

Chemical and visual sensory systems in  
feeding behaviour of the  
Antarctic fish *Ophthalmolycus amberensis*  
(Zoarcidae)

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南極産げんげ科の魚 *Ophthalmolycus amberensis* の摂餌行動における化学的、  
視覚的刺激感覚

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**要旨:** 南極産げんげ科に属する魚 *Ophthalmolycus amberensis* はサウスシェトランド諸島, キングジョージ島, アドミラルティ湾の深度 140–200 m の低照度層に出現する。この低照度環境下における摂餌行動時の行動的, 感覚的適応度を評価するために室内実験を行った。おきあみ死体, 魚肉, 端脚類生体を餌料として選んだ。摂餌行動は主として化学刺激によって誘起される。視覚刺激は摂餌行動の開始までに長時間を要する弱い誘因である。従って *O. amberensis* は大きな嗅上皮, 鼻と鼻孔附近の高密度の味蕾, 長い桿体を持ち堆体を欠く網膜を持つ。食物の選択性が認められる。これに応じた多様な形の味蕾が口唇上と口咽頭腔に存在する。光の有無にかかわらず, 食餌発見の最初の検出器官として化学的感覚系を使用する能力により, *O. amberensis* は周年海底の異なった生息環境を有効に活用することができるようになっている。

**Abstract:** The Antarctic eelpout *Ophthalmolycus amberensis* occurs in Admiralty Bay (King George Island, South Shetlands), at 140–200 m depth, where light intensity is low. To assess behavioural and sensory adaptations for feeding under these conditions, laboratory tests were undertaken. Dead krill, fish fillet, and live amphipods were the preferred food items. Feeding responses were mainly induced by chemical stimuli. Visual stimuli were weak elicitors, leading to a long delay in the initiation of feeding behaviour. These fishes present a large olfactory epithelium, a high density of taste buds on the snout and close to the nostrils, and a retina that contained long rods, but no cones. Food selection was observed. Varied types of taste buds were present on the lips and in the oro-pharyngeal cavity. The capacity to use a chemo-sensory system as first elicitor for food detection, either in the absence or presence of light, allows *O. amberensis* to efficiently exploit different habitats at the sea bottom, in all Antarctic seasons.

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## 1. Introduction

The family Zoarcidae (eelpouts) is thought to be a monophyletic group with 220 species worldwide. They have been endemic to Antarctica since the Miocene (Andrishev, 1987; Anderson, 1988). Although only 22 species inhabit the Antarctic region, zoarcids are the dominant non-notothenioid group (Anderson, 1984; Eastman, 1993). As a rule zoarcids are sluggish benthic slope dwellers (Anderson, 1984; Fisher and Hureau, 1985; Eastman, 1993) that are carnivorous epi- or in-faunal predators (Anderson, 1990). Among them, *Ophthalmolycus amberensis* has a circum-Antarctic distribution (Anderson, 1990), occurring in near-shore areas at depths characterised by low light conditions even during the Antarctic summer. Thus, one problem to be faced by these fish is that of food detection and capture under such environmental conditions.

The relationship between fish and their environment is determined by their chemo-, mechano-, and photo-sensory systems. Fishes may use one or more of these systems to detect food (Montgomery and Macdonald, 1987; Montgomery and Coombs, 1992; Løkkeborg *et al.*, 1995; Fanta *et al.*, 1994, 1999; Meyer and Fanta, 1998), although nearly all fish rely on chemoreception to some degree (Hara, 1971). In the Antarctic environment, light conditions change both seasonally and during the course of the day, implying varied stimulation of the sensory systems of fish. This also implies varied quantity and quality of available food. Consequently, fish may switch prey type, and feeding strategy, according to the capacity of their anatomical and sensory structures (Daniels, 1982; Montgomery and Macdonald, 1987; Fanta *et al.*, 1994, 1999; Grötzner and Fanta, 1998; Meyer and Fanta, 1998).

Even though it belongs to the dominant non-notothenioid group of fish in Antarctica, the behaviour of *O. amberensis* is unknown. The only available information is on their distribution (Anderson, 1984; Fisher and Hureau, 1985), and their habitat and feeding ecology (Anderson, 1984; Rakusa-Suszczewski, 1993).

Several experiments were conducted in tanks to establish their feeding habits and choice of food. Following these observations, some tests aimed to establish to what extent vision and the chemical senses are used in food detection and selection. The tests were followed by an analysis of the morphological complexity of both the visual and the chemical sensory systems.

The aim of this study is to understand the ecological significance of some of the constraints imposed not only by the Antarctic environment, but also by the morpho-functional characteristics of some sensory structures, or the feeding behaviour of the eelpouts.

## 2. Materials and methods

*Ophthalmolycus amberensis* (Tomo, Marchoff and Torno, 1977) were trapped at 140–200 m depth, in Ezcurra Inlet, Admiralty Bay, King George Island (62°09'S, 58°26'W) during the Antarctic summer of 1993/94. The fish ( $140 \pm 10$  mm standard length) were acclimatised to laboratory conditions for 30 days, in 84 L aquaria, at constant temperature

( $0 \pm 0.5^\circ\text{C}$ ), pH ( $7.5 \pm 0.3$ ), and salinity ( $33.5 \pm 0.2$  ppt), with a 20L: 4D photoperiod. During the light period, the light intensity was 7–10 lux at the bottom of the tanks, and during the dark period the aquaria were exposed to a weak red light. During acclimation a variety of food items (Amphipoda, Gastropoda, Isopoda, Polychaeta, Opisthobranchia, dead krill and fish fillet) were introduced into the aquaria. The fish were allowed to feed *ad libitum*.

### 2.1. Feeding behaviour and food preferences

Based on the food items that *O. amberensis* ingest in their natural habitat (Anderson, 1984), the amphipods *Gondogeneia antarctica*, and *Waldeckia obesa* (total length (TL) 0.2–0.8 cm), polychaetes (TL 2–4 cm), opisthobranchs and gastropods (TL 0.3–0.4 cm), isopods (TL 1.3–1.5 cm), fragments of algae, dead krill (*Euphausia superba*) (TL 2.8–3.5 cm), and fish fillet (pieces 0.09–0.25 cm<sup>3</sup>) were continuously available. The food acceptance was recorded for all fish.

### 2.2. Visual and chemical stimulation for feeding

Aquaria with the same characteristics as for acclimation were used for the tests, each one containing 5 fish. One transparent vessel containing water with the same quality as in the aquaria was centrally placed inside each test aquarium. Mobile and static food was offered, respectively amphipods (around 100 individuals of *G. antarctica*) and fish fillets (5 pieces) were added to the vessel by means of a funnel, and behind a shield, at the start time  $T_0$  (Meyer and Fanta, 1998; Fanta *et al.*, 1999). A vessel with water but no food was the blank.

Five experimental arrangements were used:

(i) Visual stimulation for feeding: there was no contact between the water of the aquarium and the water of the food container, but fish could see the food. These experiments were carried out during the light period;

(ii) Visual and chemical stimulation for feeding: the food was placed inside the food container and there was communication at the aquarium surface between the water in the food container and the water in the aquarium. This allowed the fish to detect the food visually and chemically. These experiments were carried out during the light period;

(iii) Chemical stimulation for feeding: as for (ii) except that these experiments were carried out in darkness. This allowed the fish to detect the food through the chemical sensory system;

(iv) Chemical and mechanical stimulation: food was placed directly inside the aquarium with eelpouts, allowing spatial chemical and mechanical perception of their presence, but not visual. These experiments were carried out in darkness;

(v) Entire sensory system stimulation for feeding: as for (iv) except that the experiments were carried out in light.

The reactions of all 5 fish in the aquaria were recorded for 10 min after  $T_0$ . Five repetitions were done for each experimental situation. The tests started when fish were resting, and 48 hours after the last time the fish had been fed.

### 2.3. Morphology of visual and chemical sensory organs

To analyse the structure of the nostrils, the neighbouring pores, the olfactory rosette, and the olfactory chamber, samples fixed in neutral formaline 4% and maintained in 70% alcohol were observed under a stereoscopic microscope. China-ink was carefully introduced inside the nostrils, by means of a very thin needle connected to a syringe, and the flux of ink observed. After dissection, the olfactory region was drawn under a camera-lucida. Olfactory rosettes were dissected out and measured.

For histology, under light microscopy (LM) samples of the upper and lower lips, olfactory rosette, head skin, oro-pharyngeal cavity, and of the retina were dissected out from all fish. Samples were fixed in Bouin's fluid, and embedded in Paraplast Plus<sup>®</sup>. The 2  $\mu\text{m}$  sections were stained with Haematoxylin and Eosin (HE) (Clark, 1981) and modified Mallory's triple stain (Culling *et al.*, 1985) for general morphology. Alcian blue pH 2.5 and 1.0, and P.A.S. were used for the identification of mucus secreting cells (Culling *et al.*, 1985).

The entire retina, and the nostril and olfactory rosette, were dissected out and prepared for electron microscopy (Culling *et al.*, 1985). For scanning electron microscopy (SEM) these were fixed in Karnowski's fixative and cryo-fractured with liquid nitrogen, post fixed with 1% osmium tetroxyde in 0.1 M cacodylic acid buffer, and treated with tannic acid. After dehydration and achievement of the critical point with liquid CO<sub>2</sub>, the samples were covered with gold.

For transmission electron microscopy (TEM) samples of the retina were fixed with 2.5% glutaraldehyde in 0.2M cacodylic acid buffer, routinely processed (Culling *et al.*, 1985), and embedded in Epon-812 (Luft, 1961). Some 0.5  $\mu\text{m}$  sections were obtained and stained for LM with toluidine blue.

## 3. Results

### 3.1. Food preference

Only 14.6% of the total time of behaviour observations were dedicated to feeding activity, performed by one or more fish, even in the continuous presence of food. The proportion of eaten items in relation to the captured ones was 72.7%.

The amphipod *G. antarctica* was ingested preferentially. Isopods and algae were taken rarely. Dead krill, and fish fillet, were taken several times. All other items were not apprehended. Each fish ingested around 28 amphipods every other day. Overfeeding was not observed. Once satiated, *O. amberensis* stopped showing any interest for food.

After apprehension, the potential food was retained in the mouth for 25 to 199s before acceptance or rejection. Fish sometimes caught debris, sand grains or little fragments of algae and rejected them immediately after apprehension. Often the same piece was taken and rejected several times.

### 3.2. Stimuli for feeding

#### 3.2.1. Visual stimuli

When food was offered inside the transparent vessel during the light period, and no communication was established between the water in the vessel and the water of the

Table 1. Behavioural reactions of *Ophthalmolycus amberensis* to stimuli for feeding in 28 tests. All times (mean values) were considered as delays after the initial times  $T_0$ , when food was introduced into either the vessel inside the aquarium or the aquarium.

Type of stimuli	Alert	Start swimming	Feeds
(i) visual	4 min	4 min 2 s	—
(ii) visual+chemical	1 min 25 s	3 min 33 s	4 min 10 s
(iii) chemical	1 min 12 s	2 min 1 s	5 min 16 s
(iv) mechanical+chemical	30 s	45 s	1 min 1 s
(v) visual+mechanical+chemical	1 min 25 s	1 min 28 s	4 min 3 s

aquarium with fish, there was a slow reaction by few fish, and the delay time after  $T_0$  was long (Table 1). In most cases *O. amberensis* continued to rest at the bottom of the aquarium, in the same position in which they were prior to the introduction of food.

### 3.2.2. Visual and chemical stimuli

When amphipods or fish fillets were visible inside the glass container in the aquarium and there was continuity between the water in the container with that in the aquarium, *O. amberensis* showed a quick positive reaction to the presence of prey (Table 1). They swam toward the container, and then upward, approaching the place of contact of the aquarium water and the water with food and, entering the container, ingested the food.

### 3.2.3. Chemical stimuli

In darkness when amphipods or fish fillets were placed inside the glass container in the aquarium and there was continuity between the water in the container with that in the aquarium, fish could perceive the food chemically but not visually. They showed a quick and positive reaction, with a similar delay time as in 3.2.2, and the same behaviour.

### 3.2.4. Chemical and mechanical stimuli

When amphipods were introduced in the aquarium with *O. amberensis*, in darkness, they swam in all directions and became distributed throughout the aquarium. The fish fillets sank to the bottom of the aquarium. Both were detected at once, within the shortest delay time of all tests (Table 1). If one fish started swimming to capture prey, all other individuals started to be alert or to search for food, predominantly searching at the bottom.

### 3.2.5. Visual, chemical and mechanical stimuli

When amphipods or fish fillets were placed directly into the aquarium, during the light period, all senses were stimulated. The reaction was positive, after a similar delay time as in 3.2.2 and 3.2.3, and several fish fed.

## 3.3. Morphology of sense organs

### 3.3.1. Retina

The retina of *O. amberensis* is organised in ten layers. The pigment epithelium is thin, having few large melanin granules distributed in its cells with short processes. When the rods were exposed to light, the melanin was not inside the processes, therefore not

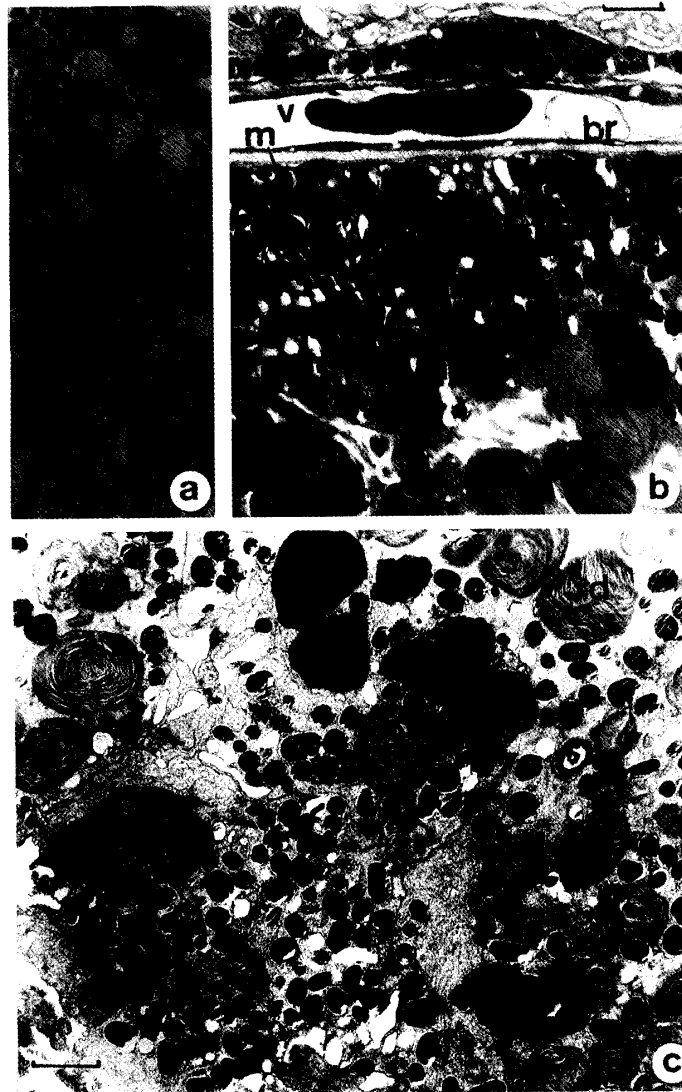


Fig. 1. Retina of *Ophthalmolycus amberensis*: a. external segment of the rods (r) and the pigment epithelium (e) (LM, stain toluidine blue, scale 5  $\mu\text{m}$ ); b. pigment epithelium cells with melanin grains (m), supported by Bruch's membrane (br) and blood vessels of the choroid (v) (TEM, scale 1  $\mu\text{m}$ ); c. epithelial cells replenished with semi-digested membrane disks of the rod tips (rd) (TEM, scale 1  $\mu\text{m}$ ).

surrounding the inner segment of the rods, but only touching their tips (Fig. 1a, b). The epithelial cells contained many partially digested rods' membranous disks (Fig. 1c).

Only one type of photoreceptor, the rods, is present (Fig. 2). Rods are long, and placed as a single layer with a mean density of 18.92 rods/100  $\mu\text{m}^2$  (range 15.28–29.16) (Fig. 2a). The outer segment is proportionally long, with a mean length of 57.96  $\mu\text{m}$  (range 56.25–65.00), in comparison with the inner segment which is 12.30  $\mu\text{m}$  long (range 10.00–15.00). The rods are cylindrical and the diameters of the outer and the inner segments are similar, 2.31  $\mu\text{m}$  (range 1.85–2.57) and 2.00  $\mu\text{m}$  (range 1.71–2.42) respectively (Fig. 2b). Their nuclei are elongated and placed at one level in the outer nuclear layer (Fig. 2b). The outer segments of the rods of *O. amberensis* have membranous lamellae, forming loose discs (Fig.

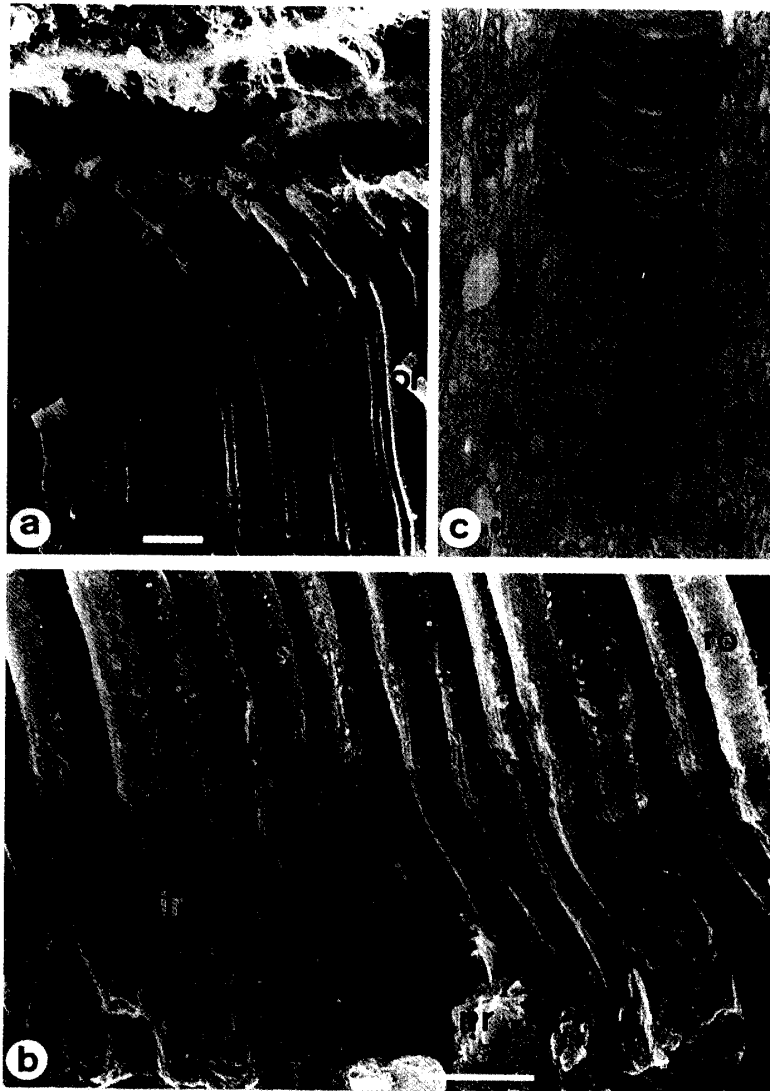


Fig. 2. Retina of *Ophthalmolycus amberensis*: a. rod layer showing mainly their parallel outer segments (or) (SEM, cryofracture, scale 1  $\mu\text{m}$ ); b. Detail of the outer (ro) and inner (ir) segments, and the nuclear layer (nr) (SEM, cryofracture, scale 5  $\mu\text{m}$ ); c. Detail of the same region as in (b), with the rod discs (rd), the connecting stalk (cs), and the inner segment with mitochondria (rm) and glycogen granules (gg) (TEM, scale 1  $\mu\text{m}$ ).

2c). The connecting stalk is short and broad. At the inner segment both the Golgi apparatus and the reticule are well developed. There is a high concentration of mitochondria and glycogen granules accumulated at their upper third, close to the outer segment (Fig. 2c).

Conducting and association neurons along with the supporting cells complete the other layers of the retina. These form only one third of its total thickness, 31.25  $\mu\text{m}$  out of 108.75  $\mu\text{m}$ , respectively.

### 3.3.2. Olfactory organ

*O. amberensis* have one cylindrical nostril at each side of the head (Fig. 3a, d). Two

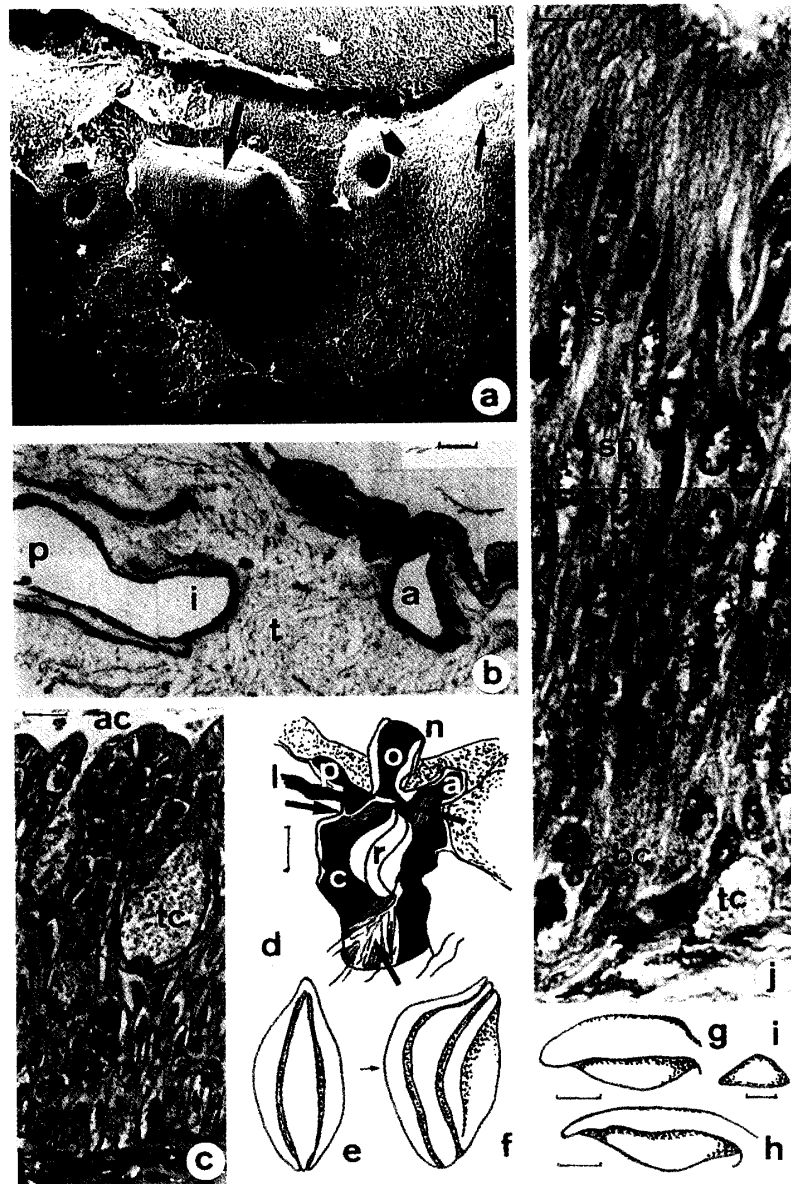


Fig. 3. Olfactory organ of *Ophthalmolycus amberensis*: a. nostril (long arrow) and lateral pores (short wide arrow) surrounded by many taste buds (small arrows) (SEM, scale 500  $\mu$ m); b. horizontal section showing a connection between the canal of the inhalant pore (i) and the neighbouring posterior pore (p), surrounded by coarse connective tissue (t). A similar connection can be seen between the olfactory chamber and the anterior pore (a) in sections at a deeper level (LM, stain HE, scale 0.1 mm); c. support stratified epithelium with connective tissue cells (tc), basal cell layer (bc) and apical irregular surface (ac) (LM, stain HE, scale 20  $\mu$ m); d. vertical section to the nostril (n) showing the olfactory bulb (o), the olfactory chamber (c), the olfactory rosette (r), the anterior pore (a), the posterior pore (p), the canal linked to the lateral line system (l), and strands of connective tissue (arrows) (scale 0.5 mm); e. frontal view of the three lamellae of the olfactory rosette; f. oblique view of the rosette, showing its position inside the olfactory chamber, with the convex part (arrow) toward the posterior region of the head; g. broad median olfactory lamella; h. thin lateral olfactory lamellae; i. triangular shape of the olfactory epithelium (scale 1 mm); j. olfactory epithelium with basal cells (bc), supporting cells (sp) and sensorial cells (sc), and connective tissue cells (tc) close to the basement membrane (LM, stain HE, scale 20  $\mu$ m).



pores are seen on slight elevations of the epithelial surface, one anterior and the other posterior to each nostril (Fig. 3a). These pores are more prominent than the pores of the lateral line system. There is internal communication among the olfactory chamber, the two pores and the canal of the lateral line system (Fig. 3b, d). As can be seen if some china-ink is added to water, the water flux enters the nostril, flows over the respiratory lamellae of the rosette, and leaves through the same pore. However, if the water that enters the olfactory bulb and the olfactory chamber is at high pressure, it can flow out through the posterior pore first, then through the anterior one and thereafter through the first two pores of the lateral line system.

The olfactory bulb is larger at the upper end, and narrower close to the olfactory chamber (Fig. 3d). The olfactory chamber is elongated, and shows some strands of connective tissue that hold the olfactory rosette in place (Fig. 3d). The olfactory rosette is oval (Fig. 3d, e, f), curved, with the convex side turned toward the posterior region of the head (Fig. 3d, f). It has three leaf shaped olfactory lamellae, with a mean length of 1.8 mm (Fig. 3e, f, g, h). The inner lamella is thicker (0.5 mm) than the two outer ones (0.2 mm) (Fig. 3g, h).

The olfactory epithelium lines the triangular sides of the lamellae (Fig. 3i). The total olfactory surface of one olfactory rosette has a mean area of 5.4 mm<sup>2</sup>. The epithelium is stratified, containing small basal cells, and very long supporting and sensory cells (Fig. 3c). Supporting cells have a dark elongated nucleus, and the sensory cells have a lighter one. The nuclei are placed at the upper 2/3 of the cell. The support epithelium is also stratified. It also has some specialised cells that are proportionally larger than the epithelial cells. They stain dark red with eosin, but not by any stain for mucopolysaccharides (Fig. 3j).

### 3.3.3. Taste organs

The taste buds of *O. amberensis* are of various sizes and shapes in different regions of the head, lips and oro-pharyngeal cavity.

Close to the nostrils and surrounding the eyes there are around 4.5 taste buds/mm<sup>2</sup> (Fig. 4a, b). They are pear-shaped with an apex that is either at the same level as the epithelial surface (Fig. 4e) or in a depression (Fig. 4d). The apical part of the taste buds has a mean diameter of 92  $\mu$ m. Taste buds are surrounded by a stratified epithelium with around 15 cell layers. The upper 5 or 6 layers of epithelial cells react positively to Alcian-blue. Some differentiated cells that stain positively with HE, but not with Alcian blue or P.A.S. (Fig. 4c), can be seen among the epithelial cells. The surface of the epithelial cells is polygonal and presents macaroni-like microridges (Fig. 4f). They retain the mucopolysaccharides that are produced by the goblet cells of the epithelium.

On the upper lips, the epithelial cell surface is covered by microridges in a homogeneous lace-pattern (Fig. 5d). Three types of taste buds were found: spindle-like with an apex of around 5  $\mu$ m diameter, at the same level as the epithelial surface, but surrounded by a circular groove (Fig. 5a); pear shaped, with an apex of around 8.5  $\mu$ m situated at the same level as the epithelial surface (Fig. 5b); and barrel-like, short but broad, all nuclei concentrated at the basal third of the bud, and the apical region of around 14  $\mu$ m diameter, situated in a concavity of the epithelium (Fig. 5c). The density was around 6 taste buds/mm<sup>2</sup>.

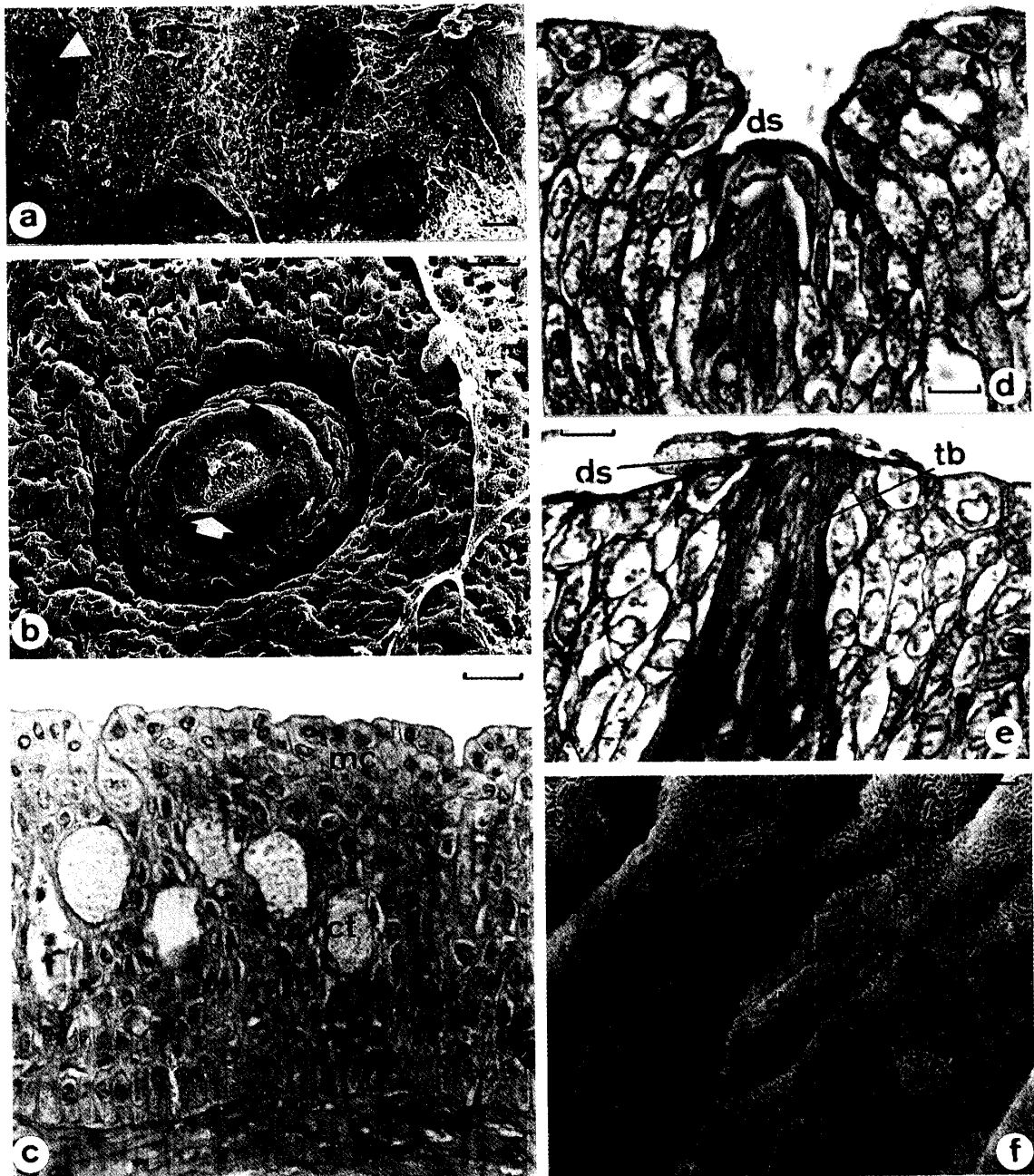


Fig. 4. Taste buds in the head epithelium of *Ophthalmolycus amberensis*: a. taste buds on the lateral surface of the head (arrow head), between the eyes and the nares (SEM, scale 200  $\mu\text{m}$ ); b. detail of one taste bud (short arrow), surrounded by concentric layers of epithelial cells (SEM, scale 50  $\mu\text{m}$ ); c. stratified epithelium from the region behind the eyes with mucopolysaccharide secreting cells (mc), migrating connective tissue cells (ct), cylindrical epithelial cells at the basal region (bc) (LM, stain HE, scale 20  $\mu\text{m}$ ); d. taste bud (\*) with apex in a depression of the epithelial surface (ds) (LM, stain HE, scale 10  $\mu\text{m}$ ); e. taste bud (tb) with apex at the same level as the epithelium surface (ds) (LM, stain HE, scale 10  $\mu\text{m}$ ); f. superficial view of the epithelial cells (EC) and depressions of the surface (DP) (SEM, scale 20  $\mu\text{m}$ ).

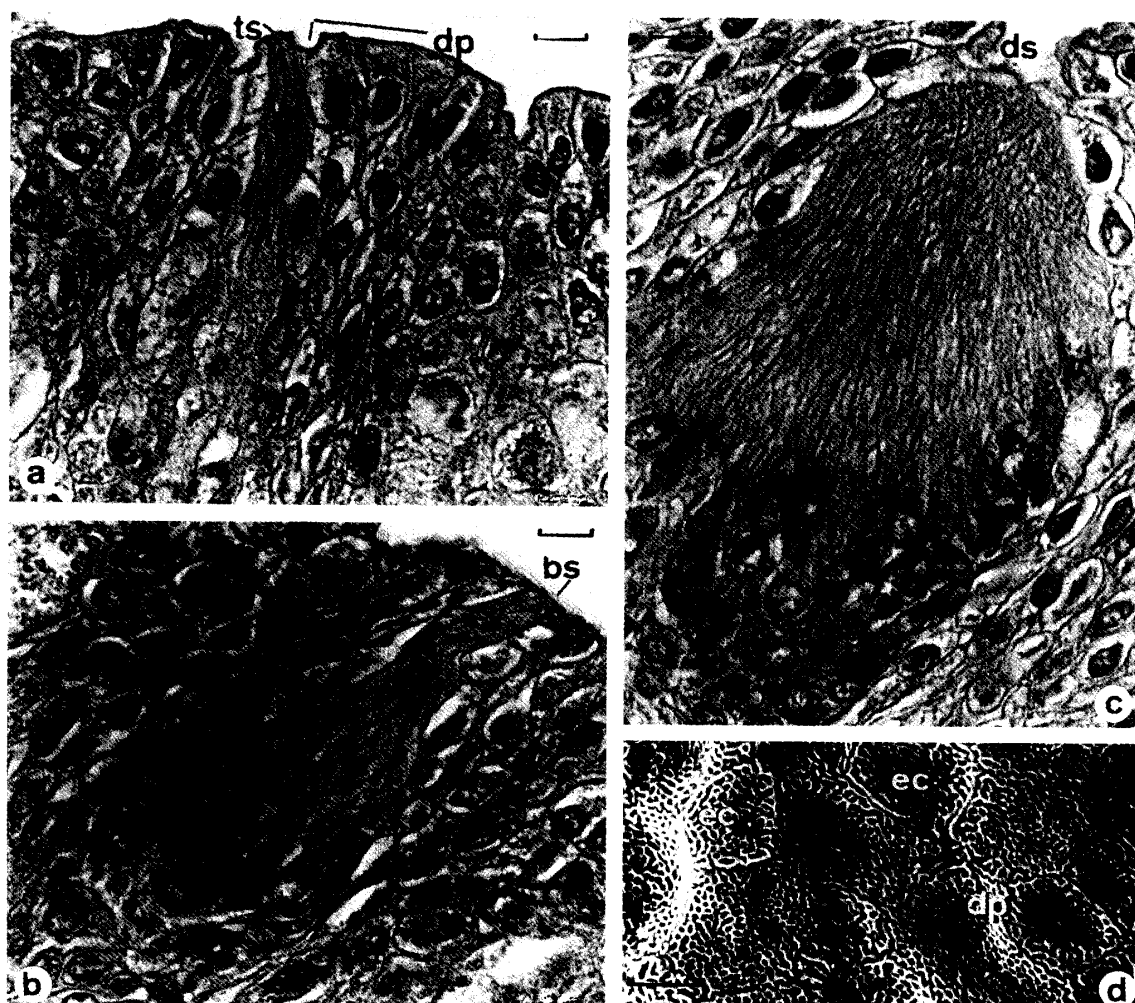


Fig. 5. Taste buds from the upper lips of *Ophthalmolycus amberensis*: a. taste bud (\*) with apex at the same level as the epithelium surface (ts), but surrounded by a depression (dp) (LM, stain HE, scale 10  $\mu\text{m}$ ); b. taste bud (\*) with apex at the same level as the epithelial surface (bs) (LM, stain HE, scale 10  $\mu\text{m}$ ); c. taste bud (ts) with apex in a depression of the epithelial surface (ds) (LM, stain HE, scale 10  $\mu\text{m}$ ); d. superficial view of the epithelial cells (ec) and depressions of the surface (dp) (SEM, scale 20  $\mu\text{m}$ ).

The epithelium of the lower lips has the same morphology as that of the upper lips except that no taste buds were seen in the preparations.

Inside the oro-pharyngeal cavity, varied taste buds were found in different locations: in front of palatal teeth where they are situated inside a depression and are surrounded by concentrically arranged epithelial cells, having an apical diameter of 200  $\mu\text{m}$  (Fig. 6a, b); behind the palatal teeth where they are arranged as a row, spaced at 400  $\mu\text{m}$  (Fig. 6a, b); at the palate (Fig. 6c) and gill arches (Fig. 6d) where they are much smaller, with an apical portion of around 30  $\mu\text{m}$ , but greater density (3.2 taste buds/ $\text{mm}^2$ ).

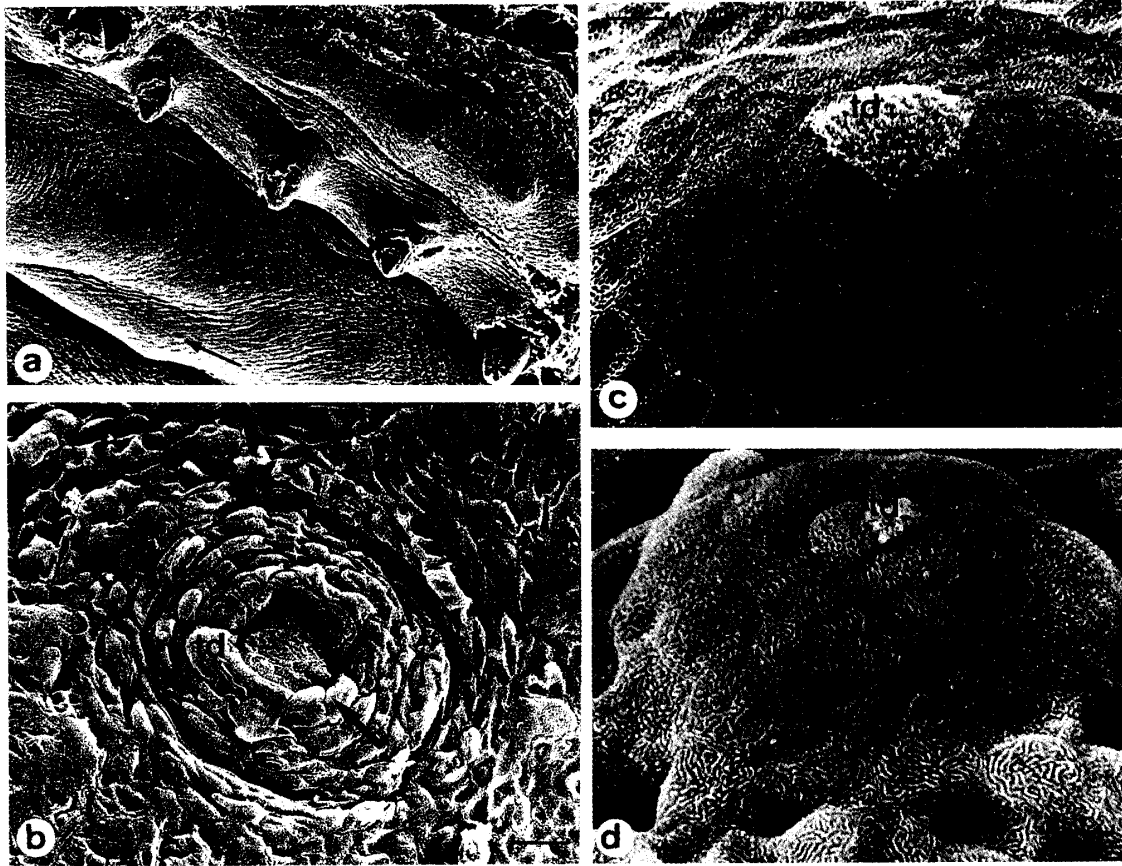


Fig. 6. Taste buds in the oro-pharyngeal cavity of *Ophthalmolycus amberensis*: a. palate teeth (\*) surrounded by numerous taste buds (arrow) (SEM, scale 200  $\mu\text{m}$ ); b. detail of taste bud from (a) (td) situated in an epithelial depression, and the concentric arrangement of epithelial cells (arrow) (SEM, scale 50  $\mu\text{m}$ ); c. taste bud at the palate, with broad apical part at an elevation of the epithelium (td) (SEM, scale 20  $\mu\text{m}$ ); d. taste bud at the pharyngeal region of the gill arch, with apical part on the top of epithelium elevation (td) (SEM, scale 20  $\mu\text{m}$ ).

#### 4. Discussion

*O. amberensis* is a zoarcid with a circum-Antarctic distribution (Anderson, 1990). They live at depths where, even during the Antarctic summer, light levels are low (Anderson, 1990), such as in Ezcurra Inlet, Admiralty Bay, King George Island (Rakusa-Suszczewski, 1993), the locality from which the experimental material used for this study was obtained. To face such varied environmental conditions these fish undergo several adaptations that are worth study.

Food was captured equally well by night as by day. However, feeding activity by *O. amberensis* was never continuous, and overfeeding was never observed, although some other Antarctic fish, such as the nototheniid *Notothenia neglecta*, frequently overfeed (Fanta and Meyer, 1998). The preferred food was the amphipod *Gondogeneia antarctica* a species eaten by several other fish in Admiralty Bay (Fanta, 1999). When there was a choice of food only these amphipods were taken, even though there are in the literature

indications of other taxa being represented in the diet of *O. amberensis* (Anderson, 1984). *O. amberensis* ingested only around two thirds of the items captured. Visual stimuli, particularly movements of the prey, caused a very slow reaction, differing from other Antarctic fish (Fanta *et al.*, 1994, 1999; Meyer and Fanta, 1998) that can be quickly stimulated by vision alone. *O. amberensis* used their vision to improve food capture, mainly after chemical stimulation, or in conjunction with odour, taste, and mechanical stimulation. Temporary retention of food within the mouth to taste or otherwise select it has also been reported in Nototheniidae (Meyer and Fanta, 1998; Rios and Fanta, 1998; Fanta, 1999). These behaviours are supported by the functional possibilities of the sensory structures of *O. amberensis*, as can be evidenced from histological examination of the retina, the olfactory organ and the taste buds.

The eye is typical of some other deep-sea fish with a similar mode of life (Marshall, 1971; Munz, 1971), where there are only rods in the retina. These are responsible for the detection of movement in low light conditions, an adaptation seen in some other zoarcids. Even at the low light intensity levels used in our experiments, rods were stimulated to activity. This can be suggested by the concentration of mitochondria and glycogen granules at their inner segment, and mainly by many partially digested disks inside the pigment epithelium cells, arising from intense renewal of the photo-sensory membrane discs, caused by photo-stimulation. Rods in *O. amberensis* are totally exposed even under illumination. Therefore, they seem to not have the adaptation of melanin migration that would protect them from incident light (Donatti and Fanta, 1999), as this is not useful in the depths where they usually live. Thus, it is clear that this species is better adapted to low light than to well illuminated environments.

The feeding behaviour of both *O. amberensis* and *Gobionotothen gibberifrons* has similarities, as have some aspects of their retinae. In comparison with *Notothenia coriiceps*, for example, *G. gibberifrons* and *O. amberensis* seem to have poor vision. Failures in detection, identification and successful capture of prey are frequent. The photosensorial cells in the retina of both species are less varied and more slender when compared to other Nototheniidae that have up to five different types of large cones (Fanta *et al.*, 1994; Grötzner and Fanta, 1998), and rods. Possibly, this causes the visual reaction of *G. gibberifrons* and *O. amberensis* to the presence of prey, to be very slow (Nicol, 1963) in comparison to other Nototheniidae. Possibly the stimulation by movement alone, as seems to be the case for *O. amberensis* in low light conditions, is a cause for delay in behavioural reactions. In contrast, a quick behavioural response is obtained after a visual stimulation that includes a sum of movement, shape and colour, in good light conditions, and that is perceived by fish that have both rods and different types of cones. On the other hand, the neuronal layer of *O. amberensis* is relatively thin in comparison to that of *Notothenia coriiceps*, *Trematomus newnesi* and *Lepidonotothen nudifrons* (Grötzner and Fanta, 1998), but similar to that of *Gobionotothen gibberifrons*. A thin neuronal layer is considered by Ingle (1971) to indicate a higher degree of evolution.

The significance of olfaction in food detection was established experimentally for many freshwater and marine species that were able to recognise their food by smell alone (Hara, 1986). Direct swimming along a chemical gradient demonstrated the importance of chemical stimuli for the perception of prey by *O. amberensis*. This may be due to

olfactory, external gustatory, and common chemical sensors, acting individually or, more probably, in concert. It is, however, difficult to distinguish between the responses of these different senses in the aquatic environment (Gerking, 1994; Valenticic and Caprio, 1994). The chemical sensory capacity is of fundamental importance in the environment where *O. amberensis* may live, because it is known that chemical signals can be perceived at a greater range than visual signals (Marshall, 1971). This enables the fish to find food in all seasons of the year and at different depths. Studies on Antarctic fish that live in more shallow water, such as *T. newnesi* (Meyer and Fanta, 1998; Fanta *et al.*, 1999), *N. coriiceps*, *N. rossii*, *Chaenocephalus aceratus* and *Parachaenychthys charcotti* (in preparation) have indicated that either the chemical or the visual sense alone was able to initiate feeding activity. Therefore they can use either one or the other, depending on the environmental conditions.

The nostrils of *O. amberensis* have one pore, connected to an elongated olfactory chamber. The olfactory rosette is relatively big and pear shaped, but has only three thick olfactory lamellae. It is different from the flat oval rosette described for *T. newnesi* (Meyer and Fanta, 1998), that has 26 thin lamellae. The number of lamellae is not necessarily related to the olfactory acuity (Eastman, 1993). According to the rosette shape classification of Hara (1971), *T. newnesi* is considered to have intermediate olfactory acuity. However, the pear shaped thick olfactory rosette of *O. amberensis* seems to be related to high olfactory perception. Even showing a connection with the anterior and posterior pores, there is no clear evidence that both have the function of exhalant pores. The chemical stimulation of food at a distance can also be done by the external taste buds that *O. amberensis* has at the head. As they present varied shape and size, one can assume that they are able to perceive different types of chemical nature of dissolved particles. The combination of poor vision and a well-developed chemosensory system is also present in *G. gibberifrons* (Fanta *et al.*, 1994).

Food selection by taste, inside the oro-pharyngeal cavity, gives the fish the chance to accept suitable food or reject unwanted particles of feed. Rejection occurs when the captured particles or organisms come into contact with the lips and/or the inside of the mouth, a phenomenon also reported in some Nototheniidae (Fanta and Meyer, 1998; Fanta, 1999). This demonstrates not only that fish sometimes misinterpret the chemical or visual stimuli, but also that they are able to make choices regarding the preference or acceptability of different types of food.

In conclusion, the chemosensory system was more important for food detection and selection than vision. Accordingly, a large olfactory epithelium and several different taste buds at the head, the lips, and the mouth cavity, are considered to be responsible for the reception of different types of chemical stimuli. Vision alone was weak in eliciting feeding behaviour. The retina is simple, showing a high concentration of long, active rods, but no cones. In combination, these features demonstrate the degree of adaptation of *O. amberensis*, not only to deeper habitats, but also to the Antarctic winter. The presence of light seems to not bring any advantage in the foraging of *O. amberensis*.

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### References

- Anderson, M.E. (1984): Zoarcidae: development and relationships. *Ontogeny and Systematics of Fishes*, ed. by H.G. Moser. Lawrence, Allen Press, 578-582.
- Anderson, M.E. (1988): Studies on the Zoarcidae (Teleostei: Perciformes) of the Southern Hemisphere. I. Antarctic and Subantarctic regions. *Biology of the Antarctic Seas*. Washington, Am. Geophys. Union, 59-113.
- Anderson, M.E. (1990): Zoarcidae. *Fishes of the Southern Ocean*, ed. by O. Gon and P.C. Heemstra. Grahamstown, Smith Inst. Ichthyol., 256-276.
- Andriashev, A.P. (1987): Review of the snailfish genus *Paraliparis* (Scorpaeniformes: Liparidae) of the Southern Ocean. *Theses Zool.*, **7**, 204.
- Clark, G. (1981): *Staining Procedures*. Baltimore, Williams & Wilkins, 512 p.
- Culling, C.F.A., Allison, R.T. and Barr, W.T. (1985): *Cellular Pathology Technique*. London, Butterworth, 642 p.
- Daniels, R.A. (1982): Feeding ecology of some fishes of the Antarctic Peninsula. *Fish. Bull.*, **80**, 575-588.
- Donnatti, L. and Fanta, E. (1999): Morphology of the retina in the freshwater fish *Metynnis roosevelti* Eigenmann (Characidae, Serrasalminae) and the effects of monochromatic light. *Rev. Bras. Zool.*, **16**, 151-173.
- Eastman, J.T. (1993): *Antarctic Fish Biology: Evolution in a Unique Environment*. New York, Academic Press, 322 p.
- Fanta, E. (1999): Laboratory test on feeding interactions of some Antarctic fish from Admiralty Bay (King George Island, South Shetlands). *Pol. Polar Res.*, **20**, 17-28.
- Fanta, E. and Meyer, A.A. (1998): Behavioural strategies for feeding of six species of the Antarctic fish family Nototheniidae (Pisces, Notothenioidei) in a tank. *Nankyoku Shiryô (Antarct. Rec.)*, **42**, 227-243.
- Fanta, E., Donnatti, L. and Freiburger, S. (1999): Visual sufficiency in food detection and initiation of feeding behaviour in the Antarctic fish *Trematomus newnesi* Boulenger, 1902. *Nankyoku Shiryô (Antarct. Rec.)*, **43**, 221-236.
- Fanta, E., Meyer, A.A., Grötzner, S.R. and Luvizotto, M.F. (1994): Comparative study on feeding strategy and activity patterns of two Antarctic fish *Trematomus newnesi* Boulenger, 1902 and *Gobionotothen gibberifrons* (Lönnberg, 1905) (Pisces, Nototheniidae) under different light conditions. *Nankyoku Shiryô (Antarct. Rec.)*, **38**, 13-29.
- Fisher, W. and Hureau, J.C., ed. (1985): *FAO Species Identification Sheets for Fishery Purposes. Southern Ocean. Vol. II*. Rome, Food and Agriculture Organisation of the United Nations,

471 p.

- Gerking, S.D. (1994): Feeding Ecology of Fish. New York, Academic Press.
- Grötzner, S.R. and Fanta, E. (1998): Comparative morphology of the retina of the Antarctic fish *Gobionotothen gibberifrons* (Lönnberg, 1905), *Trematomus newnesi* Boulenger, 1902, *Lepidonotothen nudifrons* (Lönnberg, 1905) and *Notothenia neglecta* Nybelin, 1951. *Pesq. Antarct. Bras.*, **3**, 31-47.
- Hara, T.J. (1986): Role of olfaction in fish behaviour. *The Behaviour of Teleost Fishes*, ed. by T.J. Pitcher. London, Croom Helm, 152-176.
- Hara, T.J. (1971): Chemoreception. *Fish Physiology*, Vol. 5, ed. by W.S. Hoar and D.J. Randall. New York, Acad. Press, 79-120.
- Ingle, D. (1971): Vision: the experimental analysis of visual behaviour. *Fish Physiology*, Vol. 5, ed. by W.S. Hoar and D.J. Randall. New York, Acad. Press, 59-67.
- Løkkeborg, A., Olla, B.L., Pearson, W.H. and Davis, M.V. (1995): Behavioural responses of sablefish *Anoplopoma fimbria* to bait odour. *J. Fish Biol.*, **46**, 142-155.
- Luft, J.H. (1961): Improvements in epoxy resin embedding methods. *J. Biophys. Biochem. Cytol.*, **9**, 409-414.
- Marshall, N.B. (1971): *Explorations in the Life of Fishes*. Cambridge, Harvard Univ. Press, 204 p.
- Meyer, A.A. and Fanta, E. (1998): Morphofunctional study of chemo sensorial structures of the Antarctic fish *Trematomus newnesi* Boulenger, 1902 used for food detection and selection. *Pesq. Antarct. Bras.*, **3**, 49-63.
- Montgomery, J.C. and Coombs, S. (1992): Function and evolution of superficial neuromasts in an Antarctic Notothenioid Fish. *Brain Behav. Evol.*, **44**, 287-298.
- Montgomery, J.C. and Macdonald, J.A. (1987): Sensory tuning of lateral line receptors in Antarctic notothenioid fishes: metabolic capacity and sensory performance. *Fish Ecophysiology*, ed. by J.C. Rankin and F.B. Jensen. London, Chapman & Hall, 341-374.
- Munz, F.W. (1971): Vision: visual pigments. *Fish Physiology*, Vol. 5, ed. by W.S. Hoar and D.J. Randall. New York, Academic Press, 1-32.
- Nicol, J.A.C. (1963): Some aspects of photoreception and vision in fishes. *Adv. Mar. Biol.*, **1**, 171-208.
- Rakusa-Suszczewski, S. (1993): Introduction. *The marine Antarctic Coastal Ecosystem of Admiralty Bay*, ed. by S. Rakusa-Suszczewski. Warsaw, Pol. Acad. Sci., 7-14.
- Rios, F.S. and Fanta, E. (1998): Morphology of gill rakers and their ecological functions in feeding of the Antarctic Fish *Notothenia neglecta* Nybelin. *Nankyoku Shiryô (Antarct. Rec.)*, **42**, 131-150.
- Valenticic, T.B. and Caprio, J. (1994): Chemical and visual control of feeding and escape behaviours in the channel catfish *Ictalurus punctatus*. *Physiol. Behav.*, **55**, 845-855.

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