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Description Of Protozoan Ciliate Entodinium Simplex (dogiel, 1925) From The Rumen Of Indian Cattle, (bos Indicus)

S. A. KULKARNI

Associate Professor and Head, Department of Dairy Science Adarsh College, Hingoli. (MS) (India)

Abstract: Rumen fluid samples were collected to study the taxonomy of rumen ciliate protozoa belonging to genus Entodinium, from stomach adult Indian cattle (Bos indicus). The present paper deals with the morphology of Entodinium simplex (Dogiel, 1925) and is described for the first time from rumen of Indian cattle in India. It's Body measurements, frequency distribution and Variation of characters are recorded. Critical comments are made on it's specific identity. The observations are based on a study of 50 specimens taken at random from different samples E.simplex is an medium size elongated species with an elongated macronucleus. The posterior end is slender without any indication of spine or lobes.

Keyword: Cattle, Cilites, Protozoa, Rumen, Entodinium

INTRODUCTION:

Protozoa are unicellular animals. The great majority of ciliates are free living, but a number are parasitic. The protozoa placed in the phylum ciliophora possess cilia, cirri or other compound ciliary structures which serve as organelles of locomotion. Two kinds of nuclei are present in all without exception. Nutrition is holozoic or saprozoic. Asexual reproduction is by binary fission or budding and sexual reproduction is by conjugation or autogamy in which micronuclei play an important role. The ciliates of rumen belongs to the families Buetchliidae, Isotrichidae, Paraisotrichidae, Blepharocarythidae, Ophryoscolecidae, and Cyclopostidae. The ciliates vary in size from a few micron to 2 mm or more in length. The anterior and posterior extremities are permanently differentiated, the rumen ciliates are obligate anaerobes.

Ruminants have a very complex ecosystem harboring a variety of microorganisms which are capable of bringing out diverse types of fermentation. Rumen, the largest of the four compartments of stomach in ruminants, serves as a closed fermentation vat in which ingested feed is attacked by the microflora.

The rumen microflora consist of mainly of bacteria, protozoa and fungi, which have a significant role to play in rumen fermentation. Of the total microbial biomass existing in rumen 40 to 80 percent is of protozoa origin (Jouney - 1991, Punia et al, 1992), Protozoa living in the rumen are essentially ciliates, flagellates are often less numerous, not well known and are often confused with the flagellate stage of fungi (Jouney - 1988) Fermentation of starch and soluble sugars is regulated by rumen protozoa (Mackie et al 1978) and they are held in controlling acidosis in the rumen. Rumen protozoa are generally proteolysis (Balaraman, 1996).

The ciliates are established in the rumen within three weeks after the birth of a calf (Kurar, 1996) provided that the pH is above 6.0. Entodinium population is abundant in the rumen. It increases when the diet is rich in starch. Protozoa contributes about 40 to 60 percent of total hydrolytic enzyme activity in rumen.In ruminants, protozoa were first observed by Gruby and Dalafond in 1843 (Hungate, 1978) Since then a number of protozoal species have been reported in rumen. Subsequently the taxonomic studies on the rumen protozoa was done by various workers in different parts of the world; only a few studies have been carried in domesticated Indian ruminants. Kofoid and MacLennan (1930,1932,1933) in Bos indicus, Das Gupta (1935) in Indian Goat, Ajit Banerjee (1955) in Indian Buffalo. There is much scope to do work on the taxonomy of rumen ciliates. The taxonomical work on rumen ciliates of Cattle in India is very scanty. The present research work deals with study of taxonomy of rumen protozoa from Indian cattle.

MATERIALAND METHODS

Rumen fluid samples were collected for the present study from Indian adult cattle (Bos indicus) slaughtered at abattoirs in Hingoli district of Maharashtra state in India. On the removal of stomach, rumen was slightly punctured and 10ml. rumen fluid was collected in a vial. It was centrifuged and preserved adding 1:1 glycerine:alcohol solution. A drop of this material was taken on a glass slide for observing ciliates in living condition under research microscope. The permanent slides of the samples were made in duplicate, stained by tungstophosphoric haemotoxylein stain. The staining procedure of Krier and Becker, 1987 was followed. The stained slides of ciliates were observed under research microscope for their identification and morphology.

The general features used to classify the rumen

protozoa into genus Entodinium are as follows : (Dehority - 1993)

1.The Presence of single adoral zone.
2.Lack of skeletal plates.
3.Position of the macronucleus which lies between micronucleus and closest body side.

Body measurements such as length, width, L/W ratio, diameter, length of the nucleus etc. were recorded with an ocular micrometer. Frequency distribution, body shape, location of contractile vacuole, rectum mouth are also recorded.

Taxonomical position of Entodinium Stein, 1858.

Subkingdom	:-	Protozoa
Phylum	:-	Ciliphora
Class	:-	Kinetofragminophorea
subclass	:-	Vestibuliferia
order	:-	Entodinimorphida
Family	:-	Ophryoscolecidae
Subfamily	:-	Entodniinae
Genus	:-	Entodinium

The following parameters were considered for observation.
1)Shape of the body
2)Length of the body
3)Width of the body
4)L/W ratio
5)Shape of nucleus
6)Length of nucleus
7)Position of micronucleus
8)Position of contractile vacuole
9)Location rectum and anus

RESULTS AND DISCUSSION

Entodinium simplex (Dogiel, 1925)
(Fig.1a, 1b)

During the present study Entodinium simplex is recorded for the first time in the rumen of Indian cattle (Bos indicus) in India. It's morphology is described, the body dimensions and other measurements are recorded (Table :1) The observations are based on a study of 50 specimens taken at random from different rumen fluid samples of Indian cattle.

MORPHOLOGY :

Entodinium Simplex is an medium sized elongated species with body length more than 35µm. Oral area is small with deep adoral lips. Mouth is 8.43 µm in diameter. L/w ratio is 1.74.

The dorsal and ventral body surfaces are slightly convex, The maximum diameter is in the middle half of the body (21.40 µm). The posterior end is slender without any indication of spine/lobes. The endoplasmic sack is clearly differentiated by a prominent boundary layer closely applied to the body sides Ectoplasm is thin and uniformly distributed everywhere. Rectum is located in the middle of the posterior end, which is prominent and wide; position of anus is slightly ventral.

The macronucleus is elongated wedge shaped body and is closely applied with the dorsal body surface, extending from anterior margin passes the middle half of the body. It is 21.87 µm in length and is 58.73 per cent of the body length. It's anterior end is broad and smoothly rounded (3.48 µm) where as the posterior end is small and pointed (2.29µm); small oval micronucleus is located to the left venral edge of one third length of macronucleus. Contractile vacuole is situated anteriorely left to the macronucleus just above the micronucleus.

COMMENTS :-

Entodinium simplex was firstly described by Dogiel in 1925. Thereafter various workers have reported the species. Hsiung (1932) in Chinese cattle described it as one of the small species of Genus Entodinium. Wertheim, (1935) in Bos taurus and Capra hircus, Das Gupta (1935), from Indian Goat, Lubinsky, (1958) in Reindeer from Canadian Arctic, Dehority (1978), from Cattle and Sheep, Imai (1985), from Bali cattle in Indonesia and Zebu Cattle in Srilanka, Imai and Ogimoto (1984), in zebu cattle from Taiwan and Japan, Imai, (1981) in Japanese cattle and Buffalo from Malaysia, Han (1984) Kang (1989) from Korean Cattle. Shimizu et. al (1983). Tung (1989) in Zebu Cattle from Philippines, Mukharjee (1990) from Indian Goat. Wang (1990) from Goat in Taiwan, Selim et. al (1999) from sheep, cattle and camel in Libya.

It is very difficult to differentiate the small non-caudal spinated entodinic species because of slight differences among their size and morphological characteristics. Ito et al (1993) is of the opinion that this species is similar to E.dubardi. However the shape and size of Macronuclus of E.dubardi has the same thickness in anterior and posterior ends and slight thinner in the central part, where as in E.simplex macronucleus is elongated with broad anterior end and narrow posterior end as reported by Wertheim (1935) and also found in the present study.

A Comparison of the dimensions of the species described here and those given by earlier workers is given in table : 2. Frequency distribution of various size ranges of E. simplex is given in Table : 3.

The table reveals that the species described here is smaller in size as compared to the size reported by earlier workers. The L/w ratio is closer to the ratio given by Wertheim (1935), Lubinsky (1958), and Dehority (1993). However the L/w ratio reported by Ito et al (1994) is smaller.

In the present studies this species is recorded for the first time from cattle in India.

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TABLE – 1
The Body Dimensions and other measurements of Entodinium simplex (Dogiel, 1925) are given below.
(All the measurements in microns)

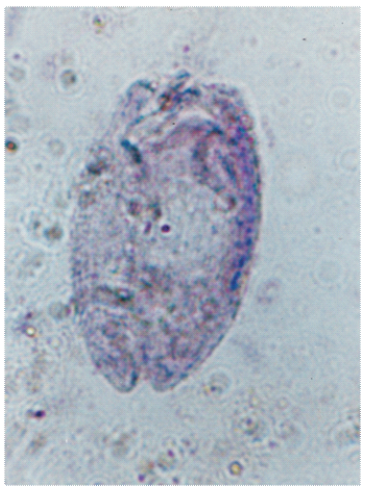
Sr. No.	Parameters	Minimum	Maximum	Average
1.	**Body**			
	Length	**34.24**	**42.80**	**37.24**
	Width	**19.26**	**25.68**	**21.40**
	Length width ratio	**1.78**	**1.67**	**1.74**
2.	**Macronucleus**			
	Length	**17.12**	**25.68**	**21.87**
	Percent length of body	**50.00**	**60.00**	**58.73**
	Dia. Ant. end.	**3.00**	**4.28**	**3.48**
	Dia. post. end.	**2.14**	**3.00**	**2.29**
3.	**Mouth**	**4.28**	**8.56**	**8.43**

TABLE – 2
Comparative Body Dimensions of Entodinium simplex (Dogiel, 1925) with closely related species of Entodinium given by earlier workers and the present dimensions

(in microns)					
Parameters	***Wertheim (1935)***	***Lubinsky (1958)***	***Dehority (1993)***	***Ito et al (1994)***	***Present Author (2013)***
Length	38-50	38-50 (44.22)	38-50 (44.00)	34.0-49.5 (44.9)	34.24-42.80 (37.24)
Width	21-29	21-29 (24.25)	21.29 (24.0)	23.8-42.5 (32.00)	9.26-25.68 (21.40)
L/W ratio	1.7-1.74	1.70-1.93 (1.82)	1.70-1.93 (1.82)	1.13-1.67 (1.41)	1.67-1.78 (1.74)
Macronucleus	--	17-31 (23.51)	--	11.9-34.0 (22.3)	17.12-25.68 (21.87)
Mouth	--	4-97 (6)	--	--	3.0-4.28 (3.48)

TABLE – 3
Frequency Distribution of Various size ranges in Entodinium simplex (Dogiel, 1925)
(In Micron)

Width	Length				
	34.24	**36.38**	**38.52**	**40.66**	**42.80**
19.26	**4**	**--**	**--**	**--**	**--**
21.40	**13**	**5**	**5**	**--**	**--**
23.54	**--**	**2**	**12**	**4**	**--**
25.60	**--**	**--**	**--**	**3**	**2**



(Fig. 1a)

PHOTOGRAPH OF Entodinium simplex (Dogiel, 1925)

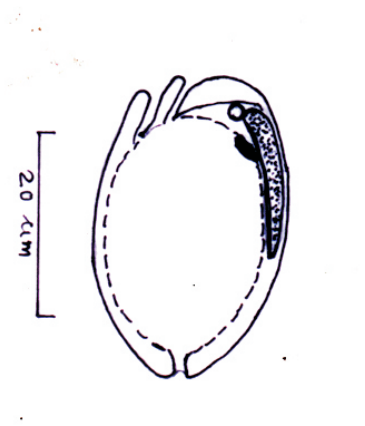


Fig. 1b: Line Drawing of Entodinium simplex (Dogiel, 1925)