ANNUAL REPORT

Fungus and Bacterial Diseases Research

Summary of 1982 Results and Research Proposal for 1983

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Introduction

This section summarizes the work done in 1982 and includes new research proposals and a budget request for 1983. Detailed reports are included in attached sections of this report for those interested in further detail.

1. Blackleg Research

Objectives

Blackleg research in 1982 continued to focus on sources of *Erwinia carotovora* which may result in recontamination of "*Erwinia* - free" potato seed and means to reduce the threat of recontamination from these sources. The main objectives of the work were:

a. to determine to what extent *Erwinia carotovora* is present in surface and subterranean water samples collected from Colorado and other areas of the United States and also to determine if *E. carotovora* populations varied during the year;

b. to determine the effect of a seedpiece treatment containing plant growth-promoting rhizobacteria (PGPR) on potato plant growth, vigor and yield;

c. to continue investigation of the ability of *E. carotovora* to persist in the rhizosphere of weed hosts;

d. to determine the effect of post harvest application of a nitrogen based experimental compound RES-SRL 444 on Erwinia populations on tuber surfaces;

e. to continue studies on the generation of Erwinia aerosols from various sources.

Results

Objective a. The presence of Erwinia carotovora in surface and subterranean water.

Surface and subterranean fresh water from sources throughout Colorado and from 11 other states was assayed for the presence of Erwinia carotovora. A total of 47 streams and reservoirs were sampled during the year as well as several wells in southcentral and northeastern Colorado. Eighty five percent of the surface water sources sampled yielded Erwinia carotovora when the water samples were enriched anaerobically or filtered to concentrate the bacterial cells prior to enrichment.

Of 19 surface water sources in Colorado and one in Wyoming which were sampled on a monthly basis from January through November, 100% of the sources yielded Erwinia during every month of the year when they were sampled. In addition, samples of sea water from the Oregon and California coasts and the Gulf of Mexico collected as far as 18 miles from land yielded Erwinia cells.

Populations in the water in Colorado ranged from less than 1 cell/ml to 123 cells/ml. Populations were generally low from January to May. They reached their peak during June to September then began to decline and reached populations well below 1 cell/ml by November.

Erwinia carotovora subsp. carotovora was the dominant organism in surface water sources and reprsented 97.6% of the isolates identified.

No good evidence was obtained to suggest that Erwinia is present in subterranean water. Nearly all samples from wells did not contain the organism and the few samples which were positive could probably be attributed to the contamination of the water after it reached the surface.

Objective b. Plant growth promoting rhizobacteria (PGPR) (flourescent Pseudomonads) were tested alone and in combination with Captan as a pre-plant potato seedpiece treatment in the San Luis Valley and northeastern Colorado. Results show that PGPR were able to colonize the roots, stolons and daughter tubers of treated plants at both locations. Data from the early season assays showed that mixing the fungicide Captan with PGPR reduced the number of PGPR colonizing the roots by a factor of approximately 100. PGPR treatment had no obvious effect on typical blackleg expression in the field, on plant vigor and on total yield.

Studies on the effect of PGPR on recontamination of blackleg-free seed were initiated in 1982 and will continue in 1983.

Objective c. Weed-root samples were collected at three times during the growing season in the San Luis Valley. Contaminated weed-roots were not found in samples collected from virgin land and only one contaminated weed-root (out of 480 samples) was collected from land cropped continuously to potatoes for 20 years. The data suggest that virgin land areas do not have weed harboring E. carotovora and that continuous cropping of potatoes possibly generated an Erwinia

suppressive soil. Paradoxically, data from one continuously cropped field suggests lowlevels of E. carotovora can remain in association with weed-roots for at least 16 years since the last potato crop.

Data from soil survival studies conducted in the greenhouse showed that E. carotovora is unable to survive in association with weed-roots for prolonged periods of time.

Objective d. Post harvest spraying or dipping of tubers with experimental compound RES-SRL 444 failed to significanly reduce the population of Erwinia on tuber surfaces or to reduce the number of soft rot pockets developing on tubers after a period of time in storage. There was a trend toward lower populations on the tuber surface as the concentration of the compound in spray application was increased from 5 - 25%. Spray applied at the rate of 5% resulted in the lowest soft rot potential in the treated tubers.

Objective e. A cooperative project was initiated with Oregon State University to monitor the generation of Erwinia aerosols from the ocean along the Oregon coast. Data to date are preliminary only.

2. Ringrot Research

Objectives

a. to evaluate foliar and tuber symptom development in recently developed potato clones;

b. to continue investigating the effect of Corynebacterium sepedonicum inoculum levels on ringrot disease development in the potato cultivars Russet Burbank and Centennial.

Results

Objective a. Seedpieces from 10 potato clones were inoculated with Corynebacterium sepedonicum and evaluated for ringrot symptom development in the San Luis Valley. All clones (except WC285-18) developed typical foliar ringrot symptoms at approximately the same time including WC230-14 and Belrus in which ringrot infection is commonly latent. However, WC230-14 and Belrus foliar symptoms were evident only during the first 2 weeks of August in a limited number of plants. Symptoms (early dwarfing) developed in Russet Burbank more quickly than in any other clone tested (July 20). A selection of stems collected from symptomatic plants from all clones, except Belrus, had a positive stem squeeze (a diagnostic test conducted in the field) on August 26, 1982. Stems collected from cv. Belrus all had a negative stem squeeze at this time. Daughter tubers harvested from clones WC285-18, WC230-14, WC567-1, WC672-2 and Russet Burbank commonly had typical ringrot symptoms. Tubers from clones WC630-2 and WC708-6 had a very limited number of tubers with typical symptoms. Tubers with typical symptoms were not found in clones BC9289-1, Belrus and WC521-12. However, only a limited number of tubers were observed for clone WC521-12 because of poor plant emergence.

Objective b. Individual Russet Burbank and Centennial seed tubers were inoculated with concentrations of the ringrot bacterium (C. sepedonicum) ranging from 10^1 cells per tuber to 10^9 cells per tuber. Definite inoculum concentration effects on foliar symptom expression and daughter tuber symptom expression was observed in both field and greenhouse grown plants. Results indicate that daughter tuber infection can occur in the absence of foliar ringrot symptom expression. Foliar ringrot symptoms were observed in the field for cv. Russet

Burbank but not Centennial. Symptom expression for the inoculation series initiated in 1981 is shown in Table 1.

Table 1. The effect of Corynebacterium sepedonicum (ringrot) tuber inoculum concentration on symptom expression in progeny plants and daughter tubers.

Cultivar	Number of cells per mother tuber	Primary foliage symptoms 1/	Daughter tuber infection	Secondary foliage symptoms 2/
Russet Burbank	6.3×10^8	-	+	+
	10^6	-	+	-
	10^4	-	+	-
	10^2	-	-	-
	10^1	-	-	-
	Buffer	-	-	-
Centennial	6.3×10^8	-	+	-
	10^6	-	+	-
	10^4	-	-	-
	10^2	-	-	-
	10^1	-	-	-
	Buffer	-	-	-

1/ Inoculated tubers were planted in the field in 1981 and symptom expression recorded.

2/ Daughter tubers harvested in 1981 were replanted in the field in 1982 and symptom expression recorded.

3. Early Blight Research

Objectives

a. to determine the effectiveness of RES-SRL444 as a foliar application for early blight control and its effect on yield;

b. to determine the relative amount of foliar early blight resistance of recently developed potato clones;

c. to continue testing the accuracy of an early blight prediction model based on day-degree accumulation.

Results

Objective a. RES-SRL444 and RES-SRL445 alone and as a combined mixture were compared to Bravo 500 as a foliar application for early blight control in the San Luis Valley. Final disease ratings show Bravo 500 offered significantly greater ($P \leq 0.005$) early blight control than the experimental materials when data for the estimated percentage of leaf area blighted and the estimated percentage of leaflets infected are compared. RES-SRL444 alone and in combination with RES-SRL445 significantly reduced the estimated percentage of defoliation observed compared with the non-treated control and RES-SRL445 alone. RES-SRL444 alone and in combination with RES-SRL445 also significantly reduced the average number of early blight lesions per leaflet compared to the non-treated control ($P \leq 0.01$). Early blight control provided by RES-SRL444 was equivalent to that offered by Bravo 500 when data for the estimated percent of defoliation and the average number of early blight lesions per leaflet are compared ($P \leq 0.01$). Generally, Bravo 500 provided the most effective early blight control followed by RES-SRL444 alone and in combination with RES-SRL445. RES-SRL445 alone did not significatly reduce disease ratings ($P \leq 0.01$) when compared to the non-treated control at any time during the growing season. Treatment with Bravo 500 significantly increased total tuber yield ($P=0.05$). There were no other treatment effects on tuber yield or grade. Data for the average number of early blight lesions per leaflet are summarized in Table 2.

Table 2. The Effect of Different Fungicide Treatments on the Infection of Russet Burbank Potato Foliage by Alternaria solani - Center, Colorado, 1982.

Fungicide Treatment[1/]	Average number of early blight lesions per leaflet[2/3/]		
	August 5	August 17	August 26
1) Non-treated	1.8 a	12.3 a	40.7 a
2) Bravo 500 (s pt/A)	0.1 c	0.3 c	5.7 d
3) RES-SRL444 (10% v/v)	0.5 bc	2.3 bc	21.2 bcd
4) RES-SRL445 (5% v/v)	1.4 ab	7.2 ab	31.0 ab
5) RES-SRL444 & 445 (10% and 5% v/v)	0.5 bc	5.2 bc	23.7 bc

1/ Fungicides were applied at 50 p.s.i. with a ground sprayer at a net rate of 90 gallons per acre. Treatments were replicated four times.

2/ Eight leaves from each treatment plot were collected and the number of leaflets and early blight lesions determined by actual counts.

3/ Treatment means with different letters differ significantly ($P \leq 0.005$) on each collection date.

Objective b. Sixteen potato clones were evaluated in the field for foliar early blight resistance in northeastern Colorado. Clones FL-1481, FL-1455, WC521-12, NY67 and NY59 were consistently more resistant to the effects of early blight, especially late in the growing season when disease pressure was greatest. Clones FL-1455 and NY67 appeared to be relatively susceptible to early blight leaf infection but they were not readily defoliated.

Objective c. A study was conducted in the San Luis Valley and Morgan County during the 1980 season to 1) further verify the day degree model developed earlier and being used in the San Luis Valley and 2) to determine if the large difference in day degrees

accumulated between planting and lesion appearance between the two areas reported in 1979 was the effect of potato variety or location. Field observations were continued in Morgan County during 1981 and 1982 to determine the day degree accumulation threshold required for initial early blight (*Alternaria solani*) lesion appearance in potatoes.

The number of day degrees accumulated from planting until the initial appearance of early blight lesions in Morgan County agreed quite closely for each year tested. The data suggested that it is possible to predict the initial appearance of early blight in both the San Luis Valley and Morgan County using the day degree model and threshold values determined for each location.

Fungicide treatment schedules initiated prior to the appearance of the first early blight lesions did not provide additional disease control. Use of the model in Colorado will enable proper timing of fungicide application and more efficient chemical usage.

Conclusions

Good progress was made towards reaching the research objectives. It was found that surface water collected from Colorado and other areas of the United States is commonly contaminated with *E. carotovora* throughout the year and that population levels are greatest during the summer months. Data suggest that the levels of *E. carotovora* present in irrigation water influence the amount of aerial blackleg lesions observed in the field.

The accuracy of an early-blight prediction model was again verified for northeastern Colorado. A similar model is being used successfully by potato growers in the San Luis Valley to time initial fungicide applications. An experimental fungicide tested for early-blight control in the San Luis Valley

compared very favorably with Bravo 500. Some recently developed potato clones were found to possess resistance to foliar early blight infection and defoliation.

Ringrot inoculation studies identified recently developed potato clones with marginal symptom development. Strong evidence was again found to support the hypothesis that latent ringrot infection can occur even in very susceptible clones.

Research Proposal 1983

1. Blackleg Research

   a. Monitoring of water sources for Erwinia contamination will occur with emphasis on increased sampling of well water and the role of stream bed sediments in maintaining Erwinia populations in water. A major effort will be directed toward determining the parameters and importance of contaminated irrigation water as a source of Erwinia recontamination of clean potato stocks and weed thizospheres. Serological methods will be developed to relate the bacteris found in tubers with those found in water sources.

   b. Studies will be made to determine if an Erwinia suppressive factor is present in a field soil collected after potato monoculture and if these organisms might affect the role of recontamination of clean stocks.

   c. Studies will be made to determine if E. carotovora aerosols can be generated on the Oregon Coast, transported inland with weather systems and deposited with precipitation. This work will require additional sources of funding which has been requested from the USDA in order to complete the work.

2. Ringrot Research

   a. The relationship between C. sepedonicum inoculum levels and symptom expression will be studied further using material from the 1981 and 1982 field plots. This work will be done in conjunction with the testing of advanced clones for ringrot reaction.

   b. Several methods including an eggplant pathogenicity test, ELISA and latex agglutination will be compared to determine their efficiency in detecting Corynebacterium in symptomless potato tubers

3. Early Blight Research

As time and funds allow:

a. New clones will be evaluated for early blight resistance.

b. The day degree model for timing fungicide applications for early blight control will be developed further.

Budget Requested

Labor	2500.00
Travel	1250.00
Supplies	1450.00
	$5200.00

The Presence of *Erwinia carotovora* in Water

Introduction

The recent discovery that *Erwinia* could be detected in surface water and that this could serve as a source of recontamination of disease-free potato seed prompted an extensive study of the presence of the organism in Colorado water. The study was expanded to include water samples from both major and minor streams in Colorado and other parts of the United States as well as samples of underground water, lakes, reservoirs and sea water.

Attempts were also made to determine the contamination of 8 major streams in various parts of Colorado from their source in the mountains well into the agricultural areas. Population counts in the water from various areas on the streams were made.

Methods

Water samples were collected at monthly intervals generally from January through December from selected streams in four distinct areas (San Luis Valley, Northeastern Colorado, Arkansas Valley and Steamboat Springs) for *Erwinia* assays. From all sites at least 50 ml of water but in many cases 1 L or more was collected. At certain selected locations up to 10 L of water was routinely collected for analysis. Additional samples from a wide range of surface water sources including streams, lakes and reservoirs were collected periodically and processed.

To determine the extent of contamination of major streams in Colorado samples from 3-6 separate locations on a particular stream beginning in the mountains near the origin of the stream as far as possible from agricultural areas and continuing at intervals down the stream well into the agricultural

areas. In all, 8 major streams including the Rio Grande River and Saguache Creek in the San Luis Valley; the South Platte and Big Thompson Rivers along the Front Range; the Elk, Yampa and Colorado Rivers in the Northwest mountainous regions; and the Arkansas River in the Southeast were studied in this way.

In addition, single or sometimes 2 sites on 12 additional streams in Colorado and 30 streams, lakes, and reservoirs in 12 other states were sampled either periodically or on a monthly basis.

All water samples were usually processed in two ways. First 50 ml of water was mixed with an equal volume of enrichment medium, incubated anaerobically for at least 48 hrs then plated on a semi-selective medium to detect Erwinia. The remainder of the water sample (if any) was passed through a bacteria proof filter to concentrate the bacterial cells. The material collected on the filters was resuspended in a small volume of water, mixed with enrichment medium and incubated anaerobically as described above.

Random Erwinia colonies were removed from isolation plates, purified, and identified using standard bacteriological methods.

For quantitative assays of Erwinia populations in water aliquots from filter concentrated or non-filtered samples were appropriately diluted, plated on a semi-selective medium and the number of Erwinia colonies which developed were counted.

Some work was also done to determine if Erwinia cells were present in the sediment at the bottom of streams at selected locations. To accomplish this two water samples were collected, one before and one after the sediment was disturbed. Erwinia populations in the two samples were compared by quantitative population assays using methods described above.

Results

Table 1 shows that every stream sampled in the state of Colorado yielded Erwinia at least at some time during the 12 month period. Those streams on which several separate sites were regularly sampled (numbers 1 through 8 Table 1) yielded Erwinia every month that they were sampled during the course of the year.

Of the water samples from 31 sources from states other than Colorado (Table 2), 24 (77.4%) yielded Erwinia. Only a single sample from most of these sources was processed, however.

Erwinia carotovora subsp. carotovora was the predominant organism isolated from water from all sources. Overall this organism represented 97.6% of the isolates identified from the water samples.

Erwinia carotovora subsp. atroseptica was rarely detected and tended to be found mainly during the early spring, late autumn and winter months.

Studies on the presence of Erwinia at various locations on 8 streams in Colorado (Table 3) showed that the organism was regularly detected throughout the year in mountainous areas, transitional areas between mountain regions and agricultural areas and in agricultural areas in the state. The single exception was one site on the South Platte River. Erwinia was never recovered from water samples collected from the highest (mountainous) location on the stream.

The frequency of detection in the various areas was similar although there was, in most cases, a tendency to detect the organism more frequently at sites located in the agricultural than in the non-agricultural areas.

Quantitative data (Table 4) show that Erwinia populations in Colorado water sources were generally quite low. Populations of less than 1 cell per ml of water were common in most of the streams during most of the year.

Table 1. Presence of *Erwinia carotovora* in Water Samples From Several Streams in Colorado and Wyoming Sampled Regularly January - December 1982

Streams	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
Rio Grande River	+	+	+	+	+	+	+	+	+	+	+	+
Saguache Creek	+	+	+	+	+	+	+	+	+	+	+	+
South Platte River	+	+	+	+	+	+	+	+	+	+	+	+
Big Thompson River	+	+	+	+	+	+	+	+	+	+	+	+
Arkansas River	+	+	+	+	+	+	+	+	+	+	+	+
Elk River	+	+	+	+	-	1/	+	+	+	+	+	+
Yampa River	-	+	+	+	+	+	+	+	+	+	+	+
Colorado River	-	-	0	+	-	+	+	+	+	+	+	+
Illinois River	-	-	0	+	-	+	+	+	+	+	+	+
Laramie River	-	-	+	+	-	+	+	+	+	+	+	+
Oak Creek	-	-	-	-	-	+	+	+	+	+	+	+
Buffalo Creek	-	0	+	-	-	-	+	+	0	0	+	+
San Luis Creek	-	0	+	0	+	+	+	+	+	0	0	0
Squaw Creek	-	0	+	0	0	0	+	+	+	+	+	+
Poncho Creek	-	-	+	0	0	+	+	+	+	+	+	+
South Arkansas River	+	0	+	+	0	+	+	+	+	+	+	+
Kerber Creek	-	0	0	0	0	0	0	+	+	+	+	+
Chalk Creek	-	0	+	+	+	+	+	+	+	+	+	+
Poudre River	-	-	-	-	-	-	+	-	-	+	-	-
Dead Horse Creek	-	-	-	-	-	-	-	+	-	-	-	-

1/ No sample collected.

Table 2. Presence of Erwinia carotovora in Water Samples Collected From States Other Than Colorado 1982

State	Number of Water Sources Sampled	Number of Water Sources Yielding Erwinia
Oregon	5	3
Washington	2	2
Wisconsin	1	1
Utah	4	4
Idaho	2	1
New Mexico	3	3
Texas	3	1
Minnesota	1	1
Kansas	1	0
Oklahoma	1	1
Wyoming	7	6
California	1	1
	31	24

Table 3. Presence of Erwinia carotovora in Water Samples collected from Various Sites on 8 Streamsin Colorado January through December 1982.

Stream	Site No.	Location[2/]	J	F	M	A	M	J	J	A	S	O	N	D
Arkansas River	1	MT	-[1/]	-	0	+	+	0	+	+	+	0	+	0
	2	MT	+	+	+	+	+	+	-	+	+	+	+	+
	3	TR	-	-	-	+	+	+	+	+	+	+	+	+
	4	TR	-	-	+	+	+	+	+	+	+	+	+	+
	5	Ag	-	-	+	+	+	+	+	+	+	+	+	+
Elk River	1	MT	-	0	0	+	-	+	+	+	+	+	+	+
	2	TR	+	+	0	+	-	+	+	0	+	+	+	+
	3	Ag[3/]	-	+	+	0	_	+	+	+	+	+	+	+
Yampa River	1	TR	-	-	-	+	+	+	+	+	+	+	+	+
	2	TR	-	+	0	-	+	+	+	+	+	+	+	+
	3	Ag[3/]	-	0	+	+	-	0	+	+	+	+	+	+
Colorado River	1	MT	-	-	0	+	-	+	+	+	+	+	+	+
	2	MT	-	-	0	+	-	+	+	+	+	+	+	+
Rio Grand River	1	MT	+	+	0	0	0	+	+	0	+	0	+	+
	2	MT	+	-	0	+	0	+	0	+	+	0	+	+
	3	TR	0	+	0	0	+	+	+	+	+	+	+	+
	4	TR	+	+	0	+	+	+	+	+	+	+	+	+
	5	Ag	+	+	0	+	+	+	+	+	+	+	+	+
	6	Ag	+	+	+	0	0	+	+	+	+	+	+	+
Saguache Creek	1	MT	+	0	-	-	-	+	+	+	+	+	+	+
	2	TR	0	0	-	+	+	+	+	+	+	+	+	+
	3	Ag	0	+	-	0	0	+	+	0	+	+	+	+
	4	Ag	0	-	-	-	-	-	0	0	0	0	-	-

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Table 3. (continued)

Stream	Site No.	Location[2/]	J	F	M	A	M	J	J	A	S	O	N	D
South Platte River	1	MT	0	0	0	0	0	0	0	0	0	0	0	0
	2	TR	+	+	+	+	+	+	+	+	+	+	+	+
	3	Ag	+	+	+	+	+	+	+	+	+	+	+	+
	4	Ag	+	+	+	+	+	+	+	+	+	+	+	+
	5	Ag	+	+	+	+	+	+	+	+	+	+	+	+
	6	Ag	+	+	+	+	+	+	+	+	+	+	+	+
Big Thompson River	1	MT	+	0	0	0	+	+	+	+	+	+	0	0
	2	MT	-	-	+	+	+	+	+	+	+	+	+	+
	3	TR	+	0	+	0	+	+	+	+	+	+	+	0
	4	TR	+	+	+	+	+	+	+	+	+	+	+	+
	5	Ag	+	+	+	0	+	+	+	+	+	+	+	+

[1/] no sample collected usually due to the fact that the stream was frozen over.

[2/] MT = Mountainous Areas; TR = Transitional Between Mountainous and Agricultural Areas; Ag= Agricultural Area.

[3/] Ranching only.

Table 4. Populations of Erwinia carotovora in Colorado Streams January - December 1982

Stream	Site No.	Location[1/]	Month and Erwinia Population (Cells/ml of water) Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
Arkansas River	1	MT	-	-	-	-	-	.25	.17	1.33	.47	.02	.02	<1
	2	TR	-	-	-	-	-	.18	.17	2.94	.15	.03	.05	<1
	3	Ag	-	-	-	-	.69	1.03	1.39	1.83	1.06	1.96	.78	-
Elk River	1	MT	-	-	-	-	-	.02	.02	.36	.26	.02	.02	.08
Yampa River	1	MT	-	-	-	-	-	.02	1.03	1.55	3.9	2.2	.17	.09
	2	TR	-	-	-	-	-	-	-	-	.17	.02	.02	.11
Oak Creek	1	MT	-	-	-	-	-	.13	.33	1.74	2.48	.92	.14	.12
South Platte River	1	MT	0	0	0	0	0	0	0	0	0	0	0	0
	2	TR	6.0	.04	2.0	3.8	1.0	.2	<1	2.0	17.0	0.1	<1	<1
	3	Ag	<1	0.8	<1	0.2	1.2	8.0	<1	0.2	15.0	3.0	5.0	<1
	4	Ag	<1	1.0	<1	<1	6.0	21.0	8.0	123.0	23.0	9.0	20.0	19.0
	5	Ag	<1	.04	<1	<1	3.4	35.0	.4	11.0	73.0	23.0	23.0	5.0
	6	Ag	<1	.02	<1	<1	2.0	11.0	3.0	57.0	39.0	8.0	5.0	6.0
Big Thompson River	1	MT	4.0	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1
	2	MT	-	-	<1	<1	<1	<1	1.0	<1	.02	<1	<1	<1
	3	TR	<1	<1	<1	<1	<1	<1	<1	<1	.2	<1	.04	<1
	4	TR	2.0	.06	<1	2.0	3.0	36.0	<1	.2	2.0	5.0	7.0	14.0
	5	Ag	.4	2.4	<1	<1	.02	5.0	2.0	1.0	11.0	20.0	4.0	5.0
Rio Grande River	1	MT	.0002	.0002	0	0	0	.0002	.0002	0	.0002	0	.0002	.0002
	2	MT	.0002	-	0	.0002	0	.0002	0	.02	.0002	0	.02	.0002
	3	TR	0	.0002	0	0	.0002	1.0	.0002	.02	.02	.02	.02	.0002
	4	TR	.0002	.0002	0	.0002	.02	4.0	.02	2.0	.2	.2	.02	.02
	5	Ag	.02	.02	0	.0002	.02	.2	1.0	2.0	.68	.54	.08	2.4
	6	Ag	.02	.02	.0002	0	0	1.4	.0002	.6	2.2	.44	.4	.08

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Table 4. (continued)

Stream	Site No.	Location[1/]	Month and Erwinia Population (Cells/ml of water) Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
Saguache Creek	1	MT	.02	0	-	-	-	.02	.2	1.0	.04	.02	.02	.0002
	2	TR	0	0	-	.0002	.0002	4.0	1.0	6.0	1.8	.04	.04	.2
	3	Ag	0	.0002	-	0	0	.02	.26	0	9.2	9.2	3.4	.0002
	4	Ag	-	-	-	-	-	0	0	0	0	-	-	0
Illinois River	1	TR	-	-	-	1.5	-	.41	29.4	10.0	18.4	17.9	8.2	2.8
Colorado River	1	MT	-	-	-	-	-	.02	.45	1.16	.06	.08	.14	.19

[1/] MT = Mountain Area, TR = Transitional Area between Mountain and Agricultural Areas, Ag = Agricultural Area.

Populations of from 1 cell/ml to over 100 cells/ml were detected at some locations during the summer period (June to October). The highest populations tended to be in streams located at lower elevations (such as the South Platte River) and at sites in the agricultural areas rather than those located in the mountain regions.

Samples of water from wells in the San Luis Valley and Northeastern Colorado yielded inconclusive results. Some difficulty was encountered in obtaining samples witout risking contamination from surface sources. Occasional samples yielded Erwinia but it is not certain if the bacteria were present in the subterranean water or originated from other sources.

Samples of sea water from the Oregon, California, Texas, Louisiana and Florida coasts were found to contain Erwinia. Of 12 samples assayed, 9 (75%) yielded Erwinia carotovora var carotovora isolates.

The effect of disturbing stream bed sediments on the population of Erwinia in the water at 5 sites on the South Platte River is shown in Table 5. The data show that disturbing the sediments often markedly increased Erwinia populations in the water. The effect was more striking at some sites than at others and also was more pronounced during the warmer part of the season (July to October) than during the cooler period (November and December).

Conclusion

Erwinia, primarily Erwinia carotovora subsp. carotovora is commonly present in surface water sources in Colorado and other states. The organisms persisted throughout the year in all of the streams sampled in Colorado. The organism is present with about equal frequency at locations in the

Table 5. The Effect of Disturbing Stream Bed Sediments at Three Sites on the Population of *Erwinia carotovora* in Water Samples

Site	Stream Bed Condition	Month and *Erwinia* Population (cells/ml) in Water					
		July	Aug	Sept	Oct	Nov	Dec
1	Normal	0	0	0	0	0	0
	Disturbed	0	0	0	0	0	0
2	Normal	0	1.06	8.69	0.1	0	0
	Disturbed	1	3.00	34.0	0	6.0	2.0
3	Normal	0	0.2	7.51	1.6	2.65	0
	Disturbed	700.00[1/]	45.0	53.0	4.0	2.0	0
4	Normal	4.38	62.39	11.85	5.34	10.61	9.65
	Disturbed	25.0	215.00	123.0	20.0	30.0	6.0
5	Normal	-	6.46	39.23	13.11	12.61	2.66
	Disturbed	-	45.0	62.0	9.0	9.0	0
6	Normal	1.6	29.84	20.49	4.8	3.06	3.49
	Disturbed	3.0	55.0	37.0	4.0	8.0	3.0

[1/] Estimated

mountains far removed from agricultural areas and in the agricultural areas. This suggests that agricultural operations are not the sources of Erwinia contamination in streams and other surface water.

Erwinia populations in the water tended to be low (1 cell/ml) during most of the year in most locations. Populations as high as 20-120 cells/ml were however detected at some locations. Maximum populations occurred during the June to October period at most locations. Populations tended to be higher in the Northeastern Colorado streams than in the San Luis Valley and mountain streams. Locations in the mountain areas tended to have lower populations than sites on the same stream located downstream, especially than in the agricultural areas. This may indicate that the bacteria increase more rapidly in the warmer water at lower elevations or that cells are deposited from crops by irrigation water.

The fact that disturbing the stream bed sediments drastically increased Erwinia populations in stream water at some times of the year at some locations suggests that this may be a major site where Erwinia multiplies in the stream to maintain populations the year around and to increase populations during the warmer parts of the season.

The Effect of Plant Growth-Promoting Rhizobacteria Seedpiece Treatment on cv Monona Potatoes in Northeastern Colorado

G.D. Franc and M.D. Harrison

Abstract

Formulated plant growth-promoting rhizobacteria (PGPR), marked by rifampicin resistance, were used to treat cut (cv Monona) seedpieces.

There was no effect of PGPR treatment on plant emergence, stand, height, vigor, typical blackleg and *Verticillium* wilt observed in the field, internal stem, external stem, and external leaf populations of fluorescent PGPR and *E. carotovora*, mother seedpiece decay, *Rhizoctonia* stem cankering, the number of potato stems and tubers per hill, the number of tubers per stem, plant weight, the number of *E. carotovora* CFU per gram of root, the number of *E. carotovora* CFU, total and nonfluorescent PGPR CFU present on daughter tuber surfaces and within the peel at harvest and, lastly, no effect on tuber yield or grade.

Data showed that PGPR readily colonized potato root surfaces under growing conditions in northeastern Colorado. Additionally, PGPR were present on daughter tuber surfaces at the time of harvest. Mixing Captan with PGPR treatments reduced colonization by a factor of ca 100X.

Inspection of bacterial CFU count data indicated that large variations in CFU occurred between replications. This made it difficult to demonstrate significant differences among treatment means. This is especially evident for total and nonfluorescent PGPR CFU count data. Data should be considered with this in mind, i.e., the statistical analysis may be very conservative.

Treatment with RHX 501 (PGPR alone) resulted in significantly more total PGPR (nonfluorescent + fluorescent) and nonfluorescent CFU per gram of root early in the growing season. This demonstrated that at least two PGPR "biotypes" or strains were isolated and they resulted from the initial PGPR treatment. This is especially evident when actual plate count data are considered since rifampicin resistant isolates were never observed in assays using material collected from non-PGPR plants. Data collected showed that three PGPR strains, based on the intensity of fluorescence, were actually present on assayed material. This is consistent with observations made during CFU assays although only the distinction between nonfluorescent and fluorescent PGPR was made.

Testing of selected isolates with the gram stain, the oxidase test and O-F (Hugh and Leifson's) test did not allow further distinction among the PGPR strains and was consistent with results expected for typical Pseudomonas isolates.

If formulated PGPR were contaminated with non-PGPR or strains developed during PGPR formulation, this is a possible explanation for the lack of treatment response in potatoes. The original PGPR cultures and subsequent formulations as well as PGPR strains isolated from treated plants need further testing.

Materials and Methods

A grower-cooperator's field near Wiggins, Colorado was selected for the study. The field had a sandy-loam soil type and was located in an area with an elevation of ca 4,500 - 5,000 msl (mean sea level). The entire field was irrigated with a center pivot overhead sprinkler irrigation system. The treatment plots were planted within a commercial potato field planted at the same time (within 24 hr) with the cultivar Monona. The treatment plots received the same fertilizer, fungicide, and pesticide applications as the commercial field. The

grower-cooperator's fertilizer and pesticide chemical application schemes are listed in Tables 1 and 2.

The entire field was pre-irrigated (2 in) before plowing. Sprinkler irrigation was initiated on June 28 and ca 2 in of water per week was applied through July and the first week of August, 1 3/4 in per week the remainder of August, and 1 in per week during the first two weeks of September. In addition, considerable rainfall fell before and during the growing season.

The area of the field used for the study had not been cropped to potatoes for an estimated 20 years. Since then, mostly corn and pinto beans had been grown. The recent cropping history was corn in 1981 and 1980, pinto beans in 1979 and corn in 1978.

On June 14 the plots received considerable hail damage. The estimated percentage of defoliation observed in the plots was determined on June 16 using the Barratt-Horsfall (0-11) scale (Table 3). Although 19-25% of the foliage was gone, the plants were very active and recovered within a short period of time.

The seed source used in the study was cv Monona, Minnesota certified blue tag seed. The bulk seedlot (ca 430 cwt) was shipped from East Grand Forks, Minnesota, on March 15, 1982. The seedlot tubers were described by the inspector as mature, firm, generally well-shaped, slightly dirty, and possessing grade defects within tolerance. Less than 0.5% soft-rot was observed at the time of shipping (Appendix 1).

Seed tubers used in the study (500 lb) were randomly selected from the bulk seedlot on April 12, 1982 at Wiggins, Colorado, and stored in sterile burlap bags at 40°F at Fort Collins until May 10. On May 10, six lots of 100 seedpieces for each treatment ($\bar{X}$ seedpiece weight = 2.1 ounces) were cut by hand from the tubers with sterile knives and placed in sterile paper bags. Freshly cut seedpieces were treated by adding the appropriate amount of formulated dust (1.0 lb/100 cut seed) to the bag and shaking. Formulated dust

Table 1. Fertilization program superimposed upon plant growth-promoting rhizobacteria (PGPR) field trial plots - Wiggins, CO, 1982.[1/]

Date	Item
	Potash - dry. 120 lb available per acre plowed down.
5/10/82	Starter used at planting time was at the rate of 10 gal per acre (Analysis: 8-20-8).
6/11/82	Side dress with liquid fertilizer (32%)(90 lb available nitrogen per acre).
6/28/82	Liquid fertilizer (32%) added through center pivot sprinkler system (20 lb available nitrogen per acre).
7/1/82	Foliar application of materials with a ground rig. Per acre: 2 gal analysis 18-5-5 1/2 pt zinc 1 oz copper 1 oz solubar 1 pt calcium 1 pt magnesium 1 pt manganese 1 1/2 pt Bravo 500 (fungicide)
7/7/82	Liquid fertilizer (32%) and 12-0-0-26 added through center pivot sprinkler system (20 lb available nitrogen per acre and 12 lb of sulfur added).
7/10/82	Same as 7/7/82.
7/13/82	Same as 7/7/82.
7/17/82	Liquid fertilizer (32%) added through center pivot sprinkler system (20 lb available nitrogen per acre).
7/21/82	Same as 7/17/82.
7/24/82	Same as 7/17/82.
8/17/82	Same as 7/17/82

[1/] Information provided by the grower-cooperator.

Table 2. Chemical spray program superimposed upon plant growth-promoting rhizobacteria (PGPR) field trial plots - Wiggins, CO, 1982.[1/]

Date	Item
6/11/82	1 1/2 qt per acre of Di-syston applied with fertilizer.
6/27/82	3/4 pt per acre of Sencor 4 flowable.
7/1/82	1 1/2 pt Bravo 500 per acre.
7/14/82	5.3 oz Pydrin, 1 1/4 lb M-45 per acre.
7/31/82	5.3 oz Pydrin, 1 1/2 lb M-45 per acre.
8/16/82	5.3 oz Pydrin, 2 lb M-45 per acre.
8/31/82	5.3 oz Pydrin, 2 lb M-45 per acre.

[1/] Information provided by the grower-cooperator.

Table 3. The amount of hail damage observed in PGPR field plots on June 16 - Wiggins, CO, 1982.

Treatment	Estimated percentage of defoliation observed[1/]
1) RHX 500 (talc alone)	19%
2) RHX 505 (Captan alone)	25%
3) RHX 504 (Captan + PGPR)	22%
4) RHX 501 (PGPR alone)	25%
	NSD[2/]

[1/] Five Barratt-Horsfall (0-11) scale ratings were made in each plot.

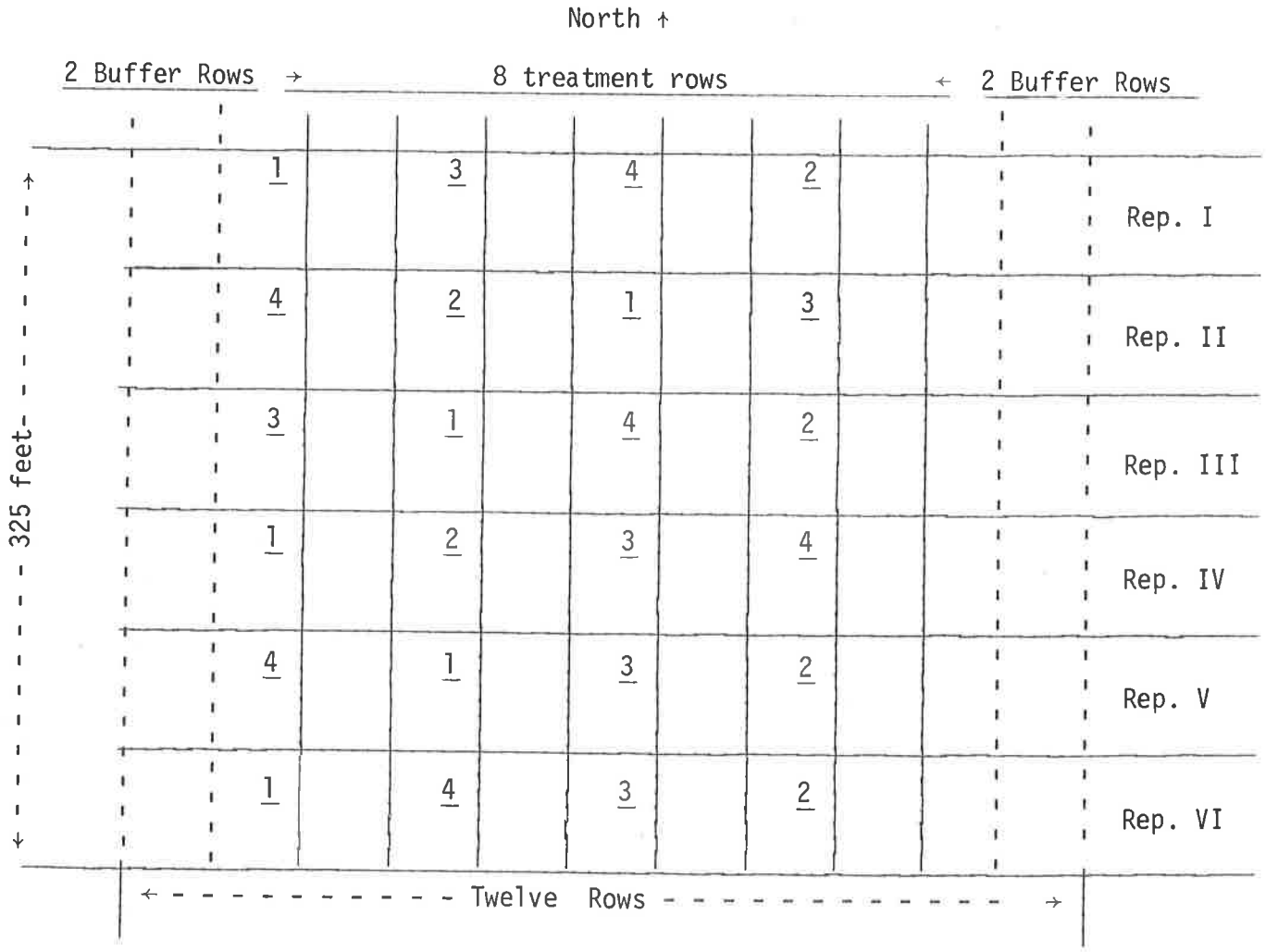
[2/] NSD - No significant differences were observed ($P \triangleq 0.05$).

was prepared by the Rohm and Haas Company. Seedpiece coverage was very uniform using this method and caking did not occur. Treated seed was immediately stored at 40°F and planted within 24 hr. The treatment plots were planted by hand at 12 inch spacing into rows hilled previously (24 hr prior) by a commercial potato planter. These rows received preplant fertilizer at the time of hilling (Table 1, 5/10/82). Treatment plots were measured and flagged in a randomized complete block design consisting of four treatments and six replications at the time of hilling. Individual treatment plots consisted of two potato rows each 50 feet long. The experimental design used is illustrated in Figure 1. Treatment and planting order was RHX 500 (control: talc alone), RHX 505 (Captan alone), RHX 504 (PGPR + Captan) and RHX 501 (PGPR alone). There was an intermittent drizzle the evening before, during and immediately after planting. Therefore, soil moisture was very adequate at the time of planting. Soil temperature at seedpiece depth 3 hr post planting was 14°C. Soil temperature was determined and recorded periodically during the growing season (Table 4).

On May 20, 15 soil cores (ca 5-7 in deep) were randomly selected from the side of the hills within the field plot and composited to determine the resident Verticillium dahlia (micro-sclerotial form) populations within the soil. Seven petri plates containing ethanol agar were inoculated with 20 mg of soil each and the Verticillium propagule concentration determined using a pre-established method (M.D. Harrison and C.H. Livingston , Pl. Dis. Reporter 50:897-899). All plates were negative thereby estimating the resident Verticillium population at less than one propagule per 20 mg of soil assayed (seven replications). This is considered to be a low soil-borne inoculum load for Colorado.

The tuber seedlot used was tested during April and May, 1982, for its soft--rot potential, the amount of internal tuber borne Verticillium, the amount of

Figure 1. Field plot plan used for plant growth-promoting rhizobacteria (PGPR) field trials - Wiggins, Colorado, 1982.1/



1/Minnesota certified seed (cv Monona) was used as the seed source. Each treatment plot consisted of two rows 50 feet long (6.5×10^{-3} acre).

2/Fifty seedpieces per row were planted. Three feet of blank row was left between treatments. Treatment designations used were:

1) RHX 500 (Control:talc alone)
2) RHX 505 (Captan alone)
3) RHX 504 (Captan + PGPR)
4) RHX 501 (PGPR alone)

Table 4. The average soil temperature in PGPR field plots - Wiggins, CO, 1982.

Date	Average soil temperature (°C)[1/]
May 11[2/]	14°C
June 9	22°C
June 16	22°C
June 21	29°C
July 1	25°C
July 12	21°C
August 2	26°C
August 18	23°C

[1/] Temperature readings were taken at several sites within the field plot at seedpiece depth.
[2/] Date of planting; May 11, 1982.

tuber borne Rhizoctonia solani sclerotia and the amount of scab (Streptomyces scabies).

Soft-rot potential was determined by wrapping tubers in wet paper towels, saran wrap, and incubating at room temperature for four days (DeBoer, S.H. and A. Kelman. 1975. Amer. Potato J. 52:117-123). The presence of Erwinia in the resulting soft rot pockets was determined by plating samples onto Stewart's MacConkey pectate media. The soft rot potential of the seedlot was calculated according to the formula: Erwinia soft rotting potential = (percentage of tubers assayed developing soft rot pockets) X (average number of soft-rot

pockets per tuber) X (percentage of tubers assayed yielding Erwinia carotovora) ÷ 100 (Harrison and Vogt. 1978. Proc. 17th Ann. Wash. Pot. Conf. pp. 41-47).

The amount of tuber-borne Verticillium was determined by aseptically excising five pieces of vascular tissue from the stem end of 60 tubers (four replications of 15 tubers each) and placing on water agar. The presence of tuber-borne Verticillium was determined by observing with a compound microscope the characteristic conidiophores formed on the surface of tuber pieces.

The estimated percentage of tuber surface area covered by tuber-borne Rhizoctonia sclerotia and surface scab was determined using the Barratt-Horsfall scale (BH scale) of 0-11. Four replications of 15 tubers each were evaluated using this technique.

Additional data was collected from the treatment plots throughout the growing season. Destructive samplings (seedpiece decay readings, root samples, etc.) were always collected from the west row of the 2-row plots. Other (nondestructive) data (vigor, height, etc.) were collected from the east row. This minimized the effect of destructive samplings on neighbor plants. The east row was harvested for total tuber and grade yields.

The percentage and rate of emergence was determined by taking total stand counts on June 9, June 16, and June 21.

The average plant height (cm) for each treatment plot was determined on June 16, July 1, and August 2. On June 16, six plants per treatment plot were measured in both the east and west plot rows. On July 1 and August 2 a total of five plants per treatment plot were measured in the east row, only. Height ratings were determined by measuring from the soil line to the terminal bud for one stem from each randomly selected plant. Measurements for each treatment plot were averaged to give one composite reading per replication.

Plant vigor in each plot was estimated seven times during the growing season using a scale of 1-10. Treatment RHX 500 (nontreated control) was arbitrarily assigned a median value of 5 for each replication. More vigorous treatment plots, based on fullness of foliage, color, uniformity, etc., (i.e., general appearance) for each treatment within a replication were rated at a value between 5 and 10 and less vigorous plants were rated at a value between 1 and 5. Readings were taken on June 9, June 16, July 1, July 12, August 2, August 18, and September 1. One vigor reading per treatment for each replication was taken on each date.

The number of typical blackleg infected (E. carotovora) plants in the east row was determined on August 2 by observation.

On August 20, the amount of Verticillium wilt was determined indirectly by estimating the relative amount defoliation observed in the plots. Three Barratt-Horsfall scale (0-11) ratings were made for each treatment plot. Four stems per treatment plot were collected on this date for direct Verticillium assays. Stems were surface disinfested and a cross section from each the bottom, middle, and top third of each stem was placed onto water agar and incubated at room temperature. Verticillium infection of stems was determined directly by observing the presence of characteristic conidiophores on any tissue surfaces 5-10 days after plating.

Isolations for internal stem populations of E. carotovora and fluorescent PGPR (100 ppm rifampicin resistant) were made from the same surface disinfested stems collected on August 20. A stem section, ca 2.5 cm long, was cut from the stem near the soil line (approximated) and split lengthwise using a scalpel previously dipped in ethanol, flamed and cooled. The stem piece was placed in a test tube containing several milliliters of sterile distilled water, vortexed rapidly for ca 10 sec and allowed to set 5-24 hr at 4°C. An aliquot of supernatant was then streaked onto 1/8 sections of both a crystal-violet pectate and King's

B (100 ppm rifampicin) petri plate and the presence of E. carotovora and PGPR (fluorescent-100 ppm rifampicin resistant) colonies was noted after 24 and 48 hr incubation at 24°C and viewing under an ultraviolet light.

On August 23, assays for the presence of PGPR (fluorescent - 100 ppm rifampicin resistant) and E. carotovora on the surface of leaves and stems was conducted in the field. Leaf surface assays for PGPR were conducted by pressing a leaflet from each the top and bottom of two plants for each treatment plot onto a petri plate containing King's B media (100 ppm rifampicin). Stem surface and additional leaf surface isolations were done by dipping sterile cotton swabs into sterile water blanks and rubbing four leaves and four stems per treatment plot. A separate cotton swab was used for each leaf and stem. The swabs were used to inoculate a 1/8 petri plate section of both crystal-violet pectate and King's B (100 ppm rifampicin) media. The presence of characteristic colonies was noted after 24 and 48 hr incubation at 24°C.

The effect of Captan and PGPR seedpiece treatment on the estimated percentage of seedpiece decay and Rhizoctonia stem cankering was evaluated on July 1 and August 2. All the stems from five hills per treatment per replication were rated on the Barratt-Horsfall scale (0-11) for the percentage of surface area below the soil line cankered by Rhizoctonia. The mother seedpiece for each hill was rated for the amount of decay observed using the same scale. Barratt-Horsfall data were converted to percentage for presentation in the tables.

The effect of seedpiece treatment on the number of stems and tubers per hill and the number of tubers per stem was determined on August 20. Data was collected from five hills per treatment per replication by direct counts.

On June 16, July 12, and August 18 one plant per replication per treatment was collected for root population assays for E. carotovora and PGPR (100 ppm rifampicin resistant) bacteria from the west row of each treatment plot. On

June 16 and July 12 the entire plant was gently lifted using a shovel and the mother seedpiece removed with minimum root contact. Individual plants were gently shaken to remove soil, placed in a new, pre-labelled, plastic bag and immediately placed in a cooler containing insulated ice-packs. A duplicate sample was shipped to Rohm and Haas personnel from the June 16 collection date. Collection on August 18 was in the same manner except plants were cut off at the soil line and the foliage discarded. Individual plants were weighed within 24 hr to determine if treatment affected plant weight. After weighing plants, roots (avoiding stolons) from each plant were snipped into a pile on clean paper towels using scissors previously dipped in ethanol and flamed. Two root subsamples of approximately 1 gram each (estimated to the nearest hundredth gram) were placed into presterilized test tubes containing exactly 10 ml of sterile buffered water (1.25 ml of 0.25 M phosphate buffer per liter of deionized water). These tubes were vortexed at high speed for 50-60 seconds to dislodge bacteria from the roots. Three serial dilutions (1 ml of solution transferred:9 ml sterile buffered water) beyond the original tube were made for each subsample. Each tube was vortexed several times prior to transfer and all transfers were done using a sterile pipette. Bacterial concentration was determined by plating onto sterile media exactly 0.1 ml using an 0.1 ml Eppendorf pipette with sterile plastic tips and spreading inoculum with a cooled glass rod previously dipped in ethanol and flamed. All sample processing and assays were done in the order of RHX 500, RHX 505, RHX 504, and RHX 501. Cross contamination between subsamples was not allowed and all platings were done in the order of the highest to lowest dilution.

The number of E. carotovora were determined by plating serial dilutions onto MacConkey pectate (Stewart's) medium (June 16 collection) or crystal-violet pectate (July 12 and August 18 collections). Each medium is a semi-selective differential medium containing pectin that is enzymatically hydrolyzed by

E. carotovora resulting in a characteristic cup-shaped depression containing a bacterial colony (*E. carotovora*). The number of *E. carotovora* colonies on each dilution plate was determined by counting at 48 hr after incubation at 22°C - 26°C. PGPR (100 ppm rifampicin resistant) bacterial counts were determined by plating dilutions onto King's B medium containing 100 ppm rifampicin. Total PGPR colonies were enumerated after 24 hr and 48 hr of incubation. At least two colony types were observed; one type was nonfluorescent and the other type was strongly fluorescent. Individual counts for the number of nonfluorescent colonies and total number of colonies (fluorescent plus nonfluorescent) were determined at 48 hr by viewing plates under an ultraviolet light.

Bacterial counts were recorded as the number of characteristic colonies present on each plate of medium and the dilution plated. Data was summarized according to the estimated standard plate count procedure (Standard Methods for the Examination of Water and Wastewater. 14th ed. APHA-AWWA-WPCF. 1975) with minor modification. If a dilution subsample series plate had 30 to 300 colonies that plate(s) was used to determine the actual number of colony forming units (CFU) per gram of root for that subsample series. If all plates within a subsample series had less than 30 CFU per plate the two plates with the lowest dilution (most concentrated) and with bacteria present were used to determine the number of CFU per gram of root. Dilution series with no bacterial counts present on the lowest dilution plate were recorded as having less than one colony and CFU per gram of root (standard plate count) were determined on this basis, i.e., CFU per gram of root were recorded on the basis of assay sensitivity. Actual plate counts (as opposed to standard plate counts) were determined as described above with one exception: if plates had no characteristic colonies present, "0" CFU (vs <1 CFU) per gram of root was recorded and averaged

into the data. Therefore, both the estimated "standard plate count" method (estimated CFU present based on assay sensitivity) and the second "actual plate count" method (estimated CFU present solely on the basis of actual counts) are presented in the tables included in this report.

Data (the average number of CFU per gram of root) were determined for each subsample series and these were averaged to give one value for each treatment and replication. Data from each date and for each method of CFU ("standard" vs "actual" counts) per gram of root determination were analyzed in a two way analysis of variance and means separated using Tukey's (HSD) test.

The east row from each plot was harvested on September 29 using a single-row harvester. Tubers were picked by hand into baskets and weighed to determine total weight harvested from 50 feet of row. Two subsamples (each ca 50 lbs) from each treatment plot were placed in individual sterile burlap sacks. One sample was used to determine the proportion of each tuber grade for each treatment.

A 15-tuber sample randomly selected over the length of each treatment plot for each replication at the time of harvest was placed into a sterile paper bag for assays determining the number of Erwinia and PGPR (100 ppm rifampicin resistant) CFU per cm^2 of tuber surface area and the number of internal CFU per gram of peel. Ten tubers from each paper bag were scrubbed with a sterile brush into sterile buffered water (ca 400 ml). The volume of wash was measured exactly and a sample placed in a sterile test tube. Three serial dilutions (1 ml transfer:9 ml of sterile buffered water) beyond the original sample were made. Crystal-violet pectate and King's B with rifampicin (100 ppm) were aseptically plated and CFU determined as described above to estimate the total number of E. carotovora and PGPR in the original wash water. The total tuber surface was determined by (1) estimating the shape of the tuber (sphere vs cylinder), (2) measuring the length of tubers that were considered to be cylinders, and (3) measuring the volume of each tuber by water displacement. Volume

measurements were used to solve for the average tuber radium which, in turn, was needed to calculate the tuber surface area in cm^2 (Appendix 2). Therefore, by knowing the total number of CFU present in the wash water for each treatment and replication and the total surface area washed (10 tubers per treatment-replication) the number (estimated standard plate count and actual counts) of CFU per cm^2 was determined. Measured tuber volume and length and calculated surface areas were analyzed to determine if treatment influenced daughter tuber size.

The internal population (within the peel) of E. carotovora and PGPR was determined by lightly peeling the tubers that had been previously washed (described above) onto clean paper towels. A sample (ca 10 grams) was weighed and placed in a blender jar, previously rinsed with ethanol, containing 100 ml of sterile buffered water. The peels were macerated at high speed for ca 15 seconds and a sample of the supernatant immediately poured into a sterile test tube. Three serial dilutions (1 ml transfer:9 ml sterile buffered water) were prepared and plated as described above. The number of CFU per gram of peel was determined for both count methods as described above. All tuber count data were analyzed in a two way analysis of variance and means separated using Tukey's (HSD) test.

Several low count serial dilution plates from the August 18 root assay series and the tuber surface wash assays were stored at 4°C until mid-December, 1982. Colonies were aseptically removed from the old plates and streaked onto freshly prepared King's B (100 ppm rifampicin) medium. Only isolates that resulted in heavy growth within 24 hr on this medium were used. Nonfluorescent and fluorescent single colony isolates were transferred periodically for purification. Bacterial smears and gram stains were prepared from purified isolates. Selected purified isolates were plated onto nutrient agar for 24 hr at 26°C and assayed for the presence of cytochrome "C" oxidase (oxidase test) and their ability to oxidatively or fermentatively utilize glucose (Hugh and Leifson's O-F test).

Results

The results of the tests for tuber borne diseases in seed used for planting the study plot are listed in Table 5. The seedlot had an E. carotovora soft rot potential rating of "272." This indicated that the tubers possessed an active resident population of E. carotovora and potential loss due to total blackleg disease (seedpiece decay, pre-emergence blackleg and typical blackleg) could be large under the proper environmental conditions. The seed tubers also had ca 5% internal tuber borne Verticillium infection present and 0.3% and 2.1% of the tuber surface covered with R. solani sclerotia and S. scabies lesions, respectively. None of the disease ratings were considered exceptional. It was stated previously (see Materials and Methods) that the soil borne Verticillium dahliae (micro-sclerotial form) inoculum potential was relatively low for Colorado (less than one propagule per 20 mg of soil).

There was no significant effect of seedpiece treatment on emergence rate and stand (Table 6) and plant height (Table 7) ($P > 0.05$). Plant vigor ratings (Table 8) were not significant on or after June 16 ($P > 0.05$) but were highly significant on June 9 ($P \leq 0.01$). On June 9, RHX 505 treatment (Captan alone) was significantly less vigorous ($P \leq 0.01$) than all other treatments. Treatments RHX 500, RHX 504 and RHX 501 were statistically equivalent on this date.

There were no significant differences ($P > 0.05$) between treatment means for the number of typical blackleg infected plants and the estimated percentage of Verticillium wilt observed in the field (Table 9). Statistical differences did not occur for the relative level of Verticillium stem infection when measured by direct plating of tissue onto water agar ($P > 0.05$). The PGPR treatments (RHX 504 and RHX 501) had the lowest proportion of stems infected by Verticillium when compared to the control (RHX 500) and Captan alone (RHX 505) (Table 9, column 3).

Table 5. Characteristics of cv Monona seed potato tubers used for PGPR study - Wiggins, CO, 1982.[1/]

Item	Value
1) *Erwinia carotovora* soft rot potential[2/]	272
2) Percentage of tubers infected with *Verticillium*[3/]	5%
3) Percentage of tuber surface covered with:[3/]	
a) *Rhizoctonia solani* sclerotia[4/]	0.3%
b) *Streptomyces scabies* scab lesions[4/]	2.1%

[1/] See Appendix 1 for inspection sheet.

[2/] *Erwinia* soft rot potential (25 tubers assayed, 17 of which had *E. carotovora* present in rot pockets) = [100% of tubers assayed developed soft rot] X (4.0 soft rot pockets per tuber) X (68% of tubers assayed had *E. carotovora*) ÷ 100 = 272

[3/] Four replications of 15 tubers each were assayed.

[4/] Estimated percentage of surface area covered using the Barratt-Horsfall (0-11) scale.

Table 6. The effect of Captan and PGPR seedpiece treatments on emergence and stand of potato plants from cut Monona seed - Wiggins, CO, 1982.

Treatment	Total stand (per 50 feet of row)[1/]		
	June 9	June 16	June 21
1) RHX 500 (talc alone)	41.4 a	42.2 a	40.9 a
2) RHX 505 (Captan alone)	42.7 a	43.2 a	42.2 a
3) RHX 504 (Captan + PGPR)	42.6 a	42.5 a	41.9 a
4) RHX 501 (PGPR alone)	44.2 a	44.1 a	43.4 a
	NSD[2/]	NSD	NSD

[1/] Seedpieces were planted by hand at one foot spacings on May 11, 1982.

[2/] NSD = no significant difference among treatment means ($P > 0.05$).

Table 7. The effect of Captan and PGPR seedpiece treatments on plant height of cv Monona potatoes - Wiggins, CO, 1982.

Treatment	Average plant height (cm)[1/]		
	June 16[2/]	July 1[3/]	August 2[3/]
1) RHX 500 (talc alone)	11.1 a	20.7 a	51.2 a
2) RHX 505 (Captan alone)	10.6 a	20.8 a	54.3 a
3) RHX 504 (Captan + PGPR)	10.8 a	20.9 a	53.6 a
4) RHX 501 (PGPR alone)	10.8 a	21.9 a	50.2 a
	NSD[4/]	NSD	NSD

[1/] Measured from ground level to terminal bud. Plots were planted on May 11, 1982.

[2/] Six plants per plot were measured.

[3/] Five plants per plot were measured.

[4/] NSD = no significant difference among treatment means ($P > 0.05$).

Table 8. The effect of Captan and PGPR seedpiece treatments on the vigor of emerged potato plants from cut Monona seed - Wiggins, CO, 1982.

Treatment	Vigor ratings on a scale of 1-10[1/]						
	Jun 9	Jun 16	Jul 1	Jul 12	Aug 2	Aug 18	Sep 1
1) RHX 500 (talc alone)	5.0a	5.0a	5.0a	5.0a	5.0a	5.0a	5.0a
2) RHX 505 (Captan alone)	3.7b	4.8a	4.8a	5.2a	5.0a	5.9a	5.7a
3) RHX 504 (Captan + PGPR)	4.7a	5.3a	5.3a	5.3a	4.7a	5.7a	5.2a
4) RHX 501 (PGPR alone)	4.8a	5.0a	5.2a	5.2a	4.8a	5.2a	5.2a
	$P \leq 0.01$ HSD=0.718	NSD	NSD	NSD	NSD	NSD	NSD

[1/] RHX 500 (talc alone) was arbitrarily assigned a vigor rating of 5.0. More vigorous treatments within each replication were given a vigor rating greater than 5 and less vigorous treatments a rating less than 5.

Table 9. The effect of Captan and PGPR seedpiece treatment on the amount of typical blackleg and Verticillium wilt observed in the field - Wiggins, CO, 1982.

Treatment	Number of typical blackleg plants observed on August 21/	Estimated percentage of Verticillium wilt observed on August 202/	Percentage of stems yielding Verticillium on August 203/
1) RHX 500 (talc alone)	0.5 a	10.5 a	12.5 a
2) RHX 505 (Captan alone)	1.0 a	7.5 a	8.3 a
3) RHX 504 (Captan + PGPR)	0.3 a	8.0 a	4.2 a
4) RHX 501 (PGPR alone)	0.5 a	10.0 a	0.0 a
	NSD	NSD	NSD

1/ The east plot row was rated (50 feet long).

2/ Three Barratt-Horsfall scale (0-11) ratings per replication per treatment were made in the east plot row (50 feet long). Verticillium was estimated indirectly by the amount of defoliation observed in the plots.

3/ Four stems were evaluated per treatment per replication. Stem discs (cross-sections) from the top, middle, and bottom of surface disinfected stems were placed onto water agar. Stems positive for Verticillium had at least one stem piece with Verticillium conidiophores present.

Seedpiece treatment with PGPR did not result in detectable PGPR populations on leaf and stem surfaces nor did it influence E. carotovora populations ($P > 0.05$) (Table 10). One fluorescent PGPR colony forming unit was detected when internal stems were assayed. However, this was not significant ($P > 0.05$). E. carotovora was found within 83.3 - 87.5 % of the stems assayed and on the surface of one stem. Treatment effects were not significant ($P > 0.05$).

The seedpiece treatment did not significantly effect ($P > 0.05$) the amount of mother seedpiece decay and Rhizoctonia stem cankering observed in the field (Table 11). Seedpiece treatment with RHX 504 (Captan + PGPR) resulted in the

Table 10. The effect of Captan and PGPR seedpiece treatment on internal stem, external stem and leaf populations of fluorescent PGPR and Erwinia carotovora - Wiggins, CO, 1982.

Treatment	The percentage of plant parts assayed containing populations of:					
	PGPR (fluorescent, 100 ppm rifampicin resistant)			Erwinia carotovora		
	on leaves[1/2/]	on stems[1/3/]	inside stems[4/]	on leaves[1/2/]	on stems[1/3/]	inside stems[4/]
1) RHX 500 (talc alone)	0a	0a	0a	0a	0a	83.3a
2) RHX 505 (Captan alone)	0a	0a	0a	0a	0a	83.3a
3) RHX 504 (Captan + PGPR)	0a	0a	0a	0a	4.2a	87.5a
4) RHX 501 (PGPR alone)	0a	0a	4.2a[5/]	0a	0a	87.5a
	NSD	NSD	NSD	NSD	NSD	NSD

[1/] Assays were conducted in the field on August 23.

[2/] Top and bottom leaves (two leaves each per treatment per replication) were imprinted onto King's B medium (100 ppm rifampicin). E. carotovora assays and additional PGPR assays were done by swabbing leaves and plating onto media.

[3/] Four stems per treatment per replication were assayed by swabbing and direct plating.

[4/] Stems (four per replication per treatment) were collected in the field on August 20. Stems were surface disinfected and a section (ca 2.5 cm) at the soil line was excised, split lengthwise, placed in sterile water, and streaked onto 1/8 of a CVP plate (E. carotovora) or King's B medium containing 100 ppm rifampicin (PGPR).

[5/] One stem in RHX 501 (replication number 6) had one fluorescent rifampicin resistant (100 ppm) colony forming unit present.

Table 11. The effect of Captan and PGPR seedpiece treatment on the estimated percentage of soft-rot observed and mother seedpieces and the amount of Rhizoctonia stem cankering observed in the field (cv Monona) - Wiggins, CO, 1982.

Treatment	Estimated percentage of mother seedpiece decay observed.[1/]		Estimated percentage of Rhizoctonia stem cankering observed[2/]	
	July 1	August 2	July 1	August 2
1) RHX 500 (talc alone)	1.8 a[3/]	86.0 a	1.0 a	1.2 a
2) RHX 505 (Captan alone)	2.5 a	93.5 a	0.6 a	0.8 a
3) RHX 504 (Captan + PGPR)	1.2 a	72.0 a	0.4 a	1.4 a
4) RHX 501 (PGPR alone)	4.0 a	91.0 a	1.8 a	3.0 a
	NSD	NSD	NSD	NSD

[1/] Five seedpieces per replication per treatment were rated on a Barratt-Horsfall (BH) scale (0-11) for the amount of seedpiece decayed.

[2/] Five stems per replication per treatment were rated on a Barratt-Horsfall (BH) scale (0-11) for the amount of stem cankering observed below the soil line.

[3/] Means with the same letters do not differ significantly ($P > 0.05$).
NSD = no significant difference.

least amount of seedpiece decay and treatment with RHX 505 (Captan alone) and RHX 501 (PGPR alone) resulted in the greatest amount of seedpiece decay. There was no treatment effect ($P > 0.05$) on the number of stems and tubers per hill or on the number of tubers per stem (Table 12).

The effect of seedpiece treatment on total plant weight on June 16 and July 12 and on plant weight below the soil line on August 18 is shown in Table 13. Significant differences between treatment did not occur on June 16 and July 12 ($P > 0.05$) but did occur on August 18 ($P \leq 0.05$). The weight of plants below the soil line treated with RHX 505 (Captan alone) was significantly greater than the other treatments ($P \leq 0.05$). RHX 500, RHX 504 and RHX 501 were all statistically equivalent on this date.

The data for the average number of E. carotovora and total PGPR (rifampicin resistant) colony forming units (CFU) per gram or root are shown in Tables 14 and 15, respectively. The number of E. carotovora CFU gram root^{-1} was not influenced by treatment for both the "standard plate count" and "actual plate count" methods ($P > 0.05$). The number of total rifampicin resistant CFU gram root^{-1} for treatment RHX 501 (PGPR alone) was significantly greater than treatments RHX 500 (talc alone), RHX 505 (Captan alone) and RHX 504 (Captan plus PGPR) on June 16 for both plate count methods ($P \leq 0.05$). On July 12 and August 18, significant differences did not occur ($P > 0.05$). The data for the actual plate counts show that rifampicin resistant CFU were never detected on roots taken from RHX 500 and RHX 505 treated plants. Total "actual" PGPR counts were consistently higher (26 to 956 times greater) for RHX 501 (PGPR alone) than for RHX 504 (Captan plus PGPR) throughout the growing season.

Data for the number of nonfluorescent PGPR (rifampicin resistant) (as opposed to "total PGPR" which included both fluorescent and nonfluorescent

Table 12. The effect of Captan and PGPR seedpiece treatment on the average number of cv Monona potato stems per hill, tubers per hill and tubers per stem - Wiggins, CO, 1982.

Treatment	Items evaluated on August 20[1/]		
	stems hill^{-1}	tubers hill^{-1}	tubers stem^{-1}
1) RHX 500 (talc alone)	2.1a	16.4a	7.9a
2) RHX 505 (Captan alone)	2.1a	18.6a	8.3a
3) RHX 504 (Captan + PGPR)	2.1a	16.0a	7.6a
4) RHX 501 (PGPR alone)	2.4a	19.0a	8.2a
	NSD	NSD	NSD

[1/] Data was collected from five hills per treatment per replication. Planting date was May 11, 1982.

Table 13. The effect of Captan and PGPR seedpiece treatments on plant weight (cv Monona) - Wiggins, CO, 1982.

Treatment	Average plant weight (grams) and date determined		
	June 16[1/]	July 12[1/]	August 18[2/]
1) RHX 500 (talc alone)	87.53 a[3/]	210.97 a	86.75 b
2) RHX 505 (Captan alone)	70.80 a	146.50 a	131.33 a
3) RHX 504 (Captan + PGPR)	56.08 a	210.28 a	94.47 b
4) RHX 501 (PGPR alone)	56.07 a	209.28 a	90.53 b
	NSD	NSD	$P \leq 0.05$ HSD = 36.42

[1/] One plant per replication per treatment was collected for root population assays. The entire plant (excluding seedpiece) was weighed prior to root assays.

[2/] Plants (one plant per replication per treatment) were broken off at the soil line and collected for root population assays. The entire plant below soil line (excluding seedpiece and daughter tubers) was weighed prior to root assays.

[3/] Column means with different letters differ significantly (Tukey's HSD test). NSD = no significant difference ($P > 0.05$).

Table 14. The effect of Captan and PGPR seedpiece treatment on the number of Erwinia carotovora cells present on plant roots during the growing season (cv Monona) - Wiggins, CO, 1982.[1/]

Treatment	The average number of E. carotovora colony forming units per gram of potato plant root based on:					
	Standard plate count[2/]			Actual plate count		
	June 16	July 12	August 18	June 16	July 12	August 18
1) RHX 500 (talc alone)	3.5×10^{2}a[4/]	8.80×10^{4}a	1.85×10^{4}a	2.92×10^{2}a	8.80×10^{4}a	1.84×10^{4}a
2) RHX 505 (Captan alone)	1.10×10^{2}a	1.55×10^{4}a	2.23×10^{3}a	2.56×10^{1}a	1.55×10^{4}a	2.20×10^{3}a
3) RHX 504 (Captan + PGPR)	5.53×10^{2}a	1.20×10^{4}a	6.97×10^{5}a	4.81×10^{2}a	1.20×10^{4}a	6.97×10^{5}a
4) RHX 501 (PGPR alone)	3.08×10^{2}a	1.29×10^{4}a	1.45×10^{5}a	4.82×10^{1}a	1.29×10^{4}a	1.45×10^{5}a
	NSD	NSD	NSD	NSD	NSD	NSD

[1/] Potato roots (1 plant per treatment plot and 2 subsamples per plant) were washed in sterile buffered water and serial dilutions prepared. Dilutions were plated onto MacConkey (Stewart's) medium on June 16 and onto crystal-violet pectate on July 12 and August 18.

[2/] Bacterial counts for dilution series plates with no E. carotovora colonies were based on the assay sensitivity (see text).

[3/] Bacterial counts for dilution series plates with no E. carotovora colonies were recorded as having "0" CFU per gram of root, i.e., actual counts were used to determine CFU per gram of root.

[4/] Column means with the same letters do not differ significantly ($P \geq 0.05$). NSD = no significant difference.

Table 15. The effect of Captan and PGPR seedpiece treatment on the number of total PGPR (rifampicin resistant) cells present on plant roots during the growing season (cv Monona) - Wiggins, CO, 1982.[1/]

Treatment	The average number of total PGPR (rifampicin resistant) colony forming units per gram of potato plant root based on:					
	Standard plate count[2/]			Actual plate count[3/]		
	June 16	July 12	August 18	June 16	July 12	August 18
1) RHX 500 (talc alone)	1.15×10^2 b[4/]	1.26×10^2 a	1.17×10^2 a	0 b	0 a	0 a
2) RHX 505 (Captan alone)	9.34×10^1 b	1.24×10^2 a	1.15×10^2 a	0 b	0 a	0 a
3) RHX 504 (Captan + PGPR)	4.45×10^2 b	2.25×10^2 a	1.49×10^3 a	4.37×10^2 b	1.14×10^2 a	9.13×10^2 a
4) RHX 501 (PGPR alone)	1.14×10^4 a	1.09×10^5 a	4.42×10^4 a	1.14×10^4 a	1.09×10^5 a	4.42×10^4 a
	$P \leq 0.05$ HSD = 0.07×10^4	NSD	NSD	$P \leq 0.05$ HSD = 1.07×10^4	NSD	NSD

[1/] Potato roots (1 plant per treatment plot and 2 subsamples per plant) were washed in sterile buffered water and serial dilutions prepared. Dilutions were plated onto King's B medium to which 100 ppm rifampicin had been added.

[2/] Bacterial counts for dilution series plates with no rifampicin resistant colonies were based on assay sensitivity (see text).

[3/] Bacterial counts for dilution series plates with no rifampicin resistant colonies were recorded as having "0" CFU per gram of root, i.e., actual counts were used to determine CFU per gram of root.

[4/] Column means with the same letters do not differ significantly ($P > 0.05$). NSD = no significant difference.

CFU counts) are shown in Table 16. Treatment with RHX 501 (PGPR alone) resulted in significantly greater CFU per gram of root on June 16 than for all other treatments ($P \leq 0.05$) for both standard and actual plate count methods. Counts on July 12 and August 18 were not significant ($P > 0.05$). Nonfluorescent "actual" PGPR counts were consistently higher (52 to 183 times greater) for RHX 501 (PGPR alone) than for RHX 504 (Captan + PGPR) throughout the growing season. The nonfluorescent PGPR counts paralleled the total PGPR counts during the growing season. "Actual" plate count data demonstrated that nonfluorescent rifampicin resistant (i.e., nonfluorescent PGPR) were never detected on roots taken from RHX 500 (talc alone) and RHX 505 (Captan alone) treated plants.

Data for the average number of E. carotovora and total PGPR (rifampicin resistant) CFU present on the surface of daughter tubers are shown in Tables 17 and 18, respectively. Significant differences between treatment means did not occur in either Table 17 or Table 18 ($P > 0.05$). The data show that the number of E. carotovora CFU cm^{-2} ranged from 2.20×10^2 up to 2.88×10^3 (actual plate counts). The data also show that PGPR were readily isolated from daughter tuber surfaces at the time of harvest. "Actual count" data show that rifampicin resistant CFU were not detected on daughter tubers when the mother seedpiece was treated with RHX 500 (talc alone) or RHX 505 (Captan alone).

Data in Table 19 show that nonfluorescent PGPR (rifampicin resistant) were also isolated from daughter tuber surfaces at harvest. Although significant differences between treatment means did not occur ($P > 0.05$), actual plate count data show that nonfluorescent PGPR were never isolated from RHX 500 and RHX 505 treatments.

The data for the average number of E. carotovora and total PGPR (rifampicin resistant) CFU $gram^{-1}$ of tuber peel (after washing) are shown in Tables 20 and 21, respectively. Significant differences did not occur between treatment means for any treatments in either table ($P > 0.05$). Actual count data show that PGPR

Table 16. The effect of Captan and PGPR seedpiece treatment on the number of nonfluorescent PGPR (rifampicin resistant) cells present on plant roots during the growing season (cv Monona) - Wiggins, CO, 1982[1/].

Treatment	The average number of nonfluorescent PGPR (rifampicin resistant) colony forming units per gram of potato plant root based on:					
	Standard plate count[2/]			Actual plate count[3/]		
	June 16	July 12	August 18	June 16	July 12	August 18
1) RHX 500 (talc alone)	1.15×10^2 b[4/]	1.26×10^2 a	1.17×10^2 a	0 b	0 a	0 a
2) RHX 505 (Captan alone)	9.34×10^1 b	1.24×10^2 a	1.15×10^2 a	0 b	0 a	0 a
3) RHX 504 (Captan + PGPR)	1.18×10^2 b	1.26×10^3 a	1.60×10^2 a	4.59×10^1 b	1.14×10^1 a	3.98×10^1 a
4) RHX 501 (PGPR alone)	2.40×10^3 a	2.12×10^3 a	6.75×10^3 a	2.39×10^3 a	2.09×10^3 a	6.75×10^3 a
	$P \leq 0.01$ HSD = 1.55×10^3	NSD	NSD	$P \leq 0.01$ HSD = 1.56×10^3	NSD	NSD

1/ Potato roots (one plant per treatment plot and two subsamples per plant) were washed in sterile buffered water and serial dilutions prepared. Dilutions were plated onto King's B medium to which 100 ppm rifampicin had been added. Fluorescence was determined after 48 hr by viewing spread plates while exposing to ultraviolet light.

2/ Bacterial counts for dilution series plates with no rifampicin resistant colonies were based on assay sensitivity (see text).

3/ Bacterial counts for dilution series plates with no rifampicin resistant colonies were recorded as having "0" CFU per gram of root, i.e., actual counts were used to determine CFU per gram of root.

4/ Column means with the same letters do not siffer significantly ($P > 0.05$). NSD = no significant difference.

Table 17. The effect of Captan and PGPR seedpiece treatment on the number of Erwinia carotovora cells present on daughter tuber surfaces at harvest (cv Monona) - Wiggins, CO, 1982.

Treatment	The average number of E. carotovora colony forming units per cm^2 of daughter tuber surface area based on:[1/]	
	Standard plate counts[2/]	Actual plate counts[3/]
1) RHX 500 (talc alone)	6.93×10^2 a[4/]	6.93×10^2 a
2) RHX 505 (Captan alone)	1.02×10^3 a	1.02×10^3 a
3) RHX 504 (Captan + PGPR)	2.88×10^3 a	2.88×10^3 a
4) RHX 501 (PGPR alone)	2.27×10^2 a	2.20×10^2 a
	NSD	NSD

[1/] Data is from 10 tubers per replication. Tuber surfaces were washed into sterile buffered water. A serial dilution series was plated onto crystal-violet pectate.

[2/] Bacterial counts for dilution series plates with no E. carotovora colonies were based on the assay sensitivity (see text).

[3/] Bacterial counts for dilution series plates with no E. carotovora colonies were recorded as having "0" CFU per cm^2 of tuber surface area, i.e., actual counts were used to determine CFU per cm^2 of tuber surface area.

[4/] Column means with the same letters do not differ significantly ($P > 0.05$). NSD = no significant difference.

Table 18. The effect of Captan and PGPR seedpiece treatment on the number of total PGPR (rifampicin resistant) cells present on daughter tuber surfaces at harvest (cv Monona) - Wiggins, CO, 1982.

Treatment	The average number of rifampicin resistant colony forming units per cm^2 of daughter tuber surface area based on:	
	Standard plate counts[2/]	Actual plate counts[3/]
1) RHX 500 (talc alone)	2.01×10^1 a[4/]	0 a
2) RHX 505 (Captan alone)	2.23×10^1 a	0 a
3) RHX 504 (Captan + PGPR)	6.22×10^1 a	5.39×10^1 a
4) RHX 501 (PGPR alone)	2.21×10^3 a	2.21×10^3 a
	NSD	NSD

[1/] Data is from 10 tubers per replication. Tuber surfaces were washed into sterile buffered water. A serial dilution series was plated onto King's B medium containing 100 ppm rifampicin.

[2/] Bacterial counts for dilution series plates with no rifampicin resistant colonies were based on assay sensitivity (see text).

[3/] Bacterial counts for dilution series plates with no rifampicin resistant colonies were recorded as having "0" CFU per cm^2 of tuber surface area, i.e., actual counts were used to determine CFU per cm^2 of tuber surface area.

[4/] Column means with the same letters do not differ significantly ($P > 0.05$). NSD = no significant difference.

Table 19. The effect of Captan and PGPR seedpiece treatment on the number of nonfluorescent PGPR (rifampicin resistant) cells present on daughter tuber surfaces at harvest (cv Monona) - Wiggins, CO, 1982.

Treatment	The average number of nonfluorescent (rifampicin resistant colony forming units per cm^2 of daughter tuber surface area based on:	
	Standard plate counts[2/]	Actual plate counts[3/]
1) RHX 500 (talc alone)	2.01×10^1 a[4/]	0 a
2) RHX 505 (Captan alone)	2.26×10^1 a	0 a
3) RHX 504 (Captan + PGPR)	2.43×10^1 a	7.58×10^0 a
4) RHX 501 (PGPR alone)	1.38×10^2 a	1.31×10^2 a
	NSD	NSD

[1/] Data is from 10 tubers per replication. Tuber surfaces were washed into sterile buffered water. A serial dilution series was plated onto King's B medium containing 100 ppm rifampicin.

[2/] Bacterial counts for dilution series plates with no rifampicin resistant colonies were based on assay sensitivity (see text).

[3/] Bacterial counts for dilution series plates with no rifampicin resistant colonies were recorded as having "0" CFU per cm^2 of tuber surface area, i.e., actual counts were used to determine CFU per cm^2 of tuber surface area.

[4/] Column means with the same letters do not differ significantly ($P > 0.05$). NSD = no significant difference.

Table 20. The effect of Captan and PGPR seedpiece treatment on the number of Erwinia carotovora cells present within the peel of daughter tubers at harvest (cv Monona) - Wiggins, CO, 1982.

Treatment	The average number of E. carotovora colony forming units per gram of daughter tuber peel based on:[1/]	
	Standard plate counts[2/]	Actual plate counts[3/]
1) RHX 500 (talc alone)	$\leq 9.92 \times 10^1$ a[4/]	0 a
2) RHX 505 (Captan alone)	$\leq 1.32 \times 10^2$ a	6.60×10^1 a
3) RHX 504 (Captan + PGPR)	$\leq 1.48 \times 10^2$ a	8.22×10^1 a
4) RHX 501 (PGPR alone)	$\leq 9.94 \times 10^1$ a	1.65×10^1 a
	NSD	NSD

[1/] A total of 10 grams of tuber peel, taken from 10 tubers per replication previously washed for surface bacteria, was macerated in 100 ml of sterile buffered water. Serial dilutions were plated onto crystal-violet pectate to determine the number of colony forming units present.

[2/] Bacterial counts for dilution series plates with no E. carotovora colonies were based on the assay sensitivity (see text).

[3/] Bacterial counts for dilution series plates with no E. carotovora colonies were recorded as having "0" CFU per gram of tuber peel, i.e., actual counts were used to determine CFU per gram of tuber peel.

[4/] Column means with the same letters do not differ significantly ($P > 0.05$). NSD = no significant difference.

Table 21. The effect of Captan and PGPR seedpiece treatment on the number of total PGPR (rifampicin resistant) cells present within the peel of daughter tubers at harvest (cv Monona) - Wiggins, CO, 1982.

Treatment	The average number of PGPR (rifampicin resistant) colony forming units per gram of daughter tuber peel based on:[1/]	
	Standard plate counts[2/]	Actual plate counts[3/]
1) RHX 500 (talc alone)	$\leq 9.92 \times 10^1$ a[4/]	0 a
2) RHX 505 (Captan alone)	$\leq 9.91 \times 10^1$ a	0 a
3) RHX 504 (Captan + PGPR)	$\leq 9.90 \times 10^1$ a	0 a
4) RHX 501 (PGPR alone)	$\leq 9.94 \times 10^1$ a	3.31×10^1 a
	NSD	NSD

[1/] A total of 10 grams of tuber peel, taken from 10 tubers per replication previously washed for surface bacteria, were macerated in 100 ml of sterile buffered water. Serial dilutions were plated onto King's B (100 ppm rifampicin) to determine the number of colony forming units present.

[2/] Bacterial counts for dilution series plates with no rifampicin resistant colonies were based on assay sensitivity (see text).

[3/] Bacterial counts for dilution series plates with no rifampicin resistant colonies were recorded as having "0" CFU per gram of tuber peel, i.e., actual counts were used to determine CFU per gram of tuber peel.

[4/] Column means with the same letters do not differ significantly ($P \geq 0.05$). NSD = no significant difference.

(rifampicin resistant) CFU were never detected when tubers were treated with RHX 500 (talc alone), RHX 505 (Captan alone), and RHX 504 (Captan + PGPR). When mother seedpieces were treated with RHX 501 (PGPR alone) daughter tubers had 33.1 PGPR CFU present per gram of peel after washing. Actual plate count data shown in Table 22 show that nonfluorescent PGPR were never isolated from daughter tubers after washing.

Data in Table 23 show that seedpiece treatment did not affect daughter tuber volume, surface area, or tuber length. Data in Table 24 show that total yield and tuber grade were not influenced by treatment ($P > 0.05$).

Characteristics of PGPR strains re-isolated from stored King's B media (100 ppm rifampicin) are shown in Table 25. PGPR-streaked plates always resulted in visible colonies within 12 hr on King's B medium (100 ppm rifampicin). Stock cultures of Pseudomonas spp. and a water extract of non-sterile soil plated onto the same medium did not result in visible colonies after 48 hr.

Fluorescent pigment production was rated visually at 24 hr (Table 25). Results show that three levels of fluorescent pigment production were represented by the PGPR (100 ppm rifampicin resistant) re-isolates tested. The gram stain, oxidase and O-F tests are characteristic for Pseudomonas spp. The production of fluorescent pigment is not a prerequisite for all Pseudomonas spp.

Discussion

There was no effect of PGPR treatment on plant emergence, stand, height, vigor, typical blackleg and Verticillium wilt observed in the field, internal stem, external stem, and external leaf populations of fluorescent PGPR and E. carotovora, mother seedpiece decay, Rhizoctonia stem cankering, the number of potato stems and tubers per hill, the number of tubers per stem,

Table 22. The effect of Captan and PGPR seedpiece treatment on the number of fluorescent PGPR (rifampicin resistant) cells present within the peel of daughter tubers at harvest (cv Monona) - Wiggins, CO, 1982.

Treatment	The average number of nonfluorescent PGPR (rifampicin resistant) colony forming units per gram of daughter tuber peel based on:[1]	
	Standard plate counts[2]	Actual plate counts[3]
1) RHX 500 (talc alone)	9.92×10^1 a[4]	0 a
2) RHX 505 (Captan alone)	9.91×10^1 a	0 a
3) RHX 504 (Captan + PGPR)	9.90×10^1 a	0 a
4) RHX 501 (PGPR alone)	9.94×10^1 a	0 a
	NSD	NSD

[1] A total of 10 grams of tuber peel, taken from 10 tubers per replication previously washed for surface bacteria, were macerated in 100 ml of sterile buffered water. Serial dilutions were plated onto King's B (100 ppm rifampicin) to determine the number of colony forming units present.

[2] Bacterial counts for dilution series plates with no rifampicin resistant colonies were based on assay sensitivity (see text).

[3] Bacterial counts for dilution series plates with no rifampicin resistant colonies were recorded as having "0" CFU per gram of tuber peel, i.e., actual counts were used to determine CFU per gram of tuber peel.

[4] Column means with the same letters do not differ significantly ($P > 0.05$). NSD = no significant difference.

Table 23. The effect of Captan and PGPR seedpiece treatment on daughter tuber size (cv Monona) - Wiggins, CO, 1982.

Treatment	Tuber dimension analyzed[1]		
	Tuber volume ($\bar{X}$)[2]	Tuber surface area ($\bar{X}$)[3]	Tuber length ($\bar{X}$)[4]
1) RHX 500 (talc alone)	270 a	226.1 a	10.3 a
2) RHX 505 (Captan alone)	260 a	222.3 a	10.2 a
3) RHX 504 (Captan + PGPR)	262 a	222.7 a	10.2 a
4) RHX 501 (PGPR alone)	270 a	224.4 a	10.4 a
	NSD[5]	NSD	NSD

[1] Treatment plots were planted on May 11, 1982 and harvested on September 29. Ten tubers per treatment per replication were assayed. Data means are on a "per tuber" basis.

[2] Measured by water displacement (mls).

[3] Calculated (cm^2) by solving for radius using volume data (spheres) and by using volume and length measurement (cylinders). See Appendix 2.

[4] All tubers were measured (cm) at what was estimated to be their longest length.

[5] NSD = no significant difference ($P > 0.05$).

Table 24. The effect of Captan and PGPR seedpiece treatment on cv Monona daughter tuber yield and grade - Wiggins, CO, 1982.

Treatment	Yield of tuber grades in cwt per acre[1/]						
		US #1					
	Total	Total	>10 oz	<10 oz	US #2	"B" grade	Culls
1) RHX 500 (talc alone)	425 a[2/]	358 a	171 a	187 a	22 a	30 a	5 a
2) RHX 505 (Captan alone)	417 a	362 a	167 a	195 a	21 a	25 a	3 a
3) RHX 504 (Captan + PGPR)	444 a	372 a	194 a	178 a	27 a	30 a	3 a
4) RHX 501 (PGPR alone)	430 a	380 a	192 a	188 a	18 a	23 a	1 a
	NSD	NSD	NSD	NSD	NSD	NSD	NSD

[1/] Single rows 50 ft long (3.25×10^{-3} acre) were harvested and all tubers weighed to determine total yield. A ca 50 lb sample was graded to determine the proportion of each grade within the sample.

[2/] Means in columns with the same letters do not differ significantly ($P > 0.05$). NSD = no significant differences observed.

Table 25. Characteristics of PGPR (rifampicin resistant) isolates subcultured from stored CFU count plates, Rohm and Haas Company, 1982.

Isolate number and source[1]	Number of single colony transfers prior to subsequent tests.[2]	Intensity of fluor-escence	Growth evident after 12 hr on King's B (100 ppm rifampicin)	Gram stain[4]	Oxidase test	O-F test[5]
1) RHX 504. Potato roots	3	+1	+	-	NT[6]	NT
2) RHX 504. Potato roots	5	+3	+	-	+	0
5) RHX 504. Potato roots	5	+1	+	-	+	0
10) RHX 501. Potato roots	3	+3	+	-	NT	NT
20) RHX 501. Daughter tuber surfaces	4	0	+	-	+	0
21) RHX 501. Daughter tuber surfaces	4	0	+	-	+	0
22) RHX 501. Daughter tuber surfaces	4	0	+	-	+	0
23) RHX 501. Daughter tuber surfaces	2	+3	+	-	NT	NT
24) RHX 501. Daughter tuber surfaces	2	+3	+	-	NT	NT
A) Pseudomonas. Stock culture	1	+3	-	-	+	0
B) Pseudomonas. Stock culture	1	+3	-	-	+	0

[1] Two replications of each isolate were made by transferring two colonies of each isolate after the initial re-isolation from stored plates. Observations were identical for each replication.

[2] Transfers were necessary to insure isolate purity.

[3] Fluorescence was rated visually (scaled 0 to +3) after 24 hr growth of purified isolates on King's B (100 ppm rifampicin) medium. Plates were viewed using an ultraviolet light source. Isolates rated as "0" had no fluorescent pigments evident at 24 hr and 48 hr.

(continued next page)

(footnotes continued)

4/ Cell morphology appeared to be typical for Pseudomonas spp.

5/ O = oxidative metabolism of glucose.
F = fermentative metabolism of glucose (Hugh and Leifson's test).

6/ NT = not tested.

plant weight, the number of E. carotovora CFU per gram of root, the number of E. carotovora CFU, total and nonfluorescent PGPR CFU present on daughter tuber surfaces and within the peel at harvest and, lastly, no effect on tuber yield or grade.

Data showed that PGPR readily colonized potato root surfaces under growing conditions in northeastern Colorado. Additionally, PGPR were present on daughter tuber surfaces at the time of harvest. Mixing Captan with PGPR reduced colonization by a factor of ca 100 X. There was no evidence that PGPR colonized stems internally.

Careful inspection of the data indicate where weaknesses might have occurred.

Delayed emergence is correlated with low initial vigor ratings. The fact that RHX 505 (Captan alone) resulted in low vigor ratings (Table 8) indicated that emergence was probably delayed. This is often observed in Colorado when potato seedpieces are treated with Captan. The fact that stand counts were not significant for Captan (Table 6) implies that subtle differences in emergence rates may have occurred for other treatments. However, since early vigor ratings for RHX 504 and RHX 501 are lower than RHX 500 this also implies that emergence, if influenced, was not earlier than for RHX 500, the nontreated control.

Inspection of bacterial CFU count data indicated that large variations in CFU occurred between replications. This made it difficult to demonstrate significant differences among treatment means. This is especially evident for total and nonfluorescent PGPR CFU count data. Data should be considered with this in mind, i.e., the statistical analysis may be very conservative.

Treatment with RHX 501 (PGPR alone) resulted in significantly more total PGPR (nonfluorescent + fluorescent) and nonfluorescent CFU per gram of root on June 16 (Tables 15 and 16, respectively). This demonstrated that at least two

PGPR "biotypes" or strains were isolated and they resulted from the initial PGPR treatment. This is especially evident when actual plate count data are considered since rifampicin resistant isolates were never observed in assays using material collected from non-PGPR treated plants. Data in Table 25 show that three PGPR strains, based on the intensity of fluorescence, were actually present on assayed material. This is consistent with observations made during CFU assays although only the distinction between nonfluorescent and fluorescent PGPR was made. Testing of selected isolates with the gram stain, the oxidase test and O-F test did not allow further distinction among the PGPR strains and was consistent with results expected for typical Pseudomonas isolates.

Pseudomonas spp. and, therefore PGPR, are able to conjugate and transfer genetic material [such as the gene(s) for rifampicin resistance] between strains. Since Pseudomonas spp. are common soil inhabitants and nonfluorescent Pseudomonas spp. occur (Bergey's Manual of Determinative Bacteriology) it is not unreasonable to expect that natural populations could acquire the gene(s) for rifampicin resistance from the PGPR. This would, out of necessity, be correlated with PGPR seedpiece treatment and could possibly explain the presence of PGPR strains (isolation based solely on rifampicin resistance) on assayed plant material. The gene(s) for rifampicin resistance would not have to be located on a plasmid although this would increase the probability of intercell transfer during conjugation.

If formulated PGPR were contaminated with non-PGPR or strains developed during PGPR formulation, this is a possible explanation for the lack of treatment response in this study. The original PGPR cultures and subsequent formulation as well as PGPR strains isolated from treated plants needs to be tested to aid in pinpointing their source and effect on potato plants after seedpiece inoculation.

Appendix 1.

U.S. DEPARTMENT OF AGRICULTURE
FOOD SAFETY AND QUALITY SERVICE

A - 199218

FEDERAL-STATE INSPECTION CERTIFICATE

This certificate is issued in compliance with the regulations of the Secretary of Agriculture governing the inspection of various products pursuant to the Agricultural Marketing Act of 1946, as amended (7 U.S.C. 1621 et seq.) and the applicable statutes of the State, and is admissible as prima facie evidence in all courts of the United States and State courts where provided by law. WARNING: Any person who knowingly shall falsely make, issue, alter, forge, or counterfeit this certificate, or participate in any of such actions, is subject to a fine of not more than $1,000 or imprisonment for not more than one year, or both.

INSPECTION POINT	CARRIER	INITIAL AND NO.
EAST GRAND FORKS, MINNESOTA	MECH ☐ CAR ☐ TRUCK ☒ TRAILER	S.D. 22578

INSPECTION BEGUN		INSPECTION COMPLETED		CONDITION OF CARRIER
HOUR	DATE	HOUR	DATE	
4:10 P.M.	MARCH 15, 1982	5:30 pm	MARCH 15, 1982	TEMP CONTROL UNIT OFF

APPLICANT NAME AND ADDRESS	☒ SHIPPER ☐ RECEIVER NAME AND ADDRESS
JEFFREY POTATO CO. EAST GRAND FORKS, MINNESOTA	SAME

PRODUCT	LOADER'S COUNT AND TYPE OF CONTAINER	CONTAINER MARKINGS	SIZE	GRADE
ROUND WHITE POTATOES	APPLICANT STATES APPROX 430 CWT	BULK	GENERALLY 1½ INCHES TO 12 OUNCES MOSTLY 2 INCHES TO 3 INCHES IN DIAMETER OFFSIZE WITHIN TOLERANCE.	MINNESOTA BLUE TAG CERTIFIED SEED GRADE 1½ INCHES MINIMUM

QUALITY AND CONDITION: MATURE, FIRM, GENERALLY WELL SHAPED, SLIGHTLY DIRTY. GRADE DEFECTS WITHIN TOLERANCE.
Less than ½ of 1% Soft Rot

REMARKS: INSPECTED DURING PROCESS OF LOADING
APPLICANT STATES VARIETY TO BE MONONA, APPROVED FOR RECERTIFICATION.

Total cwt. 430 @ .06	
Fee $ 25.80	I, the undersigned, a duly authorized inspector of the United States Department of Agriculture, do hereby certify that at the request of the applicant and on the date indicated, samples of the above described products were inspected and the quality and/or condition as shown by said samples were as herein stated.
Expense $	
Overtime 3 25	Michael Horken (INSPECTOR)
Total 25.80	29 C.S.

FVQ FORM 184-2 REPLACES FV FORM 184-2 (6-74) WHICH MAY BE USED

Appendix 2. Procedure for Tuber Surface Area Calculations

1) Determine if tuber shape most closely resembles (I) a sphere (go to 2) or (II) a cylinder (go to 3).

2)

A) Measure tuber volume (v) by water displacement and use volume measurement to solve for the tuber radius (r) in the equation:

$$V = \frac{4}{3} \pi r^3$$

B) Substitute "r" into surface area equation:

$$\text{tuber surface area (SA)} = 4 \pi r^2$$

3)

A) Measure tuber volume (v) by water displacement and tuber length (L). Use measurements to solve for tuber radius (r) in the equation:

$$V = \pi r^2 L$$

B) Substitute "r" and "L" into surface area equation;

$$\text{tuber surface area (SA)} = 2 \pi r L + 2 \pi r^2$$

The Effect of Plant Growth-Promoting Rhizobacteria (PGPR) Seedpiece Treatment on *Erwinia* Recontamination of cv Centennial Micropropagation Tubers Grown in the San Luis Valley

G.D. Franc, M.D. Harrison and J.W. Kloepper

Abstract

Studies on the effect of PGPR on growth response and on recontamination of blackleg-free seed were initiated in 1982 and will continue in 1983.

There was no obvious effect of PGPR seedpiece treatment on plant growth and yield although plant roots were readily colonized by PGPR under growing conditions in the San Luis Valley.

Data from potato root assays showed that low levels of *Erwinia* recontamination have occurred during the first growing season for plants treated with carrier alone but not for plants treated with PGPR. These preliminary results are encouraging and studies will be continued during the 1980 growing season.

Materials and Methods

Intact seed tubers were inoculated with either carrier alone (nontreated control) or PGPR. Seed tubers (cv Centennial) were generated from micropropagation plants and were *Erwinia* (blackleg)-free. Treated tubers were planted in a single row plot of 10 replications and eight tubers per replication on May 26, 1982 at the S.L.V. Research Center.

Data for potato plant growth response and yield were collected. Assays for root populations of *Erwinia* and PGPR were conducted as described in the immediately preceding section of this report entitled "The Effect of Plant Growth Promoting Rhizobacteria Seedpiece Treatment on cv Monona Potatoes in Northeastern Colorado."

Results

Data showed that there was no obvious effect of PGPR seedpiece treatment on emergence rates and stand (Table 1), plant height (Table 2), plant vigor (Table 3) and tuber yield (Table 4).

Data in Table 5 and Table 6 show that potato roots were readily colonized by PGPR by June 28 and populations were easily detected on August 17. Actual count data for Erwinia (Table 6) show that low numbers of Erwinia were detected on potato roots on August 17 in nontreated control plots. Erwinia was not detected in PGPR treated plots on June 28 or August 17 and in nontreated control plots on June 28.

Discussion

Results for 1983 are encouraging. Although effects of PGPR seedpiece treatment on plant growth were not obvious, no Erwinia recontamination occurred. Nontreated seedpieces had a low number of Erwinia detected on potato roots on August 17. Irrigation water used in the study had "naturally occurring" Erwinia populations present. This suggested that the source of Erwinia was the irrigation water and that PGPR may be of use in limiting recontamination in the field. This study will be continued in 1983.

Table 1. The effect of plant growth promoting rhizobacteria (PGPR) on potato emergence and stand in the San Luis Valley - Center, CO, 1982.

Treatment[1/]	June 19	June 23	June 28	July 8	July 19
1) Carrier (nontreated control)	1.3	4.8	6.8	7.0	7.0
2) PGPR	1.0	4.4	6.8	7.0	7.0
	NSD	NSD	NSD	NSD	NSD

1/ Planting date: May 26, 1982.

Table 2. The effect of plant growth promoting rhizobacteria (PGPR) on potato plant height in the San Luis Valley - Center, CO, 1982.

Treatment	July 8[1/]	July 19[2/]	July 27[2/]	Aug 6[2/]	Aug 17[2/]
1) Carrier (nontreated control)	8.2	15.2	23.9	31.5	31.2
2) PGPR	8.5	14.4	21.0	30.5	30.5
	NSD	NSD	NSD	NSD	NSD

1/ Four plants rated per plot.

2/ Seven plants rated per plot.

Table 3. The effect of plant growth promoting rhizobacteria (PGPR) on potato plant vigor in the San Luis Valley - Center, CO, 1982.

Treatment	July 19	July 27	Aug 6	Aug 17	Aug 26	Sept 7
1) Carrier (nontreated control)	5.0	5.0	5.0	5.0	5.0	5.0
2) PGPR	4.3	4.5	4.8	4.7	5.0	5.1
	NSD	NSD	NSD	NSD	NSD	NSD

Table 4. The effect of plant growth promoting rhizobacteria (PGPR) on total potato-tuber yield in the San Luis Valley - Center, CO, 1982.

Treatment	Average total tuber yield in kilograms[1/]
1) Carrier (nontreated control)	4.7 kg
2) PGPR	4.5 kg
	NSD

[1/] The average total tuber yield per replication for 10 replications of seven hills each.

Table 5. The effect of PGPR seedpiece treatment on E. carotovora and PGPR potato root populations on June 28 - Center, CO, 1982.[1/]

Seedpiece treatment	Average number of colony forming units (CFU) per gram of root assayed on:	
	Crystal Violet Pectate[2/]	King's B + rifampicin (100 ppm)[3/]
1) Carrier (nontreated control)	0	0
2) PGPR	0	1.22×10^5

[1/] Planting date - May 26, 1982. Assays conducted by A.G.S., Inc. and potato research personnel.
[2/] E. carotovora populations.
[3/] PGPR populations.

Table 6. The effect of PGPR seedpiece treatment on E. carotovora and PGPR potato root populations on August 17 - Center, CO, 1982.[1/]

Treatment	Average number of colony forming units (CFU) per gram of root assayed on:			
	Crystal Violet Pectate[2/]		King's B + rifampicin (100 ppm)[3/]	
	Standard	Actual	Standard	Actual
1) Carrier (nontreated control)	$<1.74 \times 10^2$	5.01×10^1	2.92×10^2	1.77×10^2
2) PGPR	$<1.28 \times 10^2$	0.00	6.63×10^4	6.63×10^4
	NSD	NSD	P = 0.01	P = 0.01

[1/] Planting date - May 26, 1982.
[2/] E. carotovora populations.
[3/] PGPR populations .

The Association of *Erwinia carotovora* on Weed Roots in the Field Greenhouse

G.D. Franc and M.D. Harrison

Abstract

Weed-root samples were collected at three times during the growing season in the San Luis Valley. Contaminated weed-roots were not found in samples collected from virgin land and only one contaminated weed-root (out of 480 samples) was collected from land cropped continuously to potatoes for 20 years. The data suggest that virgin land areas do not have weeds harboring *E. carotovora* and that continuous cropping of potatoes possibly generated an *Erwinia*-suppressive soil. Paradoxically, data from one continuously cropped field suggests that low levels of *E. carotovora* can remain in association with weed-roots for at least 16 years since the last potato crop.

Data from soil survival studies conducted in the greenhouse showed that *E. carotovora* is unable to survive in association with weed-roots for prolonged periods of time and suggested that population decline was more rapid in the "suppressive" soil. Only preliminary data from this study are presented.

Materials and Methods

Surveys for the presence of *E. carotovora* in weed rhizospheres were conducted in the San Luis Valley in 1982. Weed samples were collected from non-cropped (virgin) and cropped land. Additional samples were collected from land cropped to potatoes continuously for 20 yrs. and held fallow for the last 3 years.

Plants were removed from the soil and excess soil removed from the root system by shaking the plants. Roots were cut into short segments and placed

into culture tubes containing PT enrichment medium. The cultures were incubated for 48 hours and aliquots of the medium were streaked on Stewart's MacConkey pectate medium to detect Erwinia.

The survival of E. carotovora in soil collected from the assayed fields is currently being studied in the laboratory and greenhouse. Results of the 1982 field surveys and preliminary data for the greenhouse studies are presented.

Results and Discussion

Results were consistent with those reported for previous years. Contaminated weed-roots were not found in samples collected from virgin land and only one contaminated weed root out of 480 was detected in samples from land cropped continuously to potatoes for 20 years then held fallow since 1980. Paradoxically, data from one field continuously cropped with crops other than potatoes suggests that low levels of E. carotovora can remain in association with weed-roots for at least 16 years since the last potato crop. This field, in fact, yielded a higher percentage of contaminated weeds than the field cropped with potatoes for 20 years and thus appeared to be more "conducive" to Erwinia survival.

The data suggest that virgin land areas do not have weeds harboring E. carotovora and that continuous cropping of potatoes possibly generated an Erwinia "suppressive" soil. Perhaps E. carotovora antagonists in the soil were selected by the continuous potato cropping and resulted in the development of "suppressiveness".

Data from soil survival studies (Table 1) conducted in the greenhouse showed that E. carotovora is unable to survive in association with weed-roots for prolonged periods of time and suggested that decline was more rapid at higher temperatures and also in the suppressive soil. However, assay

dates were too far apart and the experiment will be repeated using shorter inoculation to assay time intervals.

Isolates from the study are currently being tested to determine if different subspecies of E. carotovora i.e., ECC and ECA, are better adapted for soil survival. Additional experiments using test tubes have been initiated to determine the ability of rifampicin resistent ECC and ECA to survive in "conducive" and "suppressive" soil.

Table 1. The Effect of Soil Type and Temperature on the Survival of *Erwinia carotovora* on Weed Roots - Greenhouse, 1982-1983.

Soil Source	Temperature	Date of Weed Root Assay and Percentage Contaminated with *E. carotovora* 1/ October 29, 1982	January 21, 1982
Suppresive	18°C	100%	0%
	24°C	100%	0%
Conducive	18°C	100%	2%
	24°C	100%	0%
Sterile	18°C	100%	15%
	24°C	100%	0%
Check (Non-inoculated Soil)	18°C	NT 2/	0%
	24°C	NT	0%

1/ *Kochia* weed seeds were planted in each soil type and equal numbers of *E. carotovora* subsp. *carotovora* (ECC) and *E. carotovora* subsp. *atroseptica* (ECA) cells were watered in. Inoculation date was October 14, 1982.

2/ Not Tested

Post Harvest Treatment of Potato Tubers With RES-SRL444 for Control of *Erwinia*

G.D. Franc and M.D. Harrison

Abstract

Post harvest spraying or dipping of tubers with experimental compound RES-SRL444 failed to significantly reduce the population of *Erwinia* on tuber surfaces or to reduce the number of soft rot pockets developing on tubers after a period of time in storage. There was a trend toward lower populations on the tuber surface as the concentration of the compound in spray applications was increased from 5 25%. Spray applied at the rate of 5% resulted in the lowest soft rot potential in the treated tubers.

Materials and Methods

Tubers (cv. Monona) from the 1982 growing season were used in the study. Tubers were treated by spraying with water, 5%, 10% and 25% RES-SRL444 or by dipping tubers in water or water containing 25% RES-SRL444. Non-treated tubers were included as an additional control.

The number of *Erwinia* per cm^2 of tuber surface and the Erwinia soft rot potential (internal *Erwinia*) of tuber samples was determined for treated seedlots.

Soft-rot potential was determined by wrapping tubers in wet paper towels plus saran wrap, and incubating at toom temperature for four days (DeBoer, S.H. and A. Kelman. 1975. Amer. Potato J. 52:117-123). The presence of *Erwinia* in the resulting soft rot pockets was determined by plating samples onto Stewart's MacConkey pectate medium. The soft rot potential of the seedlot was calculated according to the formula: *Erwinia* soft-rotting potential = (percentage of tubers assayed developing soft-rot pockets) X (average number of soft-rot pockets per tuber) X (percentage of

tubers assayed yielding Erwinia carotovora) ÷ 100 (Harrison and Vogt. 1978. Proc. 17th Ann. Wash. Pot. Conf. pp. 41-47).

Data for the effect of treatment on E. carotovora subsp. carotovora (ECC) vs E. carotovora subsp. atroseptica (ECA) are still being collected and will not be included in this report.

Results

The large degree of variation between replications did not allow significant differences to be observed for the various treatments. This is characteristic of data of this type since adequate sample sizes are difficult to process. Data presented in Tables 1 and 2 show definite trends, however.

Data in Table 1 show that spray treatment of tubers with increasing concentrations of RES-SRL444 consistently decreased Erwinia surface populations. Spray treatment of tubers with 25% RES-SRL444 was slightly better than the 25% tuber dip treatment.

Data in Table 2 show that treatment with 5% RES-SRL444 spray resulted in tubers with the lowest Erwinia soft-rot potential. Non-treated tubers had the greatest soft-rot potential.

Discussion

The data suggest that although 25% RES-SRL444 spray was most effective in reducing surface populations of Erwinia 5% spray was most effective in reducing the soft-rot potential of the treated seedlots.

Table 1. The Effect of RES-SRL444 Tuber Treatment on Surface Populations of *Erwinia*.

RES-SRL444 Tuber Treatment	Average Number of *Erwinia* Colony Forming Units Per cm^2 of Tuber Surface
1) H_2O spray	1.09×10^5 a[1/]
2) 5% spray	4.34×10^5 a
3) 10% spray	5.56×10^3 a
4) 25% spray	1.21×10^2 a
5) H_2O dip	4.55×10^4 a
6) 25% dip	1.40×10^3 a
7) Non-treated	1.55×10^3 a
	NSD

[1/] Means with the same letters do not differ significantly ($P > 0.05$). NSD = no significant differences.

Table 2. The Effect of RES-SRL444 Tuber Treatment on *Erwinia* Soft-Rot Potential of Treated Seedlots

RES-SRL444 Tuber Treatment	*Erwinia* soft-rot potential of treated tubers[1/]	
1) H_2O spray	172	a[2/]
2) 5% spray	107	a
3) 10% spray	270	a
4) 25% spray	238	a
5) H_2O dip	271	a
6) 25% dip	230	a
7) Non-treated	367	a
	NSD	

[1/] A higher soft-rot potential indicates a greater potential for loss due to *Erwinia* soft-rot (See text).

[2/] Means with the same letters do not differ significantly ($P>0.05$). NSD = no significant differences.

Dilution end-point assay of *Corynebacterium sepedonicum* infectivity on the potato cultivars Russet Burbank and Centennial

G.D. Franc and M.D. Harrison

Abstract

The objective of this study is to determine if potato seedpieces and daughter tubers can become latently infected with *Corynebacterium sepedonicum*. This may be an explanation for the cyclical re-occurrence of ringrot infection in inspected seed potatoes.

Russet Burbank and Centennial tubers were inoculated with serial dilutions of *C. sepedonicum* to determine if latent ringrot infection would occur in the field. Results for studies initiated in 1981 (DEP81) showed that plants failed to develop foliar symptoms even though daughter tubers became infected. Daughter tubers were planted in the field in 1982 and foliar symptoms were observed in plants receiving greater than 10^8 cells per original seedpiece.

Results for treatments inoculated in 1982 (DEP82) showed that at least 10^6 *C. sepedonicum* cells per seedpiece were needed for foliar symptom development.

Centennial failed to develop foliar ringrot symptoms in 1981 and 1982 even though daughter tubers were found to be infected. Further assays will be conducted on tubers harvested from the DEP81 and DEP82 studies.

Materials and Methods

Foundation grade, 1980, Russet Burbank and Red McClure seed potatoes purchased in the San Luis Valley and Centennial seed potatoes provided by the S.L.V. Research Center were used in the dilution end-point (DEP81) study. Tubers provided by the S.L.V. Research Center were used in the DEP82 study.

Treatments were planted in a field plot ca 33 ft x 120 ft. Inoculated tubers were planted by hand on May 28, 1981 and May 26, 1982 for DEP81 and DEP82 study plots, respectively. Treatment plots were in a randomized complete block design consisting of two cultivars, six inoculation treatments and four replications. Individual treatment plots consisted of 10 treated tubers planted at a 14 inch spacing followed by three Red McClure spacers planted at 12 inch intervals. The plots were cultivated by hand and volunteer potatoes were rogued throughout the growing season. The plot was furrow irrigated by S.L.V. Research Center personnel.

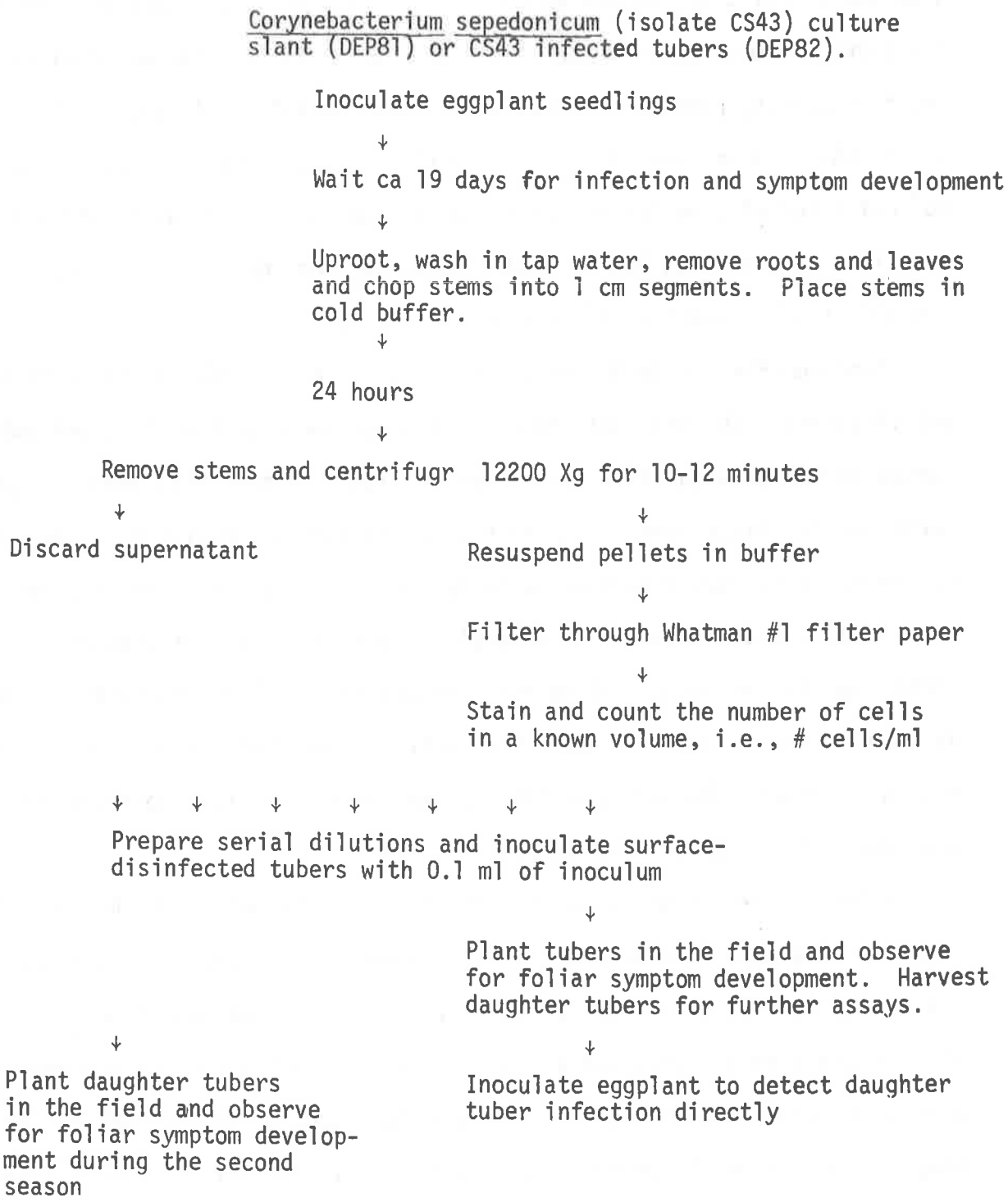
Tuber treatments for the DEP81 study consisted of five serial dilutions of Corynebacterium sepedonicum (SC43). (Original CS43 isolate, provided by S. Slack, University of Wisconsin-Madison, Madison, Wisconsin 53706.) Serial dilutions of CS43 cells were prepared as follows: On May 6, 1981, eggplant seedlings (Solanum melongena "Black Beauty") in the two-leaf stage were inoculated using a sterile 1 ml tuberculin syringe and 26G 1/2 needle containing C. sepedonicum (CS43) bacteria suspended in 0.05 M phosphate buffer, pH 7.2. The first foliar ringrot symptoms developed in ca 12 days postinoculation. On May 25, 1981, the seedlings were uprooted, washed in cold tap water and roots and leaves were removed with a knife. The remaining stems (approximately 458.5 g wet weight) were cut into ca 1 cm lengths and placed in 4-5 l of cold (40°F) 0.05 M phosphate buffer, pH 7.2 and allowed to stand overnight. Buffer was passed through cheesecloth to remove the stem segments and large debris. CS43 cells in the strained buffer were concentrated into a pellet using a Servall refrigerated automatic centrifuge and a SS34 rotor at 11 - 12 x 10^3 rpm for 10-12 minutes. Pellets were resuspended in buffer (final volume approximately 350 ml) and filtered under a slight vacuum through Watman #1

filter paper. Serial dilutions of the filtrate were made and lightly stained with Gram stain crystal violet. Stained filtrate was placed in a Petroff-Hauser counting chamber and the number of cells per 2.5 x 10^{-3} mm^2 square were counted with the aid of a microscope at 45 X. Fifty squares were counted to determine the average number of cells per 2.5 x 10^{-3} mm^2. The chamber was emptied, refilled and the counting procedure repeated (replication I:$\bar{X}$ = 3.16 cells/2.5 x 10^{-3} mm^2, S = 1.765. Replication II: $\bar{X}$ = 3.18 cells/2.5 x 10^{-3} mm^2, S = 0.625) and 6.34 X 10^9 cells per ml were determined to be present in the undiluted filtrate. Serial dilutions were carried out to produce cell suspensions of 6.34 x 10^9, 1 x 10^7, 1 x 10^5, 1 x 10^3 and 1 x 10^2 cells/ml buffer.

Inoculum for the DEP82 study was prepared in a similar manner with minor modifications. The original CS43 inoculum was prepared from infected tubers harvested the previous fall from infected plants in the DEP81 study. Eggplant seedlings (cv Black Beauty) in the 4 leaf stage were inoculated as described above on May 6, 1982. Symptoms started to develop on May 20 and stems were harvested on May 24. Bacterial cells from eggplant stems were concentrated by centrifugation on May 25 and counted (replication I: $\bar{X}$ = 0.55 cells/5 x 10^{-8} ml, II: 0.50 cells/5 x 10^{-8} ml, III: 0.60 cells/5 x 10^{-8} ml, IV: 0.60 cells/ 5 x 10^{-8} ml. S = 0.048). The cell purification and quantification procedure used in 1981 and 1982 is briefly outlined in Figure 1.

Tubers to be inoculated were surface disinfected with 10% chlorox, rinsed with cool tap water and allowed to dry. Tubers were inoculated by scooping out the stolon end with an "EKCO" fruit baller (2.8 cm diameter), pipetting 0.1 ml of inoculum directly into the depression, replacing the tuber piece and inserting a small piece of wooden toothpick to hold the tuber piece in place and immediately dipping the entire stolon end (approximately 1/4 - 1/3 of the tuber) into melted paraffin (Gulfwax). A volume of 0.1 ml was sufficient to coat the cut surface inside the tuber with inoculum when the excised tuber piece was replaced and slight

Figure 1. Flow chart outline of inoculum preparation for Corynebacterium sepedonicum dilution end-point assay - Center, CO, 1981 and 1982.



pressure applied. The paraffin sealed the excised tuber piece to the intact tuber and prevented the inoculum from drying out. Control tubers, inoculated with buffer alone, were treated in the same manner.

The treatment plots were observed throughout the growing season by Potato Virus Lab research personnel and Potato Certification inspectors for the development of foliar ringrot symptoms.

On September 21, 1981, the DEP81 plots were harvested. The center three hills in each treatment plot were dug up with a fork and the uninjured tubers were placed in a paper bag. The tubers were placed in cold storage within 12 hours of harvest for later assays. DEP81 daughter tubers were replanted in the field in 1982 at the same time DEP82 inoculated seedpieces were planted. The DEP81 and DEP82 plots were harvested in a similar manner on September 22 and 23, 1982. Tuber assays have been completed on DEP81 tubers harvested in 1981 only. Tubers harvested from the second year of the DEP81 study and first year of the DEP82 study (September 22 and 23, 1982) will be assayed in a similar manner in the spring of 1983.

Daughter tubers harvested from the DEP81 study in 1981 were divided into two lots of ca 10 tubers each. The first lot was replanted in the field and observed for symptom development in 1982 at the same time the freshly inoculated DEP82 tubers were planted (i.e., the DEP82 study is a repeat of the DEP81 study except they are staggered by one year). The second lot of DEP81 daughter tubers were tested on eggplant for the presence of ringrot infection during the spring of 1982.

Tubers were tested by lightly shaving the stolon end of each tuber and excising vascular tissue (potentially infected) with a sterile knife. The tissue was macerated with a sterile mortar and pestle to which a small volume of buffer had been added. Ten tubers per treatment per replication were assayed in this

manner. Inoculum was injected into eggplants (2 pots of ca 3-4 plants each) for symptom development. This constituted a positive tuber assay.

Results

DEP81 study initially planted May 28, 1981:

Primary foliar ringrot symptoms failed to develop at any time during the growing season. The plants were water stressed throughout the 1981 growing season and, although this was not desirable, did demonstrate that water stress can mask or delay the development of foliar ringrot symptoms when mother tubers were inoculated with as many as 6.34×10^8 C. sepedonicum cells per tuber (Table 1).

Daughter tubers harvested after the 1981 growing season were infected with ringrot according to the eggplant test (column 2, Table 1). However, daughter tuber infection was only detected in treatments for which the mother tuber was inoculated with at least 10^4 C. sepedonicum cells (Russet Burbank) or 10^6 cells (Centennial).

Ten daughter tubers harvested after the 1981 growing season were replanted in the field on May 26, 1982. Although plants were observed throughout the growing season secondary foliage symptoms were only detected in Russet Burbank plants for which the grandmother tuber had received 6.3×10^8 cells. Foliar symptoms were not observed for similarly treated Centennial plants.

DEP82 study initially planted May 26, 1981:

The only data available are from field observations made during the 1982 growing season (Table 2). Primary foliage symptoms developed in Russet Burbank plots when the mother tuber received at least 10^6 C. sepedonicum cells. Foliar symptoms failed to develop in similarly inoculated Centennial plants. Daughter tubers harvested in the fall of 1982 will be assayed for ringrot infection on eggplant during the spring of 1983 and replanted in the field. Plants will be observed for secondary foliar symptom development.

Table 1. The effect of Corynebacterium sepedonicum (ringrot) tuber inoculum concentration on symptom expression in progeny plants and daughter tubers - DEP81 study, Center, Colorado.

Cultivar	Number of cells per mother tuber	Primary foliage symptoms 1/	Daughter tuber infection 2/	Secondary foliage symptoms 3/
Russet Burbank	6.3×10^8	-	+	+
	10^6	-	+	-
	10^4	-	+	-
	10^2	-	-	-
	10^1	-	-	-
	Buffer	-	-	-
Centennial	6.3×10^8	-	+	-
	10^6	-	+	-
	10^4	-	-	-
	10^2	-	-	-
	10^1	-	-	-
	Buffer	-	-	-

1/ Inoculated tubers were planted in the field in 1981 and symptom expression recorded.

2/ Tubers were assayed for ringrot infection using the eggplant test.

3/ Daughter tubers harvested in 1981 were replanted in the field in 1982 and symptom expression recorded.

Table 2. The effect of Corynebacterium sepedonicum (ringrot) tuber inoculum concentration on symptom expression in progeny plants and daughter tubers - DEP82 study, Center, Colorado.

Cultivar	Number of cells per mother tuber[1/]	Primary foliage symptoms	Daughter tuber infection[2/]	Secondary foliage symptoms[3/]
Russet Burbank	10^9	+	NT	NT
	10^6	+	NT	NT
	10^4	-	NT	NT
	10^2	-	NT	NT
	10^1	-	NT	NT
	Buffer	-	NT	NT
Centennial	10^9	-	NT	NT
	10^6	-	NT	NT
	10^4	-	NT	NT
	10^2	-	NT	NT
	10^1	-	NT	NT
	Buffer	-	NT	NT

[1/] Inoculated tubers were planted in the field in 1981 and symptom expression recorded.

[2/] NT = not tested. Daughter tuber infection will be detected by the eggplant test.

[3/] NT = not tested. Daughter tubers will be replanted in the field in 1983 and symptom expression recorded.

Discussion

Results from both studies have been very encouraging. Data presented for the DEP81 study show that mother seedpieces can be inoculated with as many as 6.3×10^8 C. sepedonicum cells and the daughter plants will lack foliar ringrot symptoms. Eggplant assays indicate that even though foliar symptom expression was lacking, infection of daughter tubers had occurred. However, when daughter tubers are planted in the field some foliar disease expression occurred. Although daughter tubers for higher dilutions (less cells per tuber) were infected (eggplant test) symptom expression was not evident in the field during the second season. For Centennial, plants never expressed foliar symptoms in either season even though mother tubers were inoculated with sufficient cell numbers to result in daughter tuber infection. This demonstrated that ringrot inoculum could be carried through at least one tuber generation without the expression of foliar disease symptoms and that there was a definite inoculum concentration effect (dilution end-point) on ringrot disease expression. It was noted that a mother tuber (Centennial) receiving 10 cells in the DEP81 study had positive ringrot symptoms evident at harvest in the fall of 1981. Daughter tubers harvested from that treatment have shown no evidence of ringrot infection.

The same conclusions cannot be made for the DEP82 study since tuber assays are yet to be made. Foliar symptoms were observed in the field and in this respect the effect of inoculum concentration on disease expression can be seen.

Daughter tubers (DEP82) and granddaughter tubers (DEP81) will be replanted in the field in 1983 to determine how many generations ringrot infection can be carried latently in the tubers. A new study, DEP83, will be initiated once again in order to test primary foliar symptom development in the field.

FINAL REPORT

The Effectiveness of RES-SRL444 and RES-SRL445 as a Foliar Early Blight Control in the San Luis Valley

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Abstract

RES-SRL444 and RES-SRL445 alone and as a combined mixture were compared to Bravo 500 as a foliar application for early blight control in the San Luis Valley. Final disease ratings show Bravo 500 offered significantly greater ($P \leq 0.05$) early blight control than the experimental materials when data for the estimated percentage of leaf area blighted and the estimated percentage of leaflets infected were compared. RES-SRL444 alone and in combination with RES-SRL445 significantly reduced the average number of early blight lesions per leaflet compared to the non-treated control ($P \leq 0.05$). Early blight control provided by RES-SRL444 was equivalent to that offered by Bravo 500 when data for the estimated percent of defoliation and the average number of early blight lesions per leaflet were compared ($P \leq 0.05$). Generally, Bravo 500 provided the most effective early blight control followed by RES-SRL444 alone and in combination with RES-SRL445. RES-SRL445 alone did not significantly reduce disease ratings when compared to the non-treated control at any time during the growing season ($P \leq 0.05$). Treatment with Bravo 500 significantly increased total tuber yield ($P \leq 0.05$). There were no other treatment effects on total tuber yield or the proportion of tubers within each of six market grades within the harvested samples.

Materials and Methods

A sandy-loam field cropped continuously for 20 years with potatoes was selected for the study. The field was located near Center, Colorado in the San Luis Valley (a high desert valley at ca 7,600' mean sea level). Certified, Foundation grade, Russet Burbank seed potatoes produced in the San Luis Valley were used as the seed source. Each treatment plot was planted with an assist-feed 2-row tractor-drawn potato planter. Plots consisted of four potato rows each 50 ft long (1.3×10^{-2} acre). Fertilizer was applied in the row at the time of planting and plots were furrow-irrigated during the growing season. Treatment plots were planted in a randomized complete-block design with four replications and five treatments (Figure 1). Treatments consisted of a non-treated control, Bravo 500 (2 pt/A), RES-SRL444 (10% vol/vol), RES-SRL445 (2% vol/vol) and RES-SRL444 and RES-SRL445 as a mixture (10% and 2% vol/vol, respectively).

All experimental fungicide treatments were applied using a 4-row John Bean power sprayer pulled by a small tractor. Treatments were delivered at 50 psi in a total volume of 90 gallons per acre. The first fungicide applications were applied on July 8 immediately prior to the appearance of first early blight lesions. A total of six fungicide applications were made at ca 10-day intervals throughout the growing season (Table 1). The non-treated control received no fungicide applications.

Figure 1. Field plot plans used for early blight control fungicide trials - Center, Colorado, 1982.

8 buffer rows	4 treatment rows[1/] Treatment location	18 buffer rows
	I - 1 2 4 5 3 II - 2 1 3 5 4 III - 5 2 1 4 3 IV - 3 4 1 2 5	

[1/] Each individual treatment consisted of four rows 50 ft long (1.3×10^{-2} acre). All treatments were replicated four times. The treatment designations used were:

1) Non-treated control
2) Bravo 500 (2 pt/A)
3) RES-SRL444 (10% vol/vol)
4) RES-SRL445 (2% vol/vol)
5) RES-SRL444 + 445 (10% and 2% vol/vol, respectively)

Table 1. Fungicide application dates and general weather conditions at time of application - Center, Colorado, 1982.

Application Date[1/]	General weather conditions during application[2/]
July 8	Partly cloudy, cool and light breeze
July 19	Partly cloudy, cool and wind gusts
July 27	Cloudy, cool and calm; rain in the a.m.
August 6	Sunny, cool and calm
August 17	Sunny, warm and calm; drizzle in the a.m.
August 26	Partly cloudy, cool and calm

1/ Fungicide treatments were applied at 50 psi at a net rate of 90 gallons per acre.

2/ General observations were made at the time of fungicide application. For a more complete description see the weather data sheets following Appendix 5.

Foliar early blight disease severity was assessed by two methods, i.e., (1) Barratt-Horsfall (BH) field ratings using a scale of 0-11 to estimate the percentage of leaflets infected by early blight and the percentage of leaf area blighted and (2) collection of leaf samples and actual counts of the number of early blight lesions per leaflet.

Early blight foliar infection data (BH scale) were collected on August 5, August 17 and August 26. Disease assessment on each of the three dates consisted of four separate BH readings each for the top, middle and bottom third of the plant canopy on the center two rows of each treatment plot. On the same dates four leaves each from the top and bottom half of the plant canopy were collected from the center two rows of each treatment plot. Leaves were returned to the laboratory and the number of early blight lesions per leaflet was determined by direct counts. Early blight disease severity was also measured indirectly on August 17, August 26 and September 7 using BH scale field ratings to estimate the percentage of defoliation observed in each treatment plot. Five BH scale ratings were made for each treatment plot on each date.

For each method of disease evaluation all readings collected from each treatment plot on each date were averaged within each replication to give a composite disease severity index. These data are listed in Appendices 1-4. BH ratings were analyzed directly in a two-way analysis of variance and means were separated using Tukey's test (HSD). Significance occurred when $P \leq 0.05$. BH rating data were converted to percentages using the appropriate conversion table for presentation in this report. Lesion count data were expressed as the average number of lesions per leaflet and were analyzed in a two-way analysis of variance as described above.

Yield data were collected on September 22. Plots were harvested using a 2-row potato harvester. Tubers were picked into baskets by hand and the total yield from the center two rows (each 40 ft long) was determined. A random tuber sample (ca 50 lb) from each plot was retained to determine the proportion (percentage) of each tuber grade within the sample. The proportion (percentage) of each tuber grade was determined by weighing the total sample and determining the weight of each potato grade in the sample. Data for total yield and the various tuber grades are listed in Appendix 5. Yields in pounds per 80 ft of harvested row were analyzed in a two-way analysis of variance with Tukey's test. The percentage of each tuber grade in each sample was converted to an arcsin value prior to analysis. After analysis, total yield data were converted to cwt/acre and presented in the tables along with the percentage of tubers in each grade category.

Results

Data for the estimated percentage of early blight infected leaflets are shown in Table 2. Bravo 500 provided significantly greater early blight control than RES-SRL444 and 445 applied alone or as a mixture on each data collection date ($P \leq 0.05$). On August 5, RES-SRL444 alone and RES-SRL444 + RES-SRL445 as a mixture were significantly better than RES-SRL445 alone ($P \leq 0.05$). RES-SRL445 provided significantly less control than RES-SRL444 alone or in combination with RES-SRL445 and Bravo 500 ($P \leq 0.05$). On August 17, RES-SRL444 provided significantly

Table 2. The effect of different fungicide treatments on the estimated percentage of leaflets infected (cv. Russet Burbank by *Alternaria solani* - Center, Colorado, 1982.

Fungicide treatment[1/]	Estimated percentage of leaflets infected[2/]		
	August 5	August 17	August 26
1) Control (Non-treated)	9.0 a	35.0 a	99.0 a
2) Bravo 500 (2 pt/A)	0.4 d	2.5 d	46.0 d
3) RES-SRL444 (10% v/v)	3.0 c	11.5 c	95.0 abc
4) RES-SRL445 (2% v/v)	8.5 ab	28.0 ab	98.6 ab
5) RES-SRL444 + 445 (10% and 2% v/v, respectively)	3.0 c	15.0 bc	95.0 abc

[1/] Fungicide treatments were applied at 50 psi at a net rate of 90 gallons per acre.

[2/] Four Barratt-Horsfall (BH) scale (0-11) readings per plot each for the top, middle and bottom one-third of the plant canopy were averaged for each replication on each date.

greater disease control than RES-SRL445 ($P \leq 0.05$). The disease control provided by RES-SRL444 and 445 as a mixture was intermediate and statistically equivalent to the separate Allied treatments on this date. On August 26, RES-SRL444 and 445 applied separately or as a mixture provided disease control statistically equivalent to the non-treated control ($P \leq 0.05$).

Data for the average number of early blight lesions per leaflet on August 5, August 17 and August 26 are shown in Table 3. On August 5, Bravo 500 and RES-SRL444 alone and in combination with RES-SRL445 provided significantly greater early blight control than the non-treated control ($P \leq 0.05$). RES-SRL445 was not significantly different from the non-treated control at any time during the growing season ($P \leq 0.05$). However, all Allied treatments were statistically equivalent on each date. RES-SRL444 alone and in combination with RES-SRL445 provided early blight control statistically equivalent to that provided by Bravo 500 on August 5 and August 17 ($P \leq 0.05$). On August 26, Bravo 500 provided significantly greater early blight control than RES-SRL445 alone or in combination with RES-SRL444 but was not statistically better than RES-SRL444 alone ($P \leq 0.05$).

Data for the estimated percentage of leaf area blighted (Table 4) show that on August 5 RES-SRL444 alone or in combination with RES-SRL445 and Bravo 500 provided significantly greater control than RES-SRL445 alone and the non-treated control ($P \leq 0.05$). On August 17, data for Bravo 500 was statistically equivalent to RES-SRL444 alone or in combination with RES-SRL445. On August 17 and August 26, RES-SRL444 alone provided significantly greater control than did RES-SRL445 ($P \leq 0.05$). The amount of leaf area blighted in the RES-SRL445 plots was not significantly different from that in the non-treated control at any time during the growing season ($P \leq 0.05$). RES-SRL444 in combination with RES-SRL445 provided disease control intermediate to that offered by the two fungicides applied alone.

Data for the estimated percentage of defoliation observed in the field on August 17, August 26 and September 7 are shown in Table 5. Significant differences among treatment means occurred on August 26 and September 7 but not on August 17 ($P \leq 0.05$). On August 26 the Bravo 500 plots had significantly less defoliation than all other treatments. Defoliation in RES-SRL444, RES-SRL445 and RES-SRL444 + 445 plots was statistically equivalent throughout the growing season ($P = 0.05$). RES-SRL444 was the only Allied treatment that had significantly less defoliation ($P \leq 0.05$) than the non-treated control (August 26).

Total yield data are shown in Table 6. Significant differences could be shown among treatment means when an LSD test ($P \leq 0.05$) was used for mean separation but not when the more conservative HSD test was used. Treatment with Bravo 500 had the greatest total yield. The non-treated control and RES-SRL444 had the lowest tuber yields. The fungicide treatments did not significantly ($P \leq 0.05$) influence the proportion of each tuber grade within the samples.

Table 3. The effect of different fungicide treatments on the average number of *Alternaria solani* lesions on cv. Russet Burbank leaflets - Center, Colorado, 1982.

Fungicide treatment[1/]	Average number of lesions per leaflet[2/]		
	August 5	August 17	August 26
1) Control (Non-treated)	1.8 a	12.3 a	40.7 a
2) Bravo 500 (2 pt/A)	0.1 c	0.3 c	5.7 d
3) RES-SRL444 (10% v/v)	0.5 bc	2.3 bc	21.2 bcd
4) RES-SRL445 (2% v/v)	1.4 ab	7.2 ab	31.0 ab
5) RES-SRL444 + 445 (10% and 2% v/v, respectively)	0.5 bc	5.2 bc	23.7 bc

[1/] Fungicide treatments were applied at 50 psi at a net rate of 90 gallons per acre.

[2/] Four leaves each from the top and bottom half of the plant canopy were collected on each date. The number of leaflets and early blight lesions for each treatment plot were determined by actual counts.

Table 4. The effect of different fungicide treatments on the estimated percentage of leaf area blighted (cv. Russet Burbank) by Alternaria solani - Center, Colorado, 1982.

Fungicide treatment[1/]	Estimated percentage of leaf area blighted[2/]		
	August 5	August 17	August 26
1) Control (Non-treated)	3.5 a	4.0 ab	11.5 ab
2) Bravo 500 (2 pt/A)	0.2 c	0.8 c	3.0 d
3) RES-SRL444 (10% v/v)	1.6 b	1.6 bc	9.0 bc
4) RES-SRL445 (2% v/v)	3.5 a	4.0 a	13.0 a
5) RES-SRL444 + 445 (10% and 2% v/v, respectively)	1.2 bc	1.8 abc	10.5 abc

[1/] Fungicide treatments were applied at 50 psi at a net rate of 90 gallons per acre.

[2/] Four Barratt-Horsfall (BH) scale 0-11 readings per plot each for the top, middle and bottom third of the plant canopy were averaged for each replication on each date.

Table 5. The effect of different fungicide treatments on the estimated percentage of defoliation observed in the field for cv. Russet Burbank - Center, Colorado, 1982.

Fungicide treatment[1/]	Estimated percentage of defoliation[2/]		
	August 17	August 26	September 7
1) Control (Non-treated)	6.5 a	35.0 a	86.0 a
2) Bravo 500 (2 pt/A)	4.0 a	3.5 e	26.5 c
3) RES-SRL444 (10% v/v)	6.0 a	11.5 bcd	52.0 abc
4) RES-SRL445 (2% v/v)	9.0 a	22.0 ab	84.0 ab
5) RES-SRL444 + 445 (10% and 2% v/v, respectively)	6.5 a	14.0 abc	70.5 abc

[1/] Fungicide treatments were applied at 50 psi at a net rate of 90 gallons per acre.

[2/] Five random Barratt-Horsfall (BH) scale 0-11 ratings were made for each treatment plot on each date.

Table 6. The effect of fungicide treatments on total tuber yield and tuber quality (grade) of cv. Russet Burbank tubers - Center, Colorado, 1982.

Fungicide treatment[1/]	Total yield[2/3/] (cwt/A)	Percentage of total yield as:[4/]					
		US #1		Total US #1	US #2	Grade "B"	Culls
		<10 oz	>10 oz				
1) Control (Non-treated)	158.4	40.2	1.9	42.1	6.4	35.9	12.5
2) Bravo 500 (2 pt/A)	201.7	36.9	3.6	40.5	7.3	28.6	20.6
3) RES-SRL444 (10% v/v)	156.8	40.2	2.6	42.8	10.4	30.7	12.0
4) RES-SRL445 (2% v/v)	170.3	37.5	3.5	41.0	9.9	34.3	12.4
5) RES-SRL444 + 445 (10% and 2% v/v, respectively)	166.3	43.2	0.4	43.6	5.9	34.1	13.6

[1/] Fungicide treatments were applied at 50 psi at a net rate of 90 gallons per acre.

[2/] Two center rows (each 40 ft long) from each plot were harvested and total yield determined. Data were converted to cwt/acre for presentation in the table.

[3/] Significant differences occurred among treatment means ($P \leq 0.05$) on the basis of a two-way analysis of variance test. However, due to the conservative nature of the HSD value used for mean separation, grouping of means did not occur.

[4/] A sample (ca 50 lbs) was graded for each treatment per replication. Significant differences did not occur among treatment means.

Discussion

Generally, Bravo 500 provided the most effective early blight control followed by RES-SRL444 alone and RES-SLR444 in combination with RES-SRL445. RES-SRL445 alone did not significantly reduce disease ratings when compared to the non-treated control at any time during the growing season.

There were no obvious phytotoxic effects attributed to any of the treatments during the growing season. Marginal leaf necrosis or "burning" was observed to various degrees in all treatment plots during the growing season. On August 17, this was observed to be more common in the plots treated with RES-SRL444 in combination with RES-SRL445 and possibly even in plots treated with RES-SRL444 alone. However, these observations were very general and not quantitative.

The net effect of RES-SRL444 alone and in combination with RES-SRL445 on the general appearance of the treatment plots (early blight disease severity and defoliation) was visually very obvious in the field when compared to the non-treated control and even to RES-SRL445 alone. The data shown in the tables included in this report are consistent with the marked differences observed in the appearance of the plots in the field.

The fact that there was not a more pronounced effect of early blight control on yield is consistent with other reports. Other diseases commonly present (e.g. Verticillium wilt) often mask the beneficial effects of early blight control on yield.

Appendix 1. The Barratt-Horsfall ratings for the estimated percentage of leaflets infected by Alternaria solani - Center, Colorado, 1982.[1/]

Treatment	August 5, 1982					August 17, 1982					August 26, 1982				
	Rep: I	Rep: II	Rep: III	Rep: IV	Mean	Rep: I	Rep: II	Rep: III	Rep: IV	Mean	Rep: I	Rep: II	Rep: III	Rep: IV	Mean
1) Control (Non-treated)	3.6	2.3	2.1	2.5	2.6	5.5	4.3	3.3	5.8	4.7	11.0	10.2	9.8	10.8	10.5
2) Bravo 500 (2 pt/A)	0.0	0.2	0.2	0.4	0.2	0.4	0.9	1.2	1.8	1.1	3.2	5.7	6.5	5.8	5.3
3) RES-SRL444 (10% v/v)	1.2	1.3	1.3	1.3	1.3	2.8	3.1	2.7	2.8	2.9	9.2	9.3	8.7	9.3	9.1
4) RES-SRL445 (2% v/v)	2.1	2.2	2.8	3.0	2.5	3.5	4.1	4.2	5.3	4.3	10.1	9.9	10.9	10.0	10.2
5) RES-SRL444 + 445 (10% and 2% v/v, respectively)	1.1	1.2	1.1	1.6	1.3	2.6	3.3	3.4	3.8	3.3	7.9	9.2	10.4	9.3	9.2

[1/] The mean Barratt-Horsfall grade ($\bar{X}$) was converted to "estimated percentage of leaflets infected" using the appropriate conversion table for presentation in the report.

Appendix 2. The Barratt-Horsfall ratings for the estimated percentage of leaf area blighted by *Alternaria solani* - Center, Colorado, 1982.[1/]

Treatment	August 5, 1982					August 17, 1982					August 26, 1982				
	Rep: I	Rep: II	Rep: III	Rep: IV	Mean	Rep: I	Rep: II	Rep: III	Rep: IV	Mean	Rep: I	Rep: II	Rep: III	Rep: IV	Mean
1) Control (Non-treated)	1.7	1.7	0.9	1.3	1.4	2.1	0.9	1.3	1.8	1.5	3.1	2.6	3.1	2.7	2.9
2) Bravo 500 (2 pt/A)	0.0	0.1	0.0	0.3	0.1	0.2	0.4	0.4	0.7	0.4	1.3	1.3	1.4	1.1	1.3
3) RES-SRL444 (10% v/v)	0.7	0.7	0.6	1.0	0.8	0.9	0.9	0.8	0.7	0.8	2.4	2.8	2.8	2.3	2.6
4) RES-SRL445 (2% v/v)	1.3	1.7	1.3	1.3	1.4	1.2	1.2	1.8	2.0	1.6	2.8	3.1	3.3	3.2	3.1
5) RES-SRL444 + 445 (10% and 2% v/v, respectively)	0.5	0.6	0.6	0.5	0.6	0.8	0.9	0.8	0.9	0.9	2.8	2.8	2.9	2.6	2.8

[1/] The mean Barratt-Horsfall grade ($\bar{X}$) was converted to "estimated percentage of leaf area blighted" using the appropriate conversion table for presentation in the report.

Appendix 3. The Barratt-Horsfall ratings for the estimated percentage of defoliation observed in the field - Center, Colorado, 1982.[1/]

Treatment	August 17, 1982					August 26, 1982					September 7, 1982				
	Rep: I	Rep: II	Rep: III	Rep: IV	Mean	Rep: I	Rep: II	Rep: III	Rep: IV	Mean	Rep: I	Rep: II	Rep: III	Rep: IV	Mean
1) Control (Non-treated)	2.2	1.4	1.8	3.0	2.1	6.0	3.2	4.6	5.0	4.7	10.6	5.0	5.8	9.8	7.8
2) Bravo 500 (2 pt/A)	1.0	1.4	1.4	2.4	1.6	1.6	1.0	1.0	1.8	1.4	5.0	4.2	3.0	4.4	4.2
3) RES-SRL444 (10% v/v)	1.6	1.2	2.6	2.4	2.0	4.0	2.8	2.2	2.6	2.9	6.8	5.6	4.8	5.2	5.6
4) RES-SRL445 (2% v/v)	1.2	3.0	2.8	3.4	2.6	3.8	3.8	3.8	4.2	3.9	7.0	6.4	8.2	8.6	7.6
5) RES-SRL444 + 445 (10% and 2% v/v, respectively)	1.2	2.0	2.0	3.2	2.1	3.0	4.0	2.8	3.0	3.2	7.2	5.6	6.0	7.4	6.6

[1/] The mean Barratt-Horsfall grade ($\bar{X}$) was converted to "estimated percentage of defoliation" using the appropriate conversion table for presentation in the report.

Appendix 4. The average number of early blight (*Alternaria solani*) lesions per leaflet - Center, Colorado, 1982.

Treatment	August 5, 1982					August 17, 1982					August 26, 1982				
	Rep: I	Rep: II	Rep: III	Rep: IV	Mean	Rep: I	Rep: II	Rep: III	Rep: IV	Mean	Rep: I	Rep: II	Rep: III	Rep: IV	Mean
1) Control (Non-treated)	2.94	0.69	1.51	1.90	1.76	15.13	8.61	10.17	15.41	12.33	58.59	27.26	35.62	41.44	40.73
2) Bravo 500 (2 pt/A)	0.04	0.19	0.06	0.06	0.09	0.50	0.28	0.25	0.12	0.29	2.63	3.68	6.84	9.58	5.68
3) RES-SRL444 (10% v/v)	0.68	0.38	0.35	0.49	0.48	1.86	0.26	2.86	4.16	2.29	23.02	20.06	20.88	20.96	21.23
4) RES-SRL445 (2% v/v)	1.21	0.79	1.17	2.36	1.38	4.22	6.36	6.79	11.40	7.19	24.69	25.30	33.54	40.40	30.98
5) RES-SRL444 + 445 (10% and 2% v/v, respectively)	0.38	0.37	0.43	0.85	0.51	1.80	2.35	3.19	13.28	5.16	21.47	23.98	25.78	23.50	23.68

Appendix 5. The effect of fungicide treatments on tuber yield and grade - Center, Colorado, 1982.

Treatment	Total Yield (pounds/A) Rep. I	Rep. II	Rep. III	Rep. IV	Mean
Appendix 5A:					
1) Control (Non-treated)	79.9	79.8	106.6	63.2	82.4
2) Bravo 500 (2 pt/A)	100.2	107.1	113.2	99.1	104.9
3) RES-SRL444 (10% v/v)	91.5	81.4	84.3	68.9	81.5
4) RES-SRL445 (2% v/v)	83.3	96.9	88.1	86.0	88.6
5) RES-SRL444 + 445 (10% and 2% v/v, respectively)	76.6	109.7	86.2	73.5	86.5
Appendix 5B: The proportion (percentage) of total yield graded as total US #1.1/					
1) Control (Non-treated)	51.2	20.0	46.5	40.6	42.1
2) Bravo 500 (2 pt/A)	18.8	52.3	34.4	56.4	40.5
3) RES-SRL444 (10% v/v)	34.3	44.1	43.7	49.3	42.8
4) RES-SRL445 (2% v/v)	23.1	44.9	45.0	51.1	41.0
5) RES-SRL444 + 445 (10% and 2% v/v, respectively)	36.9	41.6	47.6	48.2	43.6

1/ See Appendix 5A for total tuber yield harvested from each treatment plot. A tuber sample (ca 50 lbs) from each treatment plot was graded to determine the proportion of each grade within the sample.

Appendix 5 continued next page.

Appendix 5 (continued).

Treatment	Rep. I	Rep. II	Rep. III	Rep. IV	Mean
Appendix 5C: The proportion (percentage) of total yield graded as US #1 greater than 10 ounces.1/					
1) Control (Non-treated)	4.0	0.0	1.6	2.1	1.9
2) Bravo 500 (2 pt/A)	0.0	2.9	9.4	2.3	3.6
3) RES-SRL444 (10% v/v)	0.0	2.9	2.6	5.0	2.6
4) RES-SRL445 (2% v/v)	1.6	4.0	2.8	5.6	3.5
5) RES-SRL444 + 445 (10% and 2% v/v, respectively)	1.4	0.0	0.0	0.0	0.4

1/See Appendix 5A for total tuber yield harvested from each treatment plot. A tuber sample (ca 50 lbs) from each treatment plot was graded to determine the proportion of each grade within the sample.

Appendix 5D: The proportion (percentage) of total yield graded as US #1 less than 10 ounces.1/					
1) Control (Non-treated)	47.2	30.0	44.9	38.5	40.2
2) Bravo 500 (2 pt/A)	18.8	49.5	25.0	54.1	36.9
3) RES-SRL444 (10% v/v)	34.3	41.3	41.1	44.3	40.2
4) RES-SRL445 (2% v/v)	21.4	40.9	42.3	45.5	37.5
5) RES-SRL444 + 445 (10% and 2% v/v, respectively)	35.5	41.6	47.6	48.2	43.2

1/See Appendix 5A for total tuber yield harvested from each treatment plot. A tuber sample (ca 50 lbs) from each treatment plot was graded to determine the proportion of each grade within the sample.

Appendix 5 continued next page.

Appendix 5 (continued).

Treatment	Rep. I	Rep. II	Rep. III	Rep. IV	Mean
Appendix 5E: The proportion (percentage) of total yield graded as US #2.[1/]					
1) Control (Non-treated)	5.1	11.6	2.8	6.1	6.4
2) Bravo 500 (2 pt/A)	14.7	2.9	11.7	0.0	7.3
3) RES-SRL444 (10% v/v)	21.4	10.5	4.7	5.0	10.4
4) RES-SRL445 (2% v/v)	23.1	8.0	4.4	4.3	9.9
5) RES-SRL444 + 445 (10% and 2% v/v, respectively)	6.9	6.0	9.2	1.5	5.9

[1/] See Appendix 5A for total tuber yield harvested from each treatment plot. A tuber sample (ca 50 lbs) from each treatment plot was graded to determine the proportion of each grade within the sample.

Appendix 5F: The proportion (percentage) of total yield graded as grade "B".[1/]					
1) Control (Non-treated)	40.1	38.1	33.0	32.4	35.9
2) Bravo 500 (2 pt/A)	37.1	25.9	25.5	25.9	28.6
3) RES-SRL444 (10% v/v)	30.3	28.9	35.6	27.9	30.7
4) RES-SRL445 (2% v/v)	39.0	26.5	37.1	34.6	34.3
5) RES-SRL444 + 445 (10% and 2% v/v, respectively)	37.1	35.1	29.6	34.5	34.1

[1/] See Appendix 5A for total tuber yield harvested from each treatment plot. A tuber sample (ca 50 lbs) from each treatment plot was graded to determine the proportion of each grade within the sample.

Appendix 5 (continued).

Treatment	Rep. I	Rep. II	Rep. III	Rep. IV	Mean
Appendix 5G: The proportion (percentage) of total yield discarded as culls.[1/]					
1) Control (Non-treated)	1.3	16.4	14.7	17.7	12.5
2) Bravo 500 (2 pt/A)	26.8	17.3	24.7	13.6	20.6
3) RES-SRL444 (10% v/v)	9.2	12.9	12.8	13.3	12.0
4) RES-SRL445 (2% v/v)	14.0	18.5	9.6	7.6	12.4
5) RES-SRL 444 + 445 (10% and 2% v/v, respectively)	17.0	13.4	11.9	12.3	13.6

[1/] See Appendix 5A for total tuber yield harvested from each treatment plot. A tuber sample (ca 50 lbs) from each treatment plot was graded to determine the proportion of each grade within the sample.

Station: Center SW | County: Rio Grande | State: CO | Date (Month & yr): May 1982 | Time of Observation: 8A | Standard Time in Use: MDT

RECORD OF CLIMATOLOGICAL OBSERVATIONS

G D FRANC

DATE (MAY)	TEMPERATURE °F: 24 Hours Ending at Observation Max.	Min.	At Observation Dry-bulb	Wet bulb	Dew Point	Supplemental Readings at ____ Dry-bulb	Wet bulb	Dew Point	WATER TEMP. °F: 24 Hours Ending at Observation	Soil Temperature: Type of ____ Time of Beginning 2"	Time of Ending 2"	Time of Beginning 6"	Time of Ending 6"	24 Hour Amounts: Rain, Melted snow, etc. (in. & hundredths)	Snow, Ice Pellets, Hail (in. & tenths)	At Obsn. Snow, Ice Pellets Hail, Ice on ground (in.)	WIND: Anemometer Dial Reading (Miles)	24 Hour Movement	EVAPORATION (Inches & hundredths): Gage Reading or Amount Added +	Reading When Tank Filled or Amount Removed −	Amount of Evaporation	ADDITIONAL DATA/REMARKS
									Solar Rad.	Max	Min	Max	Min									
1	64	30	39						521.16	63	46	52	51	.06				93.1				
2	65	31	42						507.40	65	46	53	51					94.4				
3	64	35	47						436.88	65	49	54	52					87.0				
4	65	30	45						345.72	63	49	54	54					88.4				
5	54	36	41						378.40	60	48	54	54					147.7				
6	54	20	33						610.60	60	43	54	52	.02				118.5				
7	59	22	34						564.16	63	43	51	51					112.7				
8	64	28	44						576.20	63	43	52	51					149.2				
9	63	25	47						657.04	63	46	52	51					148.8				
10	62	31	43						684.56	65	48	54	53					150.1				
11	60	26	43						579.64	68	50	57	54					124.7				
12	51	26	42						295.84	66	50	56	55					128.7				
13	46	27	32						333.68	55	46	54	52	.13	1" snow			149.5				
14	58	25	39	Planting Date					681.12	53	42	52	50	.01				141.7				Alfalfa Field Planted
15	59	27	37						481.60	65	42	52	50					163.0				
16	58	29	38						478.16	60	46	52	52					73.4				
17	58	25	39						288.96	62	46	53	51	.02				83.3				
18	65	25	41						507.40	60	45	52	51	.03				99.6				
19	65	26	45						692.84	63	45	53	52					151.1				
20	68	27	43						703.48	67	49	55	54					80.0				
21	69	30	45						681.12	72	48	57	54					75.5				
22	63	33	44						349.16	74	52	59	57					86.6				
23	72	34	42						522.88	64	52	57	57					77.9				
24	66	32	47						500.52	71	55	58	57					71.3				
25	67	32	39						490.20	71	55	58	58	.05				83.2				
26	77	33	45						782.60	71	55	58	58					50.3				
27	65	35	51						447.20	76	55	61	58					98.6				
28	70	34	51						715.52	65	53	60	60	Trr.				126.6				
29	68	31	42						755.08	70	50	59	58					98.4				
30	66	36	52						665.64	70	51	60	58					134.0				
31	69	32	43						749.92	68	50	60	57					57.0				
Sum	1954	913	1315						16974.68					.32				3344.3				
Avg.	63.0	29.5	42.4						547.57				Greatest	.13					Adjusted Total			

OBSERVER: SCV Res Ctr. | STATION: Center 4SSW | DATE (Month and Year): May 1982

WS FORM E-22 (4-76)

U.S. DEPARTMENT OF COMMERCE
NOAA
NATIONAL WEATHER SERVICE

Center) SW | County Rio Grande | State CO | Date (Month & year) June 8 | of Complete Observation time) 8A | Standard Time in Use MDT

RECORD OF AND CLIMATOLOG) ATION ERVATIONS

G D FRANC

ADDITIONAL DATA/REMARKS

DATE JUNE	TEMPERATURE °F: 24 Hours Ending at Observation Max.	Min.	At Observation Dry-bulb	Wet-bulb	Dew Point	Supplemental Readings at ____ Dry-bulb	Wet-bulb	Dew Point	WATER TEMP. °F 24 Hours Ending at Observation Max.	Min.	Soil Temperature Time of beginning 2" Max	Time of ending 2" Min	Time of beginning 6" Max	Time of ending 6" Min	PRECIP. 24 Hour Amounts Rain, Melted snow, etc. (in. & hundredths)	Snow, Ice Pellets, Hail (in. & tenths)	At Obsn. Snow, Ice Pellets, Hail, Ice on ground (in.)	WIND Anemometer Dial Reading (Miles)	24 Hour Movement	EVAPORATION (Inches & hundredths) Gage Reading or Amount Added +	Reading When Tank Filled or Amount Removed −	Amount of Evaporation
1	68	30	47						737	88	72	51	61	58					164.0			
2	68	30	49						727	56	72	52	61	58					70.0			
3	67	33	48						545	24	73	53	62	48					33.6			
4	70	30	48						718.	96	68	55	60	60					63.4			
5	69	35	46						760.	24	75	54	61	60					188.0			
6	68	34	48						689.	72	72	54	61	61					112.3			
7	70	28	46						743.	04	74	55	60	60					89.2			
8	71	34	53						780.	88	75	54	62	62					148.8			
9	70	30	47						744.	76	75	40	63	43					59.7			
10	71	28	46						694.	88	70	40	61	40					84.1			
11	73	35	53						550.	40	76	50	57	55					57.5			
12	73	33	35						671	40	67	48	58	55					41.4			
13	73	34	53						473.	00	68	51	58	56					29.4			
14	72	33	35						645.	64	67	50	59	56					75.9			
15	68	34	48						512.	56	68	51	57	57					43.2			
16	70	33	43						579.	64	68	52	59	57	.02				72.7			
17	71	38	53						552.	12	70	52	61	58	.04				58.0			
18	72	37	47						502.	24	70	55	61	60					68.6			
19	63	33	41						522.	88	70	54	61	59					48.9			
20	68	32	48						516.	84	66	53	61	59					48.4			
21	69	34	48						564.	16	71	52	61	59					81.5			
22	74	38	48						474.	72	70	51	64	57					25.4			
23	72	36	46						608.	88	68	50	60	57					43.6			
24	76	35	50						646.	72	71	51	61	58					15.7			
25	77	37	51						605.	44	73	53	62	60					29.9			
26	77	38	49						650.	16	72	53	63	61					45.4			
27	78	36	54						696.	60	74	54	63	61					18.3			
28	84	40	56						705.	20	77	56	65	61					56.7			
29	81	40	55						550.	40	79	58	66	63					23.2			
30	74	42	50						416.	24	77	60	66	65					28.5			
31																						
Sum	2159	1050	1441						18696	40					.06				1425.3			
Avg.	71.9	34.3	48.0						623	21					Greatest .04					Adjusted Total		

OBSERVER SLV Research Center | STATION Center 4 SSW | DATE (Month and Year) June 1982

WS FORM E-22 (4-76)

U.S. DEPARTMENT OF COMMERCE NOAA NATIONAL WEATHER SERVICE

Station: Center 4 SW | County: Rio Grande | State: CO | Date (Month & year): July 1982 | Time of Complete Observation: 8 A | Standard Time in Use: MDT

RECORD OF CLIMATOLOGICAL OBSERVATIONS

G D FRANC

DATE July	TEMPERATURE °F: 24 Hours Ending at Observation Max.	Min.	At Observation Dry-bulb	Wet-bulb	Dew Point	Supplemental Readings at ____ Dry-bulb	Wet-bulb	Dew Point	WATER TEMP. °F: 24 Hours Ending at Observation	Solar Rad.	PRECIPITATION: Soil Temperature 2 in Time of beginning	Time of ending	6 in Time of beginning	Time of ending	24 Hour Amounts: Rain, Melted snow, etc. (in. & hundredths)	Snow, Ice Pellets, Hail (in. & tenths)	At Obsn. Snow, Ice Pellets, Hail, Ice on ground (in.)	WIND: Anemometer Dial Reading (Miles)	24 Hour Movement	EVAPORATION (Inches & hundredths): Gage Reading or Amount Added +	Reading When Tank Filled or Amount Removed −	Amount of Evaporation	ADDITIONAL DATA REMARKS
											Max	Min	Max	Min									
1	75	38	55							620.92	70	59	66	65					55.2				
2	78	38	48							758.52	75	58	67	63					44.8				
3	80	38	54							743.04	80	57	68	64					31.9				
4	77	35	54							479.88	82	61	69	65					41.7				
5	73	41	44							739.60	75	61	67	66					77.6				
6	72	31	47							684.56	79	60	68	66					25.3				
7	73	34	46							476.44	81	60	68	67					60.8				
(8)	78	39	54	Application #1						677.68	75	62	68	68					41.9				
9	76	38	55							510.34	80	62	68	66					70.8				
10	79	36	45							787.76	79	60	68	67					63.9				
11	80	36	60							603.72	84	61	69	67					57.2				
12	81	39	56							660.48	84	64	70	68					33.3				
13	82	40	54							756.80	85	64	72	69					41.0				
14	80	39	55							249.40	85	65	73	69	.02				60.3				
15	81	34	53							705.20	80	60	71	69	.01				89.2				
16	80	43	58							669.08	83	49	73	51					22.0				
17	76	46	59							612.30	73	49	64	50	.08				61.8				
18	78	48	57							466.12	77	53	66	58					116.5				
(19)	80	47	55	Application #2						560.72	75	57	64	60	T				93.3				
20	83	44	56							681.12	77	57	65	62	.07				88.0				
21	84	47	60							672.52	79	58	67	63					57.5				
22	83	47	63							700.04	80	58	68	64					63.5				
23	80	49	62							694.88	81	60	70	67					65.0				
24	79	49	62							712.08	84	58	75	64					97.0				
25	80	49	62							586.52	80	59	69	65					99.4				
26	78	51	59							667.36	79	62	69	67					116.1				
(27)	74	54	56	Application #3						524.60	81	63	71	70	.09				70.4				Cool AM. Rained prior to application
28	75	56	59							436.88	78	64	69	69	.02				72.3				
29	74	54	59							407.64	79	65	69	69	.32				51.2				
30	72	54	55							538.36	79	65	69	69	.08				76.7				
31	77	46	60							674.24	79	62	69	68	.11				50.7				
Sum	2418		1340	1722						19659.30					.80				1996.3				
Avg.	78		43.2	55.5						634.82				Greatest	.32					Adjusted Total			

OBSERVER: SIV Research Center | STATION: Center 4 SSW | DATE (Month and Year): July 1982

WS FORM E-22 (4-76)

U.S. DEPARTMENT OF COMMERCE
NOAA
NATIONAL WEATHER SERVICE

Station: Center 2 W | County: Rio Grande | State: Co | Date (Month & yr): Aug. 198- | Time of Complete Observation: 8A | Standard Time in Use: MDT

RECORD OF AND CLIMATOLOGI ATION ERVATIONS

GD FRANC

DATE Aug.	TEMPERATURE °F: 24 Hours Ending at Observation Max.	Min.	At Observation Dry-bulb	Wet-bulb	Dew Point	Supplemental Readings at ____ Dry-bulb	Wet-bulb	Dew Point	WATER TEMP. °F: 24 Hours Ending at Observation	PRECIPITATION: Soil Temperature Time of beginning	Time of reading	Time of beginning	Time of reading	24 Hour Amounts: Rain, Melted snow, etc. (in. & hundredths)	Snow, Ice Pellets, Hail (in. & tenths)	At Obsn. Snow, Ice Pellets Hail, Ice on ground (in.)	WIND: Anemometer Dial Reading (Miles)	24 Hour Movement	EVAPORATION (Inches & hundredths): Gage Reading or Amount Added +	Reading When Tank Filled or Amount Removed −	Amount of Evaporation	ADDITIONAL DATA/REMARKS
									Solar Rad.	Max	Min	Max	Min									
1	79	47	58						565.88	76	61	69	68					54.5				
2	76	51	56						534.92	62	60	71	68	.20				107.3				
3	74	47	56						503.96	78	62	70	68					109.9				
4	77	52	57						541.80	78	63	70	68	.01				83.5				
5	78	48	58						564.16	77	62	70	68	T				84.9				
(6)	79	48	59	Application #4					734.44	78	62	68	68	T				49.3	Cool, sunny. Irrigated after.			
7	75	50	53						421.40	79	64	69	69	T				92.1				
8	75	44	58						464.40	76	62	68	68	.02				58.0				
9	75	52	54						684.56	78	62	69	67					124.6				
10	79	44	55						368.08	78	64	69	69					94.8				
11	74	49	55						545.24	78	63	69	69					87.8				
12	79	49	52						540.08	79	65	71	69	.05				86.2				
13	75	51	60						440.20	79	67	71	70	.02				95.6				
14	78	49	52						607.16	77	65	71	70					113.3				
15	79	51	64						402.48	81	65	72	70					62.1				
16	76	49	56						517.72	78	64	71	70	.18				62.2				
(17)	78	50	54	Application #5					555.56	79	63	71	68	.34				87.3				
18	79	50	55						495.36	79	63	70	68					97.2				
19	79	53	55						519.44	78	64	70	68	.10				96.4				
20	80	53	61						522.88	78	64	70	69	.02				87.6				
21	76	52	54						529.76	78	64	70	68	.17				91.4				
22	76	53	56						475.36	79	64	70	69	.54				84.2				
23	68	55	57						235.64	78	64	70	69	.08				66.8				
24	64	53	55						194.36	70	63	69	68	.04				67.0				
25	68	51	55						373.24	66	61	68	66	.26				55.0				
(26)	69	47	55	Application #6					465.92	71	61	66	65	T				128.2	Ptly. cloudy during application.			
27	72	45	52						430.00	70	59	65	65	T				117.9				
28	74	45	48						600.28	71	58	64	64					90.5				
29	78	44	60						519.44	73	58	64	64					115.5				
30	74	50	58						352.60	75	58	66	64					123.9				
31	78	44	59						462.68	70	60	65	65					117.1				
Sum	2341	1526	1737						15179.00									2792.				
Avg.	75.5	49.2	56.0						489.65					Greatest	2.03				Adjusted Total			

OBSERVER: [illegible] Research Center | STATION: [illegible] U.S.C. | DATE (Month and year): AUG. 1982

WS FORM E-22 (4-76) | U.S. DEPARTMENT OF COMMERCE NOAA

Station: Center 4SSW | County: Rio Grande | State: Co | Date (Month & yr.): Sept 82 | Time of Complete Observation (Local time): 8A | Standard Time in Use: MDT

RECORD OF EVAPORATION AND CLIMATOLOGICAL OBSERVATIONS

DATE Sept	AIR TEMPERATURE °F: 24 Hours Ending at Observation Max.	Min.	At Observation Dry-bulb	Wet-bulb	Dew Point	Supplemental Readings at ____ Dry-bulb	Wet-bulb	Dew Point	WATER TEMP. °F	Solar Rad.	PRECIPITATION: Soil Temperature Time of beginning (2) Max	Time of ending Min	Time of beginning (6) Max	Time of ending Min	24 Hour Amounts: Rain, Melted snow, etc. (in. & hundredths)	Snow, Ice Pellets, Hail (in. & tenths)	At Obsn. Snow, Ice Pellets, Hail, Ice on ground (in.)	WIND: Anemometer Dial Reading (Miles)	24 Hour Movement	EVAPORATION (Inches & hundredths): Gage Reading or Amount Added +	Reading When Tank Filled or Amount Removed −	Amount of Evaporation	ADDITIONAL DATA/REMARKS
1	80	48	55							571.04	74	60	65	65	.01				113.2				
2	80	45	56							528.04	76	60	65	65					73.4				
3	78	42	53							364.64	78	61	66	65					95.4				
4	77	43	58							299.28	73	60	67	66					87.7				
5	72	43	54							374.96	71	61	66	65	.02				101.8				
6	71	42	45							481.60	69	57	66	65	.07				141.5				
7	70	39	49							357.76	71	57	63	63					92.6				
8	70	43	48							359.48	68	57	63	63	.20				56.2				
9	72	44	48							397.32	68	57	62	62	.01				76.0				
10	74	41	48							517.72	71	55	62	62	.01				91.5				
11	62	45	57							256.28	73	57	62	62	.11				89.9				
12	58	46	48							197.80	65	56	63	62	.50				68.9				
13	54	41	45							201.24	61	54	60	60	.32				91.1				
14	63	31	39							519.44	57	51	59	58	.04				86.0				
15	70	38	45							634.68	63	56	58	57					105.3				
16	72	36	44							459.24	64	52	58	58	T				84.8				
17	69	43	48							344.00	67	52	60	58	.23				78.3				
18	60	42	50							127.28	64	55	60	59	.02				58.8				
19	63	33	50							311.32	61	51	59	57	.11				70.5				
20	61	43	45							258.00	62	53	58	56	.24				76.1				
21	67	36	42							500.52	61	51	57	57	.01				75.9				
(22)	72	37	46							481.60	65	51	59	57					95.9				Harvest Date
23	75	41	48							469.56	66	50	59	57					112.3				
24	76	42	49							500.52	67	50	60	58					89.0				
25	71	40	56							295.84	67	53	60	58					48.9				
26	73	48	60							466.12	65	56	60	58					157.0				
27	64	44	52							466.12	68	55	61	59					241.7				
28	53	31	39							450.64	64	51	60	59					181.8				
29	66	28	38							466.12	61	47	58	56					143.1				
30	65	38	45							230.48	63	47	56	55					187.3				
31																							
Sum	2058	1213	1460							12347.88					1.90				3071.9				
Avg.	68.6	40.4	48.7							411.60					Greatest .50					Adjusted Total			

OBSERVER: IV Research Center | STATION: Center 4SSW | DATE (Month and Year): Sept 1982

WS FORM E-22 (4-76)

U.S. DEPARTMENT OF COMMERCE NOAA NATIONAL WEATHER SERVICE

EARLY BLIGHT RESISTANCE FIELD TRIALS

G.D. Franc, M.D. Harrison and K.W. Knutson

Materials and Methods

A grower-cooperator's field near Wiggins, Colorado was selected for the study. The field had a sandy-loam soil type and was located in an area with an elevation of ca 4,500 - 5,000 feet msl (mean sea level). The study plot was planted by hand on May 28 into previously hilled rows within a commercial field planted ca 7-10 days earlier with cv Monona potatoes. The field was irrigated with a center pivot overhead sprinkler system. The entire field was pre-irrigated (2 in) before plowing. Sprinkler irrigation was initiated on June 28 and ca 2 in of water per week was applied through July and the first week of August, 1 3/4 in per week the remainder of August, and 1 in per week during the first two weeks of September. In addition, considerable rainfall fell before and during the growing season. The study plots received the same fertilizer, fungicide and pesticide applications as the commercial field. The chemical applications and dates applied are listed in Tables 1 and 2.

The clones to be tested were planted in a randomized complete block design of four replications and 16 clones. Seedpieces were cut by hand shortly before planting in a single row at ca 12 inch spacings. For clones numbered 1-6 and 7-16, 10 and 12 seedpieces per replication were planted, respectively. The plot design is illustrated in Figure 1.

The study plots were observed regularly during the growing season. Plants were damaged by hail on June 14 [estimated percentage of defoliation observed in the surrounding field (planted May 11) was 19-25%] and hail damage was still evident in the study plot on July 1 when plants were ca 12 in tall. On July 12,

Table 1. Fertilization program superimposed upon early blight resistance plots - Wiggins, CO, 1982.[1/]

Date	Item
	Potash - dry. 120 lb available per acre plowed down.
5/10/82	Starter used at planting time was at the rate of 10 gal per acre (Analysis: 8-20-8).
6/11/82	Side dress with liquid fertilizer (32%) (90 lb available nitrogen per acre).
6/28/82	Liquid fertilizer (32%) added through center pivot sprinkler system (20 lb available nitrogen per acre).
7/1/82	Foliar application of materials with a ground rig. Per acre: 2 gal analysis 18-5-5 1/2 pt zinc 1 oz copper 1 oz solubar 1 pt calcium 1 pt magnesium 1 pt manganese 1 1/2 pt Bravo 500 (fungicide)
7/7/82	Liquid fertilizer (32%) and 12-0-0-26 added through center pivot sprinkler system (20 lb available nitrogen per acre and 12 lb of sulfur added).
7/10/82	Same as 7/7/82.
7/13/82	Same as 7/7/82.
7/17/82	Liquid fertilizer (32%) added through center pivot sprinkler system (20 lb available nitrogen per acre).
7/21/82	Same as 7/17/82
7/24/82	Same as 7/17/82.
8/17/82	Same as 7/17/82.

[1/] Information provided by the grower-cooperator.

Table 2. Chemical spray program superimposed upon early blight resistance plots, Wiggins, CO, 1982.[1/]

Date	Item
6/11/82	1 1/2 qt per acre of Di-syston applied with fertilizer.
6/27/82	3/4 pt per acre of Sencor 4 flowable.
7/1/82	1 1/2 pt Bravo 500 per acre.
7/14/82	5.3 oz Pydrin, 1 1/4 lb M-45 per acre.
7/31/82	5.3 oz Pydrin, 1 1/2 lb M-45 per acre.
8/16/82	5.3 oz Pydrin, 2 lb M-45 per acre.
8/31/82	5.3 oz Pydrin, 2 lb M-45 per acre.

[1/] Information provided by the grower-cooperator.

South

↑

Row:	1	2	3	4	5	6
	I-11	I-15	I-13	I-6	I-5	I-2
	I-7	I-10	I-9	I-12	I-14	I-16
	I-3	I-4	I-8	I-1	II-7	II-11
	II-13	II-6	II-3	II-10	II-16	II-15
	II-5	II-14	II-1	II-4	II-12	II-9
	II-8	II-2	III-11	III-10	III-2	III-3
	III-9	III-1	III-5	III-7	III-8	III-15
	III-14	III-12	III-6	III-16	III-4	III-13
	IV-10	IV-13	IV-7	IV-11	IV-9	IV-8
	IV-1	IV-2	IV-15	IV-6	IV-4	IV-3
	IV-14	IV-16	IV-5	IV-12	Blank	Blank

15 ft

165 feet

Figure 1. Plot design used for early blight resistance field trials - Wiggins, CO, 1982. Clonal designations used were: (1) FL-1471, (2) FL-1481, (3) FL-1312, (4) FL-1455, (5) FL-1449, (6) FL-1207, (7) BC9289-1, (8) WC-521-12, (9) Sangre, (10) WC-6722, (11) NY63, (12) NY67, (13) NY59, (14) Rosa, (15) Hudson, and (16) Katahdin (standard). Four replications of each clone were included.

plants appeared to be very vigorous and many clones were flowering. Hail damage was not evident on July 12. Stand counts on July 1 and July 12 (Table 3) show that all clones produced very good plant stands despite hail damage and all plants had emerged by July 1.

Foliar early blight infection did not appear until after August 6 (estimated). This date was based on field observations, day-degrees accumulation and planting date. The severity of foliar early blight infection after August 6 was estimated by rating the percentage of leaflets infected, percentage of leaf area blighted and the percentage of defoliation observed in the plots.

The percentage of leaflets infected was estimated on August 18, August 24, September 1 and September 15. Visual ratings using the Barratt-Horsfall (BH) scale (0-11) were recorded. Disease ratings on each date consisted of three (four on August 18) separate BH ratings per clone per replication for each the top, middle and bottom third of the plant canopy. The percentage of leaf area blighted was estimated on September 15. Three BH scale ratings for each clone per replication were made. The percentage of defoliation observed in the plots was determined on September 15 and 29. Two BH scale ratings per replication for each clone were made on each date.

Tubers were harvested from replications one and two on September 29 using a single row potato digger. Approximately 30 lbs per clone were harvested from each replication and stored in the event they would be needed for additional data. No tuber data are included in this report.

All BH scale ratings of foliar disease severity were averaged for each clone for each replication and analyzed in a two-way analysis of variance. Means were separated using Tukey's (HSD) test. Means were also separated using the Scott-Knott cluster analysis method (Gates, C.E. and J.D. Bilbro. Illustration of a cluster analysis method for mean separation. Agron. J. 70:462-465. 1978).

Table 3. Early blight resistance plot stand counts - Wiggins, CO, 1982.

Clone designation	Total Stand Count[1/2/]	
	July 1[3/]	July 12[4/]
1) FL-1471	9.0	9.5
2) FL-1481	9.3	9.5
3) FL-1312	9.3	9.5
4) FL-1455	9.5	9.8
5) FL-1449	9.0	9.3
6) FL-1207	9.8	10.0
7) BC9289-1	11.5	10.5
8) WC-521-12	11.3	10.8
9) Sangre	12.0	12.0
10) WC-6722	11.8	11.5
11) NY63	11.8	11.5
12) NY67	12.0	12.0
13) NY59	12.0	12.0
14) Rosa	11.8	11.3
15) Hudson	12.0	11.8
16) Katahdin (standard)	11.5	11.5

[1/] Entries represent the average stand count of four replications.

[2/] For clones numbered 1-6, 10 seedpieces per replication were planted. For clones numbered 7-16, 12 seedpieces per replication were planted.

[3/] Plants were ca 12 inches tall on July 1.

[4/] Most clones were starting or already flowering on July 12.

The Knott-Scott cluster analysis never resulted in overlapping mean separation groups and therefore facilitated grouping clones into distinct early blight resistance groups. For presentation of the data in this report, all BH scale ratings were converted to percentage values using the appropriate conversion table.

Figures depicting data for the estimated percentage of leaflets infected versus date of readings are also presented in the report to facilitate the comparison of foliar disease progression among clones. The disease curves for four clones were compared to Katahdin, the reference standard, on each graph.

Results

Results for the estimated percentage of leaflets infected are shown in Table 4. Significant differences between means occurred on all evaluation dates ($P \leq 0.01$). However, mean separation was not possible for data collected on August 18 when Tukey's test or the cluster analysis were used.

On August 24, clone "Rosa" had significantly fewer leaflets infected than clones FL-1455, "Sangre" and WC-6722 ($P \leq 0.01$). All other clones were statistically equivalent on this date. On September 1 clones FL-1449, "Sangre" and FL-1312 had the most leaflets infected while FL-1481, NY67 and "Rosa" had the least. Clones NY67 and "Rosa" had significantly fewer leaflets infected than FL-1449 and "Sangre" ($P \leq 0.01$). Clone FL-1481 had significantly fewer leaflets infected than clones FL-1312, FL-1449, "Sangre" and "Hudson" ($P \leq 0.01$). Clones FL-1471, FL-1481, WC-521-12, NY63, NY67 and "Rosa" all had significantly fewer leaflets infected than the most susceptible clone on September 1 (FL-1449) ($P \leq 0.01$).

On September 15, all clones had at least 96% of their leaflets infected. Clones FL-1471, FL-1312, FL-1449, FL-1207, BC9289-1, "Sangre," NY63, "Hudson" and "Katahdin" all had greater than 99% leaflets infected. Clones FL-1481,

Table 4. The estimated severity of early blight (Alternaria solani) infection on potato foliage - Wiggins, CO, 1982.

Clone Tested	Estimated percentage of leaflets infected[1/]											
	August 18[2/]			August 24			September 1			September 15		
1) FL-1471	1.2	a[3/]	1[4/]	2.5	ab	1	8.6	bcd	1	99.2	ab	1
2) FL-1481	1.4	a	1	2.9	ab	1	5.2	d	1	96.0	d	2
3) FL-1312	2.0	a	1	5.0	ab	1	19.3	abc	1	99.8	a	1
4) FL-1455	3.5	a	1	10.3	a	1	12.8	abcd	1	98.5	abcd	1
5) FL-1449	3.7	a	1	6.8	ab	1	23.9	a	1	99.4	a	1
6) FL-1207	1.8	a	1	2.8	ab	1	10.3	abcd	1	99.9	a	1
7) BC9289-1	2.3	a	1	3.0	ab	1	10.4	abcd	1	99.4	a	1
8) WC-521-12	3.0	a	1	3.0	ab	1	8.4	bcd	1	96.8	cd	2
9) Sangre	3.3	a	1	7.3	a	1	20.6	ab	1	99.9	a	1
10) WC-6722	3.8	a	1	7.8	a	1	14.5	abcd	1	98.8	abc	1
11) NY63	1.6	a	1	2.9	ab	1	8.3	bcd	1	99.5	a	1
12) NY67	1.2	a	1	3.0	ab	1	7.0	cd	1	98.4	abcd	1
13) NY59	1.6	a	1	2.9	ab	1	8.8	abcd	1	97.0	bcd	2
14) Rosa	1.2	a	1	1.6	b	1	7.0	cd	1	98.6	abc	1
15) Hudson	2.0	a	1	4.3	ab	1	15.8	abc	1	99.4	a	1
16) Katahdin	2.2	a	1	4.2	ab	1	12.0	abcd	1	99.6	a	1

[1/] Barratt-Horsfall scale (0-11) converted to percent leaflets infected.

[2/] Mean separation by Tukey's test did not occur.

[3/] Means with different letters differ significantly based on HSD (Tukey's) test ($P \leq 0.01$).

[4/] Means with different numbers differ significantly based on Scott-Knott cluster analysis ($P \leq 0.05$).

WC-521-12 and NY59 had 97% or fewer leaflets infected. Clone FL-1481 had significantly fewer leaflets infected than clones FL-1471, FL-1312, FL-1449, FL-1207, BC9289-1, "Sangre," WC-6722, NY63, "Rosa," "Hudson" and "Katahdin" ($P \leq 0.01$). Clone WC-521-12 had significantly fewer leaflets infected than clones FL-1471, FL-1312, FL-1449, FL-1207, BC9289-1, "Sangre," NY63, "Hudson," and "Katahdin" ($P \leq 0.01$).

The Knott-Scott cluster analysis only allowed mean grouping using data collected on September 15. Clones FL-1471, FL-1312, FL-1455, FL-1449, FL-1207, BC9289-1, "Sangre," WC-6722, NY63, NY67, "Rosa," "Hudson" and "Katahdin" were located in the more susceptible group (group #1, Table 4) and clones FL-1481, WC-521-12 and NY59 were located in the more resistant group (group #2, Table 4) ($P \leq 0.05$).

Data for the estimated percentage of leaf area blighted are shown in Table 5. Clones FL-1312, FL-1449, and "Hudson" had the greatest amount of leaf area blighted compared with clones FL-1481, WC-521-12, NY67 and NY59 which had the least. Clone FL-1481 had significantly less surface area blighted than clones FL-1471, FL-1312, FL-1449, FL-1207, BC9289-1, "Sangre," WC-6722, NY63, "Hudson" and "Katahdin" ($P \leq 0.01$). Clones FL-1312, FL-1449, "Sangre" and "Hudson" were all statistically equivalent and had a significantly greater percentage of leaf area blighted than clones FL-1481, WC-521-12, NY67 and NY59 ($P \leq 0.01$). Knott-Scott cluster analysis did not allow grouping of means ($P \leq 0.05$).

The amount of defoliation observed in the plots in September is shown in Table 6. On September 15, clones FL-1312, FL-1449, "Sangre" and "Hudson" had the greatest amount of defoliation. On September 29 (harvest date) the same clones plus clones FL-1207, BC9289-1, and "Katahdin" were highly defoliated. The clones with least amount of defoliation on both September 15 and September 29 were FL-1481, WC-521-12, NY67 and NY59. On September 15, clones FL-1481, WC-521-12,

Table 5. The estimated percentage of leaf area infected by early blight (Alternaria solani) - Wiggins, CO, 1982.

Clone Tested	Estimated percentage of leaf area blighted[1/]	
1) FL-1471	18.5 ab[2/]	1[3/]
2) FL-1481	4.5 c	1
3) FL-1312	31.0 a	1
4) FL-1455	13.0 abc	1
5) FL-1449	31.0 a	1
6) FL-1207	18.5 ab	1
7) BC9289-1	22.0 ab	1
8) WC-521-12	8.5 bc	1
9) Sangre	29.5 a	1
10) WC-6722	18.5 ab	1
11) NY63	19.5 ab	1
12) NY67	8.0 bc	1
13) NY59	8.0 bc	1
14) Rosa	11.5 abc	1
15) Hudson	33.0 a	1
16) Katahdin	22.0 ab	1

[1/] On September 15, 3 BH scale (0-11) ratings per clone for four replications were made.

[2/] Means with different letters differ significantly ($P \leq 0.01$) (Tukey's Test).

[3/] Means with different letter differ significantly ($P \leq 0.05$) (Scott-Knott cluster analysis).

Table 6. The estimated severity of early blight (Alternaria solani) infection on potato foliage - Wiggins, CO, 1982.

Clone Tested	Estimated percentage of defoliation observed[1/]			
	September 15		September 29	
1) FL-1471	8.5 abcd[2/]	1[3/]	93.5 abc	1
2) FL-1481	4.0 cd	1	22.0 e	2
3) FL-1312	31.0 a	1	99.0 a	1
4) FL-1455	16.0 abcd	1	81.5 bcd	2
5) FL-1449	28.0 a	1	98.5 a	1
6) FL-1207	18.5 abc	1	99.0 a	1
7) BC9289-1	22.0 ab	1	98.0 a	1
8) WC-521-12	6.0 bcd	1	47.5 de	2
9) Sangre	33.0 a	1	99.5 a	1
10) WC-6722	12.0 abcd	1	92.0 abc	1
11) NY63	15.0 abcd	1	98.0 ab	1
12) NY67	4.0 cd	1	47.5 de	2
13) NY59	3.0 d	1	61.5 cde	2
14) Rosa	9.0 abcd	1	97.5 ab	1
15) Hudson	28.0 a	1	99.5 a	1
16) Katahdin	11.5 abcd	1	99.5 a	1

[1/] Two BH scale (0-11) ratings were made per clone for four replications.
[2/] Means with different letters differ significantly (P≤0.01) (Tukey's Test).
[3/] Means with different letters differ significantly (P≤0.05) (Scott-Knott cluster analysis).

NY67 and NY59 had significantly less defoliation than clones FL-1312, FL-1449, "Sangre" and "Hudson" ($P \leq 0.01$). On September 29, clones FL-1481, WC-521-12, NY67 and NY59 had significantly less defoliation than clones FL-1312, FL-1449, FL-1207, BC9289-1, "Sangre," NY63, "Rosa," "Hudson" and "Katahdin" ($P \leq 0.01$).

Cluster analysis did not allow mean grouping on September 15 but on September 29 clones in the most defoliated group were FL-1471, FL-1312, FL-1449, FL-1207, BC9289-1, "Sangre," WC-6722, NY63, "Rosa," "Hudson" and "Katahdin." Clones in the least defoliated group were FL-1481, FL-1455, WC-521-12, NY67 and NY59 ($P \leq 0.05$). The data are depicted graphically in Figure 2.

The disease progress curves for the growing season are depicted in Figures 3-6. The estimated percentage of leaflets infected for each evaluation date (Table 4) for four clones was plotted in each figure as well as the disease progress curve for the Katahdin standard (used as a reference). The data show that foliar infection probably started in all clones simultaneously since infection was very low on August 18. Susceptible clones showed a more rapid increase in infection after this date, resulting in a disease progress curve of greater slope than the more resistant clones. The disease curves show that susceptible clones FL-1312 (Figure 3), FL-1449 (Figure 4) and "Sangre" (Figure 5) showed a marked increase in the percentage of leaflets infected between August 24 and September 1. Clones with greater levels of resistance FL-1481 (Figure 3), WC-521-12 (Figure 4) and NY67 (Figure 5) did not show such a rapid increase in the percentage of leaflets infected during this time and thus a curve with a flatter slope resulted.

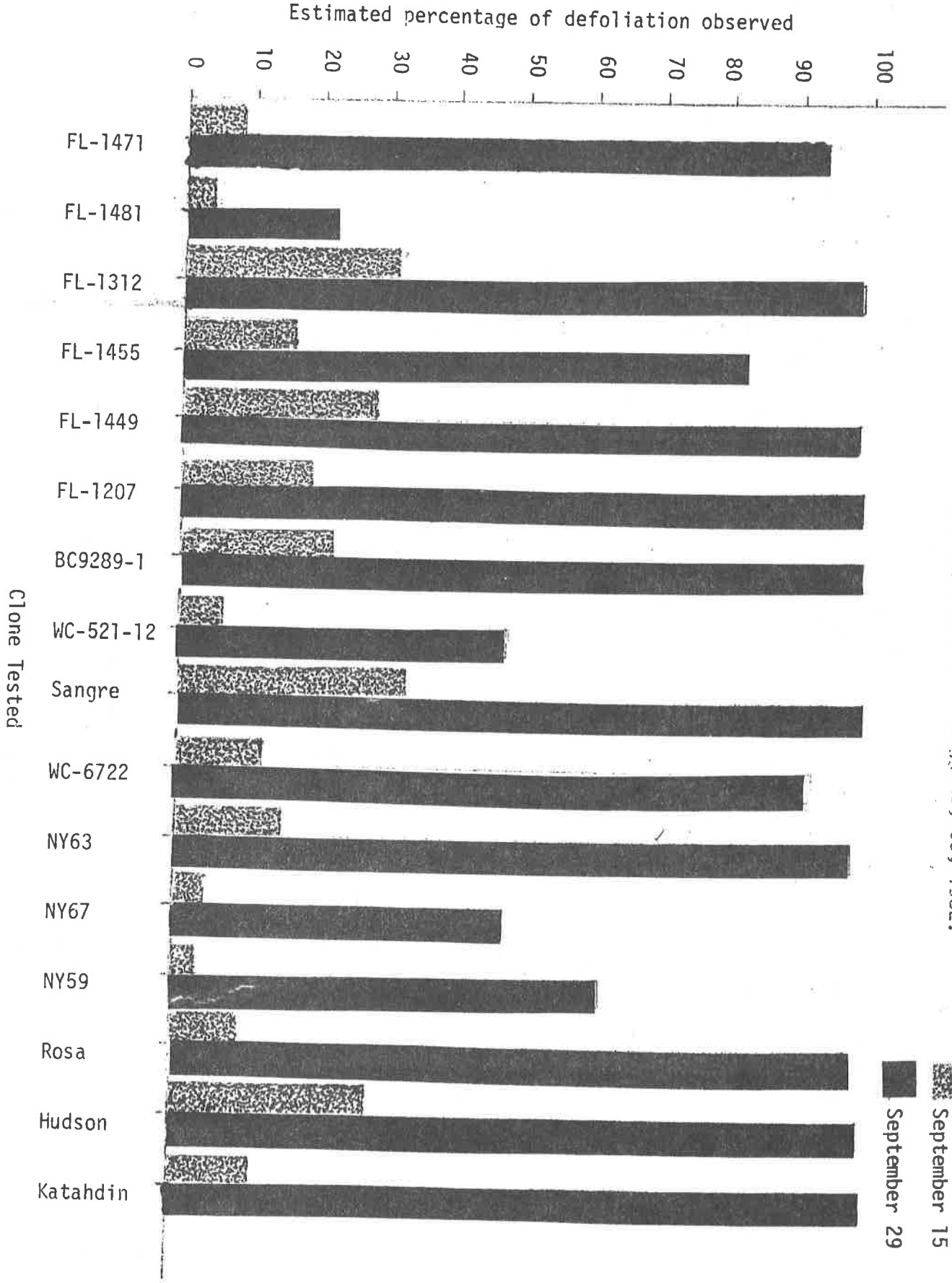


Figure 2. The estimated percentage of defoliation observed in early blight clone testing field trials on September 15 and September 29 - Wiggins, CO, 1982.

Figure 3:

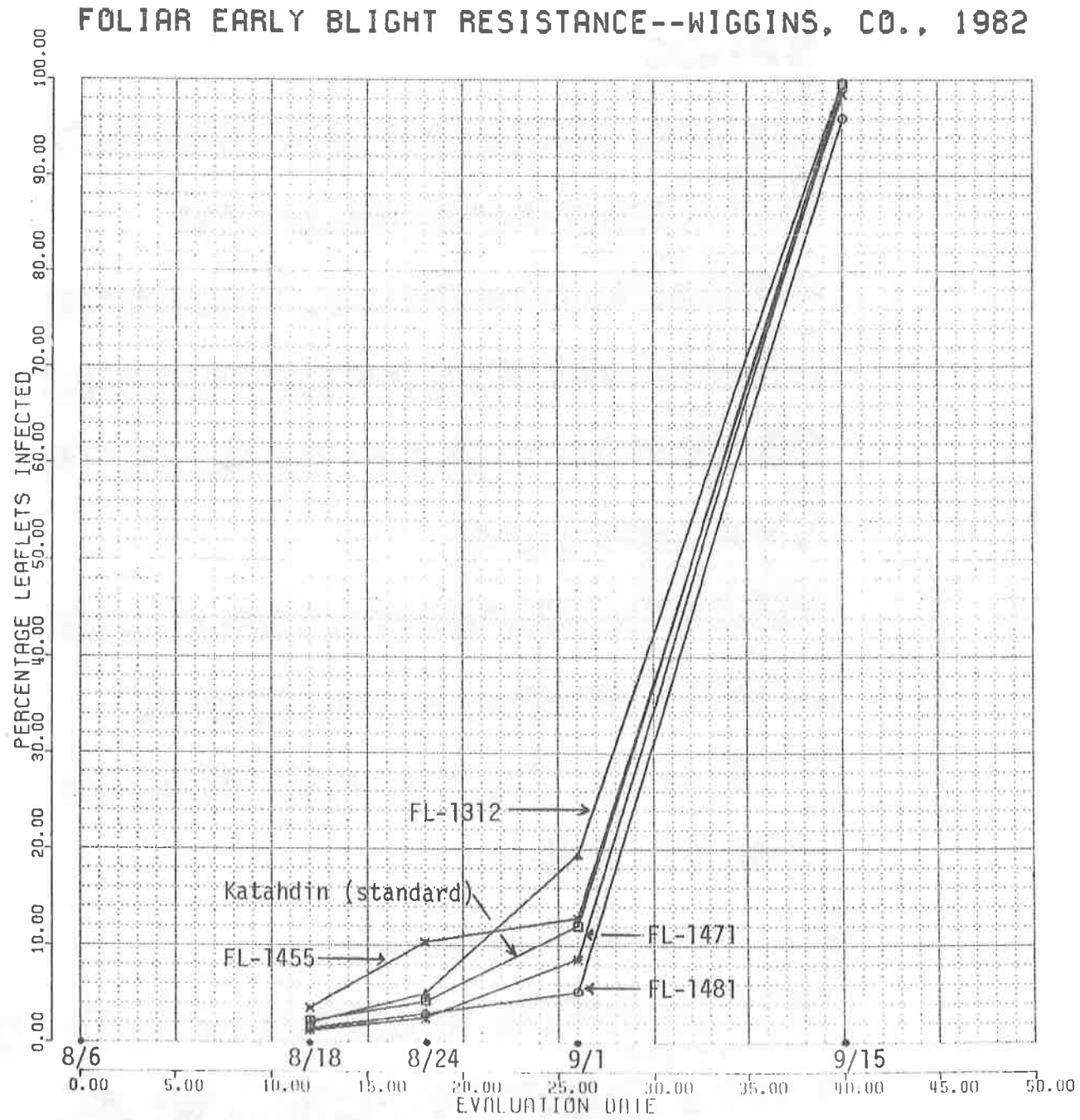

FOLIAR EARLY BLIGHT RESISTANCE--WIGGINS, CO., 1982
PERCENTAGE LEAFLETS INFECTED
0.00
10.00
20.00
30.00
40.00
50.00
60.00
70.00
80.00
90.00
100.00
FL-1312
Katahdin (standard)
FL-1471
FL-1455
FL-1481
8/6
8/18
8/24
9/1
9/15
0.00
5.00
10.00
15.00
20.00
25.00
30.00
35.00
40.00
45.00
50.00
EVALUATION DATE

Figure 4:

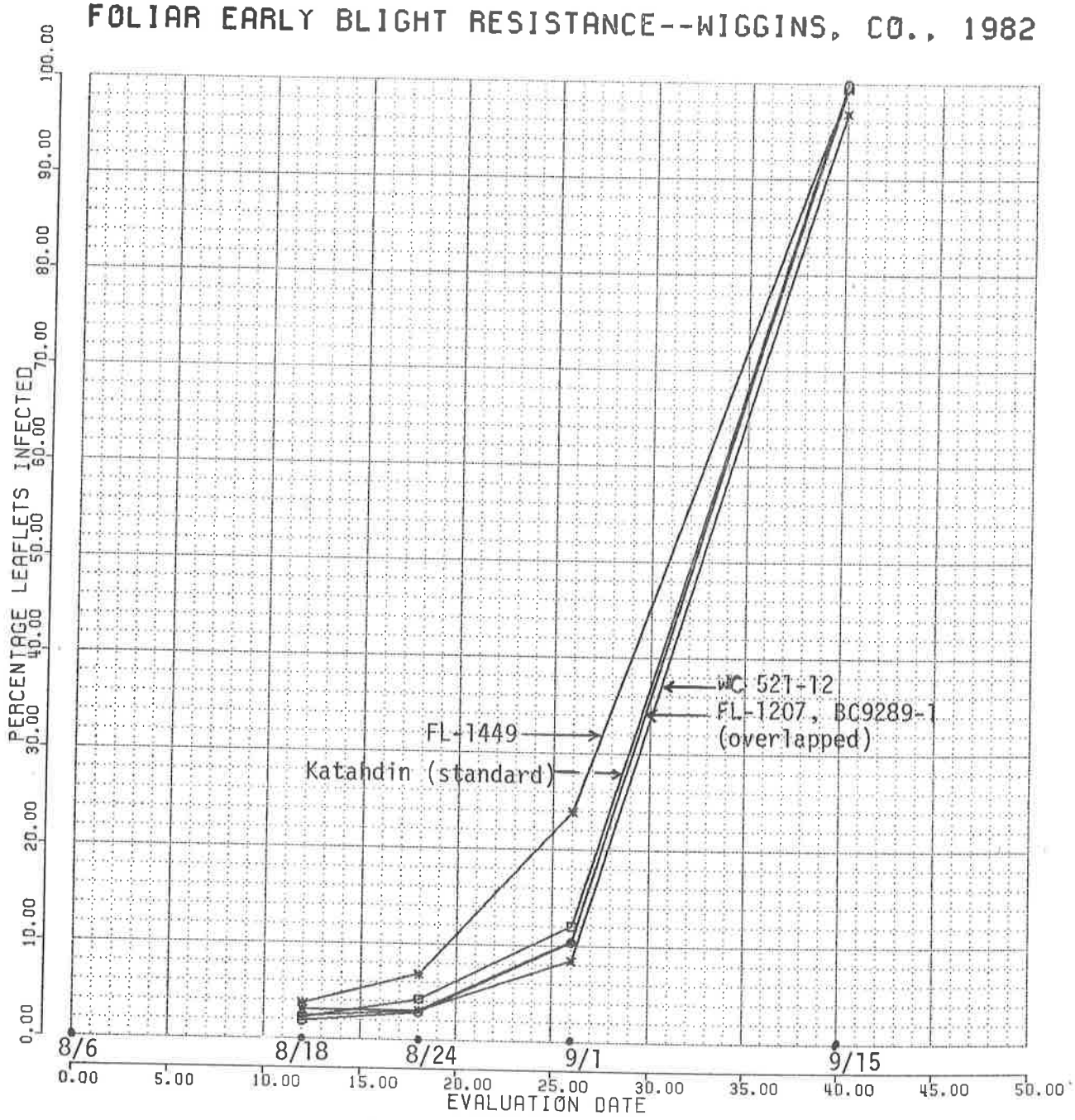
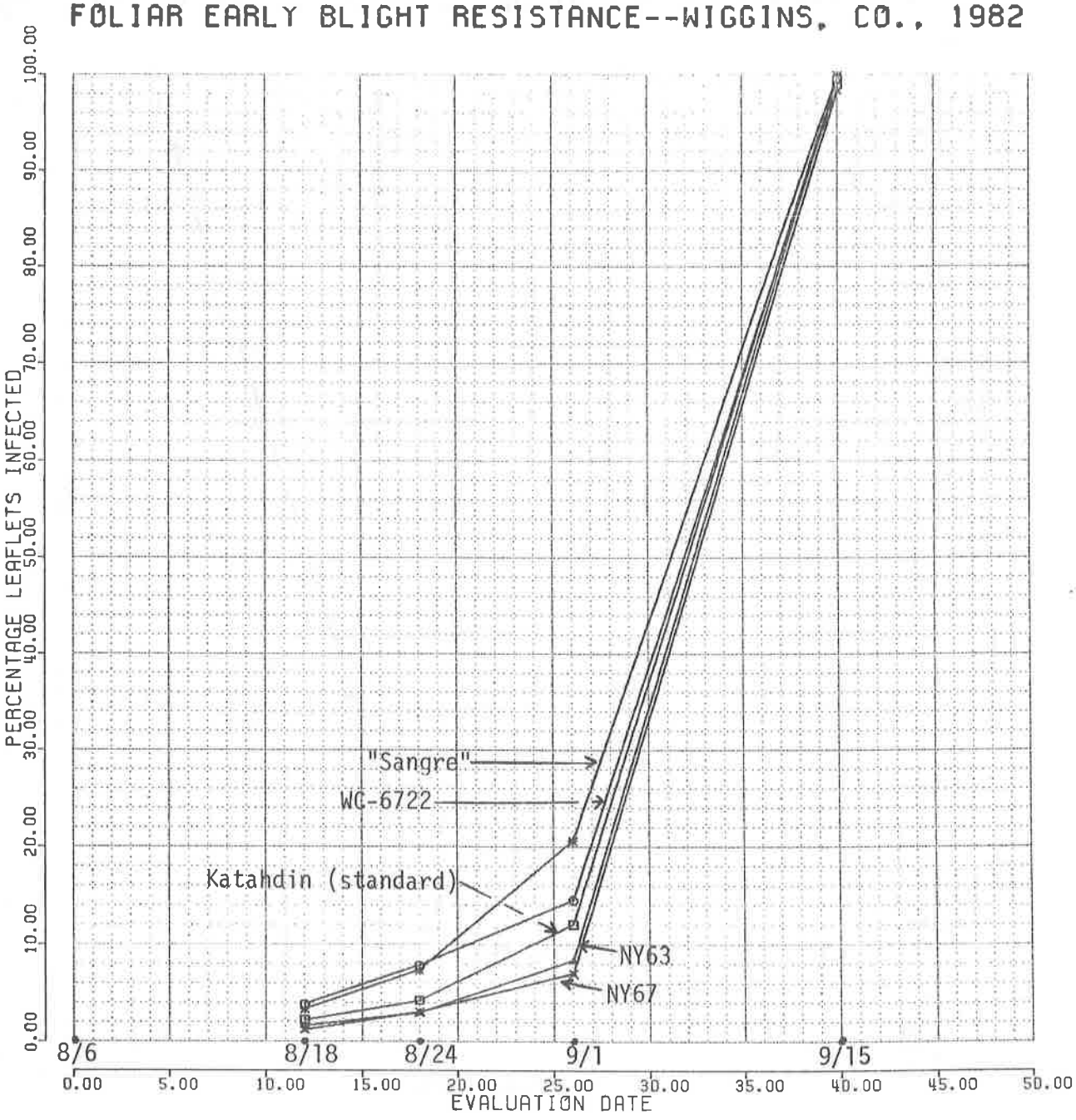

FOLIAR EARLY BLIGHT RESISTANCE--WIGGINS, CO., 1982
PERCENTAGE LEAFLETS INFECTED
0.00
10.00
20.00
30.00
40.00
50.00
60.00
70.00
80.00
90.00
100.00
8/6
8/18
8/24
9/1
9/15
0.00
5.00
10.00
15.00
20.00
25.00
30.00
35.00
40.00
45.00
50.00
EVALUATION DATE
WC 521-12
FL-1207, BC9289-1
(overlapped)
FL-1449
Katahdin (standard)

Figure 5:


FOLIAR EARLY BLIGHT RESISTANCE--WIGGINS, CO., 1982
PERCENTAGE LEAFLETS INFECTED
0.00
10.00
20.00
30.00
40.00
50.00
60.00
70.00
80.00
90.00
100.00
"Sangre"
WC-6722
Katahdin (standard)
NY63
NY67
8/6
8/18
8/24
9/1
9/15
0.00
5.00
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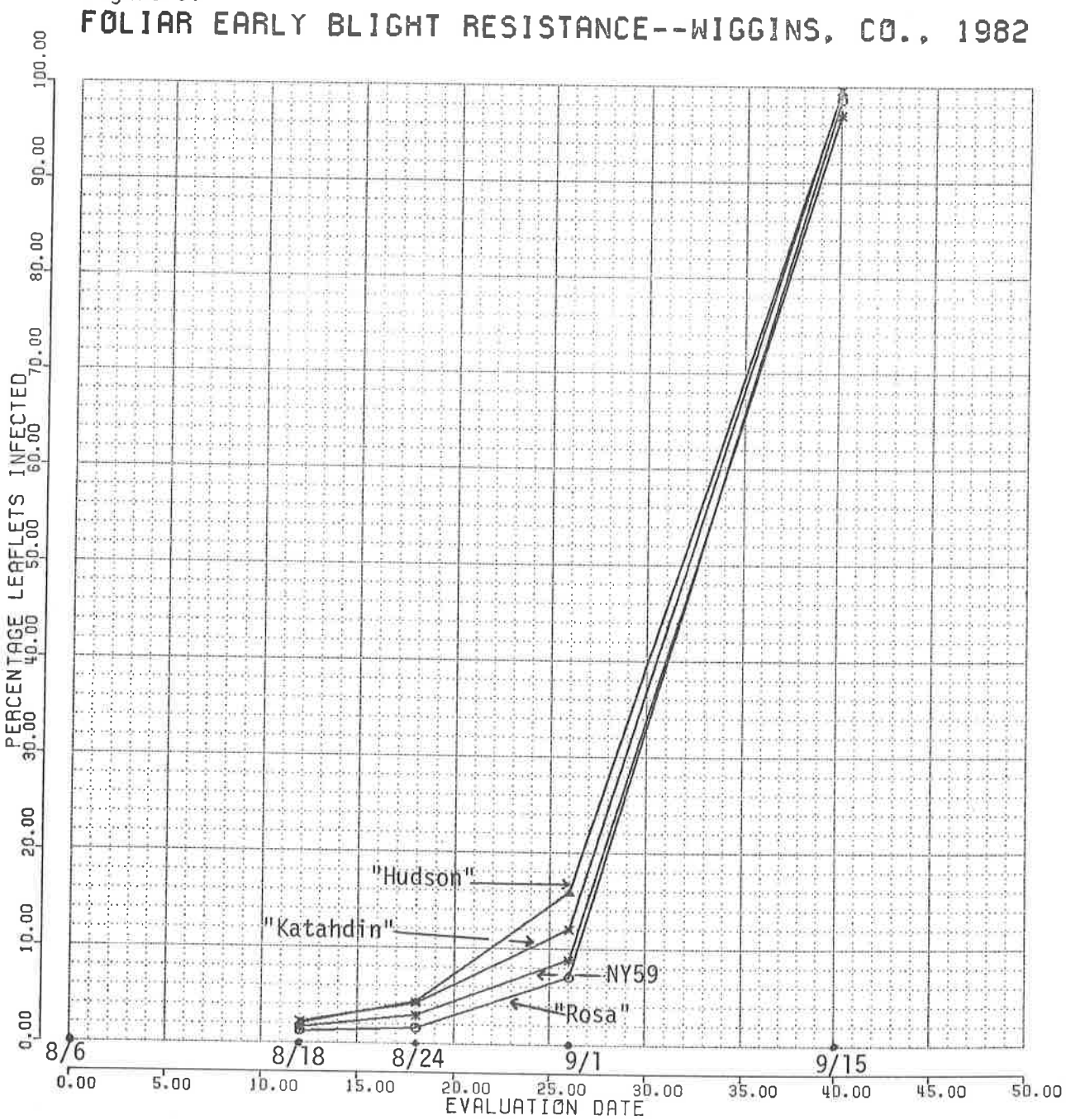
50.00
EVALUATION DATE

Figure 6:


FOLIAR EARLY BLIGHT RESISTANCE--WIGGINS, CO., 1982
PERCENTAGE LEAFLETS INFECTED
0.00
10.00
20.00
30.00
40.00
50.00
60.00
70.00
80.00
90.00
100.00
"Hudson"
"Katahdin"
NY59
"Rosa"
8/6
8/18
8/24
9/1
9/15
0.00
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45.00
50.00
EVALUATION DATE

Discussion

Clones FL-1481, FL-1455, WC-521-12, NY67 and NY59 were consistently more resistant to the effects of early blight, especially late in the growing season when disease pressure was greatest. However, since early blight is a disease of mature plants, disease resistance may simply reflect delayed maturity and, conversely, susceptibility may reflect early maturity. Observations in northeastern Colorado have consistently suggested that the amount of defoliation observed in the field is proportional to the severity of early blight infection. Although clones FL-1455 and NY67 appeared to be relatively susceptible to early blight leaflet infection (Table 4) they were not readily defoliated. Since clones FL-1455 and NY67 were relatively heavily infected but not readily defoliated the data suggest that these clones are "tolerant" or resistant to the effects of early blight infection. These clones may possess a more useful type of field resistance than clones FL-1481, WC-521-12 and NY59. The type of early blight resistance possessed by clones FL-1481, WC-521-12, and NY59 may be due to plant maturity if they are late varieties. The data collected and included in this report does not allow one to make this distinction.

Early Blight Prediction Model

G.D. Franc and M.D. Harrison

Abstract

A study was conducted in the San Luis Valley and Morgan County during the 1980 season to 1) further verify the day degree model developed earlier and being used in the San Luis Valley and 2) to determine if the large difference in day degrees accumulated between planting and lesion appearance between the two areas reported in 1979 was the effect of potato variety or location. Field observations were continued in Morgan County during 1981 and 1982 to determine the day degree accumulation threshold required for initial early blight (*Alternaria solani*) lesion appearance in potatoes.

The number of day degrees accumulated from planting until the initial appearance of early blight lesions in Morgan County agreed quite closely for each year tested. The data suggested that it is possible to predict the initial appearance of early blight in both the San Luis Valley and Morgan County using the day degree model and threshold values determined for each location.

Fungicide treatment schedules initiated prior to the appearance of the first early blight lesions did not provide additional disease control. Use of the model in Colorado will enable proper timing of fungicide application and more efficient chemical usage.

Materials and Methods

Studies to test an early blight prediction model, based on post planting day-degree accumulation, were conducted over a period of several growing seasons in two environmentally different potato producing areas in Colorado.

Study plots in 1980 consisted of three potato cultivars, Russet Burbank, Norchip and Monona, planted at the San Luis Valley Research Center near Center, Colorado on May 13 and on a cooperator's farm in Morgan County near Wiggins, Colorado on May 10. Plots in the San Luis Valley consisted of four rows for each cultivar each 100 ft long replicated four times. Plots in Morgan County consisted of four rows each 13 ft long for each of six fungicide treatment schedules replicated two times. Plots in the San Luis Valley were not treated with fungicide.

In 1981, a cooperator's commercial potato field near Wiggins, Colorado (Morgan County) was again selected for the study. The area of the field containing the research plot was planted with the cultivar Monona on approximately May 11, 1981. The treatment plots were planted in a randomized complete block design including three replications of six fungicide application schedules.

Plots in 1982 were planted by the same cooperator as in 1980 and 1981 near Wiggins, Colorado. The cultivar Monona was planted with a commercial potato planter. Fungicide was applied by airplane according to the cooperator's schedule.

Hygrothermographs and microcomputers programmed to monitor post-planting day degree accumulation above 45°F were installed at each study location. During 1980, weather vane spore traps (two per location), using greased microscope slides as the collecting surface, were also installed at each location. The greased slides were collected three times per week, returned to Fort Collins, and examined microscopically for the presence of Alternaria solani spores.

The date of foliar early blight lesion appearance in the field in relation to day degree accumulation was determined by periodically examining plants and relating the first lesion appearance with the total day degree accumulation since planting.

In order to determine the relationship of early blight control programs to day degree accumulation, different fungicide application schedules were initiated

prior to and after the appearance of the initial lesions. These studies were made in 1980 and 1981 in Morgan County. Similar studies were done in the San Luis Valley prior to 1980 (data not presented). All spray schedules were initiated sequentially at approximately 10 day intervals. Once a treatment schedule was initiated, fungicide was applied regularly at 10 day intervals. Spray schedules used in 1980 and 1981 are shown in Tables 1 and 2, respectively. Bravo 500 at the rate of 2 pt/A was delivered at 70 p.s.i. in 101 gallons per acre in 1980 and at 60 p.s.i. in a net volume of 83 gallons per acre in 1981 using a tractor drawn sprayer. All fungicide treatments were superimposed upon the grower's own fungicide application scheme.

The relative effectiveness of each fungicide treatment schedule was determined by rating the early blight disease severity in each plot periodically during the growing season.

During 1980, the severity of foliar early blight infection was determined for each treatment by estimating the percentage of leaflets infected using the Barratt-Horsfall (BH) scale (0-11) and making five ratings per treatment per replication each for the top, middle and bottom third of the plant canopy.

The relative effectiveness of each treatment during 1981 was determined by measuring the severity of early blight in the field plots, estimating the amount of defoliation observed and counting the number of early blight lesions on harvested tubers.

The severity of early blight foliar infection in 1981 was determined for each treatment by (1) estimating the percentage of leaflets infected using the BH scale and taking 20 readings per replication each for the top, middle and bottom third of the plant canopy and (2) collecting 20 leaves from the top, middle and bottom third of the plant canopy. These leaves were returned to the laboratory and the average number of early blight lesions per leaflet was determined by direct counts.

Table 1. Bravo 500 spray schedules utilized to determine the effect of spray timing on early blight control in three potato varieties, Wiggins, Colorado, 1980.

Spray application schedule	Dates of fungicide application[1]							Total number of applications
	June 25	July 3	July 14	July 25	Aug. 6	Aug. 15	Aug. 25	
1	+	+	+	+	+	+	+	7
2	-	+	+	+	+	+	+	6
3	-	-	+	+	+	+	+	5
4	-	-	-	+	+	+	+	4
5	-	-	-	-	+	+	+	3
6	-	-	-	-	-	+	+	2
7	-	-	-	-	-	-	-	0

[1] Bravo 500 (2 pt/A) was applied on the dates indicated by (+) in a total volume of 101 gal/A at 70 p.s.i.

Table 2. Bravo 500 spray schedules utilized to determine the effect of spray timing on early blight control in Monona potatoes, Wiggins, Colorado, 1980.

Spray application schedule	Dates of fungicide application[1]							Total number of applications
	July 1	July 10	July 20	July 30	Aug. 10	Aug. 20	Aug. 31	
A	+	+	+	+	+	+	+	7
B	-	+	+	+	+	+	+	6
C	-	-	+	+	+	+	+	5
D	-	-	-	+	+	+	+	4
E	-	-	-	-	+	+	+	3
F	-	-	-	-	-	-	-	0

[1] Bravo 500 (2 pt/A) was applied on the dates indicated by (+) in a total volume of 83 gal/A at 60 p.s.i.

The amount of defoliation observed in the plots in 1981 was estimated by taking 10 readings per treatment per replication using the BH scale.

Tubers were harvested on September 18, 1981. Tuber samples (125 tubers per treatment per replication) were placed in cold storage for approximately 15 weeks and the average number of early blight lesions per tuber was determined by direct counts.

All BH scale data from 1980 and 1981 were converted to percentage infection and arcsin values assigned using the appropriate conversion tables. Arcsin values were analyzed in a two-way analysis of variance and means were separated using Tukey's test. For simplicity, percentage data are presented in tables. Leaflet lesion count data and tuber lesion counts were analyzed in the same manner and are presented in the tables.

Results

San Luis Valley, 1980:

Six hundred and fifty day degrees above 45°F (actually 652.3 degrees) was reached on July 16, 1980. This was the early blight threshold determined by previous studies. Reports of scattered early blight lesions in potato fields in the San Luis Valley by potato certification personnel, others visiting fields on a regular basis and observations by our research personnel verified that the earliest lesions appeared almost simultaneously (July 15-16) with the temperature threshold.

The first *A. solani* spores were found on the greased spore trap slides on July 25, 1980. Disease severity was rated low in all three potato cultivars on July 28, 1980, and increased progressively to the end of August when the last ratings were made. The data show that the level of infection in all three cultivars on July 28 was quite similar (0.2 - 1.0% of the leaflets infected), and suggest that disease appeared simultaneously in the three varieties. The data (Table 3) verify the findings from previous years, i.e., that the

Table 3. Results of the early blight monitoring and spray-timing study in the San Luis Valley - 1980.

Date 650 day degrees had accumulated	Date first lesions were observed in the field	Date first *A. solani* spores were detected on traps	Early blight disease severity (% leaflets infected)			
			Cultivar	July 29	Aug. 13	Aug. 26
July 16	July 15-16	July 25	Russet Burbank	0.9	13.0	72.0
			Norchip	1.0	13.0	76.5
			Monona	0.2	10.5	88.0

appearance of the first early blight lesions on potato foliage in the San Luis Valley corresponds to the accumulation of ca 650 day degrees above 45°F from the time of planting. Furthermore, the data indicate that this system works equally well for the three varieties included in the 1980 study.

Morgan County, 1980:

The first early blight lesions appeared in the plots between July 14 and July 25. No lesions were visible in the plots on July 14 but they were present on July 25. Very scattered lesions were found on July 7, in an area of the grower's field planted ca 10 days earlier than the study plots. Therefore, it can be assumed that first lesions probably appeared ca 10 days later in the study area, i.e., about July 16-17. Furthermore, the first spores were found on spore trap slides on July 25. In the San Luis Valley, lesions were present 9-10 days before the first spores were captured from the air. Extrapolation for the Morgan County area using this information again indicates that the first lesions probably appeared on about July 15-16 (coincidentally with the San Luis Valley).

Table 4. Results of early blight monitoring and spray-timing studies in Morgan County - 1980.

Approximate number of day degrees accumulated when first lesions appeared	Approximate date first lesions appeared	Date *A. solani* spores were detected on traps	Early Blight Disease Severity (% leaflets infected)			
			cultivar	July 28	Aug. 18	Sept. 2
1155	July 16	July 25	Russet Burbank	0.3	4.0	10.0
			Norchip	0.2	5.0	35.0
			Monona	0.1	3.0	21.0

If the above assumptions are valid and July 16 is assumed to be the most likely date that symptoms first appeared, then 1155 day degrees had accumulated from planting to the appearance of the first lesions. Disease expression appeared in the three cultivars at almost the same time since disease readings made on July 28 showed 0.3% of the leaflets infected in Russet Burbank, 0.2% in Norchip and 0.1% in Monona (Table 4).

These data support the results reported in 1979, i.e., that considerably more day degrees (ca 1125 in 1979) must accumulate in Northeastern Colorado from planting to the beginning of disease development than is the case in the San Luis Valley (650).

Spray Schedules in Relation to Disease Control, 1980:

The data (Table 5) show that spray schedules started on or before August 6 (Schedule 1-5) controlled early blight very effectively in all three potato cultivars regardless of the total number of chemical applications. Spray

Table 5. The effect of six different spray schedules on the severity of early blight infection in three potato cultivars - Morgan County, 1980.

Schedule	Dates fungicide was applied[1]			Early blight severity (% leaflets infected)[2]								
				Russet Burbank			Norchip			Monona		
				7/28	8/18	9/2	7/28	8/18	9/2	7/28	8/18	9/2
1	6/25 7/25 8/25	7/2 8/6	7/14 8/15	0.2	0.1	0.5	0.0	0.5	1.1	0.1	0.2	1.2
2	7/25 8/25	7/3 8/6	7/14 8/15	0.1	0.1	0.9	0.0	0.8	2.5	0.0	0.3	0.7
3	7/25 8/25	8/6	7/14 8/15	0.1	0.3	0.7	0.0	0.5	2.0	0.1	0.4	1.4
4	7/25 8/25	8/6	8/15	0.3	0.3	1.1	0.2	1.0	3.0	0.2	0.4	1.1
5	8/25	8/6	8/15	0.4	0.5	1.3	0.1	1.0	1.0	0.0	0.8	3.0
6	8/25		8/15	0.1	3.0	4.0	0.1	5.0	8.5	0.2	4.5	10.0
7	None (control)			0.3	4.0	10.0	0.2	5.0	35.0	0.1	3.0	21.0

[1] Bravo 500 was applied at a rate of 2 pt/A in a total volume of 101 gal/A.

[2] Barratt-Horsfall scale (0-11) converted to percentage.

schedules started after August 6 (Schedule 6) were less effective in controlling the disease although they reduced it considerably compared to control plots which received no fungicide applications.

These data agree quite well with the spore trapping data which indicate that sprays applied before July 25 (the date spores were detected in the air) would have served little purpose and those applied afterward showed progressively lower levels of control.

Morgan County, 1981 (spray schedules in relation to disease control):

Day degree accumulation from the date of planting to the approximate date of first early blight lesion appearance (ca May 11 and July 12, respectively) was 1056-1097[1/] day degrees.

Disease readings on July 28 (Table 6) show that fungicide applications initiated on July 1 and July 10 (treatments A and B, respectively) were statistically equivalent in terms of the estimated percentage of leaflets infected and offered significantly better control ($P \leq 0.01$) than fungicide applications initiated on or after July 20 (treatments C,D,E and F). On August 13, treatments initiated on July 1 and July 10 were again not significantly different and both provided significantly better control than treatments started on or after July 30 ($P \leq 0.01$). Treatment C, initiated on July 20 provided intermediate early blight control and was not significantly different from the July 10 and July 30 treatments. On September 1 there were no statistically significant differences among treatments and all had significantly less early blight than treatment F which received no fungicide ($P \leq 0.01$).

[1/] Exact planting date is uncertain.

Table 6. Effect of Bravo 500 fungicide application schedules on the estimated percentage of cv Monona leaflets infected by early blight, Wiggins, Colorado, 1981.

Treatment	Date of first fungicide application[1/]	Total number of applications	Estimated percentage of leaflets infected[2/]		
			July 28	August 13	Sept 1
A	July 1	7	0.02 e	16.3 d	81.1 b
B	July 10	6	0.14 e	27.9 cd	90.8 b
C	July 20	5	0.47 abc	30.7 bc	82.0 b
D	July 30	4	0.50 ab	41.3 b	85.0 b
E	August 10	3	0.43 abcd	68.5 a	89.8 b
F	None applied	0	0.52 a	79.5 a	99.8 a
			P≤0.01	P≤0.01	P≤0.01

[1/] Bravo 500 was applied at a rate of 2 pt/A in a total volume of 83 gal/A at 10 day intervals.

[2/] Barratt-Horsfall scale (0-11) converted to percentage.

Data from August 14 and August 21 for the average number of early blight lesions per leaflet (Table 7) show that treatment B, initiated on July 10, provided the same level of early blight control as all treatments initiated on or before July 30 (P≤0.01). Fungicide treatments initiated after August 10 or the nontreated control (treatments E and F, respectively) provided significantly less early blight control than treatment B. On August 21, the levels of early blight control offered by all treatments initiated on or prior to July 30 (treatments A,B,C and D) did not differ significantly (P≤0.01). On this date treatment E was significantly more effective than treatment F which received no fungicide treatment (P≤0.01).

Table 7. The effect of different Bravo 500 fungicide spray application schedules on the average number of early blight lesions per leaflet (cv Monona), Wiggins, Colorado, 1981.

Treatment	Date of first fungicide application[2/]	Total number of applications	Average number of early blight lesions/leaflet[1/]	
			August 4	August 21
A	July 1	7	0.301 d	2.072 c
B	July 10	6	0.908 cd	3.619 c
C	July 20	5	1.363 abc	2.495 c
D	July 30	4	0.910 cd	3.885 c
E	August 10	3	2.024 ab	7.578 b
F	None applied	0	2.044 a	15.980 a
			P≤0.01	P≤0.01

[1/] The average number of early blight lesions per leaflet was determined by actual lesion counts on collected leaflets.

[2/] Bravo 500 was applied at a rate of 2 pt/A in a total volume of 83 gal/A at 10 day intervals. The first early blight lesions occurred on approximately July 12 (1056 day degrees accumulated on this date).

All treatments, except treatment F, on September 1 were not statistically different in degree of defoliation from treatments A and B which were initiated on July 1 and July 10, respectively (P≤0.01) (Table 8).

Tuber lesion data in Table 8 show that the different fungicide treatments had no significant effect on the average number of early blight lesions per tuber (P≤0.05).

Table 8. Effect of Bravo 500 fungicide application schedules on the estimated percentage of defoliation observed in field plots and the average number of early blight lesions on harvested tubers (cv Monona), Wiggins, Colorado, 1981.

Treatment	Date of first fungicide application[1/]	Total number of applications	Estimated percentage of defoliation September 1, 1981[2/]	Average number of early blight lesions/tuber
A	July 1	7	54.6 bc	11.41 a
B	July 10	6	61.2 bc	13.50 a
C	July 20	5	48.2 c	8.60 a
D	July 30	4	60.6 bc	12.18 a
E	August 10	3	70.4 b	10.75 a
F	None applied	0	97.6 a	10.21 a

[1/] Bravo 500 was applied at a rate of 2 pt/A in a total volume of 83 gal/A. The first early blight lesions occurred on approximately July 12 (1056 day degrees accumulated post planting).

[2/] Barratt-Horsfall scale (0-11) converted to percentage.

[3/] The data are based on actual lesion counts from 375 tubers randomly selected from harvested tuber samples.

Morgan County, 1982:

The day degree accumulation from date of planting to the approximate date of first early blight lesion appearance (May 11 to July 22, respectively) was ca 1172 day degrees. This relates quite closely with observations for Morgan County from previous years (Table 9). The accuracy (total range) of the prediction model is ca 5 days (116 day degrees) assuming ca 25 day degrees accumulated per average July day (estimated).

Table 9. The accuracy of an early blight prediction model for potatoes tested during several growing seasons - Morgan County, Colorado.

Year Tested[1/]	Approximate day degree accumulation (post planting) required for initial early blight lesion appearance
1979	1125
1980	1155
1981[2/]	1056-1097
1982	1172

[1/] Data for 1979 is not presented in this report.

[2/] Planting date uncertain.

Discussion

These studies have shown the value of using timing methods to achieve control of potato early blight with a minimum number of fungicide applications. Both day degree measurements and spore trapping appear to be acceptable means for timing initial chemical applications. The use of day degree accumulations has the advantage of predicting the appearance of the disease in fields before sporulation begins, thus allowing a reasonable period of time to apply protectant fungicides before spread occurs.

Results in the San Luis Valley in 1980 show the validity of using 650 day degrees as the measure of the beginning of disease expression. At the time the first evidence of infection was visible in fields in the San Luis Valley 652.3 day degrees had accumulated. This agreed almost exactly with the data collected in previous years and with field observations in 1981 and 1982 (data not presented). Sporulation began 9-10 days after the appearance of lesions thus providing a period of 9-10 days in which to apply controls before disease spread occurred. The 650 day degree model appeared to adequately predict symptom appearance in all three of the cultivars included in the study since the level of infection was

virtually the same in all of them when the first disease readings were made on July 29 (Table 3). This conclusion is supported by other studies that suggest that foliar resistance to early blight is more dependent on the rate of lesion formation and lesion expansion than the time of initial lesion appearance since lesions appear at approximately the same time in resistant and susceptible potato cultivars.

In Morgan County in 1980, as in 1979 (data not presented), considerably more day degrees accumulated than in the San Luis Valley before the appearance of initial symptoms in all three cultivars. These data indicate that the difference in the two areas is due to location not to potato cultivar. Based upon these results a different temperature (day degree) model will have to be developed for each area where it is to be used.

In Morgan County, 1125 day degrees above 45°F accumulated from planting to the appearance of the first early blight lesions in the Monona cultivar in 1979. In 1980 the first lesions appeared between July 14, when no lesions were observed, and July 25 when spores first appeared on the spore trap slides. Using logical interpolations, based on the appearance of the first lesions in an earlier planted part of the field and the presumed time (based on the San Luis Valley data) between the appearance of the first lesions and sporulation, the probable time of lesion appearance was about July 16. On this date, 1155 day degrees had accumulated. This compares very closely with the 1125 day degrees measured in 1979 and suggests that a model using ca 1150 days degrees above 45°F could be easily developed for the Morgan County area.

The number of day degrees accumulated from planting until the first lesion appearance in Morgan County in 1981 and 1982 agreed closely with that observed in previous years. In 1982, 1981, 1980 and 1979 the day degrees accumulated from planting to first symptom expression were 1172, 1056-1097, 1155 and 1125, respectively. The use of the day degree accumulation model may provide a simple means of timing the first fungicide application in potatoes for the control of early blight.

The 1980 and 1981 data for Morgan County support research conducted in previous years in the San Luis Valley (data not presented). Fungicide applied prior to the initial appearance of early blight provided no additional disease control than fungicide applied at the time of the first appearance of early blight lesions. Therefore, proper timing of fungicide application will allow more efficient chemical usage by decreasing the number of applications required for equivalent disease control.

The data from the San Luis Valley and Morgan County (Northeastern Colorado) showed a wide difference between the two areas in terms of required heat units accumulated before disease appeared. This suggests that some factor other than heat alone governs the appearance of lesions and that heat unit measurement is actually an indirect index which accurately predicts symptom appearance. Solar radiation may be a factor which is measured indirectly. The San Luis Valley is located at a much greater elevation (ca 7,600') than Morgan County (ca 5,000') and therefore would have a cooler environment while simultaneously having more intense sunlight radiation due to the decreased length of the light path through the atmosphere (about 2,600' less). This may account for the difference in the day degree accumulation models for each area and suggests that similar models could be established elsewhere if the proper day degree threshold is determined by temperature measurement and field observations.

The potential application of day degree prediction models for A. solani (and other fungi) infection in crops other than potatoes (e.g., tomatoes) should not be overlooked.

RESEARCH PROPOSAL FOR 1983
David G. Holm

1. The study to evaluate the effect of water stress at the time of tuberization on tuber set, yield, and grade of two potato cultivars, Centennial Russet and Russet Burbank, will be continued to verify 1982 results.
2. Cytex will be evaluated for potential usefulness in potato production. Reputable reports have indicated increased yields resulting from the use of Cytex.
3. The potato breeding and clonal development program will be continued. Advanced clones will be tested in yield trials. Development of virus-tested seed stocks of the most promising clones will be initiated.
4. The Colorado Western Regional Trial will be conducted again in 1983.
5. New protoclones of Russet Burbank will be tested for adaptability in the San Luis Valley.
6. Initial studies have shown that there are differences in the binding ability between Erwinia carotovora var. carotovora and Erwinia carotovora var. atroseptica and among Erwinia carotovora strains to potato tuber tissue. Because of this, there is a possibility that there may be differences between Erwinia varieties and potato cultivars. This may also provide a potential disease rating index for diverse potato cultivars against blackleg. Work will be performed to verify these differences as well as attempting to work this index into the potato breeding program. (In cooperation with Rob Davidson).

BUDGET REQUEST

POTATO BREEDING AND CLONAL DEVELOPMENT

Labor	$2,200.00
Travel	200.00
Equipment and Supplies	600.00
TOTAL	$3,000.00

CULTURAL AND PHYSIOLOGICAL STUDIES

Labor	$2,000.00
Travel	600.00
Equipment and Supplies	1,300.00
TOTAL	$3,900.00

GRAND TOTAL $6,900.00

dhm 3/7/83