Nematodes *Metarhabiditis* spp. as new *in vitro* model for anthelmintic drug tests

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Helminth infections have medical and veterinary importance, resulting in social and economic impacts. *Metarhabiditis* spp. are nematodes associated with parasitic activity in cattle, causing infection and inflammatory process in the auditory canal, resulting in an intense inflammatory process, and in the most serious cases, affecting the nervous system of the animal, leading to death. There are reports of human infection associated with rural workers. Current anthelmintics have limitations in the effectiveness of the treatment and control of bovine parasitic otitis caused by these nematodes. The aim of the study was to develop and standardized the *in vitro* experimental model *Metarhabiditis* spp. for anthelmintic tests. The matrix of the nematodes was obtained from the *Caenorhabiditis* Genetic Center (USA), cultivated in a Nematode Growth Medium (NGM) culture medium, supplemented with *Escherichia coli* OP50 strain, in a BOD-type incubator, according to the protocol established by Brenner and adapted by Sternagle, 2006. Initially, we standardized the *in vitro* growth of *Metarhabiditis* spp. enlarging and replicating samples. The nematodes were treated with the reference anthelmintic drugs, Albendazole 16 µM (ABZ) and Ivermectin 2,5 µM (IVM), within a period of 24 to 48 hours. Larvae (L3) and adult's motility were analyzed using video microscopy (live samples) and part of the samples were fixed in Karnovsky's solution for morphological and morphometric analyses using light microscopy (LM) and scanning electron microscopy (SEM). Drug efficacy and experimental model efficiency were evaluated by comparing results obtained with the standard drug and untreated nematode controls. The preliminary results, showed that the treatment impacted in motility of the nematodes observed using video microscopy and analyzed by ImageJ software with plugin Manual Tracking. In addition, we observed morphological and morphometric alterations in males treated with ABZ, with swelling in testicles. The mortality rate and viability of adult worms and L3 larvae in those treated with IVM, showed significant results in relation to the untreated control, however the ABZ treatment not showed the predicted impact in mortality rate and viability of adult worms. SEM images showed changes in anterior end structures, such as cephalic and labial papillae, with a cuticular retraction of the mouth, in both treatment. These results showed that this experimental system can be used as a new tool for *in vitro* testing of new molecules with anthelmintic activity. The patent of these protocols/methodology was submitted to National Institute of Industrial Property (INPI).