ECTOPARASITIC PROTOZOA (FLAGELLATES AND CILIATES)

Species affected

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.

Description taxonomy and diagnosis

Integumental ectoprotozoan genera are readily differentiated while diagnosis of species is difficult and often requires special staining.Most ectoparasitic forms are readily detected in direct microscopic examination of skin and gill scrapings from live (or freshly killed) fish.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% ethanols, brought to water, stained in a haematoxylin stain and mounted after dehydration.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.

Flagellates (Mastigophora, Kinetoplastida):

Cryptobia	free, spindle shaped, 10–30 × 3–5 µm in size (if *C. branchialis*), 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 × 20–50 µm, with 6–8 ciliary lines on each side.

C. piscicola (syn: *C. cyprini*) size 33–100 × 24–60 µm, 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.

Small trichodinids, predominantly from the gills, are bell shaped and often settle on the tips of the gill lamellae. In *Trichodinella*, the ray (the inner extension of the denticle), is totally reduced.

Genera of sessile peritrichs are differentiated by their macronuclei and scopula (attachment leg): *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; *Epistilis* cup-shaped with horseshoe-shaped macronucleus. Some *Apiosoma* also develop on stalks.

Suctoria: (*Trichophyra* and other genera) — cilia lacking, variable numbers of tentacles arise from the rounded body.

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. Both ectoparasitic flagellates, *I. necator* and *Cryptobia* spp., occur either free swimming or attached to the integument, the former through a cytoplasmic protrusion and the latter by attachment with the flagellum.

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 etached buds are ciliated. As the buds become attached to a new location on the piscine integument, cilia are shed and tentacles appear.

Water temperatures do not seem to be an important parameter, in spite of reports of low temperatures being more optimal for reproduction of *Chilodonellapiscicola* and some trichodinids. Massive infections with *I. necator*, both speciesof*Chilodonella*and the ubiquitous trichodinids and sessile species, occur in low (12–17°C) and high (25–30°C) ambient temperatures in southern Africa. 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.

A number of ciliates (species of *Tetrahymena, Ophryoglena, Glaucoma, Colpidium* and others) are facultative parasites, or opportunists which will colonise fish in special circumstances, most often when fish are stressed or traumatised. All others mentioned above are obligatory parasites which will apparently survive for only a limited time outside their hosts. Non-parasitic sessile peritrichs are different species from those colonising living organisms. Trichodinids and sessile species found on aquatic invertebrates comprise different species from those infecting fish.

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).

Among the latter, trichodinids with seemingly related morphological characteristics (e.g. *pediculus*-like, *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. 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* occurs mainly in goldfish, *T. kazubski* has been found in South African *Barbus* spp. and *T. nobilis* and *T. kupermani* mainly in asian carp.

Pathology

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.

Ichthyobodo necator attaches itself to epithelial cells and through an inserted protrusion consumes their contents, whereas *Chilodonella* spp. browse the epithelial surface. Histopathological changes in the integument 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. Secondary cellular damage due to degeneration, necrosis and desquamation results in the degradation and disintegration of the epithelial layer.

Cryptobia attachment through the flagellum does not induce any pathological or even ultrastructural cellular damage, contrary to reports of morbidities associated with this parasite.

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, and the sessilians *Apiosoma, Ambyphrya* and *Scopulata*, histopathological changes in events of massive infections by these ectoprotozoans are hardly evident, if occurring at all. *Trichodinella epizootica* in carp and *Tripartiella cichlidarum* in cichlids cause some erosion of the gill epithelium. However, food vacuoles of trichodinids revealed bacteria rather than sloughed cells. Ultrastructural observation on attached *Apiosoma* did not reveal any interference with the host cell serving as substrate or peripheral tissue response. Thus, mortalities following massive colonisation of gills by sessilians 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 haemorrhagic lesions are also contaminated with the bacterium *Aeromonas hydrophila*.

Suctorians (*Trichophyra* spp.) in certain instances cause cytological damage to the gill lamellae cells in direct contact with the parasites and subsequent hyperplasia and haemorrhages of the gill tissue.

Epizootiology (Disease occurrence)

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.

Reduced mobility facilitates parasite colonisation as well as moderating loss through detachment and drift from the integumental surface.

Defense mechanisms other than epithelial hyperplasia, and specific immune responses to integumental ectoparasites need more investigation, 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 naive or immunosuppressed.

Heavy infections by ectoparasitic protozoans are mainly found in young fish 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.

Level of infection in the fry sharply declined as fish gained in size. 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*, *T. heterodentata*, and species of *Ambyphrya* and *Scopularia*) being gradually replaced by species specific to cichlids (*Tripartiella* spp. *Trichodina centrostrigata* and species of *Apiosoma*).

Control

Treatment with formalin is still the effective means to control massive ectoparasitic infections in all warm water cultured fish species. Essential oils extracted from different plants have promising effect (garlic, moringa, etc………..).

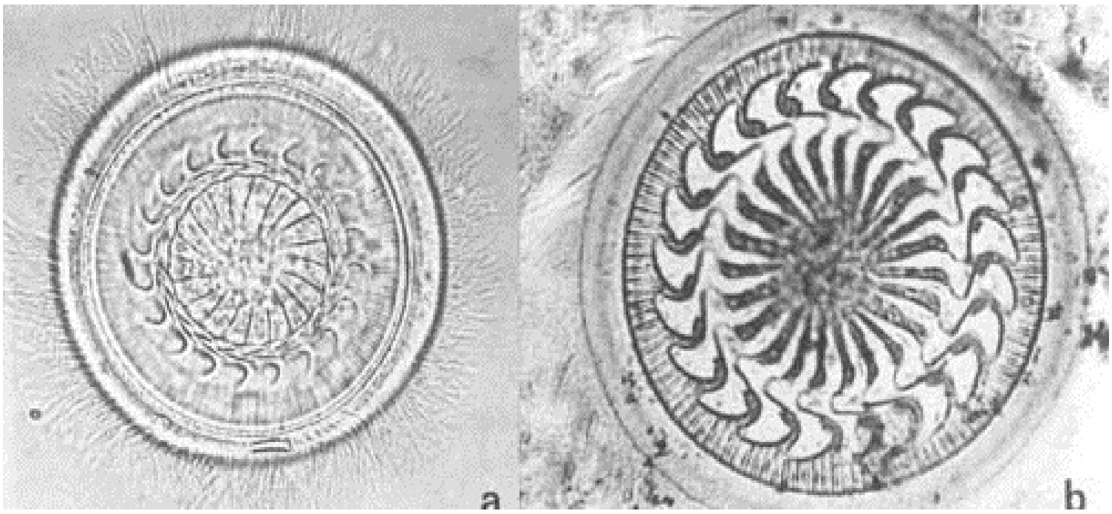

Fig. 1. *Trichodina* species

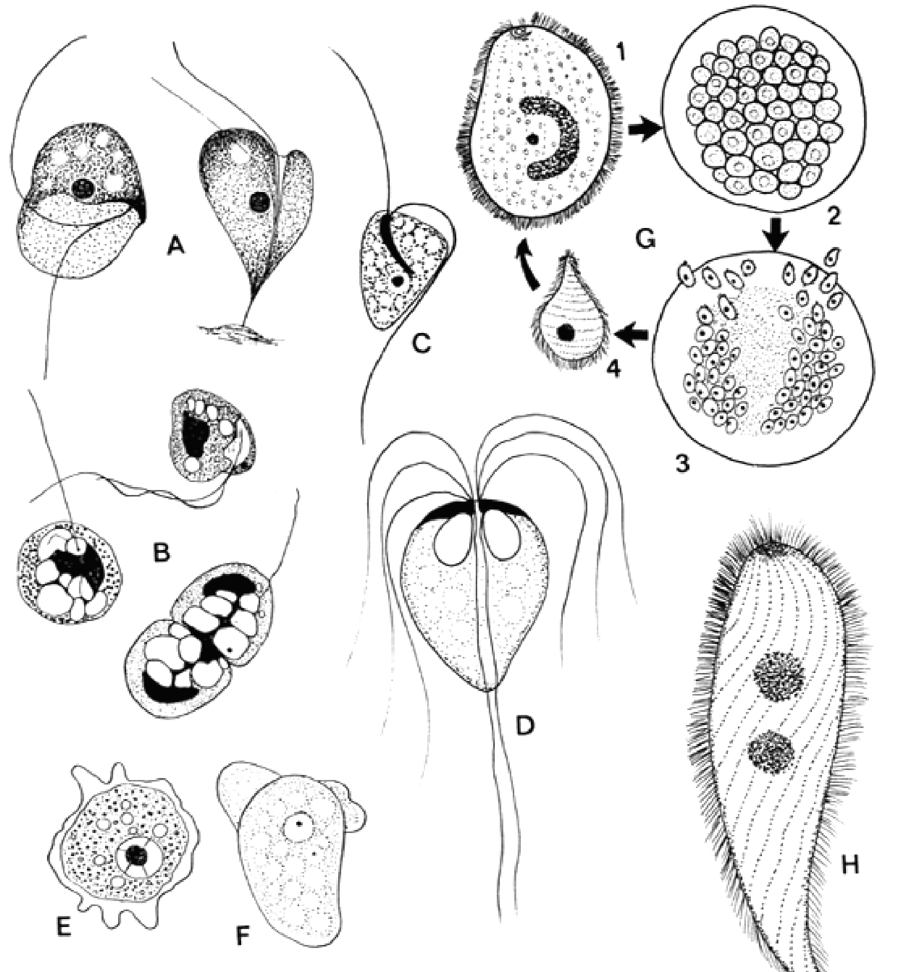

Fig. 2. Ectoparasitic and intestinal protozoa. **A.** *Ichthyobodonecator* free (left) and attached (10–15 µm long axis). **B.** *Ichthyobodo* sp. from *Aplocheilichthys gambianus* from South Ghana (10 µm long axis). **C.** *Cryptobia* (length 6–8 µm). **D.** *Hexamita* sp. from tilapia hybrid gut (7–12 µm). **E.** *Thecamoeba* (40 µm diam.) **F.** *Entameoba* (15 µm diam.). **G.** Life cycle of *Ichthyophthiriu smultifiliis*: **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).