AMERICAN MUSEUM
Novitates

PUBLISHED BY THE AMERICAN MUSEUM OF NATURAL HISTORY
CENTRAL PARK WEST AT 79TH STREET, NEW YORK, N.Y. 10024
Number 3195, 17 pp., 6 figures, 1 table May 16, 1997

Egg Surface Structure and Larval Cement Glands in Nandid and Badid Fishes with Remarks on Phylogeny and Biogeography*

RALF BRITZ[1]

ABSTRACT

Egg surface structure and larval morphology of the Nandidae and Badidae were studied with SEM, and cement organs of larval *Nandus* and *Badis* were histochemically stained using the PAS technique. The study is supplemented with data on reproductive behavior. The Asian Nandidae differ from the African–South American Nandidae in important features of reproductive behavior as well as egg and larval structure. No synapomorphy for the family Nandidae could be identified. The genera *Polycentropsis, Polycentrus,* and *Monocirrhus,* however, form a monophyletic group on the basis of the following synapomorphies: eggs with a unique surface pattern of narrow ridges running radially from the micropyle; larvae with a multicellular cement gland on top of the head; and adults with a unique spawning procedure. The genus *Afronandus* is tentatively assigned to this monophyletic group because it shares with the other three African–South American Nandidae the character of adhesive filaments at the vegetal egg pole. Comparison of egg and larval structure between the Nandidae and Badidae revealed no characters indicating a close relationship of the two families.

Lundberg's (1993) hypothesis, which explains the distribution of the African–South American Nandidae by dispersal through seawater, is rejected on the basis of the ecological preferences of these Nandidae. The age of origin of African–South American Nandidae is hypothesized to date back at least to the late Cretaceous, that is, before the separation of Africa and South America.

* This paper is dedicated to the memory of the influential zoologist Hans M. Peters who died December 13, 1996, at the age of 89.

[1] Postdoctoral Fellow, Department of Herpetology and Ichthyology, American Museum of Natural History. Present address: Lehrstuhl für Spezielle Zoologie, Universität Tübingen, Auf der Morgenstelle 28, 72076 Tübingen, Germany.

 ISSN 0003-0082 / Price $2.40

INTRODUCTION

Nandidae, or leaffishes, occur in fresh waters of Southeast Asia, West Africa, and South America (Berra, 1981; Nelson, 1994), a disjunct distribution that has been noted by a number of ichthyologists and zoogeographers (see Lundberg, 1993 for a review). In the past, seven genera, *Nandus, Afronandus, Polycentropsis, Polycentrus, Monocirrhus, Badis,* and *Pristolepis,* have been arranged in various combinations with different numbers of families (Günther, 1861; Boulenger, 1904; Jordan, 1923; Weber and De Beaufort, 1936; Berg, 1958; Greenwood et al., 1966; Liem, 1970; Lauder and Liem, 1983; Nelson, 1994). They are usually classified as members of the Percoidei (Johnson, 1984; Nelson, 1994), although sometimes a closer relationship to the Anabantoidei (labyrinth fishes) or the Channidae (snakeheads) has been postulated (Gosline, 1968, 1971; Nelson, 1969; Rosen and Patterson, 1990). Barlow et al. (1968) compared breeding behavior, egg and larval morphology, and osteology of *Badis* and *Polycentrus* and found striking differences between the two genera. As a consequence, they removed *Badis* from the Nandidae and erected a new monotypic family, Badidae. Subsequently, *Pristolepis* also was considered to be related only remotely to the Nandidae (Liem, 1970; Liem and Greenwood, 1981). Liem (1970) restricted the family Nandidae to five genera, *Nandus, Afronandus, Polycentropsis, Polycentrus,* and *Monocirrhus,* and provided an osteological definition of the group. Nevertheless, he admitted that "no single osteological feature distinguishes the family from other percoids" (op. cit. p. 82). The monophyly of the Nandidae is still questionable and its phylogenetic inter- and intrarelationships remain unresolved (Lundberg, 1993).

Aquarium breeding of three geographically separated Nandidae—the Southeast Asian *Nandus nandus,* West African *Polycentropsis abbreviata,* and South American *Monocirrhus polyacanthus*—as well as the Southeast Asian *Badis badis* provided the opportunity to investigate whether egg and larval characters reported for *Polycentrus* by Barlow et al. (1968) are actually representative of the Nandidae. These results have been supplemented by data obtained from ovarian eggs of the poorly known West African species *Afronandus sheljuhzkoi.* After evaluating the phylogenetic significance of these new data, the hypothesis recently proposed by Lundberg (1993) to explain the current distribution of the family Nandidae and its subgroups was examined.

ACKNOWLEDGMENTS

I am deeply indebted to R. Rucks (Kiel) and R. Donoso-Büchner (Dorsten) for providing eggs and larvae of *Polycentropsis, Monocirrhus,* and *Badis* and for sharing with me their profound knowledge of nandid behavior. I am grateful to the individuals at the Lehrstuhl für Spezielle Zoologie, Universität Tübingen, especially W. Maier for the opportunity to maintain specimens of *N. nandus* and *B. badis* at his institute, H. Schoppmann for his expert skills with SEM, and M. Hohloch and M. Meinert for a variety of technical advice and assistance. G. Teugels, Musée Royale de L'Afrique Centrale (MRAC; Tervuren), kindly lent a specimen of *Afronandus shelhjuhzkoi* for SEM of its eggs. At the American Museum of Natural History (New York), the help of P. Fong-Melville with SEM was greatly appreciated. The present study was supported by a grant of the Landesgraduiertenförderung Baden-Württemberg at the Lehrstuhl für Spezielle Zoologie and subsequently by a Kalbfleisch Postdoctoral Fellowship at the Department of Herpetology and Ichthyology of the American Museum of Natural History.

The manuscript was read and greatly improved by the comments of M. Toledo-Piza, G. Nelson, and J. Atz and by the reviews of S. O. Kullander and K. F. Liem.

MATERIALS AND METHODS

Freshly spawned, fertilized eggs and larvae of *N. nandus, P. abbreviata, M. polyacanthus,* and *B. badis* were prepared for SEM using the procedure described in Britz et al. (1995). They were observed and photographed with a Cambridge Stereoscan 250 Mk2.

Ovarian eggs of *A. sheljuhzkoi* were obtained from a preserved female specimen (MRAC 73-05-P-4669-4674). After removing maternal tissue by use of fine forceps, eggs were critical-point dried in a Balzers CPD 030, coated with 150 Å gold-palladium, and observed and photographed using a Zeiss DSM 950.

To confirm the position and shape of cement glands or individual cement cells, larvae of *N. nandus* (2.5 days postspawning) and *B. badis* (~3 days postspawning) were studied histochemically. The larvae were stained in toto for mucopolysaccharides in accordance with the PAS (= periodic acid Schiff reaction) technique of Peters and Berns (1982) and then photographed with a Zeiss Tessovar.

RESULTS

Egg Structure in the Nandidae

Nandus nandus: Two Southeast Asian species of the genus *Nandus,* for which information is available, spawn several thousand small, translucent eggs that adhere to plants and other substrates and show no parental care (for *N. nandus*: Parameshwaran et al., 1971; personal obs.; for *N. nebulosus*: Rucks, 1973, 1996). Respective data for the newly described third species in the genus, *Nandus oxyrhynchus* (Ng et al., 1996), are lacking. Spawned eggs of *N. nandus* measure 0.7–0.8 mm in diameter. SEM demonstrated that the animal pole of the egg adheres to the substrate (fig. 1A); to observe the micropyle, the egg had to be removed from the substrate (fig. 1B). A circular area around the micropyle bears a dense carpet of short filaments (fig. 1C, D) that appear to be primarily responsible for the attachment of the egg to the substrate. The micropyle is situated in the middle of this area and has a diameter of ~2 μm (fig. 1C, D). The remaining egg surface possesses fine, irregular wrinkles (fig. 1C) that are covered by a thin layer of unknown substance, which also may have adhesive properties inasmuch as smaller bits of plants or mud stick to it.

Afronandus sheljuhzkoi: The breeding behavior of this rare West African nandid is unknown, although some cursory remarks by Scheel (1964a) point to the existence of male parental care. Ovaries of one female contained about 70 ripe eggs with a diameter of 1.1–1.3 mm. The size of spawned, fertilized eggs may be slightly larger due to formation of the perivitelline space (Laale, 1980). The vegetal egg pole bears a tuft of filaments that originates from a circular area on the zona radiata (fig. 1E, F) and supposedly serves to attach the egg to the substrate. The micropyle is situated on the opposite pole (fig. 1G) and has a diameter of almost 4 μm. The zona radiata near the micropyle does not show any striking surface structure (fig. 1H) apart from the numerous canal openings typical of many teleost eggs (Stehr and Hawkes, 1983; Riehl, 1991).

Polycentropsis abbreviata: The male of this other West African nandid usually builds a nestlike structure of air bubbles under leaves that are floating at the water surface; eggs are attached to the underside of these leaves (Rucks, 1992). The male guards the clutch, which may consist of 300–350 eggs (Scheel, 1964b; Rucks, 1992). Eggs measure 1.3–1.4 mm in diameter. SEM reveals that their vegetal pole attaches to the leaf with the aid of a stalk of radially arranged fiber bundles (fig. 2A). Each bundle originates from the zona radiata of the egg (fig. 2B) and ends at a distance of ~0.5 cm (fig. 2A). Bundles are about 10 μm wide and consist of many smaller individual fibers that had a width of 0.2–0.3 μm each (fig. 2B). The zona radiata shows a distinct honeycomblike surface pattern where the fibers originate (fig. 2B). The animal pole bears a considerable number of radial ridges which run radially from the oval micropylar pit situated in a craterlike elevation (fig. 2C). The micropyle also has a slightly oval shape, 4 μm long and 2 μm wide (fig. 2D).

Monocirrhus polyacanthus: The South American nandid *M. polyacanthus* attaches eggs to the underside of leaves of aquatic plants. The male guards the eggs, of which there may be more than 200 (Richter, 1973). Eggs are slightly larger than those of *Polycentropsis,* measuring 1.7–1.8 μm in diameter. Their vegetal pole sits on a peduncle of fibers ~0.5 mm long that spread out distally and adhere to the substrate (fig. 2E). The peduncle originates from an area at the vegetal pole smaller than that area in *Polycentropsis,* but the zona radiata shows a similar honey-

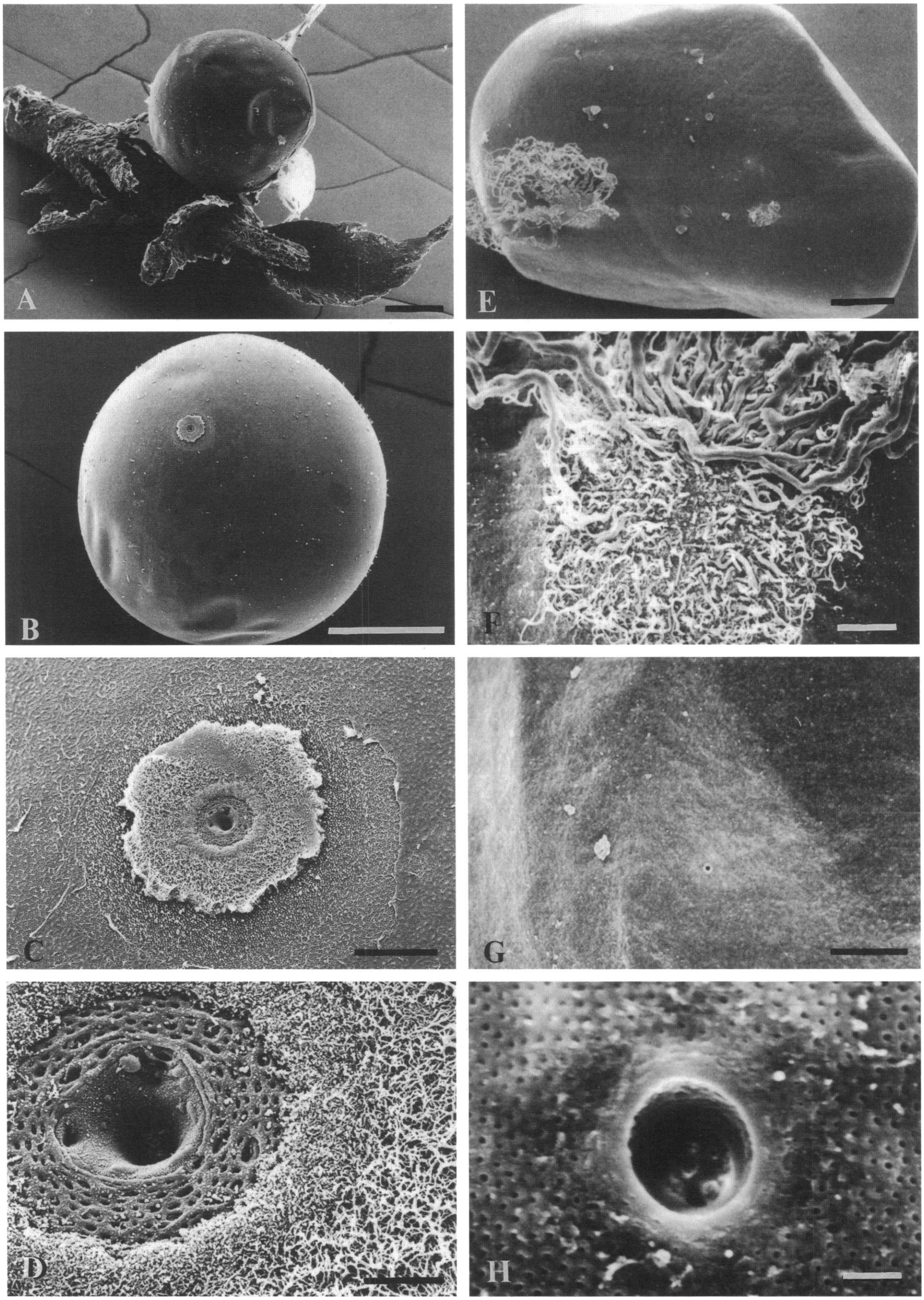

Fig. 1. A–D. *Nandus nandus*. SEM photograph of freshly spawned egg. **A,** Egg attached to *Riccia* moss, scale bar 200 μm; **B,** view of animal pole, scale bar 200 μm; **C,** micropylar region at higher magnification, scale bar 20 μm; **D,** close-up of the micropyle, scale bar 4 μm. E–H. *Afronandus sheljuhzkoi*. SEM photograph of ovarian egg. **E,** view of vegetal pole with stalk of fibers, scale bar 200 μm; **F,** region of zona radiata from which fibers originate, scale bar 20 μm; **G,** animal pole with micropyle, scale bar 50 μm; **H,** close-up of the micropyle, scale bar 2 μm.

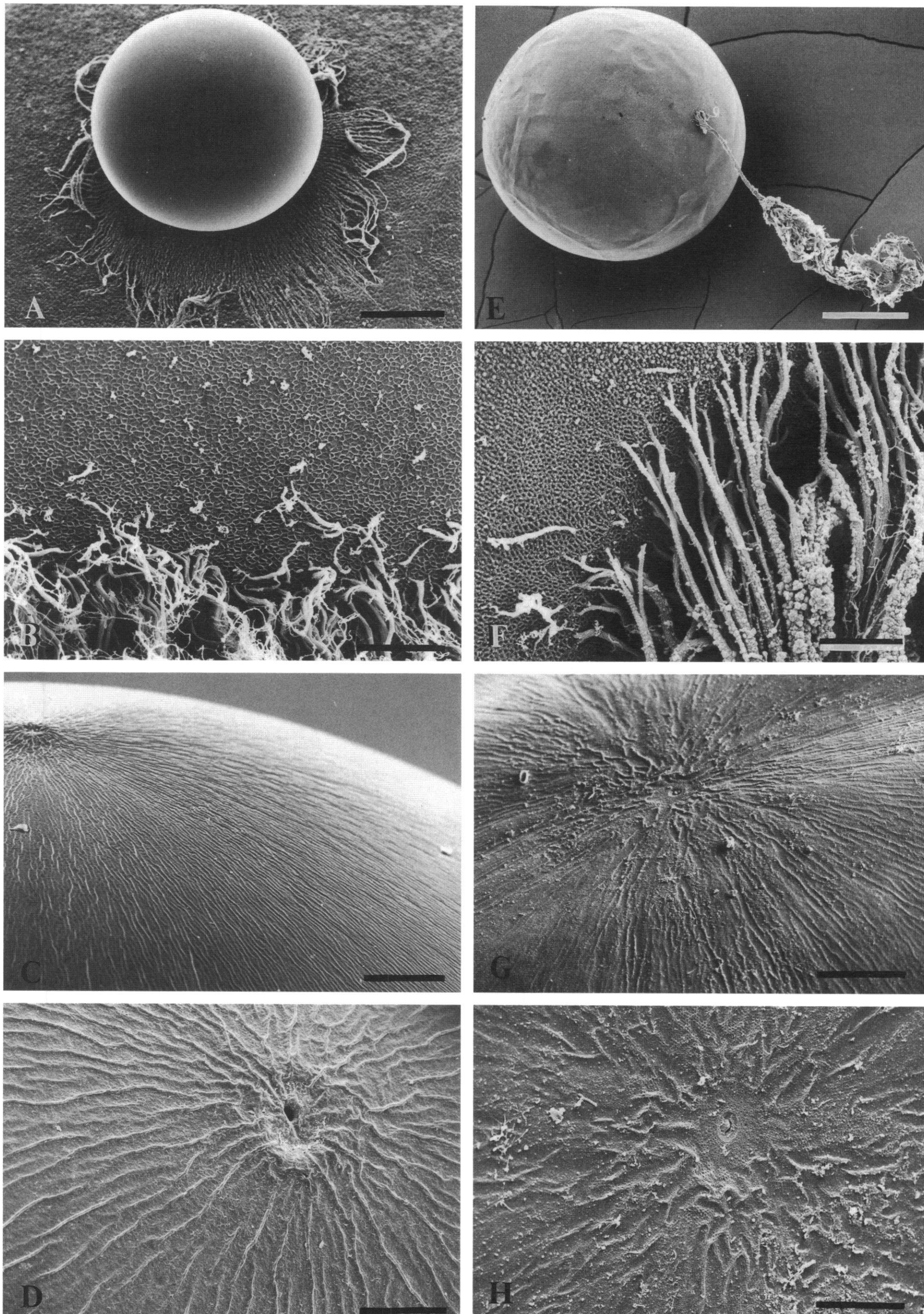

Fig. 2. A–D. *Polycentropsis abbreviata.* SEM photograph of freshly spawned egg. **A,** Egg attached to underside of *Nymphaea* leaf, scale bar 400 μm; **B,** region of zona radiata from which adhesive fibers originate, scale bar 10 μm; **C,** animal pole with micropyle, scale bar 40 μm; **D,** close-up of oval micropyle, scale bar 20 μm. E–H. *Monocirrhus polyacanthus.* SEM photograph of freshly spawned egg. **E,** view of vegetal pole of egg that has been removed from leaf, scale bar 400 μm; **F,** region of zona radiata from which adhesive fibers originate (compare 2B), scale bar 10 μm; **G,** animal pole with micropyle, scale bar 40 μm; **H,** close-up of oval micropyle, scale bar 20 μm.

combed ultrastructure (cf. fig. 2B, F). As in *Polycentropsis,* narrow ridges extend radially from the oval micropyle, which measures 3 μm in length and 2 μm in width (fig. 2G, H).

Egg Structure in the Badidae

Badis badis: The eggs of *B. badis* adhere to the substrate and are guarded and fanned by the male (Barlow et al., 1968; Richter, 1981; personal obs.). They have a diameter of ~0.8 mm. Observations made with SEM confirm the results of Barlow et al. (1968). A baglike sheath completely surrounds the egg (fig. 3A, B). The brim of this sheath adhered to the underlying substrate (fig. 3B, C). The sheath consists of a very dense network of extremely thin fibers, and its adhesiveness is affected by the individual fibers that anchored that area to the substrate (fig. 3C). Figure 3A shows a clutch of eggs of which the sheaths of two (marked with arrows) were torn to reveal the egg inside. The egg surface is irregularly wrinkled (fig. 3D). The circular micropyle, 2.5–3 μm wide, lies in a micropylar pit 15–20 μm in diameter (fig. 3D). Eggs of *Badis* that had been imported from another location exhibited a quite different surface structure surrounding the micropyle (fig. 3E). The wrinkles were confined to a small, distinct area close to the micropyle, and the remaining surface appeared to be smooth (fig. 3E).

Cement Glands of Larval Nandidae

Nandus nandus: Eggs of the Southeast Asian *N. nandus* hatch within 30–36 hours after spawning (Parameshwaran et al., 1971; personal obs.). Larvae can be seen attached to different kinds of substrate. This is also true for *N. nebulosus* (Rucks, 1996), but there is no information for *N. oxyrhynchus.* Figure 4A shows an SEM photograph of a larval *N. nandus* at 2.5 days after hatching. Cement cells can be recognized as small papillalike protuberances that bulge from the epidermis on the ventral side of the yolk sac (fig. 4A, B). A glutinous substance is released through small apertures between the epidermal cells (fig. 4B). PAS staining confirmed the SEM observation that the cement gland actually consists of scattered single cells confined to the ventral part of the yolk sac (fig. 4C, D).

Polycentropsis abbreviata: Eggs of this African nandid hatch after 120 hours at 27°C (Rucks, 1992, personal commun.). Larvae adhere to leaves with the aid of a cement gland on their head. With SEM, this gland can be recognized as a humplike structure on the dorsal area of the head of newly hatched larvae (fig. 4E, F). Between the epidermal cells, the gland has numerous openings (one marked with an arrow) through which the glutinous substance is released (fig. 4G). Unfortunately no PAS preparation could be carried out.

Monocirrhus polyacanthus: Eggs of the South American leaffish *M. polyacanthus* hatch after ~72 hours (Richter, 1973; Rucks, personal commun.) and, as in *Polycentropsis,* larvae use their cement glands to adhere to leaves, where they are guarded by the male (Richter, 1973). The cement gland is situated on top of the head (fig. 4H) and strongly resembles the gland of *Polycentropsis.*

No data on *Afronandus* were available.

Cement Glands of Larval Badidae

Badis badis: Eggs hatch after 2 days, and larvae adhere to the substrate, where they are guarded and fanned by the male (Barlow, 1964; Barlow et al., 1968; personal obs.). Adhesion is accomplished with the aid of a cement gland situated at the anteroventral tip of the larval yolk sac. This cement gland was described by Barlow et al. (1968) on the basis of light microscopy, but SEM provided further insight into its structure. Figure 5A depicts a ventral view of a larval *B. badis.* The adhesive gland can be recognized as an area on the anterior part of the yolk sac where the epidermis possesses a different sculpturing (fig. 5A, B). A roll-like elevation marks the anterior border of the gland. Here, many openings between the epidermal cells can be recognized where the glutinous substance is released (fig. 5B). Posterior to the roll, the epidermis is smoother, but a considerable number of openings are nevertheless present.

The differential staining of PAS preparation reveals that the gland consists of a considerable number of individual cells (fig.

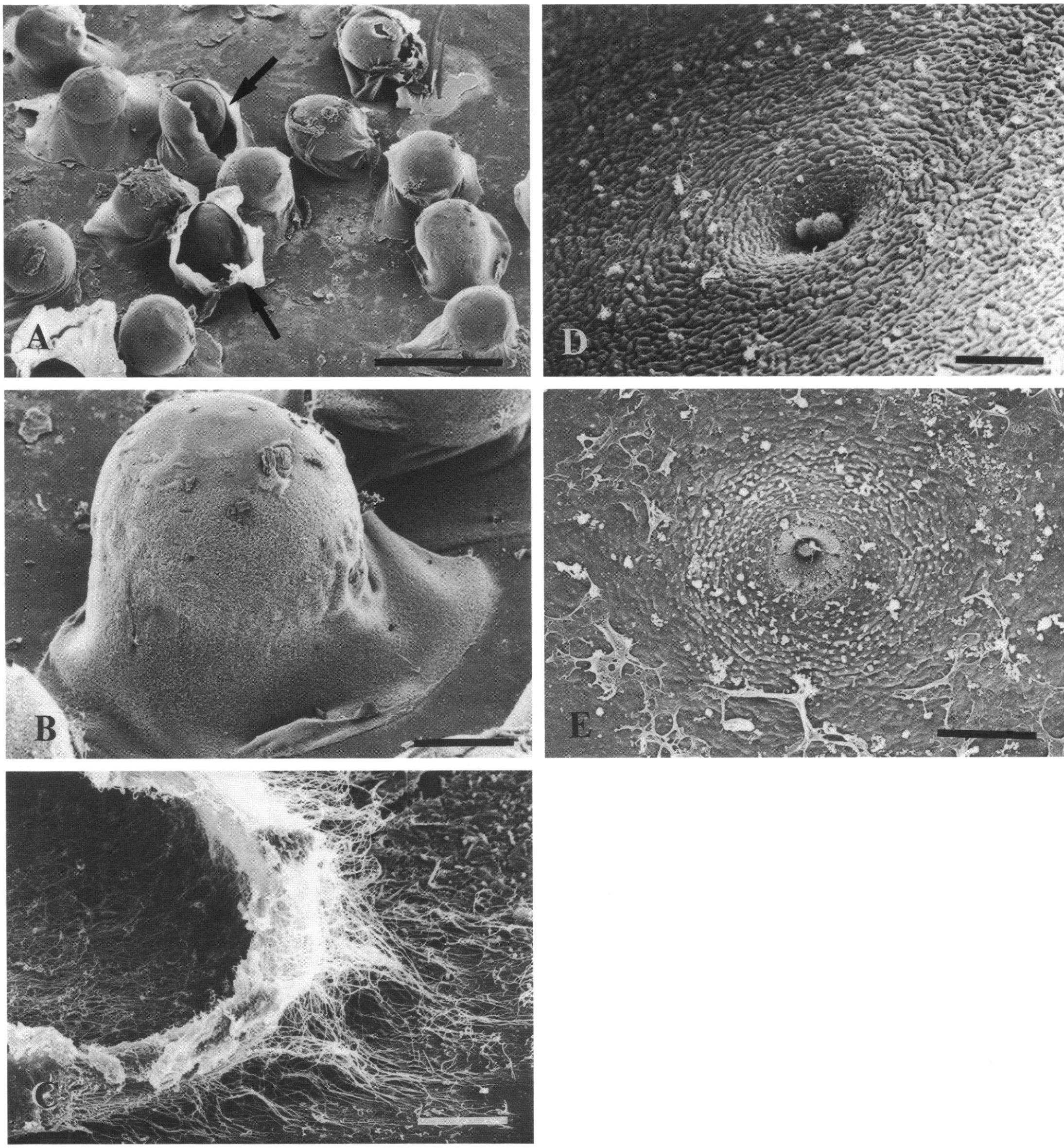

Fig. 3. *Badis badis.* SEM photograph of freshly spawned eggs. **A,** Egg clutch with adhesive sheathes of two eggs (marked by arrows) opened to reveal egg inside, scale bar 1 mm; **B,** egg with covering sheath, scale bar 200 μm; **C,** periphery of sheath with individual fibers anchored to substrate, scale bar 10 μm; **D,** micropylar region with well-developed wrinkles, scale bar 10 μm; **E,** micropylar region of egg of *Badis* from a different locality, wrinkles confined to area around micropyle, scale bar 10 μm.

5C, D). Cell density is highest at the anterior roll-like elevation of the gland and decreases toward its posterior end (fig. 5C). At the roll-like elevation the cement cells are arranged in multiple layers and possibly have collective openings through the epidermis, whereas in the posterior part they appear to be single layered with each cell having one opening. The delineation of the three-dimensional arrangement of the adhesive cells, however, requires further histological investigation.

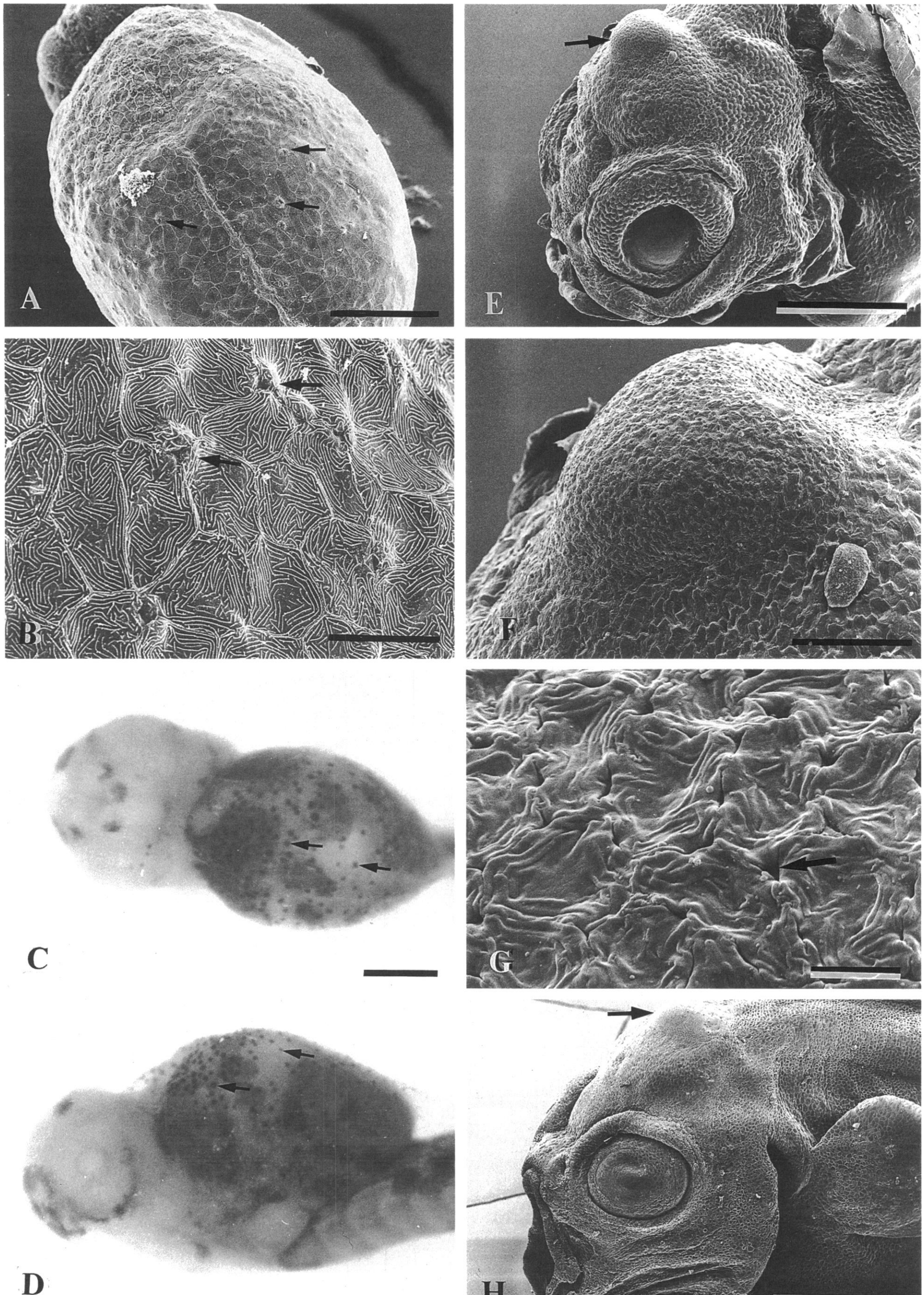

Fig. 4. Cement gland of larval Nandidae. A–D. *Nandus nandus* 2.5 days postspawning. **A,** Ventral view of larva, arrows mark bulges of individual cement cells, scale bar 100 μm; **B,** closeup of epidermis of yolk sac, cement cells indicated as bulges of the epidermis (marked by arrows) with an aperture at the tip of each protuberance, scale bar 20 μm; **C,** ventral and **D,** lateral view of PAS-stained larva, individual cement cells marked by arrows, scale bars 100 μm. E–G. *Polycentropsis abbreviata* 5 days postspawning. **E,** head of larva with humplike cement gland (marked by arrow) above eyes, scale bar 200 μm; **F, G,** cement gland at higher magnification, showing irregular openings (marked by arrows)

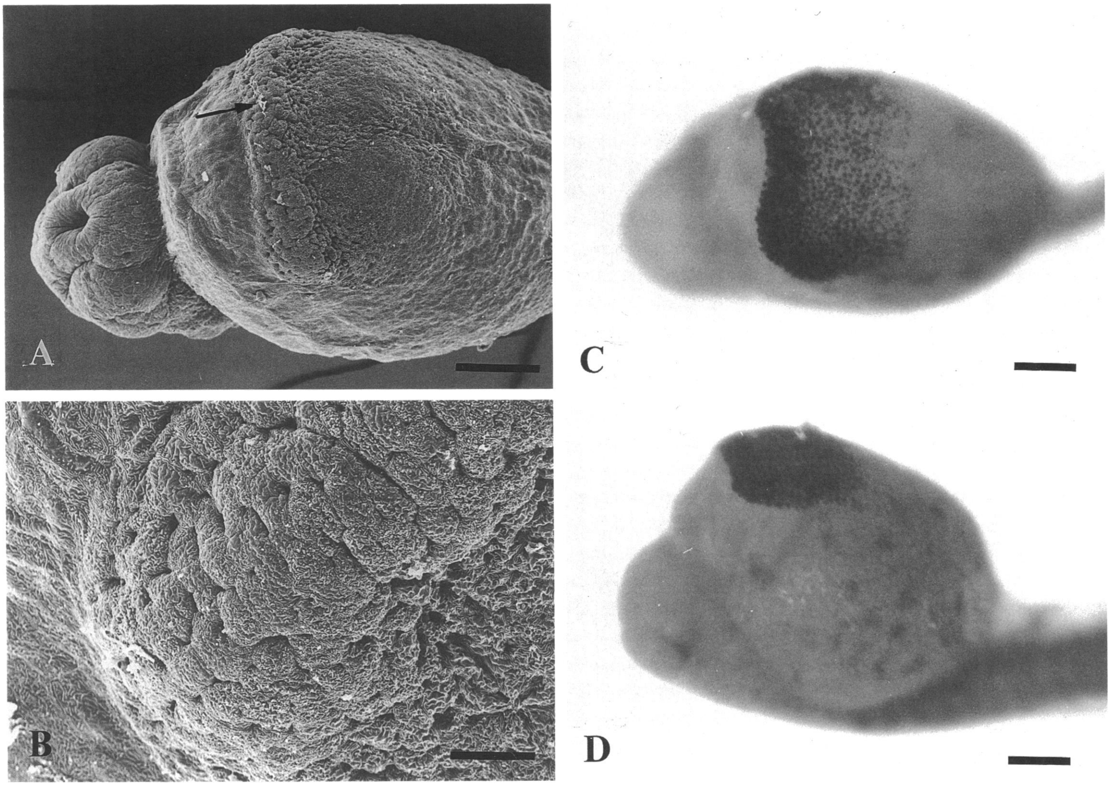

Fig. 5. Cement gland of larval *Badis badis* ~3 days postspawning. **A,** ventral view of larval yolk sac, arrow points to adhesive thread released from openings between epidermal cells, scale bar 100 μm; **B,** anterior part of cement gland at higher magnification, with numerous openings through which adhesive substance is released, scale bar 20 μm; **C,** ventral and **D,** lateral view of PAS-stained larva, scale bars 100 μm.

DISCUSSION

With the exclusion of the genus *Badis* by Barlow et al. (1968) and the genus *Pristolepis* by Liem (1970) and Liem and Greenwood (1981), the family Nandidae sensu Liem (1970) comprises only the Southeast Asian genus *Nandus,* the two West African genera *Afronandus* and *Polycentropsis,* and the two South American genera *Polycentrus* and *Monocirrhus.* Even in this restricted state, however, no synapomorphies were proposed to support the monophyly of the Nandidae and no hypotheses about the interrelationships of its five genera were available (Lundberg, 1993). Liem, in a personal communication that was cited by Cracraft (1974), claimed that the group *Polycentrus* + *Monocirrhus* is the sister-group of *Polycentropsis,* but he provided no supporting evidence for this hypothesis.

The present SEM study of egg and larval morphology, which includes four nandid species, demonstrates that the egg and larval characters reported for *Polycentrus* (Barlow et al., 1968) are not uniformly expressed within the Nandidae (see table 1). *Nandus*

←

between epidermal cells, scale bar in **F** 40 μm, in **G** 4 μm. **H,** *Monocirrhus polyacanthus* 9 days postspawning. Head region of larva with humplike cement gland (marked by arrow) above eyes, scale bar 400 μm.

TABLE 1

Comparison of Reproductive Behavior and Egg and Larval Structure of *Badis* and the Nandidae

Character	*Badis Badis*	*Nandus nandus*	*Afronandus sheljuzhkoi*	*Polycentropsis abbreviata*	*Polycentrus schomburgkii*	*Monocirrhus polyacanthus*
Parental care	Male[a,b,c,d]	None[a,e]	? Male ?[f]	Male[g,h]	Male[d,i,j,k]	Male[l]
Spawning embrace	Present	Present	?	Absent	Absent	Absent
Spawning site	Caves preferred[a,b,c,d]	Scattered among vegetation[a,e]	? Caves ?[f]	Underside of leaves at water surface[g,h]	Caves and underside of leaves[d,i,j,k]	Underside of leaves[l]
Number of eggs	~80[b]	Several thousand[a,e]	70 eggs in ovaries[a]	Several hundred (300–350)[g,h]	Several hundred (300–500)[i,j,k]	Several hundred (up to 200)[l]
Size of eggs	~1 mm[d]	0.7–0.8 mm[a,e]	1.1–1.3 mm[a]	1.3–1.4 mm[a]	~1 mm[d]	1.7–1.8 mm[a]
Ridges from micropyle	Absent[a]	Absent[a]	Absent[a]	Present[a]	Present[d]	Present[a]
Adhesive filaments	Forming sheath around egg[a,d]	Short, at animal pole[a]	Long, at vegetal pole[a]	Long, at vegetal pole[a]	Long, at vegetal pole[d]	Long, at vegetal pole[a]
Time until hatch	~24 h[a,d]	30–36 h[a,e]	?	120 h[h]	~72 h[i,j]	~72 h[l]
Cement gland	Multicellular, on yolk sac[a,d]	Scattered cells, on yolk sac[a]	?	Multicellular, on head[a]	Multicellular, on head[d]	Multicellular, on head[a]

[a] Present study.
[b] Nieuwenhuizen (1959).
[c] Barlow (1962).
[d] Barlow et al. (1968).
[e] Parameshwaran et al. (1971).
[f] Scheel (1964a).
[g] Scheel (1964b).
[h] Rucks (1992).
[i] Rucks (1960).
[j] Barlow (1967).
[k] Zukal (1971).
[l] Richter (1973).

differs from the African and South American Nandidae in several of these characters, an observation that correlates with some striking differences between the two groups in the osteological features studied by Liem (1970).

Phylogenetic Relationships Among Nandid Genera

Position of Adhesive Filaments: There is a striking similarity between African and South American Nandidae in the location of adhesive filaments at the vegetal egg pole (figs. 1E, 2A, E); in contrast, adhesive threads are developed at the animal pole in *Nandus* (fig. 1B, C).

In an attempt to polarize this character, searching among putatively related taxa proved to be of little help. Although badid eggs also adhere to the substrate, their adhesive filaments do not attach to the egg's surface at all. Anabantoidei primitively possess nonadhesive eggs that either float on the water surface, are guarded in nests of foam, or are orally incubated (Forselius, 1957; Breder and Rosen, 1966; Vierke, 1975, 1978, 1991a; Britz, 1995; Britz et al., 1995). Eggs of channids similarly float at the water surface and are not adhesive (Willey, 1910; Armbrust, 1963, 1967; Yapchiongco, 1963; Vierke, 1991b).

SEM data from other percomorph families are scarce, and wide-ranging comparisons are therefore impossible at the present. However, adhesive filaments on the vegetal egg pole have been reported for pseudoplesiopine pseudochromoids (Mooi, 1990) and for some cichlids (Wickler 1956a, 1956b; Stiassny and Mezey, 1993). Neither group is closely related to the Nandidae.

Based on this evidence, the presence of adhesive filaments at the vegetal pole of the egg in *Afronandus, Polycentropsis, Polycentrus,* and *Monocirrhus* is interpreted here as a synapomorphy of the African–South American Nandidae.

Radial Ridges Running from the Micropyle: The characteristic pattern of ridges on the micropylar region of the eggs of *Polycentropsis* (fig. 2C, D), *Polycentrus* (see Barlow et al., 1968: 442 and pl. Id), and *Monocirrhus* (fig. 2G, H) is not present in *Nandus* (fig. 1B, C, D) or *Afronandus* (fig. 1G, H) and has not been reported for any other percomorph. This unique character is considered a synapomorphy of these three genera, thus lending support to Liem's belief that *Polycentropsis* is more closely related to *Polycentrus* and *Monocirrhus* than to other Nandidae. It is unlikely that the lack of ridges in the eggs of *Afronandus* is an artifact due to the study of unfertilized ovarian eggs because several investigations have demonstrated that specific surface structures are already present in the ripe ovarian eggs of bony fishes (Stehr and Hawkes, 1983; Mooi, 1990; Mooi et al., 1990; Britz et al., 1995).

Cement Glands: In *Polycentropsis* (fig. 4E, F), *Polycentrus* (Barlow et al., 1968), and *Monocirrhus* (fig. 4H) the cement gland is a multicellular organ on top of the head. In *Nandus,* it is represented by scattered individual cement cells that are confined to the ventral and lateral parts of the yolk sac (fig. 4A–D).

Unfortunately, only limited data from other percomorphs are available for comparison. In a remarkable pioneering study of the morphology and evolution of cement glands in cichlids, Peters and Berns (1982) investigated these organs with both SEM and histochemical PAS staining techniques. In all the cichlids that have been examined, this structure consists of three pairs of glands, one situated between the eyes and the other two on top of the head (Jones, 1937; Ilg, 1952; Bennemann and Pietzsch-Rohrschneider, 1978; Peters and Berns, 1982; Hamlett, 1990). In addition to their larger number, cichlid cement glands differ in structure from those described for the three Afro-American Nandidae. Cichlid cement glands have only one wide, central, apical opening through which the adhesive substance is released, whereas the cement gland of *Polycentropsis* and *Monocirrhus* (and *Polycentrus* ?) has numerous openings. Although data about larval cement glands in other percomorphs are rare, some information can be gathered from Ilg (1952). She found multicellular glands with a central opening not only in cichlids but also in the Ambassidae, in which only one pair of glands develops.

Scattered individual cement cells have been described in the Anabantoidei *Pseudosphromenus cupanus* (Jones, 1940; Padmanabhan, 1955), *Betta splendens, Trichogaster trichopterus,* and *Macropodus opercularis* (Ilg, 1952), and *Ctenopoma damasi* and *C. muriei* (Mörike, 1977). These cells are scattered in the epidermis of the anterior part of the body and do not form multicellular glands as in cichlids or ambassids. A supplementary study by the author has confirmed the presence of such scattered individual cement cells for the Anabantoidei *Anabas testudineus, Ctenopoma weeksii, Belontia signata, Colisa labiosa, Trichogaster trichopterus, Macropodus concolor, Pseudosphromenus dayi, Parosphromenus paludicola, Trichopsis vittatus,* and *Betta imbellis,* in which the cells are scattered in group-specific patterns over the yolk sac, head, and anterior trunk region. These scattered cement cells in Anabantoidei resemble the condition in *Nandus,* but because of the lack of comparative data in other percomorphs, little can be said about their phylogenetic significance. Their occurrence in centrarchid larvae (Ilg, 1952), however, may indicate that this is a widespread character.

The only other percomorph group with larval cement glands that has been studied to a significant extent is the Badidae (Barlow et al., 1968; present study). Though also multicellular, the badid cement gland is situated at the tip of the yolk sac (fig. 5A–D). Thus, the type of cement gland of larval *Polycentropsis, Polycentrus,* and *Monocirrhus* appears to be unique among percomorphs and can be considered another synapomorphy of this assemblage. Unfortunately, nothing is known about cement glands in *Afronandus.*

It can be summarized that the investigation of breeding behavior, egg structure, and larval morphology has not yielded a single character common to all Nandidae that could be interpreted as a synapomorphy of this family. However, the genera *Polycentropsis, Polycentrus,* and *Monocirrhus* showed great similarities in three character complexes that were interpreted as synapomorphies supporting their monophyly. *Afronandus* is tentatively assigned to this group until more information about its breeding behavior and larval structure becomes available.

Breeding Behavior: Parental care exists in a variety of percomorphs; usually the male guards the eggs and often the offspring (Blumer, 1982). Among percomorph groups possibly related to Nandidae, parental care occurs in the majority of Anabantoidei (Forselius, 1957; Breder and Rosen, 1966; Vierke, 1975, 1978, 1991a; Cambray, 1990, in press; Britz, 1995), in the channids (Willey, 1910; Armbrust, 1963, 1967; Yapchiongco, 1963; Ng and Lim, 1990; Vierke, 1991b), and in the Badidae (Barlow et al., 1968). However, these taxa's reproductive styles show numerous differences and are hardly comparable to those of the genera *Polycentropsis, Polycentrus,* and *Monocirrhus.* The Anabantoidei (Forselius, 1957; Vierke, 1975; Cambray, 1990, in press; Britz, 1995), the Channidae (Yapchiongco, 1963; Vierke, 1991b), and the Badidae (Barlow et al., 1968) release eggs and sperm during a well-developed spawning embrace (fig. 6A, B, C), a behavior that seems to be present also in *Nandus* (Parameshwaran et al., 1971; Rucks, 1973, 1996). This spawning embrace is entirely lacking in *Polycentropsis, Polycentrus,* and *Monocirrhus.* In these three Nandidae, the female assumes an upside-down position and attaches eggs to the substrate while the male hovers close by in a normal position (fig. 6D, E). To fertilize the eggs, the male remains in a normal position or bends sideways, releasing his sperm and washing them to the eggs with the help of fin movements (for *Polycentrus*: Rucks, 1960; Barlow, 1967; Zukal, 1971; for *Monocirrhus*: Richter, 1973; for *Polycentropsis*: Rucks, 1992). This special spawning procedure is tentatively interpreted as another synapomorphy of the genera *Polycentropsis, Polycentrus,* and *Monocirrhus.*

Phylogenetic Relationships Between the Nandidae and Badidae

Barlow et al. (1968) justified the exclusion of *Badis* from Nandidae mainly on the basis of differences between *Badis* and *Polycentrus.* It was shown above that reproductive ethology and egg and larval morphology vary among the five nandid genera. However, *Badis* differs significantly from the leaffishes in all these characters.

Eggs of *Badis* are completely surrounded by a sheath of fibers without actually being attached to them (fig. 3A, B), a situation not

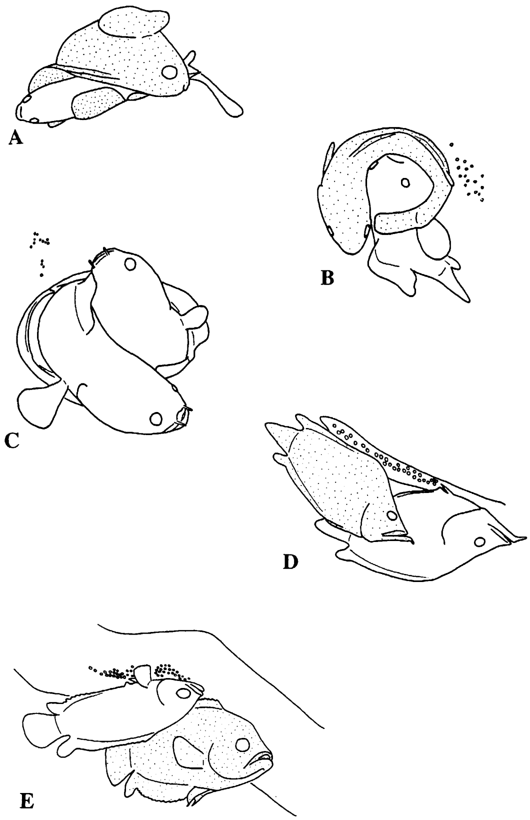

Fig. 6. Spawning posture in **A,** *Badis badis,* from Britz (1995) after photograph in Richter (1981); **B,** *Anabas testudineus,* from Britz (1995); **C,** *Channa bleheri,* from Britz (1995) after photograph in Vierke (1991b), sexes not given; **D,** *Polycentrus schomburgkii,* after photograph in Zukal (1971); **E,** *Monocirrhus polyacanthus,* after photograph in Richter (1973). Male stippled, female white.

present in any nandid. The micropylar region has neither ridges as in *Polycentropsis, Polycentrus,* and *Monocirrhus* nor the circular area of carpetlike fibers described for *N. nandus.*

The cement gland of *Badis* is multicellular but situated at the tip of the yolk sac. *Badis* shows an unusual spawning embrace (fig. 6A; Barlow, 1962; Barlow et al., 1968) that is not present in *Polycentropsis, Polycentrus,* and *Monocirrhus* (fig. 6D, E) but shared with Anabantoidei (fig. 6B). This was one of the reasons why Badidae and Anabantoidei were hypothesized to be sister groups (Barlow et al., 1968; Lauder and Liem, 1983). However, a similar spawning embrace occurs also in channids (fig. 6C; Yapchiongco, 1963; Ng and Lim, 1990; Vierke, 1991b) and seems to be present in *Nandus* (Parameshwaran et al., 1971; Rucks, 1973, 1996). Accordingly, Britz (1995) concluded that such a spawning embrace should be considered a plesiomorphic character at the level of Badidae and Anabantoidei and does not indicate a sister-group relationship between both taxa. Unfortunately, the data in this study do not further clarify the phylogenetic relationships of Badidae. No synapomorphies with Nandidae could be identified. Future study of the reproductive behavior and egg and larval structure of *Pristolepis* should yield new data and lead to a better understanding of the phylogenetic relationships of *Badis.* Moreover, the difference in egg surface structures of the *Badis* collected in two different localities in the present study might prove to be evidence for the existence of additional species of *Badis* and demonstrates the need for a thorough revision of this genus.

Biogeographic Implications

Recently Lundberg (1993) reviewed African–South American freshwater fish clades and discussed different models to explain their recent geographic distribution. As a prerequisite for building biogeographic hypotheses, the phylogeny of the taxa involved has to be satisfactorily resolved (Nelson and Platnick, 1981). Lundberg (1993) pointed out that there are no well-supported hypotheses of relationships among the different nandid genera, apart from Liem's belief (cited in Cracraft, 1974) that *Polycentropsis* is the sister group of *Polycentrus* + *Monocirrhus,* which has found supportive evidence in this study. As shown above, a monophyletic group of the three genera *Polycentropsis* + *Polycentrus* + *Monocirrhus* can be hypothesized on the basis of several synapomorphies, and therefore at least one African–South American relationship has been established.

Lundberg (1993) stressed that an origin of Nandidae in the early Cretaceous before separation of India from Africa or in the later Cretaceous before separation of Africa and

South America would "imply unacceptably ancient ages of origin for this family and its basic subgroups" (p. 187). For this reason, Lundberg (1993: 187–188) considered "postdrift dispersals of nandids through marine habitats . . . more likely." The data presented here, however, are incompatible with this hypothesis.

Observations by Scheel (1964b) and Rucks (1992) clearly show that *Polycentropsis* is a species that occurs in very soft, acidic waters. Moreover, *Polycentropsis, Monocirrhus,* and *Polycentrus* breed in captivity only when they are maintained under such conditions (Scheel, 1964b; Richter, 1973; Rucks, 1992). The most parsimonious conclusion is that the most recent common ancestor of this monophyletic group already exhibited similar ecological preferences. Thus, the ecological environment in which the recent African–South American Nandidae occur renders unlikely any dispersal through marine habitats after the separation of the African and South American landmasses.

The genera *Polycentropsis, Polycentrus,* and *Monocirrhus* show similarities in breeding behavior, egg structure, and larval morphology that are assumed to have already been present in the last common ancestor of these three genera. It seems unlikely that an ancestor with these features would have been able to survive and reproduce under saltwater conditions—an additional argument against the postdrift dispersal hypothesis of Lundberg (1993).

For the reasons given, it is hypothesized here that the distribution of the African–South American Nandidae is best explained by a simple drift vicariance model and their presence on the African–South American landmass before the final separation of the two continents. This explanation suggests a much older age of origin for the African–South American Nandidae than that assumed by Lundberg (1993). According to the recent overview of the fossil record of teleosts (Patterson, 1993a, 1993b), the earliest Percomorpha or Perciformes incertae sedis are found in the late Cretaceous. At the moment, the monophyletic taxon with the lowest rank to which the Nandidae can be assigned without doubt is the Percomorpha, although Nandidae have typically been classified as Percoidei since Regan (1913). Johnson (1984), however, pointed out that Regan's Percoidei is not supported by synapomorphies. The occurrence of African–South American Nandidae in the late Cretaceous thus appears not so unlikely as claimed by Lundberg (1993).

In addition to osteological differences cited by Liem (1970), this study has presented differences in reproductive biology and egg and larval structure between African–South American and Asian Nandidae. Therefore, inasmuch as the monophyly of Nandidae sensu Liem (1970) has not been satisfactorily demonstrated, it seems premature to speculate about models that try to explain nandid distribution on three continents.

REFERENCES

Armbrust, W.
1963. Zucht des Schlangenkopfes *Ophicephalus obscurus.* Aquarien. Terr. Z. 16: 298–301.
1967. Nachzucht beim Schlangenkopf (*Ophicephalus africanus*). Ibid. 20: 367–368.

Barlow, G. A.
1962. Ethology of the Asian teleost *Badis badis.* IV. Sexual behavior. Copeia 1962: 346–360.
1964. Ethology of the Asian teleost *Badis badis.* V. Dynamics of fanning and other parental activities, with comments on the behavior of the larvae and postlarvae. Z. Tierpsychol. 21: 99–123.

Barlow, G. W.
1967. Social behavior of a South American leaf fish, *Polycentrus schomburgkii,* with an account of recurring pseudofemale behavior. Am. Midl. Nat. 78: 215–234.

Barlow, G. W., K. F. Liem, and W. Wickler
1968. Badidae, a new fish family—behavioral, osteological, and developmental evidence. J. Zool. (London) 156: 415–447.

Bennemann, R., and I. Pietzsch-Rohrschneider
1978. The morphology of the cement gland apparatus of larval *Pterophyllum scalare* Cuv. & Val. (Cichlidae, Teleostei). Cell Tissue Res. 193: 491–501.

Berg, L. S.
1958. System der rezenten und fossilen Fis-

chartigen und Fische. Berlin: VEB Verlag.
Berra, T. M.
1981. An atlas of distribution of the freshwater families of the world. Lincoln: Univ. Nebraska Press.
Blumer, L. S.
1982. A bibliography and categorisation of bony fishes exhibiting parental care. Zool. J. Linn. Soc. 76: 1–22.
Boulenger, G. A.
1904. Teleostei. *In* S. F. Harmer and A. E. Shipley (eds.), Fishes, Ascidians, etc., pp. 541–727. London: Macmillan.
Breder, C. M., and D. E. Rosen
1966. Modes of reproduction in fishes. New York: The Natural History Press.
Britz, R.
1995. Zur phylogenetischen Systematik der Anabantoidei (Teleostei, Percomorpha) unter besonderer Berücksichtigung der Stellung des Genus *Luciocephalus.* Morphologische und ethologische Untersuchungen. Ph.D. diss., Tübingen Univ., 125 pp.
Britz, R., M. Kokoscha, and R. Riehl
1995. The anabantoid genera *Ctenops, Luciocephalus, Parasphaerichthys,* and *Sphaerichthys* (Teleostei: Perciformes) as a monophyletic group: evidence from egg surface structure and reproductive behavior. Japan. J. Ichthyol. 42: 71–79.
Cambray, J. A.
1990. Early ontogeny and notes on breeding behavior habitat preference and conservation of the Cape kurper, *Sandelia capensis.* Ann. Cape Prov. Mus. Nat. Hist. 18: 159–182.
In press. The spawning behavior and conservation of the endangered eastern Cape Rocky (*Sandelia bainsii*; Anabantidae), South Africa. Environ. Biol. Fishes.
Cracraft, J.
1974. Continental drift and vertebrate distribution. Annu. Rev. Ecol. Syst. 5: 215–261.
Forselius, S.
1957. Studies of anabantid fishes I–III. Zool. Bijdr. Uppsala. 32: 93–597.
Gosline, W. A.
1968. The suborders of perciform fishes. Proc. U.S. Natl. Mus. 124: 1–78.
1971. Functional morphology and classification of teleostean fishes. Honolulu: Univ. Press of Hawaii.
Greenwood, P. H., D. E. Rosen, S. H. Weitzman, and G. S. Myers
1966. Phyletic studies of teleostean fishes with a provisional classification of living forms. Bull. Am. Mus. Nat. Hist. 131: 339–456.
Günther, A.
1861. Catalogue of the acanthopterygian fishes in the British Museum. London: Taylor and Francis.
Hamlett, W. C.
1990. Subcellular structure and function of cement secreting glands in *Pterophyllum scalare,* a transient larval specialization. J. Submicrosc. Cytol. Pathol. 22: 27–37.
Ilg, L.
1952. Über larvale Haftorgane bei Teleosteern. Zool. Jahrb. Abt. Anat. 72: 577–600.
Jones, S.
1937. On the origin and development of the cement glands in *Etroplus maculatus* (Bloch). Proc. Indian Acad. Sci., B 6: 79–90.
1940. Notes on the breeding habits and early development of *Macropodus cupanus* (Cuv. & Val.), with special reference to cement glands of the early larvae. Rec. Indian Mus. 42: 269–276.
Johnson, G. D.
1984. Percoidei: development and relationships. *In* H. G. Moser et al. (eds.), Ontogeny and systematics of fishes, pp. 464–498. Lawrence, KS: Allen Press.
Jordan, D. S.
1923. A classification of fishes, including families and genera as far as known. Stanford Univ. Publ. Biol. Sci. 3: 77–243.
Laale, H. W.
1980. The perivitelline space and egg envelopes of bony fishes: a review. Copeia 1980: 210–226.
Lauder, G. V., and K. F. Liem
1983. The evolution and interrelationships of the actinopterygian fishes. Bull. Mus. Comp. Zool. 150: 95–197.
Liem, K. F.
1970. Comparative functional anatomy of the Nandidae. Fieldiana Zool. 56: 1–166.
Liem, K. F., and P. H. Greenwood
1981. A functional approach to the phylogeny of the pharyngognath teleosts. Am. Zool. 21: 83–101.
Lundberg, J. G.
1993. African–South American freshwater fish clades and continental drift: prob-

lems with a paradigm. *In* P. Goldblatt (ed.), Biological relations between Africa and South America: 156–199. New Haven, CT: Yale Univ. Press.

Mörike, D.
1977. Vergleichende Untersuchungen zur Ethologie zweier Labyrinthfischarten, *Ctenopoma muriei* (Boulenger 1906) und *Ctenopoma damasi* (Poll 1939) (Anabantoidei, Pisces). Ph.D. diss., Tübingen Univ. 178 pp.

Mooi, R. D.
1990. Egg surface morphology of pseudochromoids (Perciformes: Percoidei), with comments on its phylogenetic implications. Copeia 1990: 455–475.

Mooi, R. D., R. Winterbottom, and M. Burridge
1990. Egg surface morphology, and evolution in the Congrogadinae (Pisces: Perciformes: Pseudochromidae). Can. J. Zool. 68: 923–934.

Nelson, G. J.
1969. Gill arches and the phylogeny of fishes, with notes on the classification of vertebrates. Bull. Am. Mus. Nat. Hist. 141: 475–552.

Nelson, G. J., and N. Platnick
1981. Systematics and biogeography: cladistics and vicariance. New York: Columbia Univ. Press.

Nelson, J. S.
1994. Fishes of the world. (3rd ed.) New York: Wiley.

Nieuwenhuizen, A. van den
1959. *Badis badis* III. Aquarien Terr. Z. 12: 98–101.

Ng, P. K. L., and K. K. P. Lim
1990. Snakeheads (Pisces: Channidae): natural history, biology and economic importance. *In* L. M. Chou and P. K. L. Ng. (eds.) Essays in zoology, pp. 127–152. Singapore: Dept. Zool., Natl. Univ.

Ng, H. H., C. Vidthayanon, and P. K. L. Ng
1996. *Nandus oxyrhynchus,* a new species of leaf fish (Teleostei: Nandidae) from the Mekong basin. Raffles Bull. Zool. 44: 11–19.

Padmanabhan, K. G.
1955. Breeding habits and early embryology of *Macropodus cupanus* (Cuv. & Val.) Bull. C.R.I. Trivandrum. 4: 1–45.

Parameshwaran, S., S. Radhakrishnan, and C. Selvaraj
1971. Some observations on biology and life-history of *Nandus nandus* (Hamilton). Proc. Indian Acad. Sci. 73B: 132–147.

Patterson, C.
1993a. An overview of the early fossil record of acanthomorphs. Bull. Mar. Sci. 52: 29–59.
1993b. Osteichthyes: Teleostei. *In* M. J. Benton (ed.), The fossil record 2, pp. 621–656. London: Chapman and Hall.

Peters, H. M., and S. Berns
1982. Die Maulbrutpflege der Cichliden. Untersuchungen eines Verhaltensmusters. Z. zool. Syst. Evolutionsforsch. 20: 18–52.

Regan, C. T.
1913. The classification of the percoid fishes. Ann. Mag. Nat. Hist. 12: 111–145.

Richter, H.-J.
1973. DDR-Erstnachzucht von Blattfischen. Aquarien Terr. 20: 220–225.
1981. Das "Fisch-Chamäleon": Der Blaubarsch. Aquarien Mag. 446–450.

Riehl, R.
1991. Die Struktur der Oocyten und Eihüllen oviparer Knochenfische—eine Übersicht. Acta biol. Benrodis. 3: 27–65.

Rosen, D. E., and C. Patterson
1990. On Müller's and Cuvier's concepts of pharyngognath and labyrinth fishes and the classification of percomorph fishes, with an atlas of percomorph dorsal gill arches. Am. Mus. Novitates 2983: 57 pp.

Rucks, R.
1960. *Polycentrus schomburgki(i).* Aquarien Terr. Z. 13: 105–107.
1973. Zum ersten Mal (?!) gezüchtet: *Nandus nandus*—Der Nander. Aquarien Terr. Z. 26: 158–160.
1992. *Polycentropsis abbreviata.* Beobachtungen und Erkenntnisse. Aquarien Terr. Z. 45: 420–423.
1996. *Nandus nebulosus*—ein Nanderbarsch wird vorgestellt. Aquarien Terr. Z. 49: 554–556.

Scheel, J. J.
1964a. *Afronandus sheljuzhkoi* Meinken, a rare African nandid. The Aquarium (Norristown) 33(7): 6–9.
1964b. *Polycentropsis abbreviata* Boulenger 1901. The Aquarium (Norristown) 33(8): 8–10.

Stehr, C. M., and J. W. Hawkes
1983. The development of the hexagonally structured egg envelope of the C-O sole (*Pleurichthys coenosus*). J. Morphol. 178: 267–284.

Stiassny, M. L. J., and J. G. Mezey
1993. Egg attachment systems in the family Cichlidae (Perciformes: Labroidei),

with some comments on their significance for phylogenetic studies. Am. Mus. Novitates 3058: 11 pp.

Vierke, J.
1975. Beiträge zur Ethologie und Phylogenie der Familie Belontiidae. Z. Tierpsychol. 38: 163–199.
1978. Labyrinthfische und verwandte Arten. Wuppertal-Elberfeld: Engelbert Pfriem Verlag.
1991a. Brutpflegestrategien bei Belontiiden (Pisces, Anabantoidei). Bonn. zool. Beitr. 42: 299–324.
1991b. Der Regenbogen-Channa. Haltung und Zucht von *Channa bleheri*. Das Aquarium 25(266): 15–19.

Weber, M., and L. F. DeBeaufort
1936. The fishes of the Indo-Australian Archipelago. Vol. 7. Leiden: E. J. Brill.

Wickler, W.
1956a. Der Haftapparat einiger Cichlideneier. Z. Zellforsch. 45: 304–327.
1956b. Unterschiede zwischen den Cichliden-Gattungen, speziell *Geophagus* und *Biotodoma,* im Haftapparat der Eier. Naturwiss. 14: 333–334.

Willey, A.
1910. Observations on the nests, eggs and larvae of *Ophiocephalus striatus*. Spolia Zeylan. 6: 108–123.

Yapchiongco, J. V.
1963. Spawning habits of the murrell or snakehead fish, *Ophicephalus striatus* Bloch. Copeia 1963: 700–702.

Zukal, R.
1971. Bei Nacht schleicht sich der Räuber aus der Höhle . . . Der Vielstachler *Polycentrus schomburgki(i)*. Aquarien Mag. 5: 32–35.

Recent issues of the *Novitates* may be purchased from the Museum. Lists of back issues of the *Novitates* and *Bulletin* published during the last five years are available at World Wide Web site http://nimidi.amnh.org. Or address mail orders to: American Museum of Natural History Library, Department D, Central Park West at 79th St., New York, N.Y. 10024. TEL: (212) 769-5545. FAX: (212) 769-5009. E-MAIL: scipubs@amnh.org

♾ **This paper meets the requirements of ANSI/NISO Z39.48-1992 (Permanence of Paper).**