Trypanosoma kansensis sp. n. from Neotoma floridana in Kansas

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ABSTRACT: Trypanosoma kansensis sp. n. (Sarcostagi nophora: Kinetoplastida) is described from three of 23 (13%) eastern woodrats (Neotoma floridana) collected from Pottawatomie County, Kansas (USA). All bagged females found in the blood of woodrats were trypanomatiges and are larger than T. neotomae in overall dimensions, especially flagellar length and the distance between the posterior end of the organism and kinetoplast. Liver infusion-trypomastigote (L.I.T) cultures of infected whole blood resulted in the transformation of some parasites into epimastigotes; however, there was no apparent increase in parasite numbers.

Key words: Trypanosoma kansensis sp. n., Trypanosoma neotomae, Sarcocystis gophorae, Kinetoplastida, Trypanosoma, eastern woodrat, Neotoma floridana, LIT culture, survey.

The only species of trypanosore known to infect woodrats is Trypanosoma neotomae, first described by Wood (1956) from Neotoma fusipes in California (USA). Although similar to T. lewisi from Batti tus spp., T. neotomae was determined to be a distinct species since it was not transmissible to susceptible Norway rats (Battius norvegicus) and was capable of infecting Neotoma fusipes while T. lewisi was not. Also, T. neotomae was found to have a longer incubation period and persisted in the blood longer than T. lewisi in respective hosts (Wood, 1956). The woodrat flea (Ochropterus houardi = O. n. hilli (Hill)) was found to be the intermediate host with epimastigotes ("critihicks") and trypanosomites ("trypanosomes") within its intestines. Additional hosts reported to be infected with T. neotomae are N. lepida from California and N. albiculaeae from a locality north of Guaymas, Sonora, B. Mexican (Wood and Wood, 1937; Wood, 1952, 1972).

Between June and August 1986, we collected 23 eastern woodrats (Neotoma floridana) from Pottawatomie County, Kansas (USA; 39°13′N, 96°30′W) as part of a survey to determine whether Trypanosoma cruzi was present in central Kansas. All animals were trapped using Tomahawk live-traps (Tomahawk Live Trap Company, P.O. Box 523, Tomahawk, Wisconsin 54487, USA) and brought back to our laboratory at Kansas State University (Division of Biology, Ackert Hall, Manhattan, Kansas 66506, USA). Animals were killed by CO2 asphyxiation, the thoracic cavity exposed, and blood was removed from the right ventricle by needle and a 10 cc syringe containing 10 units of heparin sulfate. Approximately 0.2 ml of whole blood from each animal was placed into each of two 25 cm2 tissue culture flasks containing 5 ml liver infusion-trypomastigote (LIT) medium. The medium formulation was that described by Castellani et al. (1967), except that 0.25 mg/ml haemin was substituted for hemoglobin. All cultures were incubated at 25 C and examined at weekly intervals for a period of up to 1 mo. In addition, both thin and thick smears were made from the whole blood, air dried, fixed in 100% methanol and stained with Giemsa stain.

Although we failed to note any T. cruzi, a species similar to, but distinct from, T. neotomae was found in blood smears of 3 of 23 (13%) N. floridana. Only trypanomatiges were found in the blood of our study, which agrees with the information provided by Wood (1936) on T. neotomae except that she did note a single trypanomatige with two kinetoplasts in one blood smear. Trypanomatiges from N. floridana were found to be elongate, with a pointed, tapering posterior end and nucleus located anterior to the center of the body (Figs. 1, 2). Measurements (in μm) reported as the
mean ± SE (range) from 25 trypanomastigotes from Griesma stained smears of two individual woodrats (n = 15 and 10, respectively), are as follows and were made using a calibrated ocular micrometer and an Olympus BH-2 microscope equipped with a 100× Planar Apochromatic objective lens (Olympus Corporation, Tokyo, Japan).

Trypanosoma kamaensis sp. n. (Figs 1-3)

Description of trypanomastigote: (Sercomastigophora: kinetoplastida) with characters of the genus Trypanosoma. Total body length (TL), 32.3 ± 0.22 (29.6 to 34.0), length of body excluding flagellum (BL), 23.4 ± 6.21 (21.6 to 25.6); width of body (BW), 2.2 ± 0.05 (1.6 to 2.4); free flagellum length (FL), 9.0 ± 0.23 (0.4 to 11.2); body length (BL)/flagellum length (FL), 2.7 ± 0.06 (2.0 to 3.8); nucleus length (NL), 2.7 ± 0.06 (2.2 to 3.2); nucleus width (NW), 1.7 ± 0.02 (1.6 to 1.8); nuclear index (NI) = NL/NW, 1.6 ± 0.05 (1.2 to 2.0); posterior end to nucleus (PN), 15.1 ± 0.17 (13.6 to 17.4); posterior end to kinetoplast (PK), 5.4 ± 0.15 (4.8 to 7.6); kinetoplast length (KL), 2 ± 0.04 (0.8 to 1.6); kinetoplast width (KW), 1.0 ± 0.02 (0.9 to 1.2); kinetoplast index (KI) = KL/KW, 1.3 ± 0.05 (1.0 to 2.0).

Other stages: Unknown.

Type host: Eastern woodrat, Neotoma floridana (Rodentia: Cricetidae).

Type locality: Pottawatomie County, Kan.

Discussion: The total body length of the form we observed is slightly greater than that reported by Wood (1936) for T. neotormae (32.3 ± 20.4 mm). Additional measurements are lacking in the original description, supplemental data are given by Davis (1952) where she examined blood smears from three N. fuscipes; two of which were collected by herself and the third by F. D. Wood. The means calculated from 50 individual parasites from each host include total body length of 26.2, 28.1, and 30.9 mm, posterior end to kinetoplast length of 2.7, 3.5, and 4.4 mm, and free flagellum length of 4.9, 4.4, and 4.8 mm. Based on these data, the greatest differences between the form we observed and T. neotormae is that the kinetoplast is located further from the posterior end of the body in T. kamaensis and the flagellum of T. kamaensis is considerably longer (BL/FL = 2.7 versus 3.3, 6.4, and 6.5).

Although trypanosomes inoculated along with whole blood failed to reproduce in LIT cultures, the parasites remained viable in the medium. At 2 wk postinoculation, smears were made from two of the cultures and processed as described above for blood smears. Both trypanomastigotes and epimastigotes (Fig. 5) were noted, as were three specimens each with two kinetoplasts. Of 100 trypanosomes scored for specific morphologic type, 47% were trypanomastigotes, 42% were
epimastigotes, 10% were a form intermediate between trypanomastigote and epimastigote where the kinetoplast was immediately adjacent to the nucleus, and 1% was a form possessing two kinetoplasts but a single nucleus and flagellum. More advanced stages of division were not noted, even after cultures were held in the laboratory for 1 mo.

Based on the above measurements, we believe the form from *N. bordeus* represents a previously undescribed species of the genus *Trypanosoma* and assign it the name *Trypanosoma kansasii*, sp. n. The specific epithet is derived from Kansas, the site of the type locality.

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**LITERATURE CITED**


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