Amyloodinium, Crepidoodinium, Piscinoodinium
(protist: flagellate)

Overview

Protists are single-celled organisms with membrane-bound nuclei (eukaryotes). One protistan supergroup known as SAR comprises the Stramenopiles (with heterokont flagella), Alveolata (with cortical alveoli) and Rhizaria (with fine pseudopodia). Three major alveolate groups are recognized: ciliates, apicomplexans and dinoflagellates. Flagellates are protists that swim using one or more flagella (undulipodia); each arising from a small centriole (basal body, kinetosome) and having a microtubular axoneme core (2+9 configuration). Phytoflagellates possess chloroplasts, which require sunlight to function, so most species are free-living aquatic organisms. Dinoflagellates have a distinctive haploid nucleus (dinokaryon) where the chromosomes remain condensed during interphase and are evident as beaded strands due to low levels of histones and associated proteins (condition termed 'mesokaryotic'). Many dinoflagellates form characteristic free-swimming dinospores (thecate and/or pigmented) and various species have been associated with coloured tides/blooms, bioluminescence and even neurotoxicity in mammals (paralytic shellfish poisoning). Some species, however, are heterotrophic and parasitic in marine animals; including the blastodinids which form uninucleate trophonts (with chloroplasts) attached to the skin of fish where they may cause surface lesions (such as velvet disease).

Classification:
Domain: Eukaryota (membrane-bound nucleus)
Supergroup: SAR (Stramenopiles + Alveolata + Rhizaria)
Group: Alveolata (with cortical alveoli)
Phylum: Dinoflagellata (with unique mesokaryotic nuclei (lacking histones), autotrophs and heterotrophs)
Class: Blastodiniophyceae (extracellular athecate parasites of zooplankton, algae, crustacea and fish)
Order: Blastodiniales (uninucleate trophonts with chloroplasts)
Family: Oodiniaceae (trophont fusiform with rhizoid-like invasive organelle)
Genus: *Amyloodinium* (parasitic on skin of fish)
Genus: *Crepidoodinium* (parasitic on skin of fish)
Genus: *Piscinoodinium* (parasitic on skin of fish)
Species: various species cause lesions in fish

Parasite biodiversity and host range: Protists are unicellular eukaryotes that move using undulipodia (flagella or cilia), pseudopodia (false-feet) or a unique gliding motion. Flagellated species have one or more flagella with an internal microtubular core (in a characteristic 2+9 configuration comprising 2 single central microtubules and 9 peripheral doublets) anchored to a submembranous protein structure (known variously as a centriole, basal body, kinetosome or blepharoplast). Many types of flagellated cells have been described and recent phylogenetic studies have classified them into several disparate groups: including the metamonads (amitochondriate flagellates), heteroloboseans (amoeboflagellates), euglenozoans (euglenids and kinetoplastids), stramenopiles (heterokonts), alveolates (dinoflagellates) and cercozoans (biflagellates). While most flagellated protists are free-living organisms swimming and feeding in aquatic environments, representatives of several groups have developed symbiotic relationships with various hosts; some being endoparasitic in vertebrates (notably anaerobic metamonads in tubular organs, and heterotrophic euglenozoans occurring in blood or tissues), and some being parasitic in invertebrates (alveolates in crustacean tissues) (representatives tabulated below).

Higher taxonomy	Class or order	Family	Genera	Hosts (tissues)	Trans-mission*
Supergroup: SAR (Stramenopiles + Alveolata + Rhizaria) (3 groups unified by molecular studies)					
Group: Alveolata (with cortical alveoli)					
Phylum: Dinoflagellata (with unique mesokaryotic nuclei)	Order: Blastodiniales (uninucleate trophonts with chloroplasts)	Oodiniaceae (trophont with rhizoid-like invasive organelle)	*Amyloodinium* *Crepidoodinium* *Piscinoodinium*	fish (skin)	direct (w)
	Order: Syndiniales (multinucleate plasmodial trophonts)	Syndiniaceae (without chloroplasts)	*Haematodinium* *Ichthyodinium*	crustaceans, fish (tissues)	direct (w)
Phylum: Perkinsozoa (parasitic)	Order: Perkinsorida (released trophonts form biflagellated zoospores)	Perkinsidae (incomplete conoid)	*Perkinsus*	gastropods, bivalves (tissues)	direct (w)

Supergroup: Excavata (with conspicuous ventral feeding groove)					
Group: Metamonad (amitochondriate flagellates with karyomastigonts)					
Phylum: Fornicata (diplomonads)	Order: Diplomonadida (1-2 karyomastigonts)	Hexamitidae (2 karyomastigonts with binary axial symmetry)	*Giardia*	vertebrates (gut)	direct (f-o)
			Hexamita *Spironucleus*	vertebrates (tissues)	direct (f-o, w)
Phylum: Parabasalia (with parabasal body)	Order: Trichomonadida (3-5 anterior flagella plus recurrent flagellum)	Monocercomonadidae (costa absent, most without undulating membrane)	*Histomonas*	birds (gut, liver)	direct (f-o)
			Dientamoeba	vertebrates (gut)	direct (f-o)
		Trichomonadidae (stout axostyle, costa, undulating membrane)	*Trichomonas*	vertebrates (urogenital tract, gut)	direct (f-o, v)
		Cochlosomatidae (anterior adhesive disc)	*Cochlosoma*	birds (gut)	direct (f-o)
Group: Discoba (diverse group supported robustly by molecular studies)					
Phylum: Euglenozoa (flagella inserted in anterior pocket, heterotrophs, autotrophs)	Class: Kinetoplastea (heterotrophs, with extranuclear DNA (= kinetoplast) associated with mitochondrion)	Ichthyobodonidae (flagellar pocket continues as groove)	*Ichthyobodo* (= *Costia*)	fish (gills, skin)	direct (w)
		Parabodonidae (epizoic or endozoic)	*Cryptobia*	fish (gills, skin)	direct (w)
			Trypanoplasma	fish (blood)	indirect (v-b)
		Trypanosomatidae (monogenetic forms in insects/plants, digenetic forms in vertebrates & arthropods)	*Trypanosoma*	vertebrates (blood, tissues)	indirect (v-b)
			Leishmania	vertebrates (blood, tissues)	indirect (v-b)

*f-o = faecal-oral transmission; v-b = vector-borne transmission, w = water-borne transmission; v = venereal transmission

The group Alveolata in the supergroup SAR is characterized by the possession of cortical membranous alveoli (vesicles or sacs) underlying and supporting the cell wall, and includes many otherwise disparate groups comprising 3 large assemblages (Apicomplexa, Ciliophora, Dinoflagellata) and 6 smaller taxa (Acavomonidia, Chromerida, Colpodellida, Colponemidia, Perkinsozoa and Voromonadida). The phylum Dinoflagellata comprises a diverse group of aquatic organisms, with many free-living species being important in food chains either as producers (autotrophs) or consumers (heterotrophs), some being endosymbiotic in invertebrates (e.g. in photosynthetic corals) and some being parasitic mainly in invertebrates but also a few vertebrates (fish). Dinoflagellates have a distinctive haploid nucleus (dinokaryon) where the chromosomes remain condensed during interphase and are evident as beaded strands due to low levels of histones and associated proteins (condition termed 'mesokaryotic'). Many dinoflagellates contain chloroplasts so early classification systems have used either botanical or zoological nomenclature, whereas recent molecular phylogenetic studies on eukaryotes have recognized and characterized cognate groups and provided appropriate names. Trophonts (or dinospores) of most species have an unequal heterodynamic pair of flagella; one ribbon-like and lying in equatorial furrow (girdle or cingulum) and the other directed posteriorly and often lying in a longitudinal ventral furrow (sulcus). Many species have thecate dinospores encased in armour composed of cellulosic plates; the anterior section is termed the epitheca (epicone in unarmoured species) and the posterior section is called the hypotheca (hypocone). Most species have unique membranous cytoplasmic organelles called pusules which are thought to be associated with osmoregulatory or excretory function. Many species have coloured chloroplasts and/or coloured pigments, some even exhibiting bioluminescence when disturbed by wind, wave or boat wakes. Several pigmented species bloom under suitable environmental conditions and cause 'red tides' and some pigments are neurotoxic to mammals when concentrated in the tissues of fish or filter-feeding shellfish (causing PSP 'paralytic shellfish poisoning'). Exemplars of toxic dinoflagellates are tabulated below (for interest only):

Toxic dinoflagellates	**Toxin**	**Transvector**	**Effect**
Prorocentrum	venerupin	shellfish	human hepatotoxicity
Dinophysis	okadaic acid	shellfish	human enterotoxicity
Pyrodinium	saxitoxin	shellfish	human neurotoxicity (PSP)
Protogonyaulax	saxitoxin	shellfish	human neurotoxicity (PSP)
Gymnodinium	saxitoxin	shellfish	human neurotoxicity (PSP) & fish kills
Ptychodiscus	brevetoxin	shellfish	human neurotoxicity & fish kills
Peridinium	glenodinine	-	fish kills
Gambierdiscus	ciguatoxin	fish	ciguatera

Over 4,000 dinoflagellate species are recognized in 550 genera: the majority being free-living autotrophs in pelagic or neritic surface waters. However, some 140 species from 5 families are heterotrophic and parasitic in aquatic hosts: including Oodinidae (e.g. *Amyloodinium, Piscinoodinium* and *Crepidoodinium* on skin and gills of fish); Syndinidae (e.g. *Ichthyodinium* and *Hematodinium* in tissues of fish and crabs); Chytriodinidae (e.g. *Chytriodinium* on eggs of copepods and shrimps); Paradinidae (e.g. *Paradinium* in body cavities of copepods); and Ellobiopsidae (e.g. *Ellobiopsis* and *Thalassomyces* on exoskeletons of crustacea). Genera parasitic on fish and in shellfish that are covered in this document are tabulated below.

Taxon	Genus	Hosts	Site	Transmission
Class: Blastodiniophyceae† (uninucleate trophonts, often with chloroplasts)				
Order: Blastodiniales† (ectoparasitic on zooplankton, algae, crustacea and fish)				
Oodinidae (trophonts with rhizoid-like holdfast organelles, gymnospores)	*Amyloodinium*	fish	skin	direct (water)
	Crepidoodinium	fish	skin	direct (water)
	Piscinoodinium	fish	skin	direct (water)
Class: Syndiniophyceae (multinucleate trophonts, without chloroplasts)				
Order: Syndiniales (endoparasitic in copepods, appendicularians, crabs, radiolaria and fish eggs)				
Syndinidae (plasmodial trophonts, dinospores not of *Gymnodinium* type)	*Hematodinium*	decapods	tissues	direct (water)
	Ichthyodinium	fish	tissues	direct (water)

†In recent classifications, these names are no longer valid, and the genera are considered *incertae sedis* in the Dinoflagellata.

Ectoparasitic oodinid dinoflagellates found on fish were often first identified as *Oodinium* spp., but this genus has now been reserved for species found on tunicates and polychaetes which form sac-like trophonts without rhizoid-like attachment organelles, stomopodes, chloroplasts or starch granules. Those species infecting fish have subsequently been transferred to new genera (*Amyloodinium*, *Piscinoodinium* and *Crepidoodinium*) which all form elongate trophonts with elaborate rhizoid-like holdfast organelles attached to host cells, but differ in their host range, tissue tropism, pathogenicity, trophont size, rhizoid type, and vary in their possession of peduncles, stomopodes, chloroplasts, alveolar plates, tomont (palmella) cyst walls, dinospore (gymnospore) size and stigma (eyespots): tabulated as follows:

Character	Dinoflagellate genera ectoparasitic on fish		
	Amyloodinium	***Piscinoodinium***	***Crepidoodinium***
Hosts	marine fish	freshwater fish	marine/estuarine fish
Site of infection	skin, gills	skin, gills	gills
Trophont shape	pyriform	pyriform	fusiform
Trophont size	up to 150 µm	80-160 µm	up to 670-820 µm
Peduncle (stalk)	short	short	absent
Holdfast projections	long filiform	rod-like	finger-like
Attachment to host cells	penetrate cells	penetrate cells	do not penetrate cells
Tentacle-like stomopode	present	absent	absent
Chloroplasts	absent	present	present
Alveoli	with plates	without plates	without plates
Cytoplasm	not spongy	not spongy	spongy
Starch grains	present	present	present
Tomont (palmella) division	with common cyst wall	without common envelope	?
Number of dinospores	up to 256	up to 256	up to 2048
Dinospore stigma (eyespot)	present	absent	absent

Infections by *Amyloodinium* and *Piscinoodinium* species may cause superficial discolourations of fish skin (condition known as velvet disease) with attached trophonts leading to host cell destruction and superficial lesions. Infections occur in marine and freshwater fish respectively, particularly in species used in aquaculture and aquarium industries. Infections by *Crepidoodinium* species are less invasive and confined to the gills of several marine and estuarine fish species, although their presence may possibly interfere with respiratory function.

Parasite species	Hosts	Location (disease)	Gymnospores	Distribution
Amyloodinium ocellatum (syn. *Oodinium*)	marine and brackish-water fish – Acanthuriformes: acanthurid (doctorfish tang, Atlantic blue tang), sciaenid (silver croaker,	gills, eyes, skin, fins, sometimes internal organs	10-15 x 8-15 µm	cosmopolitan

	spot croaker, Atlantic croaker, southern kingcroaker, northern kingcroaker, sand seatrout, spotted seatrout, squeteague, red rum, banded drum, striped drum, cubbyu, meagre); Anguilliformes: ophichthid (speckled worm eel, shrimp eel); Aulopiformes: synodontid (inshore lizardfish); Batrachoidiformes: batrachoidid (Gulf toadfish, midshipman fish); Blenniformes: blenniid (striped blenny, feather blenny, freckled blenny); Carangiformes: carangid (blue runner, crevalle jack, horse-eye jack, leather jack, Atlantic bumper, pilot fish, Florida pompano, permit, greater yellowtail); Cichliformes: cichlid (Mozambique tilapia); Clupeiformes: dorosomatid (scaled sardine), engraulid (bay anchovy); Cyprinodontiformes: fundulid (saltmarsh topminnow), poeciliid (western mosquitofish); Gadiformes: morid (southern codling); Gobiesociformes: gobiesocid (skilletfish); Gobiiformes: eleotrid (spiny-cheek sleeper), gobiid (frillfin goby, naked goby, code goby, clown goby, pink wormfish), oxudercid (violet goby); Holocentriformes: holocentrid (squirrelfish); Kurtiformes: apogonid (flamefish); Labriformes: labrid (bluehead wrasse), scarid (blue parrotfish); Moroniformes: ephippid (Atlantic spadefish), moronid (striped bass, European seabass); Mugiliformes: mugilid (flathead grey mullet); Myliobatiformes: dasyatid (Atlantic stingray); Perciformes: centrarchid (bluegill), chaetodontid (foureye butterflyfish), epinephelid (rock hind, red grouper, snowy grouper), gerreid (silver mojarra), haemulid (porkfish, redmouth grunt, grey grunt, banana grunt, Spanish grunt, white grunt, blue-striped grunt), latid (barramundi), lobotid (Atlantic tripletail), lutjanid (mutton snapper, schoolmaster snapper, northern red snapper, grey snapper, dog snapper, lane snapper, yellowtail snapper), pomacanthid (gray angelfish, French angelfish), pomacentrid (sergeant major, marine clownfish), pomatomid (bluefish), serranid (rock sea bass, black sea bass, graysby, pygmy sea bass, belted sandfish, white-spotted soapfish, greater soapfish), sparid (gilthead sea bream, white sea bream, two-banded sea bream, sharpsnout sea bream, sheepshead, pinfish, porgy); Pleuronectiformes: achirid (lined sole, hogchoker), cynoglossid (blackcheek tonguefish), paralichthyid (bay whiff, fringed flounder, southern flounder), soleid (Senegalese sole); Scombriformes: stromateid (silver pomfret); Scorpaeniformes: scorpaenid (barbfish), triglid (northern searobin, striped searobin, bluespotted searobin, bighead searobin); Siluriformes: ariid (hardhead catfish, gafftopsail catfish); Syngnathiformes: syngnathid (lined seahorse, chain pipefish);	(velvet disease)		

	Tetraodontiformes: balistid (green triggerfish), diodontid (striped burrfish, spotfin porcupinefish), monacanthid (orange filefish, plainhead filefish), ostraciid (scrawled cowfish, buffalo trunkfish), tetraodontid (northern puffer, least puffer, Caribbean sharpnose puffer)			
Piscinoodinium pillulare (syn. *Oodinium pillularis, O. limneticum*)	tropical freshwater aquaria fish - Anabantiformes: osphronemid (Siamese fighting fish, paradise fish); Characiformes: characid (tetras); Cypriniformes: cyprinid (goldfish, zebrafish, barbs, danios, Crucian carp, common carp, sunbleak, tench), Cyprinodontiformes: poeciliid (guppies, mollies, platys, swordtails); Urodela: ambystomatid (axolotyl larvae); Anura: ranid (tadpoles of European common brown frogs and moor frogs)	gills, skin, fins, nasal cavity, eye orbit (velvet disease)	10-19 x 7-12 µm	cosmopolitan (aquaria, cultures)
Crepidoodinium cyprinodontum	estuarine Cyprinodontiformes: cyprinodontid (sheepshead minnow), fundulid (mummichog, spotfin killifish, striped killifish, rainwater killifish)	gills	5-9 x 2-5 µm	North America
Crepidoodinium australe	estuarine Perciformes: sillaginid (sand whiting)	gills	17 x 12 µm	Australia

Parasite morphology: The dinoflagellates *Amyloodinium*, *Piscinoodinium* and *Crepidoodinium* spp. each form 3 different types of morphological stages during their development: trophonts, tomonts and spores (each stage having acquired a variety of names over history). The trophonts (sometimes known as trophozoites) attach to host cells, beginning as small ovoid stages around 20-30 µm in diameter but then growing to becoming elongate fusiform to pyriform stages varying in size from 80-820 x 50-80 µm according to species: *Amyloodinium* ranging up to 150 µm, *Piscinoodinium* from 80-160 µm, and *Crepidoodinium* up to 670-820 µm. The trophonts are enclosed in cellulosic walls that are often rugose (with longitudinal furrows) and usually projecting perpendicularly from the host cell surface with the attached end narrowed slightly, and in the case of *Amyloodinium* and *Piscinoodinium* containing a short stalk (peduncle). They vary in colour from green-brown due to the presence of chloroplasts (in *Piscinoodinium* and *Crepidoodinium*) or remain colourless (*Amyloodinium* without chloroplasts). It should be noted that some *Piscinoodinium* isolates apparently lacking chloroplasts had elaborate membranous organelles associated with starch granules in a manner similar to that of chloroplast thylakoids, suggesting they may be degenerated chloroplasts. The trophonts were sessile and attached to the surface of host epithelial cells by discoid holdfast organelles with several elongate anchor-like projections (called rhizoids). *Amyloodinium* and *Piscinoodinium* have filiform or rod-like projections that penetrate host cells, while *Crepidoodinium* have finger-like projections that do not penetrate host cells. *Amyloodinium* also had a special tentacle-like stomopode extending basally which appears to be involved in feeding. All trophonts had well developed pusules (non-contractile vacuoles) involved in osmoregulation and/or excretion. Mature trophonts detach from the host and become spherical as they encyst to form tomont (palmella) stages which are reproductive stages as they undergo a series of internal divisions (palintomy) either within a common cystic envelope (*Amyloodinium*) or the old envelope dissolves after each division (*Piscinoodinium*). The tomonts grow up to 100-200 µm in size and they produce and release numerous free-swimming spores (known as swarmers, zoospores or dinospores which resemble those of the free-living genus *Gymnodinium,* hence the frequent use of the name gymnospores). Tomonts of *Amyloodinium* and *Piscinoodinium* may release up to 256 spores, whereas those of *Crepidoodinium* may release up to 2,048 spores. The spores have an armoured cellulosic wall and are ovoid with a somewhat flattened hamburger-shape ranging in size from 5-19 x 2-15 µm. They are biflagellated with 2 flagella: one transverse flagellum lying in an equatorial furrow (girdle or cingulum); and one longitudinal flagellum directed posteriorly and lying in a ventral furrow (sulcus). The spores often have small ventral pseudopodial projection that is used for host attachment, and the spores of *Amyloodinium* also possess a dark-coloured eyespot (stigma).

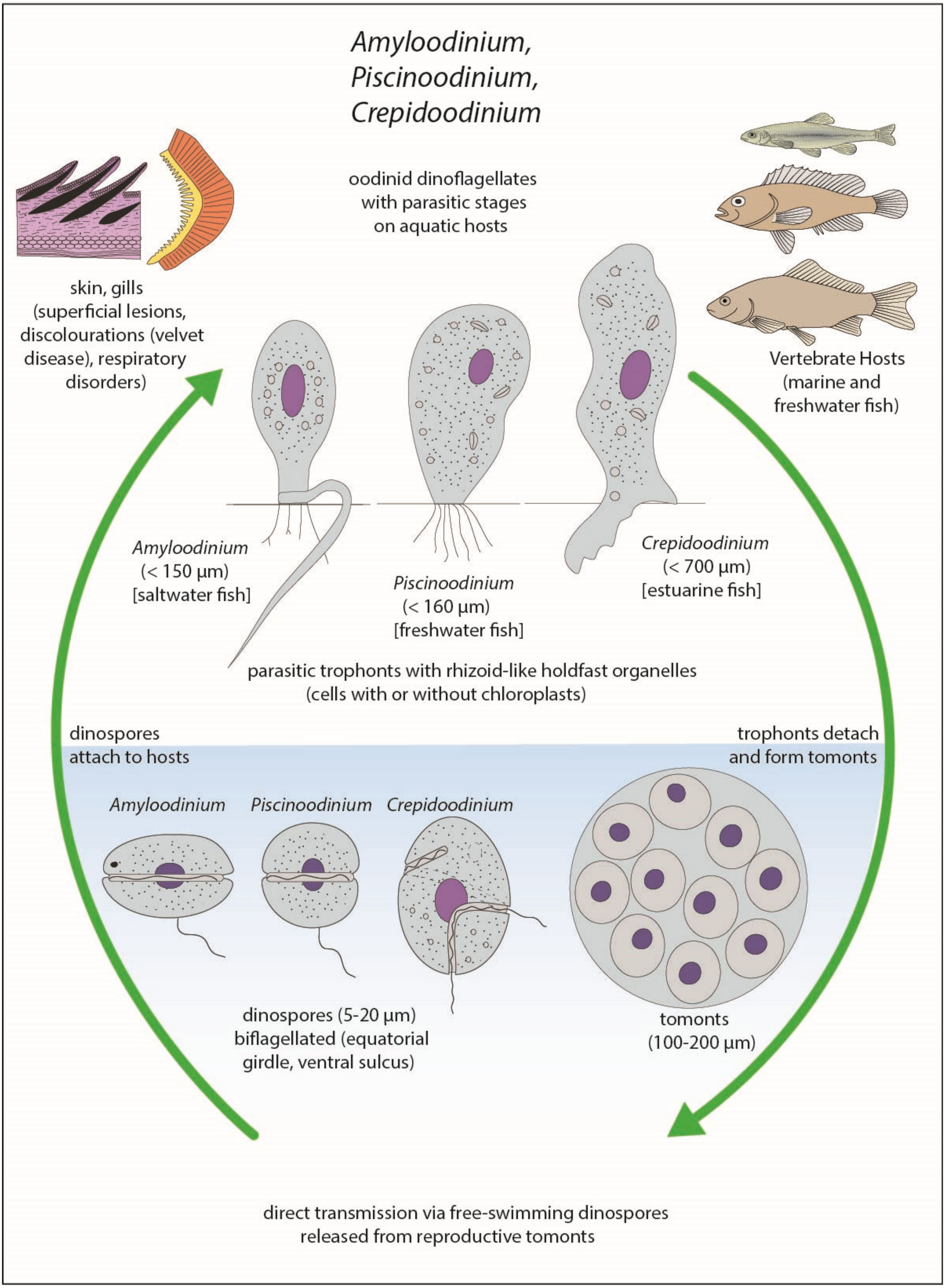
Site of infection: *Amyloodinium* and *Piscinoodinium* have ectoparasitic trophonts with rhizoid projections penetrating host epithelial cells, commonly on the gills, skin or fins of fish, but sometimes on the eyes and oropharyngeal cavity. *Amyloodinium* is cosmopolitan in brackish and seawater environments in temperate and tropical areas and has a broad host range, being found on some 123 fish species (the only dinoflagellate capable of infecting teleosts and elasmobranchs). *Piscinoodinium* is found in freshwater environments and has been recorded on almost 20 fish species, particularly small species involved in the aquarium trade. In contrast, trophonts of *Crepidoodinium* spp. do not have projections penetrating host cells but attach themselves to gill epithelia in some 6 species of estuarine fishes.

Pathogenesis: Infections by ectoparasitic trophonts may cause a skin condition known as velvet disease (sometimes called gold dust or rust disease). *Amyloodinium* causes marine velvet (amyloodinosis, previously known as oodiniosis) and *Piscinoodinium* causes freshwater velvet (piscinoodiniosis). The skin of infected fishes takes on a cream, gold or rust-coloured dusty appearance that varies in intensity with the density of attached trophonts. Skin discolouration may develop over several days to weeks with patches progressing to more dense films which may eventually present as a contiguous finely granular layer over the whole body and fins. Although attached trophonts are essentially sessile, they constantly twist and turn as their rhizoid projections penetrate epithelia slowly damaging and destroying cells causing focal lesions. Infections elicit moderate-to-intense tissue reactions with inflammation, haemorrhages, epithelial hyperplasia and necrosis. The gills may become swollen, oedematous and have excessive mucus production making breathing difficult. Infected fish experience discomfort and often show behavioural changes, such as rubbing against solid objects with jerky ('flashing') movements, clamping the fins against the body, and dyspnoea with rapid laboured breathing and gathering at the water surface gasping for air. In heavy advanced infections, the skin may begin to peel off and fish become lethargic, anorexic, lose weight and die. The causes of death may involve various combinations of respiratory insufficiency, metabolic depletion, osmoregulatory impairment, and secondary microbial infections due to epithelial disruptions. Velvet is highly contagious and thrives on fish stressed by captivity or other factors (especially poor water quality due to over-feeding, over-crowding or poor oxygenation). Infections constitute serious problems for both aquaculture and mariculture enterprises, particularly lagoon-based valliculture and inland brackish-water farming where shallow beds and poor water circulation allows parasites to proliferate over the warmer months. Infections may occur in all fish life stages, although there is some evidence to suggest that some fish may develop protective immunity after repeated or protracted exposure. Infections by *Crepidoodinium* spp. are confined to the gills of estuarine fish but the trophonts attached to gill filaments do not have rhizoid projections that penetrate and destroy host cells. Most infections are benign, and the organism is often considered to be a symphoriont rather than a parasite. However, further studies are required to determine whether heavy or persistent infections cause or predispose to gill lesions that may compromise respiratory function.

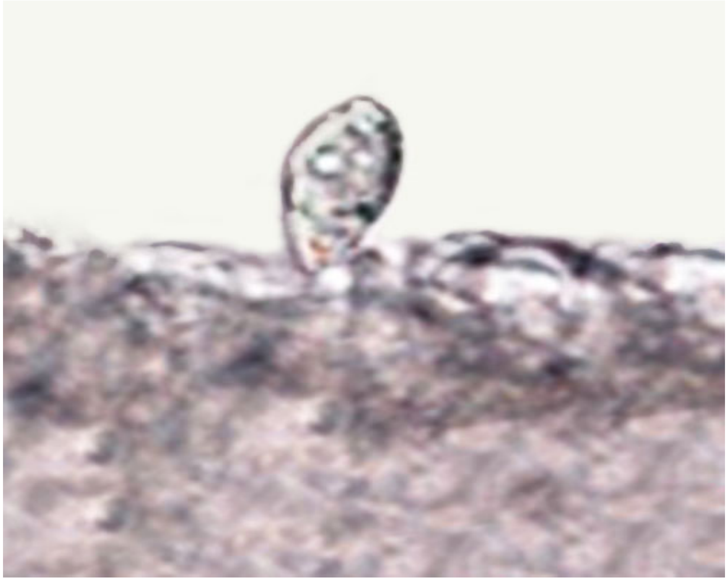
Developmental cycle and mode of transmission: These ectoparasitic dinoflagellates have similar life-cycles, involving attached feeding trophonts, detached reproductive tomonts, and free-swimming planktonic dinospores. Fish become infected when biflagellated dinospores in the water column attach themselves to epithelial surfaces using ventral pseudopodial projections. Once attached, they transform into sac-like trophonts with elaborate holdfast organelles which are disc-like with rhizoid projections attached to the cell surface or penetrating the host cell. The trophonts feed and grow, maturing in 2-6 days. When mature, the trophonts detach from the host and drop to the benthos where they round up transforming into ovoid tomonts. These stages are encysted and surrounded by a cellulosic wall, which thickens and provides good protection against unfavourable environmental conditions. The tomonts are reproductive stages and they undergo a series of asexual divisions by repeated binary fission (first division being longitudinal, and successive divisions occurring regularly and perpendicular to each other). Division may occur within a common cystic envelope, or the old envelope may dissolve after each division. In 2-4 days, tomonts produce numerous dinospores (up to 256 or more) which are released through tears or ruptures in the tomont wall. The dinospores are motile biflagellated dispersive stages which swim around in the water column. Transmission is direct and facilitated by the dinospores which are infective to susceptible fish for several days but will die if they do not find a host in that time. Once attached to a new host, the dinospore transforms into a trophont within 5-20 minutes. The duration of the life-cycle is temperature dependent, as is trophont size and tomont fecundity, and can range from 3-20 days. Parasite prevalence and abundance is greatest following rapid exponential growth in the warm months over summer.

Differential diagnosis: Infections may be suspected when fish exhibit flashing or gaping behaviours by rubbing against solid objects or struggling to breathe. Diagnoses are generally made by the visual detection of trophonts attached to the skin surface, best conducted using oblique light sources in darkened conditions. Green-brown parasites may be seen imparting a metallic yellow to velvet dusty sheen to the skin (and not as discrete white spots like those formed by ectoparasitic ciliates). Small fish may be anaesthetized and examined directly at low power under a dissecting microscope (the gills can also be examined by gently lifting opercular structures). Alternatively, skin scrapings and gill filament snips can be collected as biopsy or necropsy samples and wet mounts examined at higher powers under a compound microscope (often using Lugol's iodine to stain the starch-containing trophonts a dark brown to black colour). It is recommended that necropsy samples be collected and examined as soon as practicable, as trophonts usually detach shortly after host death. Several research studies have developed immunological tests (enzyme-linked immunosorbent assays) to detect specific host antibodies against parasite antigens, and elevated antibody titres have been associated with the development of some protective immunity. Molecular biological techniques have also been used to detect dinoflagellates in water and tissue samples by polymerase chain reaction (PCR) amplification of nuclear gene sequences (especially ribosomal RNA genes).

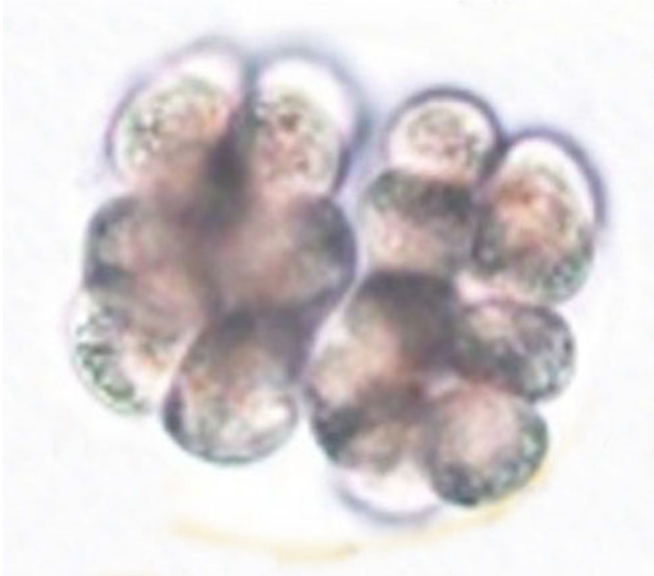
Treatment and control: Infections by ectoparasitic dinoflagellates have been successfully controlled in closed culture systems (including aquaria) by water treatment (disinfection and/or filtration) and by manipulating water quality (chemical/physical characteristics). The life-cycle stages most affected by chemical treatment are the free-living dinospores, while attached trophonts and encysted tomonts are more resistant to treatment. A range of antiprotozoal and algacidal chemicals have proven effective; including acriflavine (or trypaflavine), chloroquine, formalin, hydrogen peroxide, copper sulphate and a large number of broad-spectrum cocktail treatments are commercially available (involving mixtures of formaldehyde, cupramine, sulfathiazole, nitrofurantoin, nitrofural, quinacrine dihydrochloride, malachite green, and copper sulphate). It is important to treat infections as soon as practicable (to counter exponential growth), and to increase water aeration during treatment (to assist respiration in the case of gill infections). Fish with open sores may also benefit from antibiotic treatment for secondary bacterial infections. Caution should be exercised as some chemicals can be toxic to certain fish species (especially loaches, pufferfish, mormyrids, stingrays, and catfish) and many are lethal to aquatic invertebrates (including shrimp and snails). Studies have shown that alterations in water temperature, salinity and light levels for several days-weeks may help control infections, provided resident fish can tolerate such changes. Raising the water temperature to 29°C has been shown to speed up the parasite life-cycle, adding salt to freshwater aquaria or subjecting marine species to freshwater baths may help combat infections, and covering tanks to exclude ambient light stops parasite species with chloroplasts from accessing alternate photosynthetic pathways. When all else fails, recourse can be taken to removing all fish from aquaria, tanks or ponds for at least 7 days after which time all dinoflagellates will be dead. Other management strategies designed to prevent the spread on infections include regular screening, isolation and quarantine of new stock (particularly introduced wild fish), avoiding stress situations which may diminish natural or acquired immunity (such as inappropriate diet, poor water quality, inadequate aeration, fluctuating water chemistry/temperature, overcrowding, and aggressive behaviours such as fighting/bullying), effective water filtration, improved sanitation (disinfection of shared equipment) and preventing cross-contamination (dinospores can be spread by strong winds in aerosol droplets generated by farm equipment servicing contiguous ponds/tanks). Obviously, control programmes are difficult to implement in large-scale facilities utilizing natural aquatic features, such as coastal lagoons, fiords, estuarine swamps, inland lakes, ponds and rivers, some of which have experienced intermittent or periodic outbreaks. A case may be made for intensive studies on vaccine development as it has been consistently observed that many fish species surviving infections develop some protective immunity to subsequent challenge.



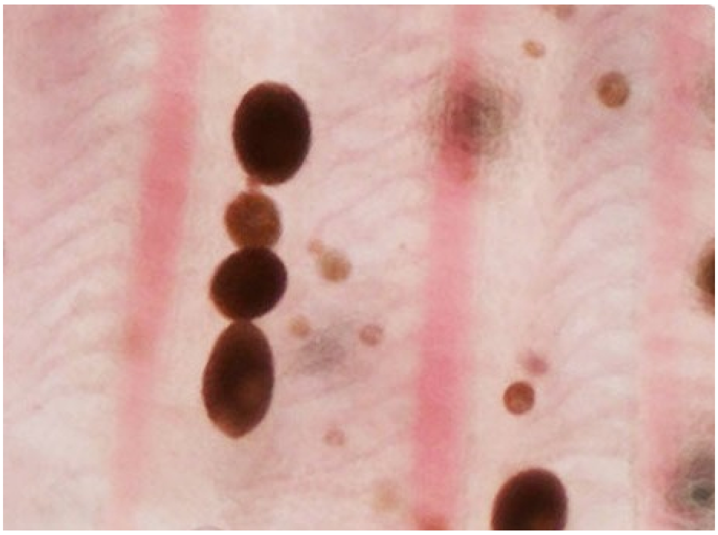
Amyloodinium, Piscinoodinium, Crepidoodinium
oodinid dinoflagellates with parasitic stages on aquatic hosts
skin, gills (superficial lesions, discolourations (velvet disease), respiratory disorders)
Vertebrate Hosts (marine and freshwater fish)
Amyloodinium (< 150 µm) [saltwater fish]
Piscinoodinium (< 160 µm) [freshwater fish]
Crepidoodinium (< 700 µm) [estuarine fish]
parasitic trophonts with rhizoid-like holdfast organelles (cells with or without chloroplasts)
dinospores attach to hosts
trophonts detach and form tomonts
Amyloodinium
Piscinoodinium
Crepidoodinium
dinospores (5-20 µm) biflagellated (equatorial girdle, ventral sulcus)
tomonts (100-200 µm)
direct transmission via free-swimming dinospores released from reproductive tomonts



Amyloodinium trophont on fish gill



Amyloodinium tomonts in fish tank



Piscinoodinium trophonts on fish gills