

Life cycle of *Metarhabditis* spp. and a proposal for a new experimental model.

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Helminths, mainly nematode infections, are a global health problem, infecting humans and other animals, affecting people of low-income countries with problems in public infrastructure of health and education, combined with a tropical weather. Parasite infections are neglected diseases without governmental and private investments. Cattle raising is one of the most important of agricultural sectors in Brazilian economy, frequently impacted with helminth infection. The nematodes *Metarhabditis* spp. are associated with parasitic infection in *Gir* cattle, causing parasitic otitis, an inflammatory process in the auditory canal, resulting in damages of the auditory tissues, pain, reduced appetite, loss of productivity, and in the serious cases, affecting the nervous system of the animal, leading to death. This and other parasites can cause losses and great economic losses due to the fall in animal production and costs with control. The administration of effective anthelmintics and in correct dosage is of great importance, because the misuse is probably the cause of infection maintenance and the emergence of resistant helminth populations. These factors contribute to the relevance of the studies of the development of new drugs/molecules test models for anthelmintic action experiments. Experimental metazoan models, to explore mammals' models (*in vivo*) or the free-living nematode *Caenorhabditis elegans* in *in vitro* experiments. An alternative *in vitro* parasite metazoan model can be open a more accessible and less expensive experiments supporting an increasing of the number of groups working with anthelmintic drug discovery. In this work, we propose to maintain the *in vitro* experimental model the species, *Metarhabditis blumi* obtained from the Caenorhabditis Genetics Center (CGC) and *Metarhabditis costai*, isolated of *Gir* cattle ear from farm at Rio de Janeiro state. We cultivate these nematodes *in vitro* in our laboratory, based on similar *C. elegans* cultures. The samples of *M. blumi* (DF5010) and *M. costai*, were washed in PBS, centrifuged and transferred to nematode growth medium (NGM) cultures in Petri dishes supplemented with *E. coli* (OP50). The plates were incubated in a BOD-type incubator at 22°C for up to 15 days. For use in drug tests, we synchronized the development stages, using hypochlorite solution (1%) bleaching of gravid worms associated with sodium hydroxide solution (NaOH, 5N), isolating only eggs. The eggs are subsequently re-cultured in NGM in a BOD incubator at 37°C for up to 48 hours. To describe the life cycle, samples were fixed in Karnovsky's solution in different time points and analyzed using light and scanning electron microscopy. We performed the morphometry and morphology characterization of eggs, the larval stages and adult worms. These results showed a detailed description of life cycle of those nematodes, helping different experiments of biological development and anthelmintic molecules drug discovery tests in our laboratory.