J. Protozool. Res., 7, 127-132 (1997)
Copyright© 1997, The Research Center for Protozoan Molecular Immunology

Ultrastructure of the nucleus system of *Balantidium coli*

BOGUMILA SKOTARCZAK

Department of Genetics, Faculty of Biology, Szczecin University, Lukasinskiego 43, 71 - 215 Szczecin, Poland.

Received 4 January 1998 / Accepted 26 June 1998

Key words : Balantidiasis; Ciliata; Nucleus system; Electron microscope; Pig.

ABSTRACT

The micronucleus in *Balantidium coli* trophozoites contains no nucleoli. The micronucleus in the electron micrographs was seen to occur close to the macronucleus.

The macronucleus of a *B. coli* trophozoites at the interphase consists of spherical nucleoli, 20 to 40 in a cross-section, irregularly distributed among chromatin aggregates. The number of nucleoli in the macronucleus of *B. coli* trophozoites, isolated from the rectum contents of acute balantidiasis-affected pigs have been higher than from pigs with asymptomatic balantidiasis.

INTRODUCTION

Balantidium coli, the protozoon belonging to Ciliata inhabits large intestine of pigs and many another mammal species.

The vegetative forms of *B. coli* can live in the lumen of large intestine without any significant pathological effect (asymptomatic balantidiasis) or invade mucosa membrane of the intestine (symptomatic balantidiasis). This protozoon can become a dangerous parasite for domestic as well as for wild pigs and for a man (Curie 1990).

Balantidium coli has a nucleus system typical of ciliates, diversified in terms of structure and function, i.e., consisting of macronucleus and micronucleus. According some ultrastructural aspects of macronucleus, this ciliate has been reported (Kraschenninnikov 1968; Rondanelli et al. 1972; Skotarczak and Leonaldi 1986) but described the micronucleus in this paper.

MATERIALS AND METHODS

Pig rectum contents were obtained from the municipal slaughter house. Trophozoites of *B. coli* were isolated from coecum samples of pigs with acute balantidiasis which contained blood. Rectum contents from pigs with asymptomatic balantidiasis showed normal intestine contents.

Sediment of intestine content after several washes in physiological fluid (0.9% NaCl) was observed under the light microscope. Suspensions with plenitude of thophozoites constituted the material for further morphological examination in electron microscopy.

The cells of *B. coli* were washed in 0.9% NaCl, fixed in 2.5% glutaraldehyde in 0.1M cacodylate buffer and stored for 24 hr at a temperature of 4°C. After several rinses in cacodylate buffer, trophozoites were fixed in buffered 1% OsO_4 for 2 hr, and then dehydrated in a graded series of ethanol and embedded in Spurr resin. Ultrathin sections were then examined in a transmission microscope (JEM 1200EX).

RESULTS AND DISCUSSION

The macronucleus of a *B. coli* trophozoite at the interphase consists of spherical nucleoli, 20 to 40 in a cross-section, irregularly distributed among chromatin aggregates (Figs. 1 to 3). The number of nucleoli in the macronucleus of *B. coli* trophozoites, isolated from the rectum contents of acute balantidiasis-affected pigs have been higher than from pigs with asymptomatic balantidiasis. The number of nucleoli in the macronucleus of *B. coli* trophozoites, isolated from the rectum contents of acute balantidiasis-affected pigs ranged from 20 to more than 30 (Figs. 2 and 3), while trophozoites from pigs with asymptomatic balantidiasis showed from 20 to 25 (Fig. 1).

The micronucleus in *B. coli* trophozoites contains no nucleoli, has chromatin in irregular aggregates visible inside. The micronucleus in the electron micrographs is seen to occur close to the macronucleus (Fig. 4).

Kraschennikov (1968) examined the macronucleus of *B. coli* under the electron microscope, and showed the macronucleus of *B. coli* trophozoites to contain three types of chromatin, differing in their electron dense materials and composition of nucleic acids. He described the macronucleus structure as being highly heterometric. It consisted of chromatin located centrally in the macronucleus; this chromatin, called the paramere, was an aggregation of DNA. Another aggregation, of DNA and RNA, was called the orthomere. The third aggregation had the form of RNA-containing nucleoli. However, both the earlier research on ultrastructure (Skotarczak and Leonaldi 1986a; 1986b) and cytophotometric study (Skotarczak 1996; Skotarczak and Zieliński 1997) and this

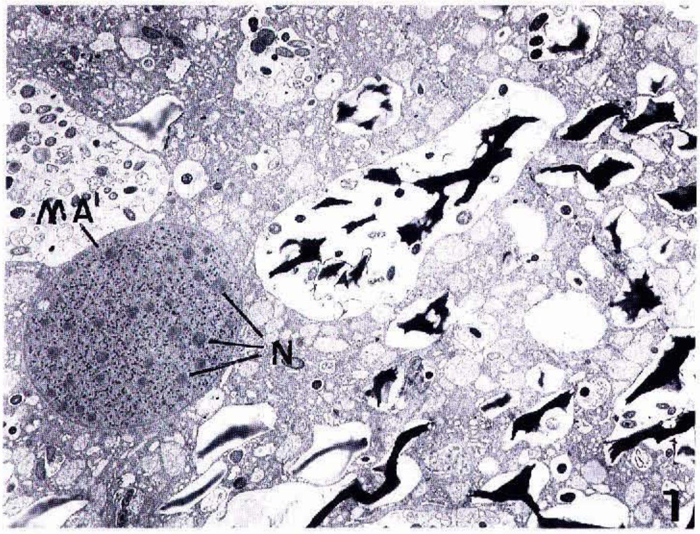

Fig. 1 The part of *Balantidium coli* trophozoite from pigs with asymptomatic balantidiasis. MA: macronucleus, N: nucleoli, magnification: x 3,800.

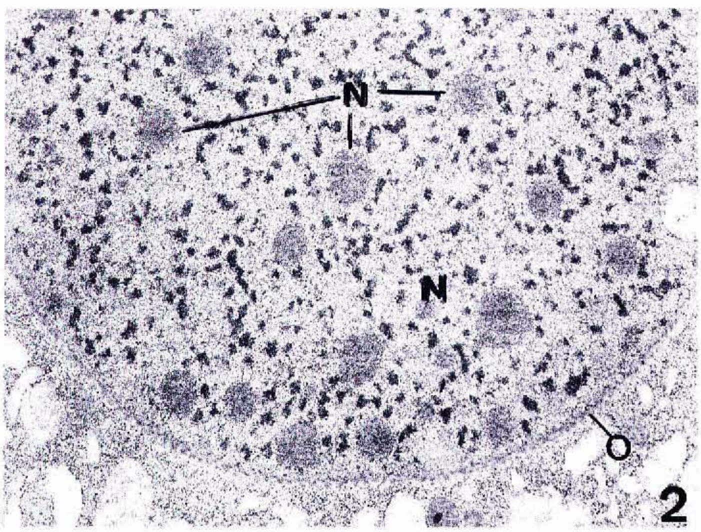

Fig. 2 The part of macronucleus of *Balantidium coli* trophozoite from pigs with acute balantidiasis. N: nucleoli, O: nucleus membrane, magnification: x 25,000.

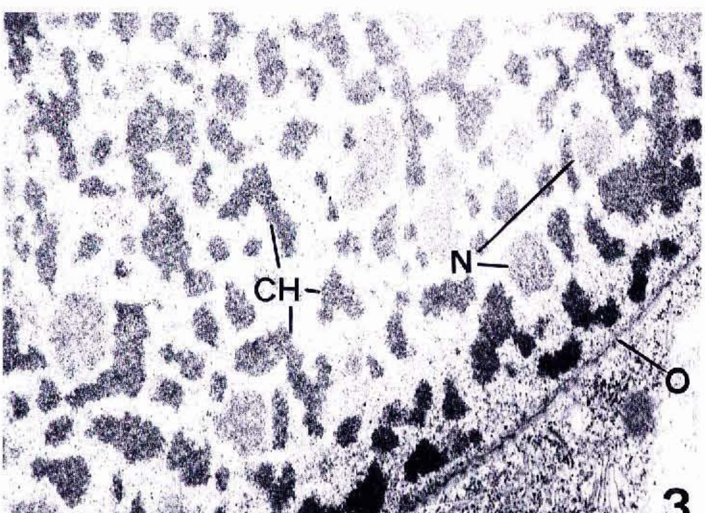

Fig. 3 The part of macronucleus of *Balantidium coli* trophozoite from pigs with acute balantidiasis. N: nucleoli, CH: chromatin,.O: nucleus membrane, magnification: x 50,000.

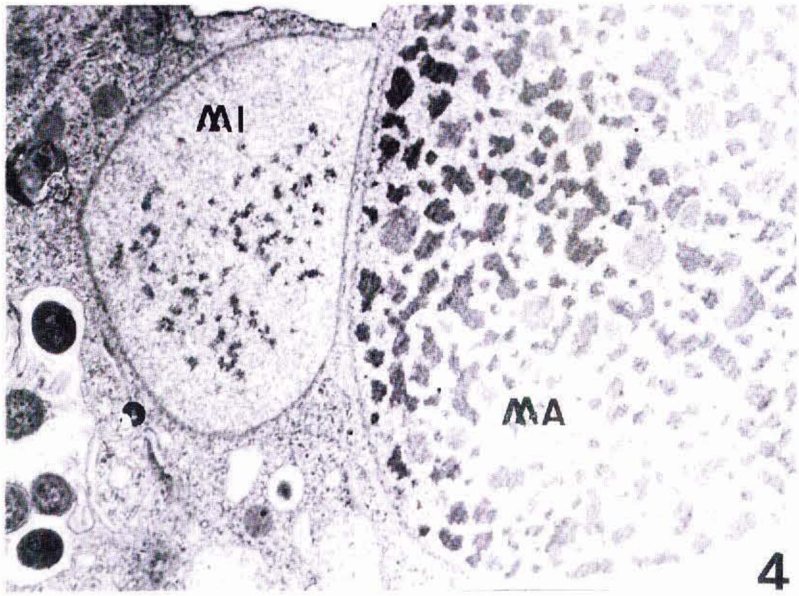

Fig. 4 The part of *Balantidium coli* trophozoite from pigs with acute balantidiasis. MI: micronucleus, MA: macronucleus, magnification: x 12,400.

study failed to demonstrate that the distribution of nucleic acids in the macronucleus of *B. coli* conformed to Kraschennikov's description. The macronucleus of a *B. coli* trophozoite at the interphase consists of spherical nucleoli, 20 to 40 in a cross-section, irregularly distributed among chromatin aggregates (Figs. 2 and 3). The number of nucleoli in the macronucleus of *B. coli* trophozoites, isolated from the rectum contents of acute balantidiasis-affected pigs ranged from 20 to more than 30, while trophozoites from pigs with asymptomatic balantidiasis showed from 20 to 25 (Fig. 1). The higher content of nucleolus RNA in line I trophozoites (i.e., those isolated from acute balantidiasis-affected pigs) seems to stem from the fact that some of the protozoa were degenerating, while others were very active metabolically.

Similarly to most other ciliates (Chakboubrty 1967; Raikhel et al. 1981), the micronucleus in *B. coli* contains no nucleoli. In many ciliate species, at the interphase, the micronucleus membrane converges with the membrane of the macronucleus (Elliott and Clemmons 1966; Elliott 1973; Kaneshiro and Hoitz 1976), which has never been observed in *B. coli*.

The ciliate micronucleus is diploid and undergoes mitotic divisions, while the macronucleus is polyploid and its divisions are most likely amitotic (Orias 1991; Prescott 1995). DNA replication is known to occur prior to the division;

e/g/. DNA synthesis in the micronucleus of *Tetrahymena* (Avilion et al. 1992) takes place immediately after a new cell has been formed, with phase G1 being skipped, and the DNA synthesis in the macronucleus occurs much later. This functional difference between the two nuclei is interesting, considering their co-occurrence in the common cytoplasm and most probably identical access to the DNA synthesis regulators.

The micronucleus ultrastructure of *B. coli* has not been described so far. The micronucleus in the electron micrographs is seen to occur close to the macronucleus. Although irregular in shape on that cross-section, it is most often spherical in other ultra-thin sections, with chromatin in irregular aggregates visible inside.

REFERENCES

Avilion, A. A., Harrington, L. A. & Greider, C. W. 1992. *Tetrahymena* telomerase RNA levels increase during macronuclear development. *Dev. Genet. 1:* 80-86.

Chakboubrty, J. 1967. Electron microscopy of Hypotricha Ciliate Osytricha phatystoma with consideration of its macronuclear organization. *J. Protozool. 1:* 59-64.

Curie, A. S. 1990. Human balantidiasis. A case report. *S. Afr. J. Surg. 28:* 23-25.

Elliott, A. M. 1973. *In:* Biology of Tetrahymena, Stradsburg, Pennsylvania.

Elliott, A. M. & Clemmons, G. L. 1966. An ultrastructural study of ingestion and digestion in *Tetrahymena pyriformis. J. Protozool. 13:* 311-323.

Kaneshiro, E. S. & Hoitz, G. G. 1976. Observation on the ultrastructure of *Uronema* spp. Marine Scuticocliates. *J. Protozool. 4:* 503-517.

NKrascheninnikov, S. 1968. A cytochemical study of *Balantidium coli. Med. Parasitol. 7:* 224-227 (in Russian).

Orias, E. 1991. Evolution of amitosis of the ciliate macronucleus: gain of the capacity to divide. *J. Protozool. 3:* 217-221.

Prescott, D. H. 1995. The DNA of ciliated protozoa. *Microbiol. Rev. 2:* 233-267.

Raikhel, N., Paulin, J. J. & Skarlato, S. O. 1981. Mitosis of micronuclei during division and regeneration in the Ciliate *Stentor coeruleus. J. Protozool. 1:* 59-64.

Rondanelli, E. G., Coros, G., Filice, G. & Scaglia, M. 1972. Ultrastruttura e citochemica di *Balantidium coli. Giornale di Malattie Infettive e Parassitarie 24:* 889-891 (in Italian).

Skotarczak, B. & Leonaldi, R. 1986a. Localizzazione ultrastrutturale della fosfatasi acida in *Balantidium coli* (Malmsten). *Esperienze di Medicina e chirurgia 2:* 109-118 (in Italian).

Skotarczak, B. & Leonaldi, R. 1986b. Ultrastruttura di *Balantidium coli* (Malmsten). *Esperienze di Medicina e chirurgia 2:* 119-125 (in Italian).

Skotarczak, B. 1996. Cytophotometric measurements of nucleic acids in *Balantidium coli*. *Wiad. Parazytol. 42:* 159-169 (in Polish).

Skotarczak, B. & Zielinski, R. 1997. A comparison nucleic acid contents in *Balantidium coli* trophozoites from different isolates. *Folia Biol.* in press.