Evaluation of NANoREG TiO₂ toxicity in Caenorhabiditis elegans

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Caenorhabditis elegans is a suitable invertebrate alternative to vertebrate assays for ecotoxicological evaluations. NANoREG TiO2 2883578/ JRCNM01001a/990407 was tested by using Nanoreg SOPToxicity test with the nematode *Caenorhabiditis* elegans for the NANoREG core nanomaterials (Kleiven and Oughton, 2015). The strain of *C.elegans* used is the wild-type N2. The pre-culturing was performed in modified S-BASE media plus cholesterol, incubate at 20°C, in darkness with shaking for 3-5 days. Age-synchronized worms were exposed to concentrations of TiO_2 0.01 to 100 mg/L. Because test solution had no good dispersion, it was sonicated during 15 min, 400 W, 20 kHz before the test. Each test concentration was assayed in 4 replicates in 24-well tissues culture plates, with a total test volume of 1 mL/well containing EPA reconstituted water, *Escherichia coli* OP50, TiO₂ and 5-20 juvenile L1 *C.elegans*. After 96 h, it was added 0.5 mL of Rose Bengal to each well and heated the multidishes in a drying oven for 10 min at 80 °C to kill the worms. To be able to estimate the growth (%) it was prepared a plate with 30 worms for the measure at time 0h. Four endpoints were determined: recovery, fertility, reproduction, and growth after 96 h of incubation. The recovery, fertility and reproduction were not affected by any of the concentrations. However, growth seems to be significantly reduced at concentration 100 mg/L (84%) in relation to control (115%). Responses in C. elegans depending not only on the concentration tested, but also on the endpoint. Growth seems to be a more sensitive endpoint to *C.elegans* than others evaluated. Acknowledgements: Embrapa, NANoREG References: Kleiven, M.; Oughton, D. Toxicity test with the nematode Caenorhabiditis elegans for the NANoREG core nanomaterials. Standard Operating Procedure, 15p, 2015.