

The Barbel-Like Specialization of the Pelvic Fins in *Ophidion rochei* (Ophidiidae)

Elisabet Codina,¹ Kéver Loïc,¹ Philippe Compère,² Branko Dragičević,³ Jakov Dulčić³ and Eric Parmentier^{1*}

¹Laboratoire de Morphologie Fonctionnelle et Evolutive, Institut de Chimie, Bât. B6c, Université de Liège, 4000 Liège, Belgium

²Laboratoire de Morphologie Ultrastructurale, Institut de Chimie, Bât. B6c, Université de Liège, B-4000 Liège, Belgium

³Institute of Oceanography and Fisheries, POB 500, 21000 Split, Croatia

ABSTRACT Pelvic fins in *Ophidion rochei* are reduced to four rod-like structures situated at the ventral jaws. While the fish is swimming, they make continuous sweeping movements on the bottom. This paper examines and describes the anatomy of the pelvic fins to determine the possible functions of these appendages in relation to the mode of life of this fish species. The pelvic fins of *O. rochei* show strong similarities with barbels because they have identical sensory cell types, (taste buds, solitary chemosensory cells, and goblet cells), innervations and sensory function. Having nocturnal habits, specialization of pelvic fins in *O. rochei* corresponds to a supporting role to the life in dark environment. J. Morphol. 000:000–000, 2012. © 2012 Wiley Periodicals, Inc.

KEY WORDS: cusk-eel; taste bud; Ophidiiforme; sensory cell; nerve; barbel

INTRODUCTION

Fox (1999) described barbels as specific elongate structures that arise from the head or branchial region. Barbels are covered with skin and in some cases are supported by a cartilaginous or bony skeleton, that are innervated by branches of the cranial nerves, and that usually have sensory components. Barbels have been gained or lost repeatedly in many distant taxa (Fox, 1999). Although barbels differ in terms of origin, location, and structure in many fish taxa, they are generally involved in taste and mechanoreception. Some barbels develop as modifications of preexisting skeletal structures. They are derived from branchiostegal rays in Mullidae (LoBianco, 1907; Gosline, 1984) and Polimixiidae (McAllister, 1968; Kim et al., 2001). The maxillary barbel arises with the maxilla in Siluriformes (Schaefer and Lauder, 1986; Geerinckx et al., 2008) and some Cypriniformes, the structure being a common ancestral feature in these groups (Fox, 1999). Barbels can also evolve as new structures such as the mental barbel of Artedidraconidae (Eastman and Eakin, 2001) or the snout, supraorbital and lower jaw barbels of some Cottidae (Sato, 1977).

Depending on the species, the barbel epithelium may contain taste buds (TBs), solitary chemosensory cells (SCCs), free nerve endings, goblet cells, club cells and/or Merkel cells (Fox, 1999). The teleost TBs are found throughout the body epithelium, especially in the oral and oropharyngeal cavity, the head, or in external organs such as the lips, barbels or fins (Hansen et al., 2002). Barbels containing TBs are reported in Mullidae (Aguirre and Lombarte, 2000), Cyprinidae (Ohkubo et al., 2005), Siluridae (Ovalle and Shinn, 1977; Fujimoto and Yamamoto, 1980), Gadidae (Harvey and Batty, 2002) and Cottidae (Sato, 1977). Chemosensory cells are found in the fish epidermis showing a single microvillus apex, or even brush-like microvilli (Kotrschal, 1991, 1996; Hansen et al., 2002; LeClair and Topczewski, 2010), in contact with the aquatic environment. Goblet cells are exocrine unicellular glands whose secretion forms a slimy protective coat (Genten, 2009). Club cells are another kind of secretory cells in the mid layer (Iger et al., 1994) or surface layer of the epidermis (Chivers et al., 2007).

The ophidiid *Ophidion rochei* Müller, 1845 lives in the Mediterranean, Adriatic and Black seas, primarily on sandy muddy bottoms, from the intertidal zone to 90 m depth (Matallanas, 1979). Like many *Ophidion* species, this carnivore (Matallanas, 1980) hides in the sand during the day and is active at night (Bacescu et al., 1957), a char-

Contract grant sponsor: University of Liège (starting grants).

*Correspondence to: Eric Parmentier, Laboratoire de Morphologie Fonctionnelle et Evolutive, Institut de Chimie, Bât. B6c, Université de Liège, 4000 Liège, Belgium.
E-mail: E.Parmentier@ulg.ac.be

Received 16 April 2012; Revised 4 June 2012;
Accepted 17 June 2012

Published online in
Wiley Online Library (wileyonlinelibrary.com)
DOI: 10.1002/jmor.20066

acteristic that could be associated with its sound-producing abilities for finding mates (Mann et al., 1997, Parmentier et al., 2006, 2010). A particular morphological characteristic of the eel-like Ophidiinae is the pelvic fins, which are located far forward and supported by an anterior extension of the pectoral girdle (Nielsen et al., 1999). Observing captive specimens of *Ophidion rochei*, these fins are obviously not used during locomotion e.g., propulsion or direction. However, they continuously conduct sweeping movements, enabling the fish to follow the bottom relief and to estimate the distance between their body and the sand (Fig. 1). Gill (1863) and Grassé (1957) proposed that modified ventral fins of the *Ophidion* were similar to the barbel structure. However, there are no morphological studies on this structure to confirm or reject this idea.

The aim of this paper was to conduct a morphological study on the pelvic fins in *Ophidion rochei* to determine the possible functions of these appendages in relation to their way of life. Results are based on data from gross anatomy (skeleton and muscles), light and electron microscopy.

MATERIALS AND METHODS

Material Collected

Ten specimens (Total Length, 217–227 mm) of *Ophidion rochei* were collected in September 2008 and May 2010 in the Cetina Estuary (temperature 12°C) near the town of Omis in Croatia (43°26' N, 16°41' E). The specimens were caught between 22:30 and 01:00 with a beach seine (22 m long, mesh size of 4 mm at the outer wing and 2 mm at the central part). All the fish were placed in 50 L carboy provided with a sandy bottom and air pump and then transported (duration: 15 h 00) to our laboratory in Liège (Belgium). In Liège, they were kept in a 250 L tank with seawater at 18°C and a 15 cm deep sandy bottom.

For morphological and histological studies, fish were euthanized with MS 222 (500-mg·L⁻¹). Six of them were fixed in 7% formaldehyde for 10 days and then transferred to 70% ethyl alcohol. Pelvic fins from the four other specimens were cut and fixed for 48 h in 2.5% glutaraldehyde for observation by transmission electron microscopy (TEM), scanning electron microscopy (SEM) and light microscopy.

Morphological Studies

Skeleton and muscles. Two formaldehyde-fixed fish were cleared and stained with alizarin red and alcian blue using the method of Taylor and Van Dyke (1985) to reveal the skeletal structures. They were then dissected, examined, and drawn using a Wild M10 binocular microscope equipped with a camera lucida. Muscle functions were inferred from their insertions and/or from manipulations with forceps on two fresh specimens.

Pelvic anatomy terminology follows Stiassny and Moore (1992) and Rosen and Patterson (1969) for osteology; Winterbottom (1974) for muscles and, Freihofer (1970) and Howes (1992) for nerves. New names were however assigned in the present study because we did not find any equivalence in the literature.

Histological Studies

Internal structure. Appendages from two glutaraldehyde-fixed fishes were postfixed in 1% osmium tetroxide, dehydrated through a graded ethanol-propylene oxide series and embedded in epoxy resin (SPI-PON 812, SPI-CHEM, Leuven, Belgium).

Journal of Morphology

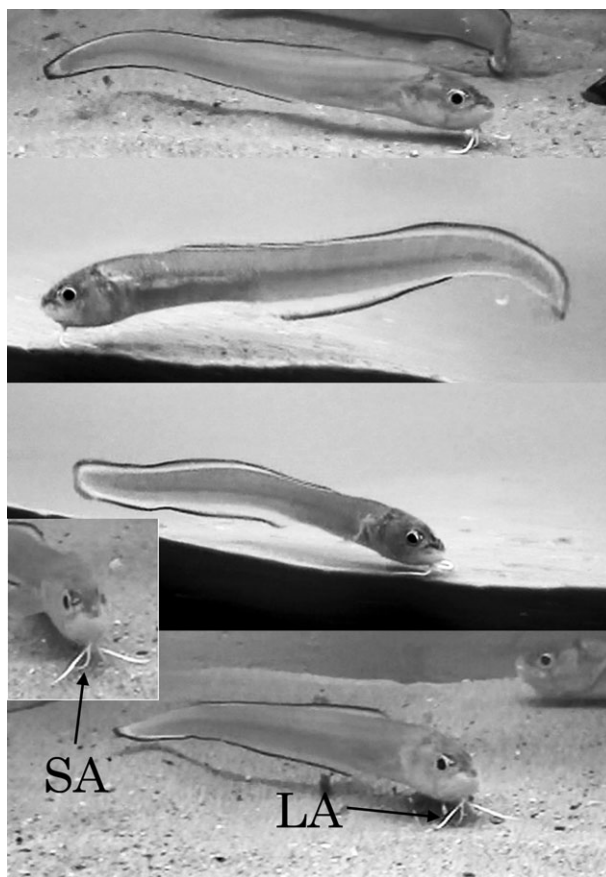


Fig. 1. Use of the pelvic fins to explore the bottom relief and to maintain a secured distance between the body and the ground in *Ophidion rochei*. The pelvic fins consist of two appendages that are each divided into two parts; the shorter (SA) ones are median and the longer (LA) ones are lateral. LA (= r1 + r3) and SA (= r2 + r4).

Semithin sections (1 µm) and ultrathin sections (60–80 nm) were cut using a diamond knife on a Reichert Ultracut E ultramicrotome. Toluidine blue-stained semithin sections were used for fin general histology and for orientation to target the area of further ultrathin sections. They were observed and photographed with a Leica MD 1000 binocular microscope equipped with a digital camera (Canon Power Shot S50, Diegem, Belgium). Ultrathin sections were classically stained with uranyl acetate and lead citrate, then viewed in a JEOL JEM 100SX transmission electron microscope (Zaventem, Belgium) at 80 kV accelerating voltage.

Surface morphology. The examination of the external morphology and distribution of TBs was carried out on eight appendages. In each case, the length and diameter were measured before fixation in 2.5% glutaraldehyde. These samples were then postfixed in 1% osmium tetroxide, dehydrated in an ethanol series, critical-point dried and platinum sputter-coated (20 nm) in a Balzers SCD-030 sputter-unit (Merksem, Belgium). Photographs were taken by a JEOL JSM 840A scanning electron microscope (Zaventem, Belgium) at a 20-kV accelerating voltage.

RESULTS

Morphology of the Pelvic Girdle

The pelvic girdle is situated ventrally to the head, in front of the cleithral symphysis and ventrally to the urohyal of the branchial basket (Fig. 2).

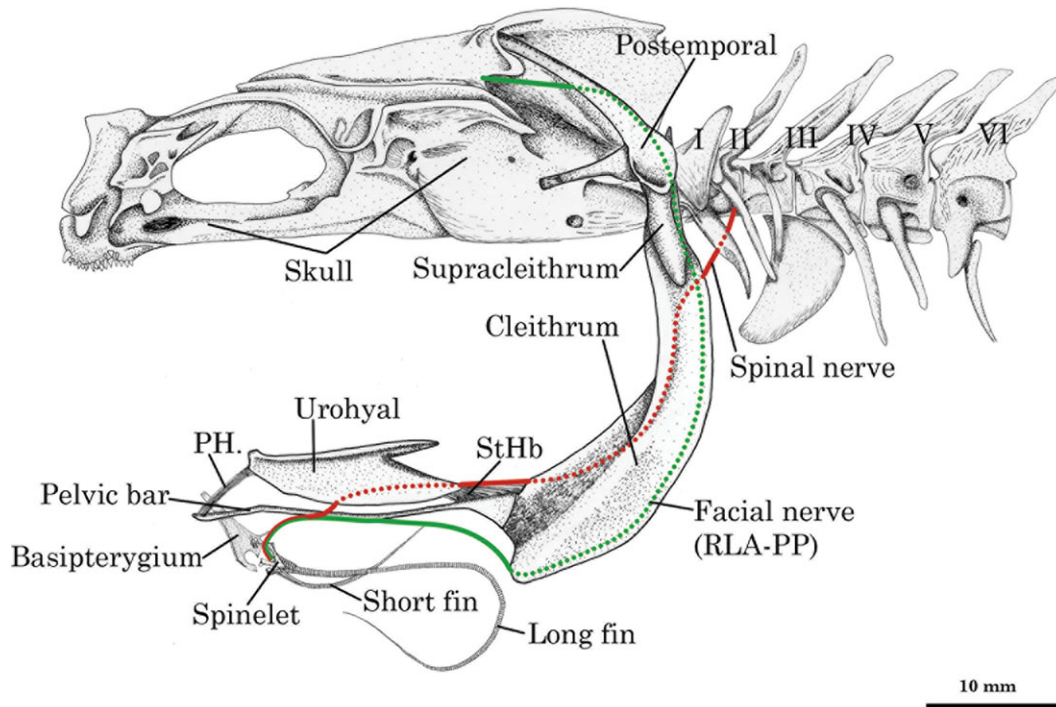


Fig. 2. Lateral view of the anterior part of the skeleton showing the skull, the first vertebrae, the urohyal, and the pectoral and pelvic girdles. The green line corresponds to the pelvic branch of the *ramus lateralis accessorius* nerve (RLA-PP) and the red line to the spinal nerve innervating the pelvic fins. A.P.H., *anterior pelvicoiyoideus*; StHb, *sternohyoideus* bundle.

Skeleton. The pelvic girdle is composed of two pelvic bars, two basipterygia and eight rays (Fig. 3).

- The pelvic bars are long horizontal shafts situated anterior to the ventral tip of the pectoral girdle. Both shafts are joined anteriorly by muscles and connective tissue, attached to the cleithra posteriorly and connected to the urohyal. Frontally, both bars enclose the tips of the basipterygia. As this is a different kind of bone from the cleithra, the term “pelvic bar” appears more appropriate than cleithral extension or bony filament (Howes, 1992; Nielsen, 1999).
- The basipterygium is a thin and long triangular bone. The structure base shows two well separated processes. Ventrally, the narrow median process (Mp) faces rays #3 and #4. Caudally, the articulation (Art) ends in a wide rounded cartilaginous area on which rays #1 and #2 are articulated. Laterally, the lateral process (Lp) is a small protuberance, situated at the level of the Art basis. Dorsally to the Art, the posterior process (Pp) is triangular, laminar and presented small foramina. Ventrally to the main body of the basipterygium, the anterior process (Ap) is a slender protuberance on the larger part of the basipterygium.
- Each ray or lepidotrichium consists of a series of numerous bony articles, making it quite flexible. Four rays (r1, r2, r3, r4) are found on each side (Fig. 3). They are grouped in pairs (r1 + r3 and

r2 + r4) in appendages of different sizes. Each of the long appendages (LA) is made up of r1 + r3 and is situated laterally while the short appendages (SA) are composed of r2 + r4 and situated medially (Fig. 3). The LAs and the SAs measured about 21.5 mm and 12.5 mm in length respectively in three fishes from 21.7 to 22.7 mm of total length. In these three fishes, the ovoid section of each appendage decreases regularly towards the distal tip, starting with about $140 \times 100 \mu\text{m}$ in the LA and $75 \times 55 \mu\text{m}$ in the SA.

- The pelvic spinelet is laterally situated at the proximal tip of the rays to which it is attached by connective tissue. The V-shaped spinelet is asymmetrical. Both branches are similar in length but the outer branch end is wider and the inner one is curved perpendicularly.

Musculature. Eight muscles are associated with the pelvic girdle in *Ophidion rochei* (Fig. 3). Working individually or in groups, they allow the appendages to move in both horizontal or vertical planes, and they raise the whole pelvic girdle. We provide here some possible functions for these muscles on the basis of their insertions and (when possible) manual tractions.

Musculus abductor superficialis pelvici (AB.S.P.). This muscle originates along the median process of the basipterygium and inserted dorso-laterally along the outer branch of the spinelet. Its

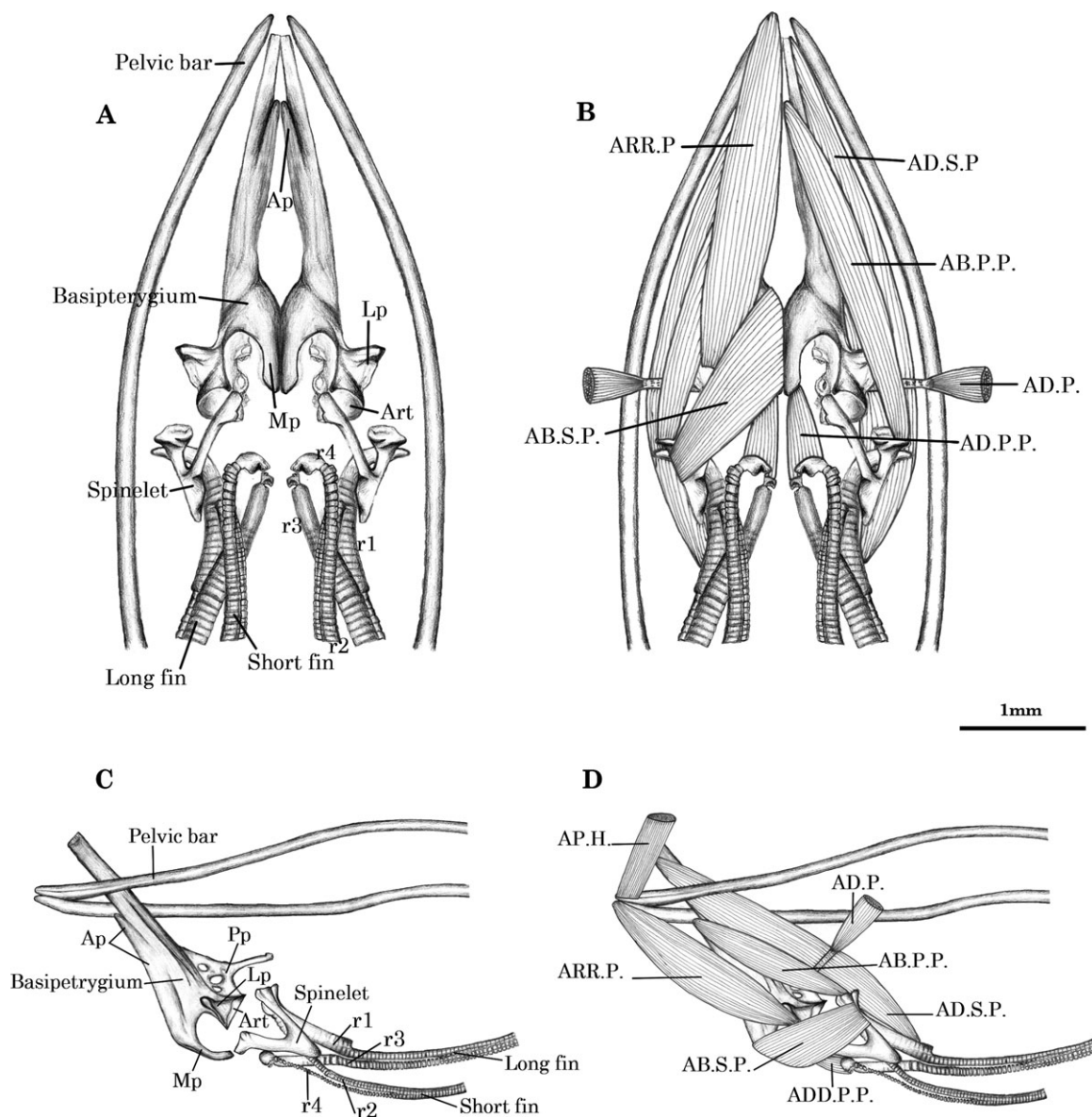


Fig. 3. Ventral (A) and left lateral (C) view of the pelvic girdle skeleton and of associated musculature (B and D) in *Ophidion rochei*. AB.P.P., abductor profundus pelvis; AB.S.P., abductor superficialis pelvis; AD.S.P., adductor superficialis pelvis; AD.P., adductor pelvis; AD.P.P., adductor profundus pelvis; Ap, anterior process; A.P.H., anterior pelvicoiyoideus; ARR.P., arrector pelvis; Art, articulation surface; Lp, lateral process; Mp, median process; Pp, posterior process; r, ray.

contraction could cause simultaneously the abduction and extension of the rays. Traction on the muscles causes lateral displacement of the rays from the median axis and their lowering.

Musculus arrector pelvis (ARR.P.). This muscle originates on the anterior end of the pelvic bar and inserts ventrally on the lateral process of the basipterygium. Traction on this muscle provokes the protraction and the elevation of the pelvic base.

Musculus abductor profundus pelvis (AB.P.P.). This muscle originates ventrally on the anterior process of basipterygium and inserts dorso-laterally on the base of the outer branch of the spinelet.

Its function could be simultaneously the abduction and elevation of the rays.

Musculus adductor superficialis pelvis (AD.S.P.). This muscle is dorsal to the AB.P.P., originating ventro-laterally on the anterior end of basipterygium and inserting dorso-laterally on the proximal region of ray #1. Its traction provokes the outward rotation of the rays, causing their flexion.

Musculus adductor pelvis (AD.P.). This is a very long straight muscle originating in the hypaxial musculature and inserting tendinously on the posterior process of the basipterygium, between the AD.S.P. and the AB.P.P. Its contraction could

pull the basipterygium caudo-dorsally, causing the elevation of the pelvic girdle.

Musculus adductor profundus pelvici (AD.P.P.). This muscle is attached dorsally on the median process of the basipterygium and inserts on the base of inner rays (#3 and #4). Its contraction seems to provoke the adduction of the appendages.

Musculus pelvicoihyoideus (PH.). This is a narrow muscle which originates dorsally on the anterior tip of the pelvic bar and inserted on the anterior part of the urohyal. Traction on this muscle brings the anterior tip of the pelvic girdle up, swinging the posterior portion down.

Musculus sternohyoideus (STHb). STHb muscle has several bundles with a paired origin. A bundle is attached on the posterior part of the urohyal and on the posterior end of the pelvic bar and the beginning of cleithrum. The *sternohyoideus* muscles have no clear function in relation to the pelvic girdle.

Nerves. The pelvic branch of the *ramus lateralis accessorius* (called *ramus recurrens facialis*) nerve (RLA-PEL) and a spinal nerve innervate the appendages (Fig. 2). The RLA-PEL is a thick nerve which left the skull via a posterolateral foramen in the parietal, at the level of the epiotic. It passes dorsally across the posterior margin of the postemporal, supracleithrum and cleithrum.

A thin spinal nerve leaves the vertebral column via a foramen in the second vertebra, following the cleithrum dorsally and the urohyal ventrally to join with the RLA-PEL between the pelvic bars. Thereafter, the common spinal and recurrent facial trunk divide each into four branches (Fig. 4) and enter into each fin through the space between the basipterygium base and the spinelet.

Histology of the Pelvic Fins

Histological cross-sections of pelvic fins provide an overview of the whole structure (Fig. 4). The description below follows an inner-outer direction. 1) The centre is occupied by two bilaterally-paired skeletal elements, the bony disks that surround diffuse connective tissue. 2) Around the axial rod, there are four main myelinated nerve bundles and blood vessels. 3) The well vascularized dermis consists of two differently compacted collagen layers. The inner *stratum spongiosum* is thicker than the outer *stratum superficiale*. Some isolated secretory tubules are observed in the *stratum spongiosum*. 4) The epidermis around the pelvic fins consists of stratified squamous epithelium of unequal thickness (Fig. 4): about 50 μm in the rostral part of the fin and about 10 μm in the caudal part. In the rostral part, the following layers of the epidermis are present from the base to the surface (Fig. 4B): a) the columnar cells of the *stratum basale*, b) the *stratum spinosum* with polygonal cells and c) the *stratum superficiale* with goblet cells. Epidermal cells appear to have numerous desmosomes and

important intercellular spaces as plasma membranes are evaginated. These evaginations seemed to form the folds of the epidermis surface. In the outer part of the epidermis, goblet cells appear to have lost their plasma and nuclear membranes. They then appear bigger than other epidermal cells before being finally shed (Fig. 4B).

Different kinds of specialized cells are found in the epidermis:

TBs. Ventrally, the thicker part of the epithelium possessed numerous bulb-shaped TBs, which are uniformly distributed along the surface epithelium (Fig. 5A). However, the density is different along-side the pelvic fins. Here, it increases from the base (mean \pm SD, $186 \pm 37 \text{ mm}^{-2}$, $n = 13$), towards the middle (ca. $283 \pm 24 \text{ mm}^{-2}$, $n = 11$) and is highest in the distal tip region (350 ± 68 , $n = 5$). On the surface, the TB microvilli form areas (TB pores) of 3.5 ± 0.9 , $n = 35$ in diameter (Fig. 5).

This intraepithelial sensory organ is composed of receptor, marginal and basal cells (Fig. 5). The receptor cells are vertically elongated, with the nucleus at the base and receptor microvilli at the apex. There are two kinds of receptor cells; some possessed a single thick microvillus whereas others have numerous thinner microvilli. The marginal cells are lengthened epidermal cells sheathing the TB and delimiting the sensory epithelium from the regular epidermal cells. Close to the basal membrane, the nuclei occupy more or less the entire basal cell volume. Numerous nerve endings and synaptic vesicles intermingle with the basal cells and with the basal region of the receptor cells.

SCCs. Spindle-shaped cells are found scattered mostly in the tip region of the pelvic fins. They are associated with nerve fibers and show a single apical receptor villus (Fig. 6).

Goblet cells. Goblet cells are found all over the epidermis. These ovoid cells are characterized by a basal core and an apical opening at the skin surface (Fig. 6). In histological sections, positive alcian blue staining indicates mucin secretion, i.e., mucopolysaccharides and glycosaminoglycans.

Club cells. Secretions from the club cells (Fig. 6) are different from those of the goblet cells but the exact nature of those secretions were not identified within the framework of this study. These cells are more numerous than the goblet cells and differ by peculiar features: an elongated shape, a round, central nucleus, a distribution in all the epidermal layers and a cytoplasm rich in mitochondria, endoplasmic reticula and vesicles. On the most superficial club cells, these vesicles are close to the apical membrane and appear to discharge substances to the outside.

DISCUSSION

The subfamily Ophidiinae, containing the cusk-eels, is characterized by pelvic fins positioned far

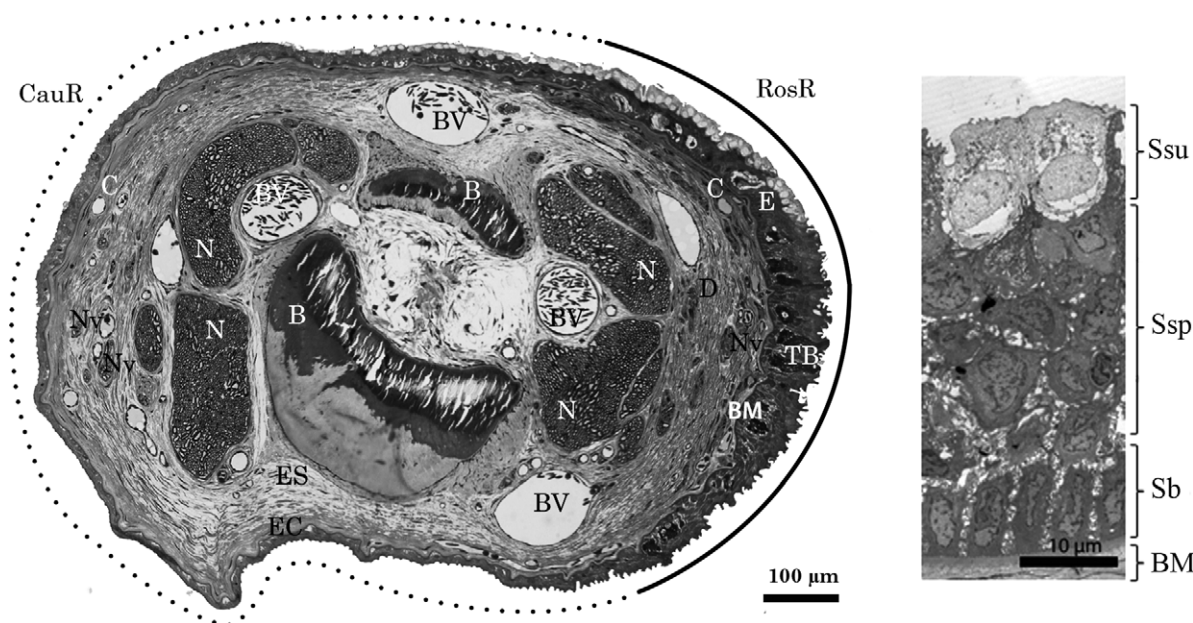


Fig. 4. Cross-section of the long pelvic fin (A) in *Ophidion rochei* showing the dermis (D) and epidermis (E) well separated by the basal membrane (BM). The dermis is divided into the stratum spongiosum (Es) and the stratum compactum (Ec). It contains blood vessels (BV), capillaries (c), nerves with sections of a different diameter (Nv, N), and bones (B). The epidermis surface is divided into a caudal (CauR, dotted line) and a rostral (RosR, full line) region, which contains the TBs. Toluidine blue staining. B: TEM micrograph of the rostral part of epidermis showing stratum basale (Sb), stratum spinosum (Ssp) and stratum superficiale (Ssu) with goblet cells.

forward and supported by a bone connected to the pectoral girdle (Howes, 1992; Nielsen et al., 1999). The major functions of fish fins are usually related to motion, such as propulsion, direction and balance. These locomotory functions are not assumed to be present in the pelvic fins in *Ophidion rochei*. The numerous muscles provide a greater degree of freedom in comparison to pelvic fins in generalized teleosts. It enables the fins to perform continuous sweeping movements and, simultaneously, to follow the surface relief (Fig. 1). This ability allows the fish adjusting its position continuously. Moreover, the reduction in pelvic fin size, the loss of pelvic fin rays, and the position of pelvic fins far forward on the body are suited for the mode of life of *O. rochei* because they may facilitate burrowing tail-first.

The cusk-eel fins have sensory functions that can be inferred from different sensory cell types and numerous free nerve endings that are innervated by the facial and spinal nerves, the combination of which allows taste and tactile reception (Davenport and Caprio, 1982; Kotrschal et al., 1993a). Some other fishes also have elongated sensory fin rays that can extend the range of perception (Fox, 1999), but in many species they do not bear TBs (Kotrschal et al., 1984; Silver and Finger, 1984; Ono, 1979) as those observed in *O. rochei*. Furthermore, the distribution of sensory cells around the pelvic fins clearly corresponds with the way the cusk-eel uses its appendages to survey the environment (Fig. 1). The lateral-rostral side that

usually faces the ground is thicker and shows a higher TB density. It confirms that this side is mainly used to explore the substrate, a function that inevitably involves rubbing of the epidermis and the adaptive development of larger squamous cells, which are regularly shed.

Taken all these previous observations into account, pelvic fins in *O. rochei* appear considerably modified and show many characteristics found in barbels (Fox, 1999): 1) they are elongated structures; 2) they are covered with skin; 3) they are supported by a bony skeleton; 4) they are innervated by a branch of the cranial nerve (*ramus lateralis accessorius*); 5) they have sensory cells in the epidermis. Although common characteristics and repeated convergences of these kinds of appendages have been found in different teleost taxa, there is a lack of homology among structures assigned the name barbel. This sensory structure is usually developed to increase exploitation of food reserves at the littoral bottom, fish with barbels feeding mainly on zoobenthos. The case of *Ophidion rochei*, however, is quite unusual because this kind of convergent adaptation is mainly associated with the pectoral girdle or the skull.

Some other benthic Ophidiiformes possess the same kind of pelvic fins and also have burrowing habits or live in deep waters where light is missing. The specialization of pelvic fins and their position far forward on the head should help these benthic fishes to explore the ocean bottom.

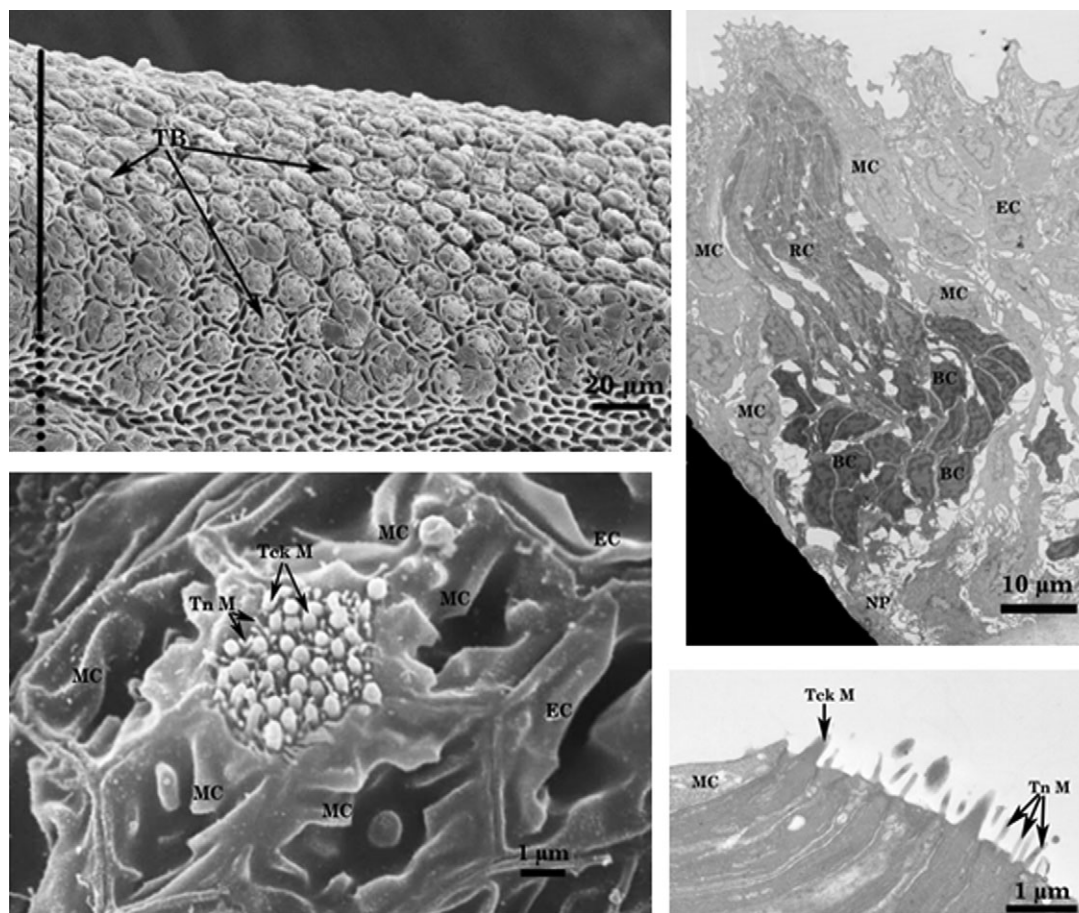


Fig. 5. Scanning electron micrographs of: long pelvic fin surface of *Ophidion rochei* showing the regular distribution of TBs (A) and detail of a TB (B). TEM micrographs showing detail of TB morphology (C) and of their terminal microvilli (D). In A, the full black line corresponds to the rostral region and the dotted line to the caudal region. BC: basal cell; EC: epithelial cell; MC: marginal cell; NP, nerve plexus; RC: receptor cell; TBs: taste buds; Tck M: thick microvillus; Tn M: thin microvillus.

In teleosts, barbels are thought to have a range of different functions. The more general and reasonably likely view is that they are gustatory, chemosensory and tactile organs mainly concerned with food location for fishes living in turbid waters (Davis and Miller, 1967; Gosline, 1984; Lombarte and Aguirre, 1997). This capability is well-known for barbels that are covered with TBs (Grover-Johnson and Farbman, 1976; Ovalle and Shinn, 1977; Fujimoto and Yamamoto, 1980; McCormick, 1993; Sakata et al., 2001; Harvey and Batty, 2002; Hansen et al., 2002; Northcutt, 2005; Zhang et al., 2006). In their comparison between two species, *Mullus surmuletus* and *Mullus barbatus*, living on rocky and muddy bottoms respectively, Lombarte and Aguirre (1997, 2000) showed that barbels in *M. barbatus* have a higher density of TBs (336 TBs mm²) than those in *M. surmuletus* (200 TBs mm²). These characteristics are associated with an increased sensitivity, as it is more difficult to locate prey in muddy bottoms where visibility is reduced and where chemical and tactile stimuli are the most important information sources for

feeding. The density of TBs on the distal tip of pelvic fins in *O. rochei* (350 TBs mm⁻²) is in the same range as that reported in *M. barbatus* by Lombarte and Aguirre (1997, 2000). *Ophidion rochei* is also a nocturnal predator inhabiting sandy muddy bottoms (Bacescu et al., 1957; Banarescu, 1964; Matallanas and Riba, 1980; Matallanas, 1981; Parmentier et al. 2010). Free nerve endings or TBs on the pelvic fins are probably used to perceive the different substrate textures allowing fish to find a hiding place.

Because of the distribution of the TBs, their specific stimuli and certain behavioral responses elicited, SCCs have been found to be distinct from TBs (Kotrschal et al., 1993a, b; Essler and Kotrschal, 1994). Electrophysiological recordings from this organ have shown a narrowly tuned chemoresponsiveness to only a few natural stimuli, such as skin surface washes from other fishes (Peters et al., 1991; Kotrschal, 1996). The function is not clear because it can vary within different taxa. In rocklings (*Ciliata mustela*), SCCs of the anterior dorsal fin can respond to heterospecific fish mucus

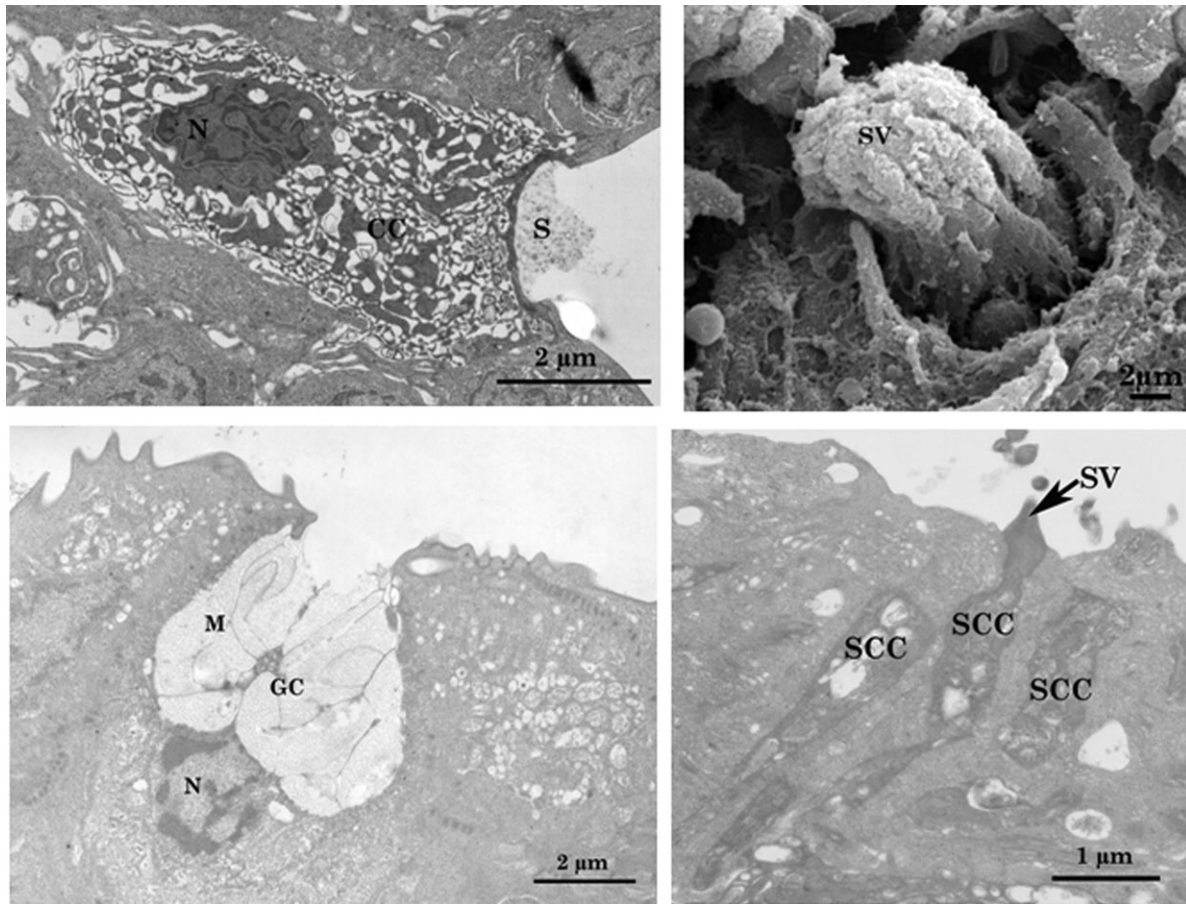


Fig. 6. Transmission electron micrographs of a club cell (A), goblet cells (C) and SCCs (D). The SEM micrograph shows the SCCs (B). CC, club cell; GC, goblet cell; M, mucin secretion; N, nucleus; S, secretion; SV, single villus.

involved in predator detection (Kotrschal et al., 1993b, Kotrschal, 2000; Peters et al., 1991). It is not possible to relate SCCs to a specific cusk-eel behavior within the framework of this study.

In fish, club cells can produce 1) alarm substances (mainly in Ostariophysi), 2) substances that promote wound healing (Pfeiffer, 1963; Smith, 1992; Ralphs and Benjamin, 1992) or 3) substances that confer protection against pathogens, parasites and UV radiation as well as against agents compromising the integrity of the epidermal layer (Chivers et al., 2007). Club cells in *Ophidion rochei* have a distribution and many ultrastructural characteristics in common with those of *Carapus acus*, member of the sister taxa Carapidae. In the latter, mucous secreting club cells do not contain alarm substances (Pfeiffer, 1963). This suggests that club cells of *Ophidion rochei* may also be related to mucous secretion and could have a protective function. Indeed, they structurally contrast with those of fishes releasing alarm substances, where club cells do not open at the surface (Pfeiffer, 1963; Suzuki and Kaneko, 1986).

CONCLUSIONS

Pelvic fins of *O. rochei* are highly modified and show strong similarities with barbels because they have identical sensory cell types, innervation and sensory functions. Based on the kinds of cells present, pelvic fins are capable at least of chemoreception and mechanoreception. These specialized pelvic fins are suited to the nocturnal mode of life and bottom dwelling habits of *O. rochei* and may allow for probing the substrate.

ACKNOWLEDGMENTS

N. Decloux kindly assisted in the microscopic study and R. Grgicevic in the fieldwork. L.K. is a PhD student of the Fonds National de la Recherche Scientifique of Belgium (F.R.S.-FNRS). The authors also thank the Centre of Applied Technology in Microscopy (ULg, Belgium) for providing access to electron microscopy equipments and two anonymous referees that kindly help to improve this manuscript.

LITERATURE CITED

- Aguirre H, Lombarte A. 2000. Distribution pattern of taste buds along hyoid barbel of *Mullus barbatus* and *M. surmuletus*. *Brain Beh Evol* 56:323–329.
- Bacescu M, Dimitrescu E, Manea V, Por F, Mayer R. 1957. Les sables à Corbulomya maeotica Mil., base trophique de premier ordre pour les poissons de la Mer Noire. *Trav Mus Hist Nat Gr Antipa* 1:305–374.
- Banarescu P. 1964. Pisces. Osteichthyes. Fauna Rep Pop Rom, Bucuresti 13:1–962.
- Chivers DP, Wisenden BD, Hindman CJ, Michalak TA, Kusch RC, Kaminskyj SGW, Jack KL, Ferrari MCO, Pollock RJ, Halbgewachs CF, Pollock MS, Alemadi S, James CT, Savaloja RK, Goater CP, Corwin A, Mirza RS, Kiesecker JM, Brown GE, Adrian JC Jr, Krone PH, Blaustein AR, Mathis A. 2007. Epidermal 'alarm substance' cells of fishes are maintained by non-alarm functions: Possible defence against pathogens, parasites and UVB radiation. *Proc R Soc B* 274:2611–2619.
- Davenport CJ, Caprio J. 1982. Taste and tactile recordings from the ramus recurrens facialis innervating flank taste buds in the catfish. *J Comp Physiol A* 147:217–229.
- Davis BJ, Miller RJ. 1967. Brain patterns in minnows of the genus *Hybopsis* in relation to feeding habits and habitat. *Copeia* 1967:1–39.
- Eastman JT, Eakin RR. 2001. Mental barbel and meristic variation in the Antarctic notothenioid fish *Dolloidraco longedorsalis* (Perciformes: Artedidraconidae) from Ross Sea. *Polar Biol* 24:729–734.
- Essler H, Kotrschal K. 1994. High resolution analysis of swim path patterns of intact and olfaction-deprived minnows (*Phoxinus phoxinus*) stimulated with food and potential predator odour. *J Fish Biol* 45:555–567.
- Fox H. 1999. Barbels and barbel-like tentacular structures in sub-mammalian vertebrates: A review. *Hydrobiologia* 403: 153–193.
- Freihofer WC. 1970. Some nerve patterns and their systematic significance in paracanthopterygian, salmoniform, gobioid and apogonid fishes. *Proc Calif Acad Sci* 38:215–264.
- Fujimoto S, Yamamoto K. 1980. Electron microscopy of terminal buds on the barbels of the silurid fish, *Corydoras paleatus*. *Anat Rec* 197:133–141.
- Geerinckx T, Verhaegen Y, Adriaens D. 2008. Ontogenetic allometries and shape changes in the suckermouth armoured catfish *Ancistrus cf. Triradiatus* Eigenmann (Loricariidae, Siluriformes), related to suckermouth attachment and yolk-sac size. *J Fish Biol* 72:803–814.
- Genten F, Terwinghe E, Danguy A. 2009. Atlas of fish histology. Enfield, NH: Science Publishers. 215 p.
- Gill T. 1863. Description of a new generic type of Ophidioids. *Proc Acad Nat Sci Phil* 15:209–211.
- Gosline WA. 1984. Structure, function and ecology in the goat-fishes (family Mullidae). *Pac Sci* 38:312–323.
- Grassé PP. 1958. Agnathes poissons anatomie, éthologie, systématique. *Traité de Zoologie*. Tome XIII: Fascicule 3. Paris: Masson et Cie. 950 p.
- Grover-Johnson N, Farbman AI. 1976. Fine structure of taste buds in the barbel of the catfish, *Ictalurus punctatus*. *Cell Tiss Res* 169:395–403.
- Hansen A, Reutter K, Zeiske E. 2002. Taste bud development in the zebrafish, *Danio rerio*. *Dev Dyn* 223:483–496.
- Harvey R, Batty RS. 2002. Cutaneous taste buds in gadoid fishes. *J Fish Biol* 60:583–592.
- Howes GJ. 1992. Notes on the anatomy and classification of ophidiiform fishes with particular reference to abyssal genus *Acanthonus* Günter, 1878. *Bull Br Mus Nat Hist (Zool)* 58:95–131.
- Iger Y, Abraham M, Wendelaar Bonga SE. 1994. Response of club cells in the skin of the carp *Cyprinus carpio* to exogenous stressors. *Cell Tissue Res* 277:485–491.
- Kim BJ, Yabe M, Nakaya K. 2001. Barbels and related muscles in Mullidae (Perciformes) and Polymixiidae (Polymixiiformes). *Ichth Res* 48:409–413.
- Kotrschal K. 1991. Solitary chemosensory cells—taste, common chemical sense or what? *Rev Fish Biol Fisheries* 1:3–22.
- Kotrschal K. 1996. Solitary chemosensory cells: Why do primary aquatic vertebrates need another taste system? *Trends Ecol Evol* 11:110–114.
- Kotrschal K. 2000. Taste(s) and olfaction(s) in fish: A review of specialized subsystems and central integration. *Pflügers Arch* 439:178–180.
- Kotrschal K, Whitear M, Adam H. 1984. Morphology and histology of the anterior dorsal fin of *Gaidropsarus mediterraneus* (Pisces, Teleostei), a specialized sensory organ. *Zoomorphology* 104:365–372.
- Kotrschal K, Whitear M, Finger TE. 1993a. Spinal and facial innervation of the skin in the gadid fish *Ciliata mustela* (Teleostei). *J Comp Neurol* 331:407–417.
- Kotrschal K, Peters R, Atema J. 1993b. Sampling and behavioral evidence for mucus detection in a unique chemosensory organ: The anterior dorsal fin in rocklings (*Ciliata mustela*, Gadidae, Teleostei). *Zool Jb Physiol* 97:47–67.
- LeClair EE, Topczewski J. 2010. Development and regeneration of the zebrafish maxillary barbel: A novel study system for vertebrate tissue growth and repair. *PLoS One* 5:8737.
- Lo Bianco S. 1907. L'origine dei barbigli tattili nel genere *Mullus*. *Atti Accad Lincei* 16:577–586.
- Lombarte A, Aguirre H. 1997. Quantitative differences in the chemoreceptor systems in the barbels of two species of Mullidae (*Mullus surmuletus* and *M. barbatus*) with different bottom habitats. *Mar Ecol Prog Ser* 150:57–64.
- Lombarte A, Aguirre H. 2000. Distribution pattern of taste buds along hyoid barbel of *Mullus barbatus* and *M. surmuletus*. *Brain Behav Evol* 56:323–329.
- Mann DA, Bowers-Altman J, Rountree RA. 1997. Sounds produced by the striped cusk-eel *Ophidion marginatum* (Ophidiidae) during courtship and spawning. *Copeia* 1997:610–612.
- Matallanas J. 1979. Algunos datos comparativos de *Ophidion barbatum* y de *O. rochei* (Pisces, Ophidiidae). *Nuevas citas de O. rochei* para el Mediterráneo occidental. *Cah Biol Mar* 20:351–359.
- Matallanas J. 1981. Regimen alimentario de *Ophidion rochei* (Pisces, Ophidiidae) en el Mediterráneo español. Comparación con el de *O. barbatum*. *Bol Inst Esp Oceanog* 4:174–185.
- Matallanas J, Riba G. 1980. Aspectos biológicos de *Ophidion barbatum* Linnaeus, 1758 y *O. rochei* Muller, 1845 (Pisces, Ophidiidae) de la costa catalana. *Invest Pesquera* 44:399–406.
- McAllister DE. 1968. The evolution of branchiostegals and associated opercular, gular, and hyoid bones and the classification of teleostome fishes, living and fossil. *Nat Mus Canada Bull (Biol Ser)* 221:1–239.
- McCormick MI. 1993. Development and changes at settlement in the barbel structure of the reef fish, *Upeneus tragula* (Mullidae). *Environ Biol Fishes* 37:269–282.
- Nelson JS. 2006. *Fishes of the World*, 4th ed. New York: Wiley-Interscience. 245 p.
- Northcutt RG. 2005. Taste bud development in the channel catfish. *J Comp Neurol* 482:1–16.
- Nielsen JG, Cohen DM, Markle DF, Robins CR. 1999. Ophidiiform Fishes of the World (order Ophidiiformes), Vol. 18. Rome: FAO. Species Catalog, 178 p.
- Ohkubo Y, Masubuchi M, Fujioka K, Tomita Y, Matsushita T, Ohsuga K, Marui T. 2005. Distribution and morphological features of taste buds in the zebrafish, *Danio rerio*. *J Oral Biosci* 47:77–82.
- Ono RD. 1979. Sensory nerve endings of highly mobile structures in two marine teleost fishes. *Zoomorphologie* 92:107–114.
- Ovalle WK, Shinn SL. 1977. Surface morphology of taste buds in catfish barbels. *Cell Tissue Res* 178:375–384.
- Parmentier E, Fontenelle N, Fine ML, Vandewalle P, Henrist C. 2006. Functional morphology of the sonic apparatus in *Ophidion barbatum* (Teleostei, Ophidiidae). *J Morphol* 267:1461–1468.
- Parmentier E, Bouillac G, Dragicevic B, Dulcic J, Fine ML. 2010. Call properties and morphology of the sound-producing

- organ in *Ophidion rochei* (Ophidiidae). J Exp Biol 213:3230–3236.
- Peters R, Kotrschal K, Krautgartner WD. 1991. Solitary chemoreceptor cells of *Ciliata mustela* (Gadidae, Teleostei) are tuned to mucoid stimuli. Chem Sens 16:31–42.
- Pfeiffer W. 1963. Alarm substances. Experientia 19:113–168.
- Ralphs JR, Benjamin M. 1992. Chondroitin and keratan sulfate in the epidermal club cells of teleosts. J Fish Biol 40:473–475.
- Rosen DE, Patterson C. 1969. The structure and relationships of the paracanthopterygian fishes. Bull Am Mus Nat Hist 141:357–474.
- Sakata Y, Tsukahara J, Kiyohara S. 2001. Distribution of nerve fibers in the barbels of sea catfish *Plotosus lineatus*. Fish Sci 67:1136–1144.
- Sato M. 1977. Histology of barbels of *Blepsias cirrhosus draciscus* (Cottidae). Jap J Ichthyol 23:4.
- Schaefer SA, Lauder GV. 1986. Historical transformation of functional design: Evolutionary morphology of feeding mechanisms in loricarioid catfishes. Syst Zool 35:489–508.
- Silver WL, Thomas EF. 1984. Electrophysiological examination of a non-olfactory, non-gustatory chemosense in the searobin, *Prionotus carolinus*. J Comp Physiol A 154:167–174.
- Smith RJF. 1992. Alarm signals in fishes. Rev Fish Biol Fisheries 2:33–63.
- Stiassny MLJ, Moore JA. 1992. A review of the pelvic girdle of acanthomorph fishes, with comments on hypotheses of acanthomorph intrarelationships. Zool J Linn Soc 104:209–242.
- Suzuki Y, Kaneko T. 1986. Demonstration of the mucous hemagglutinin in the club cells of eel skin. Dev Comp Immunol 10:509–518.
- Taylor WR, Van Dyke GC. 1985. Revised procedures for staining and clearing small fishes and other vertebrates for bone and cartilage study. Cybium 9:170–109.
- Winterbottom R. 1974. A descriptive synonymy of the striated muscles of the teleostei. Proc Acad Nat Sci Philad 125:225–317.
- Zhang G, Deng S, Zhang H, Li H, Li L. 2006. Distribution of different taste buds and expression of *a*-gustducin in the barbels of yellow catfish (*Pelteobagrus fulvidraco*). Fish Phys Biochem 32:55–62.