SEED POTATOES AS A CARRIER FOR THE VERTICILLIUM WILT ORGANISM AND CONTROL OF THIS PATHOGEN BY CHEMICAL SEED PIECE TREATMENTS [1,2]

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SUMMARY

Among 110 lots of certified seed potatoes from 10 states and Canada in 1967 and 134 lots from 9 states in 1968, 51 and 30 percent, respectively, were infected from Verticillium albo-atrum inside the tubers and 41 and 13 percent of them carried soil on the tuber surfaces infested with the fungus. The strain found inside tubers was seldom the microsclerotial type which causes Verticillium wilt in Washington; therefore, infected tubers may not be important in introducing the organism into Washington. However, high populations of the microsclerotial type of V. albo-atrum in soil attached to seed tubers may be a prime source for introducing the organism into Washington soils. Once introduced V. albo-atrum may eventually cause 20-40% reduction in yield of the Russet Burbank variety. Captan, Polyram^R, and household bleach (sodium gypochlorite) were very effective in preventing germination of the fungus in small amounts of infested field soil such as that infesting tubers of potato seed. Liquid treatments of captan and household bleach were more effective than dusts of captan and Polyram^R. Sodium hypochlorite is not registered for use as a potato seed treatment.

INTRODUCTION

Initial introduction of Verticillium albo-atrum Renke and Berth, (the microsclerotial type) into field soils by infected and/or infested seed potatoes has been implied but never proven (7). Also, it remains to be proven whether or not this organism is a natural soil inhabitant, living in low populations on symptomless hosts (7, 22). Based on previous fumigation studies, yields of Russet Burbank potatoes grown on soils infested with this fungus were reduced 20-40% (4, 5, 11).

Field surveys and experimental results from Canada (13) and Israel (10) report that potato is responsible for introducing and perpetuating Verticillium albo-atrum in soils which cause wilts of many hosts. Cherries grown in Washington soils having a cropping history of either potatoes or other susceptible hosts have a greater chance of becoming infected with the pathogen (2). The organism was found in soils near Paterson, Washington where potatoes had been grown for a single season, but not in dryland wheat or a desert soil on the Horse Heaven Hills located 3 miles north from the Paterson area (G. D. Easton, unpublished data).

The Verticillium wilt organism of potatoes (V. albo-atrum) is mainly a parasite, which doesn't reproduce in the soil (8, 12), however, it does have a resting stage, microsclerotium, which can survive for many years in relatively stable populations in the soil (6, 14, 17, 23). These microsclerotia are formed in plant parts, even on

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1/ This investigation was made possible through grants supplied by the Washington State Potato Commission and facilities of Washington State University. Chemicals were supplied by Chevron Chemical Company and Niagara Chemical Division.

2/ Any trade name of a product used in these studies does not constitute a guarantee or warranty of the product by Washington State University or that the behavior of similar products would be the same or different from the ones used.

symptomless cultivated plants and weeds (7). The fungus can be easily transported from field in irrigation water and wind-blown dust (3).

Verticillium albo-atrum has been isolated from inside tubers and stems and to a limited extent in the outside periderm of tubers form some potato seed growing areas (1, 7, 16, 18). Treatment of infested and non-infested tubers with Semesan Bel^R showed that surface infestation was more important in causing wilt that inside tuber infection (16). It has recently been reported that viability of the microsclerotial type inside potato tubers diminishes with length of storage period (19).

In this study we collected and assayed lots of certified seed potatoes, mainly in Russet Burbank and Norgold Russet varieties, to be grown in Washington for the presence of Verticillium spp. We also screened chemicals for the elimination of V. albo atrum (microsclerotial type) from infested soil such as that attached to the outside of seed potatoes.

METHODS AND MATERIALS

About 100 tubers were collected from each of 110 and 134 lots of certified potatoes to be grown in Washington in 1968 and 1969, respectively. These tubers, obtained from newly opened sacks, were placed into new paper bags and stored at about 38°F for 2-4 moths until isolation for Verticillium spp. The soil from the outside of the tubers from each lot was brushed into a polyethelene bag with a disinfected (Roccal^R -60ml/gal water) wire brush and held at 38°F. Later this soil (0.5 1.0 g) was diluted with water (1:50) and assayed by a dilution plate method (3) using strepomycin-ethanol agar (15) plus 50 ppm sodium salt of Penicillin G. The tubers, after the brushing, were stored at 38°F. The inside of the tubers was assayed for the organism by peeling about 1-1 1/2 inch length of periderm from around the stem end of each tuber with a vegetable peeler. The peeled ends were disinfected with a 20% solution of household bleach (5.25% sodium hypochlorite) for 10 minutes. A 1/8 - 1/4 inch thick disc removed by a sterile knife was plated on streptomycin-ethanol agar (15) and incubated at about 74°F for 7 - 10 days before reading.

Selected isolates of V. albo-atrum from inside tubers and from infested soil on the outside of tubers were inoculated to soil compost in pots containing eggplant var. Black Beauty to test for pathogenicity. Wilt symptoms were read at 2 weeks and 2 months after transplanting.

Tap-water dilutions of prospective seed-treatment chemicals, Captan^R 50 WP (0.93-7.50 lb/100 gal), Captan^R 4 F (1.87-7.50 lb/100 gal) and household bleach (sodium hypochlorite -5.25%), 0.01-10% solutions, for control of V. albo-atrum (microsclerotial type) were added to soil collected from a known wilt infested potato field near Othello, Washington. Two ml of each dilution were added to one gram of infested soil and after 2 minutes the thoroughly mixed chemical and soil were diluted (1:50) with tap water for propagule assay by a dilution plate method (3). Three 1:50 dilutions were made for each rate of chemical tested with 3 petri plate replications per dilution.

Dusts of Captan^R 5D and Polyram^R 7D were also applied to infested soil for control of V. albo-atrum. Infested soil (1 g) was distributed between two, sterilized, No. 1 Whatman filter papers moistened with tap water in a petri dish. Doses of captan and Polyram^R (0.175-2.0 g) were added to the top of the filter paper and allowed to remain for 2 minutes. The upper paper with chemical was then discarded and the soil on the lower filter paper was diluted with tap water (1:50) and plated by the dilution plate method for assay of propagules (3). Three 1:50 dilutions were made for each rate of chemical tested with 3 petri plate replications per dilution.

RESULTS

Among the lots of seed sampled from 10 states and Canada in 1967 (110 lots) from 9 states and Canada in 1968 (134 lots), tubers of 51 and 30 percent of the lots had internal infections, and 41 and 13 percent of the lots, respectively, had

soil surface infestations of V. albo-atrum (Tables 1 and 2).

Isolates infecting the tubers were dark mycelial, microsclerial, and rately white types of V. albo-atrum. The dark mycelial type was found more often than the microsclerotial type. Rarely did more than five and usually only one or two tubers of the 100 tubers per infected lot have either dark or microsclerotial types of V. albo-atrum. The nonpathogen, Verticillium tricorpus Isaac was commonly isolated (9).

Isolates of Verticillium spp. from soil on the outside of tubers with the culture medium used were only of the microsclerotia type. Sub-cultures of some of these isolates to potato dextrose media produced verticillate conidiophores as well as microsclerotia of V. albo-atrum, but not dark mycelia of V. albo-atrum or the yellow pigmented mycelium of V. tricorpus. The estimated population of the microsclerotial type of V. albo-atrum found in soil attached to the outside of tubers varied from 37 to over 1 million per gram of soil. Many lots carried soil with several thousand propagules per gram of dry soil.

Cultures obtained from inside and outside of tubers from infected and infested lots of seed tubers were pathogenic to eggplant (Table 3).

Treatment with Captan^{R} (WP and 4F) at rates of 0.93-7.5 active chemical ingredients per 100 gal water and household bleach (5.25% sodium hypochlorite), at 1-10% solutions prevent germination of most of the propagules of V. albo-atrum in infested field soil, while the germination in untreated soil was 944 propagules per gram (Table 4). Addition of 01.175g - 2.0g of captan and Polyram^{R} dusts also greatly reduced propagule germination.

DISCUSSION

It was shown in this study that pathogenic types of V. albo-atrum are present in and on many tubers from lots of certified seed planted in Washington (Tables 1, 2, 3). It has been implied, but not proven that seed potatoes have introduced this fungus into non-infested soil around the world and caused production losses in potatoes and many other susceptible crops (7, 10, 13). We can assume that infested and infected seed potatoes are likewise introducing the organism into our Washington soils. How quickly the population builds up in the soil will depend upon further cropping history to wilt susceptible crops such as potato, watermelon and melons, contamination of soils by susceptible weeds such as pigweed, lambs quarter and night shade, and the spread of the organism by either irrigation water or wind blown dust.

Since the microsclerotial type of V. albo-atrum was isolated from only a few of the many tubers sampled and because viability of this organism declines with length of potato storage (19), the fungus found inside seed tubers may not be as important a source of contamination as that infesting the outside of tubers. High populations of the microsclerotial type infesting soil on seed tubers could certainly be a means of introducing and then propagating the organism as the crop from such infested seed tubers matures.

Liquid treatments of either captan or sodium hypochlorite were more effective than captan and Polyram^{R} dust treatments in controlling V. albo-atrum (microsclerotial type) in infested field soil. Dust is more popular in Washington as a seed treatment but may not provide adequate concentration of active ingredient to entirely eliminate this pathogen. Captan can be applied as a liquid treatment to seed potatoes as a dip or spray at a maximum dosage of 1.5 lb of active ingredient per 100 gal of water according to the Environmental Protection Agency (EPR) (20). Common household bleach which is a 5.25% solution of sodium hypochlorite can be used to treat beans for control of bacterial blight at a dosage of 10,000 ppm chlorine according to EPR (21). A 10% solution of sodium hypochlorite (5.25%), according to titrations and calculations would be equal to about 5000 ppm chlorine. However, this treatment has not been cleared for commercial use on potato seed pieces.

ACKNOWLEDGEMENTS

The writers wish to thank the many potato seed dealers in Washington who supplied tubers for our experiments free of charge.

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Table 1. *Verticillium albo-atrum* (dark mycelial and microsclerotial types) isolated from inside potato seed.

State or country grown	Lots of Seed Potatoes 1967[1/] Number infected out of total sampled[2/]	Per cent	1968[1/] Number infected out of total sampled[2/]	Per cent
California	0-3	0	0-1	0
Canada	0-2	0	2-5	40
Idaho	7-11	63	1-5	20
Minnesota	6-14	42	8-24	33
Montana	12-17	70	14-31	45
Nebraska	2-2	100	0-1	0
North Dakota	19-29	65	10-44	22
Oregon	1-2	50	0-4	0
South Dakota	--	--	1-1	100
Utah	1-2	50	--	--
Washington	8-26	30	4-18	22
Wisconsin	0-2	0	--	--
Totals	56-110	51	40-134	30

[1/] Year grown in state or country of origin.

[2/] Identifications made on streptomycin-penicillium-ethanol media.

Table 2. *Verticillium albo-atrum* (microsclerotial type) isolated from infested soil attached to potato seed.

State or country grown	Lots of Seed Potatoes 1967[1/] Number infested out of total sampled[2/]	Per cent	1968[1/] Number infested out of total sampled[2/]	Per cent
California	1-3	33	0-1	0
Canada	1-2	50	0-5	0
Idaho	6-11	54	0-5	0
Minnesota	8-14	57	5-24	20
Montana	7-17	41	5-31	16
Nebraska	2-2	100	0-1	0
North Dakota	16-29	55	5-44	11
Oregon	0-1	0	1-4	25
South Dakota	--	--	0-1	0
Utah	1-2	50	--	--
Washington	2-26	7	1-18	5
Wisconsin	1-2	50	--	--
Totals	45-109	41	17-134	13

[1/] Year grown in state or country of origin.

[2/] Identifications made on streptomycin-penicillium-alcohol media.

Table 3. Pathogenicity of *Verticillium albo-atrum* cultures isolated from seed potatoes to eggplant.

Source of isolates	No. showing symptoms in eggplant out of total tested	Percent showing symptoms
Inside tubers	35-145[1/]	24
Soil on tubers	10-34[2/]	29

[1/] Consisted of microsclerotial and dark mycelial types.

[2/] Consisted of microsclerotial type only.

Table 4. Effect of chemicals on *V. albo-atrum* (microsclerotial type) in field soil.

Chemicals	Rates active chemical ingredients	Estimated mean viable propagules per gram oven dry soil[1/]	No. times test repeated
Captan[R] 50 WP	0.93 lb/100 gal water	20	2
	1.87	18	7
	2.81	0	2
	3.75	15	8
	5.62	0	5
	7.50	0	5
Captan[R] 4F	1.87 lb (177 ml) per 100 gal water	0	4
	3.75 lb (354 ml) "	0	4
	5.62 lb (531 ml) "	0	4
	7.50 lb (708 ml) "	0	4
Captan[R] 5D[2/]	0.175g per g soil	383	2
	0.25g "	404	2
	0.5g "	404	2
	1.0g "	343	2
	2.0g "	101	2
Polyram[R] 7D[2/]	0.175g per g soil	232	2
	0.25 "	171	2
	0.5 "	121	2
	1.0 "	141	2
	2.0 "	60	2

(table continued)

Table 4. (continued)

Chemicals	Rates active chemical ingredients	Estimated mean viable propagules per gram oven dry soil[1]	No. times test repeated
Household bleach[2] (5.25% sodium hypochlorite)	0.01% solution	342	7
	0.1	130	8
	1.0	40	8
	10%	0	8
Control	--	944	8

[1] Average number of propagules counted on 9 assay plates per treatment.

[2] Actual solution of bleach applied, not active ingredients.