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Development of the Suprarostral Plate of Pipoid Frogs

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ABSTRACT The rostral region of nonpipoid tadpoles has two sets of cartilages, the cornua trabeculae and the suprarostral cartilages, whereas the rostral region in pipoid larvae is occupied by a single and continuous cartilage, the suprarostral plate. The homology of this region in pipoid and nonpipoids tadpoles has been controversial. We examined the early formation and development of the suprarostral plate using serially cross-sectioned specimens of *Rhinophrynus*, *Xenopus*, and *Hymenochirus*. We conclude that the cartilaginous structures present in the rostral area of pipoid and nonpipoid larvae are homologous. Furthermore, we found two different developmental patterns among pipoid larvae. The chondrocranium of *Hymenochirus boettgeri* is described and illustrated to understand its developmental pattern and because of its uniqueness among pipoid chondrocrania. J. Morphol. 240:143–153, 1999.

KEY WORDS: Pipoidea; chondrocranium; *Hymenochirus*; suprarostral plate; homology

The chondrocranium of pipoid frogs is characterized by being broad and flat, by the precocious formation of the lower jaw, and by exhibiting a "high" suspensorium attachment condition. Furthermore, the rostral area—where in nonpipoid tadpoles we would find the cornua trabeculae and suprarostral cartilages—is occupied by a single, continuous, cartilaginous plate in pipoids, the suprarostral plate (Sokol, '75; Trueb and Hanken, '92).

Previous developmental work showed that the rostral elements found in pipoid and nonpipoid larvae have a common origin in the cranial neural crest (Sadaghiani and Thiébaud, '87; Olsson and Hanken, '96; Reiss, '97). However, Roček and Veselý ('89) suggested that these "are not fully corresponding structures" (not homologous). Trueb and Hanken ('92) pointed out that Roček and Veselý's ('89) suggestion would imply a diphyletic origin of anurans. Consequently, these authors argued that the suprarostral plate of pipoid larvae is formed by either the fusion and simplification of the cornua trabeculae of other Anura, or that in pipoids the cornua trabeculae are missing and a continuous cartilaginous plate develops anteriorly from the planum internasale. However, no additional evidence was provided to support the homology of rostral structures in pipoid and nonpipoid larvae.

Complete available descriptions of the chondrocrania of pipoids include those of *Xenopus laevis* (Kotthaus, '33; de Beer, '37; Trueb and Hanken, '92) and *Rhinophrynus dorsalis* (Swart and de Sá, '99). Additional information on the chondrocrania of *R. dorsalis*, *X. tropicalis*, *Pipa carvalhoi*, and *Hymenochirus boettgeri* is found in Sokol ('75, '77).

Herein, we study the formation of the suprarostral plate in pipoid tadpoles and provide developmental evidence supporting the homology of the rostral structures of pipoid and nonpipoid larvae. In order to understand the two developmental patterns found within pipid larvae, and because of the uniqueness of its chondrocrania, we must first provide a complete description of the chondrocranial anatomy of *H. boettgeri*.

MATERIALS AND METHODS

Histological serial cross-sections were used along with whole-mounted cleared and double-stained specimens to study the development of the suprarostral plate. Large specimens were embedded in paraffin and

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serial cross-sectioned at 10 µm. Paraffin sections were stained with Milligan's trichrome for differentiation of bone, cartilage, and muscle (Presnell and Schreibman, '97). Small specimens (total length <3 mm) were embedded in glycol methacrylate plastic polymer (JB4+, Polysciences, Inc.), serial sectioned at 2 µm, and stained with Toluidine blue (Presnell and Schreibman, '97).

Photomicrographs were taken under light microscopy using 35 mm film (Kodak Technical Pan developed in HC110). Chondrocranial descriptions are based on cleared and double-stained specimens complemented with observations of cross-sectioned specimens. Clearing and differential staining for cartilage and bone follows the method of Dingerkus and Uhler ('77). Drawings were obtained with the aid of a Wild M3C stereomicroscope fitted with a camera lucida attachment. Chondrocranial terminology follows that of de Beer ('37), de Jongh ('68), Sokol ('75), and Trueb and Hanken ('92).

Rhinophrymus dorsalis larvae used in this study are deposited at the National Museum of Natural History, Smithsonian Institution. They were collected at two localities: Guanacaste, Costa Rica (USNM 515945–515958) and Aguacate Lagoon, Cayo District, Belize (USNM 515959–515974). *Xenopus laevis* larvae were obtained from hormone-induced breeding of adults obtained from *Xenopus* I (Ann Arbor, MI). *Hymenochirus boettgeri* adults and larvae were obtained from the pet trade (Blue Lobster Farms, Madeira, CA). Additional larvae were obtained by natural breeding (i.e., not hormone-induced) in the laboratory. Collection numbers, measurements, and stages of material examined are given in the Appendix.

Neiuwkoop and Faber's normal table for *Xenopus* was used to stage all larvae (Neiuwkoop and Faber, '56). The similarity of external morphology of *Xenopus* and other pipoids during development makes this table preferable to Gosner's ('60) staging table. *Hymenochirus boettgeri* follows the developmental sequence of *X. laevis* up to approximately the onset of pigmentation (Neiuwkoop and Faber's Stages 38 through 40). Development of *Hymenochirus* is correlated with the staging table for *Xenopus* as closely as possible to allow for comparison with data already available for *X. laevis*. The stages of *Hymenochirus* between the completion of the retina (Stage 38) and the emergence of the hind limb bud (Stage 48) can be judged based on a few external changes, including increase in pigmentation, degeneration of the cement gland, and orientation of the mouth. Limb bud emergence is used as the main diagnostic feature for defining Stage 48. Thereafter, development of the hind limb, forelimb, and resorption of the tail are used as diagnostic staging features.

Specimens were anesthetized and killed using MS222 and fixed in 10% formalin. This procedure for euthanasia of specimens was reviewed and approved by the University of Richmond Animal Care and Use Committee (Protocol #96–3).

RESULTS

Chondrocranium of Hymenochirus boettgeri

This description of *Hymenochirus boettgeri* is based on the earliest cleared and double-stained larvae available, Stage 49. It is followed by notes on major developmental changes in chondrocranial structure through metamorphosis. A summary of the sequence of ossification is presented in Table 1.

Stage 49 (Fig. 1a,b,d)

Overall, the chondrocranium of *Hymenochirus* consists of a reduced number of robust, closely connected elements when compared to the chondrocrania of other pipoids. The lower jaw consists of a single, continuous, and inverted U-shaped mandibular cartilage. Meckel's and infrarostral cartilages are not identifiable as separate cartilages. The lateral elements of the lower jaw are

TABLE 1. Sequence of cranial ossification of Hymenochirus boettgeri

Stage	First appearance	Variation in appearance
49	Frontoparietal	St. 49–56
	Angulosplenial	St. 49–59
50	Exoccipital	St. 51–55
56	Parasphenoid	St. 57–60
61	Maxillae	St. 61–65
63	Nasal	St. 63–65
	Sphenethmoid	St. 63–64
65	Squamosal	St. 65
	Prootic	St. 65
	Dentary	St. 65
	Pterygoid	St. 65
	Tympanic annulus	St. 65–66
66	Septomaxilla	St. 66
	Premaxilla	St. 66
	Columella	St. 66
66+	Pars externa plectri	St. 66

Range of stages indicates the variation on first appearance of a given element in different specimens. 66+ indicates one month post-metamorphosis. Stages are Neiuwkoop and Faber ('56).

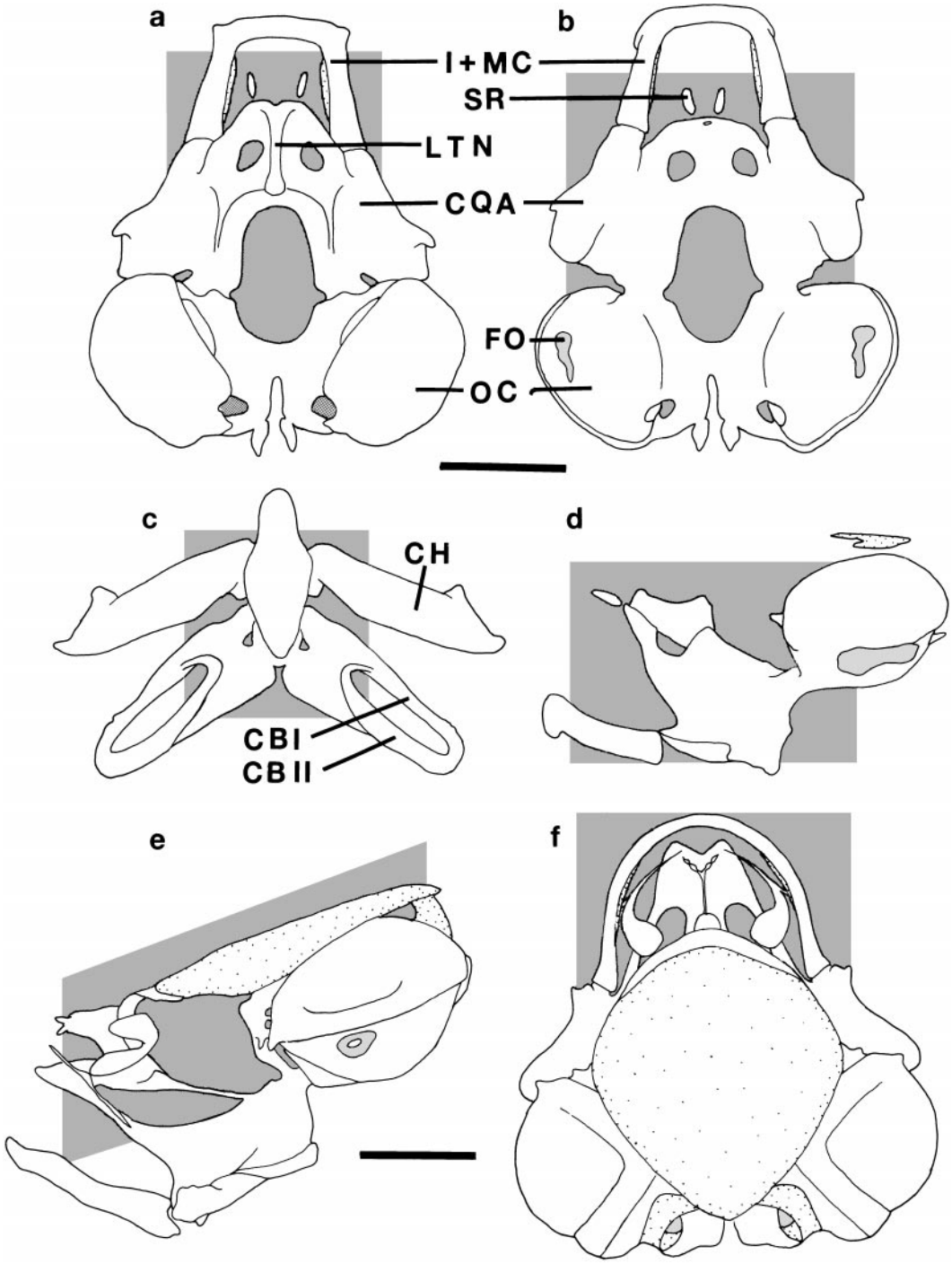

Fig. 1. Chondrocranial morphology of *Hymenochirus boettgeri* at Stages 49 (USNM 524286) and 60 (USNM 524311). **a–d**: Dorsal view, ventral view, branchial basket, and lateral view of *H. boettgeri*, Stage 49. **e–f**: Lateral and dorsal views of *H. boettgeri*, Stage 60. CBI-II, ceratobranchials I-II; CH, ceratohyal; CQA, commissura quadratocranialis anterior; FO, foramen ovalis; I + MC, infrarostrals + Meckel's cartilage; LTN, larval tectum nasi; OC, otic capsule. Bar = 1 mm.

circular in cross-section, while the anterior margin of the lower jaw is dorso–ventrally flattened.

The angulosplenials and frontoparietal have already begun ossification. The angulosplenials are present as splinters of bone developing over the ventrolateral lingual margin of the lower jaw. Overall, they are triangular, being broad posteriorly and narrow anteriorly. The frontoparietal develops as a single ossification located medial and dorsal to the otic capsules.

The suprarostrals are present as two rod-shaped cartilages projecting vertically from the anterior edge of the ethmoidal region. They are separate from each other and from other elements in the rostral region. They are embedded in noncartilaginous connective tissue.

The rostral region is divided medially by a prominent ridge, the larval septum nasi, which slopes sharply from the ethmoid plate forward to the anterior edge of the chondrocranium. Two large foramina, the foramina olfactoria, are located on the ethmoidal plate on either side of the larval septum nasi.

The processus muscularis comprises a small, lateral extension of the palatoquadrate. The pars articularis quadrati protrudes prominently at the anterior tip of the palatoquadrate and articulates with the posterior edge of Meckel's cartilage. The palatoquadrate attaches to the ventrolateral edge of the braincase only through a broad and well-developed commissura quadratocranialis anterior; no ascending or otic processes are present. The palatoquadrate does not contact the otic capsules. A subocular fenestra is also absent.

The floor of the braincase has a large, oval, basicranial fenestra. The orbital cartilages are poorly developed. The occipital condyles are present as two prominent, posterior projections of the basal plate on either side of the notochord.

The otic capsules represent approximately 40% of chondrocranial length and are fused to the chondrocranium along the entire length of their ventromedial edge. The tectum synoticum has not yet formed. The long axes of the otic capsules diverge (posteriorly to anteriorly) from the chondrocranial axis, forming an angle of approximately 40°. The ventral and lateral walls of the otic capsules are well developed and a large fenestra ovalis is present on the lateral wall.

The hyobranchial apparatus (Fig. 1c) consists of five independent elements. Lateral edges of the paired ceratohyalia articulate with the braincase. Medially, the ceratohyalia are connected to the single, medial copula. There are only two pairs of ceratobranchials (presumably I and II). Ceratobranchials I and II are fused proximally and distally forming an oval-shaped structure, which in turn is attached to the posterior edge of the single, medial copula.

Stage 53

Orbital cartilages are slightly more developed and the larval septum nasi is more robust. The foramina carotica primaria appear as lateral "notches" in the basal plate, whereas the basicranial fenestra remains open.

Stage 57

The lower jaw loses its vertical position and rotates forward, becoming horizontal to the body. At the same time, the suprarostrals also rotate from their original vertical orientation to a horizontal position.

Stage 58

A moderate amount of variation in ossification was observed at this stage. A small and ventrally projecting cartilage appears at the antero–lateral edge of the otic capsules. This could correspond to the traditional attachment of the processus anterolateralis of the crista parotica or the larval processus oticus. In addition, a small lateral process is present at the level of the pila antotica, corresponding to the remnant of the ascending process.

Stage 60 (Fig. 1e,f)

The rostral region undergoes drastic changes. The suprarostrals disappear. The adult septum nasi, tectum nasi, and lamina orbitonasalis are present in the rostral region. The tectum nasi appears as wings originating from the ethmoid plate and curving out and upward. The larval septum nasi previously present in the rostral region has eroded away and is being replaced by a newly formed adult septum nasi, which fuses to the lamina orbitonasalis.

The ceratohyals, ceratobranchials, copula, and hypobranchial plates are greatly eroded. The shape of the mandible shifts from its overall squared appearance of earlier stages to form a smooth arch. The commissura

quadratocranialis anterior erodes, leaving the palatoquadrate free along most of its length. At this stage the palatoquadrate begins to rotate and migrate posteriorly. This is accompanied by the elongation of the lower jaw. The columella is seen as a small and cylindrical cartilage projecting outwards from the fenestra ovalis.

Stage 62

The rostral region is greatly modified by the continuing development of the nasal capsules. The ceratobranchials and copula have nearly disappeared, whereas the ceratohyals remain and migrate posteriorly in association with the palatoquadrate.

Stage 63

The nasal bones begin ossification dorsal to the well-developed nasal capsules. An ossified sphenethmoid forms the antero–medial walls of the braincase. The palatoquadrate contacts the antero–medial wall of the otic capsules.

Stage 65

The ceratohyals fuse to the otic capsules posterior to the point of attachment of the palatoquadrate.

Development of the suprarostral plate
Rhinophrynus dorsalis

Serial cross-sections of specimens at Stage 45 show the ethmoid plate (anterior wall of the braincase) possessing two anteriorly projecting cartilaginous rods, the cornua trabeculae (Fig. 2a). Suprarostral cartilages are present and fused to the anterior tips of the cornua trabeculae. The cornua trabeculae are also seen fused to the suprarostral cartilages in whole-mounted, cleared and stained early tadpoles, Stages 48–51. Between Stages 48–51 a process, herein called the anterior process of the ethmoid plate (APE), grows anteriorly from the ethmoid plate between the cornua trabeculae. In cross-sections this process appears subcylindrical in shape. The APE is visible, and anteriorly fused to the suprarostral cartilages, in whole-mounted tadpoles at Stage 51.

During later development (after Stage 51), the space left between the APE and the cornua trabeculae "fills in" with cartilage, leaving two small anterior holes (behind what would correspond to the posterior edge of the suprarostrals). This cartilaginous "filling" forms a continuous plate with the APE, the cornua trabeculae, and the suprarostrals, forming the suprarostral plate of later larval stages. In cross-section, the suprarostral plate is thicker medially and laterally in the areas corresponding to the APE and the cornua trabeculae. Between these three thicker areas the suprarostral plate is seen as a thin sheet of cartilage (Fig. 2a).

Xenopus laevis

Serial cross-sections of Stage 41 specimens showed an ethmoid plate with two anteriorly projecting cornua trabeculae. The suprarostral cartilages are not yet formed. The same observations were made in specimens at early Stage 43 (Fig. 3). However, in late Stage 43 specimens, a shorter medial rod-shaped element projects from the ethmoid plate medial to the cornua trabeculae, herein identified as the anterior process of the ethmoid plate (APE). No suprarostral elements are visible at this stage but they form between Stages 43 and 45. In later development (after Stage 45), cartilage "fills-in" the area between the cornua trabeculae, APE, and the suprarostrals, forming the suprarostral plate.

Hymenochirus boettgeri

Cross-sections of the rostral region of early Stage 38 specimens showed a single medial cartilaginous rod-shaped element (the APE) projecting anteriorly from a poorly developed ethmoid plate (Fig. 4a–c). At this time, neither cornua trabeculae nor suprarostral cartilages are visible. Cross-sections of late Stage 38 specimens showed two short cartilaginous rod-shaped elements, the cornua trabeculae, developing from the ethmoid plate ventral to the APE (Fig. 4d–f). Suprarostral cartilages are not yet present.

During Stages 41 and 42, the cornua trabeculae grow anteriorly past the anterior tip of the APE. Suprarostral cartilages develop at Stage 41 and remain present until their resorption later during metamorphosis. Cross-sections of specimens at Stage 41 showed cornua trabeculae, suprarostral cartilages, and the APE (Fig. 5a–d).

Between Stages 45 and 48, cartilage "fills-in" the space between the cornua trabeculae and the distal tip of the APE, leaving a pair of small holes on either side of the medial element. From Stage 48 on, the cornua trabecula and the APE form a continuous cartilaginous plate. Furthermore, the APE grows dorsally, forming the larval septum nasi.

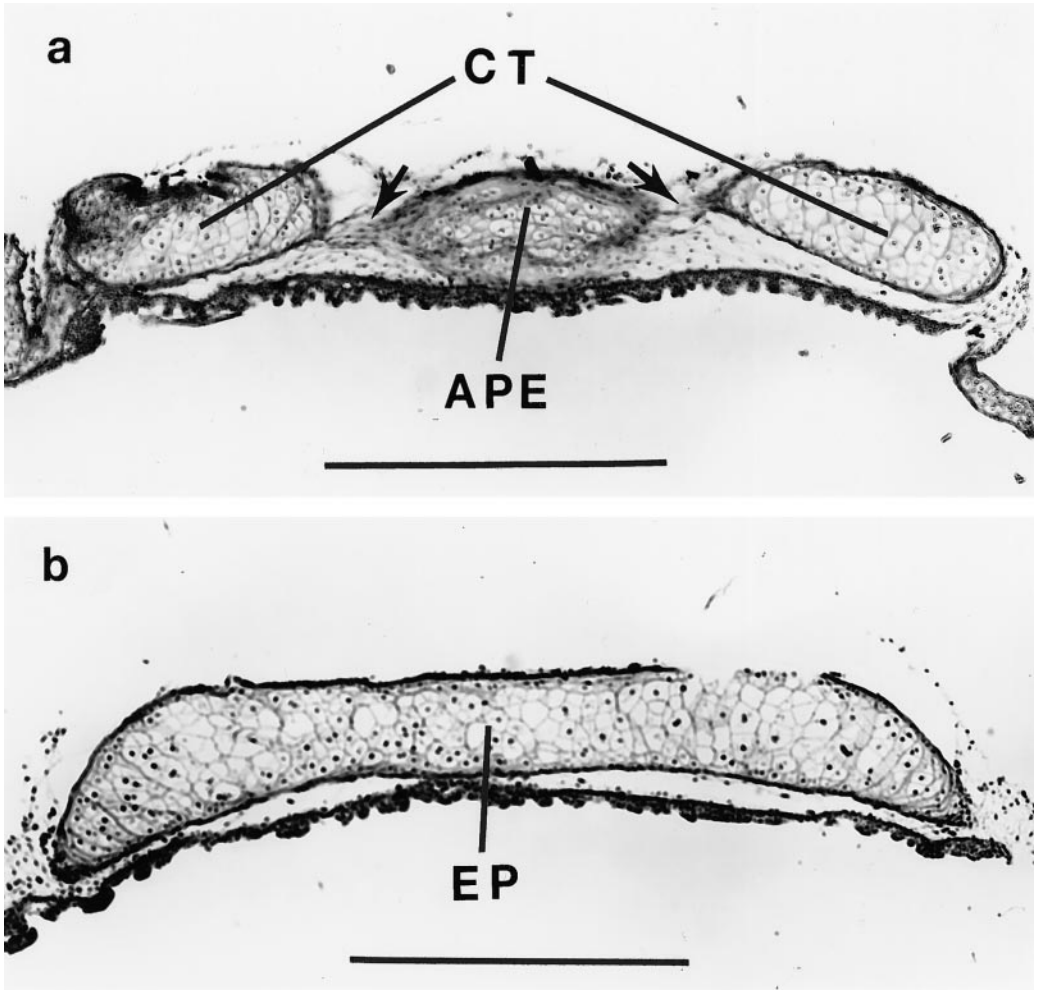

Fig. 2. Cross-sections through the suprarostral plate of *Rhinophrynus dorsalis*, Stage 51. **a**: Anterior region of the suprarostral plate. **b**: Section through the ethmoid plate. APE, anterior process of the ethmoid plate; CT, cornua trabeculae; EP, ethmoid plate. Arrows indicate thin cartilage connecting the APE and the CT and contributing to the suprarostral plate. Bar = 100 µm.

DISCUSSION

Chondrocranial morphology

There are at least five chondrocranial synapomorphies uniting pipoids: plate-like suprarostral element, broad commissura quadratocranialis anterior, reduced processus muscularis palatoquadrati, precocious formation of the lower jaw, and a well-developed larval crista parotica (Starrett, '73; Sokol '75, '77; Trueb and Hanken, '92; Swart and de Sá, '99). In addition, all pipoids except *Hymenochirus* share the following chondrocranial characteristics: presence of a lateral process of the palatoquadrate, alae of the suprarostral cartilages integrated into the suprarostral plate, presence of a muscular process of the otic capsule, slender subocular bar of the palatoquadrate, and a suspensorium with a high attachment to the braincase (Sokol '77, present study).

Hymenochirus larvae are unique because of their small size and carnivorous diet. Sokol ('77) suggested that the unique chondrocranial morphology of *Hymenochirus* was an adaptation to carnivory on small aquatic insects. Considering that *Hymenochirus* represents the smallest members among pipoids and that its tadpoles are also among the

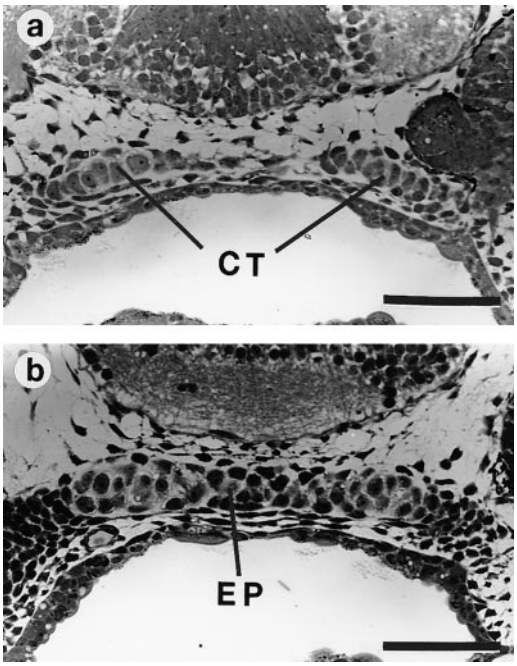

Fig. 3. Cross-sections through the rostral region of *Xenopus laevis*, Stage 43. **a:** Anteriorly projecting cornua trabeculae. **b**: Ethmoid plate. CT, cornua trabeculae; EP, ethmoid plate. Bar = 50 µm.

smallest anuran tadpoles, the highly divergent chondrocranial anatomy also may be the result of a miniaturization in this lineage. Previous studies of miniaturization among amphibians (Hanken, '83, '84; Trueb and Alberch, '85; Hanken and Wake, '93) have pointed to three traits characteristic of miniaturized taxa: reduction and structural simplification, morphological novelty, and increased variability. All three traits are observed in the chondrocranium of *Hymenochirus*. First, *Hymenochirus* has a reduced number of chondrocranial components (e.g., lack of otic and ascending processes, absence of a complete subocular bar of the palatoquadrate, loss of two ceratobranchials, etc.). Second, the absence of free suprarostral alae must be considered a plesiomorphic condition for pipoids (Roček and Veselý, '89; Trueb and Hanken, '92; Swart and de Sá, '99). Consequently, the two small and free cartilages present in the rostral region of *Hymenochirus* and previously considered to represent free suprarostral alae by Sokol ('77) represent a secondarily derived novelty in this lineage. Finally, the variation in the timing of appearance of cranial bones in *Hymenochirus* ranged from three to eleven stages for a given element (see Table 1). Variation in onset of ossification in anurans has been previously shown for other anurans (Kemp and Hoyt, '69; Hanken and Hall, '84; Trueb and Hanken, '92). Comparison across taxa is made difficult by the use of different staging tables; however, using the conversion table provided in Trueb and Hanken ('92), we can make some interesting observations. First, the highest variation in cranial ossification in *Bombina orientalis* corresponds to that of vomer and nasal bones, which appear over five Gosner stages (Hanken and Hall, '84). Second, cranial development is less variable in *Xenopus* when compared with other anurans (Trueb and Hanken, '92). The variation in cranial development observed for *Hymenochirus* exceeds that reported for all other anurans (extending over 11 Neiuwkoop and Faber stages or the equivalent of 13 Gosner stages). Furthermore, this variation differs from the previously reported conservatism in pipids, as represented by that of *X. laevis* (Trueb and Hanken, '92). However, this variation in cranial ossification resembles that reported for the miniaturized urodelan genus *Thorius* (Hanken, '84).

Development of the suprarostral plate

A suprarostral plate is characteristic of the chondrocranium of pipoids. Roček and Veselý ('89) argued that the suprarostral plate of pipoids and the cornua trabeculae of nonpipoid larvae are not homologous, i.e., they cannot be derived from one another. Subsequently, Trueb and Hanken ('92), while describing the suprarostral plate of *Xenopus laevis*, suggested that in pipoids either 1) the cornua trabeculae fail to develop and an anterior extension of the planum internasale develops in their place, or 2) the suprarostral plate corresponds to the "fusion and simplification" of the cornua trabeculae of nonpipoid tadpoles. Other scenarios could explain the formation of a suprarostral element, e.g., posterior expansion of the suprarostral cartilages to fuse with the ethmoid plate, posterior growth of the suprarostral cartilage with an anterior growth of the ethmoid plate, etc.

Serial cross-sections of early pipoid tadpoles showed that two slightly different patterns, involving the same cartilaginous elements, are present in pipoids. The first pattern is seen in *Xenopus* and *Rhinophry-*

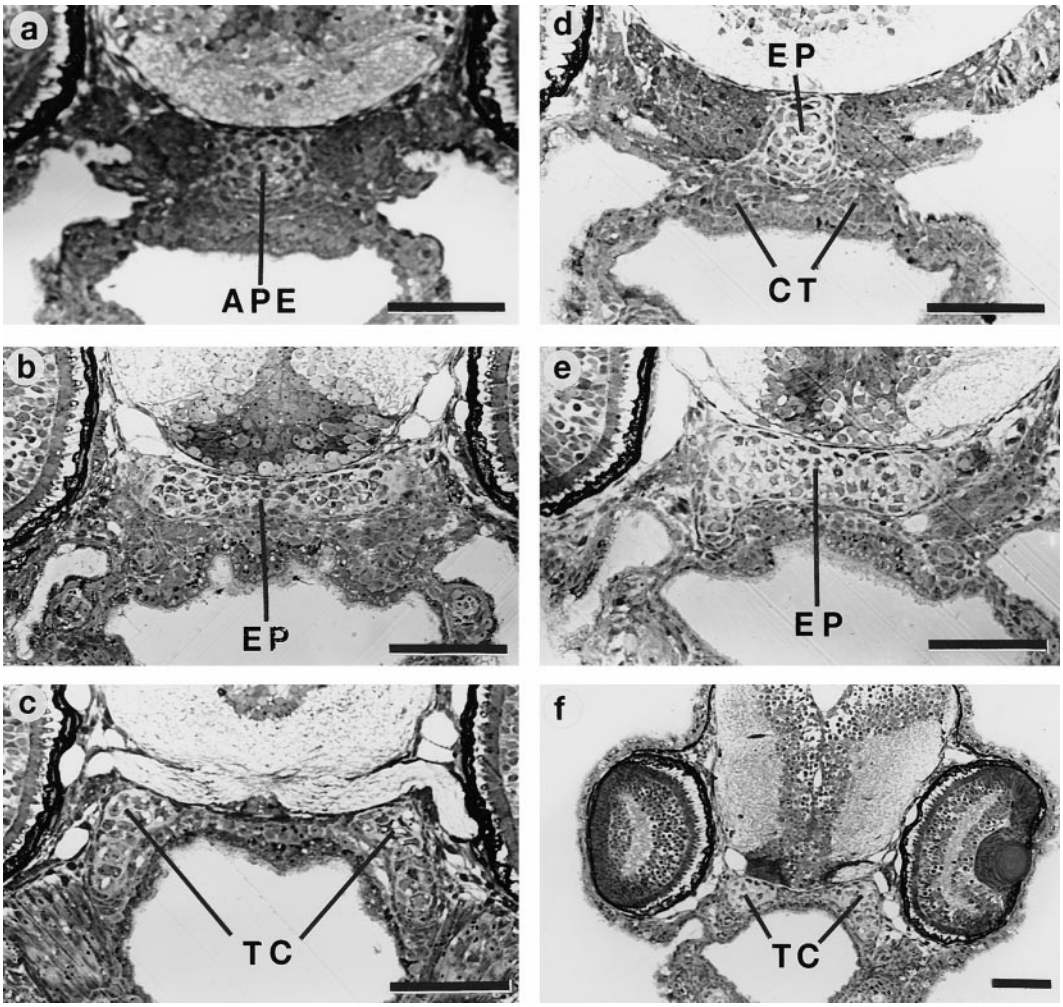

Fig. 4. Cross-sections through the rostral region of *H. boettgeri* at early Stage 38. **a**: Anterior process of the ethmoid plate. **b**: Section through the ethmoid plate. **c**: Section posterior to the ethmoid plate showing the trabecular cartilages. **d–f**: Sections through the rostral region at late Stage 38. **d**: Anterior section showing the anterior process of the ethmoid plate and the cornua trabeculae. **e**: Ethmoid plate. **f**: Section posterior to the ethmoid plate showing the trabecular cartilages. APE, anterior process of the ethmoid plate; CT, cornua trabeculae; EP, ethmoid plate; TC, trabecular cartilages. Bar = 50 µm.

nus. Initially, a pair of cartilaginous rods, herein identified as the cornua trabeculae, grow anteriorly from the ethmoid plate and fuse to the suprarostral cartilages. In addition, a third cartilaginous rod, herein called the anterior process of the ethmoid plate, also grows anteriorly from the ethmoid plate, between the cornua trabeculae, and fuses to the suprarostrals. Later, most of the space between these elements is occluded by cartilage forming the suprarostral plate.

Hymenochirus showed a slightly different pattern of development. First, the anterior process of the ethmoid plate grows anteriorly from the ethmoid plate. Subsequently, the cornua trabeculae also develop anteriorly from the ethmoid plate, but they form ventral to the anterior process of the ethmoid plate and grow past its anterior tip. Finally, new cartilage "fills-in" between the cornua trabeculae and the distal tip of the anterior process of the ethmoid plate.

Development of the rostral region of pipoid frogs has been controversial (Roček and Veselý, '89; Trueb and Hanken, '92). The first question that needs to be answered is: Do

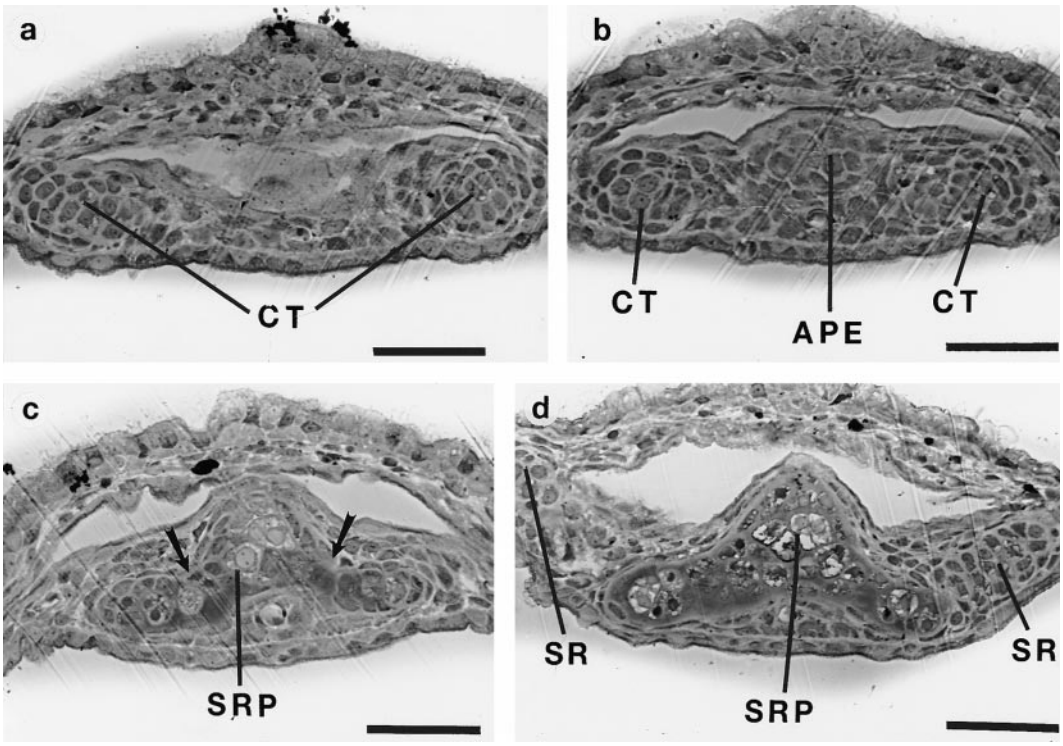

Fig. 5. Cross-sections through the rostral region of *H. boettgeri*, Stage 41. **a**: Most anterior section through the suprarostral plate showing the free anterior tips of the cornua trabeculae. **b**: Section through the suprarostral plate (mid-region) showing the anterior process of the ethmoid plate and the cornua trabeculae. **c**: Section through the ethmoid plate showing the fusion of the anterior process of the ethmoid plate with the cornua trabeculae. **d**: Posterior section showing the suprarostral alae separate from the fused cornua trabeculae and anterior process of the ethmoid plate. APE, anterior process of the ethmoid plate; CT, cornua trabeculae; SR, suprarostral cartilage; SRP, suprarostral plate. Arrows indicate areas of thin cartilage connecting APE and CT. Bar = 50 µm.

the cornua trabeculae of nonpipoid anurans appear in pipoids? Yes; the present study shows that in early stages of *Xenopus*, *Hymenochirus*, and *Rhinophrynus*, rod-like anterior projections of the ethmoid plate are present. Although they are small and transitory, we consider these structures to be homologous (i.e., in position and origin) with the cornua trabeculae of nonpipoid larvae.

The second question to be addressed is: Can the suprarostral plate of pipoid larvae be derived from the cartilaginous structures found in the rostral area of nonpipoid larvae? The data reported here clearly show that this is actually the case. Our serial cross-sections showed that during development the intertrabecular space is occluded with cartilage, forming a continuous plate between the cornua trabeculae, the suprarostrals, and the anterior process of the ethmoid plate.

Answering the previous two questions showed that: first, the rostral region of pipoids and nonpipoids is homologous, refuting Roček and Veselý's ('89) argument for a diphyletic origin of Anura. Second, the cornua trabeculae do form in pipoid larvae. Furthermore, the suprarostral plate develops not from a single fusion and simplification of the cornua trabeculae, as suggested by Trueb and Hanken ('92), but from a complex fusion of the cornua trabeculae, anterior process of the ethmoid plate, and suprarostrals, complemented by the appearance of new cartilage between these elements.

The anterior process of the ethmoid plate described here is most likely homologous to the prenasal process in *Rhinophrynus* (see Sokol, '75). A prenasal process has also been reported for *Pelodytes punctatus* (see Sokol, '77) and *Pipa pipa* (see Roček and Veselý, '89). We identify it as the anterior process of

the ethmoid plate because it forms very early in development as an anterior growth of the ethmoid plate, when the cartilages of the nasal capsules are not yet formed. This process seems to be enlarged in pipoids and will provide support to the septum nasi. A prenasal process is present in urodeles, and its purported absence in anurans was once considered a major difference between Anura, Urodela, and Gymnophiona (de Beer, '37).

Once the homology of the ethmoidal region of pipoids and nonpipoids has been established, a third question needs to be addressed: How can we explain the different developmental patterns found within Pipoidea? We propose that the cornua trabeculae and the anterior process of the ethmoid plate are homologous in *Rhinophrynus*, *Xenopus*, and *Hymenochirus*. The only difference is that the cornua trabeculae of *H. boettgeri* develop not flanking but ventral to the anterior process of the ethmoid plate. This developmental rearrangement is likely the result of a miniaturization process in *Hymenochirus* which, together with the predaceous habit of these larvae, resulted in a shift of the cornua trabeculae to a ventral position relative to the anterior process of the ethmoid plate.

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APPENDIX Stages (Nieuwkoop and Faber, '56) and measurements (in mm) of specimens used

Number	Stage	BL	TL	Status	Number	Stage	BL	TL	Status
Hymenochirus boettgeri					USNM 524328	65	10.11	10.58	C&S
USNM 524286	49	3.21	9.43	C&S	USNM 524329	65	—	9.62	C&S
USNM 524287	50	3.54	7.80	C&S	USNM 524330	65	12.32	12.40	C&S
USNM 524288	50	3.44	10.03	C&S	USNM 524331	66	—	9.85	C&S
USNM 524289	51	3.42	10.31	C&S	USNM 524332	66	—	9.80	C&S
USNM 524290	51	3.93	10.90	C&S	USNM 524333	66+	—	11.88	C&S
USNM 524291	51	3.48	9.93	C&S	USNM 524334	Adult	—	19.83	C&S
USNM 524292	51	3.54	10.82	C&S	*H. boettgeri*	35	1.03	2.11	E-G
USNM 524293	51	3.36	10.16	C&S	*H. boettgeri*	E38	1.06	2.89	E-G
USNM 524294	52	4.03	10.44	C&S	*H. boettgeri*	L38	0.96	2.56	E-G
USNM 524295	52	4.53	10.60	C&S	*H. boettgeri*	41	1.04	3.23	E-G
USNM 524296	52	4.09	11.55	C&S	*H. boettgeri*	41	1.07	3.06	E-G
USNM 524297	53	4.80	12.38	C&S	*H. boettgeri*	41	1.12	3.23	E-G
USNM 524298	53	3.73	11.65	C&S	*H. boettgeri*	42	1.18	3.11	E-G
USNM 524299	53	4.67	10.99	C&S	*H. boettgeri*	45	1.54	4.04	E-G
USNM 524300	54	4.27	10.81	C&S	*H. boettgeri*	45	1.85	4.36	E-G
USNM 524301	54	4.27	10.66	C&S	*H. boettgeri*	45	1.60	4.21	E-G
USNM 524302	54	3.62	11.00	C&S	*H. boettgeri*	45	1.45	3.95	E-G
USNM 524303	55	4.32	13.69	C&S	*Xenopus laevis*				
USNM 524304	55	5.15	15.10	C&S	*X. laevis*	41	2.75	6.14	E-G
USNM 524305	56	6.21	15.63	C&S	*X. laevis*	E43	2.12	6.03	E-G
USNM 524306	56	5.54	14.05	C&S	*X. laevis*	L43	2.08	6.40	E-G
USNM 524307	57	5.53	16.37	C&S	*Rhinophrynus dorsalis*				
USNM 524308	57	4.85	14.25	C&S	USNM 515972	45	5.23	11.04	C&S
USNM 524309	58	5.81	15.89	C&S	USNM 515974	45	4.95	10.02	C&S
USNM 524310	58	5.29	14.24	C&S	USNM 515968	48	6.47	14.50	C&S
USNM 524311	58	5.49	15.35	C&S	USNM 515973	49	6.40	14.15	C&S
USNM 524312	58	6.02	17.17	C&S	USNM 515966	50	10.14	21.21	C&S
USNM 524313	58	5.29	14.67	C&S	USNM 515967	50	10.21	18.19	C&S
USNM 524314	59	7.76	19.90	C&S	USNM 515960	51	12.37	26.17	C&S
USNM 524315	59	7.15	17.80	C&S	USNM 515962	51	9.46	20.07	C&S
USNM 524316	59	7.34	18.19	C&S	USNM 515963	51	10.17	17.98	C&S
USNM 524317	59	6.92	18.04	C&S	USNM 515964	51	11.36	19.74	C&S
USNM 524318	60	8.33	18.87	C&S	USNM 515969	51	10.42	19.07	C&S
USNM 524319	60	8.22	20.78	C&S	USNM 515970	51	10.13	21.43	C&S
USNM 524320	61	8.59	20.39	C&S	USNM 515971	51	10.05	20.21	C&S
USNM 524321	62	8.95	19.49	C&S	USNM 515952	52	14.79	34.60	C&S
USNM 524322	63	9.51	14.61	C&S	USNM 515953	52	13.58	33.64	C&S
USNM 524323	63	8.70	13.20	C&S	USNM 515965	52	11.09	20.33	C&S
USNM 524324	63	8.89	16.14	C&S	*R. dorsalis*	49	6.25	14.01	X-sect
USNM 524325	63	8.77	13.55	C&S	*R. dorsalis*	51	10.49	20.08	X-sect
USNM 524326	64	8.41	12.55	C&S					
USNM 524327	64	8.87	11.65	C&S					

BL, body length; TL, total length; C&S, cleared and stained; E-G, plastic cross-sectioned; X-sect, paraffin cross-sectioned. USNM, National Museum of Natural History, Smithsonian Institution.