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Development of the Chondrocranium of the Loggerhead Turtle, *Caretta caretta*

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ABSTRACT—To better understand the evolution and development of the amniote cranium, it is necessary to examine non-avian, non-mammalian embryos. Herein, development of the chondrocranium in the loggerhead turtle, *Caretta caretta*, is described based on whole-mount and sectioned specimens. Primitive characteristics were found to be established rather early in development; at these stages, the cranium resembled not only the early amniote chondrocrania, but also those of anamniote embryos. Several characteristics were noted in the late chondrocranium that represent shared, derived characteristics of chelonians. In particular, one amniote-specific characteristic, the tropibasic trabecula, appeared at an intermediate stage of development. In addition, developmental changes in the orbital cartilage were reemphasized, and the morphological significance of the crista sellaris was discussed in terms of the basic architecture of the vertebrate neurocranium.

INTRODUCTION

Avian and reptilian embryos exhibit a set of primitive features that are characteristic of amniotes but are missing in mammalian embryos. For example, birds and reptiles possess an extensively chondrified neurocranium composed of parachordal cartilage and broad orbital cartilage that directly surrounds the neural tube (de Beer, 1937; Kuratani, 1989). At early stages, this morphology resembles that of a shark embryo as well as the early mesenchymal condensation pattern seen in the mammalian cranium. Thus, reptiles may potentially serve as a useful model system for understanding the development and evolution of the amniote cranium. With that in mind, among the so called reptiles, chelonians are an advantageous subject for study, since they are believed to represent a less derived group, and their embryos are relatively easy to obtain (Yntema, 1966).

The morphology of the chondrocranium has been described in several chelonian species (*Emys*: Kunkel, 1911; *Dermochelys*: Nick, 1912; *Chelonia*: Fuchs, 1915; *Chrysemys*: Shaner, 1926; *Chelydra serpentina*: Rieppel, 1976; reviewed by Kamal and Bellairs, 1980). Nevertheless, there has been no description of the sequential, morphological changes in the developing skull, as have been reported for a number of other reptilian embryos by de Beer (1930) and El-Toubi and Kamal (1961a, b). It will be necessary, therefore, to characterize the morphology of the chelonian chondrocranium through a wide range of its development.

In previous works, the author described and discussed

the development of the orbital cartilage in the neurocranium of the loggerhead turtle, *Caretta caretta* (Kuratani, 1987, 1989). It was pointed out that the neurocranium of amniotes is extensively modified in association with brain development. The aim of the present study was to describe the sequence of developmental stages of the whole chondrocranium, including the viscerocranial elements, and to identify its primitive characteristics and to discuss the general morphology of the amniote cranium.

MATERIALS AND METHODS

Animals

The eggs of *C. caretta* were obtained at Shirahama, Wakayama, Japan in 1984. They had been laid at two locations on different days, so the specimens used in this study are from two separate groups of eggs. The eggs were brought to the laboratory and incubated at 36°C in moist sand. Selected eggs were excised daily. Perhaps due to varia-

Table 1. Embryo staging employed for observations of chondrocrania

Stage	Ser. No.	Car. L. (mm)	Car. W. (mm)
I	B-16	6.8	2.9
II	B-17	7.1	3.1
III	B-18	8.9	3.9
IV	A-28 B-19	9.0–9.2	4.5–4.8
V	A-29, 30 B-20	9.2–9.7	5.3–5.6
VI	A-32 B-23	11.6–12.6	7.5–8.5
VII	A-34 B-26, 27	13.1–14.1	8.9–9.9
VIII	A-36, 38, 40 B-32, 33	16.6–? 23.4–?	12.6–? 18.3–?

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tion in temperature at the original laying site, on any given incubation day, the size of the embryos varied between the two groups. The developmental stages were determined according to carapace size (Table 1), which is slightly different from the staging employed in previous works (Kuratani, 1987, 1989).

Whole-mount transparent specimens

The embryos were fixed with either 95% alcohol or 10% formalin, then skinned and eviscerated in preparation for staining carried out according to the method of Dingerkus and Uhler (1977) with slight modification. Initially the cartilage was stained with alcian blue, which reacts with the mucopolysaccharides. The embryos were then digested in a trypsin solution, and the bone was stained with alizarin red dissolved in 0.5% NaOH. Specimens thus stained were transferred through a graded series of glycerin/distilled water mixtures (30%, 60%) and finally stored and observed in 80% glycerin/distilled water.

Sectioned specimens

Specimens fixed in Bouin's fixative solution were dehydrated in a graded series of ethanol/water mixtures up to a final 99% ethanol mixture, and then transferred into 3x methylene benzoate containing 1% zelloidin where they were kept until they sank. The specimens were then transferred through two sequential, 20 min benzene washes, after which they were embedded in paraffin and cut into 10–15 µm thickness sections. After removing the paraffin, the sections were stained with hematoxylin and eosin or with Azan. When necessary, graphic reconstruction models were prepared using glass plates.

Abbreviations

an, angular; btr, basitrabecular process; can, cupola anterior; cbri, cornu branchiale I; cbrii, cornu branchiale II; ce, cartilago ectochoanalis; ch, cartilago hypochiasmatica; chy, cornu hyale; cohy, corpus hyoideus; col, columella auris; cpf, commissura prefacialis; cps, paraseptal cartilage; cs, crista sellaris; csph, commissura sphenothmoidalis; d, dentary; dca, distal portion of columella auris; del, ductus endolymphaticus; en, external nostril; epbri, epibranchial I; fap, foramen apicale; fb, fenestra basalis; fbp, fenestra basicranialis posterior; feph, foramen epiphaniale; feh, fenestra hypophysialis; fj, foramen jugulare; fh, foramen hypoglossus; fl, forebrain lamina; fm, foramen magnum; fnt, fenestra metoptica; foc, foramen for oculomotor nerve; fol, fenestra olfactoria; fpe, foramen perilymphaticum; fpp, foramen prepalatinum; fr, frontal; ios, interorbital septum; ip, incisura prootica; max, maxillary; mc, Meckel's cartilage; na, neural arches; nc, notochord; occ, occipital arch; of, primary optic foramen; ot, auditory capsule; pa, pila antotica; par, parietal; pas, processus ascendens of the palatoquadrate; pc, parachordal cartilage; pla, planum antorbitale; plp, processus lateralis posterior; pm, pila metoptica; pmp, processus maxillaris posterior; pn, paries nasi; po, orbital cartilage; pof, postfrontal; pol, polar cartilage; pp, processus pterygoideus; ppa, processus palatinus anterior; pq, palatoquadrate cartilage; prf, prefrontal; prl, processus lingualis; prt, processus retroarticularis of Meckel's cartilage; psn, processus supranarianus; pss, planum suprasedale; q, quadrate; rpo, remnant dorsal edge of the orbital cartilage; sa, surangular; sin, subiculum infundibuli; sof, secondary optic foramen; sq, squamosal; st, supratrabecular cartilage; tc, trabecula communis; tec, tectorial cartilage; tm, taenia marginalis; tn, tectum nasi; tr, trabecula cranii; tsyn, tectum synoticum;

RESULTS

Stage I

At this stage, the neurocranium of *C. caretta* consists of the parachordal and orbital cartilages and the trabecula cranii (Fig. 2a). The parachordal cartilage is seen as a pair of cartilages lateral to the notochord and fused with the ventro-

lateral wall of the auditory capsule laterally. As found in Squamata (Kamal, 1973), the parachordal cartilage is located in a mesenchyme contiguous with the cervical vertebrae. This configuration resembles that in other reptilian chondrocrania at early stages [e.g., stage 2 of *Ptyodactylus hasselquistii* (El-Toubi and Kamal, 1961a,b) or stage 1 of *Acanthodactylus boskiana* (Kamal and Abdeen, 1972)]. The parachordal cartilage does not appear to possess any foramina for cranial nerves. The absence of the hypoglossal foramen is explained by the absence of the preoccipital arch at this stage (Goodrich, 1911, 1918).

Rostral to the parachordal cartilage is found a single transverse cartilage plate (Fig. 2a, b), which can be identified as the orbital cartilage from its position caudal to the extracranial passage of the oculomotor nerve. It is situated vertically within the future cranial cavity, beneath the cephalic flexure. Thus, its location corresponds to the acrochordal connective tissue in various vertebrate chondrocrania (Goodrich 1930; Kuratani, 1989). The lateral portion of this cartilage is not actually a plate but consists of two laminae which laterally delineate an extracranial space, the cavum epiptericum (Gaupp, 1902). The dorsolateral portion of the cartilage is pierced by a single foramen on each side, the oculomotor foramen, through which the oculomotor nerve passes into the above mentioned extracranial space. The lowest margin of the orbital cartilage is pierced in the middle by the notochord, creating a foramen (Fig. 2a).

The most anterior element of the neurocranium at this stage is the trabecula cranii (Fig. 2a, b). This cartilage also consists of a pair of rods which are separated from each other. Caudally, the trabecular cartilages are connected to the orbital cartilage by means of a rather independent cartilage nodule, the polar cartilage.

A few cartilage elements are found in association with the auditory capsule: the palatoquadrate cartilage, the columella auris, and Meckel's cartilage, all of which are visceral elements. The palatoquadrate cartilage has two components, the quadrate and the pterygoid process, which are connected to each other rostrocaudally. In addition, the pterygoid process itself possesses a dorsally oriented process, the processus ascendens. Caudal to the quadrate, the columella auris is situated such that its medial end abuts the lateral wall of the auditory capsule. The proximal end of Meckel's cartilage articulates with the quadrate, while its distal end is not yet fused with its counterpart.

Hypobranchial cartilages

The hypobranchial apparatus consists of a centrally located corpus hyoideus laterally associated with several independent paired cartilages, the cornu hyale and the first and second branchial corns (Fig. 3). At this stage, the cornu branchiale I is the longest of the corns, while the cornu hyale is still in a procartilagenous state.

Stage II

The parachordal and orbital cartilages remain separate,

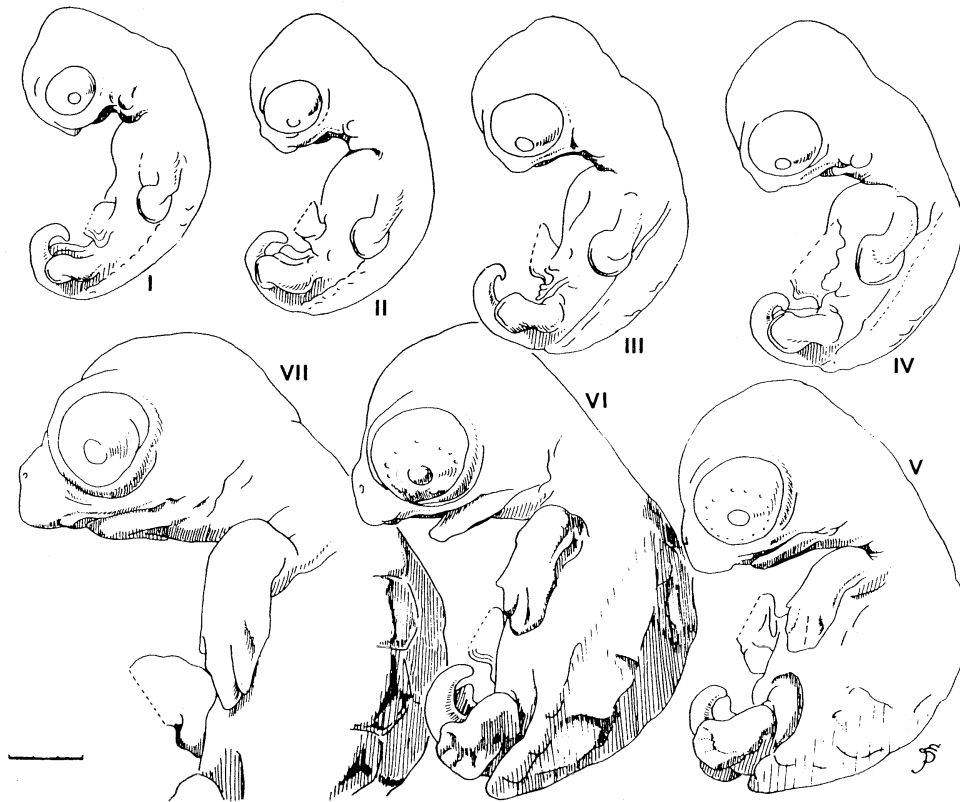
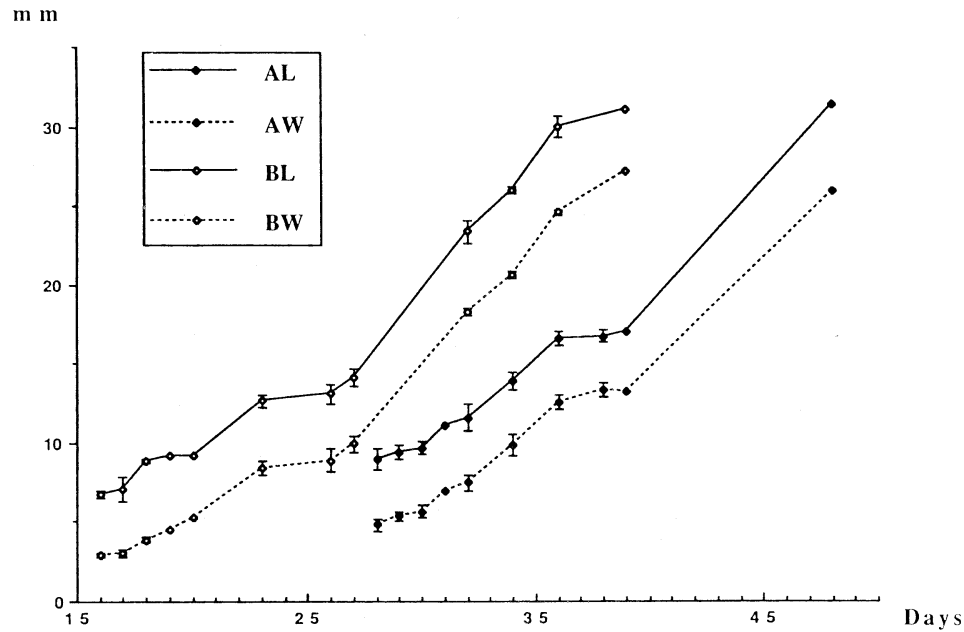


Fig. 1. Top: Correlation between carapace length and width and days of incubation. Solid circles depict the development of group A embryos taken from the south side of the peninsula; open circles denote group B embryos taken from the north side. Carapace lengths are indicated by the solid line, and the width by the broken line. Bottom: Appearances of developing embryos staged by carapace size. Stages are indicated by Roman numerals (I–VII). Bar = 3 mm.

although they are coupled by means of unchondrified connective tissue (Fig. 4a). Each of the parachordal cartilages is independent of its counterpart, leaving a space occupied by

the notochord. Caudal to the parachordal cartilage, the occipital is the first cartilage observed and thus forms an early basal plate primordium (Fig. 4). At this stage, the hypoglossal

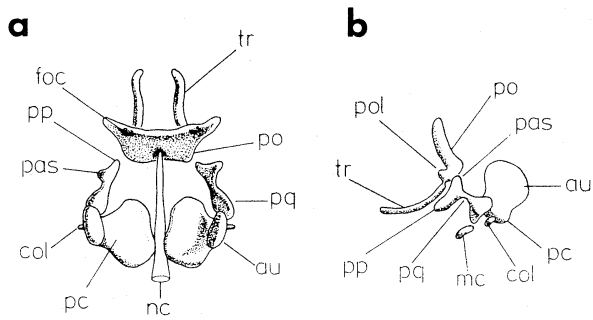


Fig. 2. Chondrocranium of *C. caretta* embryo at stage I. Dorsal (a) and left lateral (b) views are shown.

foramina are seen in the occipital cartilage for the first time; there are two on each side, which is consistent with later development (see below). Laterally, the parachordal cartilage is now secondarily separated from the auditory capsule (Figs. 4a, b), while ventrally, the parachordal cartilage has formed a ventrolateral projection at the level of the first hypoglossal foramen, delineating the occipital cartilage laterally. Longitudinally, the parachordal cartilage continues to develop, enlarging the diastema between the auditory capsule and the orbital cartilage, while rostrally, the parachordal and orbital cartilages are connected by means of a less chondrified connective tissue. The orbital cartilage possesses a ventrally pro-

jecting process on its anterior surface, indicating that the cartilaginous inner wall of the cavum epiptericum (see below) has expanded (Fig. 4b, c). This process is regarded as the earliest anlage of pila metoptica. The polar cartilage has also grown a process rostral to the hypophyseal vein that projects slightly dorsally back on itself.

The trabecular cartilages on either side have fused rostrally, and a vertical projection is now developing in the midline, forming the future interorbital septum (Fig. 4a). As a result of the trabecular cartilage fusion, a triangular space, the hypophyseal fenestra, is formed rostral to the orbital cartilage (Fig. 4). The trabecular cartilage now forms the trabecula communis and is a horizontal plate still showing a platybasic state (de Beer, 1937). The anterior portion of the trabecula communis possesses lateral projections beneath the forebrain, the forebrain lamina, which are the precursor of the planum suprasetale (Fig. 4a, c; Shaner, 1926).

Stage III

At this stage, extensive chondrification is under way in the neurocranium; the parachordal cartilage has fused with the orbital cartilage rostrally, leaving an elongated foramen, the posterior basicranial fenestra, within which is the rostral notochord (Fig. 5a, b). As a result, the foramen for the notochord has reappeared at the junction of the parachordal and orbital cartilages. The fusion of the occipital cartilage is

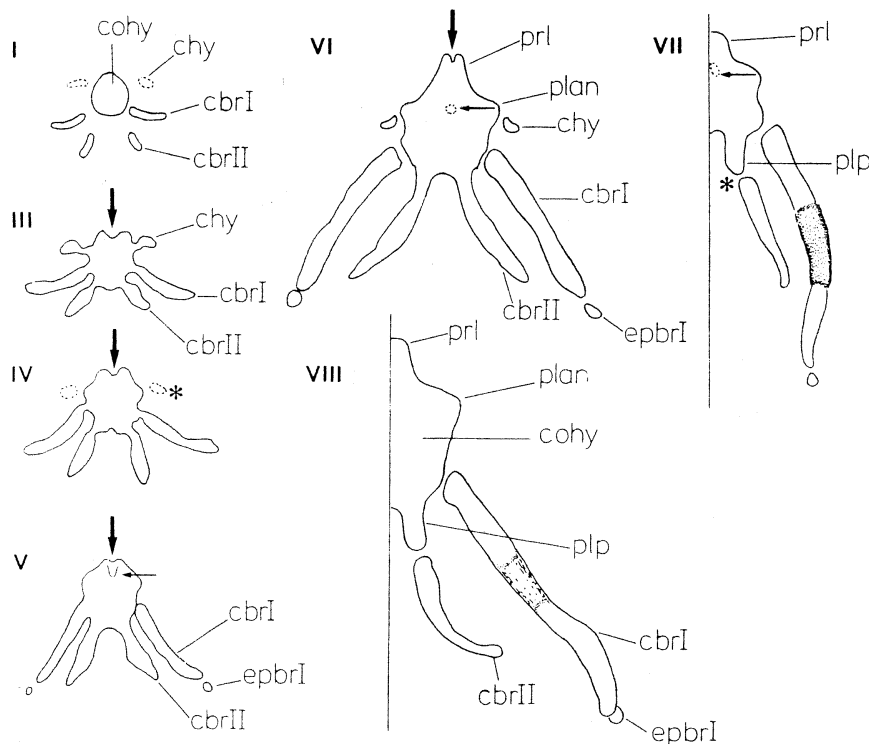


Fig. 3. Developing hypobranchial cartilages of *C. caretta* embryo at each developmental stage. Large arrows in stages III to VI indicate the medial indentation in the lingual process of the corpus hyale. The cornu hyale disappears by stage IV in many embryos (asterisk in the stage IV specimen). Small arrows in the hyoid corpse at stages VI and VII indicate the less chondrified area. Note the establishment of a secondary articulation between cornu branchiale II and the corpus at stage VII (asterisk).

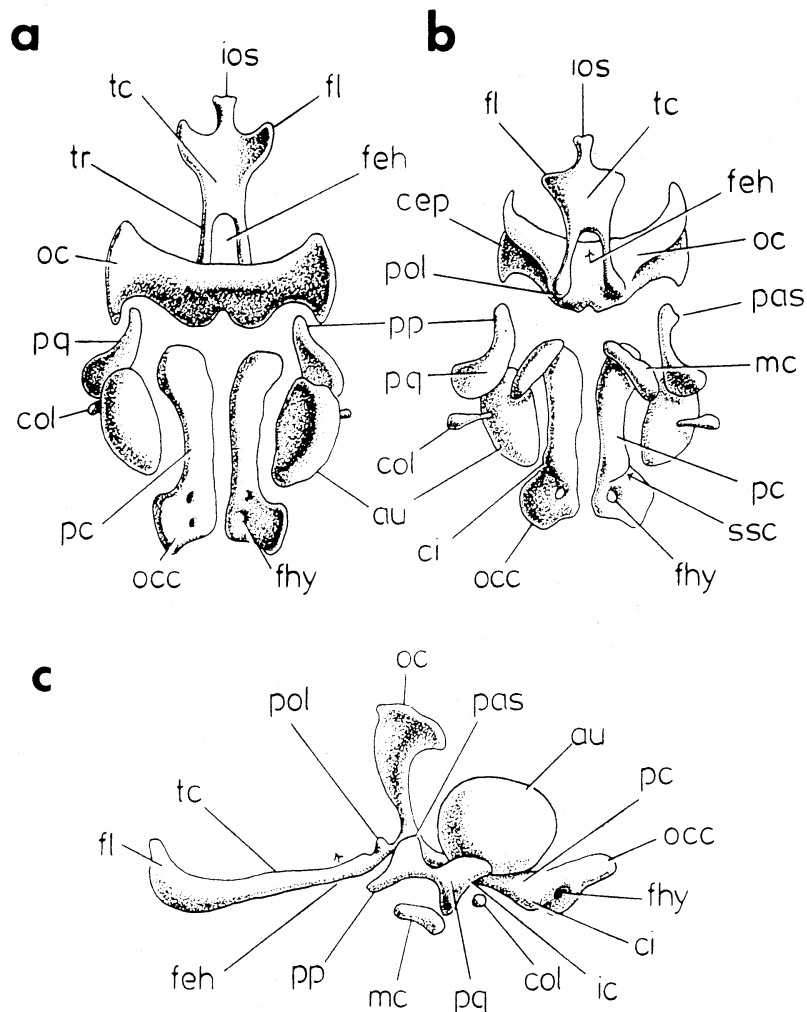


Fig. 4. Chondrocranium of *C. caretta* embryo at stage II. Dorsal (a), ventral (b) and left lateral (c) views are shown.

apparently filled with a fibrous tissue; still the diastema is not yet chondrified. Both the orbital and parachordal cartilages have developed laterally creating a notch, the incisura prootica, in which the trigeminal ganglion is located (Fig. 5a). The anterior process of the orbital cartilage seen at stage II, has now fused with the dorsal process of the polar cartilage, forming the early pila metoptica.

The trabecula communis has formed a lateral projection caudal to the interorbital septum anlage, the future planum suprasedale. The dorsal end of this process and that of the orbital cartilage are connected by a strip of cartilage, the taenia marginalis, forming a wide foramen on the lateral wall, the primitive optic foramen (Fig. 5a, b), which corresponds to what has been identified as the optic foramen in *Chrysemys* (Shaner, 1926). The dorsal end of the interorbital septum, on the other hand, has grown a pair of lateral projections, the future tectum nasi, which are the first anlage of the nasal capsule.

The auditory capsule is still separated from the parachordal cartilage medially (Fig. 5a). The columella auris has elongated laterally, and its end abuts an independent carti-

lage medially, forming the definitive ventral wall of the auditory capsule (cf. Stage V). The quadrate portion of the palatoquadrate has expanded dorsally to form an arch encircling the future middle ear cavity.

Hypobranchial cartilages

The corns are all attached to the corpus hyoideus at this stage (Fig. 3), and are all now cartilaginous. The anterior tip of the corpus hyale possesses a central indentation suggestive of the development of the future processus lingualis.

Stage IV

The parachordal cartilage has now grown two lateral processes, each projecting towards corresponding processes on the floor of the auditory capsule (Fig. 6a). The independent cartilage seen at the previous stage is now fused with the dorsolateral portion of the auditory wall to complete the floor of the capsule. The two pairs of processes found in the auditory capsule and the parachordal cartilage each represent precursors of a commissure between these cartilages. The more anterior of the parachordal cartilages is located rostral

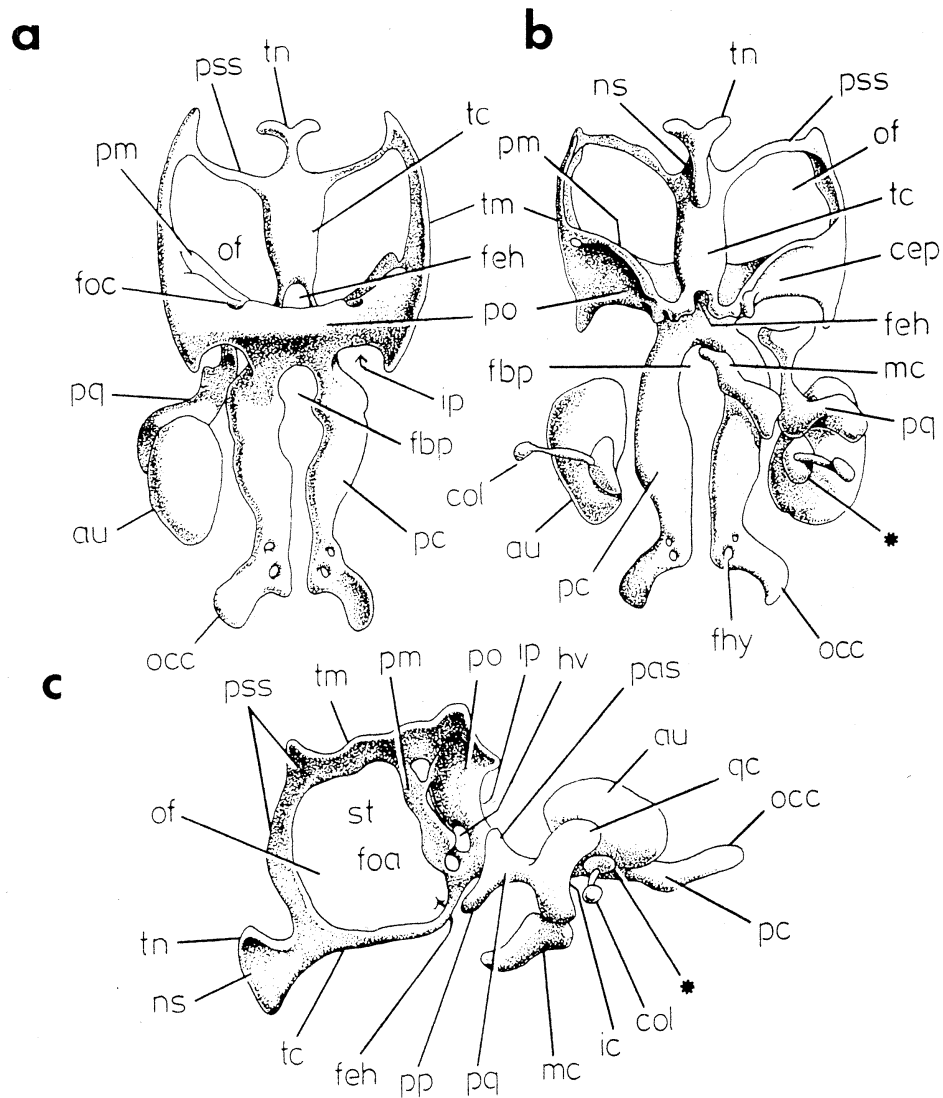


Fig. 5. Chondrocranium of *C. caretta* embryo at stage III. Dorsal (a), ventral (b) and left lateral (c) views are shown. Asterisks in (a) and (b) indicate the remnant cartilage associated with the medial end of the columella auris.

to the facial nerve outlet. Thus, unlike *Squamata* (Kamal, 1973a), the facial nerve passes through the intracapsular space in this animal. A similar passage of the facial nerve has been reported in *Chrysemys* (Shaner, 1926); however, the rostral process observed here does not represent the beginning of the prefacial commissure described by Shaner (1926), since it is formed by the floor of the auditory capsule. The true prefacial commissure lies above the cochlear capsule (Kamal, 1973a) and develops later in *C. caretta* (see below) and is homologous with the suprafacial commissure in mammals (de Beer, 1937). The caudal process of the auditory floor, on the other hand, has a foramen for the glossopharyngeal nerve (Fig. 6c).

Hypobranchial cartilages

The cornu hyale has now detached from the corpus and has degenerated. Its attachment to the corpus is suggested

by a process on the corpus (Fig. 3).

Stage V

The planum suprasedale has expanded to form a broad sheet of cartilage comprising the wall of the orbit (Fig. 7a) and has grown a rostral process, the early development of the sphenoethmoidal commissure. This process projects towards the posterior projection of the parietotectal cartilage of the nasal capsule. The pila metoptica is associated with the cartilage bar rostrally, which has grown over the trabecula cranii rostral to the ophthalmic artery (Fig. 7c).

No foramen for the notochord is observed in the orbital cartilage; at this stage, the greatest change in the orbital cartilage is the onset of absorption of its middle portion. As a result, the dorsal edge of this cartilage is now seen as a horizontal bar located in the cranial cavity (Fig. 7a). The homology between the orbital cartilage and the dorsum sellae in

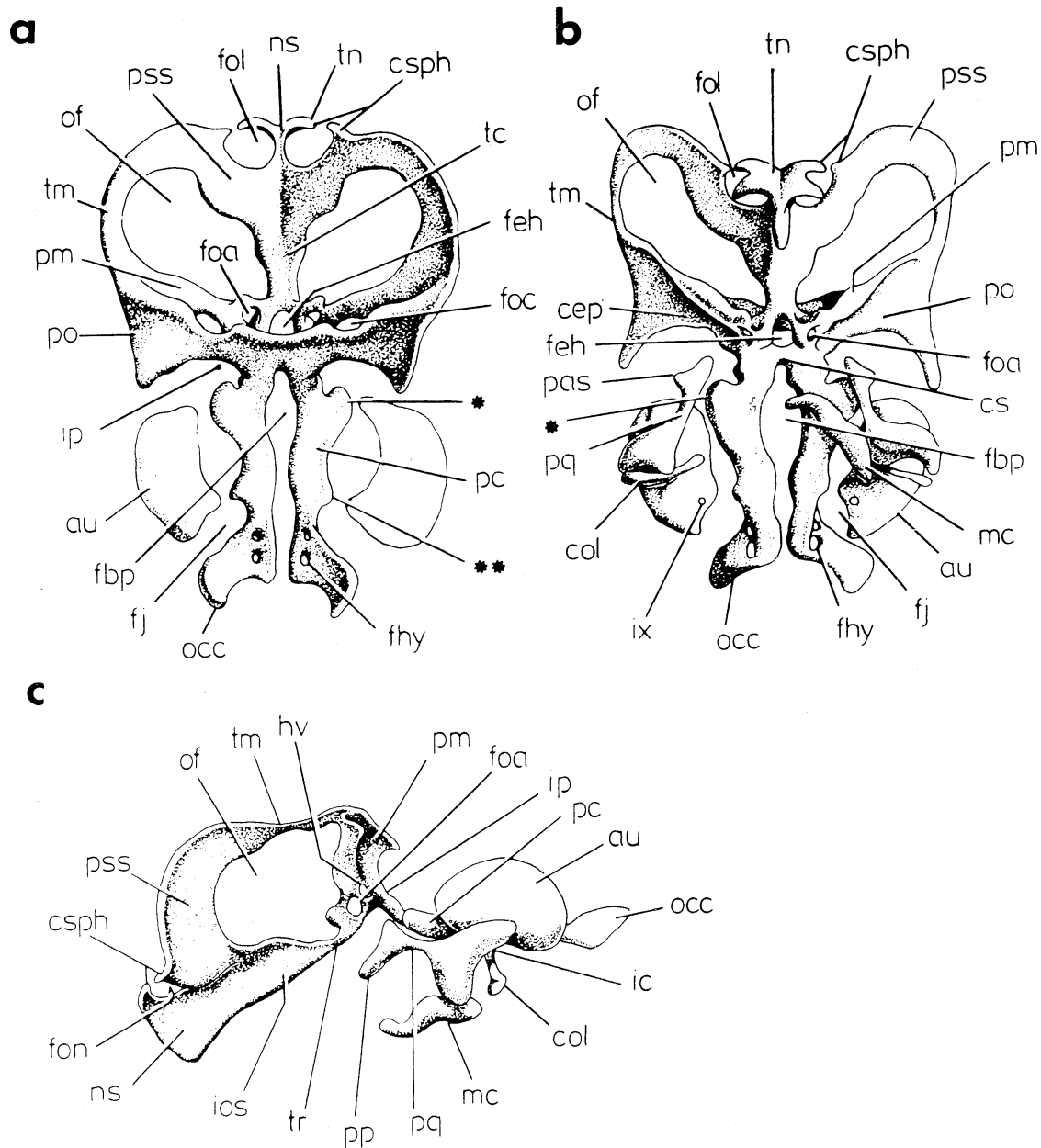


Fig. 6. Chondrocranium of *C. caretta* embryo at stage IV. Dorsal (a), ventral (b) and left lateral (c) views are shown. The single and double asterisks in a and b respectively indicate the development of the anterior and posterior cartilaginous bridges that connect the parachordal cartilage and auditory capsules.

mammals has already been discussed (Kuratani, 1989). The ventral edge of the orbital cartilage, on the other hand, now represents the crista sellaris (Fig. 7b). Thus, the orbital cartilage is not derived directly from the acrochordal cartilage itself, but rather originates from its ventral portion. From this stage onwards, the lateral edge of the orbital cartilage will be referred to as the pila antotica (Fig. 7b, c).

The floor of the auditory capsule is now connected to the anterior portion of the parachordal cartilage by two small commissures (Fig. 7a, b), the more anterior of which corresponds to what at the previous stage was referred to as the anterior process of the parachordal and the auditory capsule. The

inner wall of the auditory capsule contributes to the formation of this process, the definitive prefacial commissure (Fig. 7a). The posterior connection, the posterior margin of the facial nerve foramen, is formed exclusively by the floor of the auditory capsule. More caudally, another commissure between the two cartilages is now being formed. Both the inner wall and the floor of the auditory capsule seem to contribute to the latter commissure (Fig. 7a). The spaces left between the auditory capsule and the parachordal and orbital cartilages are referred to as basicapsular fissure and the jugular foramen, respectively. As illustrated in Fig. 7, the occipital cartilage in some specimens at this stage possesses more than two hy-

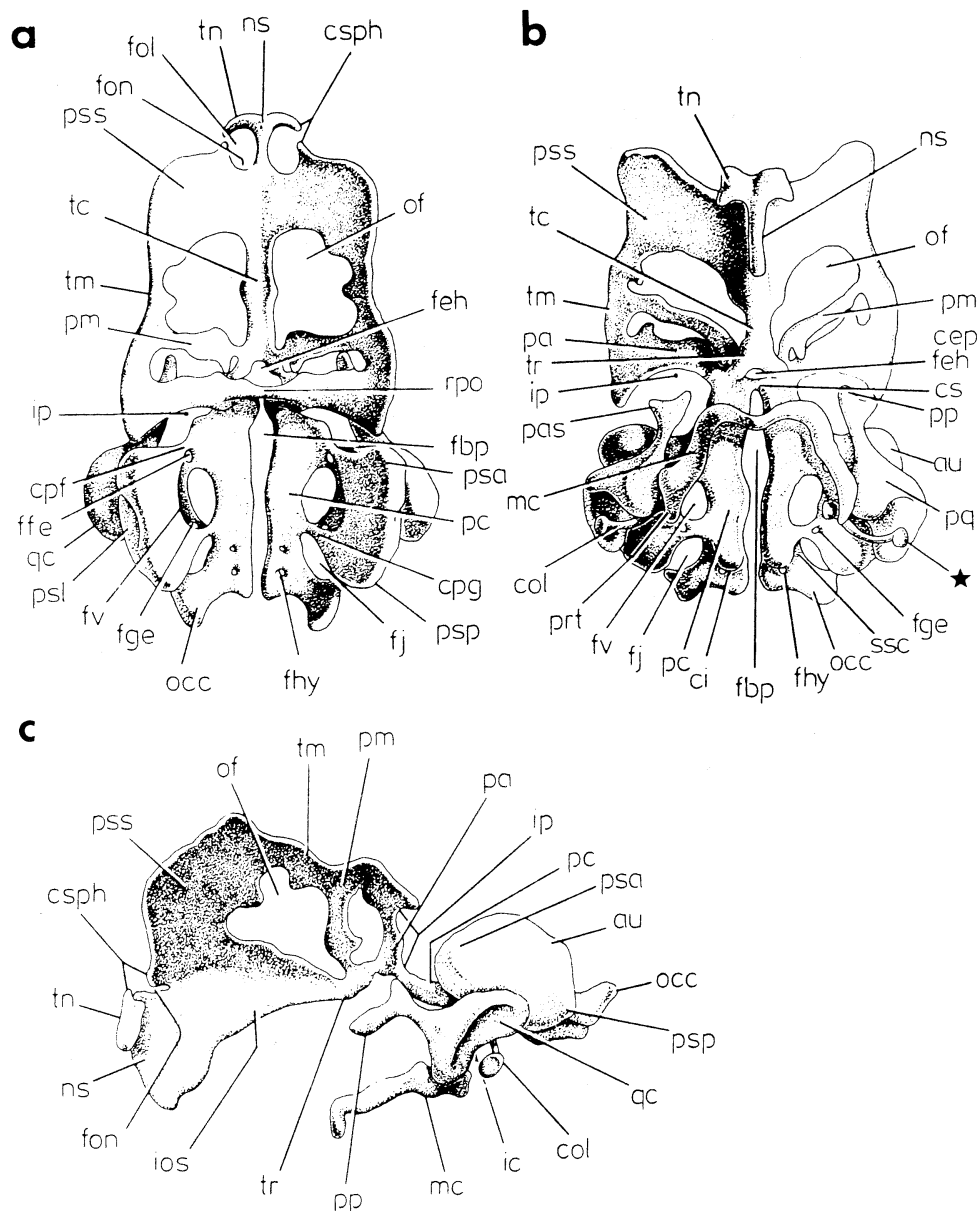


Fig. 7. Chondrocranium of *C. caretta* embryo at stage V. Dorsal (a), ventral (b) and left lateral (c) views are shown. The star in (b) indicates the earliest development of the extracolumella.

poglossal foramina, while in others there are only two.

Several morphological changes are observed in Meckel's cartilage. The distal end of the cartilage is now fused at the midline with its counterpart, forming a continuous single arch within the mandibular arch (Fig. 7c), while the caudal (or proximal) end has grown a caudally projecting process for attachment of the abductor muscle. It also possesses a slightly projecting dorsal process, the articular process, which articulates dorsally with the quadrate. Laterally, the columella auris is associated with an independent cartilage, the vestigial extracolumella (Fig. 7b, c).

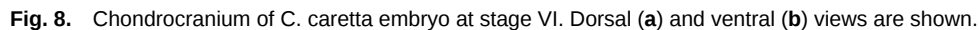
Hypobranchial cartilages

The cornu hyale has degenerated completely leaving no

trace of cartilage (Fig. 3). The process on the corpus, which will be referred to as the processus lateralis anterior, is present with the cornu hyale attached. The anterior tips on the corpus have fused to form the processus lingualis, and a less chondrified area is present in the midline of that process. The proximal end of the cornu branchiale I can be secondarily detached from the corpus hyale. On the distal tip of the branchial corn, on the other hand, there is a cartilage nodule, the epibranchial I.

Stage VI

In the nasal capsule, the paries nasi has grown a posterior projection, the processus palatinus anterior (Fig. 8b). This represents the articulation of the nasal capsule with the ante-



The chondrification of the auditory capsule is almost complete, and the fissura basicapsularis is separated from the jugular foramen. The inner wall of the auditory cartilage contributes to the connection between the parachordal cartilage and the auditory capsule, while the fissura basicapsularis now appears as a foramen in this inner wall for the passage of the

The median remnant of the orbital cartilage has disappeared by this stage (Fig. 10). Consequently, the new cranial cavity is formed by pilar cartilages, i.e. the pilae metopticae et antoticae and the planum suprasedale. Several changes are found in the nasal region: rostrally, the processus supranasalis has developed over the nasal septum (Fig. 10a); in the tectum nasi, the foramen epiphysiale is apparent (Fig.

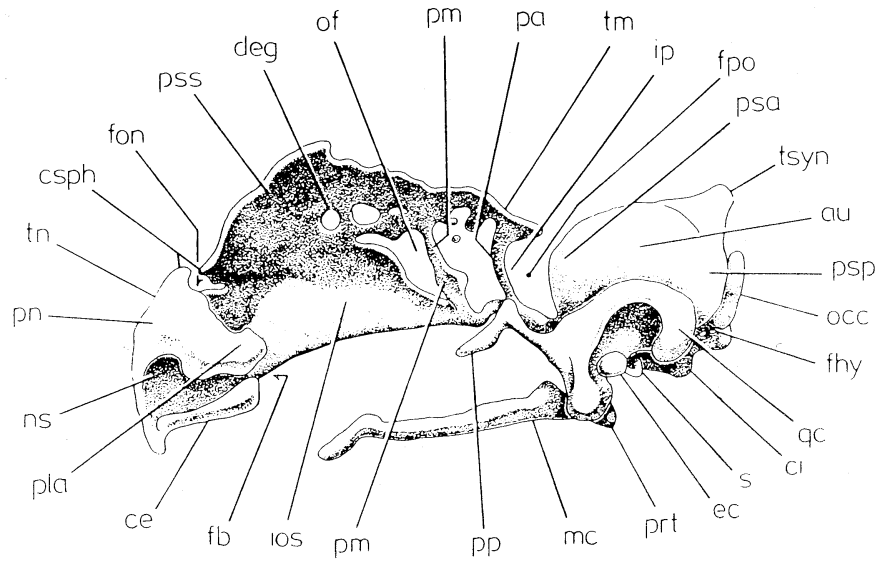


Fig. 9. Chondrocranium of *C. caretta* embryo at stage VI. Lateral view.

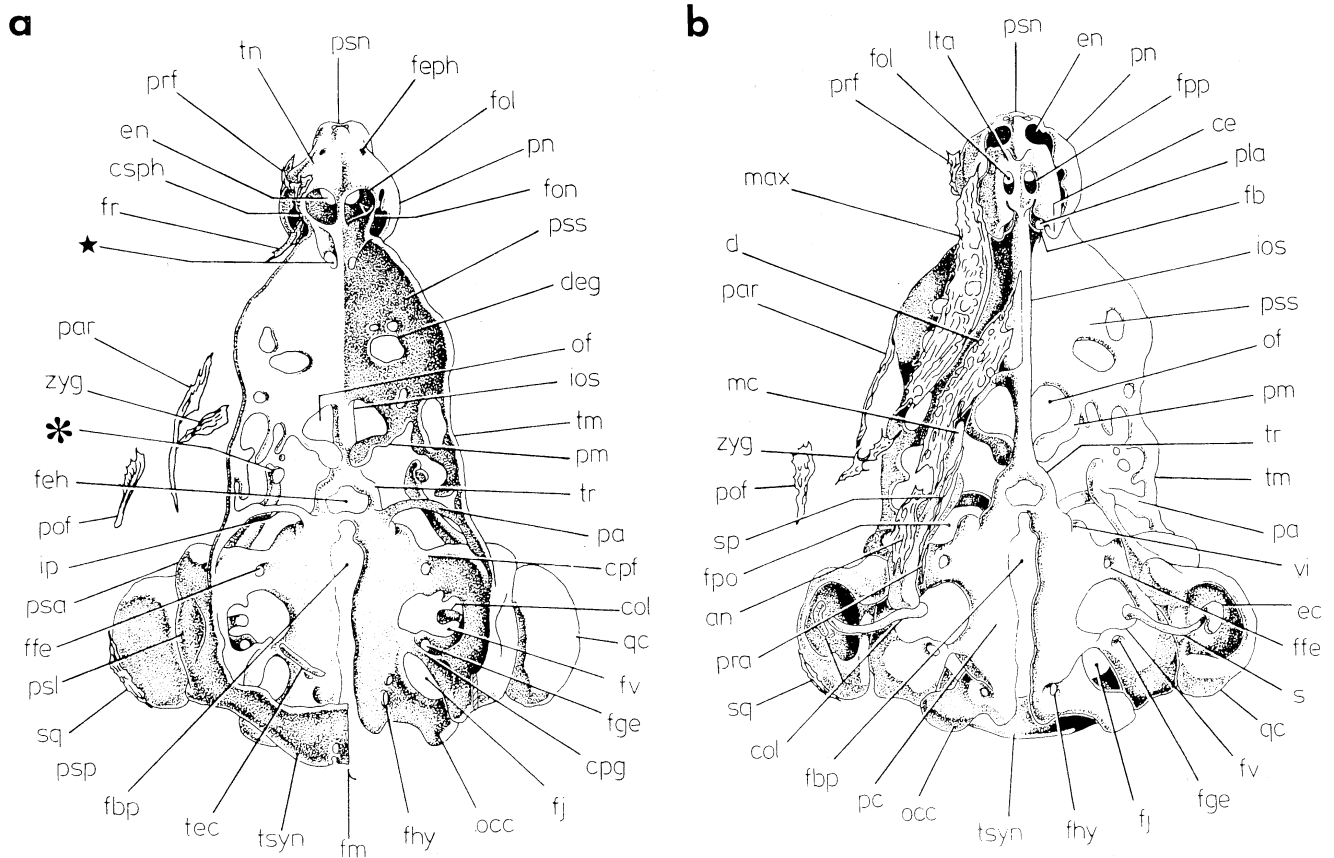
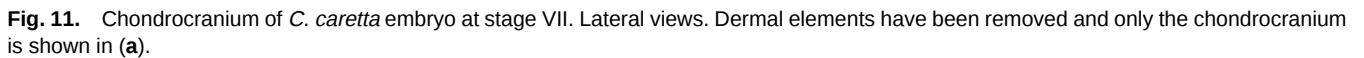


Fig. 10. Chondrocranium of *C. caretta* embryo at stage VII. Dorsal (a) and ventral (b) views are shown. The asterisk in (a) indicates a remnant chondrification in the mesenchymal lateral cranial wall, and the star indicates the secondarily established foramen that leads extracranially.

10a); and the floor of the nasal capsule is complete leaving the foramen prepalatinum (Fig. 10b).

The planum suprasedale has expanded caudally and has developed several foramina, suggesting that of the local absorption of the cartilage that began at the previous stage is

continuing (Fig. 10a, b). The ventromedial end of the pila metoptica, the root of the pila antotica, is no longer connected to the cranial base and ends freely over the central stem, medial to the abducens nerve outlet, which is seen as a notch on the basal plate (Figs. 10a, b, 11). A few cartilage nodules



The taenia marginalis is connected to the dorsal edge of the auditory capsule (Fig. 10a). The tectum synoticum is now complete and forms a cartilaginous brain case (Figs. 10a, 12). The median tectorial cartilage seen at the previous stage (Fig. 8a) is no longer present; it seems to have been incorporated into the tectum synoticum. In its place is another pair of tectorial cartilages rostral to the tectum synoticum (Fig. 12a). These cartilages can be fused with the tectum synoticum proper to

The basal plate is still incomplete, possessing an unchondrified portion in the midline. There is an independent single cartilage in the fenestra hypophysialis, medial to the internal carotid artery (Fig. 12a, b).

The morphology of the auditory capsule is little changed since the previous stage. But the quadrate has developed even more laterally to enclose the middle ear cavity (Figs. 10–12).

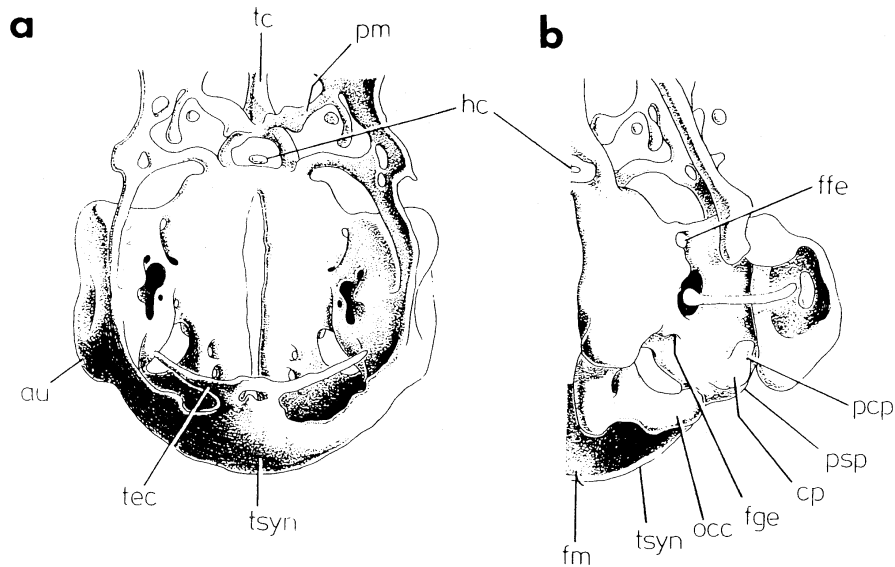


Fig. 12. Chondrocranium of *C. caretta* embryo at stage VII. Dorsal views of two different specimens. All dermal bones have been removed. Note in (a) that a pair of transverse cartilage bars are developing in connection with the tectum synoticum. Also note the late chondrification center of hypophyseal cartilage (hc) in the hypophyseal foramen.

Hypobranchial cartilages

As described above, the hypobranchial apparatus occurs in two configurations: those with the remnant cornu hyale and those without it (Fig. 3). Such a remnant cornu never fuses with the corpus. In either case, the structure of the hypobranchial apparatus is much the same as the previous stage: the cornu branchiale I forms a secondary articulation with the corpus hyale, and the cornu branchiale II is fused with the corpus. The epibranchial cartilage I is also present as in the previous stage. There is an indentation at the proximal one third of the cornu branchiale II, suggesting the future segregation of the processus lateralis posterior (see below).

Stage VIII

In the nasal capsule at this stage, the anterior palatine process is attached to the lateral wall of the capsule (Fig. 15b). This represents the anterior portion of the palatoquadrate cartilage. The pila metoptica has fused with the posterior projection of the interorbital septum, the cartilago hypochiasmatica, by means of an anterior process (Figs. 13, 14) thereby forming the secondary optic foramen. In the cranial base, the basi-cranial fenestra is still present. This indicates chondrification of the basal plate is still incomplete, which is in striking contrast to the situation in the lizard, where the parachordal cartilage is fused and widely chondrified from the very beginning of its appearance (El-Toubi and Kamal, 1961a).

The quadrate portion of the palatoquadrate cartilage has enlarged substantially, thereby forming the middle ear cavity, and is now much larger than the pterygoid process, even though the pterygoid process now has a cartilage nodule attached at its distal tip (Fig. 15b). Until this stage, no major changes were observed in the stapes, and no cartilaginous process akin to the interhyal process found in *Chrysemys* by Shaner (1926) has appeared.

The tectum synoticum is complete as a wide sheet of cartilage. A single, median tectorial cartilage is found attached to the tectum, which is much broader than was found at the previous stage.

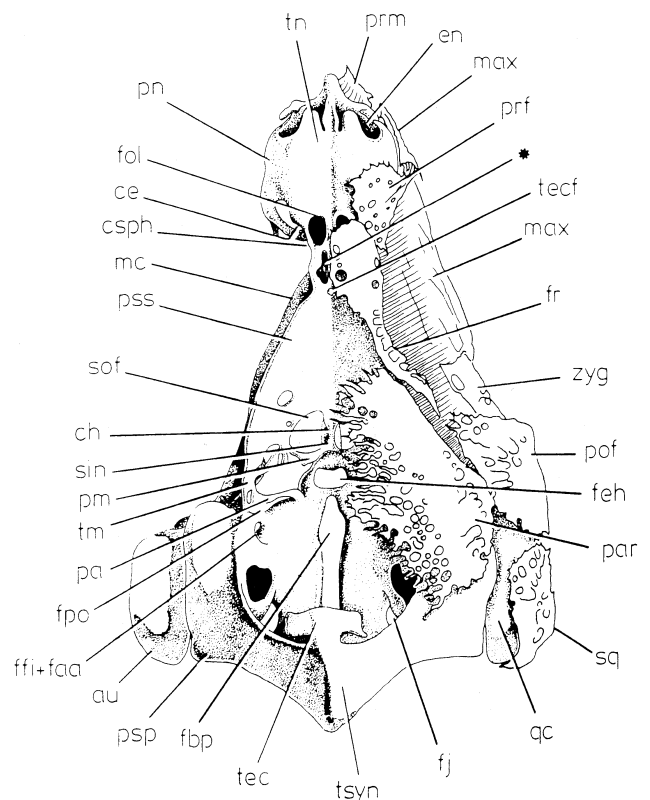


Fig. 13. Chondrocranium of *C. caretta* embryo at stage VIII. Dorsal view. Dermal bones have been removed on the left side. The asterisk indicates the same foramen shown by the asterisk in Fig. 10a.

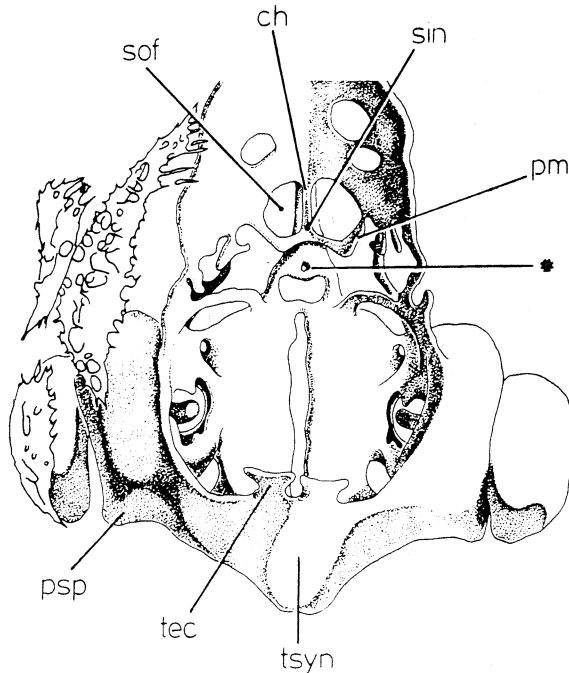


Fig. 14. Chondrocranium of *C. caretta* embryo at stage VIII. Dorsal view of the caudal half of the cranium. Dermal bones have been removed on the right side. Secondary absorption of cartilage has resulted in the development of a new foramen in the central stem (asterisk).

Hypobranchial cartilages

The corpus hyale possesses three sets of prominent processes: the processus lingualis and two lateral processes, the processus lateralis anterior and the processus lateralis posterior (Fig. 3). In contrast to the others, the processus lateralis posterior process seems to have been formed through establishment of the secondary articulation within the cornu branchiale II. The cornu branchiale I has elongated prominently as compared to the other elements, and endochondral ossification is under way in its middle portion.

DISCUSSION

Morphological characteristics of *C. caretta* chondrocranium

In the present study, the staged developmental morphology of the chondrocranium was described for embryos of *C. caretta*. This is the first precise, sequential description of the staged development of the Chelonian chondrocranium. We observed a number of developmental characteristics that are specific to *C. caretta* chondrocranium and are thus absent from virtually all other reptilian embryos (Kamal and Bellairs, 1980). These include (1) the long-term persistence of the basicranial fenestra; (2) approximately simultaneous development of the neuro- and viscerocrania; (3) exclusion of occipital cartilages from the formation of the tectum synoticum; (4) absence of the basiptyergoid process; (5) complete development of the sphenoethmoidal commissure; (6) participation of the secondary tectorial cartilages in the formation of the broad

tectum synoticum; (7) weak development of the processus ascendens; (8) lack of independent chondrification centers in the nasal capsule; and (9) the long-term persistence of the medial wall of the auditory capsule.

All of the characteristics listed above become apparent rather late in the development of *C. caretta* (Figs. 8 to 15). The early chondrocranium of *C. caretta*, like the chondrocrania of other amniotes, resembles the anamniote chondrocrania, especially those of elasmobranchs, which at early stages possess complete orbital and parachordal cartilages and paired trabecula (Figs. 2, 4, 5). The characteristics seen at these stages can be regarded as primitive, meaning that the basic architecture of the early cranium may be representative of the vertebrate cranial phylotype. The amniote-specific characteristics of the *C. caretta* cranium are particularly evident with respect to the establishment of the tropibasic trabecular cartilage, which later forms the narrow skull base and the interorbital- and nasal septa. Interestingly, this amniote morphotype becomes evident at intermediate stages in an apparent parallelism between ontogeny and phylogeny (Fig. 7). Of importance is the fact that primitive neurocranial morphology is dependent on the shape of the brain primordium; the cephalic flexure and brain vesicles determine the chondrification pattern of the mesenchyme (Thorogood, 1988; Kuratani, 1989). In this respect, the homology of some neurocranial elements, especially of those that develop rather late, should be reevaluated. One such element is the crista sellaris.

Development of the crista sellaris

The definitive crista sellaris in *C. caretta* is seen at stage VII; at that stage it is pierced by the anterior tip of the notochord. Before then, the crista sellaris is seen as the ventral edge of the degenerating orbital cartilage; thus the crista and the orbital cartilage cannot be observed simultaneously. At stage VI, the precursor of the crista sellaris is not located in the same horizontal plane as the trabecular cartilage. The configuration found after stage VII is caused by the subsequent secondary straightening of the cranial base.

If one follows the passage of the notochord rostrally, the crista sellaris appears as the anterior end of the parachordal cartilage. Careful examination at the earliest stages reveals that this is not the case, however. Topographically, at stage I the crista sellaris is in the same axial plane as the orbital cartilage, while the anterior end of the earliest parachordal cartilage does not extend to the orbital cartilage. In fact, at stage I the ventral portion of the orbital cartilage is penetrated by the anterior tip of the notochord just as the crista is at stage VII.

Although the names imply a homology, the crista sellaris is not homologous with the mammalian dorsum sellae. Instead the dorsal portion of the reptilian orbital cartilage is homologous with the crista (Kuratani, 1989). The dorsal edge of the orbital cartilage of *C. caretta* is seen within the cranial cavity at stage VI, when the definite crista sellaris is already beginning to form more ventrally at the level of the basicranium.

The crista sellaris was once identified as the cartilage

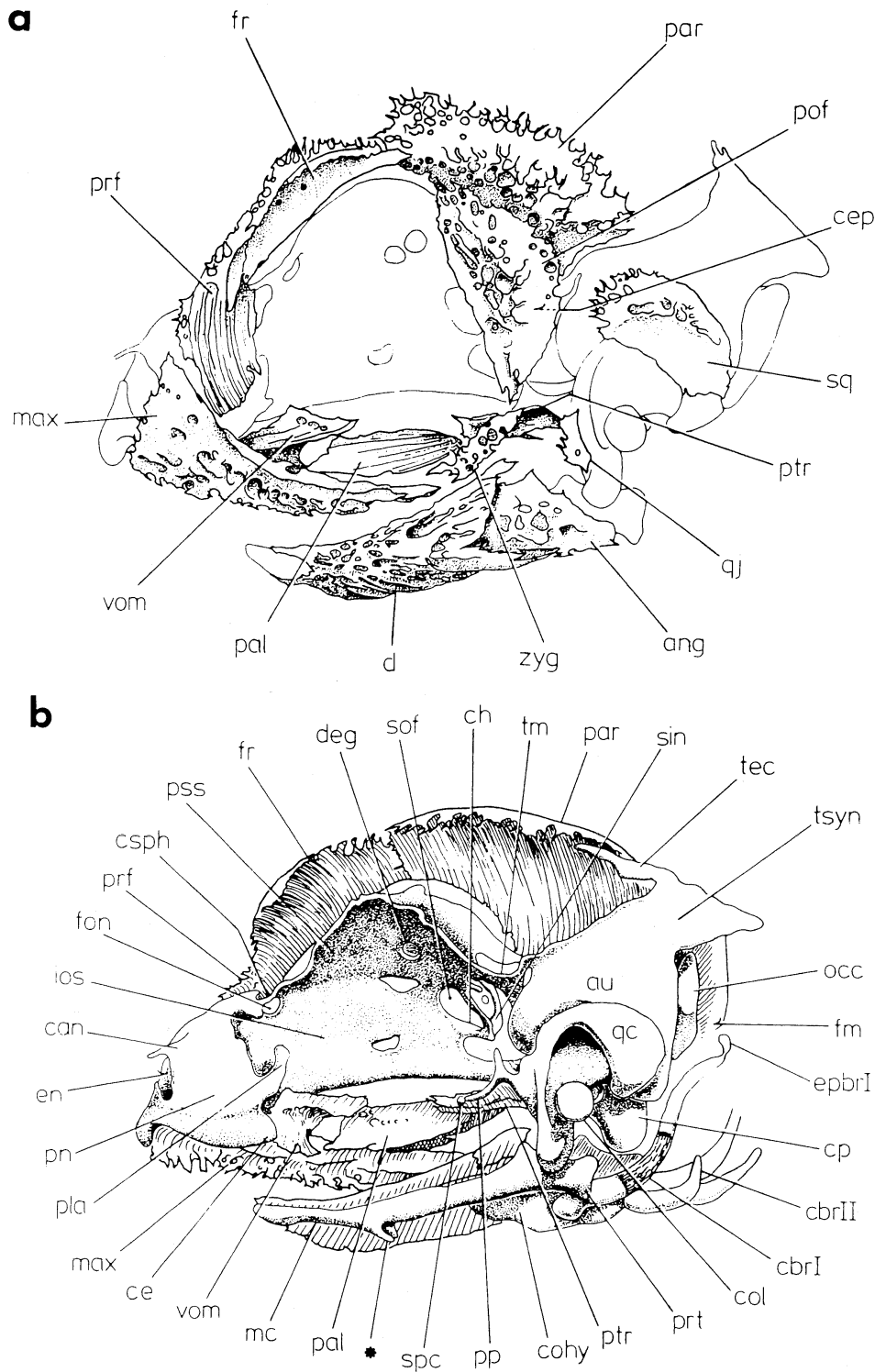


Fig. 15. Cranium of *C. caretta* embryo at stage VIII. Lateral views. Most of the dermal bones have been removed on the left side in (b). The asterisk indicates the ventrally projecting process of Meckel's cartilage.

lining the caudally-forming hypophysial fenestra (Gaupp, 1900). This caused a lot of confusion, especially with respect to young embryos still possessing the orbital cartilage. For example, the crista sellaris described in *Crocodylus* by Shiino (1914) actually represents the degenerating orbital cartilage.

From the above discussion, the term "crista sellaris" should be reserved for the structure formed secondarily as the caudal edge of the hypophysial foramen, not as a part of initial neurocranial cartilage.

General morphology of the neurocranium and its evolution

The present study reconfirmed that the main portion of the neurocranium consists of the parachordal and orbital cartilages and the trabecula. Topographically, since the rostral tip of the notochord penetrates the basal portion of the orbital cartilage, the trabecular cartilage of *C. caretta* obviously develops in the prechordal region, which is consistent with the notion that this cartilage is derived from the neural crest in the prechordal region (Couly *et al.*, 1993). Therefore, the mesoderm-derived true neurocranium (see Kuratani *et al.*, 1998) has to be sought caudal to the notochordal tip, i.e. at the orbital and parachordal cartilages.

Early on in a variety of vertebrates, the shape of the orbital cartilage exhibits a highly generalized morphology, i.e. a bending sheet of cartilage under the plica encephali ventralis. Due to topographical conservation, this cartilage is always penetrated by several cranial nerve roots. Such morphology is well described in chondrichthys (Goodrich, 1930) and even in monotremes (de Beer and Fell, 1937; reviewed by de Beer, 1937). In mammals, the orbital cartilage develops into dorsum sellae when it persists; whereas in reptiles, the main portion of the orbital cartilage is absorbed and the dorsum never persists. It is thus reasonable to assume that the chondrification of the orbital cartilage is a primitive condition, and degeneration of the cartilage may take place secondarily to various degrees in various lineages.

A homologue to the orbital cartilage has not been identified in the lamprey, the sister group of gnathostomes. Because lamprey embryos exhibit a cephalic flexure, and the rostral notochord develops at the same level as the flexure, it is possible that mesodermal cells confined ventral to the plica encephali ventralis chondrify to form the primary neurocranium. However, the only neurocranial component found in the lamprey is the parachordal cartilage located on either side of the notochord and continuing rostrally into the trabecular cartilage, whose neural crest origin has not been confirmed (Langille and Hall, 1988; reviewed by Smith and Hall, 1990). It is possible that lampreys, like bony fishes and amphibians, show secondary neurocranial degeneration due to the lack of sufficient space for mesoderm to chondrify around the neural tube.

In conclusion, the neurocrania of early amniote embryos exhibit a primitive state characterized by extensive areas of chondrification surrounding the brain primordium. *C. caretta* embryos, all of whose neurocranial elements chondrify, may therefore serve as another model system, in addition to chick, for experimental studies of cranial morphology.

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