Feeding Mechanisms in Two Treefrogs, *Hyla nana* and *Scinax nasicus* (Anura: Hylidae)

M.F. Vera Candioti,[1]* E.O. Lavilla,[1] and D.D. Echeverría[2]

[1]*Instituto de Herpetología, Fundación Miguel Lillo, San Miguel de Tucumán, 4000, Argentina*
[2]*Laboratorio de Vertebrados, Departamento de Ciencias Biológicas, Universidad de Buenos Aires, Buenos Aires, 1428, Argentina*

ABSTRACT After the description of the chondrocranium, hyobranchial apparatus, associated musculature, buccal apparatus, buccopharyngeal cavity, digestive tract, and gut contents, it was possible to define the feeding modes of *Scinax nasicus* and *Hyla nana* tadpoles (Gosner Stages 31–36). *Scinax nasicus* larvae are "typical" microphagous tadpoles, with keratodonts and robust rostrodonts appropriate for rasping surfaces and mincing of food particles; the buccopharyngeal cavity is equipped with filtering structures and has a conspicuous glandular zone and a highly developed branchial basket. In contrast, *H. nana* tadpoles have a modified buccal apparatus; the reduction of the buccopharyngeal and branchial basket structures, together with the high lever–arm ratio and the great development of the depressor muscles of the buccal floor are indicative of macrophagous feeding. J. Morphol. 261:206–224, 2004. © 2004 Wiley-Liss, Inc.

KEY WORDS: *Hyla nana; Scinax nasicus*; tadpole morphology; feeding mechanisms

The morphology and feeding habits of tadpoles have been described in numerous species of anurans. These studies usually focus mainly on morphology or ecology. Thus, studies by Savage (1955), Satel and Wassersug (1981), Ruibal and Thomas (1988), Echeverría (1992a,b, 1997a,b, 1998), Iordansky (1992), among others, provide information on morphological and anatomical features, whereas authors such as Kamat (1962), Inger (1986), Lajmanovich (1994), Lajmanovich and Fernández (1995), and Lavilla (1983) supply ecological analyses.

The hylid frogs *Scinax nasicus* and *Hyla nana* were selected because of the availability of material and the abundance of previous information. These two species live at different levels in the water column in the same lentic environments, suggesting differences in the trophic spectra and the structures involved in the feeding process.

The present study synthesizes information about the feeding modes of anuran larvae by means of morphological, anatomical, and ecological studies. Although works of this kind have been carried out with some taxa (e.g., Seale and Wassersug, 1979; Altig and Johnston, 1989; Haas and Richards, 1998; for a synthesis, see McDiarmid and Altig, 1999), this is the first integrative contribution on the subject for southern South America.

MATERIALS AND METHODS

Specimens

Twenty larvae each of *Scinax nasicus* and *Hyla nana* were collected from temporary ponds near Laguna Paiva and La Toma, Santa Fe, Argentina, in January 2000. Tadpoles were between Stages 31–36 of Gosner (1960), equivalent to Period V of Fabrezi (1988) regarding the development chondrocranium and associated musculature. Voucher specimens are housed at Fundación Miguel Lillo, Tucumán, Argentina, under the numbers FML 12172 (*S. nasicus*) and FML 12173–74 (*H. nana*).

Chondrocranium and Hyobranchial Apparatus

Five specimens of each species were cleared and stained according to Wassersug (1976a). The digestive tract was removed before clearing and staining for use in a separate analysis. Measurements of hyobranchial apparatus were recorded according to Haas and Richards (1998). Measurements were taken from digitized drawings employing UTHSCSA Image Tool software (Wilcox et al., 1995) and were transformed to ratios to eliminate absolute size as a variable.

The shapes of the hyobranchial apparatuses of both species were compared using the thin-plate spline technique with the software tpsDig and tpsSplin (Rohlf, 1997, 1998a). In this method, discrete points (landmarks) are defined in each specimen and the modifications of shape (irrespective of size, position, and rotation of the object) are depicted with respect to a reference configuration by using thin-plate splines (Rohlf and Bookstein, 1990). Selection of landmarks followed that of Haas and Richards (1998). Given that not all the structures present in *Scinax nasicus* appear in *Hyla nana*, some of the original landmarks were redefined. Figure 1 shows the landmarks and measurements considered for the analysis of the hyobranchial apparatuses.

Musculature

For the study of the mandibular and hyoid musculature, five specimens of each species at Stage 31 were prepared following the

*Correspondence to: María Florencia Vera Candioti, Instituto de Herpetología, Fundación Miguel Lillo, Miguel Lillo 251, 4000 San Miguel de Tucumán, Argentina. E-mail: florivc@hotmail.com

Published online in
Wiley InterScience (www.interscience.wiley.com)
DOI: 10.1002/jmor.10239

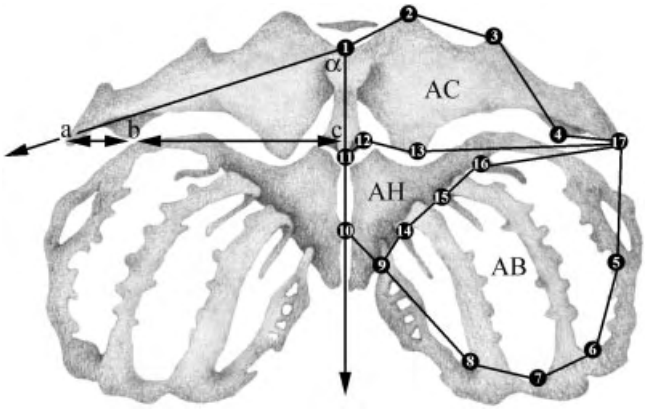

Fig. 1. Landmarks and measurements of the hyobranchial apparatus. a–c, width of the ceratohyal; a–b, width of the lateral portion of the ceratohyal; α, orientation of the ceratohyal; AC, area of the ceratohyal; AH, area of the hypobranchial plate; AB, area of the ceratobranchials. Landmarks: 1, anterior margin of pars reuniens; 2, tip of processus anterior hyalis; 3, tip of processus anterolateralis; 4, condylus articularis; 5, most lateral internal point of branchial basket; 6, most posterior point of gill slit I; 7, most posterior point of gill slit II; 8, most posterior point of gill slit III; 9, most anterior and medial point of ceratobranchial IV; 10, most posterior point of hypobranchial plates junction; 11, most posterior point of copula II; 12, lateral point of hypobranchial plates-copula II junction; 13, tip of processus posterior hyalis; 14, most posterior point of ceratobranchial III-hypobranchial plate junction; 15, most posterior point of ceratobranchial II-hypobranchial plate junction; 16, most posterior point of ceratobranchial I-hypobranchial plate junction; 17, tip of processus lateralis. Redrawn from Haas and Richards (1998).

clearing-and-staining protocol of Wassersug (1976a), with the procedure interrupted before immersion in glycerol. The specimens were then colored with Lugol, to enhance the contrast between blue cartilages and brown-reddish muscles.

The *Mm. interhyoideus* and *orbitohyoideus* were studied in detail in order to compare the buccal pumping mechanism in the two species. The muscles were freed from their attachment to the ceratohyals, placed in a drying oven at 60°C for 5 days, and then weighed with digital scales (precision = 0.000005 g) to obtain their dry weight. The ratio between the weights of *Mm. interhyoideus* and *orbitohyoideus* (IH/OH ratio) was calculated to estimate the forces involved in raising and lowering of the buccal floor (Satel and Wassersug, 1981).

Buccal Apparatus and Buccopharyngeal Cavity

Five tadpoles of each species were analyzed. In three of them the oral disc was removed whole, whereas in two the floor and the roof of the mouth were exposed for analysis of the buccopharyngeal cavity. Samples were dehydrated according to Echeverría (1997a) and then critical-point-dried and gold-coated in an ION Sputtering Balzers SCD 040 metalizer. The observations and micrographs were done with a Philips 505 scanning electron microscope.

Digestive Tract and Gut Content Analysis

Measurements of the digestive tract of 10 larvae of each species from the same pond were taken with a Craftsman caliper (precision = 0.05 mm); the gut contents were extracted to do quantitative analysis. Samples were diluted in an erythrosine-water solution (1:1) and three aliquots (0.09 ml) were evaluated by the Lackey Drop (microtransect) Method (Lackey, 1938). This technique calculates the number of food items per milliliter, applying the formula:

$$N/ml = \frac{C \times TA}{A \times S \times V}$$

where C is the number of organisms counted, TA the area of the slide cover, A the area of a row, S the number of rows counted, and V the volume of the sample under the slide cover.

Each food item was identified and measured. Some organisms, such as filamentous algae, were not quantified; we calculated their frequency of occurrence in all the larvae. The possible differences in the sizes of the food particles ingested by the two species were evaluated applying Student's *t*-test.

All drawings were made using a stereomicroscope equipped with a camera lucida. The skeletal terminology is based on that of Haas and Richards (1998) and Cannatella (1999). Latin terms are used only if English terms are not available. The nomenclature for musculature is the one suggested by Haas (2001, 2003). Terminology for the characterization of oral disc morphology and keratinized structures follows that of Altig and McDiarmid (1999) and Van Dijk (1966). The nomenclature used in the description of the buccopharyngeal cavity is the one suggested by Viertel (1982).

RESULTS

Scinax nasicus

Chondrocranium and hyobranchial apparatus (Fig. 2). The chondrocranium and hyobranchial apparatus of *Scinax nasicus* tadpoles were described by Fabrezi and Vera (1997), based on specimens at Stages 31–37 from Salta, Argentina. Examination of five specimens at Stage 31 from Santa Fe, Argentina, shows some intraspecific variation. The suprarostral cartilage has a simple corpus with a concave dorsal margin; the corpus is fused to the alae, although these can still be distinguished. Each ala bears a processus dorsalis posterior. The tadpoles examined by Fabrezi and Vera have a different suprarostral cartilage, with the corpus syndesmotically joined to the alae. The hyobranchial apparatus has a conspicuous, elliptical copula I, not recorded in Fabrezi and Vera's specimens. Table 1 shows the relative sizes of each area of the hyobranchial apparatus. The area of the ceratobranchials is highly developed, occupying more than half the whole area.

Musculature (Fig. 3).

M. mandibulolabialis. This muscle is composed of two slips, *Mm. mandibulolabialis inferior* and *superior*, both of which originate from the ventromedial region of Meckel's cartilage. The inferior slip inserts in the ventral marginal papillae and in the valleys between the tooth rows P-1–P-2 and P-2–P-3. The superior slip inserts in the lateral marginal papillae and part of the upper lip, and between them and tooth row A-1.

M. intermandibularis. From its origin on the middle part of Meckel's cartilage, muscle fibers diverge medially to meet with the opposite half, thereby

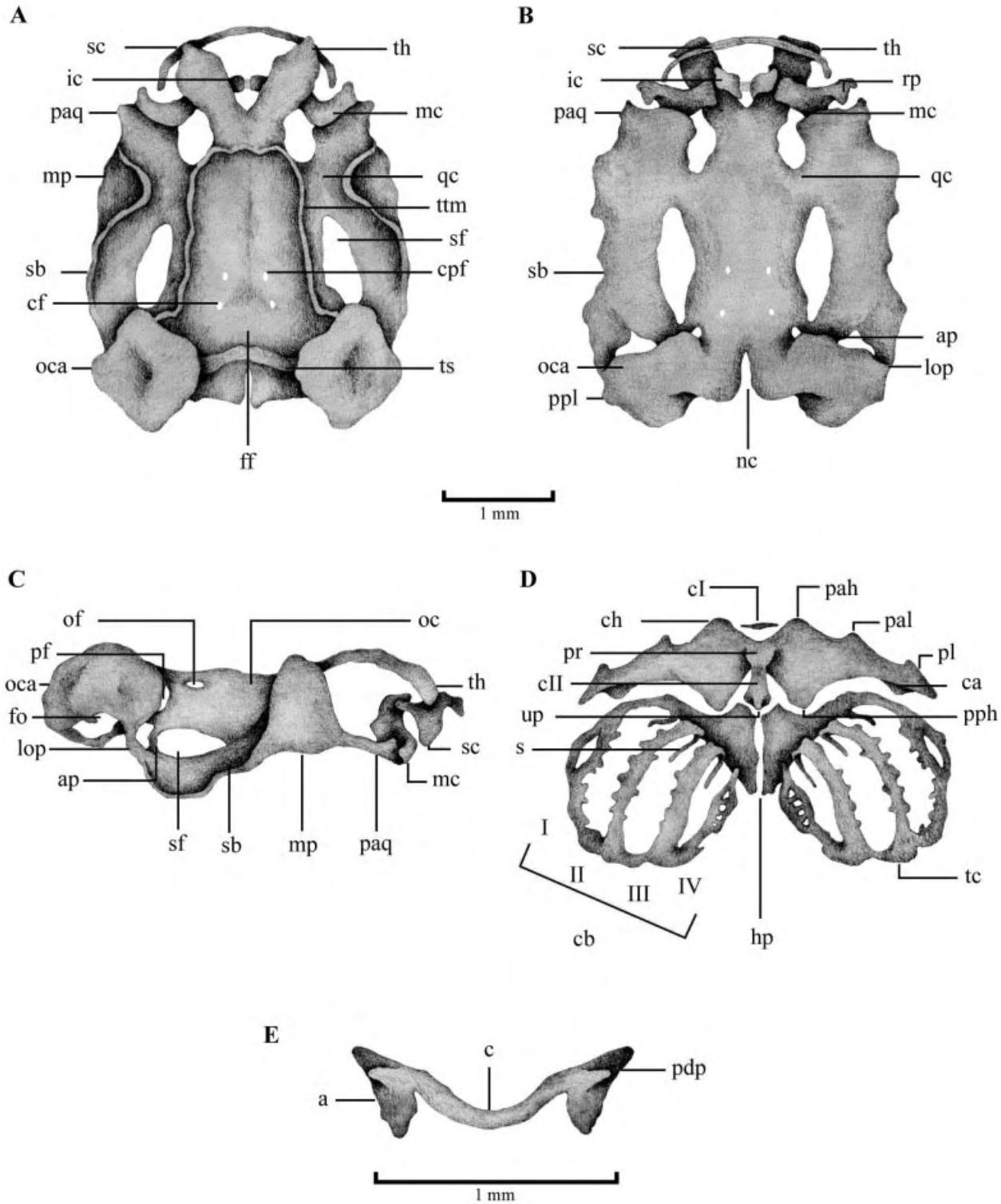

Fig. 2. *Scinax nasicus* Stage 31. Chondrocranium and hyobranchial apparatus. **A:** Chondrocranium, dorsal view. **B:** Chondrocranium, ventral view. **C:** Chondrocranium, lateral view. **D:** Hyobranchial apparatus, ventral view. **E:** Suprarostral cartilage, frontal view. a, ala; ap, ascending process; c, corpus; ca, condylus articularis; cb, ceratobranchial (I–IV); cf, carotid foramen; ch, ceratohyal; cI, copula I; cII, copula II; cpf, craniopalatine foramen; ff, frontoparietal fenestra; fo, fenestra ovalis; hp, hypobranchial plate; ic, infrarostral cartilage; lop, larval otic process; mc, Meckel's cartilage; mp, muscular process; nc, notochordal canal; oc, orbital cartilage; oca, otic capsules; of, optic foramen; pah, processus anterior hyalis; pal, processus anterolateralis; paq, pars articularis quadrati; pdp, processus dorsalis posterior; pf, prootic foramen; pl, processus lateralis; pph, processus posterior hyalis; ppl, processus posterolateralis; pr, pars reuniens; qc, quadratocranial commissure; rp, retroarticular process; s, spicule; sb, subocular bar; sc, suprarostral cartilage; sf, subocular fenestra; tc, terminal commissure; th, trabecular horn; ts, tectum synoticum; ttm, taenia tecti marginalis; up, urobranchial process.

TABLE 1. Relative values of the hyobranchial apparatuses

Species	AC/T	AH/T	AB/T	a-b/a-c	α
Scinax nasicus	0.34	0.14	0.52	0.23	71.18°
Hyla nana	0.55	0.32	0.14	0.41	47.77°

AB, area of the ceratobranchials; AC, area of the ceratohyal; AH, area of the hypobranchial plate; a-b/a-c, lever-arm ratio; T, total area; α, orientation of the ceratohyal. Values are averages of five specimens of each species, Stage 31.

forming a semicircular structure, which may be free or attached to the *M. interhyoideus* aponeurosis.

M. levator mandibulae longus superficialis. This superficial muscle of the jaw levator complex originates from the posterior border of the subocular bar and the superior border of the ascending process of the palatoquadrate. It borders the subocular fenestra externally up to the medial extreme of Meckel's cartilage.

M. levator mandibulae longus profundus. This muscle lies deep to the *M. l.m. longus superficialis.* It originates from the ascending process of the palatoquadrate and inserts via a long tendon on the ventral border of the suprarostral cartilage.

M. levator mandibulae internus. This deepest and most medial of the jaw levator muscles originates from the ventral surface of the ascending process of the palatoquadrate and the anteroventral surface of the otic capsule. It runs longitudinally through the subocular fenestra and inserts via a well-developed tendon on the lateral edge of the Meckel's cartilage, between the insertions of the *Mm. l.m. articularis* and *hyoangularis*.

M. levator mandibulae externus superficialis. This small, flat muscle has few fibers. It originates from the anterior border of the muscular process of the palatoquadrate and inserts on the dorsal margin of the ala of the suprarostral cartilage.

M. levator mandibulae externus profundus. The muscle is circular in cross section and originates from the medial, inferior surface of the muscular process of the palatoquadrate. It inserts on the ventral border of the ala of the suprarostral cartilage, together with the tendon of the *M. l.m. longus profundus*.

M. levator mandibulae articularis. This short muscle with a circular cross section originates from the ventromedial surface of the muscular process of the palatoquadrate, below the *M. l.m. externus superficialis*. It inserts on the retroarticular process of Meckel's cartilage, just medial to the attachments of the muscles of the hyoid group.

M. suspensoriohyoideus. From its origin on the posterior descending border of the muscular process of the palatoquadrate, this muscle inserts on the posterior margin of the ceratohyal.

M. orbitohyoideus. The muscle originates from both the anterior and dorsal margin of the muscular process of the palatoquadrate and inserts along the edge of the ceratohyal, covering the *M. Suspensoriohyoideus* entirely.

M. suspensorioangularis. This muscle has a circular cross section and originates from the posterolateral side of the descending border of the muscular process of the palatoquadrate. Its point of origin lies deep to the *M. orbitohyoideus*. Fibers occupy approximately the lower half of the process and extend forward to attach to the retroarticular process of Meckel's cartilage. The insertion point lies medial to that of the *M. quadratoangularis*.

M. quadratoangularis. From its origin from the ventral surface of the palatoquadrate, this muscle inserts on the retroarticular process of Meckel's cartilage; its insertion is the most lateral of the muscles attached to Meckel's cartilage. The muscle is totally concealed by the *Mm. suspensorioangularis* and *hyoangularis*.

M. hyoangularis. This cylindrical muscle originates from the dorsal side of the ceratohyal, anterior to the condylus articularis; it inserts on the retroarticular process of Meckel's cartilage.

M. interhyoideus. The parallel fibers of this muscle are oriented transversely. The muscle originates from an oblique crest located on the ventral side of the ceratohyal, near its lateral edge; fibers extend toward a medial aponeurosis and insert next to the fibers of the contralateral muscle.

M. geniohyoideus. Thin and flat, it originates in the posterior surface of the infrarostral cartilage and extends backward to insert in the hypobranchial plate of the branchial basket.

Table 2 shows the dry weights calculated for the *Mm. orbitohyoideus* and *interhyoideus* of this species.

Buccal apparatus and buccopharyngeal cavity (Figs. 4, 5). In the larvae studied (n = 5, Stage 31), the relaxed oral disc is triangular and has well-developed lateral folds. The disc is subterminal and its length 10% that of the snout–vent length. It bears a single row of marginal papillae, accompanied by smaller submarginal papillae located in the commissural region. The marginal papillae are interrupted to form a rostral gap (Figs. 4A, 5A). The jaw sheaths are wide, thick, robust, and finely serrated. The keratin sheath is wide and the frontal serrations slightly larger and more worn than the lateral ones (Fig. 5B). There are five keratodonts rows, two anteriorly (A) and three posteriorly (P). Tooth rows A-1, P-2, and P-3 are complete and extend from one commissure to the other, whereas A-2 and P-1 are interrupted medially (making the labial tooth row formula (1)(1-1)/(1-1)(2)). Each keratodont has three distinct zones: base, neck, and head. The head is convex, oblong, or slightly wider in its distal extreme, and has an average of 12 cusps (Fig. 5C).

The roof of the buccal cavity (Fig. 4B) is darkly pigmented. Numerous chromatophores occur on both sides of the buccal roof arena and on the dorsal velum. The prenarial arena contains five pustules: a

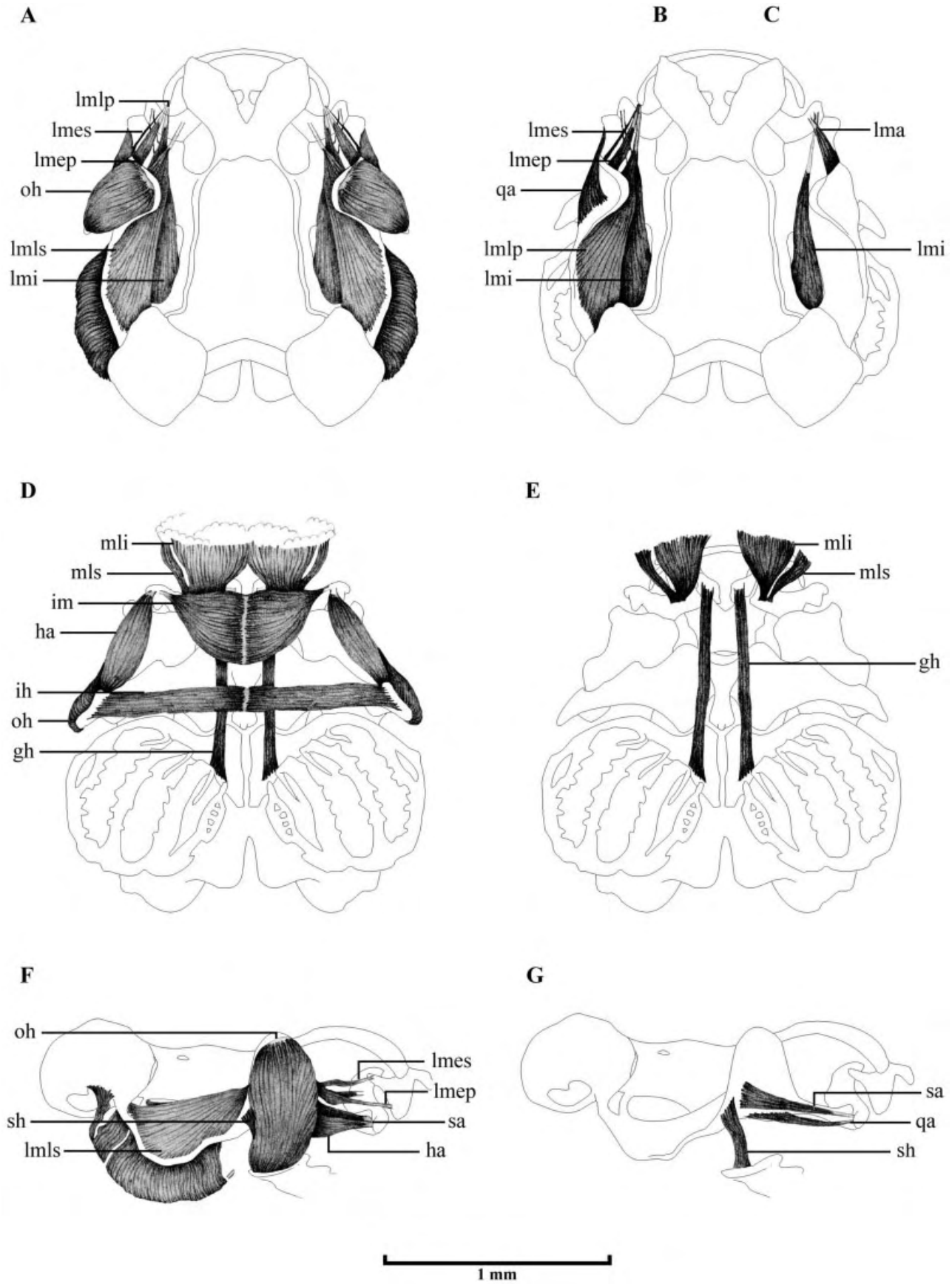
A
lmlp
lmes
lmep
oh
lmls
lmi
B
C
lmes
lmep
qa
lmlp
lmi
lma
lmi
D
mli
mls
im
ha
ih
oh
gh
E
mli
mls
gh
F
oh
lmes
lmep
sh
sa
lmls
ha
G
sa
qa
sh
1 mm

Figure 3

TABLE 2. Development of the Mm. interhyoideus (IH) and orbitohyoideus (OH)

Species	SVL (cm)	Dry weight IH (mg)	Dry weight OH (mg)	IH/OH
Scinax nasicus	1.02	0.061	0.136	0.448
Hyla nana	0.74	0.030	0.206	0.145

Values are averages of five specimens of each species, Stages 31 and 36.

central one and a pair on each side. The internal nares are oriented obliquely (Figs. 4B, 5D). Behind them, the postnarial arena contains paired, trifurcate papillae and central pustules. The median ridge is trapezoidal and its free margin is irregular, with slight projections. Between the postnarial arena and the median ridge there is a second ridge, the premedian ridge, which is less developed. The lateral ridge papillae are paired, bifurcate, and the tallest of the buccal papillae. The buccal roof arena lacks papillae, but has many pustules uniformly distributed. The dorsal velum lacks a glandular epithelium.

On the buccal floor (Fig. 4C), paired keratinized structures (spurs, according to Wassersug, 1980) are located over the commissura intramandibularis, oriented toward the exterior of the mouth (Figs. 4C, 5E). Behind them are three pustules aligned anteroposteriorly. Behind these is the orobranchial cavity, which bears small groups of pustules aligned transversely. Posteriorly, and converging toward the middle axis, there is a pair of bifurcate, nonoverlapping infralabial papillae. The tongue anlage lacks lingual papillae. Four pairs of buccal floor arena papillae are arranged symmetrically and delimit the buccal floor arena. Some of them are flat and bifurcate, whereas others are conical and small. A few pustules are distributed among these buccal floor papillae. The buccal pockets are deep, transversely oriented, and lack papillae. The ventral velum is semicircular and lacks marginal projections and a median notch. The anterior edge of the velum has a narrow band of chromatophores. There is conspicuous glandular epithelium on the free margin of the velum and the zone immediately adjacent to it (Fig. 5F).

Digestive tract and gut content analysis. The digestive tract of *Scinax nasicus* larvae is a 6D-7L coiled tube. The esophagus is short and is separated from the next section by a small constriction, followed by a decrease in diameter. A gastric region is not macroscopically differentiated, but a histological study would be necessary to document its absence. The anterior gut (from its anterior end to the switchback point, where the spiral reverses itself) is slightly thicker than the posterior portion. The diameter of the tube increases distally, forming a region called the "posterior dilatation" by Terán and Michel de Cerasuolo (1988) for *Gastrotheca gracilis* tadpoles. The gut terminates with a dextral vent tube. Meristic data are recorded in Table 3.

Filamentous algae predominate in the gut contents (Table 4). These algae appeared in all of the larvae examined, and the length of their filaments exceeded the field of view of the microscope. In most cases they were Chlorophyta of the genera *Oedogonium* and *Cladophora*, which retained their entire attaching discs and part of the substrate. Other filamentous algae found were Cyanophyta of the genera *Anabaena* and *Oscillatoria.* These were not counted due to their abundance and large size. Among the quantifiable items, euglenophyta, mainly *Lepocinclis,* were predominant. Diatoms (most of them epiphytic, such as *Gomphonema* spp.) were the second most numerous group. The size of most food particles consumed by the larvae ranged from 0.03–0.05 mm. Bigger items represented only 10% of the total (Table 5).

Hyla nana

Chondrocranium and hyobranchial apparatus (Fig. 6). The chondrocranium and hyobranchial apparatus of *Hyla nana* tadpoles were described by Fabrezi and Lavilla (1992) for specimens at Stages 31–35 from Chaco, Argentina. The specimens in our study (n = 5, Stage 31) differ in having a well-developed tectum synoticum, independent infrarostral cartilages, and no terminal commissure III between ceratobranchials III and IV.

The morphometric data obtained from the hyobranchial apparatus of *Hyla nana* tadpoles, together with those of *Scinax nasicus*, are presented in Table 1.

Musculature (Fig. 7). In order to avoid repetitions, only features that differ from *Scinax nasicus* are mentioned. Descriptions are based on four tadpoles in Stage 31 and one in Stage 36.

M. mandibulolabialis. The fibers of this muscle insert on the cutaneous flange that forms the oral tube. The muscle is not divided into two slips as it is in *Scinax nasicus*.

M. levator mandibulae lateralis. This muscle, absent in *Scinax nasicus*, originates from the surface of the nasal sac and inserts on the dorsal and external region of the pars articularis quadrati.

M. orbitohyoideus. The large size of this muscle, together with a large muscular process of the palatoquadrate attached to the lamina orbitonasalis,

Fig. 3. *Scinax nasicus* Stage 31. Musculature. **A:** Dorsal view, superficial plane. **B:** Dorsal view, middle plane. **C:** Dorsal view, deep plane. **D:** Ventral view, superficial plane. **E:** Ventral view, deep plane. **F:** Lateral view, superficial plane. **G:** Lateral view, deep plane. gh, *geniohyoideus*; ha, *hyoangularis*; ih, *interhyoideus*; im, *intermandibularis*; lma, *levator mandibulae articularis*; lmep, *levator mandibulae externus profundus*; lmes, *levator mandibulae externus superficialis*; lmi, *levator mandibulae internus*; lmlp, *levator mandibulae longus profundus*; lmls, *levator mandibulae longus superficialis*; mli, *mandibulolabialis inferior*; mls, *mandibulolabialis superior*; oh, *orbitohyoideus*; qa, *quadratoangularis*; sa, *suspensorioangularis*; sh, *suspensoriohyoideus*.

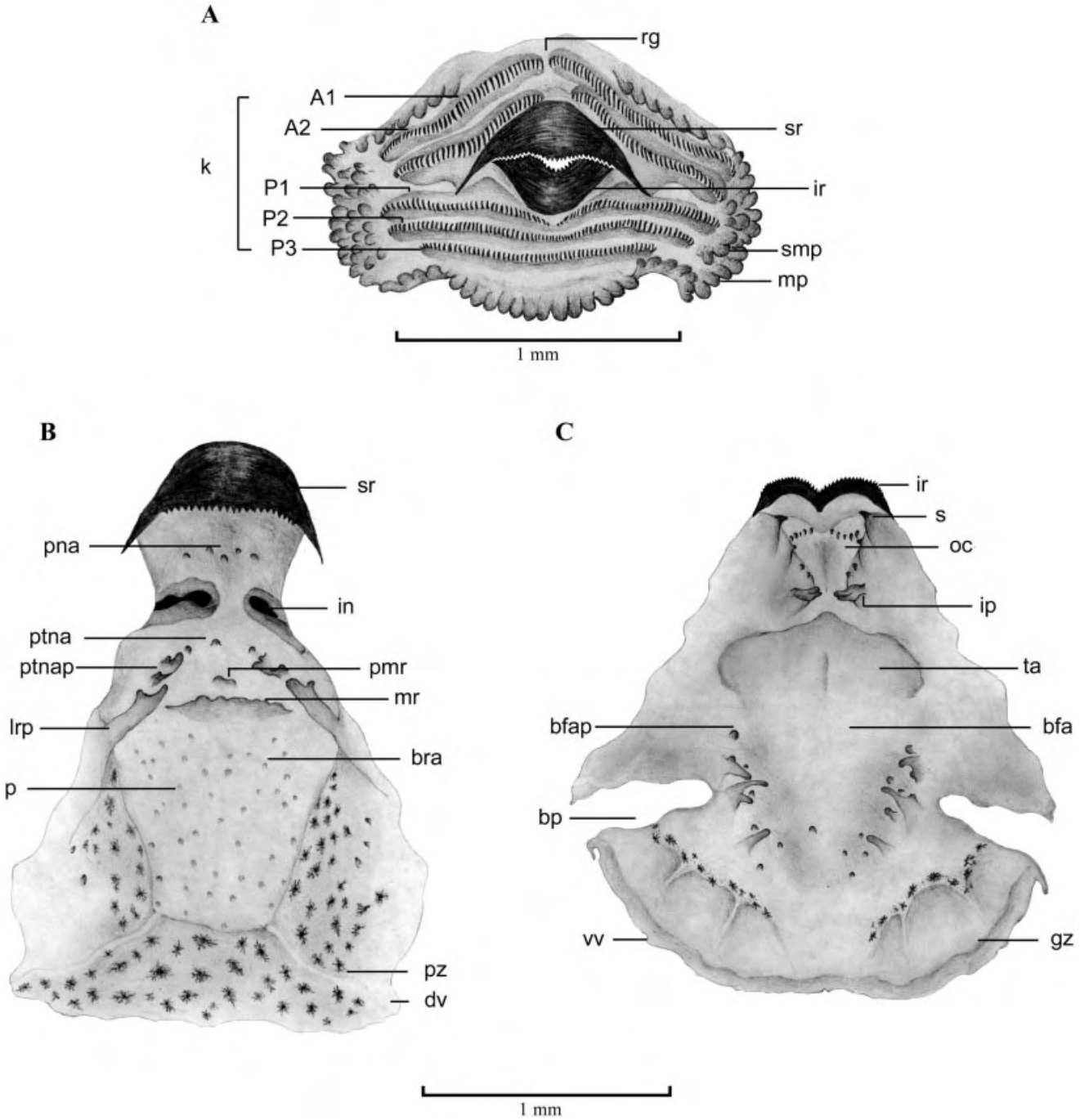

Fig. 4. *Scinax nasicus* Stage 31. Buccal apparatus and buccopharyngeal cavity. **A:** Oral disc. **B:** Buccal roof. **C:** Buccal floor. bfa, buccal floor arena; bfap, buccal floor arena papillae; bp, buccal pocket; bra, buccal roof arena; dv, dorsal velum; gz, glandular zone; in, internal nares; ip, infralabial papillae; ir, infrarostrodont; k, keratodonts (A, anterior; P, posterior); lrp, lateral ridge papillae; mp, marginal papillae; mr, median ridge; oc, orobranchial cavity; p, pustulation; pmr, pre-median ridge; pna, prenarial arena; ptna, postnarial arena; ptnap, postnarial arena papillae; pz, pigmented zone; rg, rostral gap; s, spur; smp, submarginal papillae; sr, suprarostrodont; ta, tonge anlage; vv, ventral velum.

conceals the muscles of the jaw levator complex. This unusual development derives in a very small average value of the IH/OH ratio (Table 2).

M. quadratoangularis. The muscle is more ventral in position than in *Scinax nasicus*, and is not concealed by the *M. suspensorioangularis*.

M. hyoangularis. This well-developed muscle covers the whole *M. quadratoangularis*.

Buccal apparatus and buccopharyngeal cavity (Figs. 8, 9). The buccal apparatus of *Hyla nana* was described by Lavilla (1990). The oral disc is modified into a narrow suctorial tube ($\bar{x}$ =

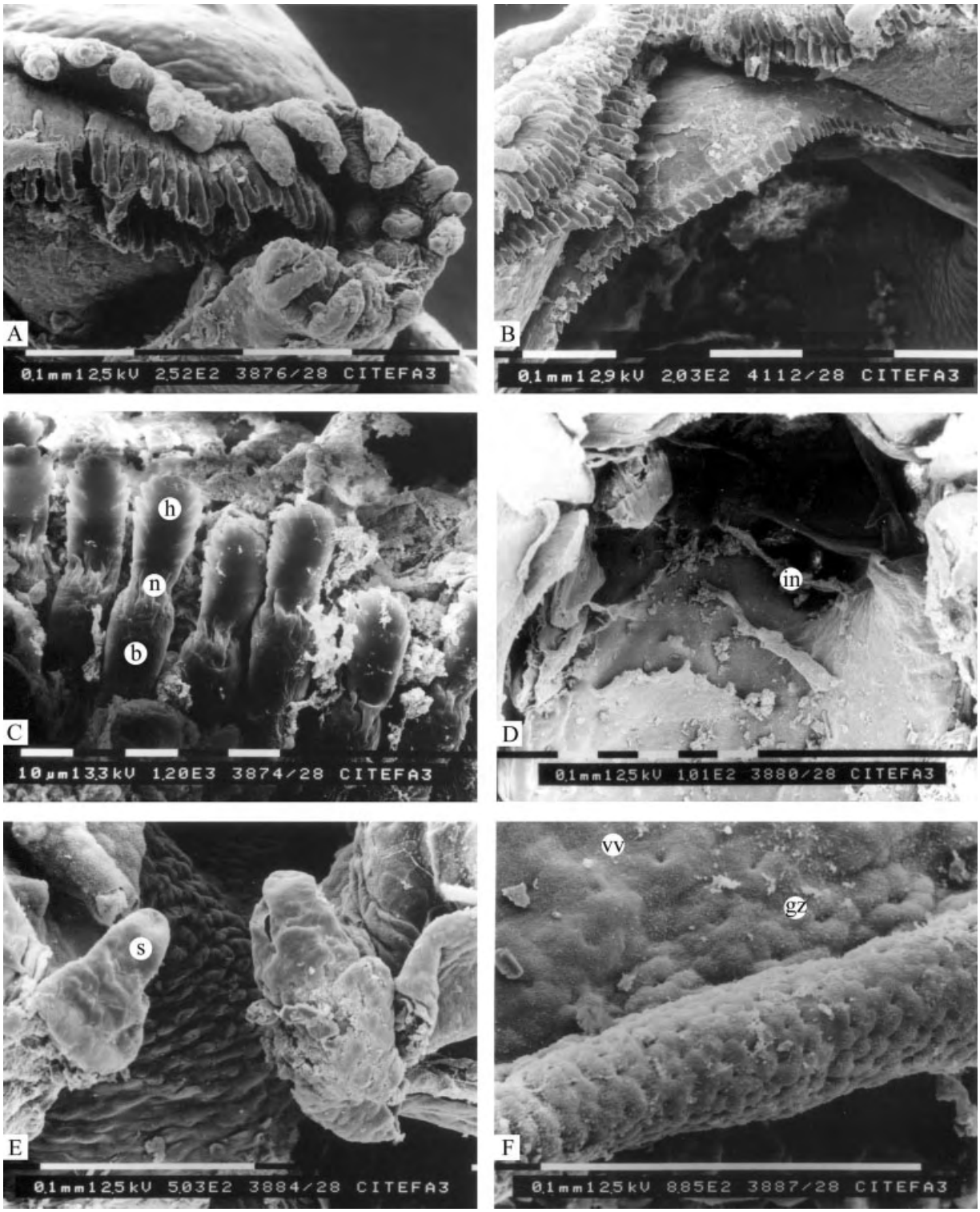

Fig. 5. *Scinax nasicus* Stage 31. Buccal apparatus and buccopharyngeal cavity. SEM. **A:** Oral disc, left commissure. **B:** Detail of suprarostrodont. **C:** Detail of keratodonts, tooth row P-2. **D:** Detail of internal nares. **E:** Detail of spurs. **F:** Detail of ventral velum and glandular zone. b, base; in, internal nares; h, head; gz, glandular zone; n, neck; s, spur; vv, ventral velum.

0.25 mm) that lacks oral papillae and keratodonts. The margins of the jaw sheaths bear fine, conical serrations. Figures 8A and 9A–C show drawings and SEM micrographs of the oral disc of the specimens examined in our study (n = 5, Stage 31).

TABLE 3. Absolute and relative lengths of the organs of the digestive tract

	Scinax nasicus		*Hyla nana*	
Measurement	x̄ (cm)	% in total length	x̄ (cm)	% in total length
Esophagus	0.43	5%	0.23	9%
Manicotto glandulare	—	—	0.27	11%
Intestine	8.17	95%	2.04	80%
Total length of the tract	8.56	100%	2.55	100%
SVL	0.97	—	0.64	—

Values are averages of 10 specimens of each species, Stages 31–36.

TABLE 4. Qualitative analysis of the gut contents, using the Lackey method

	Scinax nasicus		*Hyla nana*	
Taxa	x̄ (N°/ml)	%	x̄ (N°/ml)	%
Filamentous algae	nq	nq	11.9	5%
Crustaceans	1.1	0%	3.3	1%
Desmidiacea	3.3	0%	0	0%
Diatoms	583.3	22%	114.4	41%
Euglenophyta	1965.6	76%	134.8	48%
Oligochaeta	—	—	5*	—*
Tracheophyta remnants	3	0%	2.6	1%
Rizopoda	24.4	1%	10.4	3%
Rotifera	25.6	1%	3.3	1%
Total	2605.6	100%	280.7	100%

*Oligochaeta are not included in this analysis because their small number would make the results based on abundance misleading. nq, non quantifiable. Values are averages of 10 specimens of each species, Stages 31–36.

The buccal roof is elongated (Fig. 8B). The internal nares are longitudinally oriented, elliptical, and small (major axis: 0.2 mm) (Fig. 9D). The buccal roof bears a pair of pustules located on each side of the midline of the cavity. The posterior part of the roof, including the inconspicuous dorsal velum, has numerous chromatophores. Secretory epithelium is absent.

The buccal floor is elongated anterior to the buccal pockets (Fig. 8C). There is one pair of conspicuous, globose infralabial papillae (Fig. 9E). The tongue anlage bears a pair of pustules on its posterior edge. The buccal floor arena is not well delimited because of the lack of buccal floor arena papillae. With intense staining, two groups of five or six pustules can be detected on both sides of the longitudinal axis, on the lateral portion of the ceratohyal. The buccal pockets are small and almost unperforated. Behind them appears the ventral velum, which has a free border with a median notch and without any projections. There are no secretory structures and the glottis is not visible (Fig. 9F).

Digestive tract and gut contents analysis. Part of the digestive tract of this species was described by Lajmanovich et al. (2000). There are three macroscopically distinct sections. The most anterior is the esophagus, comparatively shorter than that of *Scinax nasicus*, and located in a medial position. Next is the manicotto glandulare, surrounded by the liver. The percentage of the total length of the tract occupied by the manicotto is similar to that occupied by the esophagus. The last section is the intestine, separated from the previous region by a sphincter-like constriction. The intestine is a tube of uniform diameter, with the exception of a small distal portion, the posterior dilatation. It is a 2D-3L coiled tube that ends in a short, dextrally oriented vent tube. Table 3 shows the measurements of the digestive tract of 10 larvae at Stages 31–36.

The most conspicuous items in the digestive tracts were oligochaetes. An average of five intact worms was found intact inside the manicotto glandulare; the mean length was 1.87 mm and the mean width 0.25 mm. It should be noted that in all of the specimens examined the intestine was almost empty, which caused low absolute values (Table 4). Between the smaller items, the predominant groups were Euglenophyta (*Euglena* spp.) and the diatoms, which together represented 83% of the total. Filamentous algae were absent, with the exception of small fragments that were considered nonquantifiable. With regard to the most predominant size range (Table 5), there was a preponderance of larger sizes; values higher than 0.1 mm represented 17% of the total. Student's *t*-test detected a significant difference ($P < 0.001$) relative to *Scinax nasicus*.

DISCUSSION

Chondrocranium and Hyobranchial Apparatus

Previous studies of hylid larvae indicate a wide variation in the chondrocranium and hyobranchial apparatus structures. Characters of several species have been analyzed by Fabrezi and Lavilla (1992), Fabrezi and Vera (1997), Haas (1996b, 2003), Haas and Richards (1998), and Lavilla and Fabrezi (1987), among others.

TABLE 5. Distribution of food particles sizes in the digestive content

	Scinax nasicus		*Hyla nana*	
Size (mm)	x̄ (N°/ml)	%	x̄ (N°/ml)	%
0.03	1870.4	65%	37.4	13%
0.04	547.4	19%	58.1	20%
0.05	181.9	6%	44.4	15%
0.06	79.6	3%	33.7	12%
0.07	79.3	3%	25.6	9%
0.08	27.4	1%	28.1	10%
0.09	14.8	1%	11.5	4%
>0.1	74.7	2%	48.2	17%
Total	2875.4	100%	287	100%

Values are averages of the counting of the digestive content of 10 specimens of each species, Stages 31–36.

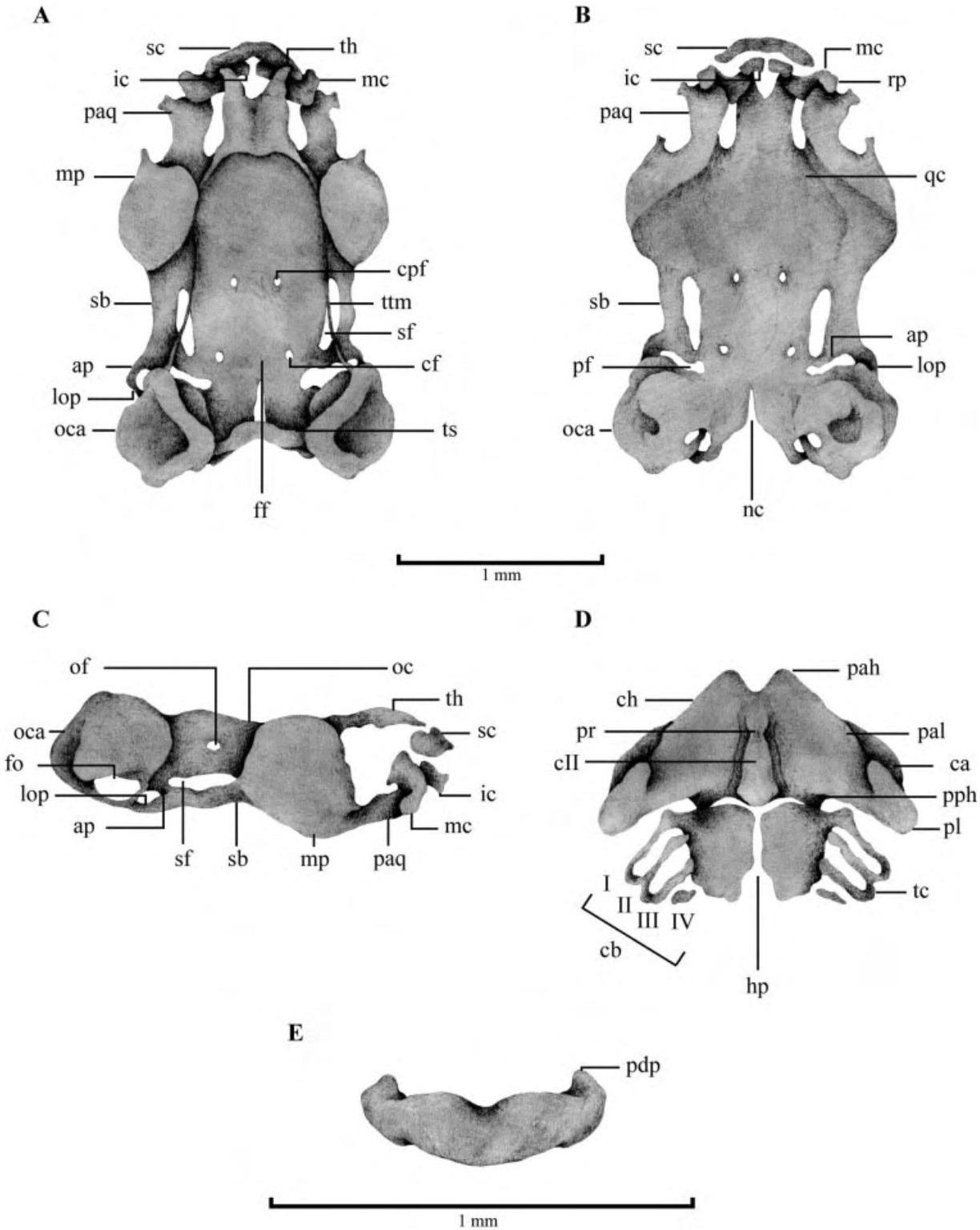

Fig. 6. *Hyla nana* Stage 31. Chondrocranium and hyobranchial apparatus. **A:** Chondrocranium, dorsal view. **B:** Chondrocranium, ventral view. **C:** Chondrocranium, lateral view. **D:** Hyobranchial apparatus, ventral view. **E:** Suprarostral cartilage, frontal view. ap, ascending process; ca, condylus articularis; cb, ceratobranchial (I–IV); cf, carotid foramen; ch, ceratohyal; cII, copula II; cpf, craniopalatine foramen; ff, frontoparietal fenestra; fo, fenestra ovalis; hp; hypobranchial plate; ic, infrarostral cartilage; lop, larval otic process; mc, Meckel's cartilage; mp, muscular process; nc, notochordal canal; oc, orbital cartilage; oca, otic capsules; of, optic foramen; pah, processus anterior hyalis; pal, processus anterolateralis; paq, pars articularis quadrati; pdp, processus dorsalis posterior; pf, prootic foramen; pl, processus lateralis; pph, processus posterior hyalis; pr, pars reuniens; qc, quadratocranial commissure; rp, retroarticular process; sb, subocular bar; sc, suprarostral cartilage; sf, subocular fenestra; tc, terminal commissure; th, trabecular horn; ts, tectum synoticum; ttm, taenia tecti marginalis.

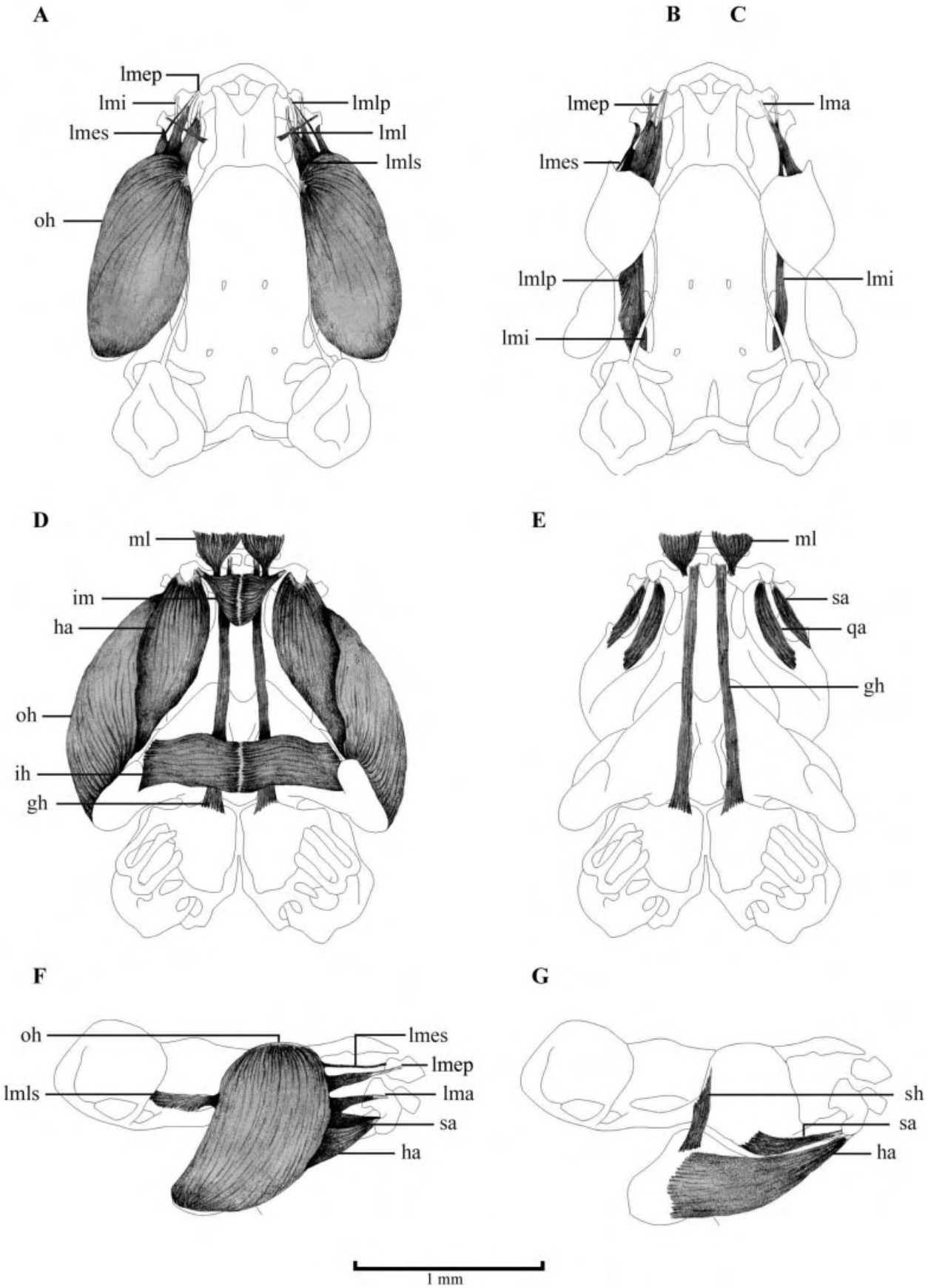
A
lmep
lmi
lmes
lmlp
lml
lmls
oh
B
C
lmep
lma
lmes
lmlp
lmi
lmi
D
ml
im
ha
oh
ih
gh
E
ml
sa
qa
gh
F
oh
lmes
lmep
lmls
lma
sa
ha
G
sh
sa
ha
1 mm

Figure 7

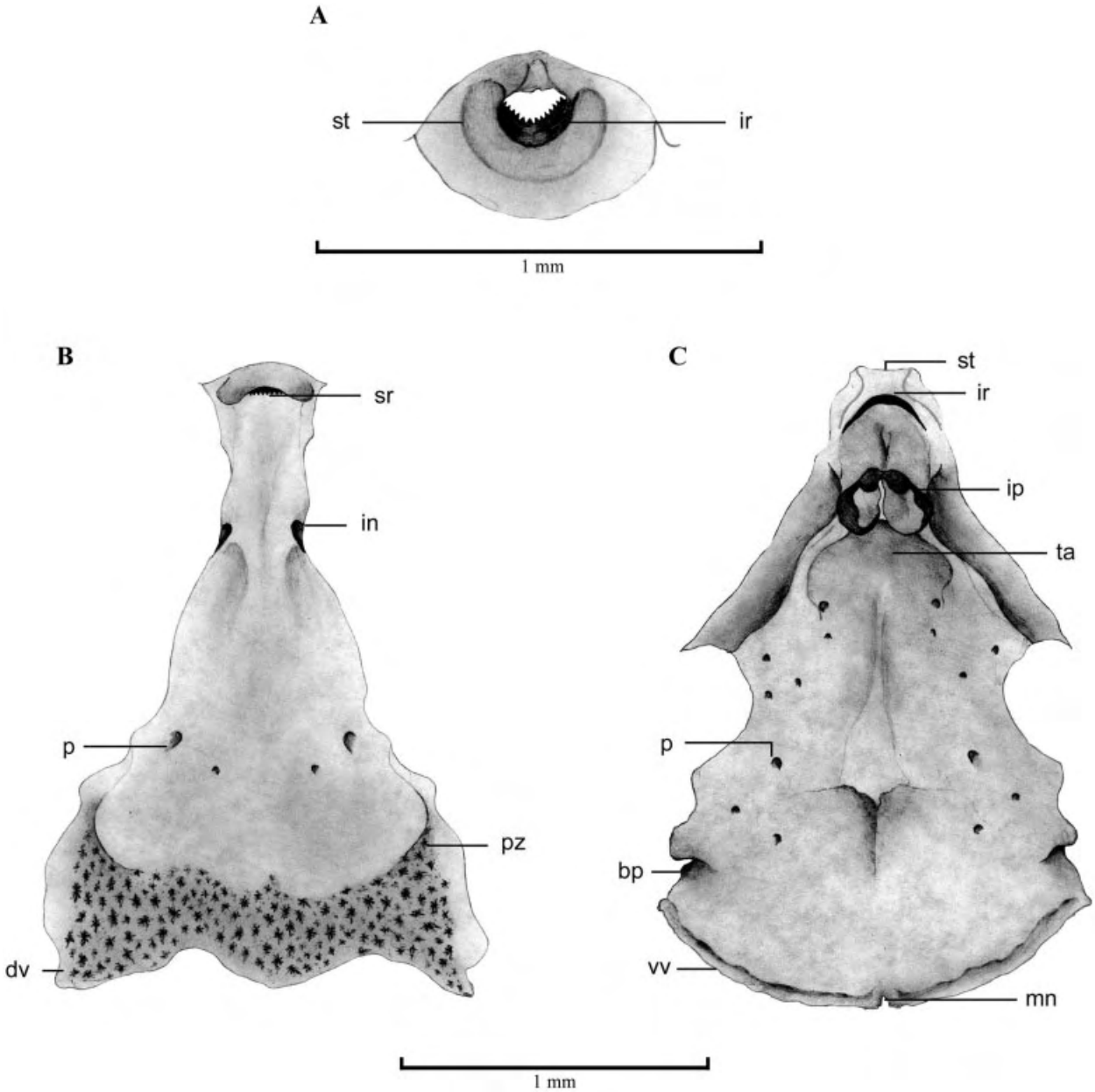

Fig. 8. *Hyla nana* Stage 31. Buccal apparatus and buccopharyngeal cavity. **A:** Oral disc. **B:** Buccal roof. **C:** Buccal floor. bp, buccal pocket; dv, dorsal velum; in, internal nares; ip, infralabial papillae; ir, infrarostrodont; mn, median notch; p, pustulation; pz, pigmented zone; sr, suprarostrodont; st, suctorial tube; ta, tonge anlage; vv, ventral velum.

Fig. 7. *Hyla nana* Stage 31. Musculature. **A:** Dorsal view, superficial plane. **B:** Dorsal view, middle plane. **C:** Dorsal view, deep plane. **D:** Ventral view, superficial plane. **E:** Ventral view, deep plane. **F:** Lateral view, superficial plane. **G:** Lateral view, deep plane. gh, *geniohyoideus*; ha, *hyoangularis*; ih, *interhyoideus*; im, *intermandibularis*; lma, *levator mandibulae articularis*; lmep, *levator mandibulae externus profundus*; lmes, *levator mandibulae externus superficialis*; lmi, *levator mandibulae internus*; lml, *levator mandibulae lateralis*; lmlp, *levator mandibulae longus profundus*; lmls, *levator mandibulae longus superficialis*; ml, *mandibulolabialis*; oh, *orbitohyoideus*; qa, *quadratoaularis*; sa, *suspensorioangularis*; sh, *suspensoriohyoideus*.

Among the taxa studied, the most marked differences involve the suprarostral cartilages, palatoquadrates, orbital cartilages, and the floor of the cranial cavity. Regarding the suprarostral cartilages, two of the four states mentioned by Fabrezi and Lavilla (1992) for that character are present: a simple corpus and alae, which are fused but still recognizable (*Scinax nasicus*), and corpus and alae fused and indistinguishable (*Hyla nana*). How differences in suprarostral cartilages correlate with the feeding mechanism has not yet been determined.

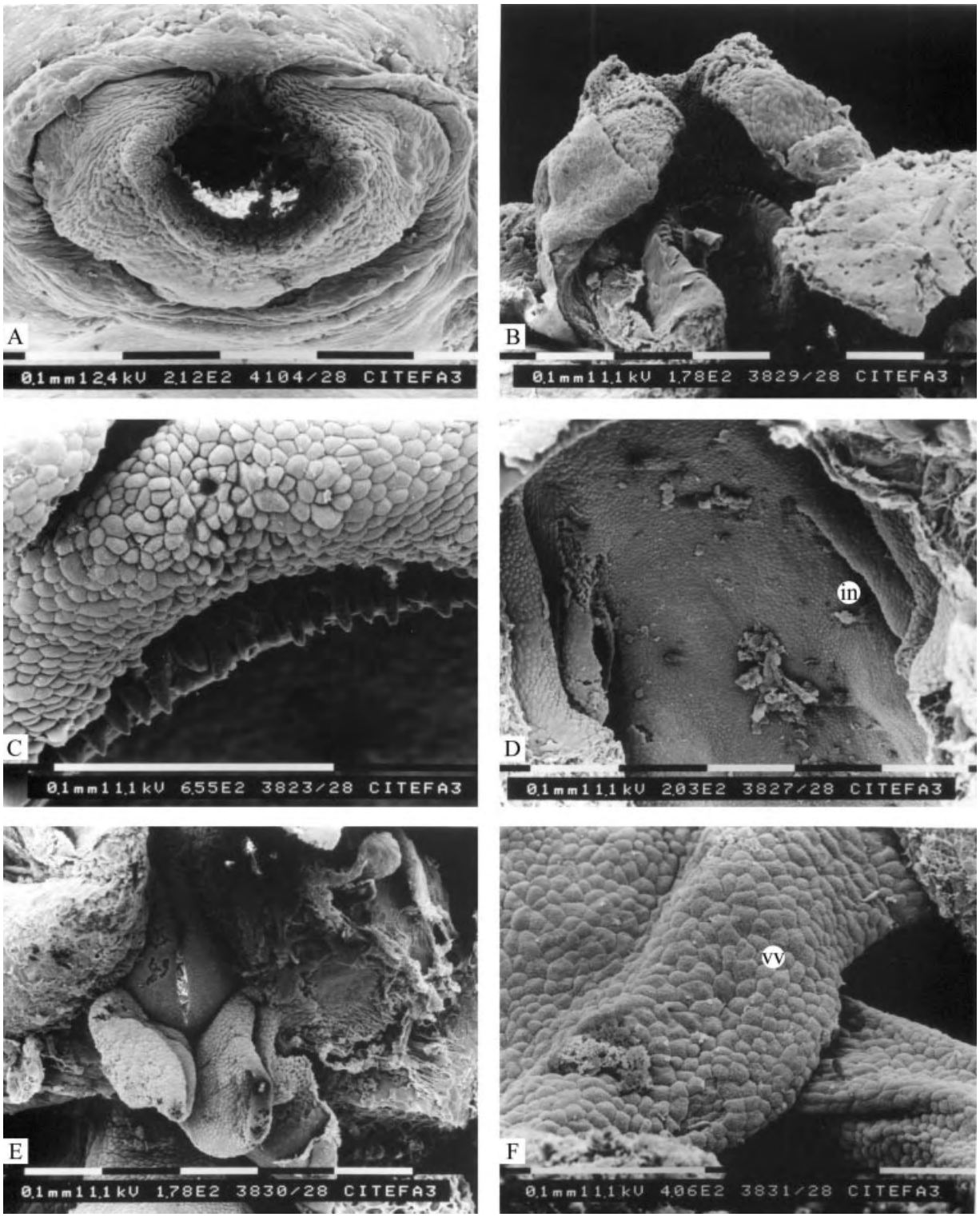

Fig. 9. *Hyla nana* Stage 31. Micrographs of buccal apparatus and buccopharyngeal cavity. **A:** Suctorial tube, frontal view. **B:** Suctorial tube and infrarostrodont, dorsal view. **C:** Detail of suprarostrodont. **D:** Detail of internal nares. **E:** Detail of infralabial papillae. **F:** Detail of ventral velum lacking of glandular zone. in, internal nares; vv, ventral velum.

Another variable character is the palatoquadrate, similar to that of most tadpoles in *Scinax nasicus* but with a peculiar quadrangular muscular process in *H. nana*. The muscular process and the posterior region of the subocular bar have clear trophic correlations in tadpoles because they serve as the attach-

ment site for muscles depressors of the larval buccal pump and adductors for jaw cartilages (Gradwell, 1968; Wassersug and Hoff, 1979). Furthermore, a recent study showed that, during ontogeny, measurements associated with these structures indicate scale with either isometry or positive allometry and a distinct expansion or widening of both of these structures with increasing size is evident. A better appreciation of the functional significance of morphological variation in palatoquadrate morphology is needed (Larson, 2002).

The orbital cartilage and the floor of the cranial cavity differ in the extent of chondrification in the two species. According to Haas (1996a) and Haas and Richards (1998), in some neotropical hylids the anterior braincase walls and the nasal septum develop late in larval life or just at metamorphosis, whereas in others these structures chondrify early. Development of these structures in *Hyla nana* seems to be delayed relative to that of *Scinax nasicus*, because in the same developmental period, *S. nasicus* has a well-developed orbital cartilage and braincase floor, whereas *H. nana* does not.

The hyobranchial apparatus shows profound morphological variation, and *Hyla nana* has the most divergent traits, relative to the generalized pond-type larva (see Table 1). In *H. nana*, the ceratohyal and hypobranchial areas are much larger than the ceratobranchials area. The small angle α reflects a difference in the orientation of the ceratohyals, the lateral edges of which are rotated posteriorly. The high value of the lever–arm ratio reflects the greater width of the lateral portion of the ceratohyal relative to its total width. Wassersug and Hoff (1979) demonstrated that high values of this ratio are characteristic of macrophagous larvae. The values calculated for *H. nana* exceed those of *H. microcephala* (which possesses a hyobranchial apparatus of similar configuration, according to Haas, 1996a) and to *Spea bombifrons*, a macrophagous carnivore. Values calculated for *Scinax nasicus* are the usual ones for generalized pond-type larvae (Haas and Richards, 1998; Wassersug and Hoff, 1979).

Morphometric differences in the hyobranchial apparatuses are summarized in Figure 10, which shows the relative larger size of the ceratohyal area and the posterior rotation of its lateral edges in *Hyla nana* tadpoles. The Cartesian deformation method has controversial applications. Fink and Zelditch (1995) employed it to detect taxonomical relationships among various species of a genus of piranhas (Teleostei). Rohlf (1998b) and Zelditch et al. (1998) discussed the validity of conclusions that concern phylogeny. Here, the method is applied only as a comparison of shapes, without establishing phylogenetic relations from the differences observed.

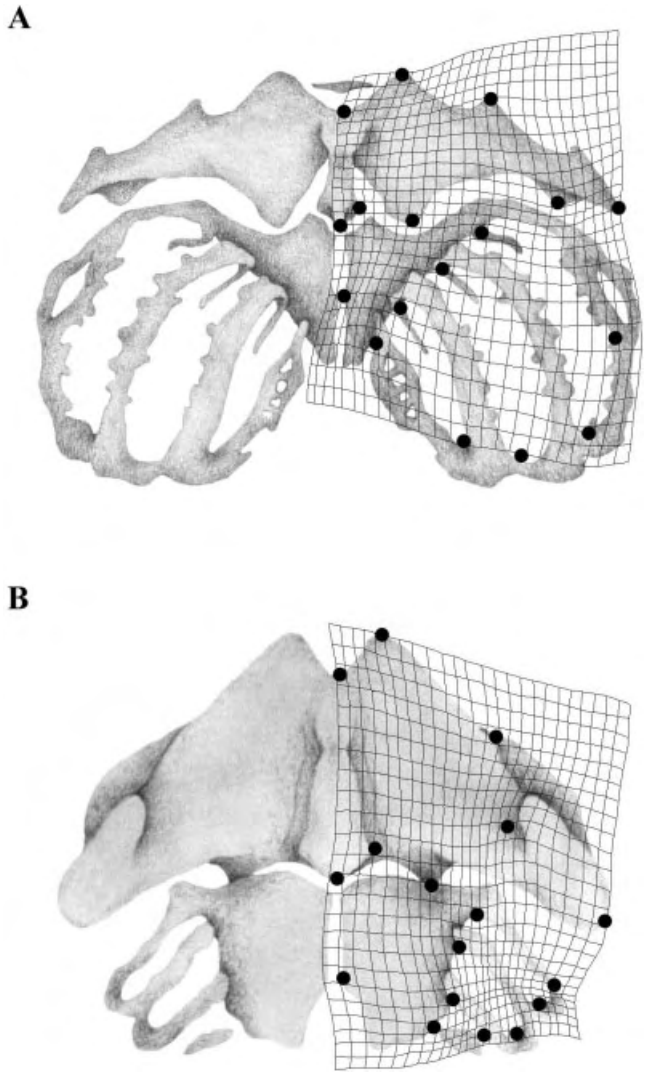

Fig. 10. Comparison of shapes of hyobranchial apparatuses of two species, applying thin-plate splines method. Drawings are not in scale. **A:** *Scinax nasicus.* **B:** *Hyla nana.*

Musculature

Cannatella (1999) compiled the available information on the mandibular and hyoid musculature of anuran larvae. Haas (2001) proposed a different nomenclature for the muscles of the mandibular group, based on the homologies of larval and adult musculature of anurans, and that of anurans compared to that of salamanders and caecilians. The functions of some muscles are known, whereas those of others remain speculative (Haas and Richards, 1998). Larson and Reilly (2003) recorded electromyographic and kinematic data from six mandibular and hyoid muscles, providing a review of muscle function in tadpoles.

In the two species examined here there is some myological variation. The *M. levator mandibulae lateralis* usually is present in hylids (Haas and Rich-

ards, 1998; Haas, 2001, 2003). However, muscle location differs from that in *Hyla nana*, because the muscle extends from the extreme of the suprarostral cartilage to the anterolateral dorsal region of the pars articularis quadrati. In tadpoles of *H. microcephala* and Pseudinae, the muscle configuration resembles that of *H. nana* (Haas, 2003; Vera Candioti, 2004, and pers. obs.). Such a configuration of the *M. l.m. lateralis* would dilate the nasal sac (Haas, 2003).

Another muscle that varies among anuran larvae is the *M. mandibulolabialis* (Carr and Altig, 1991). In the species with labial teeth, fibers insert behind each tooth ridge and contraction causes the keratodonts to rotate distally (dorsally in the upper labium and ventrally in the lower labium). Thus, the keratodonts orient at an angle that allows them to come in contact with the substrate (Taylor and Altig, 1996). According to Carr and Altig (1991), some species without labial teeth (i.e., *Hyla ebracatta*) have a well-developed *M. mandibulolabialis*, although the muscle is represented only by the inferior slip. This is also the case with *H. nana* tadpoles, in which the *M. mandibulolabialis* diverges from the general pattern, since it inserts in the skin of the suctorial tube. An electromyographic study would be necessary in order to establish the function of *M. mandibulolabialis* in these tadpoles, but it might be inferred that this muscle would be involved in the movements of the suctorial tube, at least in retraction.

The sizes of some of the muscles clearly differ. Dry weights of *Hyla nana* muscles are disproportionately greater than those of *Scinax nasicus,* particularly the *M. orbitohyoideus* (Table 2). Satel and Wassersug (1981) studied the IH/OH ratio in various species of anuran larvae. Rather than using the dry weights to quantify the ratio, these authors compared the cross sectional area of each muscle. Nonetheless, the interpretation remains the same; values near or exceeding 1 indicate a large *M. interhyoideus*, and thus large forces invested in raising the buccal floor for propelling water from the buccal cavity through the gill filters. This is characteristic of microphagous larvae. Conversely, low values, such as those of *H. nana*, reflect the presence of a large *M. orbitohyoideus*. This occurs in macrophagous and suctorial larvae because they need a powerful burst of suction, the former to retain large and moving prey, and the latter to adhere to substrate. Mean values are characteristic of generalist larvae.

The opening and closing mechanism of the jaw for gill irrigation was described by Gradwell (1972). De Jongh (1968) considered the musculoskeletal movements of this process and the feeding process to be essentially the same. According to Gradwell, the opening of the jaw occurs in three sequential phases, fundamentally by the lowering of the cartilages of the inferior jaw by muscular action. The first phase, narrow opening, is effected by the contraction of *M. quadratoangularis*. The second phase, wide opening, recruits the *M. hyoangularis*, whereas the third stage, protrusion, also requires the participation of the *M. suspensorioangularis*. Larson and Reilly (2003) confirmed many of the results of Gradwell and add new data about the *M. l.m. longus superficialis*, responsible for closing the mouth. The development of the *M. hyoangularis* in *Hyla nana* may allow these larvae to produce additional force to reach a still wider opening of the buccal orifice in order to ingest large prey.

Buccal Apparatus and Buccopharyngeal Cavity

The structure of the mouth and buccal cavities of anuran larvae are highly adaptive and correlated with feeding ecology (Wassersug, 1976b). The absence of keratodonts excludes surface-rasping in some species and, conversely, the presence of robust rostrodonts and multiple tooth rows signals food removal by surface rasping.

Wassersug (1980) found a general trend toward the progressive loss of the tooth rows associated with a reduction in the sorting and trapping structures of the buccopharyngeal cavity in the genus *Hyla*. Thus, species can be grouped by common features of their mouths and buccal cavities. To a large extent, these groups coincide with those described by Duellman (1970) for neotropical hylids, based on osteology, external structure, coloration, and adult calls.

Tadpoles of the *Hyla microcephala* group present the most extreme reduction, with a toothless mouth and the buccopharyngeal cavity devoid of sieving structures. According to Duellman (1970), in Middle America this group comprises four species: *H. microcephala, H. phlebodes, H. sartori,* and *H. robertmertensi*. In an earlier work (Duellman and Fouquette, 1968), *H. nana* was proposed to be related, although it was not added to the group due to lack of knowledge of the larvae. More recent studies added South American species, including *H. nana*, to the group (Duellman, 1972; Hero, 1990; Cruz and Dias, 1991).

The oral disc shape, the arrangement and shape of the keratodonts, and the extent of development of the rostrodonts of *Scinax nasicus* tadpoles resemble those of larvae of the local congeneric species *S. fuscovarius* (Echeverría and Montanelli, 1992), *S. eringiophilus* (Echeverría, 1997a), and *S. acuminatus* (Sandoval, 2000). Langone and Cardoso (1997) reported a labial tooth row formula of (1)(1-1)/(3) in *S. nasicus*. In the specimens examined in our work, as in Yanosky and Dixon (1997), all the larvae showed a divided P-1, making the corresponding LTRF (1)(1-1)/(1-1)(2).

Wassersug and Yamashita (2001) studied feeding kinematics in anuran larvae with a typical oral disc. They found that keratodonts help to hold the oral

disc and rostrodonts against the substrate and they are released from it in a definite order.

Other structures within the buccal cavity have well-known functions. Roof and floor arena papillae constitute a mesh that retains food particles and divides the water flux into two streams that run toward the gill filters at each side of the body (Kenny, 1969). A great number of papillae is associated with an efficient filtration capability at the level of the buccal cavity, whereas their absence presupposes either the lack of this capability or the presence of compensatory structures, such as dense gill filters (Wassersug, 1980).

The infralabial papillae would act as respiratory or sensory structures, or they might also have mechanical interactions with food and water currents. They move in concert with the jaws, erecting when the mouth is at maximum gape, and flicking caudally as the rostrodonts begin to close (Wassersug and Yamashita, 2001). Wassersug (1980) proposes that the globose papillae of *Hyla microcephala* and *H. phlebodes* would provide information on the position of the fragments of food in their buccal cavity. *Hyla nana* larvae possess similar globose infralabial papillae, with a presumably similar function.

The spurs found in *Scinax nasicus* tadpoles occur in other species. Faivovich (2002) analyzed the presence of this character within the genus *Scinax*. A possible function of assistance in the shredding food has been suggested (Wassersug, 1980).

The glandular zone of the buccopharyngeal cavity is a region the development of which varies depending on the ecological habits of the species. In general terms, the suspensivorous taxa possess a conspicuous glandular area either in the floor or the roof of the mouth, or in both. The secreted mucus aggregates and traps food particles removed from the water current. Conversely, larvae with other feeding modes lack secretory tissue (Wassersug, 1980). Of the two species studies, *Scinax nasicus* has conspicuous secretory pits in the floor of the mouth, whereas *Hyla nana* lacks any glandular zone.

Digestive Tract and Gut Content Analysis

The alimentary tract of anuran larvae has been studied by numerous authors (e.g., Barrington, 1946; Griffiths, 1961; Fox, 1984; Rada and Bello, 1988; Viertel and Richter, 1999). The general morphological pattern is that of an almost undifferentiated tube, consisting of an esophagus, a gastric region, and a long coiled intestine (Hourdry and Beaumont, 1985).

Michel de Cerasoulo and Terán (1991) correlated morphological features with different diets, recording the main differences in the cellular physiology. At a macroscopic level, longer digestive tracts are correlated with a mainly herbivorous diet, whereas shorter ones are associated with carnivorous diets (Noble, 1954; Altig and Kelly, 1974; Villée et al., 1987, in Martin et al., 1997). *Hyla nana* larvae have a more modified digestive tract (at a macroscopic level), with a conspicuous manicotto glandulare, and a shorter intestine than that of *Scinax nasicus* (Table 3).

The range of food particle size, as well as the qualitative composition of the diet in tadpoles, is highly variable, and is linked to food availability. However, Heyer (1973) and Inger (1986) mentioned interspecific variation in the sizes of food particles ingested. The latter classifies various sympatric larvae according to the size of the organisms they consume, mentioning three groups, according to the mean particle size ($\bar{x}$): macrophagous ($\bar{x} > 0.12$ mm), mesophagous ($\bar{x} = 0.074$ mm), and microphagous ($\bar{x} = 0.038$ mm). Following this criterion, *Scinax nasicus* larvae, with a mean size of particles ingested equal to 0.03 mm, would be situated among the microphagous forms. With regard to *Hyla nana*, there are alternate interpretations. The presence of oligochaeta would indicate a preference for the ingestion of large items ($\bar{x} = 1.87$ mm) and, consequently, a specialized macrophagous diet, whereas the appearance of smaller and numerous algae suggests an omnivorous feeding. However, the morphological features mentioned and the low total number of small organisms ingested, compared to that of *S. nasicus*, argue against the latter. Presumably, the presence of small items in the digestive tract is merely circumstantial (Tables 4, 5).

Although most anuran larvae are thought to feed indiscriminately, some studies postulate a possible resource selection, which would permit a differentiation in the trophic niche (Steinwascher and Travis, 1983; Taylor et al., 1995). A preliminary analysis of water samples from the site where the larvae were collected shows a smaller number of Euglenophyta in the environment than in the gut contents. This finding agrees with a study that reported a high assimilation of this kind of algae at the level of the distal zone of the intestine of another hylid larva, *Phyllomedusa hypochondrialis azurea* (Vera Candioti and Lajmanovich, 1998).

In summary, the morphological traits of *Scinax nasicus* larvae are associated with a microphagous-generalist regime. The buccal apparatus, the various groups of papillae and other sieving structures, the secretory pits, and the large branchial basket that supplies a large supporting area for the gill filters retain particles in the size range of 0.03–0.05 mm. These conclusions also find support in the mechanical features of the ceratohyals and associated musculature (low lever–arm ratio and similar development of levator and depressor muscles of the buccal floor). Although these tadpoles have been characterized as plankton feeders (Lajmanovich, 1997), they can also graze on substrate with their keratinized jaw sheaths and teeth, as evidenced by the food items in the digestive tract.

Hyla nana larvae, in contrast, have been considered macrophagous (Fabrezi and Lavilla, 1992). Many of the features mentioned in the descriptions are typical of larvae having this feeding mode. The absence of sieving structures in the buccopharyngeal cavity (papillae, ridges, etc.) and the reduction of the branchial baskets indicate that the filtration mechanism is poorly developed. The main method of food sieving is the diameter of the buccal orifice. How *H. nana* tadpoles use their unusual buccal apparatus has not yet been elucidated. Recently, Deban and Olson (2003) described the prey suction mechanism in *Hymenochirus boettgeri* larvae. Suction action is generated by hyobranchial movements, cranial elevation, and downward rotation of the lower jaw. These authors also point out several benefits of mouth protrusion during prey capture. A study focusing on food capture mechanisms in *H. nana* tadpoles has yet to be done. As previously mentioned, small planktonic organisms found inside the intestine might flow in along with the respiratory currents, and consequently be part of a secondary trophic spectrum in the tadpole. The main source of food is likely to be larger items. Gut contents and the relatively short length of the intestine indicate these larvae to be macrophagous carnivores. Further studies on enzymes will bring unequivocal conclusions in this sense. Other aspects indicating macrophagy are the high values of the ceratohyal area and the lever–arm ratio. This, along with a large muscular process of the palatoquadrate, affords a large attachment site for the *M. orbitohyoideus*, which reaches a considerable development (low IH/OH ratio). Even excluding oligochaetes, the mean size of the food particles ingested is bigger than that in *Scinax nasicus* larvae. Given the absence of an efficient retention mechanism (papillae, mucus, dense gill filters), the smaller particles would be resuspended.

Finally, considering spatial distribution within the body of water, it is clear that *Scinax nasicus* and *Hyla nana* occupy different microhabitats (Lajmanovich, 1997, 1998). Our work adds a separation at a trophic level, reflected in both size and type of the items consumed.

ACKNOWLEDGMENTS

We thank D. Jiménez from Centro de Investigaciones Técnicas de las Fuerzas Armadas (CITEFA) for technical assistance, and N. Vera Candioti and K. Warkentin for help in English translation and critical comments. Two anonymous reviewers improved earlier versions of the manuscript.

LITERATURE CITED

Altig R, Johnston GF. 1989. Guilds of anuran larvae: relationships among developmental modes, morphologies and habits. Herpetol Monogr 2:81–109.

Altig R, Kelly P. 1974. Indices of feeding in anuran tadpoles as indicated by gut characteristic. Herpetologica 30:200–203.

Altig R, McDiarmid R. 1999. Body plan. Development and morphology. In: McDiarmid RW, Altig R, editors. Tadpoles. The biology of anuran larvae. Chicago: Univ Chicago Press. p 24–51.

Barrington EJ. 1946. The delayed development of the stomach in the frog (*Rana temporaria*) and the toad (*Bufo bufo*). Proc Zool Soc Lond 116:1–21.

Cannatella D. 1999. Architecture. Cranial and axial musculoskeleton. In: McDiarmid RW, Altig R, editors. Tadpoles. The biology of anuran larvae. Chicago: Univ Chicago Press. p 52–91.

Carr KM, Altig R. 1991. Oral disc muscles of anuran tadpoles. J Morphol 208:271–277.

Cruz CA, Dias AG. 1991. Girinos do grupo "*microcephala*" do Estado do Rio de Janeiro. Rev Brasil Zool 7:679–683.

Deban SM, Olson WM. 2003. Suction feeding by a tiny predatory tadpole. Nature 420:41–42.

De Jongh HJ. 1968. Functional morphology of the jaw apparatus of larval and metamorphosing *Rana temporaria* L. Neth J Zool 18:1–103.

Duellman WE. 1970. The hylid frogs of Middle America. Museum of Natural History. Univ Kansas Monogr 1–173.

Duellman WE. 1972. The systematic status and life history of *Hyla rhodopepla* Günther. Herpetologica 28:369–375.

Duellman WE, Fouquette MJ. 1968. Middle american frogs of the *Hyla microcephala* group. Univ Kansas Publ Mus Nat Hist 17:517–557.

Echeverría DD. 1992a. Microscopía electrónica de barrido del aparato bucal de las larvas de *Melanophryniscus stelzneri* (Weyembergh, 1875) (Anura, Bufonidae). Alytes 10:137–143.

Echeverría DD. 1992b. Microscopía electrónica de barrido del aparato bucal de la larva de *Hyla pulchella pulchella* (Anura: Hylidae). Cuad Herpetol 7:24–29.

Echeverría DD. 1997a. Microscopía electrónica de barrido del aparato bucal y de la cavidad oral de la larva de *Hyla strigilata eringiophila* Gallardo, 1961 (Anura, Hylidae) con comentarios que facilitan su identificación durante la metamorfosis y postmetamorfosis. Physis Sec B 54:1–6.

Echeverría DD. 1997b. Microanatomy of the buccal apparatus and oral cavity of *Hyla minuta* Peters, 1872 larvae (Anura, Hylidae), with data on feeding habits. Alytes 15:26–36.

Echeverría DD. 1998. Microanatomía del aparato bucal y de la cavidad oral de la larva de *Bufo fernandezae* Gallardo, 1957 (Anura, Bufonidae), con comentarios acerca de la coloración in vivo y la anatomía externa. Alytes 16:50–60.

Echeverría DD, Montanelli SB. 1992. Estereomorfología del aparato bucal y de la cavidad oral de las larvas de *Ololygon fuscovaria* (Lutz, 1925) (Anura, Hylidae). Rev Mus Argent Cienc Nat Bernardino Rivadavia (Zool) 16:3–13.

Fabrezi M. 1988. Metamorfosis en *Pleurodema borellii* (Anura: Leptodactylidae). Estudio del neurocráneo, esqueleto visceral y musculatura asociada. Thesis, Universidad Nacional de Tucumán, Tucumán, Argentina.

Fabrezi M, Lavilla EO. 1992. Estructura del condrocráneo y aparato hiobranquial en larvas de algunos hílidos neotropicales (Anura: Hylidae). Acta Zool Lilloana 41:155–164.

Fabrezi M, Vera R. 1997. Caracterización morfológica de larvas de anuros del noroeste argentino. Cuad Herpetol 11:37–49.

Faivovich J. 2002. A cladistic analysis of *Scinax* (Anura: Hylidae). Cladistics 18:367–393.

Fink WL, Zelditch ML. 1995. Phylogenetic analysis of ontogenetic shape transformations: a reassessment of the piranha genus *Pygocentrus* (Teleostei). Syst Biol 44:343–360.

Fox H. 1984. Amphibian morphogenesis. Totowa, NJ: Humana Press.

Gosner KL. 1960. A simplified table for staging anuran embryos and larvae with notes on identification. Herpetologica 16:183–190.

Gradwell N. 1968. The jaw and hyoidean mechanism of the bullfrog tadpole during aqueous ventilation. Can J Zool 46:1041–1052.

Gradwell N. 1972. Gill irrigation in *Rana catesbeiana*. I. On the anatomical basis. Can J Zool 50:481–499.

Griffiths I. 1961. The form and function of the fore-gut in anuran larvae (Amphibia, Salientia) with particular reference to the manicotto glandulare. Proc Zool Soc Lond 137:249–283.

Haas A. 1996a. Das larvale Cranium von *Gastrotheca riobambae* und seine Metamorphose (Amphibia, Anura, Hylidae). Verh Naturwiss Ver Hamburg 36:33–162.

Haas A. 1996b. Nonfeeding and feeding tadpoles of hemiphractinae frogs: larval head morphology, heterochrony and systematics of *Flectonotus goeldii* (Amphibia, Anura, Hylidae). J Zool Syst Evol Res 34:163–171.

Haas A. 2001. Mandibular arches musculature of anuran tadpoles, with comments on homologies of amphibian jaw muscles. J Morphol 247:1–33.

Haas A. 2003. Phylogeny of frogs as inferred from primarily larval characters (Amphibia: Anura). Cladistics 19:23–89.

Haas A, Richards SJ. 1998. Correlations of cranial morphology, ecology and evolution in Australian suctorial tadpoles of the genera *Litoria* and *Nyctimystes* (Amphibia: Anura: Hylidae: Pelodryadinae). J Morphol 238:109–141.

Hero JM. 1990. An illustrated key to tadpoles occurring in the Central Amazon rainforest, Manaus, Amazonas, Brasil. Amazoniana 11:201–262.

Heyer WR. 1973. Ecological interactions of frog larvae at a seasonal tropical location in Thailand. J Herpetol 7:337–361.

Hourdry J, Beaumont A. 1985. Les mètamorphoses des amphibiens. Paris: Masson.

Inger R. 1986. Diets of tadpoles living in a Bornean rain forest. Alytes 5:153–164.

Iordansky N. 1992. Jaw muscles of the Urodela and Anura: some features of development, functions and homology. Zool Jahrb Anat 122:225–232.

Kamat ND. 1962. On the intestinal contents of tadpoles and algae of small ponds. Curr Sci 321:300–310.

Kenny JS. 1969. Feeding mechanisms in anuran larvae. J Zool 157:225–246.

Lackey JB. 1938. The manipulation and counting of river plankton and changes in some organisms due to formaline preservation. Pub Health Rep Washington 53:20–80.

Lajmanovich RC. 1994. Contribución al conocimiento de la alimentación de larvas de la rana criolla *Leptodactylus ocellatus* (Amphibia, Leptodactylidae) en el Paraná medio, Argentina. Stud Neotrop Fauna Environ 29:55–61.

Lajmanovich RC. 1997. Alimentación de larvas de anuros en ambientes temporales del sistema del Río Paraná, Argentina. Doñana Acta Vert 24:191–202.

Lajmanovich RC. 1998. Estructura de una comunidad larval de anuros vinculada al ecosistema del Río Paraná medio. PhD Thesis 1000-36279, Facultad de Ciencias Naturales y Museo, Universidad Nacional de La Plata, La Plata, Argentina.

Lajmanovich RC, Fernandez V. 1995. Alimentación de larvas de *Bufo arenarum* Hensel 1867 (Amphibia: Bufonidae) en ambientes del Río Paraná. Bol Mus Nac Hist Nat Chile 45:7–18.

Lajmanovich RC, Izaguirre MF, Vera Candioti MF, Casco VH. 2000. Unique structural pattern of the manicotto glandulare of *Hyla nana* tadpoles (Anura: Hylidae). Amphibia-Reptilia 21: 1–6.

Langone JA, Cardoso AJ. 1997. Morfología larval externa de *Scinax eringhiophila* (Gallardo, 1961) (Amphibia, Anura, Hylidae). Comun Zool Mus Hist Nat Montevideo 187:1–10.

Larson PM. 2002. Chondrocranial development in larval *Rana sylvatica* (Anura: Ranidae): morphometric analysis of cranial allometry and ontogenetic shape change. J Morphol 252:131–144.

Larson PM, Reilly SM. 2003. Functional morphology of feeding and gill irrigation in the anuran tadpole: electromyography and muscle function in larval *Rana catesbeiana*. J Morphol 255: 202–214.

Lavilla EO. 1983. Consideraciones sobre el régimen ileófago en larvas de *Hyla pulchella andina* de la Sierra de Metán. Bol Asoc Herpetol Argentina 1:1–3.

Lavilla EO. 1987. La larva de *Rhinoderma darwinii* D. and B. (Anura: Rhinodermatidae). Acta Zool Lilloana 39:81–88.

Lavilla EO. 1990. The tadpole of *Hyla nana* (Anura: Hylidae). J Herpetol 24:207–209.

Lavilla EO, Fabrezi M. 1987. Anatomía de larvas de *Hyla pulchella andina* (Anura: Hylidae). Physis 45:77–82.

Martin M, Gimenez H, Parisi S. 1997. Estudio morfológico e histoquímico del tracto digestivo y páncreas de larvas premetamórficas de *Hyla pulchella cordobae*, sometidas a diferentes dietas. Nat Neotrop 28:75–83.

McDiarmid RW, Altig R. 1999. Tadpoles. The biology of anuran larvae. Chicago: Univ Chicago Press.

Michel de Cerasoulo A, Terán H. 1991. Aspectos histoquímicos del tracto digestivo larval de *Gastrotheca gracilis* Laurent, en relación con la alimentación. Acta Zool Lilloana 40:69–81.

Noble K. 1954. The biology of Amphibia. New York: Dover.

Rada D, Bello R. 1988. Morfología e histología del sistema digestivo de la larva de *Pseudis paradoxus caribensis* Gallardo, 1961 (Amphibia, Pseudidae). Iheringia Ser Misc 2:89–91.

Rohlf FJ. 1997. TpsSplin v. 1.15. Department of Ecology and Evolution, Stony Brook, NY: SUNY.

Rohlf FJ. 1998a. TpsDig v. 1.18. Department of Ecology and Evolution, Stony Brook, NY: SUNY.

Rohlf FJ. 1998b. On applications of geometric morphometrics to studies of ontogeny and phylogeny. Syst Biol 47:147–158.

Rohlf FJ, Bookstein FL. 1990. Proceedings of the Michigan morphometrics workshop. Univ Michigan Mus Zool Spec Publ 2.

Ruibal R, Thomas E. 1988. The obligate carnivorous larvae of the frog *Lepidobatrachus laevis* (Leptodactylidae). Copeia 3:591–604.

Sandoval MT. 2000. Microanatomía del disco oral y cavidad bucofaríngea de la larva de *Scinax acuminatus* (Anura: Hylidae). Facena 16:93–101.

Satel S, Wassersug RJ. 1981. On the relative sizes of buccal floor depressor and elevator musculature in tadpoles. Copeia 1:129–137.

Savage M. 1955. The ingestive, digestive and respiratory systems of the microhylid tadpole *Hypopachus aguae*. Copeia 2:120–127.

Seale D, Wassersug RJ. 1979. Suspension feeding dynamics of anuran larvae related to their functional morphology. Oecologia 39:259–272.

Steinwascher K, Travis J. 1983. Influence of food quality and quantity on early larval growth of two anuran. Copeia 1:238–242.

Taylor CL, Altig R. 1996. Oral disc kinematics of four rheophilous anuran tadpoles. Herpetol Nat Hist 3:101–106.

Taylor CL, Altig R, Boyle CR. 1995. Can anuran tadpoles choose among foods that vary in quality? Alytes 13:81–86.

Terán HR, Michel de Cerasuolo A. 1988. Estudio histomorfológico del tracto digestivo larval de *Gastrotheca gracilis* Laurent (Anura: Hylidae). Neotropica 36:115–123.

Van Dijk DE. 1966. Systematics and field keys to the families, genera and described species of the Southern african anuran tadpoles. Ann Natal Mus 18:231–286.

Vera Candioti MF, Lajmanovich RC. 1998. Contribución al conocimiento de la alimentación de *Phyllomedusa hypocondrialis azurea* Cope, 1962 (Anura: Hylidae) en ambientes temporales de la Provincia de Santa Fe, Argentina. Bol Soc Biol Concepción Chile 69:203–209.

Vera Candioti MF. 2004. Morphology of premetamorphic larvae of *Lysapsus limellus* (Anura: Pseudinae). Amphibia-Reptilia 25:41–54.

Viertel B. 1982. The oral cavities of central European anuran larvae (Amphibia): morphology, ontogenesis and generic diagnosis. Amphibia-Reptilia 4:327–360.

Viertel B, Richter S. 1999. Anatomy. Viscera and endocrines. In: McDiarmid RW, Altig R, editors. Tadpoles. The biology of anuran larvae. Chicago: Univ Chicago Press. p 92–148.

Villée C, Walker W, Barnes R. 1987. Nutrición animal. In: Villée C, Walker W, Barnes R, editors. Zoología. México: Interamericana. p 147.

Wassersug RJ. 1976a. A procedure for differential staining of cartilage and bone in whole formalin fixed vertebrates. Stain Tech 51:131–134.

Wassersug RJ. 1976b. Oral morphology of anuran larvae: terminology and general description. Occ Pap Mus Nat Hist Univ Kansas 48:1–23.

Wassersug RJ. 1980. Internal oral features of larvae from eight anuran families. Functional, systematics, evolutionary and ecological considerations. Univ Kansas Misc Publ Mus Nat Hist 65:1–146.

Wassersug RJ, Hoff K. 1979. A comparative study of the buccal pumping mechanism of tadpoles. Biol J Linn Soc 12:225–259.

Wassersug RJ, Yamashita M. 2001. Plasticity and constraints on feeding kinematics in anuran larvae. Comp Biochem Physiol A 131:183–195.

Wilcox D, Dove B, McDavid D, Greer D. 1995. UTHSCSA image tool, v. 1.28. San Antonio: University of Texas Health Science Center.

Yanosky A, Dixon JR. 1997. Ecological notes and population estimates on the common frog *Scinax nasica* Cope, 1862 (Anura: Hylidae) from northeastern Argentina. Bull Maryland Herpetol Soc 33:126–138.

Zelditch ML, Fink WL, Swiderski DL, Lundrigan BL. 1998. On applications of geometric morphometrics to studies of ontogeny and phylogeny: a reply to Rohlf. Syst Biol 47:159–167.