Fetal development of the bat *Miniopterus schreibersi natalensis*

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Prenatal development in the fetus of *Miniopterus schreibersi natalensis* is discussed. Due to the long period of delayed implantation in this subspecies, the age of even the smallest fetuses, where the fore-limb buds just become visible will be around 150 days. The most reliable criterion for age determination proved to be body mass, and all fetal ages were estimated by using the equation $t = W^{1/3}/0{,}0145 + 144$.
S. Afr. J. Zool. 1981, 16: 172 – 182

Voorgeboortelike ontwikkeling van die fetus van *Miniopterus schreibersi natalensis* word bespreek. As gevolg van die lang periode van vertraagde implantering in hierdie subspesie is die ouderdom van selfs die kleinste fetusse, waar die voorste ledemaatknoppies net sigbaar word, alreeds ongeveer 150 dae. Die betroubaarste maatstaf vir ouderdom is liggaamsmassa, en die ouderdomme van al die fetusse is bereken met die formule $t = W^{1/3}/0{,}0145 + 144$.
S.-Afr. Tydskr. Dierk. 1981, 16: 172 – 182

The growth of an individual from the time of conception until birth is a continuous process but can nevertheless be divided into certain phases of development without altering the basic concept of continuity (Winters, Green & Comstock 1942). Three major periods (i.e. ovum, embryonic and fetal) are generally recognized during prenatal development (Winters *et al.* 1942) and are very concisely defined by Armstrong (1950) as follows: The period of the ovum covers the events from fertilization until implantation or attachment of the blastocyst. The period of the embryo is characterized by organogenesis and internal changes which are accompanied by some changes in external body form. The period of the fetus is characterized by growth and external differentiation.

The setting of the exact age for the ending of the embryonic period and therefore the beginning of the fetal period is a purely arbitrary, but nevertheless useful expedient (Green & Winters 1945). Therefore considerable variation can be expected amongst authors in their choice regarding the onset of the fetal period. Because external morphological changes of the fetus with age are the sole concern of this paper, the fetal period in this study is regarded from the time that all four limb buds are visible until birth. This choice is based on the fact that from the stage that all four limb buds have appeared it is relatively easy to observe external morphological changes with age.

While in most mammals the fetal period, concerned mainly with the external differentiation and growth of organ anlages, takes up the greatest part of gestation (Armstrong 1950), in *M.s. natalensis*, it accounts for only about 36% of the total gestation period (Van der Merwe 1979). This is because the blastocyst lies unimplanted in the uterine lumen for four months (i.e. about 50% of the total gestation period). The period of the embryo accounts for an additional 14% of the total gestation period.

Methods

Analysis of fetal development was taken from the stage when the limb buds just became visible based on the examination of 189 fetuses. These were collected during 1968 and 1972 to 1979 (the majority during 1974) from females in two caves: Sandspruit Cave No. 1 (24°37′S, 27°40′E) and Schurveberg Cave No. 1 (25°48′S, 28°01′E).

Description of the morphological changes occurring with age (see Table 2) are based mainly on 106 fetuses sampled during the fetal period (August – November) of

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1974. Those collected during the other years were mainly used for comparison and descriptive purposes of certain stages.

Fetuses were stored in AFA (i.e. a solution of 95% ethyl alcohol, 40% formalin, glacial acetic acid and distilled water in the ratio of 3:1:1:5). Body mass and crown-rump length were later measured. Because all fetuses are not folded similarly in the uteri, and due to slight contortion and stretching of some fetuses when out of the uterus, crown-rump lengths of fetuses of equal mass varied and were a less reliable parameter for determining age. Body mass proved to be the most reliable indicator of age as determined by using the equation $t = W^{1/3}/0{,}0145 + 144$; where t = age of the fetus in days and $W^{1/3}$ = cube root of body mass in grams (van der Merwe 1979).

The age of fetuses examined ranged from 150 to 244 days. In order to obtain meaningful age classes, the duration of fetal development was divided into periods of seven days (Table 1; column 2) to follow up gross morphological changes at weekly intervals. As the earliest age when all four limb buds were visible was 153 days, this was taken as the start of the fetal development period (Table 1; column 2). With the aid of the general equation, the expected body mass (W) for the lower and upper limits of each age class in column 2 (Table 1) was then calculated in column 3 (Table 1). However, as the use of the equation resulted in discontinuous mass classes for each age group, the data in column 3 (Table 1) were smoothed in column 4 (Table 1) to provide a continuous body mass spectrum.

All fetuses collected were then grouped according to the age classes as in Table 1, and the mean ± 1SD of their age (Table 1; column 6) and mass (Table 1; column 7) calculated for each age class. The fetuses of each group were then examined to establish gross morphological characteristics for each age class. Drawings were made with the aid of a dissecting microscope.

Results

At an average age of 154 days all four limb buds of the fetus could be distinguished as well as most of the structures participating in the formation of the face and jaws (Figure 1). From this age onwards to about 197 days of age, conspicuous external changes occurring with age, made it possible to group the fetuses into age classes according to certain morphological parameters. These parameters are summarized in Table 2. Many of these external characteristics overlapped from one age group to another, but normally the presence or absence of certain structures made it possible to distinguish fetuses of various age classes. Only from 198 to about 224 days of age was it not possible to group fetuses into age classes on pure gross morphological characteristics as, by that time, external differentiation was completed and only elongation of the appendages and increase in body mass occurred. From about 225 days of age, the appearance of other characteristics such as the deciduous teeth, again made it possible to recognise age classes.

The paired maxillary processes (Figures 1 & 3), participating in the formation of the upper jaw, gradually became more prominent and grew towards the mid-line of the oral cavity while the frontal process (Figure 3) got narrower. Due to this action the olfactory pits, originally situated more laterally (Figures 3 & 4) were gradually being pushed towards each other so that they had reached a complete anterior position (Figure 7) around 166 days of age. Elevations forming around the olfactory pits eventually gave rise to the medial nasal and lateral nasal processes (Figures 3 & 7). The auditory vesicles, situated in the vicinity of the hyoid arches (Figure 1), were quite conspicuous at an age of 154 days. However, they soon became more indistinct as the overlying skin layers thickened and were completely invisible from 162 days of age.

Table 1 Age classes with estimated lower and upper limits for age and body mass for each age class. The number (n) and mean ± 1 SD for the age and body mass of the fetuses grouped according to the estimated age classes, are included

1	2	3	4	5	6	7
Age class 1	Lower and upper limit for each age class (days) 2	W^a for limits of each age class (g)	Selected W^a values for each age class (g)	n^b	Mean ± 1 SD for: Age (days)	Mass (g)
1	153 – 159	0,002 – 0,010	0,002 – 0,011	15	156 ± 2	0,006 ± 0,003
2	160 – 166	0,012 – 0,032	0,012 – 0,034	14	163 ± 2	0,020 ± 0,005
3	167 – 173	0,037 – 0,074	0,035 – 0,078	10	169 ± 3	0,051 ± 0,016
4	174 – 180	0,082 – 0,142	0,079 – 0,148	8	178 ± 3	0,117 ± 0,027
5	181 – 187	0,154 – 0,242	0,149 – 0,251	6	184 ± 3	0,196 ± 0,043
6	188 – 194	0,259 – 0,381	0,252 – 0,393	9	190 ± 2	0,296 ± 0,031
7	195 – 201	0,404 – 0,564	0,394 – 0,580	13	198 ± 2	0,487 ± 0,053
8	202 – 208	0,595 – 0,799	0,581 – 0,818	15	205 ± 3	0,697 ± 0,090
9	209 – 215	0,837 – 1,091	0,819 – 1,115	16	213 ± 2	0,986 ± 0,091
10	216 – 222	1,138 – 1,447	1,116 – 1,476	23	219 ± 2	1,280 ± 0,080
11	223 – 229	1,504 – 1,873	1,477 – 1,907	11	226 ± 2	1,680 ± 0,201
12	230 – 236	1,940 – 2,376	1,908 – 2,415	24	233 ± 2	2,137 ± 0,176
13	237 plus	2,454 plus	2,416 plus	25	240 ± 2	2,744 ± 0,142

$^a W$ = Body mass in gram.
$^b n$ = Sample size.

Table 2 Morphological changes of the fetus at different ages

Age class	Lower and upper limit of age and body mass for each age class	Description
1	153 – 159 days 0,002 – 0,011 g	*Head and neck* General: Head has fetal appearance (i.e. incomplete and/or out of proportion). No defined rostrum. Inconspicuous neck. Eyes: Optic vesicles conspicuous. Ears: Auditory vesicles visible but no signs of external ears (Figure 1). Nose: Olfactory pits absent or may just become visible as slits laterally. Upper jaw: On both sides of the face the maxillary process becomes visible as a slight bump on the anterior proximal base of the mandibular arch (Figure 1). Branchial arches: Mandibular and hyoid arches prominent and more or less equal in size and shape from a lateral view (Figure 1). Mandibular arch (forming lower jaw) paired or left and right components uniting medially on the ventral surface (Figure 2). Third branchial arch inconspicuous or absent as it becomes incorporated in the neck. *Appendages* Anterior limbs: Forelimb buds conspicuous and may have a length equal to its width (Figures 1 & 14). Posterior limbs: Hindlimb buds visible as slight bumps at the base of the tail or may be more prominent but still shorter than broad (Figures 1 & 14). Tail: Short, terminating with a knob (Figures 1 & 14) or may have adopted a normal appearance. *Neural tube* Roofplate of the neural tube may be transparent or becoming opaque over its entire length, except for the myelencephalon area where it is still completely transparent.
2	160 – 166 days 0,012 – 0,034 g	*Head and neck* General: Head has fetal appearance. No defined rostrum. Inconspicuous neck. Eyes: Eyes open with no eyelids (Figure 6). Ears: Auditory vesicles not visible due to the thickening of the skin layers. Location of the external ears becoming obvious between the mandibular and hyoid arches (Figure 6) but no signs of a pinna or tragus. Nose: Olfactory placodes invaginate to form obvious slits laterally (Figure 4), or the olfactory pits may already be well established on the anterior surface of the face, with the lateral nasal and medial nasal processes becoming prominent (Figures 3 & 7). Upper jaw: Maxillary processes prominent and may nearly be as long as the mandibular arch from a lateral view. Arch of the upper jaw becoming completed. Branchial arches: Mandibular and hyoid arches loose similarity from a lateral view, with the mandibular arch adopting a more square appearance and the hyoid arch a more triangular appearance (Figure 5), or the hyoid arch may become more or less completely incorporated in the neck with only the outline slightly visible. Left and right components of the mandibular arch completely united medially on the ventral surface to complete the formation of the lower jaw (Figures 3 & 7). Third branchial arch not visible. *Appendages* Anterior limbs: Forelimb buds longer than broad or may be longer than broad with the handplates starting to form (Figures 4, 5, 6 & 14). Posterior limbs: Hindlimb buds of equal length and width or may be longer than broad but with no footplates visible (Figures 4, 5, 6 & 14). Tail: Normal in appearance. Patagium: Plagiopatagium may become visible as a skinfold posterior and proximal to the forelimb bud (Figures 5, 6 & 14). *Neural tube*: Roofplate of the neural tube opaque over its entire length except for the myelencephalon area where it is still relatively transparent.
3	167 – 173 days 0,035 – 0,078 g	*Head and neck* General: Head has fetal appearance. No defined rostrum. Inconspicuous neck. Eyes: Eyes open while perimeter of eyes may become pigmented. No sign of eyelids (Figure 8). Ears: Location of external ears obvious and the pinna and tragus may just become visible. Nose: External nose openings prominent and conspicuous on the anterior surface of the face. Upper jaw: Arch of upper jaw completed. Branchial arches: Hyoid arch may become completely invisible. *Appendages* Anterior limbs: Handplates well formed and may start adopting a triangular appearance (Figures 8 & 14). Posterior limbs: Footplates may start to form with roundish appearance (Figures 8 & 14). Patagium: Plagiopatagium covers a quarter of the lateral body wall between the fore- and hindlimbs (Figure 14). Uropatagium may become visible at the posterior proximal base of the hindlimbs (Figure 14). *Neural tube*: Roofplate of the myelencephalon almost opaque.
4	174 – 180 days 0,079 – 0,148 g	*Head and neck* General: Head has fetal appearance. Rostrum becoming prominent. Neck becoming prominent. Eyes: Eyes open with clear pigmented ring on the perimeter. Eyes may become slightly oval due to developing upper and lower eyelids. Ears: Pinna and tragus obvious (Figure 9). Pinna short and not covering the tragus. *Appendages* Anterior limbs: Upper and lower arms may start differentiating and becoming distinguishable from each other. Handplates triangular in shape (Figure 10) and the handbones start to differentiate. Thumbs becoming distinctive. Posterior limbs: Upper and lower legs may start differentiating and becoming distinguishable from each other. Footplates prominent and round in shape (Figure 10), and footbones may start to differentiate. Tip of toes may start to separate from each other. Patagium: Plagiopatagium covers more than half of the lateral body wall or may become united with the anterior proximal base of the hindlimbs and may even cover up to three-quarters of the anterior surface of the hind legs. Uropatagium may include from a quarter to three-quarters of the tail with the rest of the tail still naked (Figure 10). Propatagium may become visible as a skinfold anterior and proximal to the forearms or may even stretch as far as the elbow (Figures 10 & 14). Chiropatagium may become distinguishable.

Table 2 continued

Age class	Lower and upper limit of age and body mass for each age class	Description
		Neural tube: Roofplate of the myelencephalon completely opaque.
5	181 – 187 days 0,149 – 0,251 g	*Head and neck* General: Head has fetal appearance. Rostrum prominent (Figure 11). Neck prominent. Eyes: Eyes oval to slitlike because of the developing upper and lower eyelids. Ears: Pinna may just cover the tragus, or else the apex of the tragus may still be visible from a lateral view. *Appendages* Anterior limbs: Upper and lower arms prominent (Figure 11). Metacarpals as well as digits 2 to 5 differentiating and becoming distinguishable from each other. Finger joints may just become visible. Thumbs already distinct and claws may become visible. Posterior limbs: Upper and lower legs prominent (Figure 11). Metatarsals and digits differentiating with the toes becoming separated. This separation may involve only the tip of the toes, or may stretch as far as the base of the toes. Claws may become visible. Feet may become well defined from heal to toes. Patagium: Plagiopatagium covering the entire posterior part of the forelimbs stretches along the lateral body wall and may cover from half to the entire anterior side of the hindlimbs (Figures 11 & 14). Uropatagium includes half or entire tail. Propatagium may cover from half to the entire anterior side of the forearms (Figures 11 & 14). Chiropatagium conspicuous between digits 2 to 5.
6	188 – 194 days 0,252 – 0,393 g	*Head and neck* General: Head adopts adult appearance. Sagittal and coronal sutures obvious. Eyes: Eyes completely closed. Ears: Pinna flat against the head and completely covers the tragus. *Appendages* Anterior limbs: Upper and lower arms differentiated. Metacarpals and digits differentiated with the finger joints extraordinarily large. Claws on thumbs visible as blunt stubs. Posterior limbs: Upper and lower legs differentiated. Toes differentiated but may still be united at the base. Claws visible as blunt stubs. Patagium: Plagiopatagium stretches from the fifth finger to the ankle of the hindlegs (Figure 14). Uropatagium includes entire tail from the base to the tip. Propatagium is prominent and covers the entire anterior surface of the forearms. Chiropatagium is prominent and conspicuous.
7	195 – 201 days 0,394 – 0,580 g	Claws easily observable. Finger joints large (swollen appearance) (Figure 13). Third digit of hand: Second phalanx is 2–2½ times longer than the first phalanx (Figure 13). Fourth digit of hand: Second phalanx is 1–1½ times longer than the first phalanx (Figure 13). Phalanges of toes becoming conspicuous. Caudal vertebrae prominent. All the external characteristics are differentiated, and from here-on only elongation of the different parameters occur with an increase in body mass.
8	202 – 208 days 0,581 – 0,818 g	
9	209 – 215 days 0,819 – 1,115 g	
10	216 – 222 days 1,116 – 1,476 g	Up to this stage the colour of the skin is whitish due to the lack of skin pigmentation.
11	223 – 229 days 1,477 – 1,907 g	Lower deciduous incisors and canines just visible underneath the surface of the gums or may just start piercing the gums. Upper deciduous incisors and canines not visible but gums swollen. Few hairs on upper lip. Only visible under high magnification. Colour more pinkish due to skin pigmentation.
12	230 – 236 days 1,908 – 2,415 g	Lower deciduous incisors and canines through gums. Upper deciduous incisors and canines just below the surface of the gums or may start piercing the gums. Few hairs on upper and lower lips as well as the eyelids and toes.
13	237 days plus 2,416 g plus	Deciduous incisors and canines erupt through the gums, are sharp and the canines recurved. Claws well formed and sharp. Many hairs covering upper and lower lips as well as the eyelids. Not easily seen with the naked eye. Few hairs on the toes and base of the tail.

In *M.s. natalensis* the pinna and tragus of the ear (Figure 9) only became conspicuous at an age of 174 days. During development in the uterus, the ear is folded flat against the head, and grows forward against the side of the head. In this manner, the tragus eventually became covered by the pinna (Figure 12) at an age of 188 days. Only after birth do the ears become erect with the tragus again visible from the side. Development of the limbs, tail and patagium is illustrated in Figure 14 and summarized in Table 2. Technical terms used to distinguish between the different parts of the flight membrane (patagium) follow Vaughan (1978) and can be divided into four conspicuous areas: propatagium — flight membrane from the arm to the occipitopollicalis muscle i.e. the flight membrane in front of the forelimb; chiropatagium — flight membrane between the fingers i.e. between digits two to five; plagiopatagium — flight membrane extending from the body and the hind limbs to the arm and fifth digit and the uropatagium — flight membrane from hind limbs to tail.

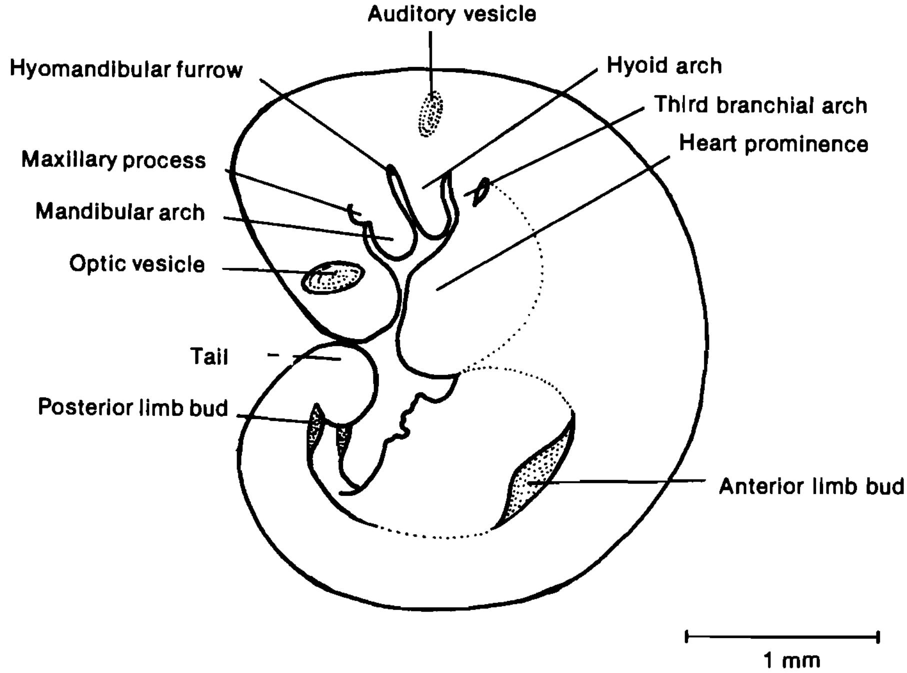

Figure 1 Drawing of a 154-days-old *M.s. natalensis* fetus showing its external appearance.

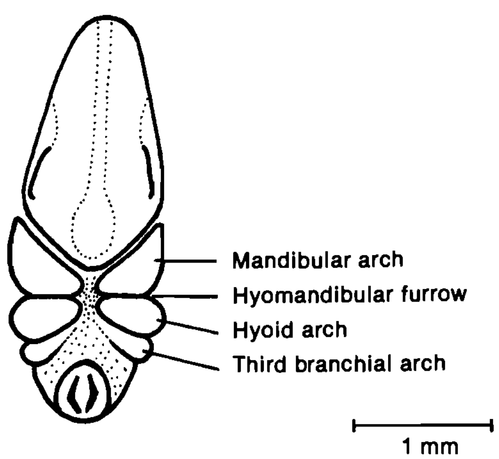

Figure 2 Drawing of the face of a 154-days-old *M.s. natalensis* fetus. Here the mandibular arch is still paired and not fused on the mid-ventral line.

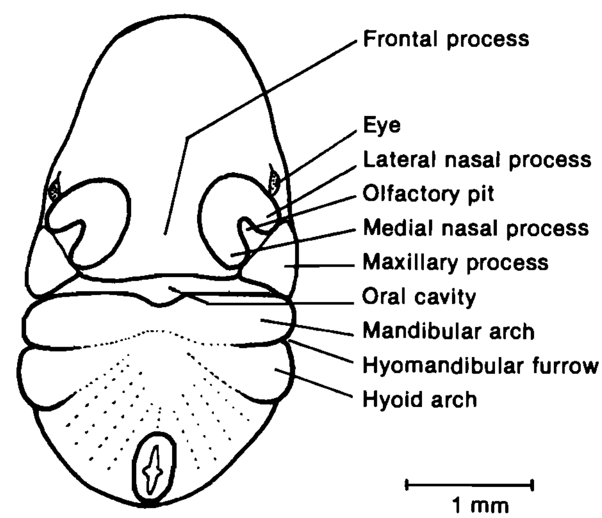

Figure 3 Drawing of the face of a 162-days-old *M.s. natalensis* fetus. Here fusion of the right and left components of the mandibular arch is completed. Both medial and lateral parts of the horseshoe-shaped nasal processes are conspicuously enlarged, and the maxillary processes are conspicuous.

Discussion

Delayed implantation in bats is not a common ordinal reproductive strategy (Fleming 1971) and mainly occurs in the hibernating members of the genus *Miniopterus* (Wimsatt 1969, 1975). Therefore, in these bats the ages of comparable fetuses will always be much older than in related species following the customary mammalian pattern. In *M.s. natalensis* the smallest fetuses will always be 150 days and older because roughly 64% of the total gestation period is occupied by events such as delayed implantation (about 50%) and organogenesis during the embryonic stage (about 14%) (Van der Merwe 1979).

Growth of the fetus is reasonably constant for a given species as it is not greatly influenced by external factors (Fairall 1969) suggesting a fairly constant gestation length for a given species. This is, however, a moot point as it has been shown beyond doubt that the length of gestation in many species is influenced by altering environmental factors. In the golden hamster gestation length increases with increased maternal age (Soderwall, Kent, Turbyfill & Britenbaker 1960) while in the rat it increases with increased environmental temperature (Pennycuik 1964). In some species again, gestation length varies inversely with litter size (Goy, Hoar & Young 1957; Dewar 1968). Racey (1973) working on pipistrelle bats has shown that the length of gestation depends on conditions of temperature and food supply during pregnancy and concludes that the length of gestation in heterothermic bats may be profoundly affected by environmental conditions. Racey (1973) feels that since bats showing similar rates of fetal development must have maintained similar body temperatures, and since the length of gestation depends on food supply and environmental temperature, the con-

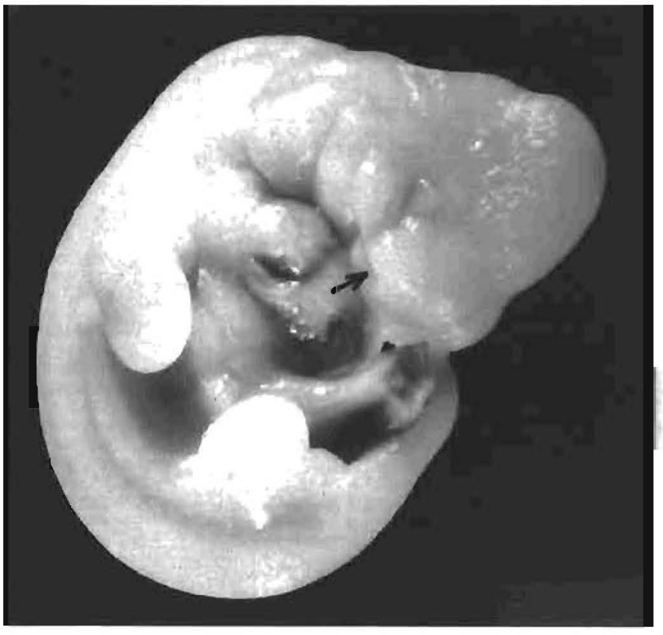

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Figure 4 Ventro-lateral view of a 164-days-old *M.s. natalensis* fetus showing the dextral (laterally situated) olfactory pit as a definitely circumscribed depression at the rostral end of the head (indicated with an arrow).

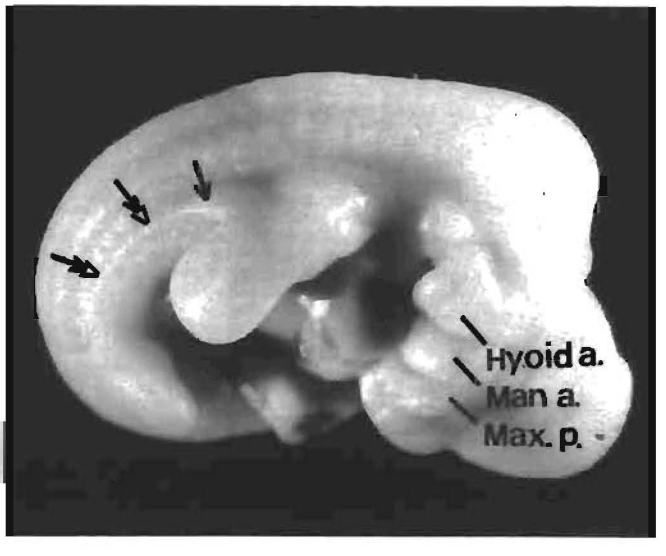

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Figure 5 Lateral view of a 164-days-old *M.s. natalensis* fetus showing the maxillary process (Max. p.), mandibular arch (Man. a.) and the triangular shaped hyoid arch (Hyoid a.). The plagiopatagium is just becoming visible posterior and proximal of the forelimb bud (arrowed) with the ridge which will give rise to the rest of the plagiopatagium conspicuous between the fore- and hindlimb buds (double pointed arrow).

cept of a fixed gestation period for heterothermic bats should be modified. From this it becomes clear that the gestation period of *M.s. natalensis* may vary from year to year and also within the same year between members of this subspecies living in different areas with different environmental conditions.

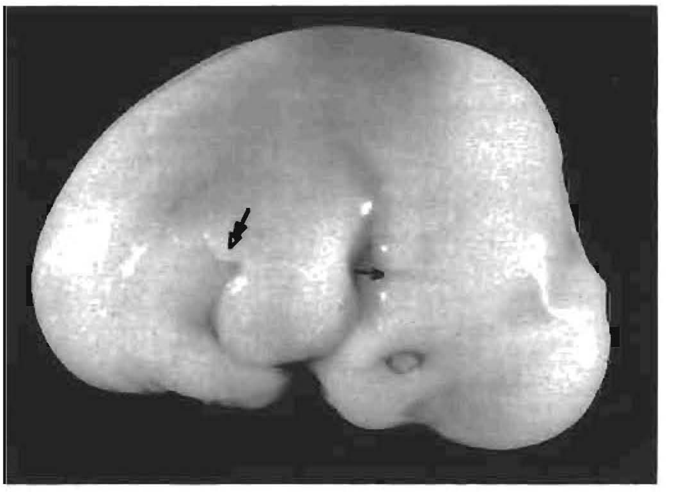

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Figure 6 Lateral view of a 166-days-old *M.s. natalensis* fetus showing the position of the external ear (arrowed). The plagiopatagium is visible as a skinfold posterior and proximal of the forelimb bud (double pointed arrow). The handplates have already started to form and are round in appearance while footplates are still not visible.

However, having an open mind on the concept of varying gestation lengths for the same species, in the same natural area, it must nevertheless be borne in mind that there is a great difference between natural and artificial conditions. Animals enclosed against their will under laboratory and experimental conditions may react differently when compared with the situation in their natural habitat. Being adapted to a certain natural area, with all its environmental fluctuations, is the result of many years of selection for the specific conditions prevailing there. Furthermore, in their natural surroundings, bats have a free choice to move to a more favourable area e.g. caves or where sufficient food sources are, should this become necessary.

In the maternity cave Sandspruit Cave No. 1 (24°37'S; 27°40'E) the first born *M.s. natalensis* were up to now, always found during the last week of October. The first ovulations and conceptions in this subspecies occur during the beginning of March — well in advance of the more unfavourable hibernation period (May to July). Therefore with ovulations and conceptions well in advance of the more unfavourable winter months there is no apparent way of predicting unfavourable winter conditions other than the normal, and therefore no reason why the ovulation conception period should be altered. Should extraordinarily severe winter conditions occur, these may result in later implantations or initial slower embryonic development or the converse under more favourable conditions, which will affect the time of parturition. Because such adverse conditions in the temperate regions are unlikely and furthermore, as in the present case, where no records are as yet available for altering parturition periods, it is doubtful whether significant variation in the mean gestation length of *M.s. natalensis*, in the present study area, can be expected.

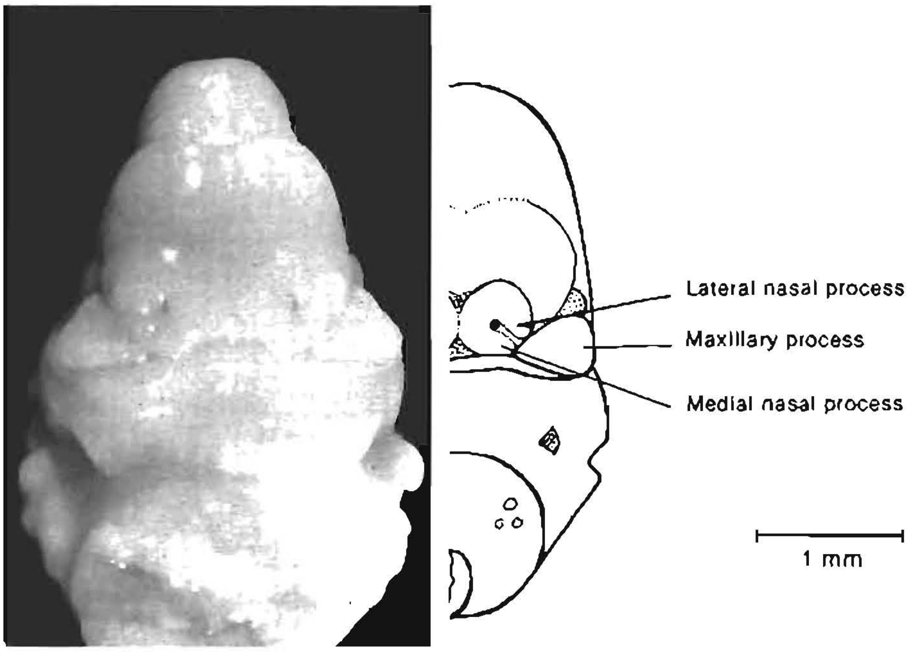

Figure 7 Photo and drawing of the face of a 166-days-old *M.s. natalensis* fetus. The medial nasal processes are more or less fused with each other on the mid-line of the face as well as with the maxillary processes to complete the arch of the upper jaw.

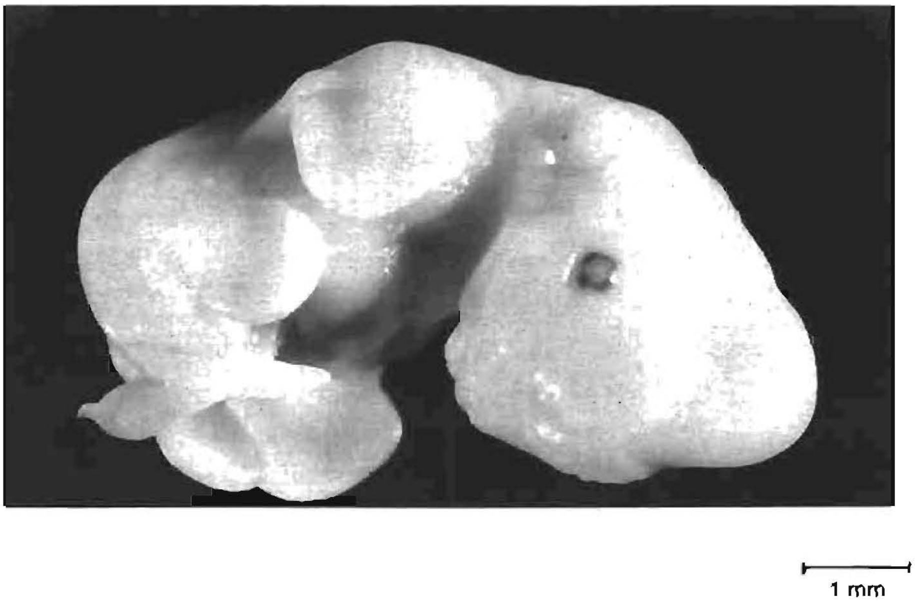

Figure 8 Ventro-lateral view of a 173-days-old *M.s. natalensis* fetus showing pigmented perimeter of the eye. The handplates are triangular in shape with the footplates round in appearance.

Adaptation to live on the southern Transvaal Highveld where the unfavourable winter months (June to August) fall somewhere within the gestation period of *M.s. natalensis* must have been accounted for over many generations. The hibernation period (May to July) in this subspecies is characterized by various types of activity (Van der Merwe 1973; Norton & Van der Merwe 1978) and does not fully coincide with the winter period (Van der Merwe 1980). Implantations nevertheless are initiated during the more unfavourable winter months (Van der Merwe 1979, 1980) and not thereafter as could be anticipated. Because there is no evidence presently available of changes in the above pattern, or changes in the parturition period of *M.s. natalensis* in the present study area, there is no reason to suspect significant variation in the mean length of the gestation period. At least in some species, even under laboratory conditions the gestation period appears to be unaffected by altering environmental temperatures as shown by Barnett & Little (1968) with various strains of mice. Sod-Moriah (1971) who has found an increased incidence of ova degeneration in heat-acclimated rats maintained at 35 °C has found no alteration in the mean duration of the gestation period. In conclusion it can be said that differences in gestation length could be expected between *M.s. natalensis* populations living in different areas under differing environmental conditions. However, in the same area only small individual variations can be expected and there is no reason at present to suspect variation in the mean gestation length.

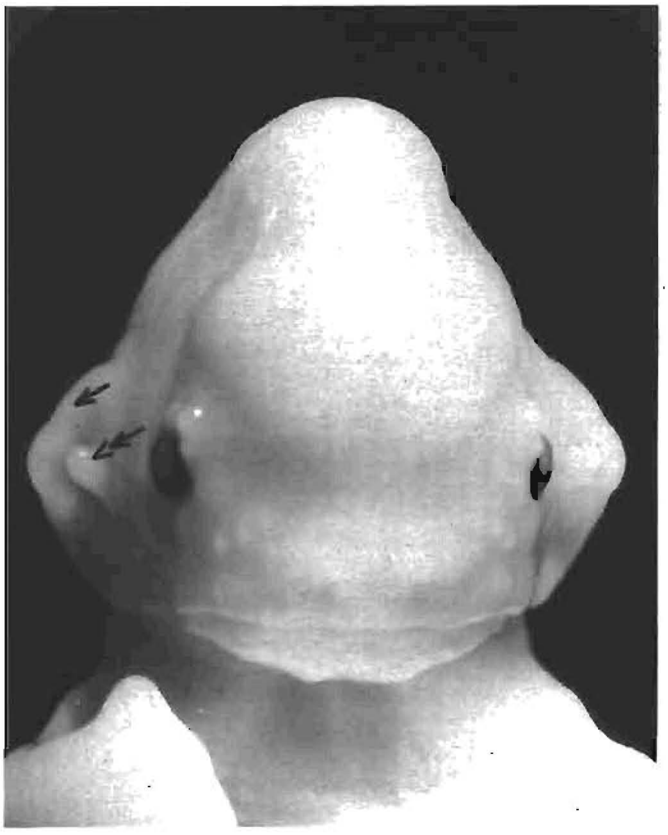

Figure 9 Face of a 177-days-old *M.s. natalensis* fetus showing the developing pinna (arrowed) and tragus (double pointed arrow).

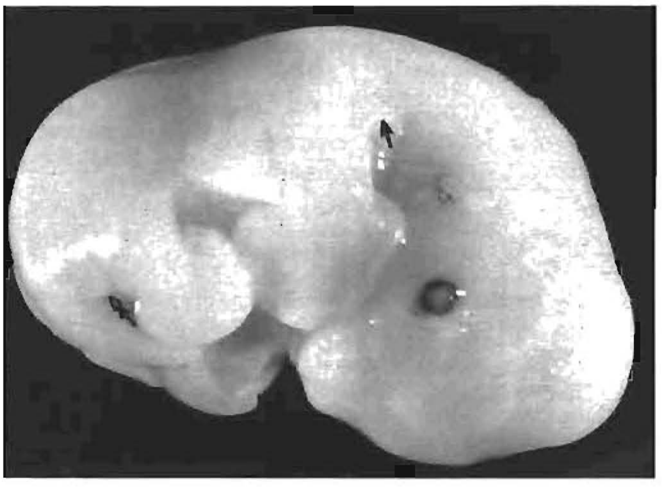

Figure 10 Lateral view of a 177-days-old *M.s. natalensis* fetus where the propatagium is just becoming visible anterior and proximal of the forearms (arrowed). The uropatagium (double pointed arrow) is visible at the posterior proximal base of the hindlimbs where it includes a quarter of the tail.

All fetal ages in the present study are approximate values subject to individual variation. Although morphological characters appear to be the best criteria for establishing the stage of development of a fetus, there are limitations to this approach (Balinsky 1975). One of these limitations is that of heterochrony (i.e. the unequal rate of development of different parts of the body) and this was also anticipated during the present study, for example, two fetuses of the same age and with similar facial structures may differ in the degree of development of their limb buds or patagium. Another problem is that fetal ages can be influenced by the way equations are used in the determination of their ages. One such problem lies in the choice of the number of decimal places used for certain calculations in the general equation, and may be a common problem for all species concerned where equations are used. In *M.s. natalensis* for example, the fetal growth velocity constant (a) was used with four decimal places (a = 0,0145). It was found that ages were not influenced when using more than four decimal places.

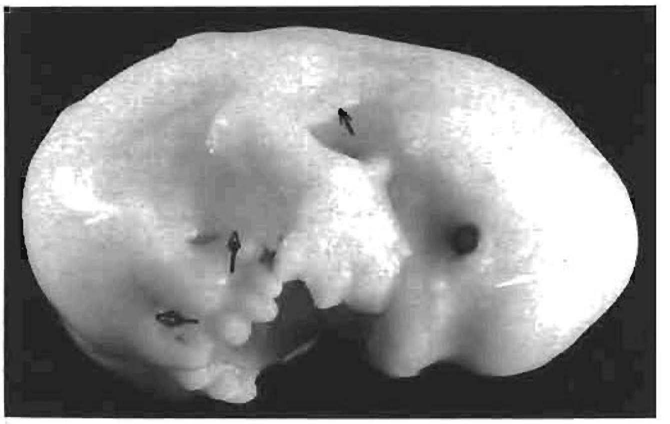

Figure 11 Lateral view of a 181-days-old *M.s. natalensis* fetus with prominent rostrum. The eyes are becoming slightly oval due to the developing upper and lower eyelids. Thumbs are distinctive and the finger joints are starting to show. The propatagium (arrowed) is conspicuous and the plagiopatagium (double pointed arrow) nearly covers the entire anterior surface of the hindlimbs. The uropatagium (triple pointed arrow) includes three-quarters of the tail. The chiropatagium stretching between the fingers is conspicuous.

Figure 12 Face of a 202-days-old *M.s. natalensis* fetus showing the pinna growing forward and thus covering the tragus.

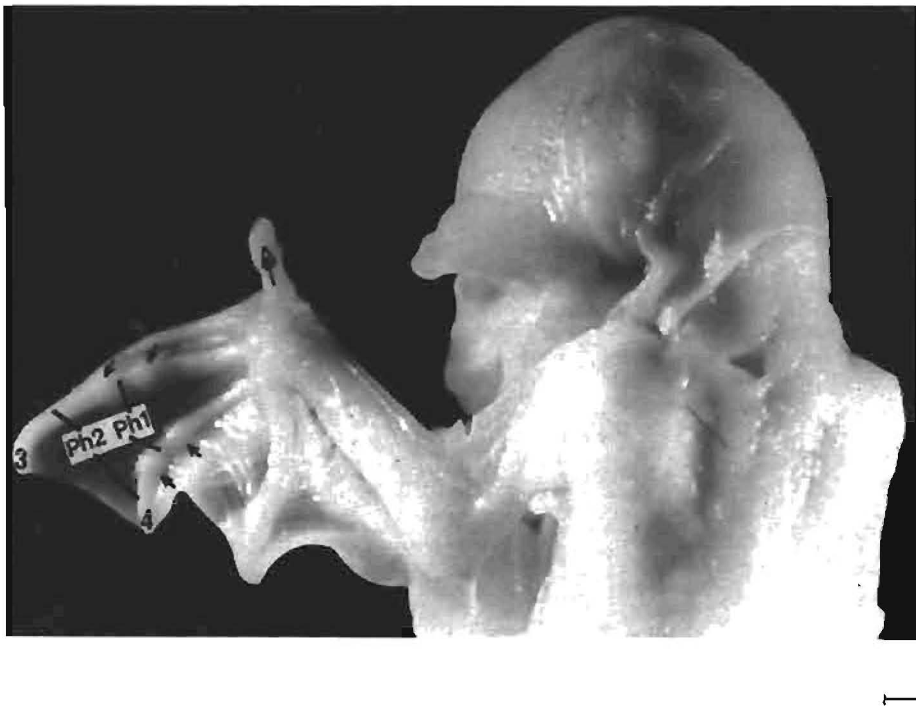

Figure 13 Outstretched arm of a 197-days-old *M.s. natalensis* fetus showing enlarged (swollen) finger joints (arrowed) and the difference in length between the 1st (Phl) and 2nd (Ph2) phalanges of the third digit (No. 3) as well as that of the fourth digit (No. 4). The claw on the thumb (double pointed arrow) is conspicuous.

However, when using three decimal places (a = 0,015) a difference of up to one day was found with small fetuses and up to three days with large fetuses. For example, the ages of fetuses with a mass of 0,002 g and 2,9 g are calculated as 153 and 242 days respectively with a = 0,0145 (or more than four decimal places). However, with a = 0,015 the ages of the same two fetuses are 152 and 239 days respectively.

By definition the period of the fetus is concerned mainly with the external differentiation and growth of organ anlages, and in the present study involved all fetuses with visible anterior and posterior limb buds. The slightly precocious development of the anterior limb buds in *M.s. natalensis* may be a common phenomenon in mammals. At least in the pig *Sus scrofa* the anterior pair of 'bud-like' tissue masses appear somewhat earlier than the posterior pair (Patten 1948). In the present study the earliest indication of anterior limb buds was found in a 150-day-old fetus. The youngest fetus however, where both the anterior and posterior limb buds were visible, was 153 days of age. Only four fetuses were younger than 153 days of age, and in all of them only the anterior limb buds were visible although very inconspicuous in the one of 150 days old. In these small fetuses there were no signs of the maxillary processes, although the mandibular and hyoid arches were well developed and prominent, differing only slightly from those of fetuses between 153 and 159 days of age.

Both pinna and tragus of the ear develop from mesenchymal tissue surrounding the meatus acusticus externus which is already a prominent opening between the mandibular and hyoid arches before any traces of the pinna or tragus can be seen. The external ear forms from the ectodermal furrow (i.e. the hyomandibular furrow) lying between the first and second branchial (i.e. the mandibular and hyoid) arches (Gilchrist 1968). Several nodular enlargements appear, some of them arising from the mandibular arch tissue and others from the hyoid arch along the caudal border of the hyomandibular furrow and by coalescence and further development of these tubercles, the pinna of the ear is then moulded (Patten 1964).

Before birth, the deciduous incisors and canines become sharp with the canines recurved. This is an important adaptation making it possible for the neonates to cling successfully to a nipple. Although the teeth are not truly external their presence or absence can be determined without sectioning. Both teeth and claws are important for anchorage onto the females, especially for the first few hours after birth where they stay attached before being deposited onto the ceiling of the cave. In *M.s. natalensis* and presumably all members of the Chiroptera, elongation of especially the front appendages is much retarded for space economy within the uterus, so that the forelimbs and digits look disproportionately small at birth. The thumbs and hind feet however, are already very well developed when born, with the hind feet very near adult dimensions (Van der Merwe 1978). The reason for the very well developed thumbs and especially the hind feet is to assist the neonates in clinging to the ceiling of the cave, as they are deposited there from only a few hours after birth. There they must stay, in juvenile clusters, throughout the juvenile period with the adult females only coming into contact with them when suckling.

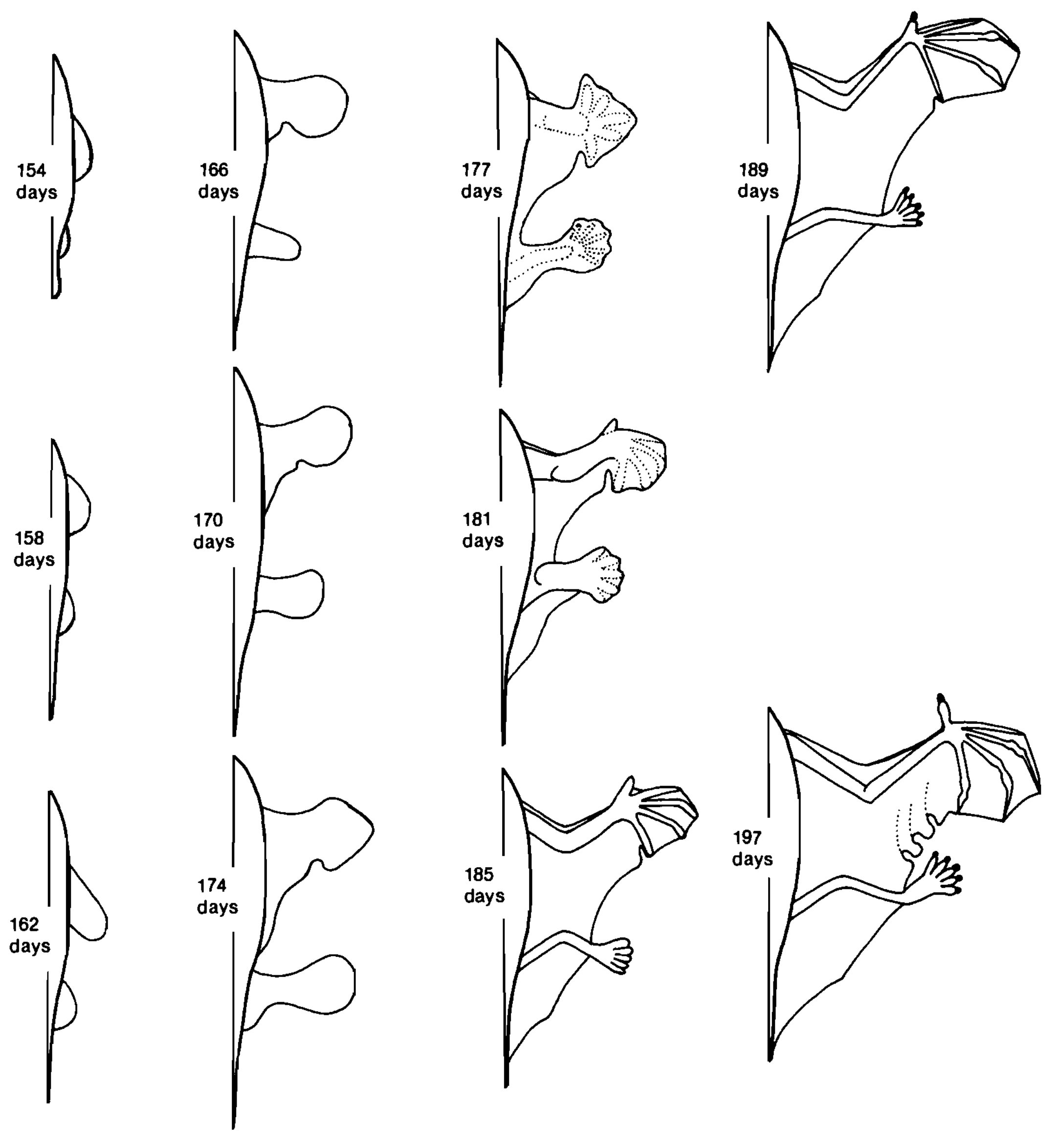

Figure 14 Schematic illustrations showing the development of the limbs and patagium in *M.s. natalensis*.

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References

ARMSTRONG, R.A. 1950. Foetal development of the northern white-tailed deer (*Odocoileus virginianus borealis Miller*). *Am. Midl. Nat.* 43: 650 – 666.

BALINSKY, B.I. 1975. An introduction to embryology, 4th edn., W.B. Saunders, Philadelphia.

BARNETT, S.A. & LITTLE, M.J. 1968. Conception and parturition of mice at two temperatures. *J. Reprod. Fert.* 15: 295 – 304.

DEWAR, A.D. 1968. Litter size and the duration of pregnancy in mice. *Q. Jl. exp. Physiol.* 53: 155 – 161.

FAIRALL, N. 1969. Prenatal development of the impala *Aepyceros melampus*. Licht. *Koedoe* 12: 97 – 103.

FLEMING, H. 1971. *Artibeus jamaicensis*: delayed embryonic development in a neotropical bat. *Science* 171: 402 – 404.

GILCHRIST, F.G. 1968. A survey of embryology. McGraw-Hill, New York.

GOY, R.W., HOAR, R.M. & YOUNG, W.C. 1957. Length of gestation in the guinea pig with data on the frequency and time of abortion and still-birth. *Anat. Rec.* 128: 747 – 757.

GREEN, W.W. & WINTERS, L.M. 1945. Prenatal development of the sheep. *Univ. Minn. Agr. Exp. Sta. Tech. Bull.* 169: 1 – 36.

NORTON, P.M. & VAN DER MERWE, M. 1978. Winter activity of bats in a Transvaal Highveld cave. *S. Afr. J. Sci.* 74: 216 – 220.

PATTEN, B.M. 1948. Embryology of the pig, 3rd edn. McGraw-Hill, New York.

PATTEN, B.M. 1964. Foundations of embryology, 2nd edn. McGraw-Hill, New York.

PENNYCUICK, P.R. 1964. The effects on rats of chronic exposure to 34 °C. IV. Reproduction. *Aust. J. biol. Sci.* 17: 245 – 260.

RACEY, P.A. 1973. Environmental factors affecting the length of gestation in heterothermic bats. *J. Reprod. Fert., Suppl.* 19: 175 – 189.

SODERWALL, A.L., KENT, H.A., TURBYFILL, C.L. & BRITENBAKER, A.L. 1960. Variation in gestation length and litter size of the golden hamster, *Mesocricetus auratus*. *J. Geront.* 15: 246 – 248.

SOD-MORIAH, U.A. 1971. Reproduction in the heat-acclimatized female rat as affected by high ambient temperature. *J. Reprod. Fert.* 26: 209 – 218.

VAN DER MERWE, M. 1973. Aspects of hibernation and winter activity of the Natal clinging bat *Miniopterus schreibersi natalensis* (A. Smith 1834) on the Transvaal Highveld. *S. Afr. J. Sci.* 69: 116 – 118.

VAN DER MERWE, M. 1978. Postnatal development and mother-infant relationships in the Natal clinging bat *Miniopterus schreibersi natalensis* (A. Smith 1834). *Proc. 4th Int. Bat Res. Conf.* (Eds.) OLembo, R.J., Castelino, J.B. & Mutere, F.A. Kenya National Academy for Advancement of Arts and Sciences. pp 309 – 322.

VAN DER MERWE, M. 1979. Foetal growth curves and seasonal breeding in the Natal clinging bat *Miniopterus schreibersi natalensis. S. Afr. J. Zool.* 14: 17 – 21.

VAN DER MERWE, M. 1980. Delayed implantation in the Natal clinging bat *Miniopterus schreibersii natalensis* (A. Smith 1834). *Proc. 5th Int. Bat Res. Conf.* (Eds.) Wilson, D.E. & Garner, A.L. Texas Tech Press. Lubbock, Texas, U.S.A. pp. 113 – 123.

VAUGHAN, T.A. 1978. Mammalogy, 2nd edn. W.B. Saunders, Philadelphia.

WIMSATT, W.A. 1969. Some interrelations of reproduction and hibernation in mammals. In: Dormancy and survival. (Ed.) Woolhouse, H.W. *Symp. Soc. exp. Biol.* 23: 511 – 549.

WIMSATT, W.A. 1975. Some comparative aspects of implantation. *Biol. Reprod.* 12: 1 – 40.

WINTERS, L.M., GREEN, W.W. & COMSTOCK, R.E. 1942. Prenatal development of the bovine. *Univ. Minn. Agr. Exp. Sta. Tech. Bull.* 151: 1 – 50.