

ABSTRACT

Gregarines (Apicomplexa: Gregarinasina) are monoxenous parasites of invertebrates. Those found in sand flies (Diptera: Psychodidae) and mosquitoes (Diptera: Culicidae) used to be considered a single eugregarine genus *Ascogregarina*. Our phylogenetic analyses of the gregarine SSU rDNA, including newly obtained sequences of three species from sand flies, showed that mosquito and sand fly gregarines are closely related to neogregarines, and most importantly, they form two disparate monophyletic groups. Based on these molecular features, accompanied by biological differences, we established a new genus *Psychodiella* for the gregarines from sand flies, reserving the genus *Ascogregarina* for the mosquito gregarines.

In the new genus, two new species *Psychodiella sergenti* from *Phlebotomus sergenti* and *Psychodiella tobbi* from *Phlebotomus tobbi* were described. They differ in the life cycles (sexual development of *Ps. sergenti* is triggered by a blood meal intake) and morphology of their life stages, mainly oocysts. The susceptibility of five sand fly species to both gregarines showed their strict host specificity, as they were able to fully develop and complete the life cycle only in their natural hosts.

The life cycle of *Ps. sergenti* was studied in detail using various microscopical methods. Oocysts are attached to the chorion of sand fly eggs. Sporozoites, with a three-layered pellicle and mucron, attach to the 1st instar larval intestine but are never located intracellularly. In the 4th instar larvae, the gregarines occur in the ectoperitrophic space and later in the intestinal lumen. In adults, the parasites appear in the body cavity, and the sexual development of *Ps. sergenti* takes place only in blood-fed females; gametocysts attach to the accessory glands and oocysts are injected into their lumen.

Psychodiella sergenti was proven to have a negative impact on its host; the infection significantly decreases the survival of various sand fly stages; however, it has no negative effect on the blood-fed female fecundity and mortality. A tenfold increase in the infection dose (5 vs. 50 gregarine oocysts per one sand fly egg) leads to roughly a tenfold, twofold and threefold increase in the number of gamonts in the 4th instar larvae, in females and males, respectively.

Even though sand fly gregarines are pathogenic parasites with interesting biology, they are not given the attention they deserve. This work attempts to fill in the missing information by giving a comprehensive insight into the sand fly gregarine taxonomy, host specificity, life cycle and pathogenicity.