

# Chirocentrids as engrauloids: evidence from suspensorium, branchial arches, and infraorbital bones (Clupeomorpha, Teleostei)

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The Chirocentridae is a family of highly specialized large predatory clupeomorphs composed of two species from coastal waters of the Indian and western Pacific Oceans. Peculiarities of the anatomy of these fishes have puzzled ichthyologists who attempted to resolve their phylogenetic relationships. Despite controversy, it is currently accepted that the Chirocentridae is a family of Clupeiformes, included with the Clupeidae in the superfamily Clupeiodea. New data support an alternative hypothesis. Seven previously unreported derived character states from the suspensorium, branchial arches, and infraorbitals strongly indicate a hitherto unsuspected sister group relationship between the Chirocentridae and Engrauloidea, which comprises approximately 140 species of the commercially important fishes known as anchovies. These are character states: (1) the anterior margin of metapterygoid located anterior to the quadrate; (2) the ventral limb of hyomandibula and quadrate not separated by the metapterygoid; (3) the posterodorsal margin of metapterygoid in line with the condyle of articulation of the hyomandibula with the opercle; (4) the presence of a laminar outgrowth of the anterior margin of the quadrate; (5) the endochondral portion of the quadrate in the shape of an isosceles triangle; (6) the presence and arrangement of autogenous tooth plates on ceratobranchials 1 to 3; and (7) posterior region of infraorbital 1 well developed and extending along the ventral margin of infraorbital 2. Three of those character states are further modified and hypothesized as synapomorphies of the Engrauloidea: (1') a substantial portion of the metapterygoid situated anterodorsal to the quadrate, (2') articulation between the ventral limb of the hyomandibula and the quadrate, and (7') infraorbitals 1 and 3 articulating by means of a well-developed laminar process of the posterior region of infraorbital 1. The separation of the dorsal, paired elements of the branchial arches of the Chirocentridae and representative Engrauloidea is apomorphic within the Clupeiodei, and constitutes circumstantial evidence for the sister group relationship between those clades. Microphagy within the Engrauloidea is secondary, homoplastic to the same condition present in other clades of the Clupeiformes. The decomposition of character complexes into discrete morphological characters and its use in phylogenetic inference is discussed. The sister group relationship between the Chirocentridae and Engrauloidea renders the Clupeiodea paraphyletic. A new classification of the Clupeiodei, with the inclusion of the Chirocentridae in the Engrauloidea, is proposed. © 2009 The Linnean Society of London, *Zoological Journal of the Linnean Society*, 2009, 156, 363–383.

**ADDITIONAL KEYWORDS:** anchovies – character complex – character interdependence – Chirocentridae – Clupeidae – Clupeiodei – Engrauloidea – Pristigasteroidea.

## INTRODUCTION

The Chirocentridae is a family of Clupeiformes comprising two morphologically similar species, *Chirocentrus*

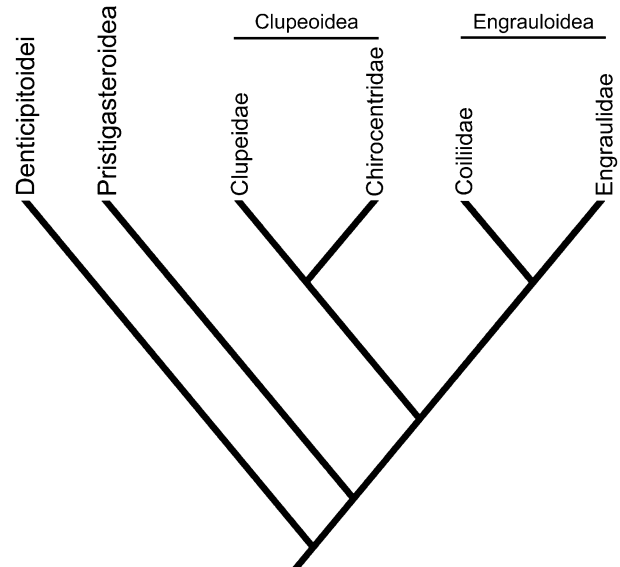
*trus dorab* and *Chirocentrus nudus* from coastal waters of the Indian and western Pacific Oceans. Whereas most clupeiforms are small to medium sized fishes forming large schools and feeding on small planktonic animals, chirocentrids are large (up to 1 m standard length) and probably solitary pelagic

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inshore predators of small fishes (Whitehead, 1985; James, 1988; Munroe, Nizinski & Wongratana, 1999). The anatomy of chirocentrids reflects a host of adaptations to a specialized predaceous life. These include the possession of an upward-directed mouth with enlarged fang-like canine teeth on both jaws, which themselves are massive and strongly articulated to the cranium, a relatively well-developed pharyngeal dentition, and a large and highly forked caudal fin (Fowler, 1931; Whitehead, 1985; Munroe *et al.*, 1999).

The phylogenetic position of the Chirocentridae has puzzled ichthyologists since the description of *Chirocentrus dorab* and its original assignment to the genus *Clupea* by Forskål (1775). The uniqueness of chirocentrids led some past authors to hypothesize them to be the sole living representatives of the Ichthyodectiformes (e.g. Woodward, 1912, 1942; Gregory, 1933; Saint-Seine, 1949; Bardack, 1965), otherwise a group composed of large marine predaceous Mesozoic basal teleosts (Patterson & Rosen, 1977; de Pinna, 1996; Arratia, 1999; Nelson, 2006). Evidence strongly indicating that the Chirocentridae is rather a family of the Clupeiformes was presented and discussed by Ridewood (1904), Gosline (1960), Whitehead (1963), Cavender (1966), Greenwood *et al.* (1966), and Nelson (1970). These authors, nevertheless, did not resolve the position of the family within the order.

The Clupeiformes, with about 360 living species, is divided into the Denticipitoidei and the Clupeoidei (Grande, 1985; Di Dario, 2004; Di Dario & de Pinna, 2006; Nelson, 2006; Lavoué *et al.*, 2007). *Denticeps clupeoides* is the sole recent representative of the Denticipitoidei (Greenwood, 1968; Grande, 1985; Di Dario & de Pinna, 2006). According to Nelson (1970), the Clupeoidei comprises four superfamilies: Clupeoidea (approximately 180 species), Engrauloidea (approximately 140 species), Pristigasteroidea (approximately 35 species), and Chirocentroidea. Most currently accepted suprageneric taxa of the Clupeoidei are poorly supported in terms of numbers of synapomorphies, and some particularly large groups are not corroborated as monophyletic (Grande, 1985; Lavoué *et al.*, 2007; Li & Ortí, 2007). Grande (1985) and Patterson & Johnson (1995) identified two apomorphic character states shared by the Chirocentroidea and Clupeoidea, the high ratio of ribs to preural vertebrae, and the fusion of epicentrals with anterior ribs. Both clades were grouped in the superfamily Clupeoidea, as the families Clupeidae and Chirocentridae (Grande, 1985; Fig. 1). Di Dario (2002) suggested that the Clupeoidea and Engrauloidea are sister groups on the basis of three putative apomorphic character states, the presence of cartilage chevrons at the tips of epicentrals, the posteriorly directed parapophyses of the second vertebra, and the



**Figure 1.** Cladogram showing the relationships in the Clupeiformes after Grande (1985), Patterson & Johnson (1995) and Di Dario (2002).

interzygapophyseal articulation. Di Dario (2002) also offered counter-evidence to that hypothesized relationship, viz., the apomorphic occurrence of the gongyloid cartilage in the branchial arches of the Engrauloidea and Pristigasteroidea.

The hypotheses of monophyly of the Engrauloidea, Pristigasteroidea, Clupeoidei, and Clupeiformes, but not the sister group relationship between the Clupeoidea and Engrauloidea, were corroborated by two recent studies based on mitogenomic and nuclear (RAG1 and RAG2) nucleotide sequences (Lavoué *et al.*, 2007; Li & Ortí, 2007). Both studies found evidence for a sister group relationship between the Chirocentridae and a clade composed of some genera traditionally assigned to the Dussumieriinae. However, Lavoué *et al.* (2007: 1103) stated that '... the higher rate of molecular evolution of *Chirocentrus* ... makes difficult the estimation of its phylogenetic position'. Li & Ortí (2007) considered their results preliminary, but indicative that a revision of the classification of the Clupeiformes is necessary. They were also not confident about the phylogenetic position of the Chirocentridae implied by their data, as topology tests failed to reject an alternative hypothesis of a sister group relationship between the Chirocentridae and Engrauloidea (Li & Ortí, 2007: 395).

The Engrauloidea is a diverse clade of the Clupeiformes and comprises fishes commonly known as anchovies, which are mainly schooling, usually small to medium-sized planktivorous fishes (Grande & Nelson, 1985; Whitehead, Nelson & Wongratana,

1988; Wongratana, Munroe & Nizinski, 1999; Nelson, 2006). Hypothesized synapomorphies of the Engrauloidea are the mesethmoid projecting anteriorly beyond the vomer and the oblique inclination of the suspensorium (Nelson, 1984; Grande & Nelson, 1985; Grande, 1985). The Engrauloidea is divided into the Coiliidae and Engraulidae (Grande & Nelson, 1985; Bornbusch & Lee, 1992; Li & Ortí, 2007). Apart from the division of the superfamily into those two families, relationships among anchovies are largely unknown. Relationships among species of the Engraulini, by far the most diverse group of the Engrauloidea, are totally unknown (Grande & Nelson, 1985).

An attempt to re-evaluate hypothesized synapomorphies of the Engrauloidea focusing on morphological components of their oblique-inclined suspensorium revealed that the Chirocentridae have a series of intermediate character states between the generalized condition present in non-engrauloid clupeiforms on the one hand, and the Engrauloidea on the other, which are herein described and properly put in a phylogenetic framework. This study furthermore presents two previously unreported character states of the branchial arches and infraorbitals that are exclusive to the Chirocentridae and Engrauloidea within the Clupeiformes. These character states together constitute strong evidence favouring a hitherto unsuspected hypothesis of a sister group relationship between the Chirocentridae and Engrauloidea.

## MATERIAL AND METHODS

Specimens were cleared and stained according to the procedure of Taylor & Van Dyke (1985) and Song & Parenti (1995). Osteological terminology follows Weitzman (1962), Nelson (1969a), Grande (1985), and Whitehead & Teugels (1985). The distribution of character states among examined taxa (Appendix) is discussed and optimized on the currently accepted phylogenetic hypothesis of relationships among components of the Clupeomorpha (Grande, 1985; Grande & Nelson, 1985; Di Dario, 2002; Chang & Maisey, 2003; Di Dario, 2004; Di Dario & de Pinna, 2006; de Pinna & Di Dario, in press).

## ABBREVIATIONS

Institutional abbreviations are: AMNH, American Museum of Natural History, New York; ANSP, Academy of Natural Sciences, Philadelphia; FMNH, Field Museum, Chicago; INPA, Instituto Nacional de Pesquisas da Amazônia, Manaus; MCP, Museu de Ciências e Tecnologia da Pontifícia Universidade Católica, Porto Alegre; MZUSP, Museu de Zoologia da Universidade de São Paulo, São Paulo; SAIAB, South African Institute for Aquatic Biodiversity, Graham-

stown; UMMZ, University of Michigan Museum of Zoology, Ann Arbor; USNM, National Museum of Natural History, Smithsonian Institution, Washington DC.

Osteological abbreviations are: ao, antorbital; b<sub>1-3</sub>, basibranchials 1–3; bh, basihyal; c<sub>1-3</sub>, ceratobranchials 1–3; ecp, ectopterygoid; enp, endopterygoid; gr, gillraker; h<sub>1-3</sub>, hypobranchials 1–3; hm, hyomandibula; ih, interhyal; ihm, medial laminar process of hyomandibula; io<sub>1-6</sub>, infraorbitals 1–6; loq, laminar outgrowth of the anterior margin of quadrate; lph, laminar process of the ventral limb of hyomandibula; mtp, metapterygoid; ohm, lateral laminar process of hyomandibula; pal, palatine; q, quadrate; s, symplectic; so, supraorbital.

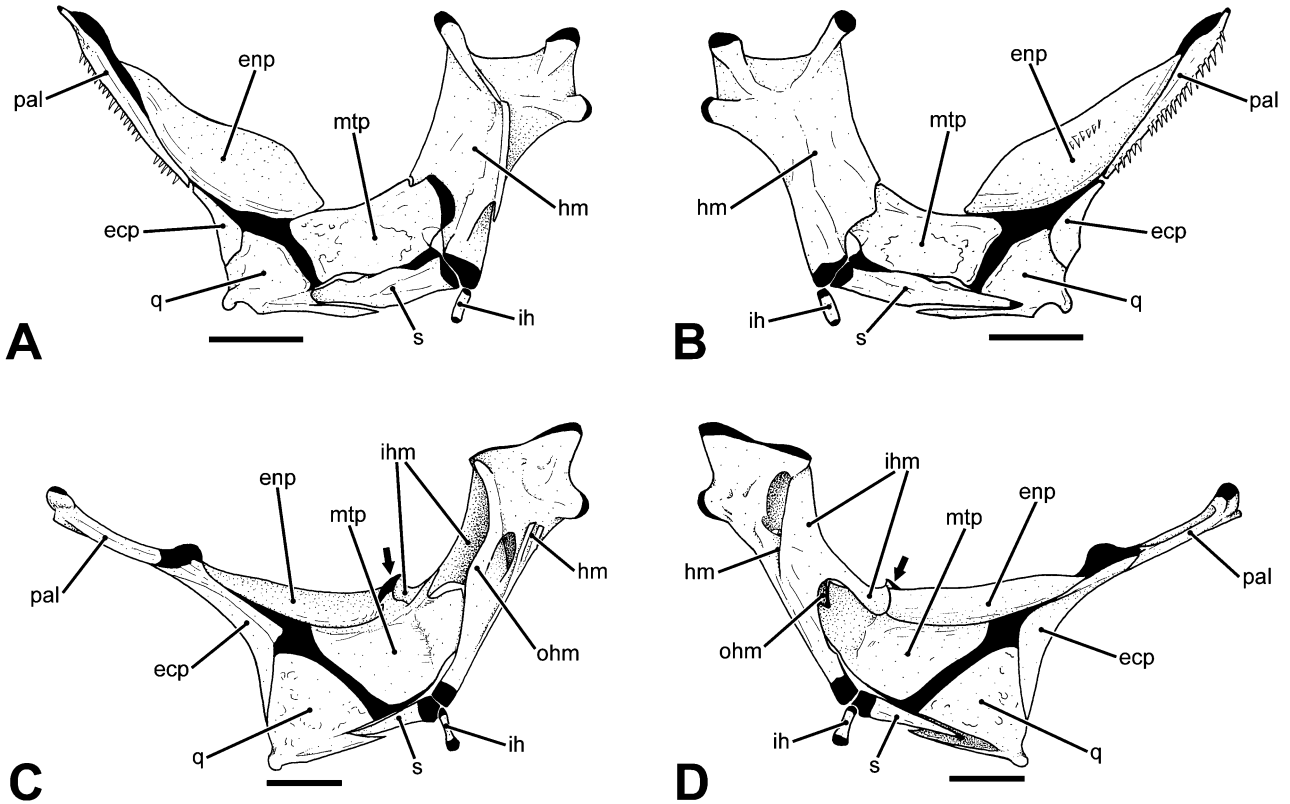
## RESULTS

### CHARACTERS

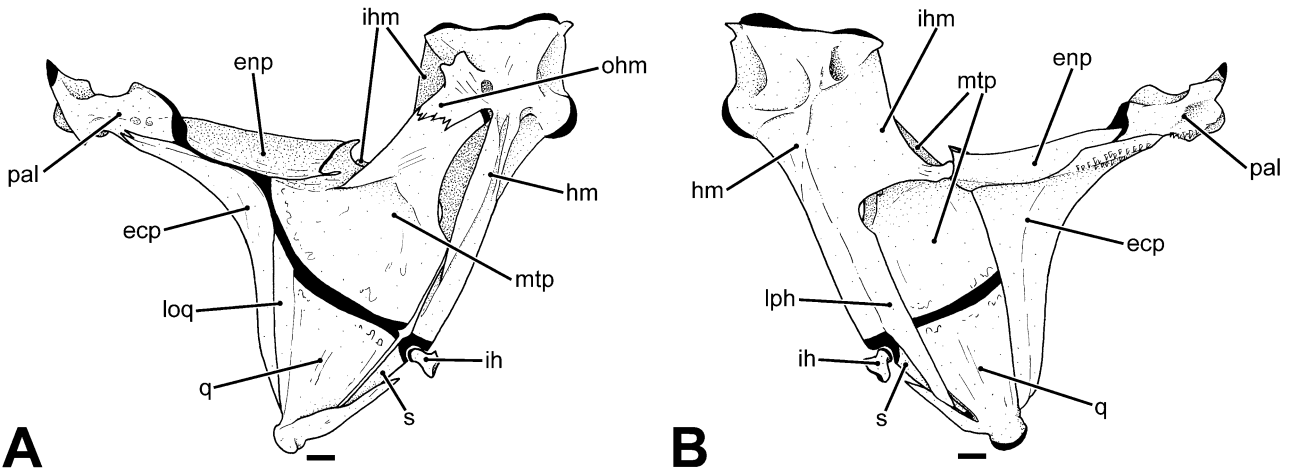
#### *1. Anterior margin of metapterygoid located anterior to the quadrate*

The metapterygoid is well developed and dorsal to the quadrate in all examined clupeiforms. In *D. clupeoides* and in the examined species of the Clupeidae and Pristigasteroidea, the anterior margin of the metapterygoid is located posterior to the anterior border of the quadrate, which, in turn, articulates with the ectopterygoid (state 0; Fig. 2). The anterior margin of the metapterygoid in these species is typically positioned midway between the anterior and posterior borders of the proximal margin of the quadrate, as in the case of *D. clupeoides* (Fig. 2A, B), or is positioned in a region slightly anterior to that (Fig. 2C, D). The metapterygoid is dorsoposterior to the quadrate in this hypothesized plesiomorphic state. Alternatively, the anterior margin of the metapterygoid is located anterior to the quadrate in the Chirocentridae and Engrauloidea (state 1; Figs 3–5). That condition results mostly from the displacement of the quadrate to a more posterior position in the suspensorium relative to state 0. A substantial portion of the metapterygoid of chirocentrids is situated dorsoposterior to the quadrate, as is the typical condition for non-engrauloid clupeiforms (Fig. 3). The metapterygoid is located in an anterodorsal position relative to the quadrate uniquely in the Engrauloidea within the Clupeiformes. This condition is hypothesized as resulting from a further displacement of the quadrate to a more posterior position in the suspensorium (state 2; Figs 4, 5).

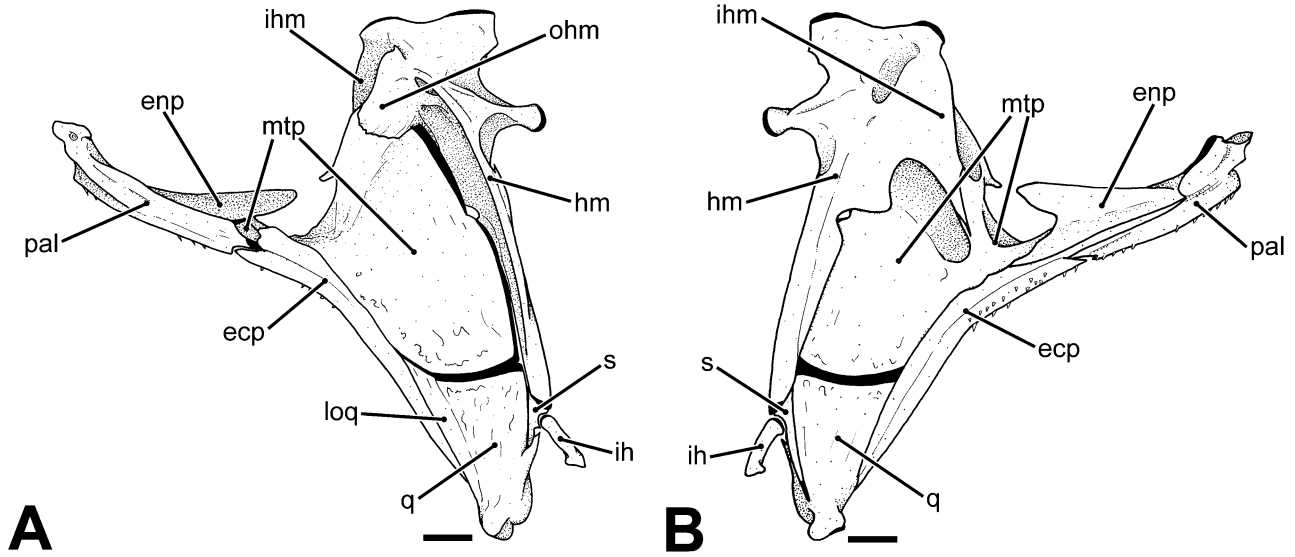
In species of the genera *Brevoortia*, *Hilsa*, and *Tenualosa* in the Clupeidae subfamily Alosinae, the metapterygoid is located in a more anterior position in the suspensorium relative to other clupeids (Segura & Díaz de Astarloa, 2004). However, as in



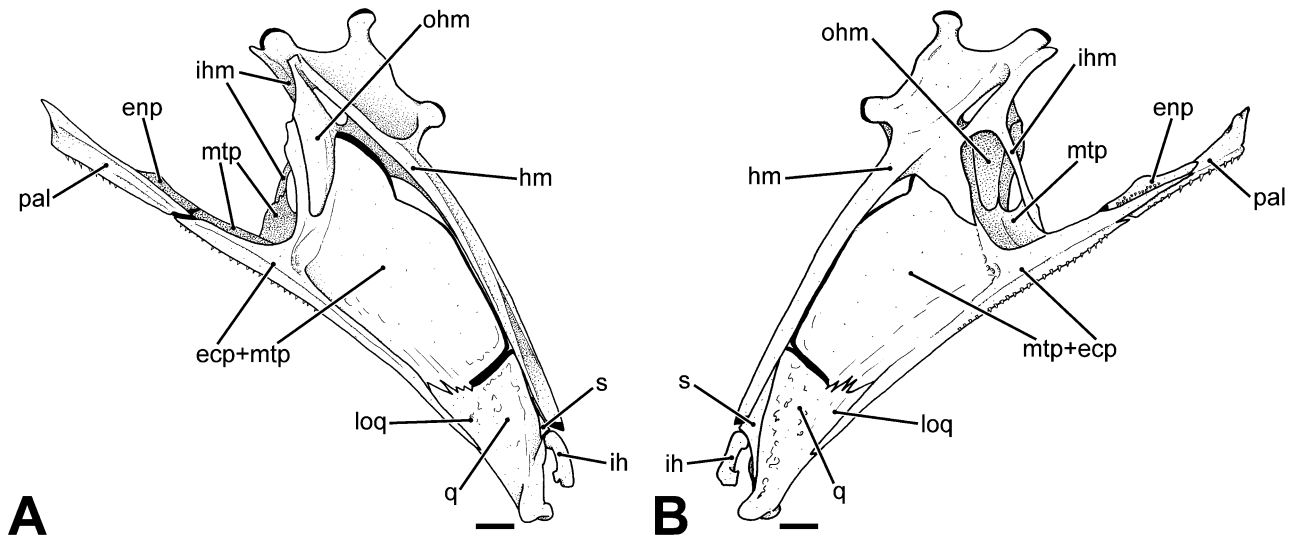
**Figure 2.** Suspensoria of the Denticipitoidei and Clupeidae showing the hypothesized plesiomorphic states of characters 1 to 5, discussed in the text. A, lateral view, and B, medial view, of the suspensorium of *Denticiceps clupeioides* (MZUSP 84776); C, lateral view, and D, medial view, of the suspensorium of *Sardina pilchardus* (MZUSP 37394). Arrows indicate scythe-like process of metapterygoid. Cartilage in solid black. Scale bars = 1 mm. ecp, ectopterygoid; enp, endopterygoid; hm, hyomandibula; ih, interhyal; ihm, medial laminar process of hyomandibula; mtp, metapterygoid; ohm, lateral laminar process of hyomandibula; pal, palatine; q, quadrate; s, symplectic.



**Figure 3.** Suspensoria of *Chirocentrus dorab* (MZUSP 62467) in lateral (A) and medial (B) views showing hypothesized apomorphic states of characters 1 to 5, discussed in the text. Cartilage in solid black. Scale bars = 1 mm. ecp, ectopterygoid; enp, endopterygoid; hm, hyomandibula; ih, interhyal; ihm, medial laminar process of hyomandibula; loq, laminar outgrowth of the anterior margin of quadrate; lph, laminar process of the ventral limb of hyomandibula; mtp, metapterygoid; ohm, lateral laminar process of hyomandibula; pal, palatine; q, quadrate; s, symplectic.



**Figure 4.** Suspensorium of *Thyryssa hamiltoni* (USNM 364595) in lateral (A) and medial (B) views showing hypothesized apomorphic states of characters 1 to 5, discussed in the text. Cartilage in solid black. Scale bars = 1 mm. ecp, ectopterygoid; enp, endopterygoid; hm, hyomandibula; ih, interhyal; ihm, medial laminar process of hyomandibula; loq, laminar outgrowth of the anterior margin of quadrate; mtp, metapterygoid; ohm, lateral laminar process of hyomandibula; pal, palatine; q, quadrate; s, symplectic.



**Figure 5.** Suspensorium of *Anchoa spinifer* (MZUSP 11454) in lateral (A) and medial (B) views showing hypothesized apomorphic states of characters 1 to 5, discussed in the text. Cartilage in solid black. Scale bars = 1 mm. ecp, ectopterygoid; enp, endopterygoid; hm, hyomandibula; ih, interhyal; ihm, medial laminar process of hyomandibula; loq, laminar outgrowth of the anterior margin of quadrate; mtp, metapterygoid; ohm, lateral laminar process of hyomandibula; pal, palatine; q, quadrate; s, symplectic.

the hypothesized plesiomorphic state, the anterior margin of the metapterygoid in these taxa does not project in advance of the anterior border of quadrate. According to Ridewood (1904: fig. 124) the anterior margin of the metapterygoid of *Alosa fallax* is located in a region anterior to the quadrate. The character is

polymorphic in *Alosa* because *Alosa caspia* has a condition similar to that described for *A. fallax* by Ridewood (1904), whereas in *Alosa pseudoharengus* the anterior margin of the metapterygoid does not project in advance of the quadrate. The occurrence of both states among species of *Alosa*, and the presence

of state 0 in other Alosinae, indicate that the anterior location of the metapterygoid in some species of *Alosa* is convergent with the condition present in the Chirocentridae and Engrauloidea. Furthermore, the Alosinae is probably a distal clade within the Clupeidae, given the presence of various hypothesized derived characters in the branchial arches of the taxa included in that subfamily (e.g. loss of tooth plates, high number of gill rakers; Di Dario, 2002). The distal position of the genera of Alosinae in a clade largely equivalent to the Clupeidae is also indicated by molecular data (Lavoué *et al.*, 2007; Li & Ortí, 2007).

## 2. Ventral limb of hyomandibula and quadrate not separated by metapterygoid

The hyomandibula of the Clupeiformes has a broad proximal body whose distal end narrows abruptly into a vertical limb distally tipped by cartilage (Greenwood, 1968; Figs 2–5). In the hypothesized plesiomorphic condition present in *Denticeps*, the Pristigasteroidea, and Clupeidae, the ventral limb of the hyomandibula and the quadrate are separated by a portion of the metapterygoid which projects ventrally between those two bones. (state 0; Fig. 2).

In the Engrauloidea and Chirocentridae uniquely within the Clupeiformes the ventral limb of the hyomandibula and quadrate are not separated by the metapterygoid (state 1; Figs 3–5). In the Chirocentridae the ventral margin of the metapterygoid is in line with the distal end of the ventral limb of the hyomandibula (Fig. 3). This state is clearly intermediate between the states described for non-engrauloid clupeiforms and the Engrauloidea, in which the ventral margin of the metapterygoid is located distinctly dorsal to the distal end of the ventral limb of the hyomandibula (Figs 4, 5). As a consequence of the relative position of those bones in the suspensorium, in the Engrauloidea the ventral limb of the hyomandibula articulates with the quadrate (state 2), whereas in other Clupeiformes, including the Chirocentridae, the vertical limb of the hyomandibula articulates exclusively with the metapterygoid.

The hyomandibula of the Chirocentridae is further distinct from those of other Clupeiformes in the presence of a laminar bony process that extends distally from the ventral limb of the hyomandibula and partially covers the medial face of the metapterygoid (Fig. 3B, lph; Ridewood, 1904; Bardack, 1965). This laminar process is uniquely present in the Chirocentridae within the Clupeiformes. The structure is therefore hypothesized to be an autapomorphy of the Chirocentridae.

Although always ventral to the distal end of the hyomandibula, the position of the ventral margin of the metapterygoid varies among examined Clupeidae. In members of the Clupeinae, Pellonulinae, Dussum-

ieriinae, and Dorosomatinae, the ventral margin of the metapterygoid is located markedly ventral to the distal end of the hyomandibula. In most members of the Alosinae the ventral margin of the metapterygoid is clearly ventrally positioned, but to a less pronounced degree, to the distal end of the hyomandibula. The distal end of the hyomandibula is aligned to the ventral margin of the metapterygoid in the specimen of *Brevoortia aurea* illustrated by Segura & Díaz de Astarloa (2004). Even if that is the case for other species of *Brevoortia*, that condition is most probably homoplastic to that observed in the Chirocentridae and Engrauloidea because of the reasons discussed for the previous character.

The proximity between the ventral limb of the hyomandibula and the quadrate of the Chirocentridae and Engrauloidea is related to a change of the relative position of the symplectic and interhyal in the suspensorium. The symplectic is a relatively small bone that inserts into the narrow space between the ventral margin and a splint-like posterior process of the quadrate in the Clupeiformes and most Teleostei (Arratia & Schultze, 1991). Typically, the symplectic is diagonally orientated relative to the longitudinal axis of the body, such that its proximal end is directed towards the distal end of the ventral limb of the hyomandibula (Figs 2–5). In the hypothesized plesiomorphic state, where the distal end of the hyomandibula is situated in a more dorsal position relative to the quadrate, the proximal end of the symplectic extends dorsally beyond the proximal margin of the quadrate (Fig. 2). As a consequence, the symplectic articulates with both the metapterygoid and quadrate, and the head of the interhyal, which articulates with the symplectic, inserts on the suspensorium in a position dorsal to the quadrate (Fig. 2).

In the Chirocentridae and Engrauloidea the symplectic and quadrate are more closely associated with each other than in other Clupeiformes, and the proximal end of the symplectic does not project dorsally beyond the quadrate. As a consequence, the proximal region of the symplectic of chirocentrids and engrauloids falls short of the metapterygoid (Figs 3–5). In the Chirocentridae the proximal tip of the symplectic is in line with the proximal margin of the quadrate (state 1; Fig. 3). In the Engrauloidea, because of the pronounced shift to a more dorsal position of the quadrate and metapterygoid relative to the hyomandibula, the proximal end of the symplectic is located in a position ventral to the proximal margin of the quadrate (state 2; Figs 4, 5). This morphology results in a marked shift ventrally of the insertion of the interhyal in the suspensorium. In engraulids such as *Anchoviella lepidentostole* and *Anchoa spinifer*, for instance, the head of the interhyal inserts on the suspensorium at a point halfway between the

extremities of the posterior margin of the quadrate (Fig. 5).

3. *Posterodorsal margin of metapterygoid reaching a relatively more dorsal position in suspensorium, in line with the condyle of articulation of hyomandibula with opercle*

The posterodorsal margin of the metapterygoid of *D. clupeioides*, the Pristigasteroidea, and Clupeoidea extends to a region located approximately halfway between the dorsal and ventral margins of the hyomandibula (Fig. 2A, B; Greenwood, 1968). In the Chirocentridae and Engrauloidea uniquely within the Clupeiformes, the posterodorsal margin of the metapterygoid reaches a relatively more dorsal position in the suspensorium, in line with the condyle of articulation of the hyomandibula with the opercle (Figs 3–5; Ridewood, 1904). Therefore, the latter condition is hypothesized as apomorphic within Clupeiformes and indicative of a sister group relationship between the Chirocentridae and Engrauloidea.

The shape of the metapterygoid and the articulations among bones of the posterodorsal region of the suspensorium in species of the Clupeoidei are complex. The variation observed in this region of the cranium has not been properly explored in a phylogenetic context, and may reveal character states potentially informative for resolving relationships within the suborder. Typically, in the Pristigasteroidea and Clupeidae, but not in the Chirocentridae and Engrauloidea, the posterodorsal region of the metapterygoid is medially curved, forming a parasagittally orientated bulge whose dorsal margin is directed towards the proximal region of the hyomandibula (Fig. 2C). The dorsal margin of the metapterygoid of non-engrauloid Clupeoidei has a scythe-like medially-directed process that originates at the base of the bulge. The anterior margin of this process is tipped with cartilage (Fig. 2C, D, arrow, and Fig 3A). The anterior margin of the hyomandibula in the Pristigasteroidea, Clupeidae, and Chirocentridae has a ventrally directed laminar process, whose distal end is laterally expanded and articulates with, or at least projects towards (as in the pristigasteroid *Opisthopterus dovii*) the scythe-like process of the dorsal margin of the metapterygoid (Fig. 2C, D, ihm). The same ventrally directed laminar process, but in this case without the laterally expanded distal end, can be identified in the Coiliidae (Fig. 4B). In the Engraulidae the process is presumably further modified as a thin, flat stretch of bone located between the main body of the hyomandibula and the metapterygoid (Fig. 5B, ihm). A third medial laminar process of the anterior margin of the hyomandibula is present in the Coiliidae, and is also further developed in the Engraulidae uniquely among the Clupeiformes.

The dorsal margin of the posterodorsal portion of the metapterygoid of the Chirocentridae and Engrauloidea is slightly bent laterally and articulates with the lateral process of the hyomandibula (Figs 3A, 4A, 5A, ohm). The lateral laminar process of the hyomandibula is probably homologous to a relatively well-developed and laminar process that projects along the outer margin of the anterior face of the hyomandibula of other species of the Clupeoidei (Fig. 2C).

The shape of the metapterygoid and the relationships among bones of the proximal region of the suspensorium of *D. clupeioides* are simplified relative to other Clupeiformes. The posterodorsal region of the bone lacks a scythe-like process, and is, consequently, completely flat and nearly rectangular (Fig. 2A, B; Greenwood, 1968). The lateral laminar process of the hyomandibula (ohm) is absent. The only laminar process of the anterior face of the hyomandibula neither has an anterior projection, nor does it articulate in a complex way to the metapterygoid as in the Chirocentridae and Engrauloidea (Fig. 2B).

The relative degree of development of the lateral laminar process of the hyomandibula (ohm) shared by the Chirocentridae and Engrauloidea seems to be apomorphic within the Clupeoidei. Other features, such as the absence of a convex bulge at the dorsal region of the metapterygoid, are probably primitive and indicative of a basal position of the Chirocentridae and Engrauloidea within the suborder. However, these character states are not conclusively optimized on the currently accepted phylogeny of Clupeiformes, pending further studies.

4. *Laminar outgrowth of anterior margin of quadrate*

The anterior margin of the quadrate of the Chirocentridae and Engrauloidea is unique within Clupeiformes in bearing a laminar outgrowth that projects anterodorsally beyond the typical cartilage bordered dorsal margin of the bone (Figs 3A, 4A, 5, loq). Developmental series of chirocentrids and engrauloids were not examined for this character. Nevertheless, the absence of cartilage bordering the dorsal margin of the outgrowth, and a marked difference in the texture and thickness between the outgrowth and the region of the quadrate posterior to it, suggest that the outgrowth develops as membrane bone following the ossification of the endochondral portion of the quadrate. A projection of the posterior margin of the ectopterygoid medially covers the laminar outgrowth of the anterior margin of the quadrate and also to a degree the metapterygoid of the Chirocentridae and Coiliidae (Figs 3B, 4B). The ectopterygoid and metapterygoid of engraulids are fused in the region where the projection of the posterior margin of the ectopterygoid in chirocentrids and coiliids articulates with the metap-

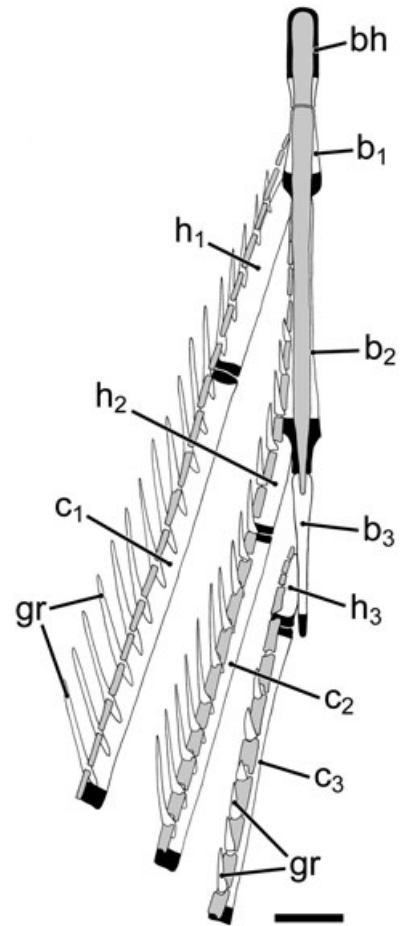
terygoid (Grande & Nelson, 1985; Fig. 5). The laminar outgrowth of the anterior margin of the quadrate is absent from examined *Denticeps*, pristigasteroids, and clupeids (Fig. 2). Therefore, the presence of this structure is hypothesized to be apomorphic to the Chirocentridae and Engrauloidea within the Clupeiformes.

##### 5. Shape of quadrate

The quadrate of the examined Clupeiformes is well developed and triangular in shape, as is usually the case in the Teleostei (Arratia & Schultze, 1991). The dorsal margin of the endochondral portion of the quadrate is somewhat arched dorsally in *Denticeps* (Fig. 2A, B), and roughly straight in observed species of the Clupeoidei (Fig. 2C, D; Figs 3–5). Regardless of the shape of the dorsal margin of the quadrate, lengths of the line segment between the vertices of the triangular-shaped endochondral portion of the quadrate in *Denticeps*, the Pristigasteroidea, and Clupeidae are approximately equivalent. As a consequence, the shape of the endochondral portion of the quadrate of those taxa is similar to that of an equilateral triangle. Alternatively, the distance between the vertices that define the dorsal margin of the endochondral portion of the quadrate of the Chirocentridae and Engrauloidea is markedly shorter than those of the approximately equal anterior and posterior margins of the bone (Figs 3–5). As a consequence, the shape of the endochondral portion of the quadrate of the Chirocentridae and Engrauloidea is similar to that of an isosceles triangle. This particular shape of the quadrate is unique within the Clupeiformes, and is therefore hypothesized to be apomorphic for the Chirocentridae and Engrauloidea.

##### 6. Presence and arrangement of autogenous tooth plates on ceratobranchials 1 to 3

Autogenous tooth plates are present on ceratobranchials 1 to 4 of the Chirocentridae. In ceratobranchials 1 to 3, they are uniform in shape and relative size, sequentially aligned, and each plate extends across the distance between two consecutive gill rakers (Figs 6, 7A). The overall shape of each tooth plate is approximately that of a laterally flattened and short siege tower (Fig. 8A). They are mostly lateral in relation to the ceratobranchials, but their dorsal third is slightly bent medially in such a way that it partially covers the oral surface of the underlying bone. The anterior and posterior tips of the dorsal region of the plates are equally prolonged relative to their middle to ventral regions. In oral view, ceratobranchials 1 to 3 of the Chirocentridae seem to be partially covered by a series of narrow, equally sized rectangular-shaped tooth plates (Fig. 6). The oral surface of each tooth plate typically bears about ten relatively well-

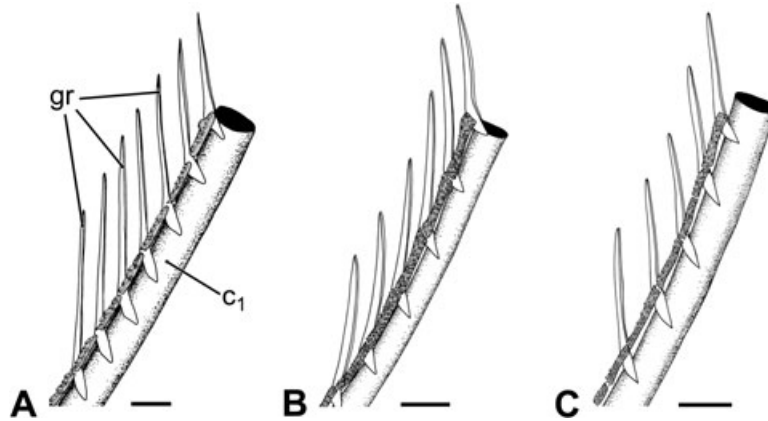


**Figure 6.** Anterior left portion of the ventral branchial arches of *Chirocentrus dorab* (MZUSP 62467) in dorsal view. Teeth and gill raker denticles not represented. Dermal tooth plates in grey; cartilage in solid black. Scale bar = 2 mm. b1–3, basibranchials 1–3; bh, basihyal; c1–3, ceratobranchials 1–3; gr, gillraker; h1–3, hypobranchials 1–3.

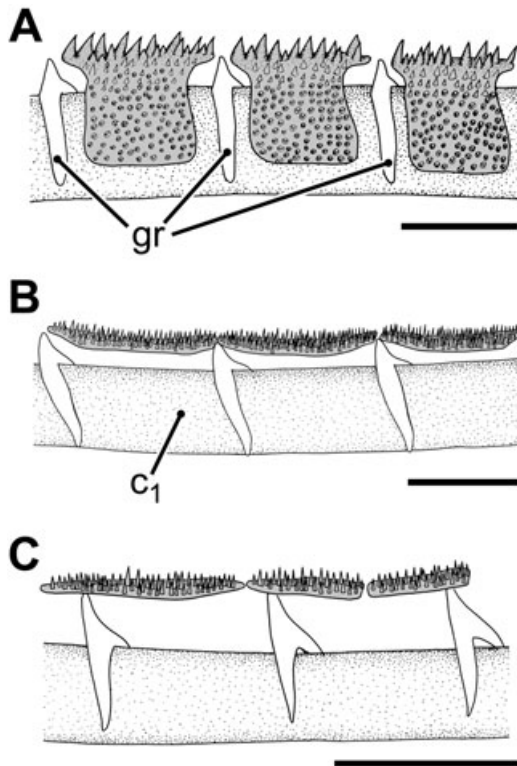
developed, posterodorsally directed, teeth (Fig. 8A). The remaining middle to ventral region of the plate is approximately square, and it is covered by less developed but evenly distributed innumerable laterally directed teeth. A gill raker projects from the short space between two consecutive plates in the lateral surface of the ceratobranchials (Fig. 8A).

Dermal tooth plates are typically distributed over most of the oral surface of the gill arches in the Engrauloidea (Nelson, 1970). Ceratobranchials 1 to 3 of the coiliids *Lycotrissa*, *Setipinna*, and *Thryssa* are densely covered by autogenous tooth plates (Fig. 7B). The arrangement and shape in oral view of tooth plates in these genera of the Coiliidae are strikingly similar to the conditions described above for the Chirocentridae (Fig. 7A). They are regularly aligned,





**Figure 7.** Proximal third of left ceratobranchial 1 of: A, *Chirocentrus dorab* (MZUSP 62467); B, *Lycothrissa crocodilus* (MZUSP 62482); and C, *Pterengraulis atherinoides* (MZUSP 18040) in dorsal view, anterior facing up. Dermal tooth plates in grey; cartilage in solid black. Gill raker denticles not represented. Scale bars = 1 mm. gr, gillraker.



**Figure 8.** Lateral view of the middle region of ceratobranchial 1 of: A, *Chirocentrus dorab* (MZUSP 62467); B, *Lycothrissa crocodilus* (MZUSP 62482); and C, *Pterengraulis atherinoides* (MZUSP 18040). Anterior facing left. Only proximal portion of gill rakers represented. Dermal tooth plates in grey. Gill raker denticles not represented. Scale bars = 1 mm. gr, gillraker.

uniform in shape and relative size, and bear a dense coat of relatively well-developed upward directed teeth (Figs 7B, 8B). As in the case of ceratobranchials 1 to 3 of the Chirocentridae (Figs 7A, 8A), each

equally sized plate uniformly extends across the distance between two consecutive gill rakers (Figs 7B, 8B). Tooth plates are relatively less developed and restricted to the oral surface of ceratobranchial 1 of *Lycothrissa*, and ceratobranchials 1 and 2 of *Setipinna* and *Thryssa*. Tooth plates on ceratobranchial 2 of *Lycothrissa*, and ceratobranchial 3 of *Setipinna* and *Thryssa* are more developed than those of ceratobranchial 1, and partially cover both the oral and lateral surfaces of the underlying bone. A variable number of small and polymorphic additional tooth plates are also present on ceratobranchials 1 to 4 of *Setipinna*, ceratobranchials 3 and 4 of *Lycothrissa*, and ceratobranchial 4 of *Thryssa*. The irregular disposition, shape, and size of those plates are evidence that they are not homologous to the regularly aligned series of tooth plates present in the anterior ceratobranchials of the Chirocentridae.

Teeth are completely absent from ceratobranchials 1 to 4 of *Coilia reventischi*, but innumerable diminutive teeth are sequentially aligned on the oral surface of ceratobranchials 1 and 2 of *C. neglecta*. The presence of a series of putatively derived anatomical features shared exclusively by species of *Coilia* indicates that the genus is monophyletic and distal in the Coiliidae (Patterson & Johnson, 1995; Di Dario, 2002). The sequentially aligned series of teeth on ceratobranchials 1 and 2 of *C. neglecta* is a condition highly reminiscent of the arrangement of autogenous tooth plates observed in other genera of the family where dentition is more developed. The distal position of *Coilia* and the occurrence of the character state in other members of the Coiliidae indicate that the presence of sequentially aligned, rectangular shaped autogenous tooth plates, each extending between consecutive gill rakers on ceratobranchials 1 to 3, is primitive for the family. The primitive arrangement of

these plates seems to be retained in the anterior ceratobranchials of species of *Coilia* where dentition is extremely reduced, albeit unquestionably present, as in *C. neglecta*.

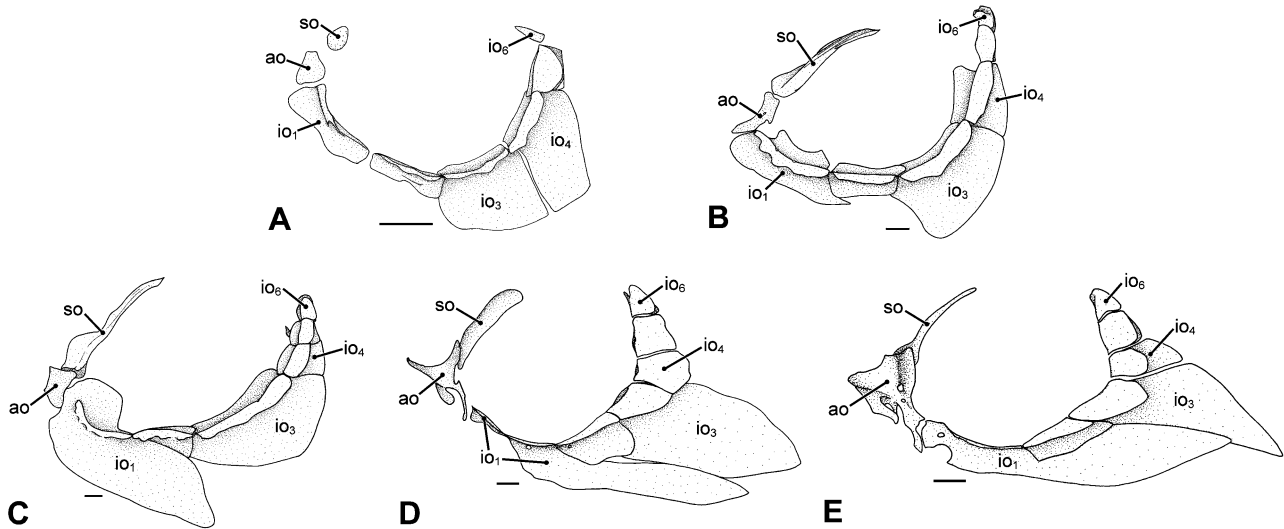
Ceratobranchials 1 to 3 of predatory engraulids such as *Anchoa spinifer*, *Lycengraulis grossidens*, and *Pterengraulis atherinoides* are partially covered by well-developed autogenous tooth plates (Figs 7C, 8C; Nelson, 1970: fig. 2A), which, in overall shape and arrangement, are strikingly similar to the ones described for the Coiliidae. All tooth plates are sequentially aligned and bear innumerable relatively small to medium sized, dorsally orientated, teeth (Fig. 8C). Teeth at the medial border of the plates are somewhat posteriorly directed. Some tooth plates in the anterior ceratobranchials of these predatory engraulids have variable lengths (Figs 7C, 8C). Ceratobranchials 1 and 2 of selective planktivorous engraulids, such as *Anchoviella lepidentostole*, which feed on larvae and small crustaceans (Cervigón, 1982; Bornbusch, 1988), are covered by reduced but sequentially aligned autogenous tooth plates. That same arrangement and a similar degree of development of tooth plates is also present on ceratobranchials 1 and 2 of examined *Stolephorus*, which feed mostly on selected small crustaceans or large zooplankton, such as amphipods, copepods, mysids, ostracods, brachyuran larvae, and euphysiids (Hardenberg, 1934; Blaber, 1979; Milton, Blaber & Rawlinson, 1990; Bornbusch & Lee, 1992). *Stolephorus* is hypothesized to be the most basal genus of the Engraulidae (Grande & Nelson, 1985).

The relative degree of development of the gill raker dentition coupled with the interpretation of feeding habits of anchovies in a phylogenetic context indicate that suspension feeding, with the typically associated reduction of tooth plates, is derived within the Engraulidae (Bornbusch, 1988; Bornbusch & Lee, 1992). Autogenous tooth plates are absent from the anterior arches of the Indo-Pacific *Encrasicholina heteroloba* and the New World *Engraulis anchoita*, *Cetengraulis edentulus*, *Anchoa* (*A. januaria*, *A. filifera*, *A. lamprotaenia*, *A. marini*, and *A. lyolepis*), *Anchovia* (*A. clupeioides*, *A. surinamensis*), *Amazonsprattus scintilla*, and *Anchoviella* (freshwater species: *A. guianensis*, *A. vaillanti*). As expected, members of those species are mostly planktivores (Bayliff, 1963; de Ciechomski, 1967; Leong & O'Connell, 1969; Blaber, 1979; James, 1988; Whitehead *et al.*, 1988; James & Findlay, 1989; Wongratana *et al.*, 1999; Gay, Bassani & Sergipense, 2002; Krumme *et al.*, 2005; Pájaro, Curelovich & Macchi, 2007). Absence of tooth plates on anterior arches of *Amazonsprattus scintilla* and other diminutive species of New World freshwater anchovies, such as *Anchoviella vaillanti* and *Anchoviella guianensis*, probably relates to their pae-

domorphic nature. A condition of tooth plates similar to those of the Chirocentridae, Coiliidae, and basal or generalized species of the Engraulidae is found even among some presumably microphagous engraulids. That is the case for the freshwater Amazonian *Jurengraulis juruensis*. Each of the anterior arches of this species bears more than 100 relatively long and slender gill rakers, which are in turn equipped with innumerable delicate dendritic projections along their inner edge (Whitehead *et al.*, 1988: fig on p. 385) A series of sequentially aligned autogenous square to rectangular tooth plates form a conspicuous flange on the lateral margin of the oral surfaces of ceratobranchials (and also epibranchials) 1 to 3. These plates are densely covered by highly reduced caniniform teeth.

In summary, autogenous tooth plates, each located between consecutive gill rakers, are present on ceratobranchials 1 to 3 of the Chirocentridae and, primitively, of the Engrauloidea. Distal members of the Engrauloidea are mostly specialized microphagists, and some New World species are pedomorphic. Those conditions probably account for the extreme reduction or secondary absence of autogenous tooth plates from their branchial arches, although some microphagous engraulids (such as *Jurengraulis juruensis*) retain the hypothesized plesiomorphic state of the character.

The Ostariophysi has been hypothesized to be the sister group of the Clupeomorpha in the Otocephala on the basis of both morphological and molecular evidence (Lê, Lecointre & Perasso, 1993; Johnson & Patterson, 1996; Lecointre & Nelson, 1996; Arratia, 1997, 1999; Inoue *et al.*, 2003). More recently, Otocephala was expanded to include the Alepocephaloidei (Ishiguro, Miya & Nishida, 2003, 2005; Lavoué *et al.*, 2005). Ceratobranchials 1 to 3 of the Gonorynchiformes and Cypriniformes, which are basal clades of the Ostariophysi (Fink & Fink, 1981, 1996), are devoid of tooth plates (Siebert, 1987; Johnson & Patterson, 1996). Autogenous tooth plates are also completely absent from ceratobranchials 1 to 3 of examined Alepocephaloidei, *D. clupeioides*, the Clupeidae, and most Pristigasteroidea (Greenwood, 1968; Di Dario, 1999; de Pinna & Di Dario, in press). The absence of tooth plates from ceratobranchials 1 to 3 is therefore more parsimoniously optimized as the primitive condition for the Clupeiformes. Conversely, the presence of autogenous dermal tooth plates on ceratobranchials 1 to 3 is apomorphic within the Clupeiformes, and indicative of a sister group relationship between the Chirocentridae and Engrauloidea. The Chirocentridae and Engrauloidea also apomorphically share the regular arrangement of autogenous tooth plates on ceratobranchials 1 to 3, where plates are sequentially aligned and each plate



**Figure 9.** Left circumorbital bones of representative Clupeiformes. A, *Denticeps clupeoides* [after Di Dario, 2002, with the addition of the antorbital based on *D. clupeoides* (MZUSP 84776); dermal denticles not represented]; B, *Alosa pseudoharengus* (MZUSP 129789); C, *Chirocentrus dorab* (MZUSP 62467); D, *Setipinna melanochir* (MZUSP 64119); E, *Anchoa filifera* (MZUSP 18528). Scale bars = 1 mm. ao, antorbital; io1–6, infraorbitals 1–6; so, supraorbital.

typically extends across the distance between consecutive gill rakers.

Autogenous tooth plates are present on ceratobranchials 1 to 3 of the South American freshwater pristigasteroids *Pellona castelnaeana* and *P. flavipinnis*, but they are absent from ceratobranchials of other pristigasteroids including the basal *Pellona harroweri* and *P. ditchella* (Di Dario, 1999; de Pinna & Di Dario, 2003). The condition present in *P. castelnaeana* and *P. flavipinnis* is therefore more parsimoniously hypothesized as convergent to that in the Chirocentridae and Engrauloidea. Autogenous tooth plates similar to those of the anterior arches of the Chirocentridae and Engrauloidea, but also hypothesized to be homoplastic according to parsimony, are present on ceratobranchials 1 to 3 of *Megalops* and *Elops* (Elopomorpha) and *Latimeria chalumnae* [Nelson, 1969a: plate 82(2)].

#### 7. Posterior region of infraorbital 1 well-developed and extending along the ventral margin of infraorbital 2

Six autogenous infraorbitals are typically present in species of the Clupeiformes, although the homology of the infraorbital 6, or dermosphenotic, of *D. clupeoides* and those of other clupeiforms is in debate (Nelson, 1969b; Grande, 1985; Di Dario, 2004; Di Dario & de Pinna, 2006). Infraorbital 1 is usually more well developed than, and always articulated with, infraorbital 2 in examined clupeiforms (Fig. 9). Infraorbital 1 of *D. clupeoides* is relatively simple, elongate and slender (Greenwood, 1968; Di Dario, 2004; Di Dario &

de Pinna, 2006). Although infraorbital 1 is slightly longer than infraorbital 2 in this species, the depth of the posterior margin of infraorbital 1 is approximately the same as that of the anterior margin of infraorbital 2 (Fig. 9A).

The ventral margin of the posterior region of infraorbital 1 of most examined species of the Clupeoidei has a process that projects underneath, and in the typical condition is partially covered by the ventral margin of the anterior region of infraorbital 2 (Fig. 9B). This process is absent from *D. clupeoides*. As a result of the presence of such a process, the posterior margin of infraorbital 1 of most species of the Clupeoidei is usually deeper than the anterior margin of infraorbital 2. In examined clupeids and pristigasteroids the posterior process of infraorbital 1 is small (e.g. *Pellonula*, *Gilchristella*, *Jenkinsia*, *Alosa*, *Clupea*, *Dorosoma*, *Pellona harroweri*, *Ilisha africana*) or absent (e.g. *Dussumieria*, *Etrumeus*). However, in the Chirocentridae and Engrauloidea, this process is strongly developed (Fig. 9C, D, E). In the Chirocentridae the posterior process of infraorbital 1 overlies the ventral margin of infraorbital 2 (Fig. 9C). In a small-sized examined *Chirocentrus dorab* (MZUSP 62467; 19.4 cm SL) the posterior process of infraorbital 1 and infraorbital 3 are separated by a short space. This is probably the usual condition for this character state in the Chirocentridae, as it was also illustrated by Ridewood (1904: fig. 119) and Bardack (1965: fig. 25). However, in one medium-sized skeleton of a *Chirocentrus dorab* (MZUSP 72930; 52 cm SL) there is a small area of

**Table 1.** Ratios of ribs to preural vertebrae (character 8 in the text), and distribution of fusion between the epicentrals and anterior ribs (character 9 in the text) in the genera of the Denticipitoidei and Pristigasteroidea

Taxa	Pleural ribs/ preural vertebrae	Epicentrals and anterior ribs	Taxa	Pleural ribs/ preural vertebrae	Epicentrals and anterior ribs
Denticipitoidei			Pristigasteroidea		
<i>Denticeps</i>	0.37–0.40*	Unfused	<i>Opisthopterus</i>	0.32–0.35*	Unfused
Pristigasteroidea			<i>Pellona</i>	0.33*–44*	Unfused
<i>Chirocentrodon</i>	0.41	Unfused	<i>Pliosteostoma</i>	0.35	Unfused
<i>Ilisha</i>	0.34–0.42*	Unfused	<i>Pristigaster</i>	0.33	Unfused
<i>Neoopisthopterus</i>	0.35*–0.41	Unfused	<i>Raconda</i>	0.28	Unfused
<i>Odontognathus</i>	0.28–0.32	Unfused	<i>Opisthopterus</i>	0.32–0.35*	Unfused

\*Values and/or taxa surveyed by Grande (1985) but not recorded in this study.

articulation between the posterior process of infraorbital 1 and infraorbital 3, such that infraorbital 2 is completely enclosed between those two bones. A similar condition was illustrated by Gregory (1933: fig. 34). Those observations indicate that the variation observed in the Chirocentridae is ontogenetic, with infraorbitals 1 and 3 articulating in fully-grown individuals. In the Engrauloidea, the posterior process of infraorbital 1 is further enlarged beyond the condition present in the Chirocentridae (Fig. 9D, E). The degree of development of the posterior process of infraorbital 1 and the disposition of bones in the infraorbital series of engrauloids result in a very unusual pattern of articulation between infraorbitals 1 to 3. Engrauloids have the posterior process of infraorbital 1 extending along most, or all, of the ventral margin of infraorbital 3. Consequently, infraorbital 2 is significantly enclosed between infraorbitals 1 and 3 (Fig. 9D, E).

The degree of development of the posterior process of infraorbital 1 of the Chirocentridae is clearly intermediate between that of *Denticeps*, the Pristigasteroidea and Clupeidae, on the one hand, and the Engrauloidea, on the other. Accordingly, the posterior process of infraorbital 1 extending along the ventral margin of infraorbital 2 is hypothesized to be apomorphic within Clupeiformes and indicative of a sister group relationship between the Chirocentridae and Engrauloidea (state 1). The presence of a relatively large area of articulation between infraorbitals 1 and 3, because of the further elongation of the posterior process of infraorbital 1, is hypothesized to be a synapomorphy of the Engrauloidea (state 2). On the one hand when present, the posterior process of infraorbital 1 projects underneath the anterior region of infraorbital 2 in the Engrauloidea, Pristigasteroidea, and Clupeidae. In the Chirocentridae, on the other hand, the posterior process of infraorbital 1 overlies the infraorbital 2. The latter condition is found exclusively in the Chirocentridae within the

Clupeiformes, and is consequently hypothesized to be autapomorphic for the family.

#### 8. Increase in ribs to preural vertebrae ratio

Values of this ratio in the genera of the Coiliidae (Engrauloidea) are low and similar to values observed in the Denticipitoidei and Pristigasteroidea (0.17–0.44; Tables 1 and 2). The range of this ratio in Engraulidae differs markedly from that observed in most Coiliidae (0.40–0.54; Table 2). Values of the ratio in representative Engraulidae are relatively high and also similar to values observed in Clupeidae (0.47–0.69; Table 3), particularly in species of the subfamily Pellonulinae. According to Grande (1985: 263), ‘this character is thought . . . to reflect a general trend of increasing relative abdominal cavity size among clupeomorphs’, a condition hypothesized as apomorphic in the Clupeiformes and proposed by Grande (1985: character 22) as the sole synapomorphy of the Clupeoidea. Grande (1985) explained the high values of ribs to preural vertebrae ratio in engrauloids as independently acquired by specialized New World forms (e.g. *Cetengraulis* and *Engraulis* species). If this is the case, then the Engrauloidea primitively had a low ribs to preural vertebrae ratio, and a high value of the ratio is indeed indicative of a sister group relationship between the Chirocentridae and Clupeidae. Nevertheless, most engraulids, including the basal *Stolephorus* and *Encrasicholina*, have a relatively high ratio of ribs to preural vertebrae. As a consequence, an alternative and equally parsimonious interpretation to the one proposed by Grande (1985) is that the Engrauloidea primitively had a relatively high ribs to preural vertebrae ratio. In this case, the Coiliidae secondarily reverted to the hypothesized plesiomorphic condition for the Clupeiformes of a low value of this ratio.

As hypotheses for either increases or decreases in the extent of abdominal cavity in the Engrauloidea are equally parsimonious, it is impossible to determine conclusively whether a low or a high ratio of ribs

**Table 2.** Ratios of ribs to preural vertebrae (character 8 in the text), and distribution of fusion between the epicentrals and anterior ribs (character 9 in the text) in the genera of the Chirocentridae and Engrauloidea (families Engraulidae and Coiliidae)

Taxa	Pleural ribs/ preural vertebrae	Epicentral and anterior ribs	Taxa	Pleural ribs/ preural vertebrae	Epicentral and anterior ribs
Clupeoidea			Engraulidae		
Chirocentridae			<i>Engraulis</i>	0.51*–0.54	Unfused
<i>Chirocentrus</i>	0.59	Fused	<i>Lycengraulis</i>	0.43*–0.45	Unfused
Engrauloidea			<i>Pterengraulis</i>	0.41	Unfused
Engraulidae			<i>Stolephorus</i>	0.46–0.50	Unfused
<i>Anchoa</i>	0.41*–0.51	Unfused	Coiliidae		
<i>Anchovia</i>	0.40	Unfused	<i>Coilia</i>	0.17–0.28*	Fused
<i>Anchoviella</i>	0.46–0.48	Unfused	<i>Lycotrissa</i>	0.33*–0.37	Unfused
<i>Cetengraulis</i>	0.44*–0.47	Unfused	<i>Setipinna</i>	0.29*–0.31	Unfused
<i>Encrasicholina</i>	0.46–0.52*	Unfused	<i>Thryssa</i>	0.41	Unfused

\*Values and/or taxa surveyed by Grande (1985) but not recorded in this study.

to preural vertebrae is the primitive state for this superfamily. If the Engrauloidea primitively had a high value of ribs to preural vertebrae ratio, then the character cannot be considered as evidence for a sister group relationship between the Chirocentridae and Clupeidae.

#### 9. Fusion of epicentrals with anterior ribs

In a survey of intermuscular bones and ligaments, Patterson & Johnson (1995) noticed that the fusion of epicentrals with the 6<sup>th</sup> to 20<sup>th</sup> anterior ribs is a feature almost unique to species of *Chirocentrus* and the genera of Clupeidae among teleosts they sampled. Such fusion does not occur in pristigasteroids and engrauloids they examined, except for *Coilia* (Coiliidae), in which only the first rib is enlarged and fused with an epicentral. They regarded *Coilia* as a specialized engrauloid, and proposed that the fusion of epicentrals with anterior ribs is additional evidence that Grande's (1985) Clupeoidea is a monophyletic assemblage.

The observations of Patterson & Johnson (1995) have been herein expanded to additional taxa. Epicentrals and ribs were found to be unfused in *Denticeps*, the Pristigasteroidea, and Engrauloidea with the exception of *Coilia* (Tables 1, 2). A fusion of epicentrals with ribs was alternatively found to occur in the Chirocentridae and in representatives of all currently accepted genera of the Clupeidae (Table 3). The distribution of this character state indicates that a fusion of epicentrals with ribs is indeed evidence for a monophyletic group composed of the Chirocentridae and Clupeidae within Clupeiformes.

## DISCUSSION

The distribution and common occurrence of five derived character states of the suspensorium, one of

the branchial arches, and one of the infraorbital series, support the hypothesis of a sister group relationship between the Chirocentridae and Engrauloidea (Fig. 10). The previous hypothesis of a sister group relationship between the Chirocentridae and Clupeidae [Grande's (1985) Clupeoidea] is supported by one non-ambiguous derived character state, the fusion of the epicentrals with anterior ribs (Grande, 1985; character 9 discussed above). The phylogenetic significance of the other evidence so far proposed for that relationship, an increase in ribs to preural vertebrae ratio (Patterson & Johnson, 1995; character 8 discussed above), requires further study because of its complex distribution in the Clupeoidei and the poorly resolved hypothesis of relationships within the Clupeidae and Engrauloidea. However, if that character is eventually demonstrated to be valid evidence of relationships, there is a much larger set of evidence for the hypothesis advocated herein. The most parsimonious distribution of all available character states strongly favours that the Chirocentridae and Engrauloidea are sister groups.

Circumstantial evidence corroborating this relationship is also present in the branchial arches of the Chirocentridae and representative Engrauloidea, particularly in the Coiliidae. Nelson (1967) first noticed a tendency of the dorsal parts of the arches of opposite sides (i.e. infrapharyngobranchials and epibranchials) to be in contact along the midline in the Clupeoidei. He observed this condition in most species of the Clupeidae, Engraulidae, and Pristigasteroidea. Infrapharyngobranchials and epibranchials of opposite sides are well separated along the dorsal midline in *D. clupeoides* and species of the Teleostei generally (Greenwood, 1968; Springer & Johnson, 2004; de Pinna & Di Dario, in press). Nelson (1967) also discovered that *Chirocentrus*, *Chirocentrodon* (Pristigas-

**Table 3.** Ratios of ribs to preural vertebrae (character 8 in the text), and distribution of fusion between the epicentrals and anterior ribs (character 9 in the text) in the genera of the Clupeidae (subfamilies Alosinae, Clupeinae, Dorosomatinae, Dussumieriinae, and Pellonulinae)

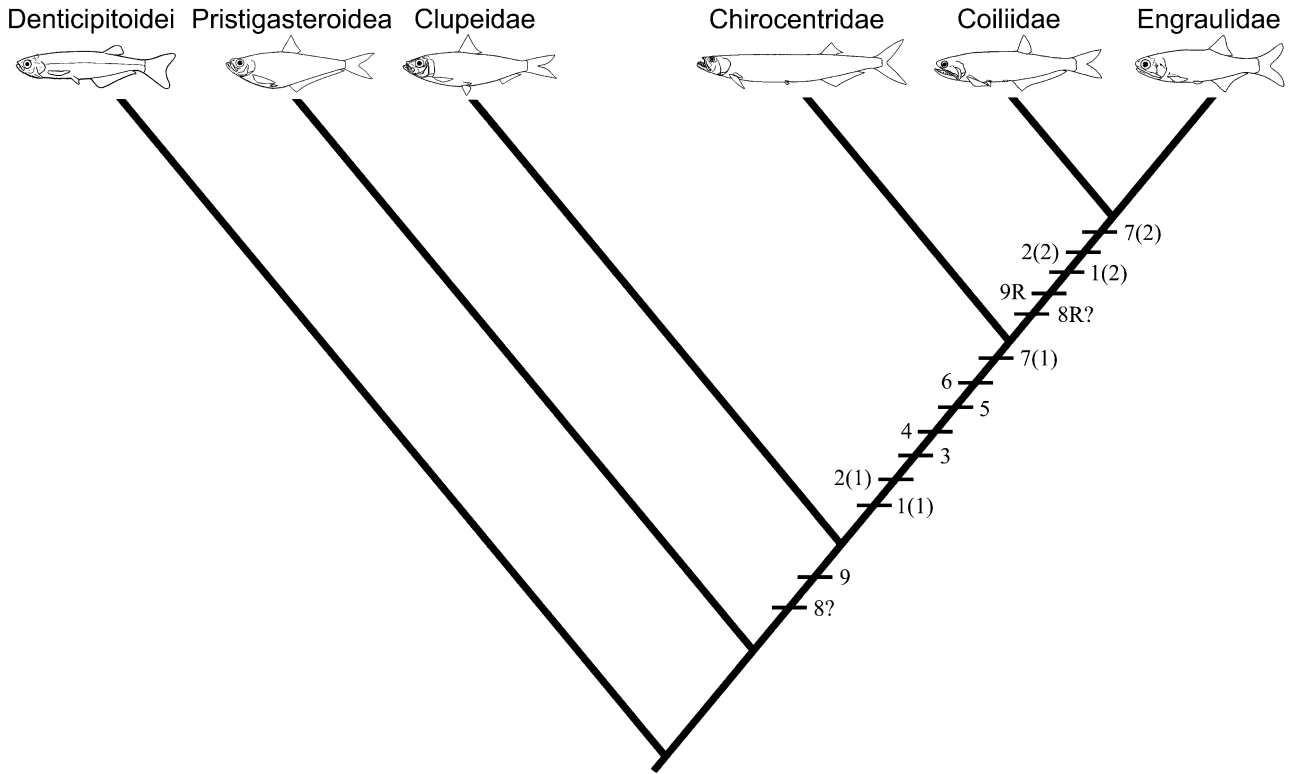
Taxa	Pleural ribs/ preural vertebrae	Epicentral and anterior ribs	Taxa	Pleural ribs/ preural vertebrae	Epicentral and anterior ribs
Clupeidae			Dorosomatinae		
Alosinae			Konosirus		
<i>Alosa</i>	0.55*–0.62	Fused	<i>Nematalosa</i>	0.66*	?
<i>Brevoortia</i>	0.54*–0.59	Fused	Dussumieriinae		
<i>Ethmalosa</i>	0.56*–0.64	Fused	<i>Dussumieria</i>	0.67	Fused
<i>Ethmidium</i>	0.63*	?	<i>Etrumeus</i>	0.70*–0.74	Fused
<i>Gudusia</i>	0.50*–0.51*	?	<i>Jenkinsia</i>	0.48–0.61*	Fused
<i>Hilsa</i>	0.56*–0.58	Fused	<i>Spratelloides</i>	0.65*–0.69	Fused
<i>Tenualosa</i>	0.58	Fused	Pellonulinae		
Clupeinae			<i>Clupeichthys</i>	0.51*–0.55	Fused
<i>Amblygaster</i>	0.57*–0.68	Fused	<i>Clupeoides</i>	0.50*–0.55	fused
<i>Clupea</i>	0.64*–0.69	Fused	<i>Congothrissa</i>	0.47*	?
<i>Clupeonella</i>	0.56*–0.60	Fused	<i>Corica</i>	0.49*	?
<i>Escualosa</i>	0.55	Fused	<i>Cynothrissa</i>	0.51*–0.55	Fused
<i>Harengula</i>	0.58*–0.61	Fused	<i>Ehirava</i>	0.56*	?
<i>Herklotsichthys</i>	0.60*–0.62	Fused	<i>Gilchristella</i>	0.51	fused
<i>Lile</i>	0.56*–0.58	Fused	<i>Hyperlophus</i>	0.49*–0.63	fused
<i>Opisthonema</i>	0.60*–0.65	Fused	<i>Laeviscutella</i>	0.52	fused
<i>Platanichthys</i>	0.53	Fused	<i>Limnothrissa</i>	0.59*	?
<i>Ramnogaster</i>	0.52*–0.57*	?	<i>Microthrissa</i>	0.47*–0.55	Fused
<i>Rhinosardinia</i>	0.55*–0.56	Fused	<i>Nannothrissa</i>	0.52*–0.57	Fused
<i>Sardina</i>	0.60*–0.64	Fused	<i>Odaxothrissa</i>	0.52*–0.53*	?
<i>Sardinella</i>	0.59*–0.66	Fused	<i>Pellonula</i>	0.50*–0.54	Fused
<i>Sardinops</i>	0.62*–0.66	Fused	<i>Poecilothrissa</i>	0.53*	?
<i>Sprattus</i>	0.58*–0.62	Fused	<i>Potamalosa</i>	0.61	Fused
<i>Strangomera</i>	0.59*	?	<i>Potamothrissa</i>	0.55*–0.57	Fused
Dorosomatinae			<i>Sierrathrissa</i>	0.56*	?
<i>Anodontostoma</i>	0.55*–0.60	Fused	<i>Spratellomorpha</i>	0.53*	?
<i>Clupanodon</i>	0.58*–0.67	Fused	<i>Stolothrissa</i>	0.59*	?
<i>Dorosoma</i>	0.49*–0.54	Fused	<i>Thrattidion</i>	0.47*	?

The pellonulines *Sauvagella* and *Dayella* were not examined. Values and/or taxa surveyed by Grande (1985) but not recorded in this study indicated by “\*”; “?” for doubtful state, taxa not surveyed in this study.

teroidea), and *Odaxothrissa* (Pellonulinae) are exceptions among the Clupeoidei, because the dorsal elements of their arches do not come together in the midline. Nelson (1967: 394) suggested that the separation between opposite dorsal elements of the branchial arches was independently acquired in these three genera. He argued that the attachment of gill arch elements along the dorsal midline prevents expansion of the pharynx for the swallowing of large prey, and consequently tends to be absent from predatory fishes such as species of those three genera. Current understanding of Clupeoidei intrarelationships corroborates the hypothesis that the separation between opposite dorsal elements of the branchial arches was independently acquired in *Chirocentrus*, *Chirocentrodon*, and *Odaxothrissa*. *Chirocentrodon* is

hypothesized to be the sister group of *Neoopisthopterus* in a distal clade within the Pristigasteroidea (Di Dario, 1999). *Odaxothrissa* is undoubtedly related to other species of the Pellonulinae (Grande, 1985; Stiassny, 2002), where the dorsal elements of the branchial arches of opposite sides are in contact along the midline as in the typical condition present in the Clupeidae.

Nelson (1970) noticed that the dorsal paired elements of the branchial arches of *Coilia* are also separated. In fact, that condition is equally pronounced in the Chirocentridae and *Coilia*. The upper arches of other examined species of the Coiliidae are widely separated posteriorly, but converge to contact anteriorly in the midline (de Pinna & Di Dario, in press). If the Chirocentridae and Engrauloidea are sister



**Figure 10.** Cladogram showing the most parsimonious hypothesis of phylogenetic relationships between the Denticipitoidei and groups of Clupeoidei based on characters discussed in text and Di Dario (2002). For characters 1, 2, and 7, state 0 represents the plesiomorphic condition (not indicated); state 1 is intermediate between states 0 and 2, synapomorphic for the clade composed by the Chirocentridae and Engrauloidea; state 2 is hypothesized as a further modification of state 1, synapomorphic for the Engrauloidea. Characters 3–6, 8, and 9, are binary, with only the hypothesized apomorphic condition indicated. Character 8 is ambiguous (as indicated by the question mark). One possible interpretation of the evolution of characters 8 and 9 is shown. In this hypothesis, both are synapomorphies of the clade composed of Clupeidae, Chirocentridae, and Engrauloidea, reversed in the Engrauloidea (indicated by ‘R’ accordingly). Character states are [1(1)] anterior margin of metapterygoid located anterior to quadrate, [1(2)] a substantial portion of metapterygoid situated anterodorsal to quadrate, [2(1)] ventral limb of hyomandibula and quadrate not separated by the metapterygoid, [2(2)] articulation between the ventral limb of hyomandibula and quadrate, (3) posterodorsal margin of metapterygoid in line with the condyle of articulation of the hyomandibula with the opercle, (4) presence of a laminar outgrowth of the anterior margin of quadrate, (5) endochondral portion of quadrate in the shape of an isosceles triangle, (6) the presence and arrangement of autogenous tooth plates on ceratobranchials 1 to 3, [7(1)] posterior region of infraorbital 1 well-developed and extending along the ventral margin of infraorbital 2, [7(2)] infraorbitals 1 and 3 articulating by means of a well-developed laminar process of the posterior region of infraorbital 1, (8) increase in ribs to preural vertebrae ratio, and (9), fusion of epicentrals with anterior ribs. Line art drawings after Whitehead *et al.* (1988), Munroe *et al.* (1999) and Nelson (2006).

groups, one possible interpretation is that the separation between dorsal elements of the branchial arches is a synapomorphy for that assemblage, convergent in the clade composed of *Chirocentrodon* and *Neoopisthopterus* in the Pristigasteroidea, and *Odaxothrissa* in the Pellonulinae. A reversion to the primitive state of the Clupeoidei, where all dorsal paired elements of the gill arches are close in the midline, would thus be necessary to account for the condition in the Engraulidae. The predatory habits of the Chirocentridae, Coiliidae, and of several probably unspe-

cialized species of the Engraulidae, suggest that macrophagy is the primitive condition for the clade Chirocentridae + Engrauloidea. In this scenario, microphagy in the Engraulidae would also be secondary and consequently convergent in relation to other species of the Clupeoidei.

The suspensorium of the Chirocentridae is unique in being composed of several morphological traits intermediate between those of generalized Clupeiformes on the one hand and the Engrauloidea on the other. A substantial portion of the evidence presented

herein for a sister group relationship between the Chirocentridae and Engrauloidea results from the decomposition of a single character complex, the obliquely inclined suspensorium of the Engrauloidea, into five discrete morphological components. Morphological components of a character complex are functionally integrated and, as such, must share variable degrees of interdependence as discussed in a series of studies (e.g., Naylor & Adams, 2001; Rieppel & Kearney, 2002; Rieppel, 2005; Kearney & Rieppel, 2006; Sereno, 2007). To decompose a functionally integrated character complex into its constituent morphological components, and to interpret each bit of the resulting information as evidence for or against a relationship is not incongruent with the principle that characters in a phylogenetic analysis must be independent. As Grant & Kluge (2004: 26) pointed out, 'what matters in individuating character[s] . . . is not the structural, developmental, or functional independence of a part, but its historical/transformational independence'. In other words, functional or developmental interdependence of parts in a character complex does not necessarily imply that all of its morphological components evolved simultaneously, as has been shown in several recent studies (e.g. Fink & Fink, 1981, 1996; Baker, Novacek & Simmons, 1991; O'Leary & Geisler, 1999; Hilton, 2001, 2003; O'Leary, Gatesy & Novacek, 2003; Di Dario, 2004; Grande & de Pinna, 2004; Di Dario & de Pinna, 2006; Schultz, 2007). To treat complex characters as a single binary character under the premise that they constitute a single evolutionary unit overlooks potential phylogenetic information (Fink & Fink, 1996; Hilton, 2001; Di Dario, 2004; Schultz, 2007). What must be avoided in the process of character argumentation is the obvious inconsistency of using logically synonymous characters as separate sources of evidence (Grant & Kluge, 2004). As succinctly expressed by Farris (1983: 20), ' . . . there is no point to using both number of tarsal segments and twice that number as characters'. Additionally, the presence of the character complex and its individual morphological components should not both be used in a phylogenetic analysis, in order not to increase artificially the support of a given clade (Hilton, 2001, 2003).

It also might be argued that the apomorphic states of the suspensorium shared by the Chirocentridae and Engrauloidea evolved independently in both groups in response to similar feeding habits. The peculiar morphology of the suspensorium shared by the Chirocentridae and Engrauloidea is, however, not strictly associated with a specific diet within those groups. The Engraulini, for instance, includes predaceous genera such as *Pterengraulis* and *Lycengraulis*, planktivores such as *Engraulis* (which is also egg cannibalistic), and highly specialized microplanktivores such as *Ceten-*

*graulis* and *Anchovia* (Bayliff, 1963; de Ciechomski, 1967; Leong & O'Connell, 1969; Blaber, 1979; James, 1988; Whitehead *et al.*, 1988; James & Findlay, 1989; Wongratana *et al.*, 1999; Gay *et al.*, 2002; Krumme *et al.*, 2005; Pájaro *et al.*, 2007). Despite this enormous interspecific variation in diet, the apomorphic states of the suspensorium shared by the Chirocentridae and Engrauloidea are present in all examined species of these assemblages with the exception of highly paedomorphic members of the Engraulidae, such as *Ama-zonsprattus scintilla* (Roberts, 1984; Nelson, 1986).

Constraints related to feeding could severely limit the range of possible morphologies of component characters of the suspensorium of the Clupeoidei, increasing the likelihood of convergent evolution. One of the presumed consequences of such constraints would be the occurrence of those same character states in other predatory or macrophagous species of the Clupeoidei, which is the trophic category hypothesized as primitive for the Chirocentridae and Engrauloidea. As discussed in the character section, conditions similar to character states 1 and 2 were observed in a few species of the Alosinae, which are microphagous and consequently have the opposite diet in terms of size of ingested items. Character states 3 to 5 are exclusive of the Chirocentridae and Engrauloidea among the Clupeoidei. Compelling evidence against the role of feeding-related constraints in the shaping of bones of the suspensorium of the Chirocentridae and Engrauloidea is found in the pristigasteroid *Chirocentrodon bleekermanus*. Like the Chirocentridae and primitively in the Engrauloidea, the carnivorous *C. bleekermanus* feeds on relatively large fishes, such as other clupeoids and shrimps up to 50% its own standard length (SL) (Sazima, Moura & Sazima, 2004; Corrêa, de Tarso Chaves & Guimarães, 2005). The Chirocentridae and *C. bleekermanus* share the presence of canine-like and relatively well-developed teeth, and the separation of the dorsal paired elements of their branchial arches among other general anatomical features presumably related to a predaceous diet. Despite the co-occurrence of these conditions, none of the five apomorphic character states of the suspensorium of the Chirocentridae and Engrauloidea discussed herein occurs in *C. bleekermanus*.

Morphological, functional, developmental, or evolutionary constraints related to feeding are insignificant or of secondary importance to the peculiar morphology of the suspensorium of the Chirocentridae and Engrauloidea according to the evidence presented. Consequently, to presume that the hypothesized apomorphic states of the suspensorium shared by the Chirocentridae and Engrauloidea evolved independently in both groups in response to similar diets is empirically unsubstantiated. Unsubstantiated ad hoc assumptions like that have been banned from phylo-



genetic argumentation early in the settling of cladistics as the current paradigm in systematics, in accordance with the logical notion that homology should be presumed in the absence of evidence to the contrary (e.g. Hennig, 1966; Wiley, 1975, 1981; Nelson, 1978; Patterson, 1982; Farris, 1983).

One direct outcome of the hypothesis of a sister group relationship between the Chirocentridae and Engrauloidea is that the Clupeoidea, as currently composed of the Chirocentridae and Clupeidae (Grande, 1985), is not a monophyletic group. Consequently, the classification of the Clupeoidei must be modified. A current estimate of higher-level relationships among subgroups of the Clupeoidei, summarized in Figure 10, can be expressed in the following sequenced classification (Nelson, 1972):

Pristigasteroidea  
 Pristigasteridae  
 Clupeoidea  
 Clupeidae  
 Engrauloidea  
 Chirocentridae  
 Engraulidae

In this revised classification, the Pristigasteroidea includes the Pristigasteridae and Pellonidae of Grande (1985) in an expanded Pristigasteridae, following de Pinna & Di Dario (2003). The Clupeoidea, previously composed of the Chirocentridae and Clupeidae, is restricted to the Clupeidae. The Engrauloidea include Grande's (1985) Engrauloidea and the Chirocentridae. The Engraulidae and Coiliidae are recognized as subfamilies Engraulinae and Coiliinae in the Engraulidae. The subfamily status of Engraulidae and Coiliidae, with the generic composition of both subfamilies as those given by Grande & Nelson (1985), was originally proposed by J. Nelson (1994). Although not universally adopted (e.g. Di Dario, 2002, 2004; Di Dario & de Pinna, 2006), it has been extensively used in several recent publications (e.g. Wongratana *et al.*, 1999; Nizinski & Munroe, 2002; Nelson, 2006; Lavoué *et al.*, 2007; Li & Ortí, 2007). Nelson's (1994) classification of the Engrauloidea is appropriate in light of the phylogenetic hypothesis presented herein. The Engraulinae includes *Stolephorus* and an unnamed clade composed of *Encracisholina* and the tribe Engraulini of Grande & Nelson (1985). Relationships within the Coiliinae are totally unknown (Grande & Nelson, 1985: fig. 4).

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#### REFERENCES

- Arratia G. 1997.** Basal teleosts and teleostean phylogeny. *Palaeo Ichthyologica* **7**: 1–168.
- Arratia G. 1999.** The monophyly of Teleostei and stem-group teleosts. In: Arratia G, Schultze H-P, eds. *Mesozoic fishes 2 – systematics and fossil record*. München: Verlag Dr Friedrich Pfeil, 265–334.
- Arratia G, Schultze H-P. 1991.** Palatoquadrate and its ossifications: development and homology within osteichthyans. *Journal of Morphology* **208**: 1–81.
- Baker RJ, Novacek MJ, Simmons NB. 1991.** On the monophyly of bats. *Systematic Zoology* **40**: 216–231.
- Bardack D. 1965.** Anatomy and evolution of chirocentrid fishes. *The University of Kansas Paleontological Contributions (Vertebrata)* **10**: 1–88.
- Bayliff WH. 1963.** The food and feeding habits of the anchoveta, *Cetengraulis mysticetus*, in the Gulf of Panama. *Bulletin of the Inter-American Tropical Tuna Commission* **7**: 399–459.
- Blaber SJM. 1979.** The biology of filter feeding teleosts in Lake St. Lucia, Zululand. *Journal of Fish Biology* **15**: 37–59.
- Bornbusch AH. 1988.** Gill raker morphologies of anchovies (Teleostei: Engraulidae) from the Río Orinoco, Venezuela. *Copeia* **1988**: 174–182.
- Bornbusch AH, Lee M. 1992.** Gill raker structure and development in Indo-Pacific anchovies (Teleostei: Engrauloidea), with a discussion of the structural evolution of engrauloid gill rakers. *Journal of Morphology* **214**: 109–119.
- Cavender T. 1966.** The caudal skeleton of the Cretaceous teleosts *Xiphactinus*, *Ichthyodectes*, and *Gillicus*, and its bearing on their relationships with *Chirocentrus*. *Occasional Papers of the Museum of Zoology, University of Michigan, Ann Arbor* **650**: 1–15.
- Cervigón F. 1982.** La ictiofauna estuarina del Caño Mánamo y áreas adyacentes. In: Novoa D, ed. *Los recursos pesqueros del Río Orinoco y sur explotación*. Caracas: Corporación Venezolana Guyana, 205–260.
- Chang M-M, Maisey JG. 2003.** Redescription of *Ellimma branneri* and *Diplomystus shengliensis*, and relationships

- of some basal clupeomorphs. *American Museum Novitates* **3404**: 1–35.
- de Ciechowski JD. 1967.** Investigations of food and feeding habits of larvae and juveniles of the Argentine anchovy *Engraulis anchoita*. *California Cooperative Oceanic Fisheries Investigations* **11**: 72–81.
- Corrêa CE, de Tarso Chaves P, Guimarães PRB. 2005.** Biology of *Chirocentron bleekermanus* (Poey, 1867) (Clupeiformes: Pristigasteridae) in a continental shelf region of Southern Brazil. *Brazilian Archives of Biology and Technology* **48**: 419–427.
- Di Dario F. 1999.** Filogenia de Pristigasteroidea (Teleostei, Clupeomorpha). Unpublished MS Dissertation, Universidade de São Paulo.
- Di Dario F. 2002.** Evidence supporting a sister group relationship between Clupeoidea and Engrauloidea. *Copeia* **2002**: 496–503.
- Di Dario F. 2004.** Homology between the *recessus lateralis* and cephalic sensory canals, with the proposition of additional synapomorphies for the Clupeiformes and the Clupeoidei. *Zoological Journal of the Linnean Society* **141**: 257–270.
- Di Dario F, de Pinna MCC. 2006.** The supratemporal system and the pattern of ramification of cephalic sensory canals in *Denticeps clupeoides* (Denticipitoidei, Teleostei): additional evidence for monophyly of Clupeiformes and Clupeoidei. *Papéis Avulsos de Zoologia, Museu de Zoologia da Universidade de São Paulo* **46**: 107–123.
- Farris SJ. 1983.** The logical basis of phylogenetic analysis. In: Platnick NI, Funk VA, eds. *Advances in cladistics*, Vol. 2. New York: Columbia University Press, 7–36.
- Fink SV, Fink WL. 1981.** Interrelationships of the ostariophysan fishes (Teleostei). *Zoological Journal of the Linnean Society* **72**: 297–353.
- Fink SV, Fink WL. 1996.** Interrelationships of the ostariophysan fishes. In: Stiassny MLJ, Parenti LR, Johnson GD, eds. *Interrelationships of Fishes*. San Diego: Academic Press, 209–249.
- Forskål P. 1775.** *Descriptiones animalium avium, amphibiorum, piscium, insectorum, vermium; quae in itinere Orientali observavit Petrus Forskål*. Hauniae: Mölleri.
- Fowler HW. 1931.** A synopsis of the fishes of China, part II. The herrings and related fishes. *Hong Kong Naturalist* **2**: 49–59.
- Gay D, Bassani C, Sergipense S. 2002.** Diel variation and selectivity in the diet of *Cetengraulis edentulus* (Cuvier 1828) (Engraulidae-Clupeiformes) in the Itaipu Lagoon, Niterói, Rio de Janeiro. *Atlântica, Rio Grande* **24**: 59–68.
- Gosline WA. 1960.** Contributions toward a classification of modern isospondylous fishes. *Bulletin of the British Museum, Natural History* **6**: 327–365.
- Grande L. 1985.** Recent and fossil clupeomorph fishes with materials for revision of the subgroups of clupeoids. *Bulletin of the American Museum of Natural History* **181**: 231–372.
- Grande L, Nelson G. 1985.** Interrelationships of fossil and Recent anchovies (Teleostei: Engrauloidea) and description of a new species from the Miocene of Cyprus. *American Museum Novitates* **2826**: 1–16.
- Grande T, de Pinna MCC. 2004.** The evolution of the Webberian apparatus: a phylogenetic perspective. In: Arratia G, Tintori A, eds. *Mesozoic fishes 3 – systematics, paleoenvironments and biodiversity*. München: Verlag Dr Friedrich Pfeil, 429–448.
- Grant T, Kluge AK. 2004.** Transformational series as an ideographic character concept. *Cladistics* **20**: 23–31.
- Greenwood PH. 1968.** The osteology and relationships of the Denticipitidae, a family of clupeomorph fishes. *Bulletin of the British Museum (Natural History) Zoology* **16**: 215–273.
- Greenwood PH, Rosen DE, Weitzman SH, Myers GS. 1966.** Phyletic studies of teleostean fishes, with a provisional classification of living forms. *Bulletin of the American Museum of Natural History* **131**: 339–456.
- Gregory W. 1933.** Fish skulls: a study of the evolution of natural mechanisms. *Transactions of the American Philosophical Society* **23**: 75–481.
- Hardenberg JDF. 1934.** Some remarks on the genus *Stolephorus* Lacépède in the Indo-Australian Archipelago. *Treubia* **14**: 313–375.
- Hennig W. 1966.** *Phylogenetic systematics*. Urbana: University of Illinois Press.
- Hilton EJ. 2001.** Tongue bite apparatus of osteoglossomorph fishes: variation of a character complex. *Copeia* **2001**: 372–381.
- Hilton EJ. 2003.** Comparative osteology and phylogenetic systematics of fossil and living bony-tongue fishes (Actinopterygii, Teleostei, Osteoglossomorpha). *Zoological Journal of the Linnean Society* **137**: 1–100.
- Inoue JG, Miya M, Tsukamoto K, Nishida M. 2003.** Basal actinopterygian relationships: a mitogenomic perspective on the phylogeny of the ‘ancient fish’. *Molecular Phylogenetics and Evolution* **26**: 110–120.
- Ishiguro NB, Miya M, Nishida M. 2003.** Basal euteleostean relationships: a mitogenomic perspective on the phylogenetic reality of the ‘Protacanthopterygii’. *Molecular Phylogenetics and Evolution* **27**: 476–488.
- Ishiguro NB, Miya M, Nishida M. 2005.** *Sundasalanx* (Sundasalanxidae) is a progenetic clupeiform, not a closely-related group of salangids (Osmeriformes): mitogenomic evidence. *Journal of Fish Biology* **67**: 561–569.
- James AG. 1988.** Are clupeid microphagists herbivorous or omnivorous? A review of the diets of some commercially important clupeids. *South African Journal of Marine Science* **7**: 161–177.
- James AG, Findlay KP. 1989.** Effect of particle size and concentration on feeding behavior, selectivity and rates of food ingestion by the Cape anchovy *Engraulis capensis*. *Marine Ecology Progress Series* **50**: 275–294.
- Johnson GD, Patterson C. 1996.** Relationships of lower euteleostean fishes. In: Stiassny MLJ, Parenti LR, Johnson GD, eds. *Interrelationships of fishes*. San Diego: Academic Press, 251–330.
- Kearney MM, Rieppel O. 2006.** Rejecting ‘the given’ in systematics. *Cladistics* **22**: 369–377.
- Krumme U, Keuthen H, Barletta M, Villwock W, Saint-Paul U. 2005.** Contribution to the feeding ecology of the predatory wingfin anchovy *Pterengraulis atherinoides* (L.)

- in north Brazilian mangrove creeks. *Journal of Applied Ichthyology* **21**: 469–477.
- Lavoué S, Miya M, Inoue JG, Saitoh K, Ishiguro NB, Nishida M. 2005.** Molecular systematics of the gonorynchiform Fishes (Teleostei) based on whole mitogenome sequences: implications for higher-level relationships within the Otocephala. *Molecular Phylogenetics and Evolution* **37**: 165–177.
- Lavoué S, Miya M, Saitoh K, Ishiguro NB, Nishida M. 2007.** Phylogenetic relationships among anchovies, sardines, herrings and their relatives (Clupeiformes), inferred from whole mitogenome sequences. *Molecular Phylogenetics and Evolution* **43**: 1096–1105.
- Lê HL, Lecoindre G, Perasso R. 1993.** A 28S rRNA-based phylogeny of the gnathostomes: first steps in the analysis of conflict and congruence with morphologically based cladograms. *Molecular Phylogenetics and Evolution* **2**: 31–51.
- Lecoindre G, Nelson G. 1996.** Clupeomorpha, sister group of Ostariophysi. In: Stiassny MLJ, Parenti LR, Johnson GD, eds. *Interrelationships of fishes*. San Diego: Academic Press, 193–207.
- Leong RJH, O'Connell CP. 1969.** A laboratory study of particulate and filter feeding of the Northern anchovy (*Engraulis mordax*). *Journal of Fisheries Research Board of Canada* **26**: 557–582.
- Li C, Ortí G. 2007.** Molecular phylogeny of Clupeiformes (Actinopterygii) inferred from nuclear and mitochondrial DNA sequences. *Molecular Phylogenetics and Evolution* **44**: 386–398.
- Milton DA, Blaber SJM, Rawlinson NJF. 1990.** Diet and prey selection of six species of tuna baitfish in three coral reef lagoons in the Solomon Islands. *Journal of Fish Biology* **37**: 205–224.
- Munroe TA, Nizinski MS, Wongratana T. 1999.** Family Chirocentridae. In: Carpenter KE, Niem VH, eds. *FAO species identification guide for fishery purposes. The living marine resources of the Western Central Pacific*, Vol. 3. Batoid fishes, chimaeras and bony fishes part 1 (Elopidae to Linophrynidae), 1397–2068. Rome: FAO, 1771–1774.
- Naylor GJP, Adams DC. 2001.** Are the fossil data really at odds with the molecular data? Morphological evidence for Cetartiodactyla phylogeny reexamined. *Systematic Biology* **50**: 444–453.
- Nelson GJ. 1967.** Gill arches of teleostean fishes of the family Clupeidae. *Copeia* **1967**: 389–399.
- Nelson GJ. 1969a.** Gill arches and the phylogeny of fishes, with notes on the classification of vertebrates. *Bulletin of the American Museum of Natural History* **141**: 474–552.
- Nelson GJ. 1969b.** Infraorbital bones and their bearing on the phylogeny and geography of osteoglossomorph fishes. *American Museum Novitates* **2394**: 1–37.
- Nelson GJ. 1970.** The hyobranchial apparatus of teleostean fishes of the families Engraulidae and Chirocentridae. *American Museum Novitates* **2410**: 1–30.
- Nelson GJ. 1972.** Phylogenetic relationship and classification. *Systematic Zoology* **21**: 227–231.
- Nelson GJ. 1978.** Ontogeny, phylogeny, paleontology, and the biogenetic law. *Systematic Zoology* **27**: 324–345.
- Nelson GJ. 1984.** Notes on the rostral organ of anchovies (Family Engraulidae). *Japanese Journal of Ichthyology* **31**: 86–87.
- Nelson GJ. 1986.** Identity of the anchovy *Engraulis clarki* with notes on the species-groups of *Anchoa*. *Copeia* **1986**: 891–902.
- Nelson JS. 1994.** *Fishes of the world*, 3rd edn. New York: John Wiley & Sons, Inc.
- Nelson JS. 2006.** *Fishes of the world*, 4th edn. Hoboken, NJ: John Wiley & Sons, Inc.
- Nizinski MS, Munroe T. 2002.** Family Engraulidae. In: Carpenter KE, ed. *FAO species identification guide for fishery purposes and American Society of Ichthyologists and Herpetologists Special Publication No 5. The living marine resources of the Western Central Atlantic*. Vol. 2. Bony fishes, part 1 (Acipenseridae to Grammatidae), 601–1373. Rome: FAO, 764–794.
- O'Leary MA, Gatesy J, Novacek MJ. 2003.** Are the dental data really at odds with the molecular data? Morphological evidence for whale phylogeny (re)reexamined. *Systematic Biology* **52**: 853–864.
- O'Leary MA, Geisler JH. 1999.** The position of Cetacea within Mammalia: phylogenetic analysis of morphological data from extinct and extant taxa. *Systematic Biology* **48**: 455–490.
- Pájaro M, Curelovich J, Macchi G. 2007.** Egg cannibalism in the northern population of the Argentine anchovy, *Engraulis anchoita* (Clupeidae). *Fisheries Research* **83**: 253–262.
- Patterson C. 1982.** Morphological characters and homology. In: Joysey KA, Friday AE, eds. *Problems of phylogenetic reconstruction*. London: Academic Press, 21–74.
- Patterson C, Johnson GD. 1995.** The intermuscular bones and ligaments of teleostean fishes. *Smithsonian Contributions to Zoology* **559**: 1–78.
- Patterson C, Rosen DE. 1977.** Review of ichthyodectiform and other Mesozoic teleost fishes and the theory and practice of classifying fossils. *Bulletin of the American Museum of Natural History* **158**: 85–172.
- de Pinna MCC. 1996.** Teleostean monophyly. In: Stiassny MLJ, Parenti LR, Johnson GD, eds. *Interrelationships of fishes*. San Diego: Academic Press, 147–162.
- de Pinna MCC, Di Dario F. 2003.** Family Pristigasteridae (Pristigasterids). In: Reis RE, Kullander SO, Ferraris CJ Jr, eds. *Check list of freshwater fishes of South and Central America (CLOFFSCA)*. Porto Alegre: EDIPUCRS, 43–45.
- de Pinna MCC, Di Dario F. In press.** The branchial arches of the primitive clupeomorph fish, *Denticeps clupeoides*, and their phylogenetic implication (Clupeiformes, Denticipitidae). In: Schultze H-P, ed. *Festschrift in honor of G. Arratia*. Munchen: Verlag Dr. F. Pfeil, in press.
- Ridewood WG. 1904.** On the cranial osteology of the clupeoid fishes. *Proceedings of the Zoological Society of London* **2**: 448–493.
- Rieppel O. 2005.** The philosophy of total evidence and its relevance for phylogenetic inference. *Papéis Avulsos de Zoologia, Museu de Zoologia da Universidade de São Paulo* **45**: 77–89.

- Rieppel O, Kearney M. 2002.** Similarity. *Biological Journal of the Linnean Society* **75**: 59–82.
- Roberts T. 1984.** *Amazonsprattus scintilla*, new genus and species from the Rio Negro, Brazil, the smallest known clupeomorph fish. *Proceedings of the California Academy of Sciences* **43**: 317–321.
- Saint-Seine P. 1949.** Les poissons des calcaires lithographiques de Cerin (Ain). *Nouvelles Archives du Muséum d'Histoire Naturelle de Lyon* **2**: 1–357.
- Sazima C, Moura RL, Sazima I. 2004.** *Chirocentrodon bleekermanus* (Teleostei: Clupeiformes: Pristigasteridae), a small predaceous herring with folded and distinctively oriented prey in stomach. *Brazilian Journal of Biology* **64**: 165–168.
- Schultz JW. 2007.** A phylogenetic analysis of the arachnid orders based on morphological characters. *Zoological Journal of the Linnean Society* **150**: 221–265.
- Segura V, Díaz de Astarloa JM. 2004.** Análisis osteológico de la saraca *Brevoortia aurea* (Spix) (Actinopterygii: Clupeidae) en el Atlántico suroccidental. *Revista de Biología Marina y Oceanografía* **39**: 37–52.
- Sereno PC. 2007.** Logical basis for morphological characters in phylogenetics. *Cladistics* **23**: 565–587.
- Siebert DJ. 1987.** Interrelationships among families of the order Cypriniformes (Teleostei). Unpublished PhD Thesis, The City University of New York.
- Song J, Parenti LR. 1995.** Clearing and staining whole fish specimens for simultaneous demonstration of bone, cartilage and nerves. *Copeia* **1995**: 114–118.
- Springer VG, Johnson GD. 2004.** Study of the dorsal gill-arch musculature of teleostome fishes, with special reference to the Actinopterygii. *Bulletin of the Biological Society of Washington* **11**: 1–235.
- Stiassny ML. 2002.** Revision of Sauvagella Bertin (Clupeidae; Pellonulinae; Ehiravini) with a description of a new species from the freshwaters of Madagascar and diagnosis of the Ehiravini. *Copeia* **2002**: 67–76.
- Taylor WR, Van Dyke GA. 1985.** Revised procedures for staining and clearing small fishes and other vertebrates for bone and cartilage study. *Cybio* **9**: 107–119.
- Weitzman SH. 1962.** The osteology of *Brycon meeki*, a generalized characid fish, with an osteological definition of the family. *Stanford Ichthyological Bulletin* **8**: 1–77.
- Whitehead PJP. 1963.** A contribution to the classification of clupeoid fishes. *Annals and Magazine of Natural History* **13**: 737–750.
- Whitehead PJP. 1985.** FAO species catalogue. Clupeoid fishes of the world (suborder Clupeoidei). An annotated and illustrated catalogue of the herrings, sardines, pilchards, sprats, anchovies and wolfherrings. Part 1 – Chirocentridae, Clupeidae and Pristigasteridae. *FAO Fisheries Synopsis* **7**: 1–303.
- Whitehead PJP, Nelson GJ, Wongratana T. 1988.** FAO species catalogue. Clupeoid fishes of the world (suborder Clupeoidei). An annotated and illustrated catalogue of the herrings, sardines, pilchards, sprats, anchovies and wolfherrings. Part 2 – Engraulididae. *FAO Fisheries Synopsis* **7**: 305–579.
- Whitehead PJP, Teugels GG. 1985.** The West African pygmy herring *Sierrathrissa leonensis*: general features, visceral anatomy, and osteology. *American Museum Novitates* **2835**: 1–44.
- Wiley EO. 1975.** Karl R. Popper, systematic, and classification: a reply to Walter Bock and other evolutionary taxonomists. *Systematic Zoology* **24**: 233–243.
- Wiley EO. 1981.** *Phylogenetics: the theory and practice of phylogenetic systematics*. New York: John Wiley & Sons.
- Wongratana T, Munroe TA, Nizinski MS. 1999.** Family Engraulidae. In: Carpenter KE, Niem VH, eds. *FAO species identification guide for fishery purposes. The living marine resources of the Western Central Pacific*, Vol. 3. Batoid fishes, chimaeras and bony fishes part 1 (Elopidae to Linophrynidae), 1397–2068. Rome: FAO, 1698–1753.
- Woodward AS. 1912.** The fossil fishes of the English Chalk. *Palaeontographical Society, London* **56**: 1–264.
- Woodward AS. 1942.** Some new and little known Upper Cretaceous fishes from Mount Lebanon. *Annals and Magazine of Natural History* **9**: 537–568.

## APPENDIX

## COMPARATIVE MATERIAL EXAMINED

Number of examined specimens follows catalogue number:

Denticipitidae: *Denticeps clupeoides*, MZUSP 62480, 1; MZUSP 84776, 20.

Chirocentridae: *Chirocentrus dorab*, MZUSP 62467, 1; MZUSP 72930, 1(skl); USNM 359341, 1.

Clupeidae: *Alosa caspia*, USNM 143891, 1; *A. pseudoharengus*, MZUSP 62471, 1; *Anodontostoma chacunda*, MZUSP 62476, 1; *Brevoortia aurea*, MZUSP 11729, 1; *B. pectinata*, MCP 7722, 1; MCP 7725, 1; MCP 7727, 1; *Clupea harengus*, ANSP 157065, 1; *Clupeichthys aesarnensis*, MZUSP 62465, 3; *C. bleekeri*, MZUSP 63114, 1; *C. perakensis*, MZUSP 63104, 3; *Dorosoma cepedianum*, MZUSP 62481, 3; USNM 272899, 6; *Dussumieria acuta*, MZUSP 62468, 3; *Etrumeus teres*, MZUSP 62469, 3; USNM 188934, 3; *Gilchristella aestuaria*, MZUSP 64115, 3; *Harengula clupeola*, MZUSP 18672, 1; *H. jaguana*, MZUSP 11269, 2; MZUSP 10791, 1; *Herklotsichthys dispilonotus*, MZUSP 63115, 1; *Lile piquitinga*, MZUSP 11215, 1; *Ophistonema oglinum*, MZUSP 10844, 3; *Pellonula afzeliusi*, UMMZ 195029, 2; *Platanichthys platana*, MZUSP 10629, 3; MCP 19409, 3; *Rhinosardina amazonica*, MZUSP 11231, 2; MZUSP 11452, 1; *Sardina pilchardus*, MZUSP 37394, 3; MZUSP 12123, 1; *Sardinella albella*, MZUSP 63117, 2; *S. aurita*, USNM 272875, 2; *S. brasiliensis*, MZUSP 12124, 1; MZUSP 11418, 2; *S. maderensis*, MZUSP 37382, 1; *Tenualosa ilisha*, USNM 276407, 2; *Spratelloides delicatulus*, MZUSP 62470, 3.

Coiliidae: *Coilia neglecta*, USNM 357380, 6; *C. reben-tischii*, MZUSP 62483, 1; *Lycotricha crocodilus*,

- MZUSP 62482, 1; *Setipinna melanochir*, MZUSP 64119, 1; *Thryssa baelama*, ANSP 63228, 1; *T. hamiltoni*, USNM 364595, 1; *T. mystax*, ANSP 60578, 1.
- Engraulidae: *Amazonsprattus scintilla*, MZUSP 93481, 30; *Anchoa filifera*, MZUSP 18528, 2; *A. lamprotaenia*, MZUSP 11508, 2; *A. januaria*, MZUSP 10630, 2; *A. lyolepis*, MZUSP 11476, 3; *A. marinii*, MZUSP 11489, 1; *A. spinifer*, MZUSP 11454, 3; *A. tricolor*, MZUSP 11547, 2; *Anchovia clupeioides*, MZUSP 11559, 1; *A. surinamensis*, MZUSP 11570, 1; *Anchoviella brevirostris*, MZUSP 11578, 2; *A. carrikeri*, MZUSP 5728, 3; *A. guianensis*, MZUSP 5726, 1; MZUSP 58400, 2; *A. lepidentostole*, MZUSP 51826, 2; *A. vaillanti*, MZUSP 11587, 2; *Cetengraulis edentulus*, MZUSP 11634, 3; *Encrasicholina heteroloba*, MZUSP 63118, 2; *Engraulis anchoita*, MZUSP 18393, 10; *Jurengraulis juruensis*, MZUSP 27857, 1; *Pterengraulis atherinoides*, MZUSP 11723, 1; MZUSP 18040, 2; *Stolephorus indicus*, MZUSP 63112, 2; MZUSP 63113, 1; MZUSP 63106, 1.
- Pristigasteroidea: *Chirocentrodon bleekermanus*, MZUSP 11097, 3; *Ilisha africana*, MZUSP 62463, 4; *I. amazonica*, MZUSP 33266, 2; *I. elongata*, UMMZ 219537, 1; *I. kampeni*, MZUSP 62477, 2; *I. megaloptera*, MZUSP 62472, 2; *I. melastoma*, MZUSP 62473, 2; *Neoopisthopterus tropicus*, MZUSP 62478, 2; *Odontognathus mucronatus*, MZUSP 11264, 3; MZUSP 11267, 3; MZUSP 10835, 2; *Opisthopterus dovii*, MZUSP 62462, 3; *O. equitorialis*, MZUSP 62479, 2; *O. tardoore*, MZUSP 62475, 2; *Pellona castelnaeana*, INPA 4959, 1; MZUSP 5492, 1; *P. ditchela*, ANSP 63526, 1; USNM 189996, 1; *P. flavipinnis*, MZUSP 40063, 1; MZUSP 18728, 1; *P. harroweri*, MZUSP 11283, 3; MZUSP 11308, 1; MZUSP 11374, 3; *Pliosteostoma lutipinnis*, FMNH 2818, 2; *Pristigaster cayana*, MZUSP 30338, 3; *P. whiteheadi*, INPA 8555, 3; *Raconda russeliana*, MZUSP 62466, 4.
- Gonorynchiformes
- Chanidae: *Chanos chanos*, MZUSP 62601, 1; USNM 347536, 9.
- Gonorynchidae: *Gonorynchus* sp., MZUSP 63663, 1.
- Kneriidae: *Kneria auriculata*, MZUSP 63121, 4; USNM 290762, 2.
- Cypriniformes
- Catostomidae: *Catostomus commersoni*, USNM 238094, 2; USNM 238111, 2; USNM 340759, 2.
- Cyprinidae: *Opsariichthys uncirostris*, USNM 87445, 2; *Zacco platypus*, MZUSP 62597, 1.
- Gyrinocheilidae: *Gyrinocheilus aymonieri*, USNM 271692, 1.
- Characiformes
- Citharinidae: *Citharinus latus*, MZUSP 84480, 1.
- Distichodontidae: *Xenocharax spilurus*, MZUSP 50358, 1.
- Characidae: *Hollandichthys multifasciatus*, MZUSP uncatalogued, 5.
- Curimatidae: *Curimata roseni*, MZUSP 55740, 1.
- Argentiniformes
- Argentinidae: *Argentina striata*, USNM 188212, 2.
- Alepocephalidae: *Searsia koefoedi*, USNM 206873, 1; *Talismania aphos*, USNM 215540, 1.
- Salmoniformes
- Galaxiidae: *Galaxias auratus*, USNM 344893, 3; USNM 344895, 1.
- Osmeridae: *Osmerus mordax*, MZUSP 64116, 1.
- Salmonidae: *Salmo* sp., MZUSP uncatalogued.
- Esociformes
- Esocidae: *Esox americanus*, USNM 237257, 2; USNM 120051, 2.
- Umbridae: *Dalia pectoralis*, USNM 034033, 1; *Umbra pygmaea*, USNM 333152, 3; USNM 345523, 1.
- Elopomorpha
- Albulidae: *Albula vulpes*, MZUSP 10625, 2.
- Elopidae: *Elops* sp., MZUSP 60346, 1.
- Megalopidae: *Megalops cyprinoides* USNM 173580, 3.
- Osteoglossomorpha
- Hiodontidae: *Hiodon tergisus*, MZUSP 28450, 1; *H. alosoides*, USNM 350554, 1.
- Notopteridae: *Chitala* sp., MZUSP uncatalogued, 1.