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MICROBIAL DEGRADATION OF ALDICARB IN RENIFORM NEMATODE INFESTED COTTON FIELD SOILS

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Abstract

The degradation of aldicarb (Temik 15G) was examined in the greenhouse and field using soil from four cotton fields with a history of aldicarb use. In the greenhouse, soils were sterilized by autoclaving or not autoclaved (natural) and aldicarb was added at 5lb/A or no aldicarb was added. *Rotylenchulus reniformis* was incorporated back into the autoclaved soil treatments to return the autoclaved soil to the original natural soil populations. The addition of aldicarb at 5 lb/A to natural soil decreased *R. reniformis* numbers only 36 % compared to 95% in the same soils which were autoclaved. Autoclaving the soils restored aldicarb toxicity. The use of increasing rates of aldicarb did not increase the efficacy of aldicarb in the natural soils. Bacterial populations were increased in the natural soils with aldicarb compared to the soils without aldicarb. However, no bacterial species were consistently associated with aldicarb degradation. The degradation of aldicarb to aldicarb sulfoxide and then to aldicarb sulfone was determined using HPLC. Complete degradation of aldicarb occurred within 12 day after planting the MS and LM field soils. Complete degradation did not occur within 43 day after planting in the CL field.

Introduction

Accelerated microbial degradation has been reported in recent years for a growing number of soil - applied pesticides. This enhanced biodegradation is usually linked to monoculture production regimes treated with repetitive applications of biodegradable pesticides which results in a loss of effectiveness against the target pests. Aldicarb, (Temik 15G) is a systemic, broad-spectrum insecticide/nematicide registered for use on multiple crops. In cotton, aldicarb is applied as a granule at planting and is released by soil moisture. Aldicarb degradation begins immediately with microorganisms oxidizing aldicarb to the metabolite aldicarb sulfoxide, which is oxidized slowly to aldicarb sulfone. Aldicarb sulfoxide and sulfone are of less nematicidal value compared to aldicarb. Loss of efficacy of aldicarb has been observed in mid-south cotton production fields infested with *Rotylenchulus reniformis*. Beginning in 1998, Arkansas, Alabama, Mississippi, and Louisiana have all reported a lack of response to aldicarb.

This study examines the toxicity of aldicarb in natural and autoclaved cotton field soils as measured by HPLC, *R. reniformis* numbers and corresponding effects on the soil mycoflora.

Materials and Methods

Soils were collected from three cotton fields in Alabama, (LM, ES, and CL) and one field from Mississippi (MS) where aldicarb had been applied in previous years and a loss of efficacy had been observed in three of the fours fields. In greenhouse tests, treatments were arranged as a 2 x 2 factorial, (i) soils were sterilized by autoclaving or remained natural (not autoclaved) and (ii) aldicarb added at a rate of 5lb/A or no aldicarb added. Nematodes were extracted by gravity screening and sucrose centrifugal flotation and enumerated with a dissecting microscope. *Rotylenchulus reniformis* from each soil was incorporated back into the autoclaved soil treatments to return the autoclaved soil to the original natural soil populations. Each test was planted with DPL1218 B/RR cotton, placed in a factorial arrangement of a RCBD with 5 reps. Treatments were destructively harvested every 3 days for 21 days, then every 7 days for 42 days concluding with a final harvest at 60 days. In field tests, two of the cotton fields were selected for further evaluation. Aldicarb added at a rate of 5 lb/A or no aldicarb was added and these treatments were replicated four times. Soil samples were collected every three days for 21 days.

The degradation of aldicarb was monitored by HPLC with UV detection at 247 and 220 nm. Sample elution was achieved using a Nova Pak C18 column and a gradient elution consisting of solvent A (5% acetonitril, 5% methanol, and 90% water)

and B (40% acetonitril, 40% methanol, and 10% water) at 1 ml/min. For the first 40 min, solvent A was reduced from 100 to 40% while solvent B was increased from 0 to 60% using a linear curve. From 40 to 45 min, solvent B was brought to 100% and maintained at that level for another 5 min before the mobile phase was returned to the initial conditions.

Microorganisms were isolated from serial dilutions of 10^{-1} to 10^{-4} by plating with a spiral plater on PDASA and TSA media. The bacterial and fungal colonies were enumerated from both dilutions and mediums. One representative bacterial colony morphology was isolated for further identification. Each bacterium was identified by analysis of fatty acid methyl-esters of total cellular fatty acids. Fungal colonies were identified after 7 days.

Greenhouse and field tests were repeated at least twice. All data were subjected to ANOVA and means compared using Fishers least significant difference test ($\underline{P} \leq 0.05$).

Results and Discussion

In three of the soils tested, (MS, LM and ES) the capacity of aldicarb to decrease *R. reniformis* numbers was reduced in natural soil when compared to the autoclaved soil. In natural soil without aldicarb, *R. reniformis* numbers increased in all soils. The addition of aldicarb to the natural soil reduced *R. reniformis* numbers 0, 26, and 49% in the MS, LM, and ES soils, respectively. Only in the CL soil, were *R. reniformis* numbers reduced 90%. Aldicarb was inconsistent in reducing *R. reniformis* population numbers in natural soil. In autoclaved soils without aldicarb, *R. reniformis* numbers increased as previously observed in the natural soil. However, the addition of aldicarb to the autoclaved soil reduced *R reniformis* numbers 97% in all four soils. This indicates that microbial degradation may be responsible for the loss of efficacy to *R. reniformis*.

Sixty-five species of bacteria from 37 genera were identified from all treatments. No bacterial or fungal genus or species was consistently associated with the presence of aldicarb. However, bacterial populations were increased in the natural soil + aldicarb in the MS, LM, and ES soil compared to the CL soil.

In CL soil, where aldicarb was efficacious, HPLC analysis indicated the aldicarb level dropped from the initial 318 ppm to 64.5 ppm by 15 DAP and was not detected after 22 DAP. Aldicarb sulfoxide was detected from 0 to 43 DAP from 4 to 45 ppm. Aldicarb sulfone was detected from 6 to 43 DAP with rates increasing from 1.6 to 10 ppm. In the LM soil, were aldicarb was not efficacious, the aldicarb level dropped from the initial 304 to 3.5 ppm at 6 DAP. Aldicarb sulfoxide was detected from 0 to 12 DAP from 2.8 to 8 ppm. Aldicarb sulfone was not detected at any date. The MS soil HPLC analysis was similar to the LM analysis. Aldicarb dropped from the initial 277 to 3 ppm at 6 DAP. Aldicarb sulfoxide was detected at 3 and 6 DAP from 13 to 18 ppm. Aldicarb sulfone was detected from 0 to 12 DAP with the highest concentrations occurring at 3 and 6 DAP. Complete degradation of aldicarb occurred within 12 DAP in the MS and LM field soils. Complete degradation did not occur within 43 DAP in the CL field. Aldicarb sulfoxide and sulfone were present at 16 and 10 ppm, respectively, when sample analysis was completed.