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Study of the Systematics of Free Living Protozoan from Mumbai Region Maharashtra

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ABSTRACT

Protozoan's are unicellular, eukaryotic organisms that are placed in animal kingdom protista. The word protozoa come from the Greek word Protos and Zoon meaning "First animals are eukaryotic protists. The study of protozoa is of great practical important to man, several species form highly virulent parasites of men and animals, causing various dreadful infectious diseases particularly in the tropical countries. No less than twenty to twenty five different species of parasitic protozoa are known to live in man alone. The knowledge of these parasites is useful from the medical point of view. The knowledge of the portals of entry and means of transmission of these parasites is of vital important from the stand point of preventive medicines. Thus a general study of the phylum protozoa is most essential to understand the parasitic forms and to fight out their menace to mankind, domestic animals and crops.

KEYWORDS: Protozoa, Protista, Protozoology etc.

Introduction:

Protozoan's are unicellular, eukaryotic organisms that are placed in kingdom Protista. The word protozoa comes from the Greek word 'protos and zoon 'meaning "first animal, are eukaryotic Protists. They occur generally as single cells and may be distinguished from other eukaryotic Protists by their ability to move at some stages of their life cycle and by their lack of cell wall Protozoa are predominantly microscopic in size. The majority are between 5 and 250 um in diameter. Colonies of protozoa also occur in a protozoan colony, the individual cells are joined by cytoplasmic threads or are embedded in a common matrix. Thus colonies of protozoa are essentially aggregates of independent cells. The study of these eukaryotic Protists called Protozoology.

Protozoa are found in all moist habitats. They are common in the sea, in soil and fresh water. Free-living protozoa have even been found in the polar regions and at very high altitude. Parasitic protozoa may be found in association with most animals groups. Many protozoa survive in dry conditions by the formation of a resistant cyst, or through dormant stage. For example, the soil amoeba is a resistant cyst in dry weather, is a naked amoeba in moist soil and becomes flagellated when flooded with water. Parasitic protozoa can modify their morphology and physiology to cope with a change in host.

Free-living protozoa are found in a variety of habitats. The factors which influences the distribution and number of free-living protozoa in habitat are, moisture, temperature, light, available nutrient, and other physical and chemical conditions. The vegetative or trophic stages of free-living protozoa occur in every type of salt water, freshwater, sand, soil and decaying organic matter. Evidently, for those protozoa which bear chromatophores and carry out photosynthesis, sunlight is essential.

The protozoa population in an aquatic environment is influenced by the chemical constituents of the water. Some protozoa thrive in water rich in oxygen but low in organic matter (mountain springs, brooks or pounds); others require water rich in minerals. Some grow in water where there active oxidation and degradation of organic matter (the majority of fresh water protozoa such as the ciliates). Still others prefer water with little oxygen but many decomposition products (e.g.; black bottom slim and sewage). Some species have been found to live in both salt water and fresh water.

The chemical nature of protozoa is another important factor that influences the very existence of protozoa in a given body of water. Individual protozoan species requires a certain chemical composition of the water it can be cultivated under experimental conditions, although this may be more or less variable among different forms [Needham et al., 1937].

The unit of measurement employed in protozoology is as in general microscopy, 1 micron [u]which is equal to 0.001mm. The knowledge on morphology of protozoa is rapidly increasing because of improvement in the microscope and of introduction of new techniques and devices. While the ordinary microscopes remain a basic tool for observation, the phase contrast microscopy is widely used as it reveals various intracellular structures in living organisms. Furthermore, since the introduction of thin sectioning, electron microscopy is revealing hitherto unknown structural detail of various protozoa. On the basis of their habitat, protozoa may be divided into free-living and parasitic forms. The vegetative stages of free-living protozoa occur is every type of fresh and salt water.

The body form of protozoa is even more varied, and because of its extreme plasticity it frequently does not remain constant. Furthermore the form and size of a given species may vary according to the kind and amount of food. From a small spheroidal mass up to large highly complex forms, all possible body forms occur.

Structurally, the nucleus is of two types: Vesicular and Compact. The vesicular consist of a nuclear membrane which is sometimes very delicate but distinct nucleoplasm, achromatin and chromatin. Many small amoebac contain a vesicular nucleus. The compact nucleus, on the hand, contains a large amount of chromatin substance and a comparatively small amount of nucleoplasm, and is thus massive.

Cytoplasm is the extra nuclear part of the protozoan body. It is composed of a colloidal system, which may be homogenous, granulated, in the majority of protozoa, the cytoplasm is differentiated into the ectoplasm and the endoplasm. The ectoplasm is a crotical zone which is hyaline and homogenous in Sarcodina and Sporozoa. In the Ciliophora, it is a permanent and distinct part of the body and contains several organelles. The endoplasm is more voluminous and fluid. It is granulated and contains various organellac.

MATERIAL AND METHOD:

The water samples were collected from various water bodies from Mumbai city and its vicinity.

Water sample were collected in plastic bottles and plastic bags or in plastic cans. Care should be taken that water must be collected along with submerged plants, decaying leaves or any other detritus material. Most of the samples were collected in morning time

as the temperature affects the abundance of protozoa and they found more abundant at low temperature.

These samples were brought to laboratory and examined under the microscope for the further study and observation. Water samples were observed directly by taking a water drop on a slide and it was covered with cover slip so that water cannot be dry.

Protozoa are usually swim rapidly in water and hence unable to identify. To immobilize those, 10% methyl cellulose was added to the water drop on slide. This slows the movement of organism without immediate death or bursting.

Culture method:
When protozoa are less abundant in the water sample their population can be increased by culturing them. For cultivation of these organisms following methods are used.

i) Hay infusion ii) Wheat infusion iii) Rice infusion

RESULT AND DISCUSSION:
Systematics of Protozoa:
The protozoa have two subkingdom Biciliata and Sarcomastigophora. The Biciliata includes infrakingdom Alveolata, Exacavata and Rhizaria. The infrakingdom Alveolata has two phyla; phylum Ciliophora and phylum Mycozoa, while infrakindom Exacavata has four phyla, Euglenozoa, Percolozoa, Luokozoa and Metamonada. The subkingdom Sarcomastigophora has two phyla; phylum Amoebozoa and phylum Choanozoa.

The phylum Ciliophora possess cilia, cirri or other compound ciliary structures which serve as organelles of locomotion. Macro and micronucleus are present in all without exception. The phylum Ciliophora has two subphyla; i) Intramacronucleata ii) Postciliodes-matophora.

This subphylum has following eight classes.

1. Spirotrichea
2. Phyllopharyngea
3. Litostomatea
4. Oligohymenophorea
5. Colpodea
6. Prostomatea
7. Nassophorea
8. Plagiophylea

1. Class Spirotrichea (Butschli, 1889)
Body is usually large (Cheng, 1973), Cilia are usually sparse, buccal ciliature is conspicuous with adoral zone typically composed of many membranelles, winding clockwise to cytostome. This class includes subclass Hypotrichia and Stichotrichia. The members of subclass Hypotrichia are flattened and strong cilia or cirri are restricted to ventral surface. The peristome is very large with well developed adoral zone. The cirri or cilia on the ventral surface are called, according to their location frontal (frontal cirri), Ventral (ventral cirri), marginal (marginal cirri), anals (anal cirri) and transversal (transverse cirri).

The subclass Hypotrichia includes order Euplotida. The body of these individual is from small to medium. The shape of these individual is ovoid to rectangular. They are usually free swimming. Frontoventral cirri are present. Oral structure, as for subclasses; during cell division, only the ventral somatic infraciliature is replaced while replication of the dorsal ciliature typically occurs within an equatorial band and within the Parental Kineties. This order has suborder Euplotina, this suborder includes family Aspidiscidae and family Euplotidae. Members of family Aspidiscidae have poor developed adoral zone. The cirri are reduced in number and limited to frontal and anals. Genus Aspidisca belongs to this family. In family Euplotidae have large peristome & ventral. Anal fins with cirri are conspicuous. It includes genus Euplotes.

The subclass Strichotrichia has order Sporadotrichida and order Stichotrichida. Body of the individuals of the order Sporadotrichida is sometimes elongate, even tailed, but often oval to elliptical. The ventral cirri are typically heavy and conspicuous. Marginal cirri are often absent or else reduced. This order include family Oxytrichidae. The individual of this family have clear – cut rows of right and left marginal cirri. Frontoventral and marginal cirri are typically heavy and distinctive. The adoral zone of membranelles generally anterior third of relatively body. The family Oxytrichidae includes the subfamily oxytrichinae and Stylonychinae. Subfamily Oxytrichinae includes genus Oxytricha and Subfamily Stylony chinae includes genus Stylonychia. Members of the order Stichotrichida, often elongate, sometimes very drawn out posteriorly. Ventral cirri are generally small and quite in conspicuous and typically arranged in 3 – 12 longitudinal rows. Marginal cirri are common, transverse cirri are sometimes absent. This order Stichotrichida includes family Spirofilidae, in these individuals Ventral cirri are inconspicuous and in quite numerous or helically spiral rows. The genus Hypotrichidium belongs to this family.

2. Class Phyllopharyngea (de Puytorac et al., 1974)
It is class of ciliate Protozoa, including some which are extremely specialized. The cilia are restricted to the ventral surface, or some part there of arising from monokinetids with a characteristic ultrastructure. Chonotrichs, found. In crustaceans and vase-shaped with, cilia restricted to a funnel leading down into the mouth. The Suctoria lack cilia and were not classified as ciliates.

The mouth of Phyllopharyngea is surrounded by microtubular ribbons, called phyllae. Sub class – Phyllopharyngia, most of which are free living. Body frequently dorsoventrally flattened, with somatic ciliature restricted to ventral surface and often an adhesive organelle at the posterior end. The sub class includes order Chlamydodontida in which Ventral. Ciliature of the organisms is thigmotactic, with no prominent specialized adhesive organelle. Body is broad and flattened dorsoventrally. Body ciliation is confined to ventral surface. It includes family Chilodonellidae has small to large sized. They are free swimming. The anterior preoral kinetal arcs of right ventral somatic kineties are continuous with the more posterior part of those Kineties. Oral ciliature is typically and two circum oral Kineties, but some variations. Macronucleus is centric heteromerous, globular to ellipsoid. Micronucleus is present. Contractile vacuole may be multiple. They are found in fresh, marine and terrestrial habitats, free living but some Chilodonella species as facultative parasites of fishes. This family Chilodonellidae includes genus Chilodonella. During present study one species of this genuss were recorded, Chilodonella unicinata.

Subclass Suctoria has different forms of the body in different species, may be approximately spherical, conical, clubbed – shaped, cylindrical and irregularly branching. Tentacles may be distributed over the surface or they may arise in clusters or extensible arms. Cilia are absent in the mature stage. They are found in fresh and marine water. Tentacles are supported by microtubules and phyllae and have toxic extrusomes called haptocysts at the tip which attach to prey. Suctoria reproduce through conjugation. The genus Astophrya belongs to this sub class.

3. Class Litostomatea (Small & Lynn, 1981)
This ciliated is small to large and their shape is varied. They are free swimming and alveoli are poorly to well developed. Somatic ciliation, holotrichous to sparse in pleurostomes and some endosymbionts. Somatic mono kinetids, typical with laterally directed kinetodesmal fibril that does not overlap those of adjacent kineties, slightly convergent postciliary ribbon and two transverse ribbons, one which is tangential to the kinetosome perimeter and extends anteriorly into the somatic ridge to the left of the kinetid while the other transverse ribbon is radial to the kinetosome perimeter and extends transversely into the adjacent somatic ridge. Oral ciliature as simple kinetids from which nematodesmata arise to support the cytopharynx, but nematodesmata may arise from so called "oralzied" somatic kinetids adjacent to the oral region and in some symbionts, oral ciliature is organized into polykinetid like structures called syncillia.

This class include subclass – Haptoria. The individuals of this subclass has small to large body size and shape is variable. These are free swimming and poorly developed alveoli. Somatic ciliation, holotrichous, but reduced to grirdles in some forms and sparse

in pleurostomus. Somatic kinetids are as for the class, but postciliary microtubules over lapping longitudinally. Oral region typical oval or slit like rarely permanently open, so that the chytopharynx becomes eversible In some species oval dikinetids, rarely monokinetids, on border of cytopharynx. Cytopharynx, typically with outer or posterior kinetosome with a transverse microtubular ribbon that extends anteriorly and reflects posteriorly to support the cytopharynx. Toxicysts localized in or near the oral area, typically between the oral transverse ribbon and bulge microtubular of the rhabdos. Subclass Haptoria include order Pleurostomatida. The members of this order has cytostome ventral or slit like, along edge of laterally compressed body, with nearby ciliature showing left right differentiation. Micronuclei are possibly of low ploidy number. These are carnivorus, voracious and facultatively interstitial (Corliss, 1974). This order includes the family Litonotidae and Amphileptidae. They have slit like cytostome, located anteriorly but not at the anterior tip. Body is flask sheped and laterally compressed. Ciliation is uniform. This family includes genus Amphileptus. The family Litonotidae as synonym for Amphileptidae. The genus Litonotus belongs to the family Litonotidae.

4. Class Oligohymenophorea (de Puytorac et al, 1974)

The class Oligohymenophorea is a larger class of ciliate protozoa. It is typically ventral groove contain the mouth and oral cilia. These include a preoral membrane to the right of the mouth and membranelles, usually three in number, to its left. The cytopharynx is inconspicuous and never forms the complex cyrtos found in similar classes. The body cilia arise from monokinetids, with dikinetids occurring in limited distribution over part of the body. It includes sub class Peniculia, Peritrichia and Scuticociliata.

The subclass Peniculia, oral ciliature is characterized by peniculi located deep in the buccal cavity. They are often with ciliated vestibule. Body is contained uniformly cilia (Kudo, 1966). This subclass Peniculia has the order Peniculida, in which peniculi of oral ciliature deep in buccal cavity. Distinct pre and post oral suture and nematodesmata, but not mucocysts of common occurrence. They are monomorphic, generally microphagous forms without cysts. This order Peniculida includes the family Parameciidae. In these, the oral groove leads into buccal cavity. Buccal ciliature is composed of endoral membrane. The genus Paramecium belongs to this family.In sub class Peritrichia, the Peritrichs shows a much enlarged disc with prominent adoral ciliature which winds counter clockwise to the Cytostome. Telotrochs or free swimming larvae are provided with a ciliary gridle. A sexual reproduction is by binary fission and sexual reproduction by conjugation. This sub class has order Sessilda in which mature individuals are attached to the substrate with Contractile Stalks. This order Sessilida includes family Vorticellidae. In members of family Vorticellidae, all organisms have bell shaped body with contractile stalks, sessile when mature and often colonial. This family includes genus Systilis and genus Vorticella.

The members of sub class Scuticociliata have body typically small to medium in size. Ciliation is uniform, though sometimes sparse, often including specific thigmotactic areas and one or more caudal cilia. The oral ciliature, which may lie in buccal cavity. Stomatogenesis is buccokinetal with complex morphogenetic movements and unique involvement of a scutica. It includes order Philasterida in these members scutica – sestige separate and posterior to paroral. Anterior end is bluntly tapered. It has the family Uronematidae.

5. Class Colopodea (Small & Lynn, 1985)

The colpodea are a class of ciliates, of about 200 species common in fresh water and soil habitats. The body cilia are typically uniform, and are supported by dikinetids of characteristic structure, with cilia on both kinetosomes. The mouth may be opical or ventral with more or less prominent associated polykinetids. Many are asymmentrical, twisting sideways and then untwisting again prior to division, which often takes place within cysts. Most ciliateds placed were originally considered advanced trichostomes, on the assumption that they lacked true oral cilia. It has order Colopodida and the individuals of this order possess some vestibular ciliature organized into two fields of "cirromem-branelles". Stomatogenesis sometimes which discrete perimordium, basically telokinetal. Somatic Kinetids are typically with kinetosomes in pairs, both ciliferous. Macronucleus is possibly of low ploidy. They are found in fresh water habitats.

6. Class Protostomatea (Schewiakoff, 1896)

Somatic monokinetids are usual, with radial transverse ribbon, slightly convergent postciliary ribbon and anteriorly directed kinetodesmal fibril that does not overlap those of other kinetids. The cytostome is apical to sub apical. Oral kinetids are radial to tangential to perimeter of oral area. These individual have rhabdos like cytopharyngeal apparatus. This class has two orders; Prostomatida and Prorodontida. The order Prorodontida, cytostome is apical to sub apical, "Brosse" of kinetid units often as three or more files of dikinetids, varying from parallel to perpendicular to body axis. This order includes family Colepidae. They have brosse as three inconspicuous short files of dikinetids. Body is often with anterior end or posterior spines. The genus Coleps belong to this family.

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