Haemogregarina
(protist: apicomplexan)

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. Apicomplexan cells possess a distinctive apical complex of organelles, comprising a conoid, polar ring, rhoptries, micronemes and subpellicular microtubules, which facilitate entry into host cells as they are obligate intracellular parasites for most of their life-cycles. There are three main apicomplexan groups: gregarines, coccidia and haematozoa. Three groups of coccidia are recognized: coelotrophiid coccidia in marine annelids; adeleid coccidia (including the blood parasites paradoxically known as 'haemogregarines') in marine and terrestrial animals; and eimeriid coccidia in vertebrates. Haemogregarines are heteroxenous (2-host) parasites where merogony (= schizogony) and gamogony occurs in the vascular system of a vertebrate host while sporogony occurs in a haematophagous invertebrate host (vector). They are common blood parasites in reptiles and some occur in fish, amphibians, birds and mammals. Four main assemblages are recognized: haemogregarinids, karyolysids, hepatozoids and dactylosomatids. Haemogregarinids (*Haemogregarina*, *Cyrilia* and *Desseria*) produce oocysts with naked sporozoites in the guts of leech vectors. Vertebrates (mainly aquatic animals) are infected by vector bite, and schizogony occurs in host erythrocytes (although infected cells tend to be sequestered in viscera) prior to the development of intraerythrocytic gamonts. Infections are generally considered to be nonpathogenic, even when high parasitaemias are present. Given the paucity of information on the developmental biology of most species, it has been suggested that all members of the genus *Haemogregarina* in snakes, crocodiles, lizards, amphibians, birds and mammals (but not tortoises or fish) be transferred to the genus *Hepatozoon* pending determination of their vectors and developmental cycles.

Classification:

Domain:	Eukaryota (membrane-bound nucleus)
Supergroup:	SAR (Stramenopiles + Alveolata + Rhizaria)
Group:	Alveolata (with cortical alveoli)
Phylum:	Apicomplexa (with apical complex, all parasitic, sexual development (gamogony))
Class:	Coccidiomorphea [Conoidasida] (with conoid)
Subclass:	Coccidia [Coccidiasina] (small intracellular gamonts)
Order:	Eucoccidiorida (cyclic merogony (schizogony), gamogony, sporogony)
Suborder:	Adeleina (syzygy, 1-4 microgametes)
Family:	Haemogregarinidae (ookinete, gamonts in blood cells, invertebrate vectors)
Genus:	*Haemogregarina* (vector-borne haemoparasites)
Species:	various species cause infections in blood/tissues of reptiles/amphibia/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. Cells with different modes of locomotion do not form separate monophyletic assemblages as previously thought, but rather are distributed across several disparate supergroups (as evidenced by recent molecular phylogenetic analyses). One protistan supergroup known as SAR comprises the Stramenopiles (with heterokont flagella), Alveolata (with cortical alveoli) and Rhizaria (with fine pseudopodia). Three diverse alveolate groups are recognized: Ciliophora (with cilia), Dinoflagellata (with flagella) and Apicomplexa (with gliding motion, some also with flagellated microgametes). Over 4,000 species of Apicomplexa have been described as obligate parasites from vertebrate and invertebrate hosts. At some stage in their development, these possess unique cytoskeletal and membrane-bound organelles (conoid, rhoptries, micronemes, subpellicular microtubules) forming an apical complex that facilitates host cell invasion. Apicomplexans undergo cyclic development involving up to three different divisional processes: asexual merogony (schizogony) either by fission (splitting of maternal cell) or endogeny (internal formation of daughter cells); gamogony involving formation of gametes (macrogametes = female, microgametes = male) which undergo fertilization to recombine by fusion (syngamy) with or without paired alignment (syzygy); and sporogony (formation of infective sporozoites).

Three main apicomplexan groups are recognized: haematozoa, gregarines, and coccidia. Haematozoa are small blood-borne parasites in vertebrates which complete their development in blood-sucking invertebrate vectors; with pleomorphic haemosporidia being transmitted by insects and pear-shaped piroplasms being transmitted by ticks. Gregarines are lumen-dwelling parasites that form large extracellular (sometimes septate) gamonts with an anterior holdfast organelle (mucron or epimerite) used to attach to the gut or body cavity of invertebrates. Coccidia are tissue-invading parasites that form small intracellular gamonts (lacking a mucron or epimerite) and most species undergo sexual reproduction by anisogamous fusion without syzygy forming non-motile resistant spores (oocysts) containing infective sporozoites usually confined within secondary spores (sporocysts). Three groups of coccidia are recognized: coelotrophiid coccidia in marine annelids; adeleid coccidia in marine and terrestrial animals (including blood parasites paradoxically known as 'haemogregarines' in reptiles and amphibians with leech or arthropod vectors); and eimeriid coccidia in vertebrates. Many eimeriid coccidia are monoxenous gut parasites undergoing faecal-oral transmission, but some are heteroxenous alternating between enteric stages in predators and encysted stages in prey (there are also a few enigmatic 'haemococcidia' in the blood of reptiles and birds).

Higher taxonomy	Family	Genera	Hosts	Site	Trans-mission*
Class: Gregarinomorphea (gregarines, trophonts with specialized attachment epimerite or mucron, syzygy)					
Subclass: Cryptogregaria (epicellular parasites of vertebrates with feeder organelle but lacking apicoplast)					
	Cryptosporidiidae (naked sporozoites)	*Cryptosporidium*	vertebrates	gut, lungs	direct (f-o)
Class: Coccidiomorphea [Conoidasida] (with conoid)					
Subclass: Coccidia [Coccidiasina] (small intracellular gamonts)					
Order: Eucoccidiorida (cyclic merogony (schizogony), gamogony, sporogony)					
Suborder: Adeleina (syzygy, 1-4 microgametes)	Haemogregarinidae (ookinete, gamonts in blood cells, invertebrate vectors)	*Haemogregarina*	reptiles, amphibia, fish	tissues, blood	indirect (v-b)
		Hepatozoon	mammals, reptiles	tissues, blood	indirect (v-b)
	Klossiellidae (sporocysts)	*Klossiella*	mammals	kidney	direct (f-o)
Suborder: Eimeriorina (no syzygy, >4 microgametes)	Eimeriidae (monoxenous, endogenous merogony and gamogony, exogenous sporogony)	*Caryospora*	birds, reptiles	gut	direct (f-o)
		Cyclospora	mammals, reptiles	gut	direct (f-o)
		Isospora	birds, reptiles	gut	direct (f-o)
		Eimeria	vertebrates	gut, tissues	direct (f-o)
		Epieimeria	fish	gut	direct (f-o)
		Goussia	fish	gut	direct (f-o)
	Sarcocystidae (heteroxenous, 1:2:4 oocyst:sporocyst:sporozoite configuration)				
	subfamily Cystoisosporinae (monozoic cysts)	*Cystoisospora* (no Stieda bodies)	carnivores, omnivores	gut, tissues	direct (f-o), indirect (p-p)
	subfamily: Sarcocystinae (thick-walls, metrocytes)	*Sarcocystis* (*Frenkelia*)	mammals, birds, reptiles	gut, muscles	indirect (p-p)
	subfamily: Toxoplasmatinae (thin-walled cysts without metrocytes)	*Besnoita*	mammals, reptiles	gut, tissues	indirect (p-p)
		Hammondia	mammals	gut, tissues	indirect (p-p)
		Neospora	herbivores, dogs	gut, tissues	indirect (p-p)
		Toxoplasma	vertebrates, cats	gut, tissues	indirect (p-p)
Class: Aconoidasida (asexual stages without conoid)					
Subclass: Haematozoa (clade of vector-borne spore-forming haemo-protozoa)					
Order: Haemosporida (pleomorphic blood stages, insect vectors, motile ookinete)	Plasmodiidae (schizogony in tissues then blood cells, haemozoin pigment)	*Plasmodium*	mammals, birds, reptiles	liver, erythrocytes	indirect (v-b)
	Haemoproteidae (schizogony in tissues, haemozoin pigment)	*Haemoproteus*	birds	endothelia, erythrocytes	indirect (v-b)
	Leucocytozoidae (schizogony in tissues, no haemozoin pigment)	*Leucocytozoon* (*Akiba*)	birds	tissues, leucocytes	indirect (v-b)
Order: Piroplasmorida (pear-shaped blood stages, tick vectors)	Babesiidae (merogony in erythrocytes, trans-stadial + trans-ovarian transmission)	*Babesia*	mammals	erythrocytes	indirect (v-b)
	Theileriidae (merogony in leucocytes, trans-stadial transmission in ticks)	*Theileria*	ruminants	leucocytes, erythrocytes	indirect (v-b)

* f-o = faecal-oral transmission; p-p = predator-prey transmission; v-b = vector-borne transmission.

Haemogregarines are heteroxenous (2-host) parasites where merogony (= schizogony) and gamogony occurs in the vascular system of a vertebrate intermediate host while fertilization and sporogony occurs in a haematophagous invertebrate definitive host (vector). They are common blood parasites in reptiles and fish and some occur in amphibians, birds and mammals. Infections are generally considered to be nonpathogenic, even when high parasitaemias are present. Haemogregarines are classified together with other coccidia, specifically with the adeleorine coccidia in marine and terrestrial animals. These parasites undergo sexual reproduction involving the formation of macro- and micro-gametes which associate pairwise in syzygy so comparatively few microgametes are produced (usually 1-4). The resultant zygote forms an oocyst containing infective sporozoites which are transmitted to vertebrates when infected vectors are eaten (consumptive) or when they feed on vertebrate blood (inoculative). Over 450 haemogregarine species have been described in vertebrate blood, most records being confined to the detection of intra-erythrocytic stages (almost exclusively gamonts) which do not produce haemozoin pigment. Numerous species were described simply on the basis of host occurrence but little is known about their actual host specificity or developmental cycles. Despite early confusion, there has been growing acceptance of the recent classification of haemogregarines into four families: Haemogregarinidae containing the genera *Haemogregarina*, *Cyrilia* and *Desseria*; Hepatozoidae containing the genus *Hepatozoon;* Karyolysidae containing the genera *Karyolysus* and *Hemolivia*; and Dactylosomatidae containing the genera *Dactylosoma* and *Babesiosoma* (in addition to 3 other adeleine families: Adeleidae, Klossiellidae and Legerellidae). It has been suggested that the chelonian (tortoise) haemogregarines be regarded as *Haemogregarina sensu stricto* (*s.s*., in the strictest sense) whereas fish haemogregarines be classified as *Desseria* or *Cyrilia* or left as *Haemogregarina sensu lato* (*s.l*., in the broadest sense). It was also suggested that all members of the genus *Haemogregarina* in reptiles (snakes, crocodiles, lizards), amphibians, birds and mammals be transferred to the genus *Hepatozoon* pending determination of their vectors and studies on their developmental cycles. All haemogregarines use vertebrates as intermediate hosts for cyclic merogony (often forming macro- then micro-merozoites) in tissues and gamont development in blood cells (mostly red, sometimes white, blood cells). Gamogony is completed in invertebrate definitive hosts (leeches, acarines or dipterans) followed by fertilization and oocyst formation in the gut or haemocoel. The oocysts undergo sporogony resulting in sporozoites lying directly within oocysts (for Dactylosomatidae), within sporocysts (for Hepatozoidae), within tissues (for Haemogregarinidae) or within eggs (for Karyolysidae). The four families are currently differentiated as follows:

- Haemogregarinids produce oocysts with naked sporozoites in the gut of leech vectors, vertebrates are infected by vector bite, and schizogony occurs in host erythrocytes (although infected cells tend to be sequestered in viscera prior to the development of intraerythrocytic gamonts); genera comprise *Haemogregarina s.s.* in turtles and *Haemogregarina s.l., Desseria* and *Cyrilia* in fish.
- Hepatozoids produce oocysts containing numerous sporocysts in the gut or haemocoel of various vectors (leeches, insects, acarines), vertebrates are infected by ingesting vectors, and schizogony occurs in vascular endothelial cells in host tissues (cystic stages also observed contributing to chronic infections and possible paratenic transport); the genus *Hepatozoon* occurs in mammals, reptiles (crocodiles, snakes, lizards), amphibians and birds.
- Karyolysids produce sporokinetes and motile spores in the gut and ova of mite vectors, vertebrates are infected by ingesting mites, and schizogony occurs in host vascular endothelial cells; genera comprise *Karyolysus* and *Hemolivia* in reptiles and amphibians.
- Dactylosomatids produce oocysts which bud sporozoites directly within epithelial cells in the intestines of leech vectors, vertebrates are infected presumably by vector bite and schizogony occurs in host erythrocytes; genera comprise *Dactylosoma* and *Babesiosoma* in reptiles, amphibians and fish.

Family	**Genus**	**No. species**	**Site of development in vertebrate**		**Vertebrate hosts**	**Invertebrate vector**
			meronts	**gamonts**		
Haemogregarinidae	*Haemogregarina*	*s.s.* 38; *s.l.* 42	erythrocytes	erythrocytes	*s.s.* in turtles; *s.l.* in fish	leeches
	Cyrilia	3	erythrocytes	erythrocytes	fish	leeches
	Desseria	35	exo-erythrocytic	erythrocytes	fish	leeches
Hepatozoidae	*Hepatozoon*	341*	viscera	erythrocytes, leucocytes	mammals, reptiles, birds, frogs, fish	leeches, arthropods
Karyolysidae	*Hemolivia*	4	RE cells	erythrocytes	lizards, tortoises, toads	ticks
	Karyolysus	11	viscera ± erythrocytes	erythrocytes	lizards, frogs	mites
Dactylosomatidae	*Dactylosoma*	10	erythrocytes	erythrocytes	reptiles, amphibians, fish	leeches
	Babesiosoma	12	erythrocytes	erythrocytes	fish, amphibians	leeches

*Includes some 200 former *Haemogregarina* spp. from crocodiles, snakes, lizards, amphibians and birds following their mass transfer to the genus *Hepatozoon* pending further characterization

Early phylogenetic studies used cladistics analyses of multiple phenotypic characters (morphology, biology) to reveal several clades apparently associated with different vectors (notably ticks, flies and leeches) rather than particular vertebrate hosts, thus suggesting vector-first evolution. However, such patterns may have arisen due to the environments in which the hosts occurred; e.g. marine hosts with leeches, and terrestrial hosts with ticks or flies. Molecular phylogenetic studies using small subunit (SSU) ribosomal RNA sequences revealed any associations between haemogregarines and their vertebrate hosts to be polyphyletic with many mixed clades associated with parasites in snakes, lizards, amphibians and mammals. Detailed analyses revealed five major clades: two *Hepatozoon* clades (clade A associated with carnivores and clade C with reptiles and frogs) separated by an intermediary *Hemolivia* clade in lizards, and two distant clades, one containing *Haemogregarina* species from tortoises and the other containing *Dactylosoma/Babesiosoma* species from fish and frogs. The five major clades were also well separated with respect to vectors: the former three clades using ticks and flies as vectors and the latter two using leeches. The *Hepatozoon* species in mammals (clade A) were more closely related to *Karyolysus* species from lizards, while the *Hepatozoon* species in frogs and reptiles (clade C) were more closely related to *Hemolivia* species from tortoises and lizards. Several interesting biological associations were evident between polyphyletic clades. *Haemogregarina* s.s. and dactylosome clades use leech vectors without sporocyst formation, and they only formed micro-merozoites in their turtle, frog or fish hosts. In contrast, members of the *Hepatozoon*, *Karyolysus* and *Hemolivia* clades use tick vectors with sporocyst (or sporokinete) formation, and they formed macro-merozoites in their mammal or reptile hosts.

Haemogregarina spp. have been described from a wide variety of vertebrate hosts; including reptiles (chelonians, crocodiles, snakes, lizards), amphibians (toads, frogs, salamanders), fish, and birds. The type species (*H. stepanowi*) infects chelonians and leeches, so the genus *Haemogregarina s.s.* is considered to contain all species in chelonians, although it has yet to be unequivocally shown that they all have leech vectors in which oocysts produce 8 naked sporozoites. Species of *Haemogregarina s.s.* have been found around the world in a range of freshwater turtles and terrestrial tortoises. Other species infecting fish have been classified as *Haemogregarina s.l.* as little is known about their vectors and sporogonic cycles. Many haemogregarines have been found in bony fish from marine habitats as diverse as intertidal rock pools to submarine canyons, and several species have also been detected in cartilaginous fishes, such as sharks, skates and rays. A smaller number of species have been found in bony fish from freshwater habitats as diverse as ditches, rivers and lakes, with several species are found in catadromous or anadromous fish (such as eels and some salmonids) which spend some of their life-cycles at sea. The taxonomic classification of haemogregarines in lizards, snakes, crocodilians and birds is confounded because not enough information is known about their developmental cycles, especially sporogonic development in their vectors (required for assignment to correct family and genus). Every *Haemogregarina* spp. described from non-chelonian reptiles, amphibians and birds for which sporogonic development was subsequently discovered, had multisporocystic oocysts and they were consequently transferred to the genus *Hepatozoon* in keeping with generic defining characters. It was further suggested that all other *Haemogregarina* spp. from non-chelonian reptiles, amphibians and birds (around 200 species) be similarly transferred to the genus *Hepatozoon* pending identification of their vectors and developmental cycles. This follows the earlier precedents of mass transfers of *Haemogregarina* spp. described from birds and mammals, as well as many non-avian *Leucocytozoon* spp., to the genus *Hepatozoon*. The situation is far from clear, and the various public databases continue to list many *Haemogregarina* spp. from non-chelonian reptiles and amphibians. In this document, those species are listed under the genus *Hepatozoon* (origins and status indicated by synonyms).

***Haemogregarina* species**	**Gamont dimensions (µm)**	**Hosts**	**Location***	**Vectors**	**Distribution**
Haemogregarina s.s. (species in Testudines, with leech vectors)					
H. bagensis	25-30 x 4-5	geoemydid (Spanish pond turtle)	rbc		Tunisia
H. balli	8-14.5 x 3-6.5	chelydrids (snapping turtle), emydid (spotted turtle, painted turtle); geoemydid (Mediterranean pond turtle)	rbc	Hirudinea: glossiphoniid (*Placobdella parasitica, ornata*)	Canada
H. billeti (syn. *H. choudhuryi, gangetica, laverani, malabarica, mesnili, vittatae, xavieri*)	8-9.6 x 2-4.8, elongate forms 17-26	trionychid (softshell turtles, flapshell turtles), emydid (pond turtles)	rbc	Hirudinea: glossiphoniid (*Helobdella nociva*)	India, Sri Lanka
H. bongaonensis	6.5-13.6 x 1.3-4.4	trionychid (softshell turtle)	rbc		India
H. botuliformis	16 x 6	geoemydid (Chinese pond turtle)	rbc		China
H. chelodinae (syn. *H. clelandi, H. dentata*)	7-12 x 7-9	chelid (eastern long-necked turtle, northern snake-necked turtle, broad-shelled river turtle, Murray River turtle, Krefft's turtle, saw-shelled turtle, northern snapping turtle)	rbc		Australia
H. chinemydis	17-19 x 4-5	geoemydid (Chinese pond turtle)	rbc		China
H. clelandi	7-20 x 4-12	chelid (snake-necked turtles)	rbc		Australia
H. clemmydis		emydid (spotted turtle), trionychid (flapshell turtle), geoemydid (Japanese pond turtle)	rbc		Japan

H. cuorae	16-18 x 2-3	geoemydid (Asian box turtle)	rbc		China
H. dimorphon		testudinid (tortoise)	rbc		
H. emydae	8 x 4	trionychid (Chinese softshell turtle)	rbc		Japan
H. fitzsimonsi [molecular evidence suggests close relationship with *Hepatozoon*]	17-18 x 3-4	testudinid (Bell's hinged tortoise, angulate tortoise, leopard tortoise)	rbc	Acari: ixodid (*Amblyomma sylvaticum, A. marmoreum*)	South Africa
H. galeata	11-13 x 5	trionychid (Chinese softshell turtle)	rbc		China
H. ganapatii	6-11.5 x 3-4.5	trionychid (flapshell turtle)	rbc		India
H. hubeiensis		trionychid (Chinese softshell turtle)	rbc		China
H. hydromedusae		chelid (snake-necked turtle)	rbc		Australia
H. ibera		testudinid (tortoise)	rbc		Europe
H. labbei		emydid (spotted turtle), chelid (twist-necked turtle)	rbc		South America
H. macrochelysi		chelydrid (alligator snapping turtle)	rbc		North America
H. maputensis		pelomedusid (side-necked turtle)	rbc		Africa
H. nicorae	10	geoemydid (Indian black turtle)	rbc	Hirudinea: ozobranchid (*Ozobranchus shipleyi*)	India, Sri Lanka
H. parvula		testudinid (Bell's hinged tortoise)	rbc		South Africa
H. pellegrini	two forms: 15-19 x 6-9, 11-15 x 4-6	geoemydid (Mekong snail-eating turtle), platysternid (big-headed turtle)	rbc		Thailand, China, Vietnam
H. pelusiensi	12.5-24 x 3.4-9	geoemydid (Caspian turtle), pelomedusid (serrated hinged terrapin)	rbc	Hirudinea: glossiphoniid (*Placobdella multistrigata*)	South Africa
H. podocnemis		podocnemidid (yellow-spotted Amazon River turtle)	rbc		South America
H. pseudemydis		emydid (sliders, cooters, box turtle, map turtle), kinosternid (eastern mud turtle), chelydrid (common snapping turtle), trionychid (spiny softshell turtle)	rbc, wbc	Hirudinea: glossiphoniid (*Placobdella parasitica*)	Central and North America
H. rara	15 x 2-3	geoemydid (Chinese pond turtle)	rbc		East Asia
H. reichenowi		trionychid (softshell turtle)	rbc		Africa
H. sacaliae	12-15 x 6-8	geoemydid (four-eyed turtle)	rbc		Vietnam
H. sinensis	11-12 x 4-5	trionychid (Chinese softshell turtle)	rbc	Hirudinea (*Mooreotorix cotylifer*)	China
H. stepanowi [type species]	29-37 x 3-5.5	emydid (European pond turtle, Sicilian pond turtle, painted turtle, Blanding's turtle, river cooter, Big Bend slider), chelydrid (common snapping turtle), testudinid (hinge-back tortoise, marginated tortoise), kinosternid (mud turtle, musk turtles), trionychid (softshell turtles)	rbc	Hirudinea: glossiphoniid (*Placobdella catenigera, costata*)	Europe, North America, Africa

H. stepanowiana	18-20 x 5	geoemydid (Chinese pond turtle)	rbc		South-East Asia
H. sternotheri		pelomedusid (side-necked turtle)	rbc		Africa
H. striata		trionychid (Aubry's flapshell turtle)	rbc		Africa
H. sundarbanensis		trionychid (Indian flapshell turtle)	rbc		India
H. testudinis	11-13 x 5-6	testudinid (Asian forest tortoise)	rbc		Indo-Malaysia
H. trionyxis		trionychid (African softshell turtle)	rbc		Africa
Haemogregarina s.l. (species in fish; with leech and isopod vectors)					
species in marine fish producing 2 or more gamonts per erythrocyte ('schizo-haemogregarines')					
H. bigemina (syn. *H. fragilis, Desseria fragilis*)	paired gamonts 10 x 2	Acanthuriformes: acanthurid (surgeonfish, tangs); Anguilliformes: muraenid (morays); Aulopiformes: synodontid (lizardfish); Beloniformes: belonid (needlefish), hemiramphid (halfbeaks); Blenniformes: blenniid (shanny, tompot, Montagu's blenny), clinid (bluntnose klipfish, highfin klipfish, orange clinid); Carangiformes: carangid (trevallies, amberjack), coryphaenid (dolphinfish), rachycentrid (kingfish); Cypriniformes: cyprinid (bream); Gobiiformes: gobiid (gobies); Istiophoriformes: istiophorid (sailfish), sphyraenid (barracuda); Labriformes: labrid (wrasse), scarid (parrotfish); ; Mugiliformes: mugilid (mullet); Perciformes: gerreid (mojarras), haemulid (grunts), kyphosid (chub), lutjanid (snapper, emperor), malacanthid (tilefish), mullid (goatfish), percid (perch), pomacanthid (angelfish), pomacentrid (sergeant-major); serranid (seabass, groupers); Scombriformes: scombrid (frigate tuna, mackerel); Scorpaeniformes: cottid (sculpins); Spariformes: sparid (porgies); Tetraodontiformes: balistid (triggerfish); Trachiniformes: pinguipedid (sandperch)	rbc, wbc	Isopoda: gnathiid (*Gnathia maxillaris, aureamaculosa*?)	Europe, North America, South Africa, Red Sea, South Pacific
H. polypartita	16 gamonts	Gobiiformes: gobiid (rock goby)	rbc		Europe
H. quadrigemina	4 gamonts 18 x 17	Callionymiformes: callionymid (common dragonet)	rbc		France
H. rubrimarensis	paired gamonts 12 x 1.5	Acanthuriformes: acanthurid (surgeonfish); Labriformes: scarid (parrotfish)	rbc		Red Sea
H. sachai	paired gamonts 5.5-10.9 x 1.1-2.0	Pleuronectiformes: scophthalmid (turbot)	wbc, rbc (myeloid leucosis)		Scotland
H. simondi	4, 6, 8 gamonts 9-19 x 1.5-3.6	Pleuronectiformes: soleid (common sole)	rbc, wbc	Hirudinea: piscicolid (*Hemibdella solea*), Copepoda:	Europe

				pennellid (*Lernaeocera*)	
species in marine fish producing one gamont per erythrocyte (with or without polar cap))					
H. balistapi		Tetraodontiformes: balistid (lagoon triggerfish)	rbc	Isopoda: gnathiid (*Gnathia aureamaculosa*)	Australia
H. bathysauri	13.8-17.6 x 3.3-4.3	Aulopiformes: synodontid (lizardfish)	rbc		Cape Verde Islands
H. binucleata		Callionymiformes: callionymid (dragonet)	rbc		England
H. blanchardi		Gobiiformes: gobiid (goby)	rbc		Europe
H. callionymi		Callionymiformes: callionymid (dragonet)	rbc		Europe
H. clavata	32 x 2.5	Pleuronectiformes: soleid (common sole)	rbc		Europe
H. coelorhynchi	polar caps	Gadiformes: macrourid (javelin fish), morid (cod)	rbc		New Zealand
H. curvata	6.3-7.9 x 0.9-2.3	Blenniformes: blenniid (horned blenny), clinid (bluntnose klipfish)	rbc	Hirudinea: piscicolid (*Zeylanicobdella arugamensis*)	South Africa
H. gobii		Gobiiformes: gobiid (goby)	rbc		Europe
H. hartochi		Gobiiformes: gobiid (goby)	rbc		Europe
H. hoplichthys	12.6 x 3.8, with polar cap	Scorpaeniformes: platycephalid (flathead)	rbc		New Zealand
H. koppiensis	6.1-9.8 x 2.0-3.6	Tetraodontiformes: tetraodontid (pufferfish)	rbc		South Africa
H. labri		Labriformes: labrid (wrasse)	rbc		Europe
H. leptoscopi	polar caps	Trachiniformes: uranoscopid (stargazer)	rbc		New Zealand
H. marzinowskii		Gobiiformes: gobiid (black goby)	rbc		Mediterranean
H. michaeljohnstoni	13.8-16.7 x 3.2-3.3	Ophidiiformes: ophidiid (brotulas)	rbc		Cape Verde Islands
H. minuta		Gobiiformes: gobiid (sand goby)	rbc		Mediterranean
H. parmae		Perciformes: pomacentrid (damselfish)	rbc		Australia
H. platessae (syn. *H. achiri, bothi, flesi, laternae*)	7.7 x 1.4	Pleuronectiformes: paralichthyid (flounder), pleuronectid (European plaice), scophthalmid (turbot), soleid (common sole)	rbc		France, North America
H. pollachii		Gadiformes: gadid (cod)	rbc		Europe
H. roelofsi		Scorpaeniformes: scorpaenid (black rockfish)	rbc		North America
H. rovignensis		Scorpaeniformes: triglid (streaked gurnard)	rbc		Africa
H. salariasi		Blenniformes: blenniid (blenny)	rbc		Fiji
H. scorpaenae		Scorpaeniformes: scorpaenid (scorpionfish)	rbc		Europe
H. tetraodontis	7.5-8.5 x 1-1.5	Tetraodontiformes: tetraodontid (pufferfish)	rbc		Australia
H. wladimirovi		Gobiiformes: gobiid (goby)	rbc		Europe
H. yakimovikohli		Gobiiformes: gobiid (goby)	rbc		Europe
species in marine cartilaginous fish (Chondrichthyes)					
H. carchariasi	34	Lamniformes: odontaspidid (sand tiger shark)	rbc		Australia
H. delagei	11.3 x 3.7 some polar caps	Rajiformes: rajid (skates); Squaliformes: squalid (dogfish sharks)	rbc		?
H. hemiscyllii	22-23.5 x 7-9.5	Orectolobiformes: hemiscylliid (epaulette shark)	rbc		Australia
species in freshwater fish					

H. acipenseris	5.9 x 2.4	Acipenseriformes: acipenserid (sturgeon)	rbc		Volga River
H. daviesensis		Ceratodontiformes: lepidosirenid (South American lungfish)	rbc		South America
H. escoi (syn. *Hepatozoon, Leucocytogregarina*)		Esociformes: esocid (pickerel)	rbc		Europe
H. majeedi		Cypriniformes: cyprinid (binni)	rbc		Middle-East
H. meridianus		Mugiliformes: mugilid (mullet)	rbc		Middle-East
H. thyrsoideae		Anguilliformes: anguillid (eels)	rbc		India

*rbc = red blood cells (erythrocytes), wbc = white blood cells (leucocytes)

Haemogregarines from fish that have been shown to form 16 or more sporozoites in leech vectors (Glossiphonidae and Piscicolidae) have been allocated to the genera *Cyrilia* (with erythrocytic merogony) and *Desseria* (with exo-erythrocytic merogony, although the exact location of schizogony has not been determined for many species and infected blood cells may be sequestered in visceral organs).

Parasite species	**Gamont dimensions (µm)**	**Hosts**	**Location**	**Vectors**	**Distribution**
Genus ***Cyrilia*** (merogony and gamogony in blood of freshwater fish, glossiphonid leech vectors)					
C. lignieresi (syn. *C. gomesi*) [type species]		Synbranchiformes: synbranchid (swamp eel)	wbc, rbc	Hirudinea: glossiphoniid (*Haementeria lutzi*)	Central and South America
C. nili (syn. *Desseria nili*)		Anabantiformes: channid (obscure snakehead); Cichliformes: cichlid (tilapia); Siluriformes: bagrid (catfish)	rbc	Hirudinea: glossiphoniid (*Batracobdelloides tricarinata*)	Africa
C. uncinata	24.6 x 5.7, polar caps	Scorpaeniformes: zoarcid (eelpout)	rbc	Hirudinea: piscicolid (*Johanssonia* sp.)	North America
Genus ***Desseria*** (merogony in tissues, gamogony in blood of fish, piscicolid leech vectors)					
species in freshwater teleost fishes					
D. baueri		Scorpaeniformes: cottid (sculpins)	rbc		Asia
D. catostomi	14.5 x 4.5	Cypriniformes: catostomid (suckers)	rbc		North America
D. colisa		Anabantiformes: osphronemid (gouramis)	rbc		West Bengal
D. cotti (syn. *Haemogregarina*)		Scorpaeniformes: cottid (sculpins)	rbc		Europe
D. irkalukpiki	17.2 x 3.2	Salmoniformes: salmonid (char)	rbc		Canada
D. lepidosirensis (syn. *Hg. bertoni*)	capsule	Ceratodontiformes: lepidosirenid (lungfish)	rbc		South America
D. ninakohlyakimovae		Cypriniformes: cyprinid (barbels)	rbc		Europe
D. parasiluri		Siluriformes: bagrid (catfish)	rbc		Asia
D. tilapiae		Cichliformes: cichlid (tilapia)	rbc		Africa
D. turkestanica		Siluriformes: bagrid (catfish)	rbc		Asia
D. vltavensis	12.5 x 3.4	Perciformes: percid (European perch)	rbc		Czechoslovakia
species in marine teleost fishes					
D. acanthoclini (syn. *Hepatozoon*)		Perciformes: plesiopid (roundheads, New Zealand rockfish)	rbc		New Zealand
D. aeglefini (syn. *Hg. urophysis*)	8-14.9 x 1.4-4.4, with polar cap	Gadiformes: gadid (haddock, cod, pollock), merlucciid (hake)	rbc		Northwest Atlantic
D. anarhichadis	polar caps	Scorpaeniformes: anarhichadid	rbc	Hirudinea: piscicolid	Europe

		(wolffish)		(*Platybdella anarrhichae*)	
D. brevoortiae		Clupeiformes: clupeid (menhaden)	rbc		North America
D. cataphracti		Scorpaeniformes: agonid (hooknoses)	rbc		Europe
D. dakarensis		Perciformes: haemulid (grunts)	rbc		Europe
D. georgianae (syn. *Haemogregarina*)		Perciformes: bathydraconid (Antarctic dragonfish)	rbc		Antarctica
D. gilbertiae		Perciformes: latid (seaperch)	rbc		Australia
D. harriottae	7.4-9.6 x 2.7-3.5	Chimaeriformes: rhinochimaerid (narrownose chimaera)	rbc		Cape Verde Islands
D. leptocotti (syn. *Hg. reolofsi*)	6.1-2.1	Scorpaeniformes: cottid (Pacific staghorn sculpin), sebastid (rockfish)	rbc		North America, South Africa
D. londoni	capsule	Blenniformes: blenniid (blennies)	rbc		Europe
D. marshalllairdi	10.1-14.9 x 3-3.9	Gadiformes: macrourid (rattails, grenadiers); Notacanthiformes: halosaurid (abyssmal halosaur)	rbc		Cape Verde Islands, North America
D. mavori		Scorpaeniformes: zoarcid (ocean pout)	rbc		North America
D. moringa		Anguilliformes: muraenid (spotted moray eel)	rbc		South America
D. mugili		Gobiiformes: gobiid (gobies); Mugiliformes: mugilid (mullet)	rbc		South America
D. myoxocephali [type species]	7.8 x 2.4	Scorpaeniformes: agonid (sculpins)	rbc	Hirudinea: piscicolid (*Malmiana nuda, scorpii*)	Northwest Atlantic
D. nototheniiae		Perciformes: nototheniid (cod icefish)	rbc		Antarctic
D. rovignensis	polar cap	Scorpaeniformes: trigilid (gurnard)	rbc		Italy
D. thyrsoideae		Anguilliformes: muraenid (moray eel)	rbc		India
D. zei		Zeiformes: zeid (dories)	rbc		South Africa
D. zeugopteri		Pleuronectiformes: scophthalmid (turbot)	rbc		Europe
species in marine cartilaginous fishes					
D. dasyatis (syn. *Haemogregarina*)		Myliobatiformes: dasyatid (southern stingray)	rbc		North America
D. heterodonti		Heterodontiformes: heterodontid (bullhead shark)	rbc		Asia
D. torpedinis (syn. *Hg. lobianci*)		Torpediniformes: torpedinid (electric rays)	rbc		Europe

Parasite morphology: Haemogregarines form 4 different types of developmental stages: meronts and gamonts in intermediate hosts; and gametes and oocysts in definitive hosts (vectors). Meronts (sometimes called schizonts) appear as rounded basophilic bodies ranging in size from 9-24 µm that are located intracellularly in erythrocytes (*Haemogregarina*, *Cyrilia*) or host viscera (*Desseria*). They undergo internal division to form several (2-8 for *Haemogregarina* and *Cyrilia*, 12-25 for *Desseria*) slender merozoites ranging in size from 9-13 x 2-3 µm. A few early descriptions reported some larger schizonts with up to 70-140 merozoites, but they have not been observed since. Gamonts appear as broad fusiform bodies usually located intracellularly within host erythrocytes (sometimes with more than one gamont per erythrocyte), although they may occasionally be detected in leucocytes or extracellularly in blood smears. They may be crescent-, sausage- or club-shaped, range in size from 8-15 x 2-7 µm, and some appear to have small polar caps. Intra-erythrocytic gamonts are often larger that the host cell nucleus and usually displace it laterally. Gamonts have a single conspicuous nucleus, which is generally discrete and central, but sometimes may be diffuse or fragmented. Microgamonts (male) have club-shaped nuclei tapering posteriorly, the cytoplasm stains blue with red inclusions, and many possess a short-recurved tail. Macrogamonts (female) have a central rounded nucleus, the cytoplasm stains blue with flocculent material, and they do not possess tails. While many haemogregarine species have been described solely on the basis of host occurrence and gamont morphology, their correct allocation to genera is dependent on differentiating oocysts formed in invertebrate vectors (data lacking for most species). In the vector, ingested gamonts form macrogametocytes (female) and microgametocytes (male) which associate pairwise (syzygy) and develop into anisogamous (dissimilar) gametes. Macrogametes remain as single rounded cells, while microgametocytes lose their tails and produce 1-4 microgametes, most reported to have a single terminal flagellum (although those of some species appear to have no flagella, and other rarely 2 flagella). Fertilization produces a non-motile zygote that begins oocyst formation within a flexible membrane, young undifferentiated oocysts being subspherical (6-9 x 5-8 µm) and growing in size as they mature (10-22 x 8-15 µm). Parasite species that form 4-16 (usually 8) naked sporozoites (not enclosed within sporocysts) have been allocated to the genus *Haemogregarina* (*s.s.* in turtles, s.l. in fish), while those that form 16 or more sporozoites have been allocated to the genera *Cyrilia* and *Desseria* (genera differentiated on the basis of the site of merogony in fish: erythrocytic in *Cyrilia,* and exo-erythrocytic in *Desseria*). The sporozoites are elongate (8-14 x 3-6 µm) tapering posteriorly, and possess a central nucleus and several crystalloid inclusion bodies. Ultrastructural studies have demonstrated the presence of typical apicomplexan organelles in various developmental stages: with most containing polar rings and micropores, and all non-gamete stages bearing an anterior conoid. The cells possessed cristate mitochondria but lacked amylopectin granules (present in *Hepatozoon*).

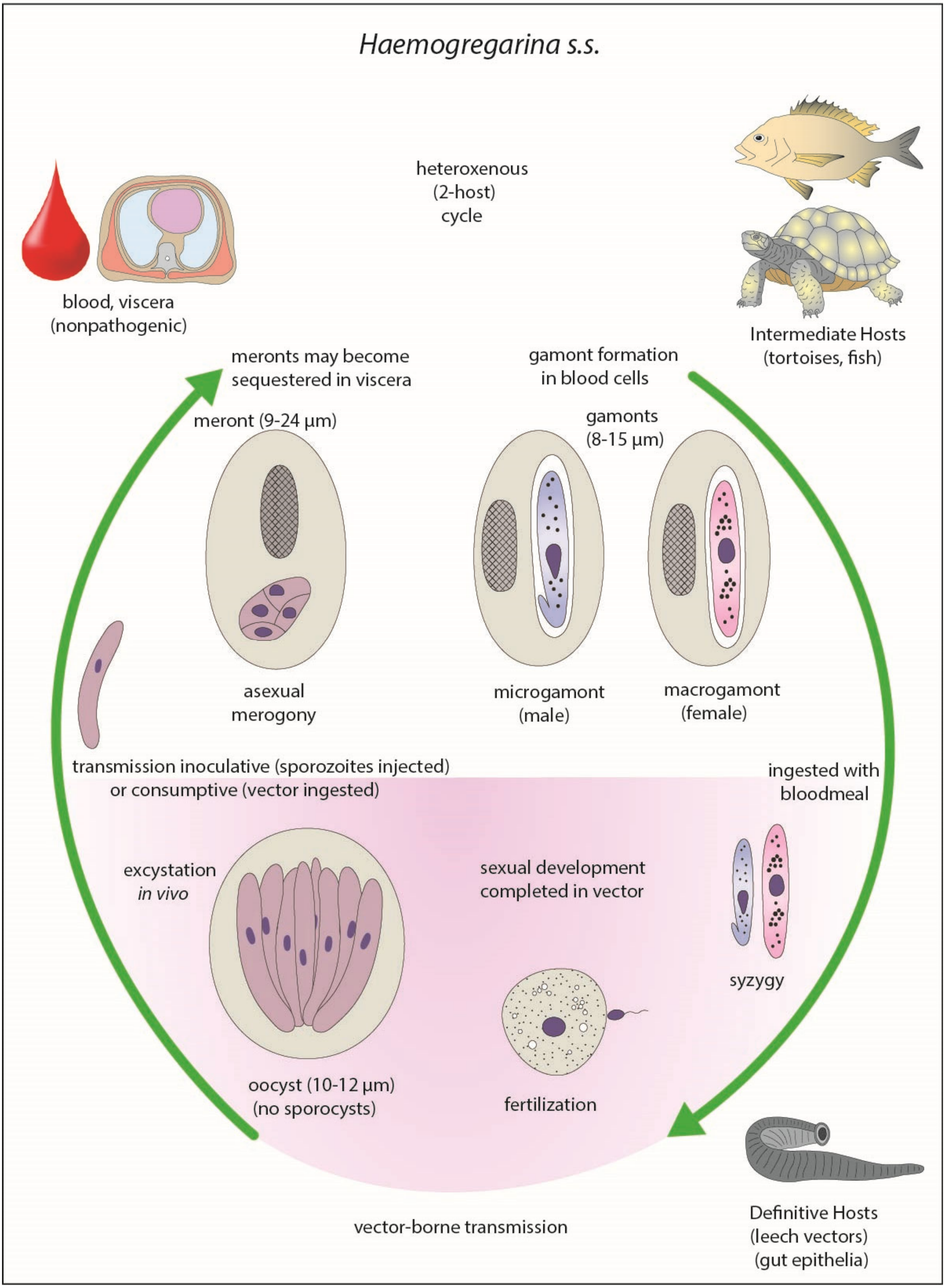
Site of infection: Hundreds of haemogregarine species have been described from a wide range of vertebrate hosts, including reptiles, amphibians, fish, birds and mammals. Over time, studies on parasite developmental cycles indicated that several genera may be involved. It was suggested that haemogregarines from mammals, birds, snakes, lizards, crocodiles and amphibians be transferred *en masse* to the genus *Hepatozoon* pending further studies. This left haemogregarines in tortoises as members of the genus *Haemogregarina s.s.* (*sensu stricto*, in the strictest sense), while haemogregarines of fish were left in the genus *Haemogregarina sensu lato* (*s.l*., in the broadest sense) or allocated to the sibling genera *Cyrilia* or *Desseria.* Both *Haemogregarina* and *Cyrilia* spp. form erythrocytic meronts, while *Desseria* spp. form exo-erythrocytic meronts. Infected blood cells often become trapped or sequestered in visceral organs, including the liver, lungs, spleen, bone marrow and occasionally the heart and brain. Gamonts of all 3 genera are then found within erythrocytes (sometimes leucocytes) of their chelonian or piscine hosts. Some 35 species of *Haemogregarina s.s.* have been recorded in around 50 tortoise species (belonging to 10 families). Around 38 species of *Haemogregarina s.l.* have been reported in fish, including 32 species in marine fish belonging to 65 species (in 49 families in 25 orders), and 6 species in freshwater fish belonging to 6 species (in 6 families in 6 orders). Three *Cyrilia* spp. have been found in freshwater fish belonging to 5 species (in 5 families in 5 orders), and 35 *Desseria* spp. have been reported, with 24 species occurring in marine fish belonging to 31 species (in 26 families in 15 orders) and 11 species in freshwater fish belonging to 9 species (in 9 families in 8 orders). In those species for which vectors have been determined, gametogony and oocyst formation occur in the gut epithelia of aquatic invertebrate hosts, predominantly leeches, but including some isopods and rarely copepods.

Pathogenesis: Infections by haemogregarines in vertebrates have not been associated with any specific clinical disease, neither during the asexual merogonous proliferation of parasites in blood cells or tissues, nor during the carriage of gamonts in blood cells (even when some parasitaemias exceed 30%). While merogony ultimately leads to host cell lysis to release the contained merozoites, the extent of damage to the host appears to be minimal and usually does not lead to clinical signs. Gamonts that develop in host erythrocytes may persist for long periods and while they may cause variation in erythrocyte morphology (modest hypertrophy, turgidity and nuclear displacement), they are not thought to cause significant haemolysis. Nevertheless, several studies have reported changes in haematological parameters in a few hosts (mainly freshwater fish), but they have often been contradictory, e.g. red cell counts decreased in sturgeon but elevated in other species, and variable but inconsistent white cell counts in several species (esp. lymphocytes, neutrophils and monocytes). Long-standing infections in pond turtles in a European zoo were linked to poor health, general weakness, anorexia, skin haemorrhages, visceral hyperaemia (lungs, liver, kidneys, spleen), reduced hematocrits, and shell lesions often with secondary bacterial or fungal infections. It has often been suggested that subclinical infections in fish may still affect fish health, growth and survival, even resulting in reduced productivity in commercial fisheries and aquaria (particularly in fish stressed by poor water quality and crowded conditions). Some leucocyte infections in cultured marine fish (mostly turbot) have been associated with a proliferative disease (myeloid leucosis or lymphoma-like condition), with lesions comprised entirely of encapsulated aggregates of merozoites or infected macrophages embedded in granulomatous tissue.

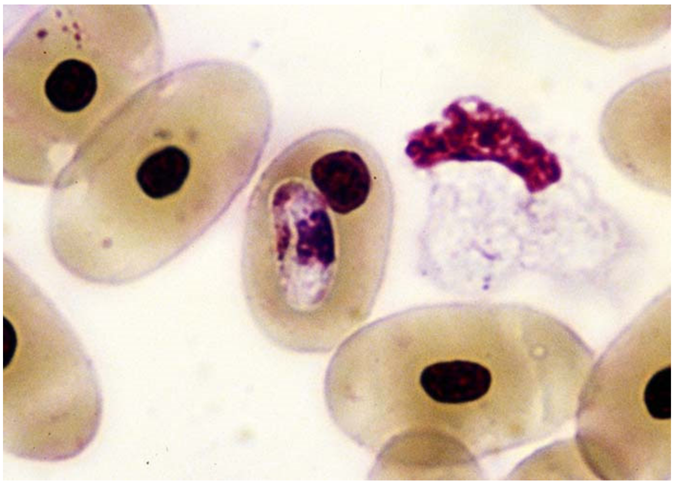
Developmental cycle and mode of transmission: The complete developmental cycles have only been determined for a handful of haemogregarine species, but they have involved obligative heteroxenous (2-host) life-cycles with asexual merogony and gamont formation occurring in the vascular system of vertebrate hosts (intermediate hosts) and sexual gamogony and oocyst formation occurring in the gut of haematophagous invertebrate hosts (vectors or definitive hosts). Vertebrate hosts become infected when sporozoites invade tissues where they form intracellular meronts (schizonts) within blood cells or vascular endothelial cells. An intracellular pre-meront (or trophozoite) stage has occasionally been observed for some species. Most *Haemogregarina* and *Cyrilia* spp. infect erythrocytes, although some species may also infect leucocytes. Infected cells tend to be sequestered in visceral organs. *Desseria* spp. infect endothelial cells in various host tissues. The parasites proliferate asexually by endogenous division (an internal budding process) with meronts producing numerous merozoites. Studies have implicated multiple meront generations in the parasite proliferative cycles, with some species forming macro-meronts producing small numbers of large merozoites, and other species forming micro-meronts producing larger numbers of smaller merozoites. Several studies have indicated that some species with persistent tissue-phase meronts may possibly be transmitted horizontally between vertebrate hosts by predation (when infected fish are eaten by larger predators), but such transmission remains to be proven. Eventually, merozoites invade erythroblasts or erythrocytes and develop into gamonts, with many studies also describing pre-gamont stages. Haemogregarines have been allocated to 2 broad groups depending on whether cells undergo division immediately before gamont formation: with 'schizo-haemogregarines' or those of the '*bigemina*-group' dividing to form paired (or multiples of two) undifferentiated gamonts; and those of the '*rovignensis*-group' not dividing but forming single monomorphic or sexually dimorphic gamonts, some with deeply staining polar caps. The intra-erythrocytic gamonts are infective to haematophagous vectors (mainly leeches, and some isopods and copepods) when they feed on host blood. The gamonts differentiate in the gut and undergo extracellular gametogenesis often on the microvillous surface of epithelial cells. Female macrogametocytes and male microgametocytes pair up (in syzygy) and form single 'egg-like' macrogametes and several 'sperm-like' microgametes. Fertilization occurs by gamete fusion resulting in a non-motile zygote that initiates oocyst formation within a flexible membrane (rather than a heavy oocyst wall like enteric coccidia). Developing oocysts undergo sporogony forming multiple sporozoites that are not enclosed within secondary cysts (sporocysts) but lie 'naked' within the oocyst. *Haemogregarina* spp. form oocysts with 4-16 (usually 8) naked sporozoites, while *Cyrilia* and *Desseria* spp. form oocysts with more than 16 sporozoites. The oocysts excyst endogenously releasing sporozoites which may be found in various vector tissues, especially the salivary glands. Sporozoites from some species are also thought to undergo one round of merogonous division in vector tissues. The sporozoites and/or merozoites are infective to vertebrate hosts, with transmission being inoculative (sporozoites/merozoites injected during feeding) or consumptive (infected vector eaten during grooming). Some fish haemogregarines have demonstrated strong seasonal patterns of infection, with heavy infections developing in marine hosts when temperatures were higher in spring and summer, while heavy infections developed in freshwater hosts when water temperatures were lower. Besides water temperature, other factors influencing parasite distribution and abundance including host and vector biogeography and population densities, migratory behaviours, and host stress factors (relating to nutritional state and spawning behaviours).

Differential diagnosis: Infections in turtles and fish are predominantly asymptomatic and resistant parasite stages are not voided from hosts. Parasites are usually detected incidentally by the microscopic detection of characteristic intra-erythrocytic gamonts in blood smears stained with Giemsa. Specific and generic identification based solely on gamont morphology is not possible as *Haemogregarina*, *Cyrilia* and *Desseria* spp. form similar gamonts which often includes sexual dimorphism. Meronts (schizonts) are usually detected by the microscopic examination of blood smears and histological sections. *Haemogregarina* and *Cyrilia* spp. form intra-erythrocytic meronts, while *Desseria* spp. form exo-erythrocytic schizonts, but the sequestration of infected cells in host tissues can confound the actual site of infection. Infections in leeches are usually detected by the microscopic examination of fresh squash preparations or fixed histological sections of gut epithelia looking for developing oocysts. Mature oocysts of *Haemogregarina* spp. form oocysts with 4-16 (usually 8) naked sporozoites, while *Cyrilia* and *Desseria* spp. form oocysts with more than 16 naked sporozoites. More recently, molecular biological techniques have also been used to characterize parasites and infer phylogenetic relationships by the polymerase chain reaction (PCR) amplification of nuclear gene sequences (small subunit (18S) ribosomal DNA).

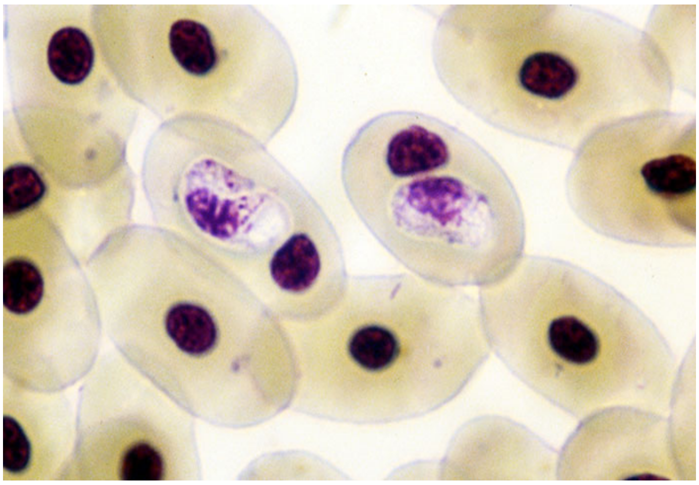
Treatment and control: Treatment is generally not required as most infections are benign and do not produce clinical disease, so there are few historical reports involving chemotherapy. However, several captive turtles with shell lesions had concomitant haemogregarine infections that were tentatively linked to anaemia, low haemoglobin, basophilia, eosinophilia, heterophilia and azurophilia. Haemogregarine numbers declined in some turtles following combination therapies involving the systemic application of antiprotozoals (metronidazole), anthelmintics (albendazole, mebendazole) and antibiotics (enrofloxacin) together with debridement and scrubbing of lesions with disinfectants. Various preventive measures were instituted in captive or culture facilities to eliminate potential sources of infection (routine pond cleaning, vector control) and ensure healthy rearing conditions to avoid chronic host stress and hence susceptibility (avoid over-handling and over-crowding, maintain water quality, and provide good nutrition without contributing to organic enrichment). The eventual elimination of long-standing haemogregarine infections in some zoo turtle collections was attributed to the regular mechanical removal of leeches.



Haemogregarina s.s.
heteroxenous
(2-host)
cycle
blood, viscera
(nonpathogenic)
Intermediate Hosts
(tortoises, fish)
meronts may become
sequestered in viscera
gamont formation
in blood cells
meront (9-24 µm)
gamonts
(8-15 µm)
asexual
merogony
microgamont
(male)
macrogamont
(female)
transmission inoculative (sporozoites injected)
or consumptive (vector ingested)
ingested with
bloodmeal
excystation
in vivo
sexual development
completed in vector
syzygy
oocyst (10-12 µm)
(no sporocysts)
fertilization
vector-borne transmission
Definitive Hosts
(leech vectors)
(gut epithelia)



Haemogregarina s.s. gamont in tortoise blood



Haemogregarina s.s. gamonts in tortoise blood