## 3D microscopy applied to the study of helminth taxonomy

Ana Paula Nascimento Gomes<sup>1,3</sup>, Eduardo José Lopes Torres<sup>3</sup>, Kildare Miranda<sup>1,2</sup>

<sup>1</sup> Laboratório de Ultraestrutura Celular Hertha Meyer, Instituto de Biofísica Carlos Chagas Filho and Centro Nacional de Biologia Estrutural e Bioimagem - UFRJ, Rio de Janeiro, RJ, Brazil

<sup>2</sup>Centro Nacional de Biologia Estrutural e Bioimagem CENABIO, Unidade de Microscopia Avançada – UMA.

<sup>3</sup>Laboratório de Helmintologia Romero Lascasas Porto, Departamento de Microbiologia, Imunologia e Parasitologia, UERJ, RJ, Brazil.

The study of the helminth parasites taxonomy has been carried out with different microscopy techniques, providing morphological and ultrastructural information that are important to the morphological characterization and species identification. However, these tools present limitations, especially with helminth thick and complexity morphology, for example in depth of focus, and unable viewing of the internal and external structures of the parasite simultaneously. Furthermore, the images obtained are two-dimensional (2D) axis, highlighting the difficult to resolve complex and multidimensional structures. The present study aims to show the advantage of using fluorescence stereomicroscopy with structured illumination, as a microscopy tool that produce results and images with simultaneous registration of the surface and internal structures of helminths, as well enabling a 3D reconstruction and modeling. We analyzed the nematode Globocephalus sp. (Ancylostomidae) and the trematode Echinostoma paraensei (Echinostomatidae) using the fluorescence stereomicroscopy with structured illumination. Our results showed images of the surface and internal structures simultaneously, as well the 3D reconstruction and modeling of the species. Structures with taxonomic importance of Globocephalus sp., such as anterior end, giving a ventral and dorsal view of the rounded cephalic end, circular opening of the mouth and buccal capsule with two pairs of teeth. Echinostoma paraensei it was also possible to visualize the internal structures of the ventral and dorsal faces, such as the seminal vesicle, cirrus, oral and ventral suckers, spines on the peristomal sucker and spines along the body. This technique brought important advances in taxonomy, which allowed a better characterization of the surface and internal structures of helminths, enabling the 3D modeling of the parasite. Exploring 3D modeling that can help to explore in more detail morphological features easily, especially in more complex descriptions of cryptic species. Our study shows that fluorescence stereomicroscopy can open new possibilities to study of helminths, especially with thick specimens, in addition to enabling the reconstruction/modeling of 3D structures of the sample.