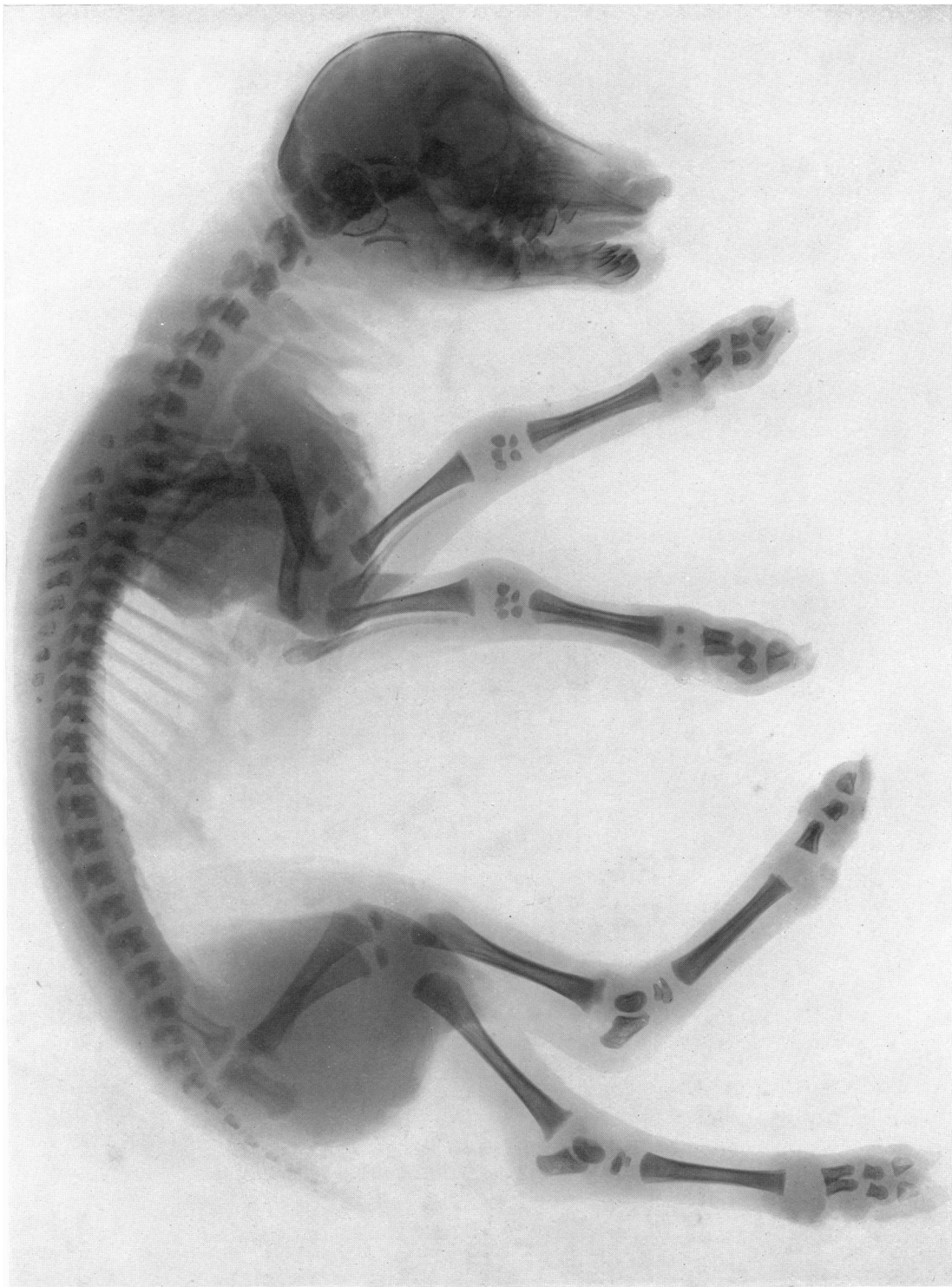
My thanks are due to Mr J. A. F. Fozzard for valuable assistance with the illustrations.

EXPLANATION OF PLATE I

Plate I. Radiograph of a lamb of 104 days, crown-rump length 26·5 cm.

The semicircular canals are heavily ossified. The centre for the distal epiphysis of the femur is present and for the distal epiphysis of the cannon bone of the forelimb.

The precocious bony centres for the spines of the upper dorsal vertebrae are present.



HARRIS—Foetal Growth of the Sheep

REFERENCES

(1) BUNGE, G. (1874). "Der Kali-, Natron- und Chlorgehalt der Milch, verglichen mit dem anderer Nahrungsmittel und des Gesammtorganismus der Säugethiere." *Zeits. f. Biol.* vol. x, p. 326.

(2) HARRIS, H. A. (1933). *Bone Growth in Health and Disease*, p. 211. London.

(3) BARCROFT, J., BARRON, D. H. & WINDLE, W. F. (1936). "The genesis of respiratory movements in the foetus of the sheep." *J. Physiol.* vol. LXXXVIII, p. 56.

(4) PEARSON, K. & DAVIN, A. G. (1921). "On the sesamoids of the knee joint. Part II. Evolution of the sesamoids." *Biometrika*, vol. XIII, pp. 350–400.

(5) HAMMOND, J. (1932). *Growth and Development of Mutton Qualities in the Sheep*, p. 369. Edinburgh.

(6) HARRIS, H. A. (1929). "A preliminary note on the relation of skeletal ossification in the hindlimb to the index of cerebral value of Anthony and Coupin." *J. Anat.*, Lond., vol. LXIII, pp. 267–76.

(7) HARRIS, H. A. (1936). "Atrophy, burial, suppression or total loss in evolution." *Nature*, Lond., vol. CXXXVIII, p. 928.

(8) SEDGWICK, A. (1905). *A Student's Textbook of Zoology*, pp. 579–80. London.

(9) EDGEWORTH, F. H. (1935). *The Cranial Muscles of Vertebrates*. Cambridge.