

**FIB-SEM REVEALS THE MORPHOLOGICAL PECULIARITIES OF
TRYPANOSOMA BRUCEI DIFFERENTIATION IN THE PROVENTRICULUS OF
TSETSE FLY**

**Moara Lemos, Eloise Bertiaux, Adeline Mallet, Thierry Blisnick, Brice Rotureau,
Philippe Bastin**

**Trypanosome Cell Biology & INSERM U1201
Institut Pasteur, 25-28 Rue Dr. Roux, Paris
E-mail: moara.lemos@pasteur.fr**

KEY WORDS: *Trypanosoma brucei*, tsetse fly, developmental stage, cell morphology, FIB-SEM, cell cycle

Trypanosoma brucei is the protozoan flagellated parasite responsible for sleeping sickness in Sub-Saharan Africa. There is no vaccine available and drug treatments are difficult to apply in rural areas and can promote severe side effects. *Trypanosoma brucei* possesses a complex life cycle alternating between tsetse flies and mammals including cattle and humans. Flies become infected during a blood meal on an infected mammalian host. In the fly, trypanosomes undergo several steps of proliferation and differentiation in different tissues. Here, we have characterized the stages of life cycle that take place in the proventriculus of tsetse flies by Focused Ion Beam Scanning (FIB-SEM). This is a critical phase of the life cycle because it prepares trypanosomes for the migration to the salivary glands, where mammal-infective parasites will be produced. We turned to FIB-SEM a powerful technique that allows trimming a fixed parasites over several μm to provide the 3D reconstruction in a larger number of cells. Trypanosomes possess a nucleus and a single mitochondrion whose DNA is condensed in a structure named kinetoplast and their relative positions defines the trypanosomes morphotypes. In the proventriculus, we observed a large range of trypanosome cell morphologies, the long trypomastigotes in which the kinetoplast is posterior to the elongated nucleus that occupies a large extension of the cell body. The epimastigotes in which kinetoplast is located between the nucleus and the anterior end of the cell, possess a long or short cell body with drastic differences in flagellum length, originated by the asymmetric division. The proventricular trypanosomes undergo an intense proliferative cell cycle by duplication of the single-copy organelles.