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# **Review Article**

# Major Ectoparasitic Protozoa of Fish and Other Aquatic Animals: With Particular Emphasis on Morphology, Biology, Epidemiology, Pathology, Diagnosis, Prevention and Control

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### Abstract

The Phylum protozoa brings together several organisms evolutionarily different that may act as ecto or endoparasites of fishes and other aquatic animals over the world being responsible for diseases, which, in turn, may lead to economic, social and health impacts in many countries. Though ectoparasitic protozoans affect a wide variety of fish species and pose tremendous impact on the economy and health of people worldwide, only little attention is given towards the disease. Therefore, this review highlights on the most important protozoan ectoparasites infecting fish and other aquatic animals: with particular emphasis on morphological features, distribution, epidemiology, pathology, pathogenic effects, clinical importance, diagnosis, and prevention and control.

# **INTRODUCTION**

Fish is important to human populace in trade and economy; it is of importance in the diet of different countries especially in the tropics and subtropics where malnutrition is a major problem [1]. As the human population inevitably increases, the demand for fish as source of protein also grows. In recent times, there has been tremendous increase in the development of fish farming and culture attributable to the increased need for affordable animal protein especially in the tropics [1,2].

However, parasitic infections and diseases are some of the factors that hindering the high productivity in fish farming [1,3]. According to Klinger and Franci-Floyd [4], protozoa are a vast assemblage of eukaryotic organisms and that most of the commonly encountered fish parasites are protozoa, which with practice, are the easiest to identify and easiest to control. In general protozoa are one of the major sectors of fish parasites that have been long neglected because of its inherent difficulty in studying compared to other larger parasites. Among protozoa, ecto- and endo-parasitic protozoa occupy a very important sector as one of the hazardous threats to fish health. These parasites attack the fish, causing massive destruction of skin and gill epithelium. Even moderate infection of these organisms on small fish may prove a fatal disease, since the infection may cause the fish to stop feeding [1,5].

Some fish parasites would develop in humans if the fish is eaten raw, but none would be harmful if the fish is thoroughly cooked. All reports of people being infested with fish parasites were because of ingestion of raw fish or insufficiently cooked fish [1]. Most fish especially in the wild population are likely to be infested with parasites, but in the great majority of cases, no significant harm to the host may be ensued or identified; thus, there are only few reports of parasites causing mortality or

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serious damage to the fish populations, but this may be largely because such effects go unnoticed [1,6]. Fishermen or consumers often observe parasites in wild fish only when they are so obvious as to lead to rejection of fish [1]. In culture fish population, on the other hand, parasites often cause serious outbreak of disease. The presence of dense populations of fish kept in particular environmental conditions may favor certain parasites so that the parasite population increases to a very high level. According to Roberts et al. [7], parasites are the most diverse and common pathogens the aqua culturist may likely encounter, and parasitic diseases are very common in fish all over the world and are of particular importance in the tropics [1].

Parasite of fish can either be external or internal. Parasitic infections often give an indication of the quality of water, since parasites generally increase in abundance and diversity in more polluted waters [1,8]. Parasites are capable of causing harm to the fish host notwithstanding the spp., either through injury to the tissues or organs in the process of burrowing or consuming food or the removal of digested food in the gut of the fish as well as the secretion of proteolytic enzymes [1].

Fish parasites result in economic losses not only mortality, but also from treatment expenses, growth reduction during and after outbreak of disease and this militate against expansion of aquaculture. Protozoan parasites cause serious losses in fishponds and wild fish, and their lesions render the fish unmarketable. Fish carrying protozoa parasites are capable of passing on the infective disease to man after its consumption. In general, ectoparasitic protozoans are common tropical freshwater fish parasites that affect public health and cause losses to fishes [1].

A number of prophylactic and curative measures have been suggested, although many of the recommended chemicals may be overused or misused by aqua cultural workers, leading to parasite's potential drug resistance and negative impact on the aquatic environment. In this way, it must be highlighted the importance of disease impact in order to elaborate efficient strategies for early diagnosis and fast intervention in management practices in fish farms so as to obtain a stable and sustainable production [9]. Therefore, this review focuses on the most important protozoan ectoparasites infecting fish and other aquatic animals with particular emphasis on morphological features, distribution, epidemiology, pathology, pathogenic effects, clinical importance, diagnosis, and prevention and control strategies.

# LITERATURES REVIEW

# **Morphological Features and Taxonomy**

**Host Range:** Host specific species are associated with a wide range of fish species from most families. Ubiquitous or opportunistic species (*Ichthyobodo necator, Chilodonella* spp., and some species of *Trichodina, Ambyphrya* and *Scopulata* (*Scyphidia*) are particularly common in juvenile Cichlids and Carp [11] [Table 1].

Distribution and Epidemiology: The ubiquitous

<b>Table 1:</b> Morphological features of ectoprotozoan parasites of fish and other aquatic
animals

Order	Characteristic Features		
Flagellates [Mastigophora, Kinetoplastida]:			
Cryptobia	Free, spindle shaped, $10-30 \times 3-5 \mu m$ in size (if <i>C. branchialis</i> ), or pyriform when attached to the integument, with two flagellae, one wholly or partly adjunct to the body, kinetoplast rod-shaped or round.		
Ichthyobodo	Free, 13–26 $\times$ 2–7 $\mu m$ in size, or attached to the integument; with four flagellae.		
Ciliates [Ciliophora]:			
Chilodonella	Rounded to oval, cytostome distinct, macronucleus round and cilia on the concave ventral surface are arranged in several concave parallel rows:		
C. hexasticha	Size 30–65 $\times$ 20–50 $\mu m$ , with 6–8 ciliary lines on each side.		
C. piscicola (syn: C. cyprini)	Size 33–100 $\times$ 24–60 $\mu m,$ with more than 10 ciliary lines on each side.		
Trichodina	Cup shaped: $20-100 \ \mu m$ in diameter with concentric rows of cilia and a crown of denticles. The denticle shape is a distinct taxonomic feature; for differential specific diagnosis of African spp.		

ectoprotozoans are cosmopolitan or transcontinentally dispersed via translocation of their cultured fish hosts (carp and tilapia in particula)(Ichthyobodo necator, Cryptobia branchialis, Chilodonella hexasticha, C. piscicola, Trichodina acuta, T. heterodentata and T. pediculus, T. reticulata, T. mutabilis, T. centrostrigata, Trichodinella epizootica [12-20]. Distribution of the more specialized host specific species follows that of their hosts, but may also be more restricted, sometimes to only one or a few watersheds. There is evidence for the presence of Ichthyobodo, Chilodonella, and in particular trichodinids and sessilians in a number of water systems in tropical Africa (Lake Volta and East African lake systems [21-24], but taxonomic data are limited to only a few locations [25,26]. The most comprehensive taxonomic data on trichodinids in African fish are from Southern Africa and the Zambezi river system. Data are also available from Israeli fish [10,13,15,27].

**Life Cycle and Biology:** Most ectoprotozoans, flagellates as well as ciliates, have simple life histories. Species of *Cryptobia* are ectoparasites as well as intestinal and vascular parasites. It has been shown that an ectoparasitic phase occurs in two vascular species [28]. Both ectoparasitic flagellates, *I. necator* and *Cryptobia* spp., occur either free swimming or attached to the integument, the former through a cytoplasmic protrusion [29] and the latter by attachment with the flagellum [30].

Reproduction is usually by binary fission. Conjugation is sometimes observed in ciliates. Sessile species also bud and give birth to a free swimming mobile generation, reminiscent of mobile peritrichs, which settle on suitable substrates (fish). The sessile suctorians reproduce by internal and external budding, the detached buds are ciliated. As the buds become attached to a new location on the piscine integument, cilia are shed and tentacles appear [12]. Spores or other forms of waiting stages are unknown; the suggested existence of waiting stages, such as encysted forms of *Chilodonella hexasticha* in the gills [31] or free cysts [32], has to be confirmed. Water temperatures do not seem to be an important parameter, in spite of reports of low temperatures being more optimal for reproduction of *Chilodonella piscicola* and some trichodinids [32]. Massive infections with *I. necator*, both species of *Chilodonella* and the ubiquitous trichodinids and sessile species, occur in low (12–17°C) and high (2530°C) ambient temperatures in southern Africa and Israel. Most freshwater ectoparasitic protozoans disappear in ponds with increased salinities (above 2000 ppm chlorinity); only *I. necator* and some *Cryptobia* are tolerant and become the predominant parasites in fish of such ponds. There are also halophilic species of *Ambyphrya* and *Scyphidia* which infect fish (grey mullet) in estuaries [11].

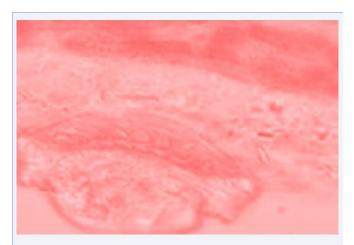
A number of ciliates (species of *Tetrahymena, Ophryoglena, Glaucoma, Colpidium* and others (see Hoffman [12]) are facultative parasites, or opportunists which will colonize fish in special circumstances, most often when fish are stressed or traumatized [12]. All others mentioned above are obligatory parasites which will apparently survive for only a limited time outside their hosts. Nonparasitic sessile peritrichs are different species from those colonizing living organisms. Trichodinids and Sessile species found on aquatic invertebrates comprise different species from those infecting fish [14,33,34]. There are, however, a few documented exceptions: *T. pediculus* being reported from both hydra and fish, and *T. diaptomi* a parasite of a calanoid copepod, which temporarily invaded hatchery grown fry of *Clarias gariepinus* [13,35].

There are several degrees of adaptation of trichodinids to their piscine hosts: ubiquitous species, of an opportunistic nature, which are always found on the fish skin but never on the gills (T. pediculus and T. acuta); other ubiquitous species occur both on gills and skin (T. heterodentata); additional, seemingly ubiquitous, widespread species appear to have a variable degree of predilection for one fish family or another (cichlids or cyprinids) [11]. Among the latter, trichodinids with seemingly related morphological characteristics (e.g. pediculuslike, acuta like and nigra like), in different geographical regions, demonstrate definite affinities to a particular group of hosts and may in fact comprise diverse species [15]. Host specific trichodinids, are all, with only a few exceptions, gill parasites: *T*. centrostrigata and great numbers of small trichodinids mainly species of Tripartiella, are associated with Cichlidae; T. reticulata occursmainly in goldfish, T. kazubski has been found in South African Barbus spp. and T. nobilis and T. kupermani mainlyin asian carp [13-15,18].

**Pathology and Pathological Effects:** Ectoparasitic protozoa are variable in their effect on their hosts. Pathological effects are density dependent, when both the size of the parasite population and the nature of the tissue responses are modulated by the physiological (clinical) condition of the fish. Hostile environments (stressful conditions) compromise the fishes' capacity to counteract infection [11].

*Ichthyobodo necator* attaches itself to epithelial cells and through an inserted protrusion consumes their contents [29]; whereas *Chilodonella* spp. browse the epithelial surface [36] (Figure 1,2). Histopathological changes in the integument

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**Figure 1** *Trichodina* spp. on the skin of *C. gariepinus.* Source: Omeji et al. [1].



**Figure 2** *Chilodonella* spp. on the skin of *C. gariepinus* Source: Omeji et al. [1].

following infection by Chilodonella spp. and I. necator are an outcome of two counteracting cellular processes hyperplasia of the epithelial cells, including mucus cells and chloride cells, versus a progressive cellular destruction. Cellular destruction primarily occurs due to direct action of the parasites, and later by enhanced abrasion of the peripheral cells after the depletion of mucus forming cells. The production of mucus cells is limited. Accelerated mucus cell production stimulated by the infection apparently exhausts resources for mucus production, and the infected fish become "dry". Some parasites seem to yield cytotoxins or proteolytic enzymes which could be the cause of spongiosis, which affects both the proliferated and unchanged epithelial layer [36,37]. Secondary cellular damage due to degeneration, necrosis and desquamation results in the degradation and disintegration of the epithelial layer [11]. Cryptobia attachment through the flagellum does not induce any pathological or even ultra-structural cellular damage [30], contrary to reports of morbidities associated with this parasite [28].

Although there are a number of reports on poor condition and mortalities, particularly of fry, coinciding with massive infestation of trichodinids, Trichodinella epizootica in particular [38], and the sessilians Apiosoma, Ambyphrya and Scopulata [39-43], histopathological changes in events of massiveinfections by these ectoprotozoans are hardly evident, if occurring at all [44,45]. Trichodinella epizootica in carp [38] and Tripartiella cichlidarum in cichlids [43] cause some erosion of the gill epithelium. However, food vacuoles of trichodinids revealed bacteria rather than sloughed cells [45]. Ultrastructural observation on attached Apiosoma did not reveal any interference with the host cell serving as substrate [38,44,46] or peripheral tissue response. Thus, mortalities following massive colonization of gills by sessilians [39] could result from the dense cover of sessilians disrupting gas exchange through the respiratory epithelium. The only exception among these infections are the colonies of the stalked sessilia Heteropolaria (Epistilis) which cause lesions ("red sore") at the stalk attachment to the fish skin, these inflamed hemorrhagic lesions are also contaminated with the bacterium Aeromonas hydrophila [47,48]. Reported localized infection above the opercular bone (in cultured tilapia in Israel) resulted in aggravation of the lesion into a wide (6 mm in diam.) perforation of the bone [43]. Suctorians (Trichophyra spp.) in certain instances cause cytological damage to the gill lamellae cells in direct contact with the parasites and subsequent hyperplasia and hemorrhages of the gill tissue [49].

Clinical Importance: Clinical manifestation comprises nonspecific signs such as respiratory difficulty, loss of equilibrium and appetite. Apart from these alterations, whitish lesions on the gills, scaleness, darkened skin, skin and fins ulcers, and haemorrhagic areas are common in mixed infection with bacteria [9]. In highly infested fish by *Epistylis* spp. it can be seen fin erosion and skin ulcers related to bacterial enzymes activity. On histological sections, degeneration and epithelial necrosis adjacent to the colonies, intense desquamation, as well as increased mucus production and inflammatory infiltrate are also related. The fish death occurs generally after secondary invasion by opportunistic bacteria thus causing systemic infection. In Ichthyophthirius multifiliis, the main clinical sign is the presence of white spots on the fish surface including skin, fins, eyes, buccal cavity and gills. It is common to observe fish with respiratory difficulty, flashing behavior in ponds and aquaria, and in cage reared fish flashing on the sieve cages, leading to muddy water in earth ponds. In Trichodinidae, clinical signs are not specific and diseased fish may present darkness of the skin, whitish areas in the gills, hypoxia and flashing on the ponds or aquaria surface. Lethargy and erratic swimming on the body surface are frequently found in diseased fish larvae. Most other flagellates, more or less, have similar clinical pictures [9].

**Epizootiology:** The course of infection by ectoparasitic protozoans is determined either individually or by the interaction of the following factors: (a) Mobility of the fish; (b) The fish's capacity to activate its defense systems [11]. Reduced mobility facilitates parasite colonization as well as moderating loss through detachment and drift from the integumental surface

[11]. Defense mechanisms other than epithelial hyperplasia, and specific immune responses to integumental ectoparasites have not yet been studied (except in *I. multifiliis*), although spontaneous recovery from infection has been frequently observed. Juvenile fish and fish under stress (and at below optimum ambient temperature) have both limited mobility and apparently immunological incompatibility, being either naïve or immunosuppressed [50,51].

Heavy infections by ectoparasitic protozoans are mainly found in young fish (less than one year old) when overcrowded and confined to restricted habitats, and under stress conditions. In these circumstances opportunistic and ubiquitous species are involved. Infections otherwise, in grown-up fish, are very low and host-specific species predominate [11]. New born cichlids, as soon as they were weaned from parental care and sometimes before, became heavily infected by trichodinids and sessilians of the genera Ambyphrya and Scopulata. Infestation reached its climax level in fish 10 to 12 mm long. Such infections occur in natural habitats (lakes), man-made impoundments, as well as in hatchery installations [21,41,45]. Heavy infections, however, were not found in all the breeding habitats of the investigated lake system. Conditions for infestation varied with habitat and ambient conditions and were positively related to the abundance of fry schools [11].

Level of infection in the fry sharply declined as fish gained in size [45]. The decline in infection also coincided with changes in parasite species composition, the ubiquitous, generalists and opportunists (*T. pediculus* [=? *T. migala*], *T. acuta* [=? *T. compacta*, Basson & Van As, 1989], *T. heterodentata*, and species of *Ambyphrya* and *Scopularia*) being gradually replaced by species specific to cichlids (*Tripartiella* spp. *Trichodina centrostrigata* and speciesof *Apiosoma*) [13,15,25-27]. Heavy infections with ubiquitous trichodinids (*T. pediculus*) and sessile peritrichs (mainly *Scopulata* spp.) also occur in carp fry in hatcheries and nursery ponds, and likewise as fish grow, are replaced by more specialized species (such as *T. nigra*, *T. mutabilis* and *Apiosoma* spp.) [13,17,18].

Heavy infections (by trichodinids and sessile species) accumulate in fish small spp. Of *Barbus, Alestes,* cyprinodontids and juvenile cichlids and *Clarias* spp. crowded in residual pools in rivers drying out during the dry season. In larger water bodies in Africa, infections with both trichodin and sessilians in fish other than fry may be common but low [22,35,52]. Low temperature stress plays an important role in epizootic outbreaks of ectoprotozoan infections in cichlid fish outside the limits of the tropical environment and of populations introduced to non-tropical countries such as the southern USA [11]. Heavy infections by skin and gill protozoa, predominantly of *Chilodonella* spp., are a frequent occurrence in overwintering stocks of cultured tilapia hybrids (*Oreochromis aureus x niloticus*) inIsrael, and *O. mossambicus* in ponds and dam reservoirs in southern Africa [53-55].

In small ponds (1 hectare) fish are not spared even in relatively mild winters, with minimum temperatures above

13°C. Fish in lakes and large reservoirs on the other hand, become severely affected only in extremely cold winters, with temperatures declining to 10°C and below. Mortalities often occur from the cumulative effects of ectoprotozoans, dermal saprolegniases and systemic bacterial diseases, all mediated by the stress of low temperature [11]. In addition to temperature stress, overwintering tilapia in ponds are often stressed by overcrowded stocking and inadequate feeding. Intermittence of higher and lower ambient temperatures, characteristic of the Mediterranean type winters, increases the unpredictability of food demand by fish and thus complicates feeding schedules [11].

Infestation levels rise by late fall, with increased abundance of trichodinids and *C. hexasticha*. Fish succumbing in early winter were predominantly hyperinfected by *C. hexasticha*. Late winter and early spring mass mortalities (even when temperatures were already rising above 15°C) were associated with *C. piscicola* hyperinfections. *C. piscicola* is abundant in carp in some ponds already by early winter; however, it will only infect tilapia at the end of the cold season when they become compromised by prolonged stress [11].

*Ichthyobodo necator* hyperinfections are morbid to cichlids as well as to fish of other families. Mortalities occur in fish overcrowded in holding tanks, ponds and in both warm and cold water conditions. Natural infection was also revealed in *Aplocheilichthys gambiae* from a pool in Ghana. In Israel *Cryptobia* spp. occasionally swarm the gills of tilapia, goldfish and silver carp and also, in the latter, in low saline waters (8–10 ppt salt) but data from Africa are lacking [11]. *O. mossambicus* appears to be more tolerant to low temperatures in water of higher salinities, and also where most ectoparasites are excluded (except *l. necator*). Members of the genus *Tilapia*,in Israel (*T. zillii*) and in southern Africa (*T. rendalli & T. sparmanii*) are also less affected in freshwaters by low temperatures and are rarely heavily parasitized [11].

Few instances of mortalities coincided with heavy infections, concomitantly or exclusively, by trichodinids, sessilians (*Apiosoma*), *Chilodonella* spp. And *I. necator*, in overwintering carp, but occurred more often in relation to other stress factors such as high levels of overcrowding or high nitrite concentrations [39,45,56]. Heavy infections by *Chilodonella* spp. Seems to have an excluding effect on other integumental protozoans. Otherwise, skin and gill ectoparasites coexist, and are even synergistic, with metazoan ectoparasites (*Gyrodactylus* and *Argulus*) and skin lesions (epithelioma) [45,56,57]. Mass mortalities of farmed *Clarias gariepinus* (in the Central African Republic) were associated with mass infestation by *Chilodonella hexasticha*. *Epistilis* infections, including red sore and opercular perforations only occur sporadically with no particular link to overwintering [11].

**Diagnosis:** Integumental ectoprotozoan genera are readily differentiated [58,59], while diagnosis of species is difficult and often requires special staining [11]. Most ectoparasitic forms are readily detected in direct microscopic examination of skin and gill

scrapings from live (or freshly killed) fish [11]. Flagellates may be further detected in air dried, methanol fixed, Giemsa stained smears. Smears containing ciliates should be air dried, fixed in Bouin for 20 min., destained in 70% ethanol, brought to water, stained in a haematoxylin stain and mounted after dehydration [11] (Figure 3, Figure 4). Trichodinids for specific differentiation should be impregnated with silver. Air dried smears should be placed in 2% silver nitrate for 7–9 min. in the dark, rinsed in water and exposed to the sun or UV for 5–10 min (Figure 3 and 4) [11] [Table 2].

Small trichodinids, predominantly from the gills, are bell shaped (Tripartiella and Paratrichodina) and often settle on the tips of the gill lamellae [15], for generic division of Trichodinidae). In Trichodinella, the ray (the inner extension of the denticle) [15], is totally reduced, and in the other two genera is delicate or rudimentary [25,27]. Genera of sessile peritrichs are differentiated by their macronuclei and scopula (attachment leg) [33,34]: Scopulata (Scyphidia) round macronucleus and wide scopula; Apiosoma (Glossatella) pyriform nucleus, small scopula; Ambyphrya Ribbon shaped macronucleus and wide scopula. Stalked sessile peritrichs Heteropolaria with elongate body and curled macronucleus [60]; Epistilis cup-shaped with horseshoe shaped macronucleus [33]. Some Apiosoma also develop on stalks. Suctoria: (Trichophyra and other genera) cilia lacking, variable numbers of tentacles arise from the rounded body [11]. Source: Van As and Basson [10].

**Control and Prevention:** Treatment with formalin is still the only effective means to control massive ectoparasitic infections in all warm water cultured fish species. In Israeli fish farms, ponds are sprayed with formalin up to concentrations of 25 or 40 ppm (of the 37% commercial product) [56,61]. Efficacy of formalin treatments is affected by ambient temperatures, water quality, including salinities and parasites treated. Product quality is variable, and is particularly affected by storage, resulting in accumulation of polymerized (paraformaldehyde) sediment.

Table 2: Important diagnostic features of ectoprotozoan parasites of fish and other aquatic animals

Order	Diagnostic Features	
Flagellates [Mastigophora, Kinetoplastida]:		
Cryptobia	Free, spindle shaped, $10-30 \times 3-5 \mu m$ in size (if <i>C. branchialis</i> ), or pyriform when attached to the integument, with two flagellae, one wholly or partly adjunct to the body, kinetoplast rod-shaped or round.	
Ichthyobodo	Free, 13–26 × 2–7 μm in size, or attached to the integument; with four flagellae.	
Ciliates [Ciliophora]:		
Chilodonella	Rounded to oval, cytostome distinct, macronucleus round and cilia on the concave ventral surface are arranged in several concave parallel rows:	
C. hexasticha	Size 30–65 $\times$ 20–50 $\mu m,$ with 6–8 ciliary lines on each side.	
C. piscicola (syn: C. cyprini)	Size 33–100 $\times$ 24–60 $\mu\text{m},$ with more than 10 ciliary lines on each side.	
Trichodina	Cup shaped: 20–100 µm in diameter with concentric rows of cilia and a crown of denticles. The denticle shape is a distinct taxonomic feature; for differential specific diagnosis of African spp.	

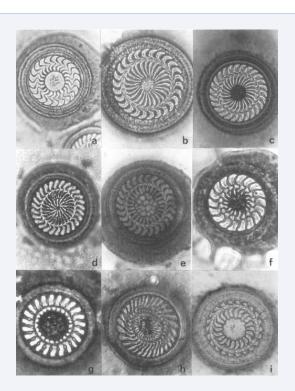
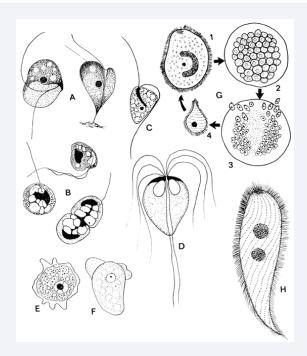


Figure 3 Ectoparasitic Protozoa: Trichodinids. a,b. Trichodina heterodentata: a, live, carp, Israel; silver impregnated, O. aureus x niloticus, Israel. c. Scanning electron microscopic (SEM) view of T. Reticulata of goldfish. d,e. SEM view of O. aureus x niloticus gill infection with Tripartiella cichlidarum; f. same as d, in histology. Source: FAO [11].



**Figure 4** Ectoparasitic and intestinal protozoa of fish. **A.** *lchthyobodo necator* free (left) and attached (10–15 μm long axis). **B.** *lchthyobodo* sp. from *Aplocheilichthys gambianus* from South Ghana (10 μm long axis). **C.** *Cryptobia* (length 6–8 μm). **D.** *Hexamita* sp. from tilapia hybrid gut (7–12 μm). **E.** *Thecamoeba* (40-μm diam.) **F.** *Entameoba* (15-μm diam.). **G.** Life cycle of *lchthyophthirius multifiliis*: **1.** Trophont; **2.** Dividing tomont; **3.** End of division – tomites (theronts) escape from the cyst residues; **4.** Tomite (theront). **H.** *Protoopalina* (150–350 μm long). Source: FAO [11]

Trichodinids were readily eradicated with treatment by 25 ppm, while elimination of *Chilodonella* was achieved after treatment with 40–50 ppm. Van As et al. [62], also demonstrated differential efficacy with the type of fish treated, e.g. 25 ppm per 24 hrs was effective in cleaning infected carp, while with tilapia fry it has been achieved with 45 ppm per 24 hrs [11].

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