
Supplementary information

The oldest tadpole reveals evolutionary stability of the anuran life cycle

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SUPPLEMENTARY INFORMATION

The oldest tadpole reveals evolutionary stability of the anuran life cycle

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1. Geology and paleoenvironment

MPM-PV 23540 was found in a quarry of the Estancia La Matilde, located 100 km SW of Puerto Deseado city, Santa Cruz Province, Argentina (Extended Data Fig. 1). This quarry was named “Estancia La Matilde locality” in the literature¹⁵ and it is the type locality of *Notobatrachus degiustoi*. The rocks exposed at this locality belong to the La Matilde Formation¹⁵.

The La Matilde Formation, a volcanoclastic unit that crops out in several localities in Santa Cruz Province, and is laterally and vertically interbedded with the Chon Aike Formation, a pyroclastic unit covering an area of some 100000 km²(^{16, 63}). The La Matilde Formation was considered Bathonian–Callovian in age (Middle Jurassic) based on fossil floral remains by Stipanovic & Bonetti⁶⁴. However, the age of the La Matilde Formation is also established based on the radiometric dating of volcanic rocks of the interbedded Chon Aike Formation. Whole-rock K-Ar dates ranging from 161 ± 10 (Oxfordian, Late Jurassic) to 138 ± 10 Ma (Valanginian, Early Cretaceous) were obtained from samples in the Gran Bajo de San Julián area⁶⁵. Rb-Sr whole-rock analyses were also carried out on several lavas and associated ignimbrites from the eastern outcrops of the Chon Aike Formation, resulting in an isochron of 168.2 Ma (Bathonian, Middle Jurassic^{66,67}).

MPM-PV 23540 comes from fine-grained, light-grey, tuffaceous sandstones deposited in alternating thin light and dark layers, which correspond to the “Fossiliferous level I” recognised by Stipanovic and Reig¹⁵ in their original description of the “Estancia La Matilde locality”. Geological data for the La Matilde Formation indicate that these sediments were deposited in lacustrine environments, scattered across the floodplain and with pyroclastic material input under seasonally dry-wet climatic conditions^{16,17}. The

Estancia La Matilde locality has yielded hundreds of adult specimens of *Notobatrachus degiustoi* preserved as impressions, but also contains diverse plants, branchiopods, bivalves, and insects (Extended Data Fig. 1). Carapaces of the clam shrimp *Eosolimnadiopsis? santacruzensis*¹⁸ are frequently found in the sandstones along with adult specimens of *N. degiustoi*. It is noteworthy that clam shrimps (Spinicaudata) currently live in temporary water bodies^{68,69}. Interbedded between the frog-bearing levels are abundant vegetal detritus provided by surrounding forests composed of Equisetales, Osmundales, Coniferales, Araucariaceae, and Podocarpaceae^{19,20,70–72}. Taken together, sedimentological and fossil data suggest that the fossil-bearing levels at Estancia La Matilde locality were deposited in shallow, ephemeral lentic environments with a high input of plant debris.

2. Additional anatomical information

MPM-PV 23540 represents an incomplete tadpole preserved in one slab in dorsal aspect, with most of the right half of the body and the proximal part of the tail (Extended Data Figs. 2–3). There is a whitish area delimiting the contour of the specimen, which clearly has a polished surface that contrasts with the rugose surface of the slab, allowing us to distinguish soft parts of the body and tail (Extended Data Figs. 2–3). In general, preserved structures exhibit a high degree of anatomical relationship, with skeletal elements in articulation and soft parts in natural position.

Skeletal elements are represented only by impressions, mirroring the taphonomic mode observed in most adult specimens of *Notobatrachus degiustoi*. Some elements (e.g., cleithrum and femoral shaft) left a striated imprint, probably due to the incipient ossification stage of these bones. Several soft parts, including eyes and nerves, were

preserved as dark imprints, easily identified by their respective anatomical positions (Extended Data Fig. 2).

Both nasals are present, each represented by an anteriorly concave ossification lying close to the anterior end of the frontoparietals (Extended Data Fig. 4). Poorly defined dark areas anterior to the nasal ossifications might correspond either to nasal cartilages or to rostral cartilage. The ossification corresponding to the parasphenoid bone is visible in the median region of the cranium, flanked by the paired frontoparietals, the latter of which extend as longitudinal thin strips of bone widely separated from each other along the midline and markedly pointed anteriorly. The posterior region of the chondrified neurocranium is preserved as imprints, whereas the right otic region is represented by a subtriangular and knob-like prominence, the crista parotica. A large circular notch is present laterally in the crista parotica, which corresponds with the fenestra ovalis (Extended Data Fig. 4). The crista parotica exhibits a simple structure, lacking anterolateral (*processus anterolateralis*) and posterolateral projections, like in early-diverging crown-anurans such as *Ascaphus* and discoglossids⁴⁵. Exoccipitals (Extended Data Fig. 4) may be present in the midposterior region of the skull, although their identity is not certain.

The right hyobranchial skeleton is well preserved in MPM-PV 23540. A dark robust plate represents a ceratohyal positioned lateral to the otic region, with its medial and lateral regions delimited by a slight constriction. The medial part of the ceratohyal exhibits both a *processus anterior* and a *processus posterior*, whereas the lateral part shows a *processus lateralis* (Fig. 2). A *condylus articularis* is also distinguishable, implying the presence of an articulation with the unpreserved palatoquadrate cartilage. The overall morphology of the ceratohyal strongly resembles that of extant tadpoles, in which processes of this element provide attachment surfaces for muscles that power the buccal pump^{5,30,31}.

Slightly ornamented ceratobranchialia are preserved in MPM-PV 23540 as four dark mediolaterally oriented bands, with bright ellipsoid fields between them representing branchial slits (Extended Data Fig. 2). Ceratobranchial I is partially overlapped by the ceratohyal. Ceratobranchials II–IV are proximally connected by commissurae proximales and distally by commissurae terminales. Similar proximal fusions (commissurae proximales) are found in tadpoles of Discoglossoidea and Pipoidea, in contrast to numerous species of Neobatrachia where some or all commissurae proximales are absent^{30,45,73,74}. Notably, medial and lateral projections are distinguished along the shafts of ceratobranchialia II and III. These projections represent novel structures of the anuran hyobranchial skeleton, which support the gill filters⁴³. An oblique rod-like element is placed medioventral to ceratobranchial I (Fig. 2); it might be assigned to the lateral projection of the hypobranchial plate, although its identity is doubtful.

The neural arches of nine presacral vertebrae, the sacrum, and three postsacral vertebrae are present (Extended Data Fig. 4). The ossified contralateral halves of the neural arch laminae of vertebrae V–VIII contact at the midline, and their postzygapophyses are well-developed. Free ribs are present on the right side of presacrals II–IV. The lengths of these ribs diminish anteroposteriorly. The rib of vertebra III possesses a posterior uncinate process at its mid-length, like in adults of *Notobatrachus degiustoi* and early-diverging crown anurans, such as *Ascaphus*, *Leiopelma* and discoglossids^{75,76}. Notably, adult specimens of *N. degiustoi* bear a fourth pair of free ribs in vertebra V, although it was not still ossified in young postmetamorphic individuals¹³. Vertebrae V–VIII show short transverse processes on both sides, whereas the ninth vertebra preserves only the left transverse process (Extended Data Fig. 4). The sacral diapophyses are narrow and not expanded distally. There are three postsacral vertebrae represented by neural arches, those

of postsacral 1 exhibit minute transverse processes (Extended Data Fig. 4). Extant anurans also bear three postsacral vertebrae, which fuse during metamorphosis, forming the coccyx, one of the elements that constitute the urostyle⁷⁷. Thus, the *N. degiustoi* tadpole demonstrates a conservative morphogenetic developmental pathway for the urostyle, a key evolutionary feature of the anuran *bauplan*¹.

The pectoral girdle is only represented by the cleithrum, which is an elongated bar overlapping the rib of vertebra II. The right forelimb is preserved within the body limits, lateral to the hyobranchial skeleton (Extended Data Figs. 2–3). It is represented by the radio-ulna and the manus. The radius and ulna are fused, although the suture is still clearly distinguishable. Metacarpals II–V are represented as the imprints of cylindrical shafts, with slight lateromedial overlapping between them. Metacarpal IV is the longest and stoutest. The proximal phalanges of the four digits are short cylindrical elements with a slight constriction around mid-length, whereas the central phalanges are only present in digits IV and V.

The posterior appendicular skeleton is represented by the ossifications of the right ilium and femur, placed posterolaterally to the third postsacral vertebra (Extended Data Fig. 3). The right iliac shaft is preserved in lateral view, with the dorsal acetabular expansion already ossified. The femur extends laterally, with its diaphysis oriented almost perpendicular to the axial skeleton, and its proximal epiphysis is slightly overlapped by the ilium (Extended Data Fig. 3).

2.1 Ossification state and ontogenetic stage of the *Notobatrachus degiustoi* tadpole

MPM-PV 23540 is at an advanced stage of development, just prior to the beginning of metamorphosis, as indicated by the presence of the forelimb within the body contour and

the state of development of cranial and postcranial ossifications (Extended Data Fig. 4). During development, the parasphenoid, frontoparietals and exoccipitals are the first skull ossifications to be differentiated^{78–82}. These ossifications are present in the *Notobatrachus degiustoi* tadpole, demonstrating a conservative onset of ossification in anurans. Regarding the vertebral column, ossification of sacral diapophyses starts during metamorphic climax²², whereas the ossification of the coccyx is only established late during metamorphosis^{1,77}. This is congruent with the developmental stage of the vertebral column of the *N. degiustoi* tadpole, allowing us to infer a stage previous to metamorphosis. Furthermore, the presence of an ossified central phalanx, fused radio-ulna, and ilium not contacting the sacral vertebra allows us to refer MPM-PV 23540 to a late developmental stage. In sum, we interpret the tadpole of *N. degiustoi* to be at Gosner Stage 40–41²⁴.

2.2 Soft parts

The exquisite preservation of MPM-PV 23540 sheds light on some remarkable aspects of the soft anatomy of the tadpole of *Notobatrachus degiustoi*. Both eyes are preserved, but only the right is complete (Extended Data Fig. 2). The eyes are dorsally positioned; the lens within the right eye is clearly distinguishable in the anterolateral region of the eyeball, which is indicative of the eye's anterolateral facing direction.

Several nerve impressions are distinguishable on the right side of the cranial region (Fig. 2). Based on comparisons with extant crown-anurans such as *Discoglossus pictus*⁴⁴ and several neobatrachians⁸³, we differentiate three different rami of the trigeminal nerve anterior to the eyeball. The truncus maxilo-mandibularis of the trigeminal nerve diverges around the middle of the orbit into the medial maxillary ramus (V₂) and the lateral mandibular ramus (V₃). Posterior to the eyeball, a thickened area is observed (Fig. 2). This

structure is identified as a ganglionic complex, whose neural components cannot be clearly determined, but likely corresponds to ganglia of nerves V and VII^{73,84}. Posteriorly to this ganglionic complex, there is a prominent hyomandibular trunk (truncus hyomandibularis) that extends toward the ceratohyal in the ramus hyoideus of the facial nerve (VII). The hyomandibular trunk also diverges distally into two main rami: the internal mandibular ramus of the facial nerve and the external mandibular ramus. The latter corresponds to a composite of fibres belonging to the facial nerve and fibres belonging to the anteroventral lateral line in extant anuran tadpoles^{44,84,85}.

2.3 Bromalite

On the right side of the axial skeleton, from the level of vertebra V to postsacral 3, there is a compact, yellowish accumulation of particles enclosed within the tadpole body limits (Extended Data Figs. 2–3). It has rounded boundaries and a roughly ovoidal shape. It extends posteriorly as a narrow, diffuse accumulation that ends near the base of the tail, at the level of the anterior end of the ilium. Several crystalline grains are observed within the accumulation, thus contrasting with the rest of the very fine-grained slab containing the specimen. We consider this accumulation as representing in situ intestinal matter or a cololite⁸⁶. The preserved cololite is not in the form of a spiral, but instead suggests a sack-like structure for the digestive system. This could be explained by the developmental stage of the *Notobatrachus degiustoi* tadpole, which is at the end of prometamorphosis, when the coiled intestine of filter-feeding tadpoles undergoes deep structural changes to become a sac-like gut in adults³⁶.

3. Morphometric analyses

To obtain an estimation of the total length of the *Notobatrachus degiustoi* tadpole, we built a linear regression model of body length against total length ($F_{1,106} = 913.5$, $p < 2.2e-16$, multiple R-squared = 0.896, slope = 2.8995, intercept = 0.764) where both traits were highly correlated ($r^2 = 0.947$; $p < 2.2e-16$). The body length of the *N. degiustoi* tadpole is 54.7 mm, thus the estimated total length is 159 mm.

As we established a threshold for gigantism based on the dataset of extant tadpoles (see Methods), those tadpoles having a TL equal to or higher than twice the mean of the TL of the sampled species ($X = 50.94$; $n = 190$; $SD = \pm 28.2$) are considered giants (i.e., $TL \geq 101.8$ mm). Thus, the *N. degiustoi* tadpole is within the size range of extant giant tadpoles (Extended Data Fig. 9). Notably, the largest postmetamorphic specimens of *N. degiustoi* reached a body size (SVL) of 150 mm¹³, which also is within the range of giant adult anurans (>127 mm SVL)³⁴. We found a weak correlation ($r^2 = 0.44$; $p < 2.825e-10$) between tadpole and adult body size in contrast to Emerson³², but in agreement with a recent comprehensive study about the evolution of body size in tadpoles and adult anurans⁸⁷.

4. Phylogenetic analyses

We employed the morphological matrix of Haas⁴⁵, excluding characters 12, 83, 116, and 117 following Roelants et al.⁸, resulting in a matrix of 152 characters and 86 terminals with the inclusion of the *Notobatrachus degiustoi* tadpole.

4.1 Character list (modified from Haas [2003]⁴⁵)

- 1 Epidermal melanocytes elongated and forming orthogonal mesh (0); melanocytes of irregular shape not forming reticulation (1)
- 2 Larval subdermal serous glands absent (0) present (1).
- 3 Keratodonts absent (0) present (1).
- 4 Labial ridges with single row of keratodonts (0) ridges each with double (or more) rows of keratodonts (1).
- 5 Distal anterior labial ridge and keratodont-bearing row broad (0) very short and median (1).
- 6 Keratinized jaw sheaths absent (0) present (1).
- 7 Oral lips absent (0) present (1).
- 8 Upper lip papillation continuous (0) broad diastema present (1).
- 9 Lower lip papillation complete without gap (0) diastema in papillation (1).
- 10 Oral orifice wide (0) narrow (1).
- 11 Eye position dorsolateral (0) lateral (1).
- 12 Single-layered dermis (0) double-layered dermis (1).
- 13 Vena caudalis dorsalis absent (0) present (1).
- 14 Paired venae caudales laterales short (0) long (1).
- 15 Operculum not fused to abdominal wall (0) fused to abdominal wall (1).
- 16 Opercular canal absent and spiracles paired (0) canal present and spiracle single (1).
- 17 Spiracle position median anterior (0) sinistral (1) median posterior (2).
- 18 M. geniohyoideus origin from basibranchial (proc. urobranchialis) (0) ceratobranchial I/II (1) ceratobranchial III (2) planum hypobranchiale (3) from connective tissue lateral to glandula thyroidea (4).
- 19 M. tympanopharyngeus absent (0) present (1).

- 20 Insertion of m. levator arcuum branchialium IV restricted to distal parts of ceratobranchial IV (0) insertion reaching medially and extending on proximal parts of ceratobranchial IV (1).
- 21 M. transversus ventralis IV absent (0) present (1).
- 22 M. interhyoideus posterior absent (0) present (1).
- 23 M. interhyoideus posterior thin but complete sheath (0) loosely spaced fibers in restricted areas of the operculum (1) extensive and strongly developed (2) (see character 23).
- 24 M. diaphragmatopraecordialis absent (0) present (1).
- 25 M. diaphragmatopraecordialis meeting m. interhyoideus posterior in smooth arch (0) both muscles meeting in specific angled pattern (1).
- 26 M. constrictor branchialis I absent (0) present (1).
- 27 Interbranchial septum III musculature absent (0) present (1).
- 28 Interbranchial septum IV musculature absent (0) lateral fibers of m. subarcualis rectus II?IV invade septum (1) origin of m. subarcualis rectus II?IV completely far lateral (2).
- 29 M. subarcualis rectus II?IV one flat tract of fibers (0) split into medial and lateral separate muscles (1).
- 30 Single m. subarcualis obliquus originates from ceratobranchial II (0) two clearly separate heads from ceratobranchialia II and III respectively (1) three portions from ceratobranchialia II III IV (2).
- 31 M. subarcualis rectus accessorius absent (0) present (1).
- 32 M. subarcualis rectus I portion with origin from ceratobranchial I absent (0) present (1).
- 33 M. subarcualis rectus I portion with origin from ceratobranchial II absent (0) present (1).

- 34 M. subarcualis rectus I portion with origin from ceratobranchial III absent (0) present (1).
- 35 Insertion site of the m. subarcualis rectus I on medial parts of ceratohyal (0) ventral muscle portion inserts laterally (1).
- 36 Anterior insertion of m. subarcualis rectus II?IV on ceratobranchial I (0) ceratobranchial II (1); ceratobranchial III (2).
- 37 M. subarcualis rectus II?IV continuous (0) discontinuous (1).
- 38 Insertion of m. rectus cervicis far anteriorly [basibranchial] (0) at processus branchiales II or III (1) on proximal ceratobranchialia III and IV (2) on ceratobranchial IV only (3).
- 39 Mm. levatores arcuum branchialium I and II narrow wide gap between them (0) very wide small gap between them or fused (1).
- 40 M. levator arcuum branchialium III not split (0) split into two crossing bundles (1).
- 41 Larval jaw depressors originate from neurocranium (otic capsule) and dorsal fascia (0) from palatoquadrate (1).
- 42 M. hyoangularis absent (0) present (1).
- 43 Jaw depressors or portions thereof with post- or suborbital origin (0) jaw depressors originating exclusively preorbitally (1).
- 44 M. suspensoriohyoideus absent (0) present (1).
- 45 Origin of m. suspensoriohyoideus from posterior base of processus muscularis (0) from posterior palatoquadrate (1) from otic capsule (2).
- 46 M. interhyoideus and m. intermandibularis in close proximity (0) well separated by a gap (1) X-configuration (2).
- 47 M. mandibulolabialis absent (0) present (1).

- 48 M. mandibulolabialis inserting in soft tissue of lip (0) inserting exclusively on cartilago labialis inferior (1).
- 49 M. mandibulolabialis superior absent (0) present (1).
- 50 M. mandibulolabialis superior originating from medial edge of cartilago meckeli (0) from posteroventral tip of cartilago labialis inferior (1).
- 51 Origin of m. intermandibularis along a line on the ventral side of the cartilago meckeli corpus (0) restricted to the medial face of the cartilago meckeli (1).
- 52 Upper jaw without insertion of jaw muscles (0) upper jaw cartilages powered by jaw muscles. (1).
- 53 Larval m. levator mandibulae externus present as one muscle body (0) two portions (profundus and superficialis) (1).
- 54 Larval m. levator mandibulae externus inserts exclusively on lateral side of mandible (0) main portion inserts on upper jaw cartilages (1) soft tissue insertion (2).
- 55 Functional larval m. levator mandibulae lateralis absent (0) present (1).
- 56 M. levator mandibulae lateralis inserts in tissue close to posterodorsal process of suprarostril cartilage or adrostral tissue (0) nasal sac insertion (1).
- 57 M. levator mandibulae internus high (0) low (1) anterior (2).
- 58 Insertion of the larval m. levator mandibulae internus in relation to jaw articulation medial (0) lateral (1) broadly across articulation (2).
- 59 M. levator mandibulae longus originates dorsally on skull (0) from posterior palatoquadrate (1) exclusively from arcus subocularis (2).
- 60 M. levator mandibulae longus in one portion (0) superficialis and profundus portions (1).

- 61 Profundus and superficialis portions of m. levator mandibulae longus broadly overlapping at insertion (0) not overlapping parallel (1).
- 62 Ramus maxillaris (cranial nerve V2) lateral to m. levator mandibulae longus (0) medial to the muscle (1).
- 63 Ramus mandibularis (c.n. V3) posterior (ventral) to m. levator mandibulae longus (0) between portions of muscle (1) anterior (dorsal) to the muscle (2).
- 64 Ramus mandibularis (c.n. V3) posterior (ventral) to m. levator mandibulae externus group (0) runs through the externus group (1) anterior (dorsal) to the externus group (2).
- 65 Processus anterolateralis of crista parotica absent (0) present (1) forms larval processus oticus (2).
- 66 Posterolateral projections of the crista parotica absent (0) process present (1) expansive flat chondrifications (2) processus otobranhialis (3).
- 67 Posterior palatoquadrate curvature flat or slightly concave (0) clearly concave with bulging and pronounced margin (1).
- 68 Palatoquadrate connection to trabecula cranii caudal (0) rostral (1).
- 69 Position of jaw articulation suborbital (0) preorbital (1).
- 70 Suspensorium high (0) intermediate (1) low (2) ultralow (3).
- 71 Processus ascendens stout (0) thin (1).
- 72 Confluence between posterior palatoquadrate and ventral otic capsule absent (0) processus basalis present (1) palatobasal connection present (2).
- 73 Commissura praefacialis absent (0) present (1).
- 74 Commissurae craniobranhiales absent (0) present (1).
- 75 Processus suboticus quadrati absent (0) present (1).
- 76 Processus pseudopterygoideus absent (0) short ($l \leq d$) (1) long ($l > d$) (2).

- 77 Dorsal connection from processus muscularis to neurocranium ligament only (0)
ligament and pointed processus antorbitalis (1) commissura quadrato-orbitalis (2) high
commissura quadrato-orbitalis (3).
- 78 Processus muscularis absent (0) present (1).
- 79 Anterolateral base of processus muscularis without conspicuous projection (0) bearing
ventrolateral process (1).
- 80 Arcus subocularis with smooth and sharply delimited lateral margin (0) irregular margin
(1) three distinct processes (2) processus lateralis posterior (3).
- 81 Shape of arcus subocularis in cross section with horizontal or rising lateral margin (0)
margin sloping ventrad laterally (1) round in cross section (2).
- 82 Cartilago labialis superior (suprarostral cartilage) absent (0) present (1).
- 83 Articulation of cartilago labialis superior with cornu trabeculae by pars corporis (0) by
pars alaris (1) fused into rostral plate (2).
- 84 Pars alaris and pars corporis fused forming smooth margin distally (0) separated by deep
distal notch (1).
- 85 Partes corpores medially separate (0) distal confluence present (1) forming medial body
(2).
- 86 Posterior processes of pars alaris: double (0) single (1).
- 87 Posterior dorsal process of pars alaris tapering conically or ending bluntly not expanded
terminally (0) expanded terminally almost rectangular in lateral view (1).
- 88 Adrostral cartilage absent (0); present but small (1) very large and elongate (2).
- 89 Partes alares and corpores of cartilago labialis superior in line (0) concentric lines (1).
- 90 Lower jaw cartilage uniform (0) differentiated into distal cartilago labialis inferior and
proximal cartilago meckeli (1).

- 91 Orientation of lower jaw U-shaped (0) transverse (1).
- 92 Distal end of cartilago meckeli cylindrical (0) with stout dorsal and ventral processes forming a shallow articular fossa (1) expanded and flattened no fossa (2) broad and flat processus dorsomedialis absent no fossa (3).
- 93 Admandibular cartilage absent (0) present (1).
- 94 Cartilaginous roofing of the cavum cranii absent (0) taenia transversalis present only (1) taeniae transversalis et medialis (fenestrae parietales) present (2) formation of tectum parietale (3) tectum of cavum cranii almost completely chondrified (4) taeniae tecti medialis only (5).
- 95 One perilymphatic foramen (0) two foramina (1).
- 96 Parasphenoid without processus alares (0) processus alares present (1).
- 97 Vertebral centra formation perichordal (0) epichordal (1).
- 98 Distinct medial ossification center of vertebral centra ventral to notochord absent (0) present (1).
- 99 Number of presacral vertebrae more than nine (0) nine (1) eight (2) less than eight (3).
- 100 Larval ribs absent (0) present (1).
- 101 Ceratobranchial I and hypobranchial I fused (0) articulation present (1).
- 102 Hypobranchial skeletal parts as two cylindrical hypobranchialia (0) planum hypobranchiale (1).
- 103 Free basihyal absent (0) present (1).
- 104 Basibranchial long (0) short (1) indistinguishably fused to hyobranchial elements (2) absent (3).
- 105 Hypobranchial plates articulate (0) fused (1).

- 106 Processus urobranchialis absent (0) short not reaching beyond hypobranchial plates (1) very long reaching beyond hypobranchial plates (2).
- 107 Commissura proximalis I: absent (0) present (1).
- 108 Commissura proximalis II absent (0) present (1).
- 109 Commissura proximalis III absent (0) present (1).
- 110 Spicula short or absent (0) present and long (1).
- 111 Reticulating cartilaginous connections between spicula III and IV absent (0) present (1).
- 112 Processus branchialis open (0) closed (1).
- 113 Ceratohyal without diarthrotic articulation with palatoquadrate slender medial part (0) diarthrotic articulation present medial part broad (1).
- 114 Processus postcondylaris of ceratohyal absent (0) present (1).
- 115 Cartilaginous projections along ceratobranchialia absent (0) present (1).
- 116 Accessory longitudinal bars of cartilage dorsal to ceratobranchialia II and III absent (0) present (1).
- 117 Endolymphatic spaces extend into vertebral canal for less than half of the vertebral column (0) extending into more than half of vertebral canal (presacral vertebra IV or beyond) (1).
- 118 Intracranial endolymphatic system: anterior intracranial processus ascendens absent (0) anterior processus ascendens present (1).
- 119 Cleft between hyal arch and branchial arch I closed (0) open (1).
- 120 Internal gill filaments absent (0) present (1).
- 121 Ligamentum cornuquadratum absent (0) present (1) cartilaginous connection (2).

- 122 Ligamentum cornuquadratum inserting on ala of cartilago labialis superior (0) inserting on cornu trabeculae (1).
- 123 Ligamentum mandibulosuprarostrale absent (0) present (1).
- 124 Ventral valvular velum absent (0) present (1).
- 125 Posterior margin of ventral velum continuous (0) discontinuous (1).
- 126 Glottis position posterior (0) anterior (1).
- 127 Nostrils open in larval stages (0) closed in larval stages (1).
- 128 Forelimb erupts out of limb pouch outside peribranchial space (0) limb erupts from pouch into peribranchial chamber (1).
- 129 Larval lungs rudimentary or absent (0) present and functional (1).
- 130 Branchial food traps absent (0) present (1).
- 131 Branchial food traps continuous (0) divided crescentic (1).
- 132 Secretory ridges absent (0) present (1).
- 133 No transport of larvae (0) larvae are picked up at oviposition site and transported to body of water adhering to dorsum of adult (1).
- 134 Amplexus absent (0) present (1).
- 135 Amplectic position inguinal (0); axillary (1); cephalic (2).
- 136 Advertisement call absent (0); inspiratory (1); expiratory (2); nonairflow (3).
- 137 Clutches: clumps of eggs or scattered submerged eggs (0); strings (1); floating eggs/surface film (2).
- 138 Guiding behavior absent (0); present (1).
- 139 Pupil shape vertical (0) horizontal (1) inverted drop-shaped (triangular) (2) round (3).
- 140 Shoulder girdle arciferal (0) firmisternal (1) pseudofirmisternal (2).
- 141 Clavicula absent (0) overlapping scapula anteriorly (1) not overlapping (2).

- 142 Palatine absent (0) present (1).
- 143 Parahyoid bone absent (0) present (1).
- 144 Cricoid as two cartilages (0) closed ring (1) ring with ventral gap (2) ring with dorsal gap (3).
- 145 Tongue absent (0); present (1).
- 146 Tibiale and fibulare separate (0) elongate but not fused at ends (1) elongate and fused at ends (2) elongate and fully fused (3). *We modified this character in order to describe the condition of Notobatrachus, which bears elongated tibiale and fibulare but they are not fused in the proximal and distal ends.*
- 147 Intercalary element absent (0) present (1).
- 148 M. caudalipuboischiotibialis absent (0) present (1).
- 149 M. sartorius partially or fully fused with m. semitendinosus (0) discrete muscle (1).
- 150 Bidder organ absent (0) present (1).
- 151 Musculus intermandibularis simple undifferentiated (0) with supplementary apical portion (1) supplementary posterolateral portion (2).
- 152 Terminal phalanges rounded (0) claw-shaped (1) bifurcated T- or Y- shaped (2).

4.2 Calibration points

Discoglossidae-*Iberobatrachus angelae*⁴⁷

La Huérguina Formation, Early Cretaceous (120 Ma)

Pipidae-*Cratopipa novaolindensis*⁴⁸

Crato Formation, Early Cretaceous (113 Ma)

*Anomocoela-Aerugoamnis paulus*⁴⁹

Fossil Butte Member of Green River Formation, early Eocene (52 Ma)

Pyxicephalidae-*Thaumastosaurus servatus*⁵⁰

Phosphorites du Quercy, late Eocene (39.5 Ma)

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