

# Comparison of the oral cavity architecture in surgeonfishes (Acanthuridae, Teleostei), with emphasis on the taste buds and jaw “retention plates”

Lev Fishelson · Yakov Delarea

Received: 4 September 2012 / Accepted: 25 March 2013  
© Springer Science+Business Media Dordrecht 2013

**Abstract** The present study summarizes observations on the skin plates (“retention plates”) and taste buds (TBs) in the oropharyngeal cavity (OC) of 15 species of surgeonfishes (Acanthuridae), all of which are predominantly herbivorous. Two phenomena mark the OC of these fishes: the presence of skin-plates rich in collagen bundles at the apex of the jaws, and cornified papillae on the surface. It is suggested that these plates help in retaining the sections of algae perforated at their base by the fishes’ denticulate teeth. The TBs, especially type I, are distributed across the buccal valves, palate and floor of the OC, forming species-specific groupings along ridges established by the network of sensory nerves. The number of TBs in the OC increases with growth of the fish up to a certain standard length, especially at the posterior part of the OC, and differs among the various species: e.g., *Zebbrasoma veliferum* possesses 1420 TBs and *Parcanthurus hepatus* 3410. Species of *Naso* show a higher number of TBs than most species of *Acanthurus*, possibly connected with their more diversified diet. The pharyngeal region of these fishes is expanded through lowering the base of the gill-arches, and together with the occurrence of high

numbers of TBs in this region, indicates the importance of the posterior region of the OC in herbivorous fishes for identification of the engulfed food particles prior to swallowing. The discussed observations shed light on the micro-evolutionary developments of the OC within the family Acanthuridae and contribute to the taxonomic characterization of the various species.

**Keywords** Surgeonfishes · Oral cavity comparison · Herbivore · Retention plate · Taste buds

## Introduction

The primary organs for food chemoreception in fish are the taste buds (TBs), formed by modified epithelial cells found within the oral cavity and on the barbels around the oral opening. Each TB comprises a group of sensory cells with exposed microvilli rising above the surrounding epithelium (Kinnamon 1987; Fishelson et al. 2004, 2010 and citations therein; Fishelson 2005a, b). From these sensory centers the facial (VII), glossopharyngeal (IX) and vagus (X) sensory nerves transfer the received signals to the relevant brain centers (Finger et al. 2000; Doty 2003; Hansen and Reutter 2004; Northcutt 2004, 2005; Reutter and Hansen 2005). TB cytology has been studied in various families of fishes (Tagliafierro and Zaccone 2001; Fishelson et al. 2004, 2010; Northcutt 2005, and citations therein). The works by Hara (2006),

---

L. Fishelson (✉) · Y. Delarea  
Department of Zoology and Electron Microscopy Unit,  
George S. Wise Faculty of Life Sciences,  
Tel Aviv University,  
Tel Aviv, Israel  
e-mail: fishelv@post.tau.ac.il

Qgawa and Caprio (2010), and Kasumyan (2011) (and citations therein), summarize most of our knowledge on TB chemistry and the transfer of information by these organs; while the study of Sakata et al. (2001) illustrates the structure of the nervous network in the barbels of the catfish, *Plotosus lineatus*. Recent studies have revealed that the number and distribution of TBs in the oral cavity differ among the various species, even among species belonging to the same genus (Fishelson et al. 2004; Gon et al. 2010), frequently irrespective of the body-length of the fish.

To date almost all studies of fish TBs have focused on carnivorous species, Apogonidae and Synodontidae, with a few dealing with omnivorous ones, such as the blennies (Fishelson and Delarea 2004). It was thus of interest to study these structures in herbivorous fish, and for which we selected species of the surgeonfish, family Acanthuridae (Table 1), a group of fishes with a 50 myl year presence. The importance of surgeonfishes as reef dwellers, especially as species partly feeding on algae growing on the reef surfaces was already recognized in the classical studies of Hiatt and Strasburg (1960), Hobson (1974), and Sano et al. (1984). According to Winterbottom and McLennan (1993), this family is of Indo-Pacific origin. Studies on their phylogeny are controversial: some, e.g., Tyler et al. (1989) and Guisasu and Winterbottom (1993), recognize *Acanthurus* spp. and related genera as a monophyletic clade according to their synapomorphic characters; whereas Clements et al. (2003), based on their genetic studies, reject this hypothesis, arguing for several periods of rapid speciation. During the last two decades additional papers have been published on the evolution and biogeography of surgeonfishes (Tang et al. 1999), as well as on their growth (Choat and Axe 1996). The feeding of these fish on reef surfaces often strongly impact the living cover of these hard structures (Wainwright and Bellwood 2002; Choat et al. 2004; Schuhmacher et al. 2008).

The majority of surgeonfish are herbivorous, feeding on algae growing in the shallow and rocky environments, and partly on shallow-water detritus of tropical and subtropical seas (Fishelson 1977; Fishelson et al. 1987; Montgomery et al. 1989). Species of this family have undergone ecomorphological adaptations to various diets, such as *Acanthurus* spp. (sub-family Acanthurinae), that generally are grazers and partly detritivorous; and several species of *Naso* (sub-family Nasinae), which feed as sub-adults on algae and as adults on zooplankton. As demonstrated by Robertson

and Gaines (1986), Fishelson et al. (1987), and Tilghman et al. (2001), some species demonstrate selective algal diets at different ages and distribution. Three types of feeding behavior have been observed in these fishes: species such as *Ctenochaetus striatus*, *Zebrasoma veliferum* and *Z. xanthurum* forage along the reefs singly, in pairs, or in small groups, without connection to specific sites; while species such as *Acanthurus nigrofasciatus* demonstrate cooperative behavior, migrating to feed in larger groups at specific sites, and returning from these to their night quarters (Robertson 1983; Fishelson et al. 1987); and species like *A. sohal* occupy permanent territorial rocky surfaces in groups, protecting their algae lawns from invasion by other algivorous fish. *A. nigrofasciatus* in the Gulf of Aqaba alter their diet during the year: in summer they feed on micro-algae, such as *Polysiphonia*, *Ceramium*, whereas from November the diet consists of fleshy and larger green-algae, such as *Ulva*, *Enteromorpha* and *Cladophora*. This switch is concomitant with their recrudescence of gonads (Fishelson et al. 1987).

The life-history and behavior of surgeonfishes has been studied by several authors (Randall 1967; Barlow 1974; Robertson 1983; Myrberg et al. 1985; Fishelson et al. 1987). Some of the species, such as *A. nigrofasciatus* and *A. sohal*, inhabiting coral reefs, were identified as the primary grazers, abrading the rocky surface with their teeth during feeding (Robertson and Gaines 1986; Montgomery et al. 1989; Choat et al. 2004). The study by Purcell and Bellwood (1993) on the oral cavity skeleton of *C. striatus* and *A. nigrofasciatus* demonstrated that during feeding, when the mouth opens and the dentale descends, an upward motion of the premaxilla is induced, and both jaws, gliding on the angular joint, reveal the teeth and inside of the jaws. In the studied species of surgeonfishes the mobile parts of the jaws occupy ca. 10 % in length of the OC, with the lower jaw being narrower than the upper one, so that the teeth of the premaxilla partly cover the teeth of the dentale.

The present study is the first to focus on the special “retention plate” structure and the taste buds (TBs) in the oral cavity (OC), of 15 species of surgeonfishes belonging to the sub-family Nasinae and several tribes of Acanthurinae (Table 1), and to compare them with the closely-related *Siganus luridus* and *S. rivulatus* belonging to the Siganidae. Comments on the teeth and pharyngeal bones are also appended.

**Table 1** Species studied and collection sites

	Species	Collection site	No.	SL (mm)
	<i>Acanthurus guttatus</i>	Hawaii	1	210
	<i>Acanthurus achilles</i>	Hawaii	1	136
	<i>Acanthurus nigricans</i>	DA	3	150–210
	<i>Acanthurus nigrofuscus</i>	GA, Hawaii	16	12–150
	<i>Acanthurus sohal</i>	GA, SR	6	20–230
	<i>Acanthurus triostegus</i>	Hawaii	1	168
	<i>Zebrasoma veliferu m</i>	GA, DA	6	18–200
	<i>Zebrasoma xanthurum</i>	GA	3	120–130
	<i>Paracanthurus hepatus</i>	Philippines	1	200
	<i>Prionurus scalprum</i>	Shanghai	1	130
	<i>Ctenochaetus striatus</i>	GA	5	115–130
	<i>Naso unicornis</i>	RS, Philippines	3	120–140
GA Gulf of Aqaba, DA Dahlak Archipelago, Southern red Sea, RS Red Sea, SR Southern Red Sea	<i>Naso brevirostris</i>	Hawaii	1	235
	<i>Naso hexacanthus</i>	Hawaii	1	146
	<i>Naso lituratus</i>	GA, Hawaii	3	170–200

## Materials and methods

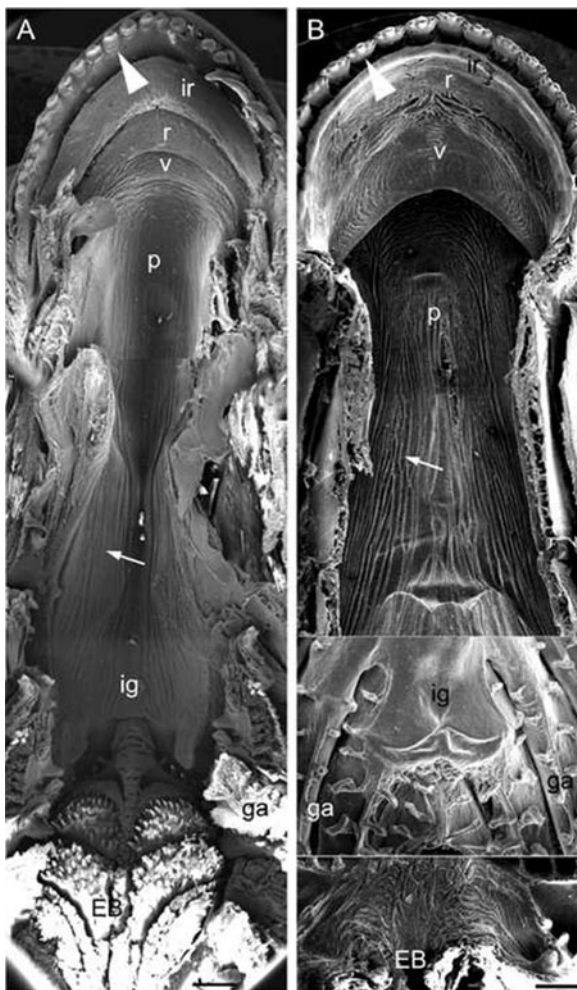
The studied species of surgeonfish were sampled from various sites (Table 1) and donated for the present study by the fish collection of the Smithsonian Natural History Museum, Washington, USA; the Bernice Bishop Museum of Hawaii, the Hebrew University, Jerusalem; and the ichthyological collection of the National Natural History Museum, Tel Aviv University. In most instances the fish had been fixed in formalin and preserved in ethanol. For scanning electron microscopy (SEM) the open dissected heads of the separated jaws of the fish were passed along ascending grades of ethanol, saturated with CO<sub>2</sub>, critical point-dried in a Balzer Union Point Dryer (BU-11120, Balzer, Lichtenstein), sputter-coated with gold using a Polaron E5100 Sputter Coater, and observed with a JSM840A scanning microscope. Some samples were air-dried following immersion in absolute ethanol. Larger jaws were photographed with a digital Nikon camera. For transmission electron microscopy (TEM), the samples were immersed for 24 h in glutaraldehyde 3.5 % (pH 7.2), post-fixed in osmium tetroxide, embedded in Epon, and their micro-sections were stained with uranyl acetate and lead citrate. These sections were studied with a JEOL 1200 EX TEM, and photographed with an attached digital camera. To count the TB numbers micrographs were produced (20×20 cm) and the counted organs were marked with ink dots.

## Results

### The jaws and oral cavity

With the shortening of the jaws in the surgeonfishes, the base of the gill-arches moved forward and their lower posterior part, formed by the gill-ossicles, flattens. This morphological modification enlarged posteriorly the flat regions of the OC, the dorsal palate, and the ventral floor (Fig. 1a, b). This consequently increased the volume of the OC, and provides both a larger space for accumulation of the algal morsels prior to engulfing them, and larger sites for TB development (see below). The surface membranes of the OC epithelial cells are provided with the fingerprint pattern of mazes (Fig. 2a) typical for fish ectoderm (Fishelson 1984). The protruding hillocks of TBs between these cells cause irregularity in these patterns, especially at sites with dense populations of TBs organs (Figs. 2b and 5e). Of all the studied species, only *A. nigrofuscus*, *A. nigricans* and *Ctenochaetus striatus* displayed a black melanized buccal valve, and only *A. nigricans* and *C. striatus* revealed a population of melanocytes dispersed across the tongue epithelium of the OC; in all other species the cavity was pale, not pigmented.

With the exception of *Naso* spp, the teeth of which are pointed and bear only small rudiments of sera (Fig. 2c), the dentale and premaxilla of all the other



**Fig. 1** Upper jaw (internal). **a** *Naso unicornis*, bar 2 mm; **b** *Acanthurus sohal*, bar 1.8 mm. EB, epipharyngeal bones; ga, gill arches; ig, inter-gill region; ir, inflated edge of the retraction plate; p, palate; v, buccal valve; r, retention plate;; arrowhead, teeth; arrow, ridges

studied surgeonfishes feature a row of serrated teeth, each with a long shaft deeply embedded within the soft tissue of the jaws. The exposed, serrated, blade-like part of the tooth differs among the various species in form and number of lateral denticles. For example, in the genus *Acanthurus* they form a uniform line, each tooth denticulate laterally along both edges (Fig. 2d); whereas in *Ctenochaetus* the blade bears denticles only along one edge, with all the teeth on both jaws leaning with their smooth edge toward the midline of the jaw (Fig. 2e, f). The denticulation also differs among the various species of *Acanthurus*. Thus, in *A. nigrofuscus* each tooth bears one apical

denticle and four others on each side (Fig. 2g); its denticulated part is 300–400  $\mu\text{m}$  long and each denticle is 100–120  $\mu\text{m}$ , increasing in size from ventral to apical. In *A. sohal* the teeth are more robust and larger (Fig. 2h); the blade is ca. 1 mm long and over 1 mm wide, with thick denticles, 3–4 on each side, all of approximately similar length. The teeth of *Zebrasoma* spp. are similar to those of *A. sohal*. The most robust teeth are those of *A. achileus* (Fig. 3a). The lips in all species are narrow and devoid of TBs.

#### The buccal (breathing) valve region

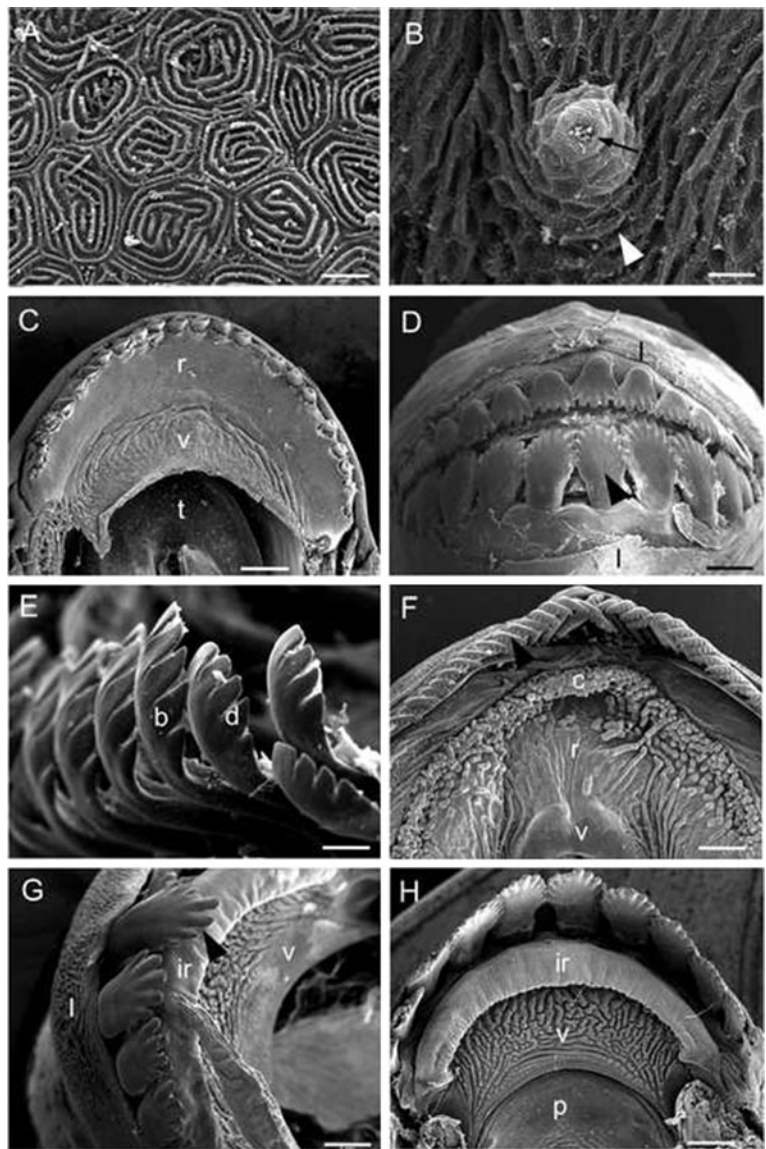
Posterior to the teeth the jaws display a complex and unique organization of two protruding skin-folds. The anteriorely directed one is termed here a “retention plate”, while the posteriorly-directed one forms the buccal (breathing) valve (Fig. 1a, b), a structure common to most fishes, and functioning, together with the operculum, to regulate water-flow across the gills.

#### The “retention plates”

These plates constitute a flat and relatively thick skin-extension inside both jaws, attached posteriorly along the junction line with the buccal valve. Their anterior edges are free, extending along the teeth. This free edge of these plates differ between the upper and lower jaws: along the free edge of the upper jaw, in all species the plate is round-inflated, prominent in *Acanthurus* spp. (Fig. 3b), and especially inflated in *Zebrasoma veliferum* (Fig. 3c), covered by densely packed papillae. The plate of the lower jaw is less thick than that of the upper jaw and in most species not inflated (Fig. 3d), covered with papillae. The first signs of development of the retention plates are visible in 12 mm SL *A. nigrofuscus* and *Z. veliferum*, and in 16 mm SL *A. sohal*. Outgrowths of the skin are covered by a multilayer epithelium with strongly interdigitating membranes, held together by long junctions (Fig. 3e and inset). Starting from the basal membrane a continuum across the epithelium, these cells produce an amorphous substance (cornification?), and their nuclei gradually disintegrate (Fig. 3f). From the line of attachment at the buccal valve base the surface cell-membranes of the plates gradually lose their micro-ridge mazes (Fig. 3g) which become replaced by cornified papillae (Fig. 3h). Below the basal membrane this epithelium is



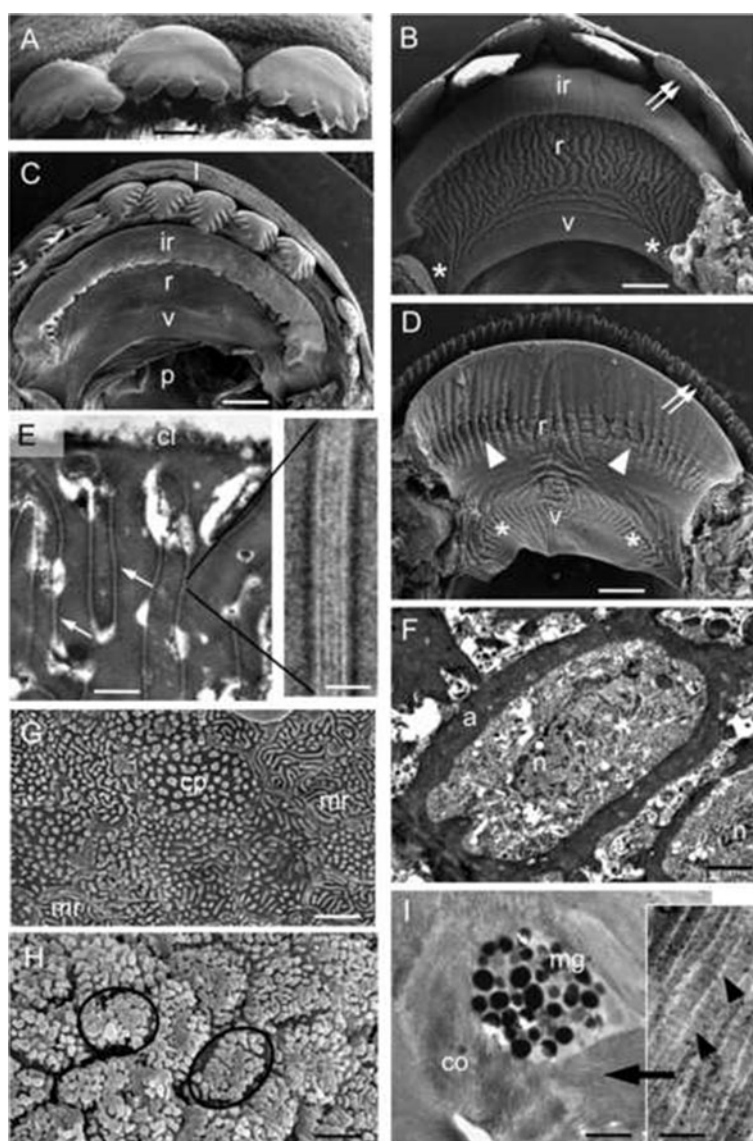
**Fig. 2** Comparison of oral parts of surgeonfishes. **a** Ridge mazes on epithelial cells of *Acanthurus nigrofuscus*, bar 12  $\mu$ m; **b** Bending of epithelial ridges induced by TB papilla in *A. nigrofuscus*, bar 10  $\mu$ m; **c** Lower jaw of *Naso hexacanthus*, bar 0.8 mm; **d** Front of *A. nigrofuscus* head, bar 1 mm; **e** Blades of teeth of *Ctenochaetus striatus*, bar 80  $\mu$ m; **f** Apex of upper jaw of *C. striatus*, bar 1 mm; **g** Upper jaw of *A. nigrofuscus*, bar 0.5 mm; **h** Upper jaw of *A. sohal*, bar 1 mm. c, cornified papillae; b, blades of teeth; d, denticles; ir, inflated free edge of retention plate; l, lip; p, palate; t, tongue; arrow, sensory microvilli of TB; white arrowhead, bending ridges on anterior part of palate; black arrowhead, teeth; star, ridges on the buccal valve



strengthened by dense bundles of collagen fibers (Fig. 3i and inset), frequently with lines of melanophores and melanin granules (Fig. 3i). In the studied species the plates are relatively thick compared with the buccal valve, and their surface is crossed by longitudinal ridges that break down to merge with the buccal valve (Fig. 4a). The number of ridges increases with growth of the fish: for example, *A. nigricans* of 115 mm SL possess 25 ridges and of 200 mm SL possess 36 ridges. Close to the buccal valve the ridges bear type III TBs submerged in the surrounding epithelium (Fig. 4b).

The form and morphology of the cornified papillae is especially prominent in *Ctenochaetus striatus* (Fig. 2f).

In *Naso* spp. the lower plate is smooth, with only partly cornified epithelial membranes (Fig. 4c). On both jaws the free anterior edges of the plates extend to only slightly lower than the teeth and in the closed mouth the upper and lower plates are closely juxtaposed, forming a retention organ behind the jaws. The dimensions of these plates, as well as of the buccal valves, increases with growth of the fish, relative to the width of the jaws, being larger on the upper jaw than on the lower. For example, in 40 mm SL *A. sohal* the retention plate is ca. 2 mm wide and 1.8 mm long, while in fish of 230 mm SL it is 14 mm wide and 4.5 mm in length on the upper jaw and 12×3 mm on lower one.



**Fig. 3** Structure and surface of the oral retention plates. **a** Teeth of *Acanthurus achilles*, bar 0.5 mm; **b** Apical part of upper jaw of *A. nigricans*, bar 0.3 mm; **c** Apical part of upper jaw of *Zebrasoma veliferum*, bar 1 mm; **d** Apical part of lower jaw of *A. nigrofuscus*, bar 1.2 mm; **e** Interdigitation of epithelial cells of the retention plate in *Naso lituratus*, bar 0.5 mm; inset, junctions between neighboring cells, bar 70  $\mu$ m; **f** Surface-close epithelial cells of the retention plate of *A. nigricans*, bar 2  $\mu$ m; **g** Splitting of ridge mazes on cells of *A. sohal*, bar 8  $\mu$ m; **h** Cornified papillae on cells of *Ctenochaetus striatus*, bar 8  $\mu$ m; **i** Collagen

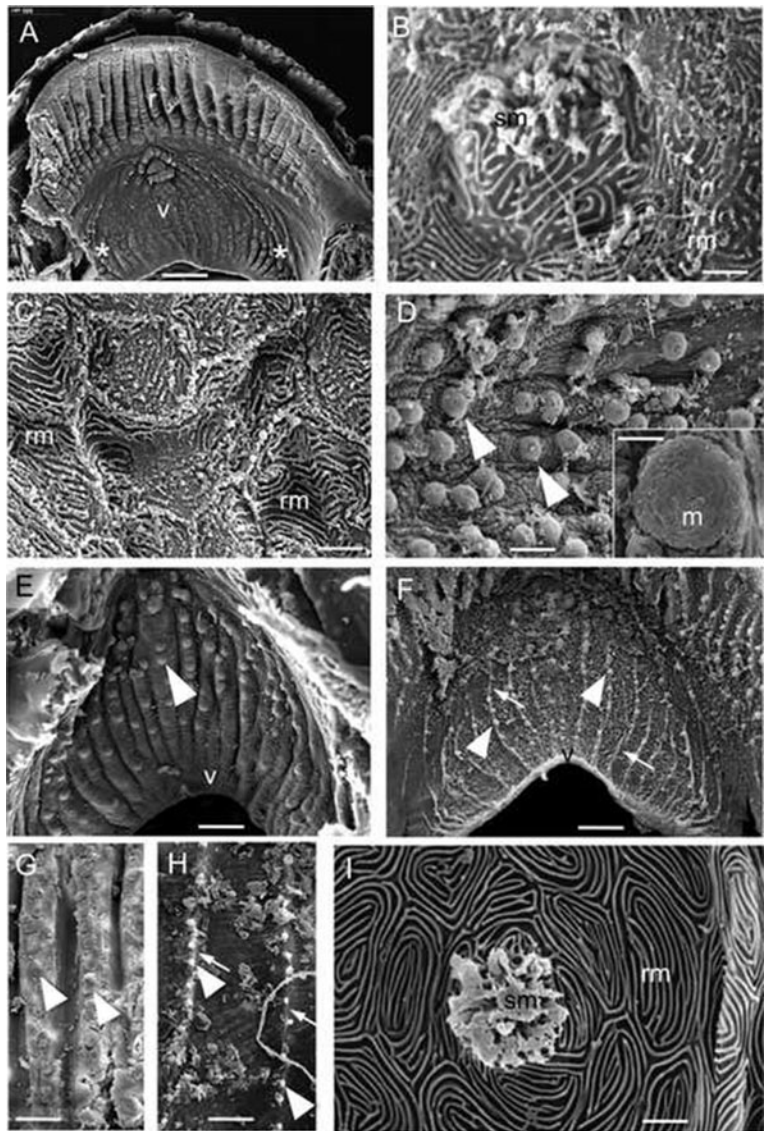
bundles in the retention plate of *A. nigricans*, bar 5  $\mu$ m; inset, collagen fibers of the same, bar 250 nm. a, amorphous material; cl, cornified outer layer of retention plate; co, collagen fibers; cp, cornified papillae; mg, melanin granules; mr, rudiments of micro-ridges; n, nucleus; ir, inflated free edge of retention plate; r, retention plate; v, buccal valve; ring, cornification on single cells; arrow, interdigitation; double arrow, teeth; arrowhead, cross-striation of collagen fibers; white arrowhead, splitting of ridges; star, ridges on the buccal valve

In *C. striatus* of 160 mm SL the plate is 16 $\times$ 8 mm on the upper jaw and 9 $\times$ 3 mm on the lower one; the anterior part of the plate is densely covered by conical cornified papillae.

In *A. sohal* the retention plate bears 16–18 thick ridges, while in *Z. xanthurum* the number is much higher, and the ridges are very narrow and dense. Lines of melanocytes extend between the ridges in *A.*



**Fig. 4** Oral retention plates and TBs in OC of surgeonfishes. **a** Splitting of ridges on retention plate of *Naso brevirostris* as they merge with the buccal valve bar 0.8 mm; **b** Type III TB on the junction between plate and buccal valve of *Acanthurus nigricans*, bar 1  $\mu$ m; **c** Partial mazes and cornifications on the retention plate of *N. brevirostris*, bar 6  $\mu$ m; **d** Lines of TB-papillae on the buccal valve of juvenile *A. nigrofuscus*, bar 40  $\mu$ m; inset, enlarged papilla, bar 10  $\mu$ m; **e** Buccal valve of *A. sohal* with lines of TBs papillae, bar 80  $\mu$ m; **f** Buccal valve of *A. sohal* after surface decomposition, bar 1 mm; **g** Ridges with TBs on tongue of *Zebrasoma flavicauda*, bar 200  $\mu$ m; **h** Ridges on tongue of *Z. flavicauda* following surface decomposition, bar 1 mm; **i** Type II TB on palate of *A. nigricans*, bar 2  $\mu$ m. r, retention plate with ridges; rm, micro-ridge mazes; sm, sensory villi of TB; v, buccal valve; white arrowheads, TBs; white arrow, nerves connecting the TBs; star, ridges on the valve



*nigricans*. The form of these ridges seems to be species-specific, possibly connected with the type of diet of the fish (see [Discussion](#)).

#### TBs on the buccal valves

The buccal (breathing) valves appear early on during morphogenesis, formed by skin extension, and revealed in post-larvae of 10–12 mm SL. In *A. nigrofuscus* at this stage both valve and plate of the lower jaw are melanized. In 14 mm SL fishes the valve already bears numerous type I TBs, with papillae 30  $\mu$ m high and 20  $\mu$ m in diameter (Fig. 4d). Their number and organization on the

valve progresses from this stage on, particularly with formation of the surface ridges.

In *A. nigrofuscus* and *A. sohal* of 18–22 mm SL, the buccal valves are covered by ridges. Dissolving the epithelial cover here with trypsin (or simply washing down with a jet of water over the valve or the entire surface of the OC), reveals each line of TBs to be situated along connecting nerve branches of the innervating central nerves (Fig. 4e, f), as also reflected along the tongue (Fig. 4g, h). Like the retention plates, the breathing valves too enlarge with growth of the fish, attaining 8×4 mm in *Z. veliferum* of 180 mm SL and 14×5 mm in 230 mm SL *A. sohal*. With growth the valve surface becomes more ridged, and in larger *S.*

*sohal* 32–34 longitudinal ridges of epithelium cross the valve. In some species, e.g. *A. nigrofuscus* and *A. sohal*, the posterior part of this valve is smooth. In adult fish the number of ridges and lines or groups of TBs along them differs in the various species and often in different-size specimens of different size of the same species (Table 2). Comparing the number of TBs with the SL of the fish revealed that the major development of the TB network takes place up to 150–180 SL of the fish, after which and despite the growth of the fish, only very minor changes occur in TB number.

In *A. nigrofuscus* the TBs of the valve are organized in groups of 5–6 on mini-islets separated by a swollen edge from the surrounding epithelium (Fig. 5a). Most of them are of type I, situated on hillocks 60 µm high and ca. 10–20 µm wide; their exposed sensory microvilli form tufts 5–6 µm in diameter (Figs. 4i and 5c). Most TBs on the lower jaw are of type I, with some type II that are only slightly raised above the surrounding skin (Fig. 5b).

#### TBs on tongue and palate

The wide flat tongue covers the ventral site of the OC, extending from below the breathing valve, caudally joining the flat base between the gill-arches (Fig. 1a, b). Covered by the fingerprint mazes of the epithelial cells, the tongue and palate reveal delicate ridges that at the anterior end cross the site perpendicular to the body axis (Fig. 5d), extending longitudinally between the gills (Fig. 5e, f). The number of these ridges increases with growth of the fish: for example, in 30 mm SL *Ctenochaetus striatus* the palate bears 18 ridges and 24

ridges on the tongue and the floor of the OC, respectively; in 20 mm SL *Acanthurus nigrofuscus* there are 14 ridges on the palate and 18 on the floor. In contrast, in adult fishes of most of the studied species 30–40 ridges extend along both the upper and lower regions of the OC. These ridges bear type I TBs along their entire length; in juvenile *Acanthurus* there are 40–50 on each ridge and in adults there are 60–70. In juvenile *Acanthurus* the TB papillae occur individually, while in adults they form bundles, 2–4 in *Acanthurus* spp and 5–7 in *C. striatus* (Fig. 5g and inset). Just as the number of folds differs among the various species, so too do the number of TBs on the palate and floor of the OC (Table 2). At the base of the gill-arches, the ridges are less prominent, but feature numerous types II and III TBs. For example, in *A. sohal* there are 200 TBs while *Paracanthurus hepatus* possesses ca. 4,000 (Fig. 6a, b).

#### Epipharyngeal and hypopharyngeal bones

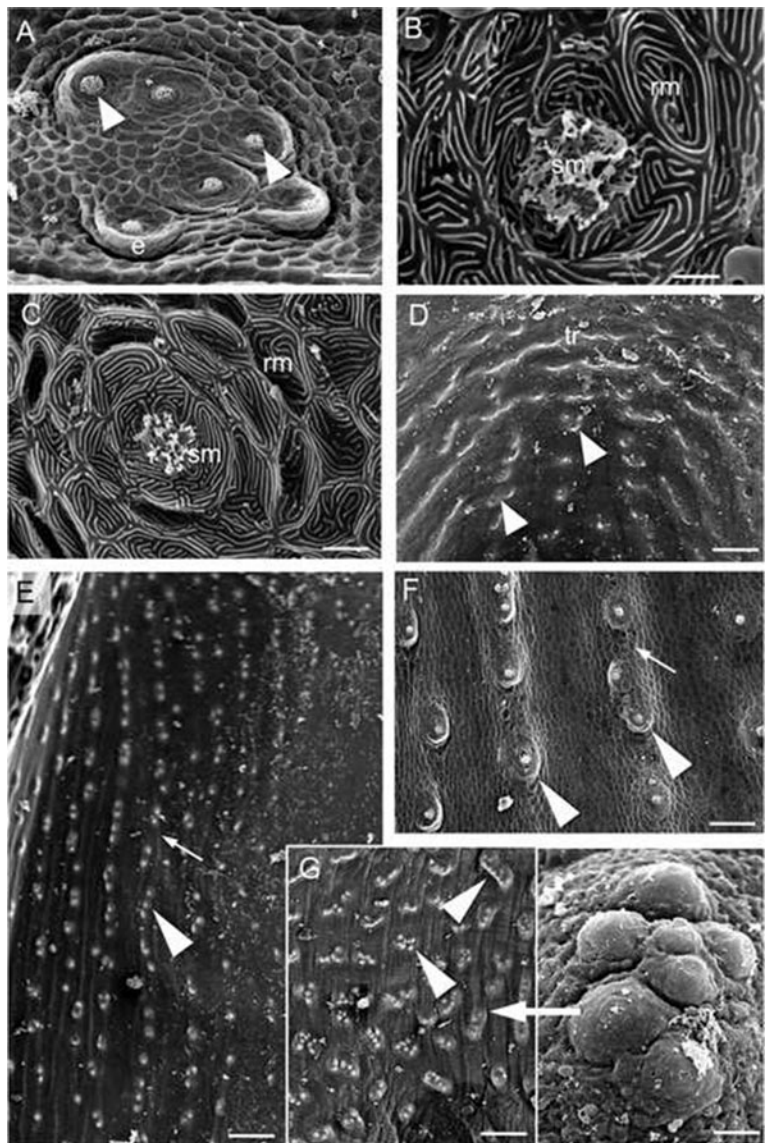
The epipharyngeal bones (EB), and hypopharyngeal bones (HB), namely the 2–4 pharyngobranchials and the 5th ceratobranchial, form the pharyngeal jaw apparatus situated at the entrance to the esophagus. These bones, especially the EB differ in form in the various species (Fig. 6c, d). The HB display transverse lines of sharp teeth, but without TBs between them (Fig. 6e). In the EB rows of round large papillae extend between the ossicles, each bearing 1–2 type III TBs. Of special interest is the observed morphology of the secondary gill-lamellae of the 4th and 5th ceratobranchials. Namely, in *Acanthurus* spp. and *Ctenochaetus* the inner row of

**Table 2** Number of TBs on different sites of the oral cavity in the largest fish studied of the various species

Species	Standard length (mm)	Upper valve	Palate	Lower valve	Tongue	Inter-Gill site	Total
<i>Acanthurus achilles</i>	136	210	750	180	580	260	1770
<i>A. nigricans</i>	210	300	1120	460	700	600	3180
<i>A. nigrofuscus</i>	150	160	1000	120	600	600	2480
<i>A. sohal</i>	230	360	960	500	800	150	2770
<i>Zebrasoma veliferum</i>	200	120	600	100	480	120	1420
<i>Z. xanthurum</i>	130	300	560	180	380	220	1640
<i>Ctenochaetus striatus</i>	130	440	560	420	200	620	2240
<i>P. Paracanthurus hepatus</i>	200	90	900	120	1100	1200	3410
<i>Naso unicornis</i>	140	360	800	400	750	760	3070
<i>N. lituratus</i>	200	140	1500	160	800	700	3300
<i>N. hexacanthus</i>	146	110	1800	140	900	450	3400



**Fig. 5** Taste buds form and distribution in the OC. **a** TBs in groups of *Acanthurus nigrofuscus*, bar 12  $\mu\text{m}$ ; **b** Type II TB of the inter-gill region of *A. sohal*, bar 2  $\mu\text{m}$ ; **c** Type I TB of *A. achilles*, bar 4  $\mu\text{m}$ ; **d** Transverse TB-bearing ridges on the palate of *A. nigrofuscus*, bar 220  $\mu\text{m}$ ; **e** Longitudinal TB-bearing ridges on palate of *A. nigricans*, bar 260  $\mu\text{m}$ ; **f** Enlarged part of E, bar 50  $\mu\text{m}$ ; **g** Bundles of TBs along ridges of *Ctenochaetus striatus*, bar 120  $\mu\text{m}$ ; inset, a magnified bundle of TBs, bar 20  $\mu\text{m}$ . e, elevated rim of epithelia; rm, micro-ridge mazes; sm, sensory microvilli of TBs; tp, taste buds papillae; tr, transverse ridges with TBs; white arrowheads, TBs; white arrows, ridges (nerve) connecting the TBs; star, ridges on the buccal valve



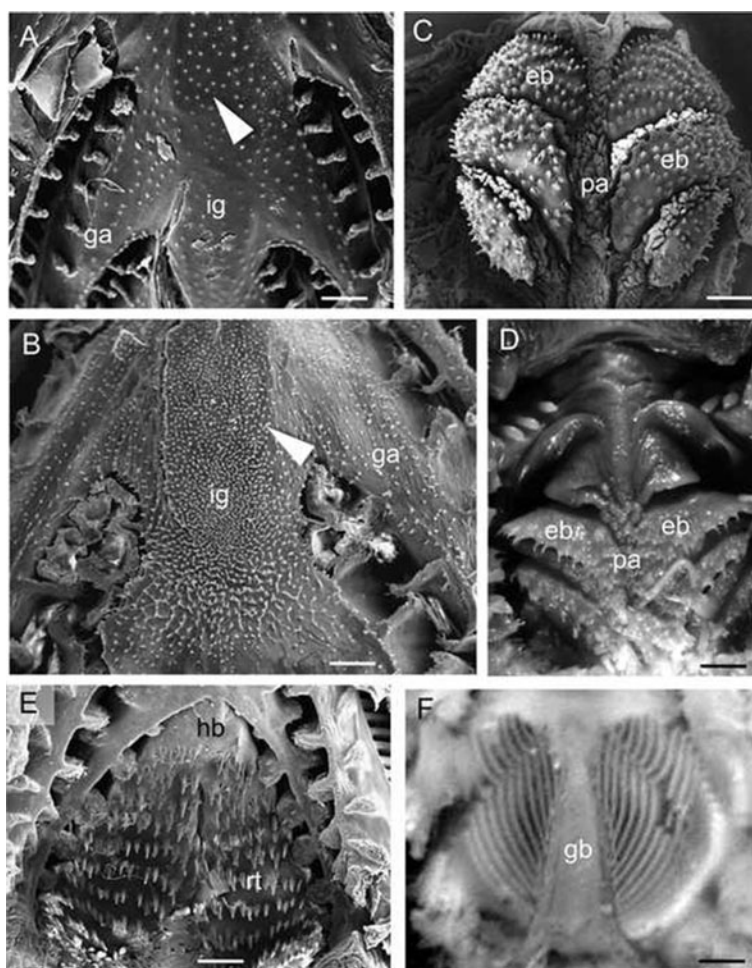
lamellae are oriented transverse to the body axis and, interweaving in the middle band with the hypobranchial ossicles, they form a kind of basket, a dense sieve, especially prominent in *C. striatus* (Fig. 6f), and able to retain the smallest food-particles collected. In species of the genus *Naso* these gill-lamellae basket is not formed.

## Discussion

During the last three decades several papers have been published on the diet and feeding in surgeonfishes (Robertson and Gaines 1986; Montgomery et al. 1989;

Purcell and Bellwood 1993; Wainwright and Bellwood 2002; and references therein), incorporating data on the mechanism of feeding, and stressing the extreme shortening of the biting jaws. Most of the species of this family are algivorous, with various microalgae and macroalgae being consumed by *Zebrafish* spp. and *Acanthurus* spp., macroalgae and zooplankton by *Naso* spp., and detritus by *Ctenochaetus* spp. and some *Acanthurus*. According to several authors, recognized zoophagy is a derived character. *A. sohal*, *A. nigrofuscus* and *Naso lituratus*, usually algivorous fish, were observed (L.F.) to swim-up into the open water and feed on zooplankton when macro-plankton swarms were dense.

**Fig. 6** Taste buds and pharyngeal bones. **a** Inter-gill region with TBs of *Acanthurus sohal*, bar 5  $\mu$ m; **b** Inter-gill region of *Paracanthurus hepatus*, bar 0.5 mm; **c** Epipharyngeal bones of *Naso unicornis*, bar 300  $\mu$ m; **d** Epipharyngeal bones of *A. gutattus*, bar 300  $\mu$ m; **e** Hypopharyngeal bone of *Naso hexanthus*, bar 0.8 mm; **f** Gill-basket of *Ctenochaetus striatus*, bar 0.8 mm. eb, epipharyngeal bones; ga, gill arches; gb, gill basket; hb, hypopharyngeal bone; ig, inter-gill region; pa, papillae with TBs; rt, rows of teeth



When engulfing the collected algae, the surgeonfish also take in the epifauna on them, including colonies of Hydrozoa, Bryozoa, numerous Foraminifera, micro-worms and crustaceans (personal observations). However, until now, no observations have been available on the soft tissues and sensory centers of the oral cavity of surgeonfishes, the function of which is to recognize the qualities of the engulfed material.

Following the extreme short jaws and mouth-opening, the gill-bases in surgeonfishes are situated more anterior than in predatory fish, and flattened, increasing the volume of the OC, and thus providing more space for food-accumulation and for the development of TBs. The widening of the basal part of the gill-arches expands the “oral basket” enabling the temporary accommodation of a large amount of food-particles, which then are sorted by the TBs and either discarded or swallowed. Especially prominent is

this basket in the detritivorous *Ctenochaetus striatus*. In carnivorous fish this activity is centered in the more anterior part of the OC, and expansion of space for the sorting of food items is thus not required.

A line of serrated teeth extends along the premaxillae and dentale of the studied species. These teeth are flat, laterally with prominent dentition in the genera *Acanthurus* and *Ctenochaetus*, and sharply pointed with rudimentary serration in the studied *Naso*. The various species reveal differences in the robustness of their teeth, which according to Wainwright and Bellwood (2002) reflects their different diets. Those authors suggested that the delicate comb-like teeth of *C. striatus* serve to brush-down epiphytic growths from the blades of macro algae or, as observed by us, to brush detrital material into the mouth cavity, whereas the more robust teeth of *A. nigrofusus* bite and shear algae. They also demonstrated that when biting down on the algal blades, the

fish “shakes” its head laterally. The feeding behavior of *A. nigricans* and *A. sohal* is similar, with both species possessing very robust teeth. During feeding the function of the “retention plates”, closely attached to the teeth on both jaws, becomes important. It would appear that here, where the teeth only perform initial perforation the algae, the plates close, holding the algae fast, while a shaking head motion by the fish detaches the required portion from the source. The plicate and strongly-papillated surface of the epithelium at this site helps to hold the food item in place during the head movement that separates it from its base. Such head-shaking, as observed by Purcell and Bellwood (1993), was observed in the Gulf of Aqaba in *A. nigricans*, *A. nigrofuscus* and *A. sohal*, but was less prominent in *Zebrosoma* spp., although the retention plates are well developed in the latter species. The uniserial denticulated and delicate teeth of *C. striatus* would be adapted to transport the detritus into the OC, where it is held and possibly ground by the plates. The major segregation of the edible material, for either engulfing or discarding, occurs deeper in the OC, above the larger fields of TBs and the basket sieve of the gills. Might the different food-items and differential feeding have been derived secondarily from an algivorous ancestor? Examination of the OC in *Siganus luridus* and *S. rivulatus*, belonging to the Siganidae, an herbivore family related to the Acanthuridae, revealed the presence of retention plates also in these fishes too. Their teeth, featuring three small denticles, the OC with numerous longitudinal ridges bearing TBs, strongly resemble those of the surgeonfishes. Might this indicate that they branched out evolutionarily after the surgeonfishes have developed their oral plates? Yashpal et al. (2009) described uncini on the upper jaw of the carp, *Cirrhinus mrigula*, which, like the plates in acanthurids, seem to function in food-holding or scraping; and Moron (2011) in his MSc. thesis (unpublished) described the inside of the jaws in *Kyphosus*-fishes, which resemble that in surgeonfishes (also personal observation, LF). In conjunction with this, it will be of interest to study other families of herbivorous fishes, some of which are closely related to the surgeonfishes.

As revealed by various studies, the number of OC TBs increases with growth of the fish, albeit partly remaining species-specific (Fishelson et al. 2004, 2010). For example, in *Paracanthurus hepatus* and *Acanthurus nigrofuscus*, of equal 180 mm SL, their OC feature 6200 and 2480 TBs, respectively. A similar

phenomenon was observed in other families of fishes, such as the cichlids: in *Labotropheus trewavasae* of 110 mm SL they number 3400, whereas in *Tilapia zillii* of the same SL they number 17 500; in the cardinal fish *Fowleria variegata* of 75 mm there are 24 600 TBs, and in *Cheilodipterus caninus* of 160 mm SL only 3270 TBs (Fishelson et al. 2010, 2012). Similar phenomena were observed in surgeonfishes, even in closely-related species. For example, in *A. nigricans* and *A. nigrofuscus* the number of TBs is 4780 and 2480, respectively. There is also a difference in location of the majority of TBs between predatory and herbivorous fishes: in the former, as seen in the lizardfishes (Fishelson et al. 2010), most TBs are situated in the frontal part of the OC, whereas in the studied surgeonfishes they are concentrated more at the posterior, inter-gill and pharyngeal sites. This may indicate that the predatory fishes are more visually selective in regard to their prey, whereas in the herbivorous fish the selection is more chemically oriented, occurring within the OC. This correlation between neural branches and TB position is well illustrated in the work by Sakata et al. (2001) on barbels of catfishes.

In the studied surgeonfishes, as in other fishes (Fishelson et al. 2010), the number of TBs/1mm SL of the body also reveals a certain constancy during growth of the fish, until reaching a certain standard length. For example, in 180 mm SL *Acanthurus nigricans* it is 26 TBs/mm SL, and in *A. nigrofuscus* it is ca. 14 TBs/mm SL, whereas in *Paracanthurus hepatus* of the same SL 30 TB/mm can be observed. This is certainly low compared to 38 TBs/mm SL in *Starksia sluiteri* of 38 mm (Labrisomidae), or 40 TBs/mm SL in *Clinus superciliosus* of 100 mm SL (Clinidae). This constancy illustrates the specificity of TB numbers in the various species, as well as a possible ongoing correlation between growth of the body, increase in brain size, and genesis of the peripheral sensory network that induces TB formation.

Comparing the number of TBs in the single available SL-series of *A. nigrofuscus*, revealed a gradual increase, from 200 in juveniles of 14 mm SL to ca. 2480 in adult fish of 180 mm SL. A subsequent counting of TB numbers in larger fish revealed very minimal changes, and the number of TBs remained almost unchanged, asymptotic to the increase of SL of the fish, up to 220 mm. A similar trend was observed in the small number of *A. sohal* specimens of various sizes. Might this indicate that the maturation of the neural network, which induces and establishes the



base for TB formation, occurs before the fish attain their maximal standard length? Interestingly, a similar phenomenon was observed by Choat and Axe (1996) in regard to increase in size of the sagittal otoliths in acanthurids, namely: the sagittas reach asymptosis in growth (and not in weight) before the fish attain their full growth.

The subtle differences in OC morphology, especially of TB distribution, as well as the various forms of the retention plates, are eco-morphological adaptations correlated with the variety of diets of the different species. The present study of the soft tissues in the OC of surgeonfishes, especially the difference in the retention plate structures and TB numbers and position, presents an additional and unexploited source for comparisons of cyto-morphological adaptations. As such, it is expected to contribute to the understanding of the phylogenies associated with diet and feeding that contribute to the separation of the various species.

**Acknowledgments** The authors are grateful to Daniel Golani of the Hebrew University, (Jerusalem), Carole Baldwin of the Natural History Museum Smithsonian Institution (Washington, USA), and to L. O'Hara and A. Suzumoto of the Bernice Bishop Museum (Hawaii), for the contribution of specimens for this study. We also thank Naomi Paz for her editorial assistance and Varda Vexler for help in artwork. Thanks are due to the anonymous reviewers.

## References

- Barlow GM (1974) Extraspecific imposition of social grouping among surgeonfish's. *J Zool (Lond)* 174:333–340
- Choat H, Axe LM (1996) Growth and longevity in acanthurid fishes; an analysis of otolith increments. *Mar Ecol Prog Ser* 134:15–26
- Choat JH, Robbins WD, Clements KD (2004) The trophic status of herbivorous fishes on coral reefs. *Mar Biol* 145:445–454
- Clements KD, Gray RD, Choat JH (2003) Rapid evolutionary divergences in reef fish family Acanthuridae (Perciformes: Teleostei). *Mol Phylogenet Evol* 26:190–201
- Doty RL (2003) Handbook of olfaction and gestation, 2nd edn. Marcel Dekker, New York
- Finger TE, Silver WL, Restrepo D (2000) The neurobiology of taste and smell. Wiley-Liss, New York
- Fishelson L (1977) Sociobiology of feeding behavior of coral-reef fishes of the Gulf of Eilat, Red Sea. *Israel J Zool* 26:114–134
- Fishelson L (1984) A comparative study of ridge-mazes on surface epithelial cell membranes of fish scales. *Zoomorphology* 104:231–238
- Fishelson L (2005a) Comparison of taste bud types and their distribution on the lips and oropharyngeal cavity, as well as dentition in cichlid fishes (Cichlidae, Teleostei). In: Reutter K, Kapoor BG (eds) *Fish chemosenses*. Science Publishers, Enfield, pp 247–275
- Fishelson L (2005b) Histogenesis of the oropharyngeal cavity taste buds and the relevant nerves and brain centers in substrate-brooding and mouth brooding cichlid fishes (Cichlidae, Teleostei). *Anat Embryol* 209:179–192
- Fishelson L, Delarea Y (2004) Taste buds on lips and mouth of some blennioid and gobiid fishes: comparative distribution and morphology. *J Fish Biol* 65:651–665
- Fishelson L, Montgomery LW, Myrberg AA Jr (1987) Biology of surgeonfish *Acanthurus nigrofasciatus* with emphasis on changeover of diet and annual gonadal cycle. *Mar Ecol Prog Ser* 39:37–47
- Fishelson L, Delarea Y, Zverdling A (2004) Taste bud form and distribution on lips and in the oropharyngeal cavity of cardinal fish species (Apogonidae, Teleostei), with remarks on their dentition. *J Morphol* 259:316–327
- Fishelson L, Golani D, Galil B, Goren M (2010) Comparison of taste bud form, number and distribution in the oropharyngeal cavity of lizardfishes (Aulopiformes, Synodontidae). *Cybio* 34:260–277
- Fishelson L, Baldwin CC, Hastings PA (2012) Comparison of the oropharyngeal cavity in the Starksiiini (Teleostei: Blenniiformes: Labrisomidae): taste buds and teeth, including a comparison with closely-related genera. *J Morphol* 273:618–628
- Gon O, Fishelson L, Delarea Y (2010) Comparative morphology of the oropharyngeal cavity in clinid fishes (Perciformes: Clinidae), with particular attention to the form, number and distribution of taste buds and dentition. *Am J Mar Sci* 29:283–298
- Guiaes RC, Winterbottom R (1993) Osteological evidence for the phylogeny of recent genera of surgeonfishes (Percomorpha, Acanthuridae). *Copeia* 1993(2):300–312
- Hansen A, Reutter K (2004) Chemosensory systems in fish: structural, functional and ecological aspects. In: Von der Emde G, Magdans J, Kapoor BG (eds) *The senses of fish: adaptation for the reception of natural stimuli*. Narosa Publishing House, Kluwer Academic Publishers and Springer-Verlag, Dordrecht, pp 55–89
- Hara TJ (2006) Gustation. In: Zielinski B, Hara TJ (eds) *Sensory systems neurosciences*. Fish Physiol 25: 45–96
- Hiatt RW, Strasburg DW (1960) Ecological relationship of the fish fauna on coral reefs of the Marshall Islands. *Ecol Monogr* 30:65–127
- Hobson ES (1974) Feeding relationships of teleostean fishes on coral reefs in Kona, Hawaii. *U S Fish Bull* 72:915–1031
- Kasumyan AO (2011) Functional development of chemosensory systems in fish ontogeny. *Ontogenesis* 42:205–212
- Kinnamon JC (1987) Organization and innervations of taste buds. In: Finger TE, Silver WL (eds) *Neurobiology of taste and smell*. Wiley-Interscience, New York, pp 277–297
- Montgomery WL, Myrberg AA, Fishelson L Jr (1989) Feeding ecology of surgeonfishes (Acanthuridae) in northern Red Sea, with particular reference to *Acanthurus nigrofasciatus*. *J Exp Mar Biol Ecol* 132:179–207
- Moron C (2011) Curious food procurement in the Eastern North Pacific Kyphosidae. California State University, Monterey bay (MS-Thesis)

- Myrberg AA Jr, Fishelson L, Montgomery WL (1985) The reproductive behavior of the surgeonfish *Acanthurus nigrofasciatus* on the reefs of Eilat, Israel (Gulf of Aqaba, Red Sea), with emphasis on migration and spawning. 19th Ethological Conference, Toulouse, France pp 1: 25
- Northcutt RG (2004) Taste buds: development and evolution. *Brain Behav Evol* 64:198–206
- Northcutt RG (2005) Taste bud development in the channel catfish. *J Comp Neurol* 482:1–16
- Purcell SW, Bellwood DR (1993) A functional analysis of food procurement in two surgeonfish species, *Acanthurus nigrofasciatus* and *Ctenochaetus striatus* (Acanthuridae). *Environ Biol Fishes* 37:139–159
- Qgawa K, Caprio J (2010) Major differences in the proportion of amino acids fiber types transmitting information from oral and external regions in the channel catfish. *J Neurophysiol* 103:2062–2073
- Randall JE (1967) Food habits of reef fishes of the West Indies. Proceedings of the International Conference on Tropical Oceanography. Univ Miami Stud Trop Oceanogr 5:665–847
- Reutter K, Hansen A (2005) Subtypes of light and dark elongated taste bud cells in fishes. In: Reutter K, Kapoor BG (eds) Fish chemosenses. Science Publishers, Enfield, pp 211–230
- Robertson DR (1983) On the spawning behavior and spawning cycles of eight surgeonfishes (Acanthuridae) from the Indo-Pacific. *Environ Biol Fishes* 9:193–223
- Robertson RD, Gaines SD (1986) Interference competition structures habitat use in a local assemblage of coral reef surgeonfishes. *Ecology* 67:1372–1383
- Sakata Y, Tsukahara J, Kiyohara S (2001) Distribution of nerve fibers in the barbels of sea catfish *Plotosus lineatus*. *Fish Sci* 67:1136–1144
- Sano M, Shimizu M, Nose Y (1984) Food habits of teleostean reef fishes in Okinawa Island, southern Japan. *Univ Tokyo Bull* 25:1–128
- Schuhmacher H, Krone R, van Treeck P (2008) Underestimated eroders among reef fishes-experimental comparison between *Ctenochaetus striatus* and *Acanthurus nigrofasciatus* (Acanthuridae). Proceeding of the 11th International Coral Reef Symposium (Florida), 331–334
- Tagliafierro G, Zaccone G (2001) Morphology and immunochemistry of taste buds in bony fishes. In: Kapoor BG, Hara TJ (eds) Sensory biology of jawed fishes. IBH, New Delhi, pp 335–345
- Tang KL, Berendzen PB, Wiley EO, Morrissey JF, Winterbottom R, Johnson GD (1999) The phylogenetic relationships of the suborder Acanthuroidei (Teleostei: Perciformes) based on molecular and morphological evidences. *Mol Phylogenet Evol* 11:415–425
- Tilghman GC, Klinger-Bowen R, Francis-Floyd R (2001) Feeding electivity indices in surgeonfish (*Acanthuridae*). *Aquar Sci Conserv* 3:215–223
- Tyler JC, Johnson D, Nakamura I, Collette B (1989) Morphology of *Luvarus imperialis* (Luvaridae) with phylogenetic analysis of the Acanthuridae (Pisces). *Smithson Contrib Zool* 485:1–78
- Wainwright PC, Bellwood DR (2002) Ecomorphology of feeding in coral reef fishes. In: Sale PF (ed) Coral reef fishes, dynamic and diversity in a complex ecosystem. Acad Press, Orlando, pp 33–55
- Winterbottom R, McLennan DA (1993) Cladogram versatility: evolution and biogeography of Acanthurid fishes. *Evolution* 47:1557–1571
- Yashpal M, Kumari U, Mittal S, Mittal AK (2009) Morphological specialization of the buccal cavity in relation to food and feeding habit of a carp *Cirrhinus marigala*: a scanning electron microscope investigation. *J Morphol* 270:714–728