In vitro and *in vivo* effects of MultiGuard Protect®, CropGuard® and other sugarcane based test products on the biology and respiration of root-knot nematodes (*Meloidogyne* species)

Fourie, H.,* Van Aardt, W.J., Venter, C & Tiedt, L.R.

[1]Unit for Environmental Sciences and Management, North-West University, Private Bag X 6001 (Internal Box 264), Potchefstroom 2520, South Africa
[2]Laboratory for Electron Microscopy, North-West University, Private Bag X 6001, Potchefstroom 2520, South Africa.

Progressive withdrawal of Class 1 synthetic nematicides from world markets creates an urgent demand for the exploitation and investigation of environmentally-friendlier products to manage plant-parasitic nematode population levels in agricultural/horticultural cropping systems.

Although various plant-derived- and other biological-control products have been studied to date to determine their effect on the biology and mortality of plant-parasitic nematodes, baseline knowledge about the mode of action of furfural-based products, such as MultiGuard Protect® and CropGuard® s well as other sugarcane-based test products are lacking. The effects of MultiGuard Protect®, CropGuard® and four sugarcane-based test products, using a range of concentrations, have thus been evaluated *in vitro* (in a temperature-regulated growth chamber) as well as *in vivo* (in a greenhouse) at 26°C. Freshly-hatched second-stage juveniles (J2) of *M. javanica* and *M. incognita*, respectively, were suspended in 2ml-aliquots of MultiGuard Protect®, CropGuard® and the test products and their concentrations in separate trials. Data on J2 motility, mortality, ultrastructure and specific oxygen consumption rate (($\dot{M}O_2$) were recorded 24, 48, 72 and 96 h after onset of trials. *M. javanica* J2 that were exposed for 96h to different concentrations of one of the test products were inoculated on roots of two-leaf-stage tomato seedlings (cv. Rodade), planted in 4-ℓ capacity plastic pots that were filled with a Telone II-fumigated, sandy-loam soil (4 % clay, 0 % silt and 96 % sand) with a pH (KCl) of 4.69. N. After J2 inoculation, the tomato plants were left in a greenhouse for 56 days to enable the J2 to penetrate, develop and reproduce at least two generations. Untreated controls, consisting of sterilised tap water were included in all trials.

The untreated controls for the different trials resulted in a significant ($P \geq 0.05$) number of J2, ranging between 92 %-97 %, being motile. On the other hand, significantly higher numbers of J2 were immotile after suspension in MultiGuard Protect®, CropGuard® and the other test products, especially 48 h onwards. However, for some concentrations of some of the test products the majority of J2 regained motility after 72 to 96 h. Furthermore, staining of immotile J2 with a 2 % Trypan blue solution resulted in less than 10% being identified as dead after suspension in the different test products and concentrations for 96h.

Scanning Electron Microscopy results indicated that the lateral line structures of J2 suspended in the CropGuard® showed a “sunken” appearance. Ultimately, $\dot{M}O_2$ measurements confirmed that the respiratory physiology of J2 was adversely affected after suspension in CropGuard® and the other test products. The latter results thus supported motility studies that showed a similar trend with regard to the biology of these organisms. Partial respiratory recuperation of only *Meloidogyne incognita* J2 occurred when transferred to sterile tap water after being suspended in the lowest CropGuard® concentrations. Also, magnetic stirring during $\dot{M}O_2$ measurements of J2 did not affect the oxygen consumption rates during a 20 minute period, using 5 000 individuals in the respiration chamber .The $\dot{M}O_2$ stayed at a constant rate (30.2 micromole O_2 hr^{-1} g^{-1}) at 25 ºC with speeds varying from 25 rpm up to 1 000 rpm.

Reproduction of *M. javanica* on the susceptible tomato cultivar Rodade in a greenhouse study showed that significantly lower numbers of eggs and J2 were obtained for the two highest concentrations of one of the test products compared to those for the lowest concentrations. In terms of Rf values, the latter three low concentrations had values higher than 1 (indicating the final egg and J2 population levels exceeded the initial inoculation levels), while the two highest concentrations had Rf values lower than 1. The latter trend indicated that exposure of J2 to these two concentrations resulted in a lower reproduction capacity of these nematodes when subsequently inoculated on tomato roots to such an extent that their final population levels were lower than their initial inoculated ones.

Results from our study demonstrated that two of the test products as well as MultiGuard Protect and CropGuard® had an adverse effect on the biology and structure of root-knot nematode J2 populations used in these experiments.