

Susceptibility of Tsetse to Trypanosome Infection in Relation to the Levels of Midgut Trypsin, Trypsin-like Enzymes and Lectins

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Abstract—Post feeding midgut trypsin and trypsin-like enzyme levels, trypanosome transformation and infection rates were compared in *Glossina morsitans*, *G. longipennis* and *G. fuscipes fuscipes*. The abilities of midgut homogenates of different tsetse species to agglutinate *Trypanosoma brucei* was also studied. The results showed that peak enzyme levels in the three *Glossina* species occurred between 48 and 72 h after feeding, with *G. morsitans* the lowest. No significant differences were observed among the three species in their abilities to transform *T. brucei*. Midgut homogenates of *G. longipennis* and *G. fuscipes* had greater capacities to agglutinate trypanosomes than that of *G. morsitans*. Mature *T. brucei* infection rates in *G. longipennis* were less than those in *G. morsitans* and *G. fuscipes fuscipes*. It was concluded that differences in trypanosome infection rates between the *Glossina* species may be related to levels of blood meal-induced trypsins and lectins.

Key words: *Glossina* spp., trypanosomes trypsin, lectin

Introduction

Biomedical factors secreted in the *Glossina* midgut in response to a bloodmeal are involved in the establishment of *Trypanosoma* species in the midgut. These include proteolytic enzymes (Cheeseman and Gooding, 1985), lectins/ agglutinins (Stiles *et al.*, 1990) and trypanolysins (Molyneux and Stiles 1991). Trypsin induces trypanosome differentiation at optimal levels and destroys the parasites at high concentrations (Imbuga *et al.*, 1992). Trypsin constitutes over 50% of the total proteolytic activity in the midgut (Gooding, and Rolseth 1976). Separation of tsetse midgut homogenate by ion exchange chromatography gives two trypsin activity peaks. The unbound trypsin and the bound, trypsin-like enzyme fractions (Abubakar *et al.*, 1995)

The role of midgut trypsin on trypanosome development has been established. However its functional differences among *Glossina* species is still not fully understood. In this study the midgut trypsin levels and infection rates were

compared in the *Glossina morsitans*, *G. longipennis* and *G. fuscipes fuscipes*.

Materials and Methods

Glossina morsitans and *G. fuscipes* were obtained from a colony at the International Centre of Insect Physiology and Ecology (ICIPE). The *G. longipennis* were provided by the International Livestock Research Institute (ILRI). The flies were maintained on rabbit blood *in vivo* at 25-26°C, 65 – 70% RH and a 12h light: dark cycle. *Trypanosoma brucei brucei* EATRO 1969 was used in this study. The trypanosomes were maintained in Wistar rats and the parasites isolated in a Percoll gradient and suspended in a phosphate saline glucose (PSG: 0.15 M phosphate, 0.1M NaCl containing 10% glucose).

Midgut samples of flies fed on infected blood were teased in 0.1 Tris / HCL buffer, Ph 8.0 and the supernatant fraction used in trypsin assays. Enzyme activity was determined using Chromozym - TRY, Serva (Imbuga *et al.*, 1992)

Agglutination assays were carried out using the methods described by Abubakar *et al.*, (1995). Infection rates were also assessed in the three different species.

Results and Discussion

Glossina species showed significant variation in peak activities, which were, 12.0- 21.25-, and 30.0- $\mu\text{mol/mg/min}$ in *G. morsitans*, *G. longipennis* and *G. fuscipes fuscipes* respectively. However, the profiles were similar up to 24 h post-feeding. Peak activities in all the three species occurred between 48 and 72 h after a bloodmeal. Midgut homogenates of *G. morsitans*, *G. longipennis* and *G. fuscipes* agglutinated both bloodstream and procyclic *T.b.brucei*. Agglutination titres for bloodstream form trypanosome were 128, 512, and 1024 for *G. morsitans*, *G. fuscipes* and *G. longipennis* respectively. The titres for the procyclic parasites were 512, 4096 and 4096 respectively.

Lectin activity in the midguts of different *Glossina* species, as indicated by the ability of their midguts homogenates to agglutinate trypanosomes, was correlated with midgut trypsin activity. It has been observed that trypsin and lectin are secreted together in the tsetse midgut as a bifunctional molecule, and this may be a reconciliation of the views regarding the roles of lectins and trypsins in

trypanosome differentiation and lysis (Maudlin, 1991, Abubakar *et al.*, 1995)

There were variations among *Glossina* species in their infection rates with trypanosomes. Mouthparts infections with *T.b. brucei* were greater in *G. morsitans* than either *G. longipennis* or *G. fuscipes*. However midgut infection rates were similar in all the species ($P > 0.05$). Although *G. longipennis* had lower rates of salivary gland infections than *G. morsitans*, the infection rates in the latter were similar to those in *G. fuscipes*. It is suggested that the higher trypsin and lectin activities in the midguts of *G. longipennis* and *G. fuscipes*, compared to *G. morsitans* midgut may be related to the greater refractoriness of the former two species to trypanosomes, compared with *G. morsitans*.

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