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EFFECT OF SEED-TREATMENT FUNGICIDES ON CONTROL OF ASCOCHYTA RABIEI IN CHICKPEA SEED INFECTED WITH THE PATHOGEN

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ABSTRACT

The incidence of Ascochyta blight (Ascochyta rabiei) in chickpea seedlings was greatly reduced and emergence markedly increased when infected seed was treated with certain chemical compounds, especially the systemic benzimidazoles, benomyl and thiabendazole. Treatment of healthy chickpea seed with different fungicides prior to planting did not protect the foliage of seedlings against infection by A. rabiei when plants were inoculated 2-3 weeks after emergence. No adverse effects were noted on germination, growth, flowering or pod set of chickpea plants whose seeds had been treated with various fungicides.

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Blight of chickpea (Cicer arietinum) caused by Ascochyta rabiei (Pass.) Labr. is a serious and important disease in areas of the Middle East and Southeast Asia where rainfall or relative humidity, or both, is high during part of the growing season (2). The fungus affects all foliar parts of the plant and is seedborne (1, 2, 4, 5, 6). The seed is usually infected while it is still in the pod (1, 4, 5), but sometimes the seed is contaminated by the pathogen during threshing (5). Tests in Iran showed that the pathogen can survive for a long time as mycelia and pycnidia in infected chickpea tissues (2, 3). In areas where environmental conditions are favorable for disease epidemics, seed transmisison is an important mechanism in spread and survival of A. rabiei (2, 6).

These studies were carried out to observe the control of seedborne and foliar infection by A. rabiei by treating chickpea seeds with different fungicides.

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MATERIALS AND METHODS

In the inoculation studies, seeds of a white-seeded chickpea variety from Ghazvin, Iran were used. The seeds were from healthy plants grown the previous year at Karaj, Iran, where Ascochyta blight does not occur.

A mixed spore inoculum of four Iranian isolates of A. rabiei was used to inoculate chickpea seeds and seedlings. The isolates of the pathogen were from Dezful, Ghazvin, Gorgan and Saqez, Iran.

The various fungicides used in these studies were: 2-(4-thiazolyl)-benzimidazole (thiabendazole), methyl 1-(butylcarbamoyl)-2-benzimidazolecarbamate (benomyl), tetrachloroisophthalonitrile (chlorothalonil)(Daconil), 5,6-dihydro-2-methyl-1,4-oxathiin-3-carboxanilide (carboxin) (Vitavax), zinc ethylenebis[dithiocarbamate] (zineb), cis-N-[(1,1,2,2-tetrachloroethyl)thio]-4-cyclohexene-1,2-dicarboximide (Dfn.) (Difolatan), a coordination product of zinc ion and manganous ethylenebis[dithiocarbamate] (zinc + maneb), N-(ethylmercuri)-p-toluenesulfonanilide (Cer. M) (Ceresan M), 2,4-dichloro-6-(o-chloroanilino)-s-triazine (Dy.) (Dyrene), manganous ethylenebis[dithiocarbamate] (maneb), N-[(trichloromethyl)thio]-4-cyclohexene-1,2-dicarboximide (captan), 2,6 dichloro-4-nitroaniline (DCNA), pentachloronitrobenzene (PCNB), 1,4-dichloro-2,5-dimethoxybenzene (chloroneb) (Demosan), 5-ethoxy-3-(trichloromethyl)-1,2,4-thiadiazole (ETMT) (Terrazole) + pentachloronitrobenzene (PCNB) (ETMT + PCNB), copper oxychloride (Cupravit Blue), 5,6-dihydro-2-methyl-1,4-oxathiin-3-carboxanilide-4,4-dioxide (oxycarboxin) (Plantvax), and sodium p-(dimethylamino)benzenediazosulfonate (Dex.)(Dexon).

Before inoculation with the pathogen, seeds were washed in running tap water, surface sterilized in 0.5% sodium hypochlorite for 10 min, and rinsed in running tap water. Surface-sterilized seeds were submerged either in distilled water or in a spore suspension (10^9 to 10^{10} spores/cc) of A. rabiei contained in separate desiccators. A vacuum was increased and released several times before being maintained for 3 hr.

Inoculated seeds were placed on clean paper towels to dry. Air from a fan was circulated over the seeds to facilitate drying. Noninoculated seeds were treated the same way.

Seeds inoculated with A. rabiei on December 23, 1970, were stored in the laboratory at room temperature until March 13, 1971, when they were treated with 18 different fungicides. Treated and nontreated inoculated seeds and nontreated, noninoculated seeds were planted in pasteurized soil on March 15 and April 10, 1971. Pots were randomly distributed on a greenhouse bench where temperatures ranged from 10-30° C and relative humidity seldom exceeded 50%. Periodic observations were made of emergence and infection of the stem, petiole, and leaves by A. rabiei.

In another series of experiments, healthy seeds were treated with six fungicides (benomyl, thiabendazole, carboxin, oxycarboxin, chloroneb, and captan at 3 dosage rates/fungicide) before they were planted in pasteurized soil. Seedlings were inoculated with a spore suspension of A. rabiei (10^9 spores/cc) when plants were 10-15 cm tall (3-4 weeks after planting). Immediately after inoculation plants were incubated in a moist chamber for 4 days and then removed to a greenhouse bench. Plants were graded for infection 3-5 weeks after inoculation.

RESULTS

Effect of seed-treatment fungicides on emergence and incidence of disease in Ascochyta-infected chickpea seed: Germination and emergence of chickpea seeds infected with A. rabiei were reduced in many treatments as a result of pre- and post-emergence damping-off (Table 1 and Fig. 1 and 2). In treatments with a high incidence of disease, lesions frequently formed on the hypocotyls, stems, and foliage of seedlings, causing stunting, yellowing, or death of diseased plants. Pycnidial formation was often abundant on infected tissues. Conidia oozed from the pycnidia at times, especially when the humidity was high. The pathogen was isolated consistently from diseased seedlings when surface-sterilized pieces of infected tissue were cultured on potato-dextrose agar or water agar. Emergence and number of healthy plants were increased in all but one instance by fungicidal seed treatment (Table 1). In the nontreated inoculated control, seedling emergence was reduced by 54%, and 94% of the surviving plants were infected with the pathogen (Table 1 and Fig. 1). Most seedlings in the nontreated inoculated control group were stunted and chlorotic, and the root systems of these plants were poorly developed. Some seed-treatment compounds, however, particularly the systemic fungicides, benomyl and thiabendazole, greatly increased emergence and healthy stand counts. Emergence of healthy seedlings treated with each of these fungicides was 90% or more (Table 1), and lesions on infected plants were restricted in size and number. No phytotoxic responses were noted in

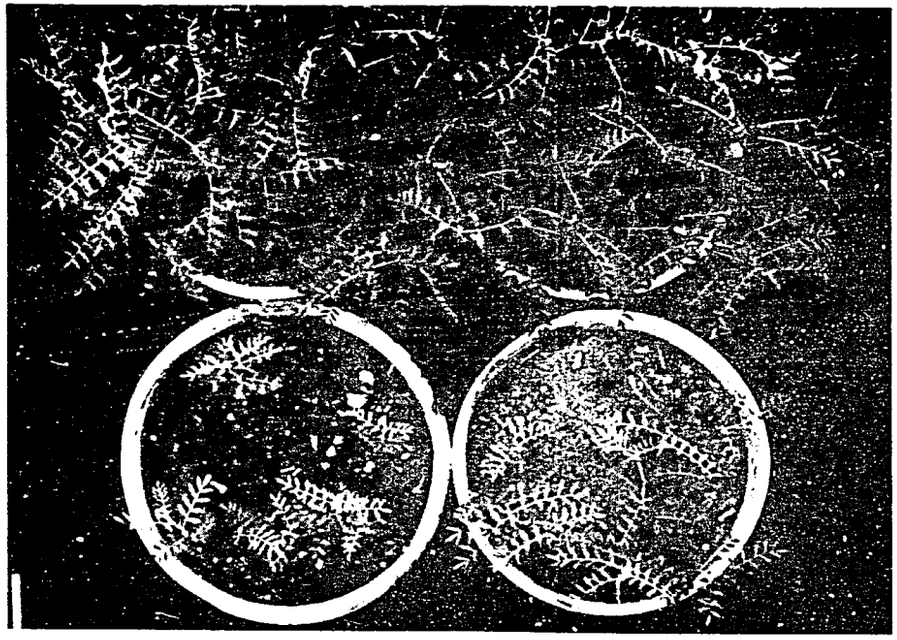

FIGURE 1. Effect of seed-treatment fungicides on emergence of chickpea seed infected with Ascochyta rabiei. Pots were planted with 25 seeds. Treatments were: Control (nontreated, noninoculated), upper left; zinc + maneb, upper right; oxycarboxin, lower right; and control (nontreated, inoculated), lower left.

FIGURE 2. The apical part of a germinating chickpea seed infected with Ascochyta rabiei is discolored (arrow) as a result of infection by the pathogen. Black spots on seed coat are pycnidia of the fungus.

FIGURE 3. Treatment of healthy chickpea seed with various fungicides before planting was ineffective in preventing heavy infection, dieback, or death of plants inoculated 2-3 weeks after emergence (middle, right). Healthy nontreated, noninoculated plants, left.

Table 1. Effect of seed-treatment fungicides on emergence of chickpea seed infected with Ascochyta rabiei[a].

Fungicide	Rate of Application (g/kg seed)	% Emergence[b]	% Healthy
Control (nontreated, noninoculated)	--	99	99
thiabendazole 60 W	3.0	94[c]	91
benomyl 50 W	3.0	93.5	90
chlorothalonil 75 W	3.0	79	56.5
carboxin 75 W	3.7	78.5	54.5
zineb 75 W	3.0	78.5	53
Dfn. 65 W	3.0	78	60.5
zinc + maneb 80 W	3.0	77.5	70
Cer. M 7.7 W	2.0	76.5	69
Dy. 50 W	3.7	76	56.5
maneb 80 W	3.0	74.5	65
captan 75 W	3.0	74	63.5
DCNA 75 W	3.0	74	58
PCNB 75 W	2.0	69	56
chloroneb 65 W	3.7	62.5	42
ETMT + PCNB L 21	2.0	60	27
copper oxychloride 35 W	3.7	57	8.5
oxycarboxin 75 W	3.7	51	9.5
Control (nontreated, inoculated)	--	46	6
Dex. 70 W	2.0	35.5	4.5

[a]Seeds inoculated with a heavy spore suspension of four Iranian isolates of A. rabiei. [b]Results are the average of two experiments consisting of four replications/experiment with 25 seeds/replication. [c]Results of one experiment.

chickpea seedlings, the seeds of which had been treated with different fungicides at the dosage rates indicated. Emergence of healthy plants in the nontreated noninoculated control was 99% (Table 1 and Fig. 1).

Effect of seed treatment fungicides on incidence of foliar infection by A. rabiei: None of the seed-treatment fungicides applied to healthy chickpea seeds before planting appeared to protect the foliage of seedlings which were inoculated with the fungus 3-4 weeks after planting (Table 2). Infection of inoculated seedlings was extensive in all treatments, regardless of the fungicide or dosage rate used to treat the seed. Within 3 weeks of inoculation, most inoculated seedlings were killed or heavily infected and stunted (Table 2 and Fig. 3). The different fungicides or dosage rates did not adversely affect seed germination or subsequent growth of chickpea seedlings before inoculation.

DISCUSSION

In several countries where Ascochyta blight is a serious disease of chickpea, breeding programs have been established to locate sources of resistance to strains of the pathogen (3). Until resistant varieties are developed, chemical control may be required to avoid crop failures in areas where the disease occurs in epidemic proportions. Unfortunately, very little information is available on use of fungicides to control foliar or seedborne infection by A. rabiei. Ascochyta is usually introduced into farmers' chickpea fields on infected seed (2, 6). A chemical seed treatment that prevents development of the disease from infected seed would be an important factor in restricting the spread of the pathogen into or within chickpea plantings. Halfon-Meiri (1) has shown that germination of chickpea seed infected with A. rabiei was usually reduced and that infected seedlings that survived often had poorly developed root systems and thin stems. In our experiments we found that the germination of nontreated, inoculated chickpea seed was reduced by more than 50% and that over 90% of the seedlings were diseased. Emergence of seedlings treated with certain fungicides, particularly the benzimidazoles, was over 90%, and less than 4% of the plants were infected with A. rabiei. Possibly, because of translocation of these systemic compounds from the treated seed into the stem tissues, the few lesions that developed on hypocotyls of infected seedlings were reduced in size, and plants grew normally.

Table 2. Effect of treating healthy chickpea seed with various fungicides on control of foliar infection by Ascochyta rabiei[a].

Fungicide	Rate of Application (g/100 g seed)	Disease Rating[b]
benomyl 65 W	0.125	3.4
do.	0.250	3.4
do.	0.500	3.5
thiabendazole 60 W	0.125	3.6
do.	0.250	3.6
do.	0.500	3.3
carboxin 75 W	0.125	3.7
do.	0.250	3.8
do.	0.500	3.6
oxycarboxin 75 W	0.125	3.6
do.	0.250	3.8
do.	0.500	3.8
chloroneb 65 W	0.125	3.1
do.	0.250	3.3
do.	0.500	3.5
captan 75 W	0.125	3.8
do.	0.250	3.6
do.	0.500	3.9
Control (nontreated, inoculated)	--	3.8
Control (nontreated, noninoculated)	--	0

[a]Plants were inoculated with a heavy spore suspension of four Iranian isolates of A. rabiei 3-4 weeks after planting.
[b]Disease rating, scale 0-4: 0, no infection; 4, all plants dead. Results are the average of two experiments consisting of six replications/experiment with 10 seeds/replication.

Systemic fungicides like thiabendazole and benomyl, which are effective in controlling seed-borne infection of A. rabiei, might also be translocated to the aerial parts of susceptible seedlings in sufficient quantities to protect them from infection for a period of time after planting. At the high inoculum levels of the pathogen used in our experiments, we did not obtain control of foliar infection by treating seeds with different dosage rates of several systemic and conventional fungicides. Perhaps, at lower inoculum levels, there would be a measure of control of the disease.

Literature Cited

1. HALFON-MEIRI, A. 1970. Infection of chickpea seeds by Ascochyta rabiei in Israel. Plant Dis. Reptr. 54: 442-445.
2. KAISER, W. J. 1972. Occurrence of three fungal diseases of chickpea (Cicer arietinum) in Iran. FAO Plant Prot. Bull. 20(4): 73-78; 98.
3. KAISER, W. J. 1973. Factors affecting the growth, sporulation, pathogenicity, and survival of Ascochyta rabiei. Mycologia. (In press.)
4. LUTHRA, J. C., A. SATTAR, and K. S. BEDI. 1935. Life-history of gram blight (Ascochyta rabiei (Pass.) Lab. =Phyllosticta rabiei (Pass.) Trot., on Cicer arietinum L.) and its control in the Punjab. Agr. & Live-Stock, India 5: 489-498.
5. SATTAR, A. 1933. On the occurrence, perpetuation, and control of gram (Cicer arietinum L.) blight caused by Ascochyta rabiei (Pass.) Labrousse, with special reference to Indian conditions. Ann. Appl. Biol. 20: 612-632.
6. ZACHOS, D. G., C. G. PANAGOPOULOS, and S. A. MAKRIS. 1963. Reserches sur la biologie, l'épidémiologie et la lutte contre l'anthracnose du pois-chiche. Ann. Inst. Phytopath. Benaki N.S. 5: 167-192.