

Parasite Taxonomy

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Learning Objectives

- 1. To have a broad overview of the existing classification of parasites.
- 2. To understand the basis for the classification.
- To have a knowledge about the modern methods for taxonomy and the associated changes in classification of some parasites.

Introduction

Ever since the first classification of living beings into two kingdoms—Animalia and Plantae by Carl Linnaeus in 1758—new information and discoveries have resulted in increasing complexity and complications in designing a proper classification. The earlier classification systems relied heavily on the morphological aspects of the organisms. The advent of ultrastructural details, their enzymatic pattern and genetic makeup have been instrumental in the re-classification of many of these parasites. Recent advancements in gene sequencing and other methodologies have found that some earlier phylogenetic classifications do not necessarily fall in line with the evolutionary past. Hence, new changes and modifications are necessary as new discoveries come to light. There is a need to understand the taxonomical classification of parasites from two points of view: the traditional and the modern. While most scientists are familiar with the older and conventional classification, the modern-day system using more sophisticated data has classified and re-classified the existing parasites, and new nomenclature has even been assigned to them. This has created understandable confusion among the various stakeholders. Thus a compromise is necessary between the current evolutionary thinking and the more practical need for a system of nomenclature which will allow scientists from diverse backgrounds to effectively communicate with each other and retrieve relevant information from archival and historical data.

The Evolution of Classification Systemics

The broad division of all living beings into two kingdoms, Animalia and Plantae, in 1758 by Linnaeus, marked the advent of taxonomical classification. The discovery of numerous unicellular organisms with the invention of the microscope prompted scientists like Haeckel in 1876, credited with the creation of a third kingdom, Protista, to include these life forms. Subsequently, four

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kingdoms were proposed by Copeland in 1949 (Animalia, Plantae, Protoctista and Mychota). The removal of fungi from the plant kingdom necessitated the addition of the fifth kingdom of fungi. Jahn and Jahn in 1949 modified further the kingdoms which formed the basis of a five-kingdom classification of Whittaker in 1969. This classification included Monera (prokaryotes), Animalia, Plantae, Fungi and Protista. Corliss (1994) proposed six kingdoms in the empire Eukaryota retaining the old Plantae, Fungi and Animalia and introducing three kingdoms of unicellular organisms of Archezoa, Protozoa and

Taxonomy of Protozoal Parasites

The unicellular eukaryotic organisms have been given various names: Protozoa, Protista or Protoctista. Each name has its own proponents and followers. Protozoa and Protista are the favourites among parasitologists and protozoologists, respectively. When additional kingdoms were introduced, the status of Protozoa rose to the level of the kingdom. The Protozoa was first classified by Goldfuss in 1818 into three groups: amoebae, flagellates and ciliates based on their organs of locomotion. Subsequently, the sporozoans were included in the kingdom by Butschli in 1883. Since then, numerous classification systems and re-classifications have been suggested.

Cavalier-Smith (2003) Classification of the Kingdom Protozoa

The kingdom Protozoa as proposed by Cavalier-Smith (2003) is based on certain traits which distinguish them from other unicellular living organisms. The classification proposed by Cavalier-Smith suggests that the kingdom Protozoa includes 11 phyla of which only a few are pathogens in humans and animals. The kingdom Protozoa includes more than 200,000 protozoa species, of which only about 10,000 (0.5%) are parasites, with or without any pathogenic potential. Phyla Amoebozoa, Trichozoa, Percolozoa, Euglenozoa, Miozoa and Ciliophora are the only phyla of 11 phyla in the kingdom Protozoa that contain potentially pathogenic species for humans and animals (Table 1):

- 1. Amoebozoa: These include protozoa that have pseudopodia as locomotory organs or are motile by protoplasmic flow. Flagella, if present, are restricted to one particular life stage. They reproduce asexually by fission; sexual reproduction is associated with free living amoebas. Mitochondrial cristae tubular or mitochondria and peroxisomes are absent.
- 2. **Euglenozoa:** Protozoa included in the group have flagella, often with the presence of paraxial rod. They also have discoidal mito-chondrial cristae and cortical microtubules and show persistence of nucleoli during meiotic division.
- 3. **Percolozoa:** Percolozoa have heterotrophic flagella or amoeboflagella and discoid mitochondrial cristae. They commonly alternate between a flagellate phase with pellicle and a main non-ciliate trophic amoeboid phase.
- 4. **Trichozoa:** These protozoa are flagellates or, rarely, amoebae consisting of hydrogenosomes and prominent Golgi dictyosomes. They exhibit closed mitosis with extra-nuclear mitotic spindle.
- 5. **Miozoa:** The Miozoa consists of protozoa which commonly or ancestrally feed by the process of myzocytosis. These protozoa therefore pierce the cell wall or cell membrane of the host with a conoid or feeding pipe and suck out the cellular contents.
- 6. **Ciliophora:** These protozoa are parasites of digestive tracts. They have cilia and cortical alveoli and, typically, have two types of nuclei (*heterokaryotic*). The Ciliophora protozoa may exhibit sexual phenomenon of *conjugation* or *autogamy* and *cytogamy* or asexual reproduction by transverse fission. Contractile vacuoles are present.

Chromista.

 Table 1
 Revised detailed classification of pathogenic protozoan parasites (After Cavalier Smith, 2003)

Kingdom: Protozoa								
A. Subkingdom: Sarcomastigota								
Phylum: Amoebozoa								
Subphylum 1: Protamoebae								
Class 4: Variosea: Acanthamoeba, Balmuthia								
Subphylum 2: Archamoeba								
Class: Archamoeha: Entamoeha: Endolimax								
B. Subkingdom: Biciliata								
Infra-kingdom: Excavata								
Phylum 2: Metamonada								
Subphylum: Trichozoa								
Superclass 1: Parabasalia								
Class 1: Trichomonadea: Trichomonas, Lophomonas								
Superclass 3: Eopharyngea								
Class 1: Trepomonadea:								
Subclass 1: Diplozoa: Giardia								
Class 2: Retortamonadea: <i>Retortamonas, Chilomastix</i>								
Superphylum 1: Discicristata								
Phylum 1: Percolozoa								
Class 1: Heterolobosea: Naegleria								
Phylum 2: Euglenozoa								
Subphylum 2: Saccostoma								
Class 1: Kinetoplastea: Trypanosoma, Leishmania.								
Infra-kingdom: Alveolata								
Phylum 1: Miozoa								
Subphylum 3: Apicomplexa								
Infraphylum: Sporozoa								
Class 1: Coccidea: Toxoplasma, Cryptosporidium								
Class 2: Hematozoa: <i>Plasmodium</i> , <i>Babesia</i> .								
Phylum 2: Ciliophora								
Subphylum 2: Intramacornucleata								
Class 2: Litostomatea: Balantidium								

Kingdom	Phylum	Class	Order	Agent		
Archezoa	Metamonada	Trepomonada	Diplomonadida	Giardia		
			Enteromonadida	Enteromonas		
Retortamonada		Retortamonadida	Retortamonas, Chilomastix			
	Microspora	Microsporea	Microsporida	Encephalitozoon, Enterocytozoon, Nosema, Septata		
Protozoa	Percolozoa	Heterolobosea	Schizopyrenida	Naegleria		
	Parabasalia	Trichomonadia	Trichomonadida	Trichomonas		
	Euglenozoa	Kinetoplastidea	Trypanosomatida	Trypanosoma, Leishmania		
	Ciliophora	Litostomatea	Vestibuliferida	Balantidium		
	Apicomplexa	Coccidea	Eimerida	Cryptosporidium, Cyclospora, Toxoplasma, Isospora, Sarcocystis		
		Haematozoa	Haemosporida	Plasmodium		
			Piroplasmida	Babesia		

 Table 2
 Utilitarian classification of pathogenic protozoa (After Corliss, 1994)

Corliss (1994) Classification of the Kingdom Protozoa

Classification by Corliss (1994) is another simplified system of classification. This classification encompasses both conventional and molecular characteristics of parasites. This system also retains the older names of parasites for the sake of simplicity and familiarity. This system of classification is highly useful for medical and veterinary parasitologists and is of practical importance (Table 2).

As per the classification by Corliss (1994), pathogenic potential protozoal species that can cause infections in humans and animals are included in the following phyla:

- 1. **Metamonada:** These protozoa are parasites of the intestinal tract. They have two or more flagella and contain hydrogenosomes instead of mitochondria.
- Microspora: These are unicellular spore-like structures containing one or two nuclei with sporoplasm and a polar filament. They lack mitochondria and peroxisomes but have 70S ribosome.
- 3. **Parabasalia:** These protozoa have multiple flagella. They have parabasal fibres which arise at the kinetosomes. The parabasal

apparatus is analogous to the Golgi apparatus. They lack mitochondria.

4. Apicomplexa: The protozoa belonging to this phylum have a unique structure known as the apical complex. The complex comprises a polar ring, micronemes, rhoptries, conoid and subpellicular tubules. They have cortical alveoli and represent the sporozoans described in the old classifications of protozoa.

Taxonomy of Helminthic Parasites

Classification of helminths into cestodes, trematodes and nematodes is a working classification of convenience, more familiar among parasitologists in the field of medical and veterinary sciences.

A zoological system of classification of helminths includes the subkingdom Bilateria in the Animalia kingdom that consists of helminthic parasites. The infra-kingdom 1 (*Ecdysozoa*) comprises the nematodes (Table 3), while the infra-kingdom 2 (*Platyzoa*) contains the trematodes and cestodes (Table 4):

1. Nematodes: Nematodes are typically bilaterally symmetrical and are elongated with tapering ends. They possess a body cavity or *pseudocoel*. The digestive system

	bers	inella spiralis	uris trichiura	lostoma duodenale, Necator americanus	ris, Toxocara	tkis	unculus	iereria, Brugia, Onchocerca, Dirofilaria, Mansonella	tocerca	hostoma	ostrongylus	-obius	gyloides	ylonema	phagostomum, Ternidens	nomonoganus	ızia	ostrongylus
	Mem	Trich	Trich	Ancy	Asca	Anise	Drac	Wuch	strep	Gnat	Angia	Enter	Stron	Gong	0eso,	Mam	Thel	Trich
S	Family	Trichinellidae	Trichuridae	Ancylostomatidae	Ascarididae	Anisakidae	Dracunculidae	Onchocercidae		Gnathostomatidae	Angiostrongyloidae	Oxyuridae	Strongyloididae	Gongylonematidae	Chabertidae	Sygamidae	Thelaziidae	Trichostrongyloidae
	Superfamily	Trichinelloidea		Ancylostomatoidea	Ascaridoidea		Dracunculoidea	Filarioidea		Gnathostomatoidea	Metastrongyloidea	Oxyuroidea	Rhabditoidea	Spiruroidea	Strongyloidea		Thelazioidea	Trichostrongyloidea
nic Nemathelminth parasite	Class	Adenophora	(Aphasmidea)	Secementea	(Phasmidea)													
tion of pathoger	Infraphylum	Nematoda																
Table 3 Classificat	Phylum	Nemathelminthes																

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Phylum	Class	Order	Family	Members				
Platyhelminthes	Digenea	Strigeida	Diplostomadae	Diplostomum				
			Schistosomatidae	Schistosoma				
			Clinostomatidae	Clinostomum				
		Echinostomatida	Echinostomatidae	Echinostoma				
			Fasciolidae	Fasciola				
				Fasciolopsis				
			Zygocotilidae	Gastrodiscoides hominis				
				Watsonius watsoni				
		Plagiorchiida	Dicrocoeliidae	Dicrocoelium dendriticum				
			Heterophyidae	Heterophyes, Metagonimus				
			Opisthorchiidae	Opisthorchis (Clonorchis)				
			Lecithodendriidae	Phaneropsolus				
			Paragonimidae	Paragonimus				
			Plagiorchiidae	Plagiorchis				
			Troglotrematidae	Nanophyetus salmincola				
	Cestoidea	Pseudophyllidea	Diphyllobothridae	Diphyllobothrium, Spirometra, Sparganum				
		Cyclophyllidea	Anoplocephalidae	Bertiella				
			Davaineidae	Raillietina				
			Dipylidiidae	Dipylidium caninum				
			Hymenolepididae	Hymenolepis (Rodentolepis) nana,				
				Hymenolepis diminuta				
			Mesocestoididae	Mesocestoides				
			Taeniidae	Taenia, Echinococcus, Multiceps				

Table 4 Classification of pathogenic Platyhelminthes

comprises the mouth, pharynx and anal opening. The digestive canal is tri-radiate. The body does not possess cilia or flagella but has a variety of *sensilla* as sensory organ. The worms are dioecious with separate male and female adults. Females are normally larger and mostly oviparous. The ventral vulva represents the opening of the female reproductive system, while in the male, it opens into a cloaca along with the digestive system.

2. **Platyhelminthes:** They are called flatworms since they have a dorso-ventrally flattened bilaterally symmetrical body. They do not have a body cavity. The body is covered with tegument. Most of the body is made up of parenchyma and muscle fibres can be found in parenchyma. The digestive system is a blind sac-like structure with a mouth at the anterior end. The flame cells represent the excretory system of the worms. Most members are monoecious and can fertilize their own eggs.

Platyhelminthes are classified into two groups, trematodes and cestodes:

- Trematodes: They are hermaphrodite worms. They are also known as flukes and have a leaflike body and two suckers, one at the anterior and another at the posterior end. Trematodes have a digestive system. They require definitive hosts harbouring the adult stage and two intermediate hosts harbouring the larval stages of miracidium, sporocysts and cercaria.
- 2. Cestodes: The cestodes have three embryonic layers: ectoderm, mesoderm and endoderm. The head or scolex present at the anterior end of the body helps in attachment of the cestode to tissue of the host. The body or strobila is segmented and is unique to these parasites. The strobila consists of a linear series of male and female reproductive organ systems, and the surrounding area is known as the segment or proglottides. New proglottides or segments are found at the anterior end, while gravid proglottides are found at the posterior end. The gravid proglottides contain branched uterine structures filled with eggs. They lack a digestive tract and absorb all nutrition from the external covering or tegument with high metabolic activity.

Modern Methods for Classification of Parasites

While the nineteenth century and the first half of the previous century relied almost exclusively on light and later on electron microscopy to classify parasites, newer techniques were gradually introduced to study the relationship between these life forms at the molecular level. The need for these techniques has arisen due to multiple reasons (Fig. 1).

One of the first such methodologies to be applied was to study the isoenzyme profiles. This was very useful to distinguish between closely related organisms, and the classical example was to differentiate pathogenic and non-pathogenic forms of the Entamoeba histolytica. It was similarly used for Toxoplasma gondii and to identify the subspecies of Trypanosoma brucei. In recent years this technique has also been used for phylogenetic

classification of Plasmodium falciparum and Cryptosporidium hominis. Subsequently, the new DNA and RNA technological advances overshadowed all other methods, and they now remain the most commonly used methodology for systemic classification, particularly for resolving taxonomical and phylogenetic controversies and problem-solving. Historically, the small subunit of ribosomal RNA was first utilized to create a phylogenetic tree in the 1980s. At present the 16S and 18S small nuclear RNAs and DNA probes are extensively used in taxonomy works, and they are particularly useful to find the evolutionary distance between the strains and create phylogenetic trees. Molecular karyotyping is another method which helps in determining the chromosomal size differences. It has been employed for the agents of cutaneous leishmaniasis and helped in geographical grouping of the strains. The study of whole genome sequencing may help in assigning some atypical or unclassified members of a genus



Fig. 1 The 'diagnostic cascade'. The driver to circumscribe a particular parasite taxon may be driven by 'biological' or 'policy' reasoning. (From: Stentiford G,

Feist S, Stone DM, Peeler E, Bass, D. Policy, phylogeny, and the parasite. Trends in parasitology. 2014. 30. https://doi.org/10.1016/j.pt.2014.04.004)

to novel subspecies or sub-genus levels. In addition to these, the emerging field of *proteomics* may open up new corridors in the classification of organisms. Genomic studies are useful to determine the evolutionary trends as well as giving indications about the level of genetic differences, but they do not take into account post-transcriptional regulation of protein expression and cannot determine the degree of crossreactivity between parasite species. It is possible that proteomic comparisons can give a better indication of phenotypic differences between different parasites.

The above mentioned newer technologies are gradually changing the way we look at these parasites and bringing up new and useful information. Further refinements in the taxonomy of parasites are expected in the next few decades, which may result in re-classification of existing parasites and creation of new classes or genera of these organisms.

Case Study

Taxonomical Position of Microspora

The Microspora has traditionally been considered as protozoans, but research findings in the last decade have found otherwise. In the fungal zygomycetes group, the sex locus is a syntenic gene cluster that governs sexual reproduction and comprises a high mobility group (HMG) gene, flanked by a triose phosphate transporter and a RNA helicase gene. The microsporidian genomes harbour a sex-related locus with the same genes in the same order. Moreover, genome-wide analysis of synteny reveals multiple other loci common to microsporidia and zygomycetes. These findings support the hypothesis that microsporidia are true fungi that descended from a zygomycete ancestor and suggest the microsporidia may have a genetically controlled sexual cycle. On the basis of these findings, Microspora is no longer

considered a protozoan parasite but is designated as a fungus.

- 1. Give one or two examples where similar re-classification of parasites has been made.
- 2. Describe the methods available for identifying a new parasite which shows some similarity with a known parasite.
- 3. Define a hybrid parasite. Name one common parasite which exhibits this feature.

Research Questions

- Like microsporidium, are there other protozoa which do not belong to the parasite group, but belong to fungi or some hitherto unknown group?
- 2. Is it possible to separate some algal forms, euglenids and dinoflagellates from protozoa which have totally different biology?
- 3. How can one create a more refined taxonomy of helminths based on the newer methods of classification?

Further Readings

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