



## **Research Report**

**Title:** Armorex Laboratory Assay Against Nematodes

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**Location:** Iowa, USA

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### **Abstract:**

The purpose of this study was to evaluate the biopesticide potential of Armorex against select local nematode isolate commonly found in soil. In vitro growth chamber experiments were used to study the effect of Armorex on the mortality of third-stage juveniles (J3). Armorex has good potential as a biopesticide against J3s. At an application rate of 800 ppm, almost all nematodes were killed within six hours.

### **Methods:**

Polystyrene 24 well microliter plates were used for the study. Using a micropipette 1,000  $\mu$ L of distilled water was placed into each well. Twelve wells were used for the trials and six for the controls. One row of six wells was left empty between the test and control sections to reduce any potential spillage or cross contamination. The nematodes were purchased in nutrient media that was shaped into small beads. Each bead contained approximately 7,000 nematodes. One bead was placed into each well of the microliter plate and the bead was broken up to allow the nematodes to move freely.

The J3s are the only stage of this species which exist freely in soil. They are considered to be more resistant to environmental conditions than the other three stages, meaning that a dose that can kill J3s would likely be lethal to the other stages. The mobility of the J3 stage of makes them good candidates for study. In the laboratory this tendency to move is useful because changes in motion or complete cessation of movement indicate impairment or death.

The nematodes were allowed to sit in their well for 30 minutes, and then were checked to ensure that they were moving about and appeared healthy. Photo 1 shows untreated nematodes. Note the curvature of the nematode bodies in this picture, which was taken while they were in motion prior to treatment. To begin the test, 800  $\mu$ L of Armorex was placed into a beaker containing 1000 mL of distilled water to give a dilution rate of 800 ppm. Since Armorex

already has surfactant in it, no additional surfactant was needed to dissolve it in water. The solution was applied at 500  $\mu\text{L}$  to each of the 12 test wells containing the nematodes. The six control wells were left alone. The microliter plates were then left on a level surface at 22 $^{\circ}$  C for six hours.

After six hours had passed, each well was checked. 200  $\mu\text{L}$  of the contents were placed onto a slide and then observed using a stereo microscope to count how many nematodes had ceased movement. This procedure was repeated 3 times per well. The results were considered to be representative of entire 1,500  $\mu\text{L}$  well. Nematodes that had become rigid were considered to be dead, while those that were still moving, however slowly, were counted as alive. The controls were evaluated first, followed by the test wells. The percentage of dead nematodes was averaged to give a lethal dose (LD) percentage. For example, if half of the nematodes were found to be dead, then the well was counted as having a LD 50. The LD results from all 12 wells were averaged.

### **Results:**

After six hours, the six control wells had a LD 0; all nematodes observed were still mobile and appeared healthy. After six hours with the 800 ppm Armorex treatment, the 12 test wells had an average LD of 97.5; only 2.5% of the nematodes were still moving. The lowest LD among the 12 trials was 90%, and the highest 100%. Photo 2 shows nematodes that have been treated with Armorex and have ceased moving completely. In most cases their bodies have straightened into large arcs.



Photo 1: Before treatment



Photo 2: After treatment

### **Conclusions:**

Armorex has good potential as a biopesticide against third-stage juvenile nematodes. At an application rate of 800 ppm, almost all nematodes were killed within six hours.