Organophosphorous Insecticides Effects on Legume Growth And Soil Nitrification

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In recent years the agricultural use of organophosphorous insecticides has increased greatly. Much of this is because the chlorinated hydrocarbon insecticides, such as DDT, chlordane, aldrin and lindane are extremely resistant to degradation in plants and soil. Furthermore, when taken into the body, these insecticides tend to accumulate in animal tissues. This characteristic has resulted in extensive legal restrictions on their use. The organophosphorous group, which includes some compounds highly toxic to man and animals, generally has the advantage that they decompose rapidly enough in soils to avoid excessive buildup of residues which might enter the food chain.

The addition of pesticidal chemicals to soil involves certain risks. Even small changes in the chemical structure of compounds can change their biological effects greatly and unpredictably. An example is the common herbicide 2,4-D, which breaks down quite rapidly in soil as the microbial population uses it for nutrition. However, the addition of only a single additional chlorine molecule, forming the compound 2,4,5-T, results in an herbicide which resists decomposition in the soil indefinitely. Also, pesticides added to soil may prove to have unexpected resistance to degradation, or may have toxic effects on plants or on essential soil bacteria.

In view of this, the Department of Bacteriology, in cooperation with the Department of Entomology at North Dakota State University has tested the effects of several organophosphorous insecticides, centering the investigation on their effects on soil bacteria such as those which carry out nitrification, and on the bacteria of the **Rhizobium** group, which are necessary for legumes to fix nitrogen.*

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Nitrification experiments

Nitrification is the conversion by soil bacteria of nitrogen, as found in decomposing plants, into nitrates. The nitrates, in turn, are the nitrogen form best assimilated by growing plants. Only a limited group of bacteria are capable of carrying out this process in the soil. The effects of three insecticides on the ability of soil bacteria to convert ammonium sulfate into nitrate is illustrated in Figure 1.



Figure 1. Effect of insecticides on nitrification rates in soil. FR = Field rate 10 X = Ten times field rate 100 X = Hundred times field rate

The insecticides were applied at normal field rates and at ten and 100 times field rates. As shown, phorate (Thimet) did not inhibit nitrification. Dasanit (Bayer 25141) inhibited nitrification considerably for several days at high concentrations, but not when applied at normal field rates. By the eleventh day this inhibition had disappeared. Application of Diazinon stimulated nitrification. The chemical formula of this insecticide includes a nitrogen-containing ring structure which apparently breaks down rapidly in the soil and releases urea. The urea is then rapidly converted into additional nitrate by nitrifying bacteria, accounting for the apparent stimulation.

Field experiments in which the numbers of nitrifying organisms in the soil were statistically estimated showed that there was no important depression in the numbers of the organisms.

Nitrogen fixation by legumes

Leguminous plants such as alfalfa, clover, and soybeans are capable of using the nitrogen in the air for growth. However, the plants require bacteria of the **Rhizobium** group to form the root nodules necessary for this. Pesticides toxic to **Rhizobium spp.** would be expected to inhibit proper legume nodulation and subsequent plant growth.

An experiment to determine the effects of an insecticide on **Rhizobium trifolii** is illustrated in Figure 2. Various concentrations of the liquid insecticide were applied to paper discs. These discs then were placed on the surface of a nutrient medium which had been seeded with the test bac-

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Figure 2. An example of inhibition of growth of a bacterium by Dasanit (Bayer 25141) A. 2 \cup 1 B. 5 \cup 1 C. 10 \cup 1 D. 20 \cup 1.



Figure 3. Effect of Dasanit (Bayer 25141) on growth of alfalfa.

terium. The clear area showing immediately next to the disc indicates that the bacteria have failed to grow at all concentrations of this insecticide. Table 1 shows the results of these experiments with a number of important soil bacteria.

Applications of the test insecticides were made to alfalfa, sweetclover, and soybeans in greenhouse pot experiments. Rates of applications were at normal field rates and also at ten and a hundred times normal field rate.

Table 1. Disc Inhibition Tests of Effects of Insecticides on Bacteria.

+ = Inhibition	- = No effect		
Organism	Dasanit	Diazinon	Phorate
Azotobacter vinelandii	+		+
Bacillus subtilis	+	_	+
Pseudomonas fluorescens	+		-
Rhizobium japonicum	+	-	-
Rhizobium leguminosarum	+		+
Rhizobium meliloti	+	-	_
Rhizobium trifolii	+	13	+



Figure 4. Effect of Diazinon on growth of alfalfa.



Figure 5. Effect of phorate (Thimet) on growth of alfalfa.

Soybeans were not greatly affected by any of the insecticides at normal field rates or even at ten times normal field rate applications. At one hundred times normal field rate the numbers of nodules decreased sharply and the plants showed some stunting and chlorosis. These effects were particularly noticeable with phorate.

Sweetclover was not damaged by Diazinon at any of the concentrations tested. Higher concentrations of Dasanit and phorate resulted in fewer nodules and affected the general growth somewhat. Phorate, again, was more damaging. At the highest concentration it almost completely eliminated nodulation on these plants.

Alfalfa proved much more sensitive to insecticides. Phorate inhibited nodulation and growth at all levels of application. Diazinon and Dasanit were apparently damaging only at the highest concentrations. (Figures 3, 4, and 5).

Similar experimental work with other insecticides is continuing.