Translocation of Benomyl, Carboxin, and Chloroneb in Soybean Seedlings

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ABSTRACT

Use of ^{14}C-labeled and nonlabeled fungicides as seed treatments of soybean showed that benomyl and chloroneb initially tended to localize in cotyledons, while carboxin did not. At 4 days after treatment, chloroneb was redistributed into the hypocotyl and cotyledons, whereas benomyl moved only into the epicotyl. Carboxin was distributed uniformly throughout the seedling, with higher concentrations in the epicotyl. Phytopathology 61:1301-1302.

Additional key words: systemic fungicides, *Rhizoctonia solani, Penicillium atrovenetum.*

Benomyl, chloroneb, and carboxin are taken up by germinating soybean seed, and benomyl or a related compound is localized in the cotyledons (9). Benomyl, chloroneb, and Thiabendazole [2-(4-Thiazolyl)-benzimidazole] seedlings, and benomyl tends to localize in the cotyledons after uptake by roots (5). ^{14}C-labeled and nonlabeled Thiabendazole moves unaltered into aboveground parts of soybean seedlings after root uptake, and the concentration increases in epicotyl and root tissues with increased exposure time, but not in hypocotyl tissues (6). We report further information on the translocation and distribution of three nonlabeled and two ^{14}C-labeled fungicides in soybean seedlings after application as seed dressings.

Commercial formulations of three fungicides were used: benomyl (Benlate 50 WP, E. I. duPont de Nemours & Co., Inc., Wilmington, Del.); carboxin (Vitavax 75 WP, UniRoyal Chemical Co., Bethany, Conn.); and chloroneb (Demosan 65 WP, E. I. duPont de Nemours & Co., Inc., Wilmington, Del.). ^{14}C-carboxin, mol wt 235, was uniformly labeled in the aniline ring, had a specific activity of 0.2 μc/mmole, and was provided by the UniRoyal Chemical Co. ^{14}C-chloroneb, mol wt 207.6, was randomly labeled in the benzene ring, had a specific activity of 0.447 μc/mmole, and was provided by the E. I. duPont de Nemours & Co., Inc.

Rhizoctonia solani Kuehn, isolate T (ATCC-18184), and *Penicillium atrovenetum* G. Smith were used for bioassays. Inoculum was obtained from cultures of the fungi grown on commercial potato-dextrose agar (PDA-D) prepared by standard procedures.

Seed treatment and bioassay techniques used were similar to those of Allam et al. (1), Erwin (3), Gray & Sinclair (5, 6), and Thapliyal & Sinclair (9). Seed (*Glycine max* [L.] Merr. 'Calland') were dusted with each of the labeled or nonlabeled fungicides at 0.125, 0.25, or 0.5 g/100 g (2, 4, or 8 oz/100 lb.). Comparable lots of seed not treated with fungicide also were planted. Seedlings were grown in a growth chamber at 24 C, 70% relative humidity, and a 12-hr light of 8,000 ft-c. Samples were taken from plants grown from benomyl-treated seed at 4, 5, 6, 7, 8, 9, 10, and 17 days after planting. Plants from chloroneb- and carboxin-treated seed were taken at 8, 9, 10, 14, and 25 days. Four plants were used at each sampling. Root, hypocotyl, cotyledons, and leaves were bioassayed separately.

The techniques used for the study of uptake and translocation of ^{14}C-chloroneb and -carboxin were similar to those described by Kirk et al. (8). Radioautographs were made of both nontreated and treated seedlings. Extracts from plants treated with labeled compounds were prepared for liquid scintillation counting, using methods described by Bray (2).

Penicillium atrovenetum was more sensitive to benomyl than was *R. solani* when used in bioassays. The mean zone of inhibition (area around a tissue section that did not allow growth of the test fungus) of *P. atrovenetum* was 20 to 23 mm in diam at 14 and 17 days, respectively, from roots of plants from seed treated with 0.5 g, while no growth inhibition was shown in *R. solani* bioassay plates. Leaf discs from plants grown for 4 to 17 days inhibited growth of *P. atrovenetum,* but zones of inhibition of *R. solani* occurred only around leaf discs from plants grown for 14 to 17 days. There was no inhibition of growth of either fungus by extracts from hypocotyl tissues. Discs or extracts from cotyledons inhibited growth of *P. atrovenetum* or *R. solani,* respectively, at all rates and at all days.

Extracts from roots of seedlings from seed treated with chloroneb at 0.5 g showed no activity against *R. solani* until 25 days after treatment with a reading of 3.5 cm. Readings for hypocotyl extracts at 8, 9, 10, 14, and 25 days were 5.0, 5.0, 4.9, 4.9, and 4.6 cm, respectively, indicating comparatively low activity. Similarly, for leaf extracts, the readings for the respective days were: 5.0, 5.0, 4.8, 4.6, and 4.5 cm. The highest activity was found in the cotyledons with readings of 2.9, 3.1, 3.5, and 3.8 cm for 8, 9, 10, and 14 days, respectively. There was no inhibition by root extracts of seedlings from seed treated with carboxin at 0.5 g at any time, while hypocotyl extracts showed slight activity with readings of 4.9, 4.8, 4.6, 4.9, and 4.4 cm at 8, 9, 10, 14, and 25 days, respectively. Inhibition by leaf extracts increased with increased days with readings of 4.8, 4.6, 4.4, 3.8, and 3.5 cm, respectively. Highest activity was found in cotyledon extracts, which gave readings of 2.0, 2.1, 2.8, and 2.8 cm at 8, 9, 10, and 14 days, respectively.

Radioautographs showed that the ^{14}C-chloroneb was localized in cotyledons, lower hypocotyl, and roots.

There was no redistribution of fungicide after 9 and 14 days. The radioactivity in hypocotyls and cotyledons of seedlings from seed exposed to fungicides in the germinator for 24 and 48 hr was higher than the activity in these same tissues from seed exposed for 12 hr or dormant seed. ^{14}C-carboxin was uniformly distributed within the different seedling parts after 24- and 48-hr exposure. In seed germinated for 12 hr, the compound was not present in the epicotyl. At 14 days, ^{14}C-carboxin moved from cotyledons into the leaves.

Bioassays gave indirect evidence that the fungicides entered seedling tissues. More direct evidence of seed absorption and seedling translocation was obtained using ^{14}C-chloroneb and -carboxin. Results using chloroneb were similar to those reported for carboxin when used as seed dressing on cottonseed (1).

When extracts of plant parts were counted using liquid scintillation techniques, the data indicated that ^{14}C-chloroneb was localized in cotyledons at 9 and 14 days after treatment, whereas ^{14}C-carboxin after 14 days was distributed in both cotyledons and leaves. The counts/min (cpm), corrected for background, at 9 and 14 days from seed treated with ^{14}C-chloroneb, respectively, were: roots, 1 and 5; hypocotyl, 8 and 10; cotyledons, 522 and 537; and leaves, 7 and 5. The cpm at 9 and 14 days/sample of seedlings from seed treated with ^{14}C-carboxin, respectively, were: roots, 2 and 5; hypocotyls, 23 and 25; cotyledons, 504 and 189; and leaves, 19 and 363.

The systemic movement of benomyl almost exclusively into cotyledons may limit its use as a seed dressing not only in soybean, but in other crops as well. Harper (7) reported benomyl was ineffective as a seed dressing in peas. This also may be due to cotyledonary localization. Benomyl apparently had limited movement from the cotyledons into epicotyl tissues, but no downward movement. This is the first report of chloroneb being localized in soybean cotyledons. Activity of chloroneb, on the other hand, was detected only in the lower hypocotyls and roots of seedlings. ^{14}C-chloroneb was detected in hypocotyl and root tissues at 7, 9, and 14 days after seed treatment, and suggested possible downward movement. The data were not sufficient to show whether the roots and hypocotyl tissues absorbed the compound directly or translocated into these tissues from the cotyledons. ^{14}C-chloroneb was shown to accumulate in root and hypocotyl of bean and cotton seedlings grown in treated soil (4). Kirk et al. (8) reported limited downward movement of ^{14}C-chloroneb in cotton seedlings, and found the chemical had little tendency to spread to plant parts above the cotyledonary node. In contrast to their report (8) that ^{14}C-carboxin was localized in the lower portions of cotton seedlings, carboxin apparently did not become localized in soybean cotyledons, but moved fairly uniformly to all parts of seedlings.

Redistribution (or lack of it) of these fungicides from cotyledons to hypocotyl and epicotyl tissues may be important for control of infections of these plant parts. Chloroneb has the potential of being more useful for control of root and lower-stem infections because it becomes localized in this region where seedling infections are most apt to occur. In contrast, benomyl and carboxin would be less effective against this type of infection, and more effective against pathogens invading the epicotyl tissues.

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