Effect of Four Nematocides on Activities of Microorganisms in Soil

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Tests were conducted to determine the effects of four nematocides, Dasanit, carbofuran, D-D, and Vorlex on microbial activities in a loamy sand. The results indicated that bacterial and fungal populations initially decreased with some nematocide treatments but recovered rapidly to levels similar to those in the controls. In some instances, ammonium production from added peptone increased in the nematocide-treated soils, whereas mineralization of soil organic nitrogen and nitrification and oxidation of elemental sulfur were depressed. Oxygen consumption generally increased in proportion to the concentration of nematocide in the soil. However, with Vorlex, an increase in respiration was evident at the lower concentration, whereas an inhibitory effect occurred at the higher concentration. The study indicated that indigenous soil microorganisms can tolerate these chemicals used for control of nematodes in soil.

Pesticides are often applied directly to the soil for pest control. Some of the organophosphorus and carbamate pesticides have been shown to be moderately persistent (4) and to have some effect on microbiological activities in the soil (1, 8). For many years, fumigants such as D-D and Vorlex have been widely used for control of nematodes. Other compounds such as Dasanit and carbofuran have also shown promise when applied at relatively high rates of application. Little is known about the effects of these chemicals on the beneficial soil microbes that are important in soil fertility. This paper reports the effects of four nematocides on the activities of microorganisms in the soil.

MATERIALS AND METHODS

The experiments were conducted with Delhi loamy sand, a typical agricultural soil in southwestern Ontario. Composite soil samples were taken to a depth of 6 inches and analyzed for chemical, mechanical, and physical characteristics. The soil contained 0.81% organic matter and 0.03% total nitrogen. The moisture holding capacity was 27%, and the pH was 8.2.

Sufficient amounts of Dasanit (O,O-diethyl O-[p-(methylsulfinyl)-phenyl] phosphorothioate) and carbofuran (2,3-dihydro-2,2-dimethyl-7-benzo-furanyl methylcarbamate) to give final concentrations of 1 and 5 μg/g of soil were mixed with measured amounts of carrier sand by dissolving them in a distilled petroleum ether-acetone solvent (1:1; v/v). After the solvent was evaporated, the sand-nematocide mixture was incorporated with the soil. The required amounts of fumigant D-D (mixture of 1,2-, 1,3-, 2,3-, and 3,3-dichloropropene) at rates of 120 and 600 μg/g and Vorlex (20% methyl isothiocyanate-chlorinated C, hydrocarbon mixture) at rates of 30 and 180 μg/g (14 and 84 GPA) were mixed into the soil. Chemical purities of nematocides were at least 94.5%. Reagent grade peptone and elemental sulfur powder were added to each soil sample at 1,000 μg of nitrogen or sulfur per g for ammonification and sulfur oxidation, respectively. Oxidation of ammonium from soil organic nitrogen was studied by nitrification. The additives were thoroughly mixed with the soil. The mixtures and controls were transferred to 0.236-liter (0.5 pint) milk bottles, which were closed with 0.0381-mm (1.5 mil) thick polyethylene film. Soil moisture was maintained at 60% of the moisture-holding capacity. The treatments, in duplicate, were incubated in the laboratory at 28 C for appropriate periods, i.e., 1 week for ammonification, 1 and 2 weeks for nitrification, and 4 weeks for sulfur oxidation. Changes in the population of soil microorganisms were determined after 1, 2, 4, 8, and 12 weeks. Rose Bengal-streptomycin-agar was used for determination of changes in fungi in the soil, and sodium albuminate-agar was used for bacteria and actinomycetes. Procedures for chemical, physical, microbial, and statistical analyses of the soil samples have been described elsewhere (8).

Respiratory studies were conducted with Warburg
reaction flasks containing 0.15 ml of 20% KOH solution in the center wells to absorb CO$_2$. An 8-g sample (oven-dry weight) of each soil and nematocide mixture was placed in a flask with 0.70 ml of distilled water to bring the soil to 60% of its moisture holding capacity. One hundred micrograms of glucose-C per g was added and mixed into each soil sample. Oxygen consumption was measured at 30 C for 80-hr periods with a Gilson differential respirometer.

RESULTS AND DISCUSSION

The plate count data obtained indicated that none of the materials tested affected the fungal population drastically, although statistically significant differences were observed in many cases (Table 1). Because of the limitations of the plate technique for determining populations of soil fungi, the results can be taken only as an indication of what is likely to occur in the field. There was a slight depression in population after the first week with 30 $\mu$g of Vorlex per g, 600 $\mu$g of D-D per g, 5 $\mu$g of carbofuran per g, and Dasanit at both concentrations; after the second week, there was a slight depression at both concentrations of the two fumigants and with carbofuran at 5 $\mu$g/g (Table 1). An inhibitory effect was also observed in the samples treated with 5 $\mu$g of Dasanit per g and both concentrations of carbofuran after 4 weeks of incubation. All populations subsequently recovered to the level of those in the controls.

The plate counts also indicated that the bacterial populations decreased significantly during the first week of incubation (Table 1) with samples treated with 5 $\mu$g of carbofuran per g, 120 $\mu$g of D-D per g, 180 $\mu$g of Vorlex per g, and at both concentrations of Dansanit. Populations subsequently recovered to levels at or above those found in the controls.

During the 12-week incubation period, plate counts in the controls showed a decrease in the populations of fungi and bacteria (Table 1) in the soil. These declines probably resulted from the fact that over a long incubation period aeration was inadequate, the sources of available nutrients were depleted, and waste metabolites had accumulated (2, 7). In general, organisms capable of forming spores or of existing at low metabolic levels would survive (7). In the later part of the incubation period, populations in the nematocide-treated soils exceeded those found in the controls (Table 1). *Trichoderma* species became dominant. These species may be more tolerant to the toxic pesticides and thus, after partial sterilization of the soil, may be reestablished rapidly in the less competitive condition. It has been shown that *Trichoderma* species can tolerate and degrade pesticides (5).

The results indicated that, in some instances, ammonium production from added peptone-N increased by 1 to 10% in the nematocide treatments (Table 2). Similar responses with herbicides and organophosphorus insecticides have been reported (8, 9). The fumigants temporarily depressed ammonification of soil organic nitrogen.

Vorlex depressed nitrification of ammonium from soil organic nitrogen slightly during the first week after treatment (Table 2). However, nitrate production was equal to or better than the control after two weeks of incubation with all four nematocides. Similar responses have been demonstrated with other organophosphorus and carbamate pesticides and fumigants (1, 3, 10).

Oxidation of added elemental sulfur ranged from 20 to 40% (Table 2). With the exception of Dasanit, the nematocides caused a slight

| Treatment  | Rate ($\mu$g/g) | Fungi ($\times 10^4$/g of soil) | Bacteria ($\times 10^4$/g of soil) |
|------------|----------------|--------------------------------|----------------------------------|
|            | 1  | 2 | 4 | 8 | 12 | 1  | 2 | 4 | 8 | 12 |
| Control    |    |   |   |   |    |  9 | 7 | 5 | 4 | 2 |  7 |  9 | 7 |  6 |  3 |
| Dasanit    | 1  | 7 | 5 | 4 | 3 |  7 | 4 | 3 |  7 |  6 |  3 |  9 |  8 |  5 |  6 |
| Carbofuran | 1  | 7 | 5 | 4 | 3 |  7 | 4 | 3 |  7 |  6 |  3 |  9 |  8 |  5 |  6 |
| Carbofuran | 1  | 7 | 5 | 4 | 3 |  7 | 4 | 3 |  7 |  6 |  3 |  9 |  8 |  5 |  6 |
| D-D        | 10 | 7 | 5 | 4 | 3 |  7 | 4 | 3 |  7 |  6 |  3 |  9 |  8 |  5 |  6 |
| Vorlex     | 30 | 7 | 5 | 4 | 3 |  7 | 4 | 3 |  7 |  6 |  3 |  9 |  8 |  5 |  6 |
| Vorlex     | 180| 7 | 5 | 4 | 3 |  7 | 4 | 3 |  7 |  6 |  3 |  9 |  8 |  5 |  6 |

*Values within each column followed by the same letter are not significantly different at the 5% levels determined by Duncan's multiple range test.

*Weeks of incubation.
but significant decrease in sulfur oxidation.

Soil pH was not significantly altered by any of the nematocide treatments. Sulfur oxidation lowered the pH by 1.6 units, whereas ammonification of the added peptone increased pH to 9.2, thus preventing nitrification which is inhibited above pH 8 (6).

The effect of the nematocides on the soil microbial population as a whole is illustrated by changes induced in respiration as a result of the nematocide treatments (Fig. 1). With the exception of Vorlex at the higher concentration, oxygen consumption from the decomposition of native organic matter was greater in the treated soils than in the controls. In addition, oxygen consumption increased with increasing concentration of Dansanit and carbofuran, both in soils with and without supplemented glucose-C. Similar results were obtained with some organophosphorus insecticides (8). Respiration was depressed with both D-D treatments and with the higher level of Vorlex in the glucose-treated soils (Fig. 1). Soil that received the lower concentration of fumigants consumed a greater amount of oxygen.

The temporary depression of soil respiration by the fumigants was apparently the result of partial sterilization, as indicated by the plate counts (Table 1).

This study has pointed out that the indigenous soil microorganisms can tolerate these chemicals used for control of soil nematodes. Temporary inhibition of mineralization of soil organic nitrogen, nitrification, and oxidation of elemental sulfur in soil occurred, but the microorganisms recovered rapidly. Increased respiration associated with the higher levels of

### Table 2. Microbial activities after soil was treated

| Treatment | Rate (µg/g) | Ammonification | Nitrification of ammonium from soil organic nitrogen | Sulfur oxidation of elemental sulfur |
|-----------|------------|----------------|--------------------------------------------------|-----------------------------------|
|           |            | Soil organic nitrogen | Peptone-N | 1* | 2 |                                      |
| Control   |            | 36 a | 688 b | 5 ab | 7 d | 397 a |
| Dasanit   | 1          | 27 abc | 696 b | 5 ab | 9 bc | 340 ab |
|           | 5          | 36 a | 640 c | 5 ab | 10 ab | 333 ab |
| Carbofuran| 1          | 25 bc | 720 b | 5 ab | 8 cd | 241 de |
|           | 5          | 29 ab | 680 b | 5 a | 11 a | 198 e |
| D-D       | 120        | 20 bcd | 725 b | 4 bc | 7 cd | 305 bcd |
|           | 600        | 16 cd | 784 a | 4 ab | 9 bc | 262 cde |
| Vorlex    | 30         | 8 d | 704 b | 4 cd | 8 cd | 312 bc |
|           | 180        | 8 d | 716 b | 4 cd | 7 d | 276 bcd |

*Within each column, values accompanied by the same letter are not significantly different at the 5% level with Duncan’s multiple range test. Values are expressed as micrograms of (NH₄⁺, NO₂⁻, NO₃⁻)-N, (NO₂⁻, NO₃⁻)-N, and SO₄²⁻-S (for ammonification, nitrification, and sulfur oxidation, respectively) per gram.

*Weeks of incubation time.

![Fig. 1. Effect of four nematocides on soil microbial respiration.](image_url)

Dasanit and carbofuran might result from the degradation of these chemicals by the soil microorganisms.

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