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Research Note

ISOLATION OF *TRYPANOSOMA FREITASI* (KINETOPLASTIDA: TRYPANOSOMATIDAE) FROM *PSYCHODOPYGUS CLAUSTREI* (DIPTERA: PSYCHODIDAE)

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Trypanosoma (Megatrypanum) freitasi is a stercorarian trypanosome occasionally found in the blood of the opossums *Didelphis albiventris* (syn. *Didelphys paraguayensis*) (Rego et al., 1957, *Rev. Brasil. Malariol., 9:* 277-284) and *D. marsupialis* (Deane, 1964, *Rev. Inst. Med. Trop. São Paulo, 6:* 225-232). In the latter species reproductive stages have been reported from the lumen of the anal scent glands (Deane & Jansen, 1986, *Mem. Inst. Oswaldo Cruz, 81* (Suppl.): 53). As with most species of the subgenus, little is known of the vectors, except that all attempts to infect triatomine bugs have failed.

One example of the phlebotomine *Psychodopygus claustrei* caught in a rodent-baited castor oil trap in Balbina, Amazonas State, had a moderate infection with trypanosomatid flagellates in the hind triangle and posterior portion of the midgut. There was no vertebrate blood in the midgut and development of the ovaries corresponded to Christophers' stage III. The flagellates were isolated in NNN Difco rabbit blood agar culture medium. Thirty five days after isolation, 0.5 ml of first passage culture with abundant parasites was inoculated subcutaneously in a 350 g *D. marsupialis.* Haemoculture (10 tubes) and microscopy were negative prior to inoculation. This animal had been captured as a 19 g juvenile still in the mother's marsupium.

On day 3 after inoculation a patent parasitaemia was detected in fresh blood films. Although smaller than in the original description, parasites were identified as *T. freitasi* on the basis of the experimental host and morphology of the bloodstream forms (see Figs 1-5): length including distinct free flagellum 29.5-47.2 μm, width 3.0-6.9 μm, nucleus central, kinetoplast marginal and far from posterior end of body, kinetoplastic index 3.0 (2.6-3.6). Parasitaemia reached a peak of 3-6 organisms per microscope field (X 40 objective) on day 9, and was still patent on day 25, but repeated examinations after day 30 were negative, as was haemoculture after 3 and 6 months and culture of secretion from the anal glands after 6 months.

We consider the present findings strong circumstancial evidence that Phlebotominae are intermediate hosts and vectors of *T. freitasi.* It should be noted, however, that Rego et al. *(loc. cit.)* considered a similar trypanosome from *Didelphis marsupialis aurita* to represent a distinct (unnamed) species. The transient nature of the experimental infection in the opossum may explain why this trypanosome is not more frequently seen in the vertebrate host.

Parasite material including cryopreserved culture deposited under the reference code IM-3145; insect host as slide number 377 Balbina 29/04/87; both in the authors' laboratory. Fig. 1 by Artêmio Coelho da Silva, from Giemsa-stained slides.

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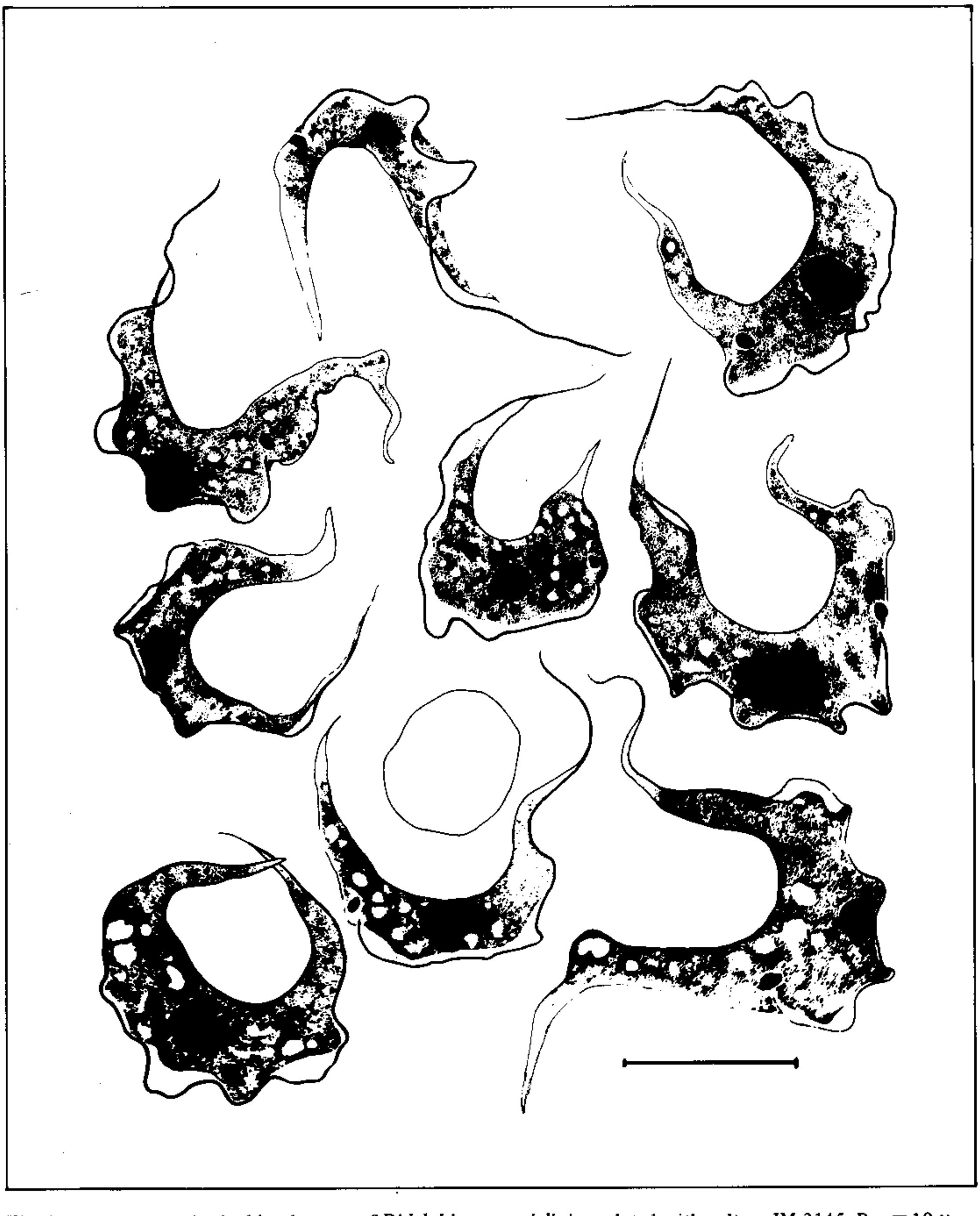
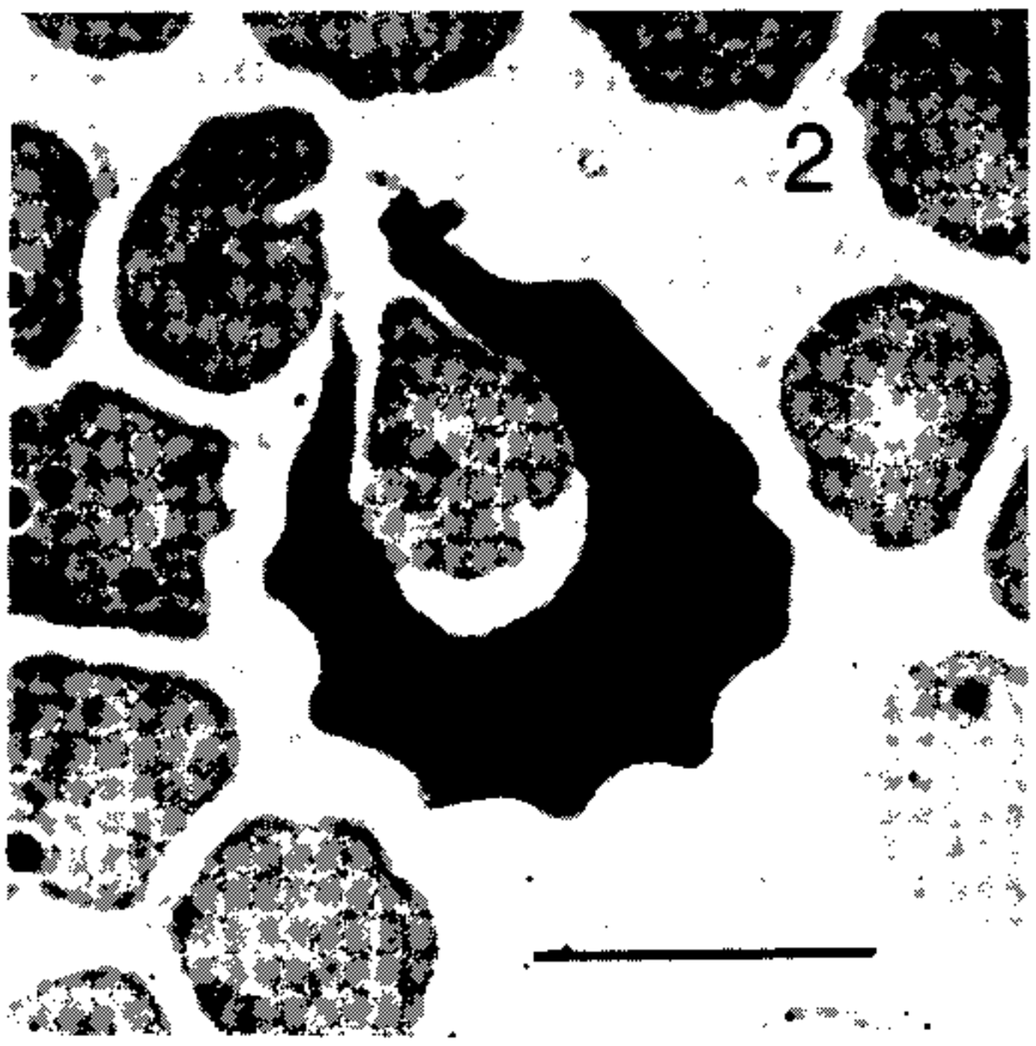
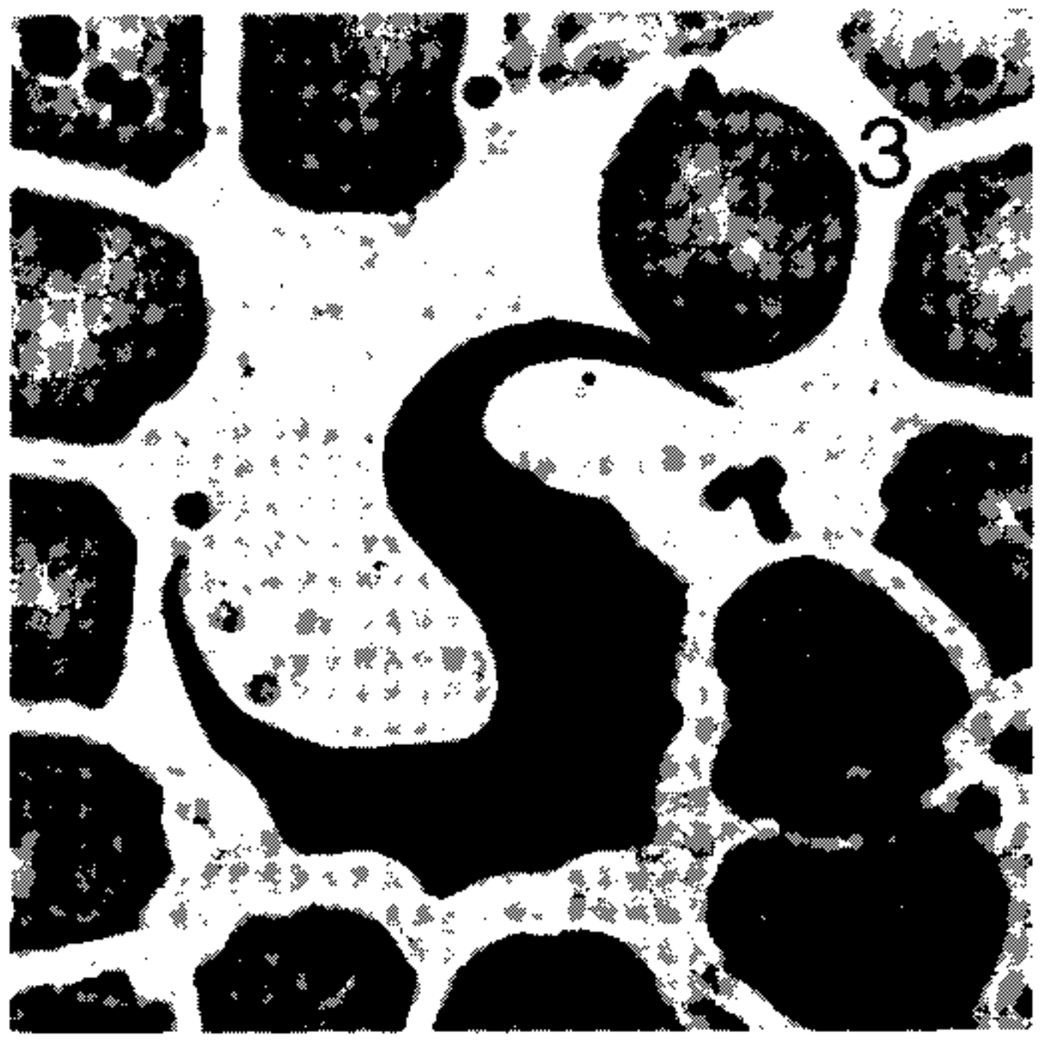
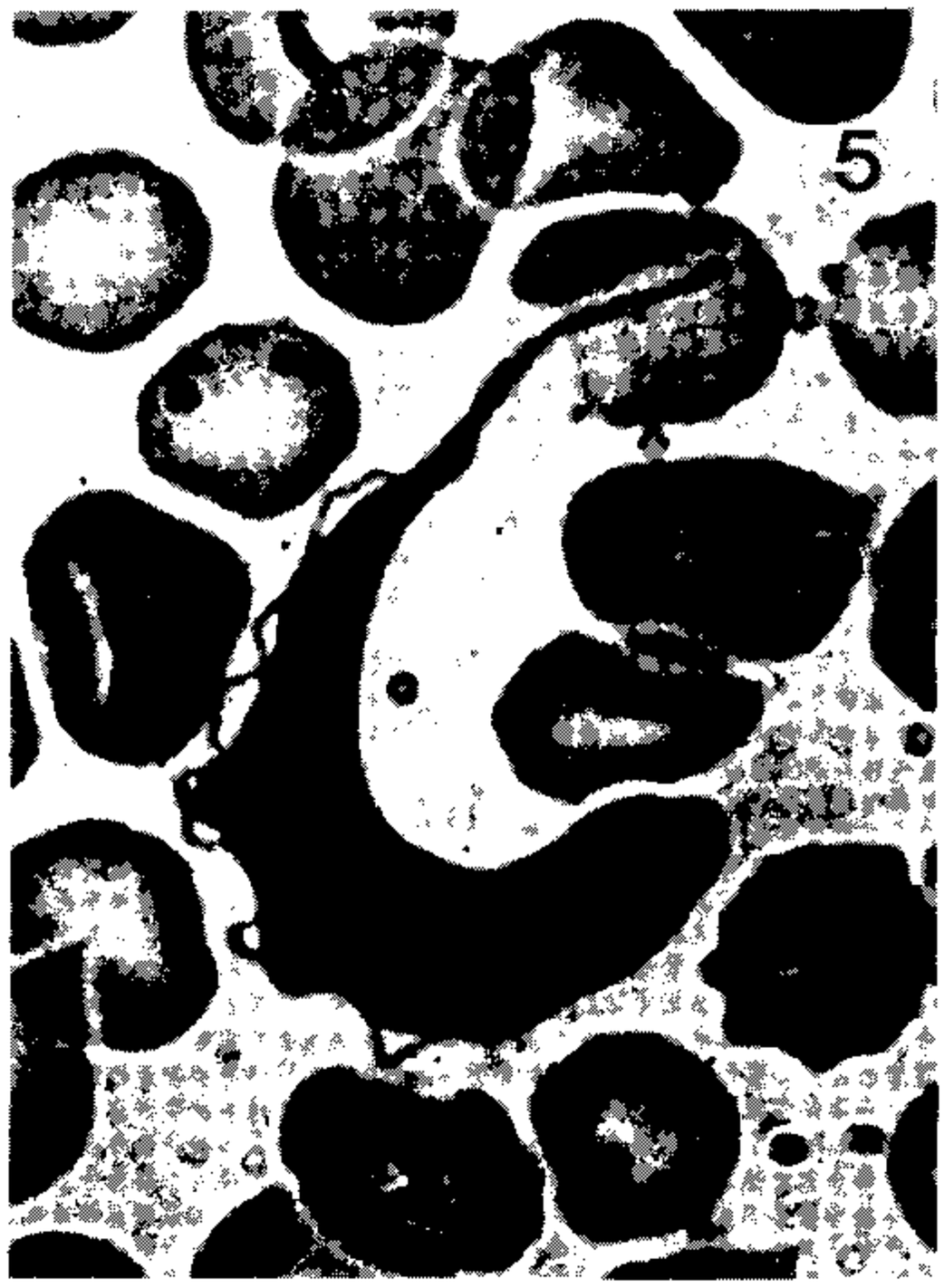


Fig. 1: trypanosomes in the bloodstream of *Didelphis marsupialis* inoculated with culture IM-3145. Bar = 10 μm.







Figs 2-5: trypanosomes in the bloodstream of *Didelphis marsupialis* inoculated with culture IM-3145. Giemsa. All to same scale. Bar in Fig. 2 = 10 μm.