CYTOCHEMICAL EVIDENCE FOR THE PRESENCE OF STRONG ACID GROUPS OF POLYSACCHARIDE IN A CILIATE PROTOZOA, *CONCHOPHTHIRIUS LAMELLIDENS AMICRONUCLEATUM*

POLYSACCHARIDE of mammals have been classified into acidic and basic groups according to their capacity to bind the basic dye, *i.e.*, methylene blue[2] at various pH[1]. Presence of weak acidg roups has been reported in mammalian polysaccharide[2] and in the polysaccharide of a ciliate protozoan *Tetrahymena pyriformis* (Strain W)[3]. As a part of a detailed cytochemical study on this ciliate, the study of metnylene blue staining was carried out to assess the quality and characteristics of the polysaccharide.

Conchophthirius lamellidens amicronucleatum is a commensal ciliate in the mantle cavity and on the gills of the common fresh water mussel, *Lamellidens marginalis*[1]. These mussels were collected from Moosi River in Hyderabad, and dissected immediately. The mantle cavity and gills were washed and the ciliates were picked up with a fine pipette, air dried on slides and fixed in 10% neutral formalin. Solution of methylene blue B (5 mM) . was prepared using veronal-actate buffer ranging in pH from 3·0 to 9·0 and the slides were immersed in it for 24 hours.

Capacity to bind methylene blue was demonstrated over a wide range of pH values. Binding of the dye was intense at the pH levels tried. The macronucleus, ectoplasm and basal granules were unstained, the peristomal region and the anterior thigmotactic region had a few bluish granules but the posterior endoplasm was intensity blue. The details are given in Table I.

The mammalian polysaccharide gives maximum staining with methylene blue at an alkaline pH and as the acidity is increased the dye uptake is decreased or absent[2]. In *Tetrahymena pyriformis* (Strain W) birding of methylene blue was very interse at pH levels 9·0, 8·0, 7·0, less at pH 6·0 and 5·0 and absent at pH 4·0 and below[3]. But in the present study the binding capacity of methylene blue in this commersal ciliate protozoan is uraltered as the acidity is increased. It is assumed that acid polysaccharide exists in this organism so that dissociation of acid groups is not suppressed at pH 4·0, 3·5 and 3·0.

TABLE I

Specific areas of methylene blue staining

	pH 9·0	pH 8·0	pH 7·0	pH 6·0	pH 5·0	pH 4·0	pH 3·5	pH 3·0
Macronucleus	–	–	–	–	–	–	–	–
Basal granules	–	–	–	–	–	–	–	–
Ectoplasm	–	–	–	–	–	–	–	–
Peristomal region	+	+	+	+	+	+	+	+
Anterior thigmotactic region	+	+	+	+	+	+	+	+
Region surrounding the macronucleus	++	++	++	++	++	++	++	++
Posterior endoplasm	+++	+++	+++	+++	+++	+++	+++	+++

– Negative; + Positive; ++ Strong; +++ Intense.

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