Benomyl-Soil Microbial Interactions

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ABSTRACT

Benomyl added to soil caused no changes in bacterial populations, but did reduce numbers of fungi and actinomycetes. Decomposition (16-34%) of ^{14}C ring-labeled benomyl, during 6 and 12 months incubation periods, occurred only in nonsterilized soil. Ring cleavage of the benzimidazole nucleus and metabolism of this moiety to CO_2 is apparently related to the presence of soil microorganisms.

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Additional key words: fungicide, soil microflora, metabolism.

In a recent study (1) the "half-life" of benomyl was reported to be 3-6 months in cropped soil and 6-12 months in bare soil. On the other hand, benomyl had little effect on soil microbial populations under either greenhouse (3) or field conditions (4). This laboratory study reports the effect of benomyl on microbial populations and the metabolism of this fungicide in soil.

A Maury silt loam soil, collected in October 1972 and May 1973 from an area not treated with any of the benzimidazole fungicides, was screened through a 2-mm sieve. A portion of the soil was oven dried, and the percent moisture capacity in the remaining soil was determined. Fifty grams of soil (oven-dry-weight basis) was placed in 300-ml flasks. Some flasks were autoclaved three times for 1 hour during a 48-hour period. One ml of acetone containing either 500 μg of ring-labeled benomyl (3.6 μCi, sp. act. 2.01 mCi/mmole) or nonlabeled benomyl and either water or nutrients (1% glucose and 0.5% yeast extract, w/w), to bring the soil to 70-75% field capacity, was added to each flask. The cotton-plugged flasks were shaken for 1 hour.

The flasks containing nonlabeled fungicide, nonsterilized and nonnutrient-amended soil were incubated in the dark at 25 C for 30-35 days, after which microorganisms were determined by a dilution plate technique (2). Fungi were isolated on the dextrose-peptone-yeast extract agar of Papavizas and Davey (2), and bacteria and actinomycetes on Hutchinson's agar as modified by Bhat and Shetty (2).

The breakdown of ^{14}C benomyl in amended and nonamended, and in sterilized and nonsterilized, soil was determined by trapping the radioactive CO_2 produced in a solution containing 10 ml of ethylene glycol:ethanolamine (1:1, v/v) (5). Traps were changed every 4 days and 1.0-ml portions of the trapping solution were counted by liquid scintillation to determine the amount of $^{14}CO_2$ evolved. The soils were monitored for $^{14}CO_2$ evolution for up to 340 days, after which they were dried and the total ^{14}C remaining in the soil was determined by oxygen combustion.

All experiments were run twice with triplicate treatments.

TABLE 1. Effect of benomyl on soil microbial populations[a]

Treatment	Microorganisms per gram of soil: Fungi ($\times 10^3$)	Bacteria ($\times 10^6$)	Actinomycetes ($\times 10^6$)
Fungicide			
Oct 73	12.7 ± 4.1	43.5 ± 3.5	1.3 ± 1.1
May 74	24.2 ± 7.2	29.4 ± 3.2	1.6 ± 1.8
Control			
Oct 73	36.2 ± 3.8	40.1 ± 2.8	8.7 ± 1.5
May 74	98.5 ± 6.1	24.8 ± 4.0	6.8 ± 1.4

[a]Five-hundred micrograms (22.4 kg/hectare) of benomyl was uniformly mixed with 50 g (oven-dry-weight basis) Maury silt loam soil. Fungicide-soil mixture was incubated for 30-35 days and microbial populations determined by dilution plate and agar-medium isolation techniques.

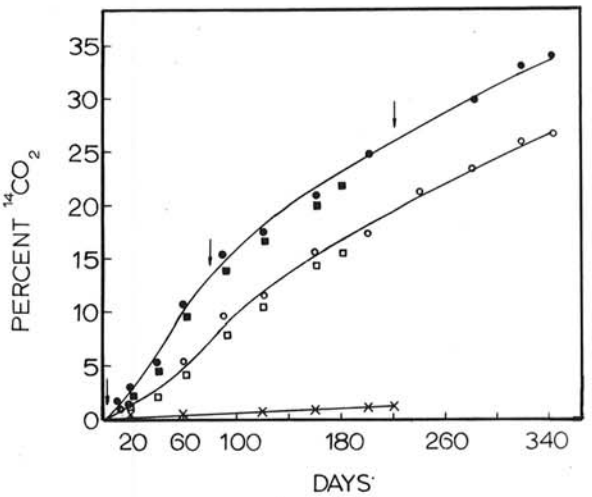

Fig. 1. Rate of release of $^{14}CO_2$ from ring-labeled benomyl incubated in soil for either 180 or 340 days. Five-hundred micrograms (3.6 μCi) of ^{14}C benomyl was uniformly mixed with 50 g (oven-dry-weight basis) of Maury silt loam soil, and $^{14}CO_2$ was trapped in ethylene glycol: ethanolamine (1:1, v/v) solution. Legend: •—•, incubation 340 days, in nutrient-amended [1.0% glucose + 0.5% yeast extract (w/w)] soil [nutrients added three times at (↓)]; ■, same as above, incubation for 180 days; o—o, incubation for 340 days in nonnutrient-amended soil; □, same as above, incubation for 180 days; x—x sterilized soil, amended and nonamended with sterile nutrients.

In short-term (30-35 days) studies of benomyl in soil there were 2- to 3-fold reductions in fungal and actinomycete populations (Table 1). However, there was no effect on bacterial populations. These results agree with those in greenhouse (3) and field studies (4), except for the changes in fungal and actinomycete populations. Large scale changes (> 10-fold) in soil microbial populations did not occur in these short-term benomyl-soil experiments.

A uniform decomposition of ^{14}C benomyl to $^{14}CO_2$ occurred in the two separate experiments (Fig. 1). In amended soil 21 and 34% of the label was collected during the 180- and 340-day experimental periods, respectively.

In nonamended soil, 16 and 27% of the label was collected during the same intervals, respectively. Only the initial nutrient amendment of nonsterilized soil apparently stimulated the breakdown of ^{14}C-labeled fungicide. Soil microflora are the apparent cause of the decomposition of benomyl since less than 1.25% of the label was collected as $^{14}CO_2$ from sterilized soils.

The amount of ^{14}C remaining in the soil at the end of each experiment added to that lost as $^{14}CO_2$ closely approximated the total activity of the labeled benomyl used.

These data suggest that, through microbial activity, ring cleavage of the benzimidazole nucleus occurred with the resulting metabolism of the labeled carbon to $^{14}CO_2$.

LITERATURE CITED

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