

COMPARATIVE ACTION OF CERTAIN
FUNGICIDES ON THE SCLEROTIA
OF RHIZOCTONIA SOLANI KÜHN
ON POTATO TUBERS

Thesis for the Degree of M. S. MICHIGAN STATE COLLEGE George Lewis Barnes 1950

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COMPARATIVE ACTION OF CERTAIN FUNGICIDES ON THE SCLEROTIA OF RHIZOCTONIA SOLANI KUHN ON POTATO TUBERS

Ву

George Lewis Barnes

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INTRODUCTION

Economic Importance of the Potato Crop

The production of potatoes is limited largely to the areas of the north temperate region of the world where the climate is fairly cool and moist. The potato plant is grown largely for its edible subterranean tubers which supplies a cheap, nutritious, carbohydrate food to at least a third of the world's total population. Hardenburg (13) states that the potato ranks first in terms of volume (23.4 per cent) of the fresh product and is outranked only by rice in terms of population fed.

Eighty per cent of the total potato crop of the entire world is grown in Europe alone due to the cool growing season and cheap hand labor in that region. By contrast, according to Hardenburg (13), the United States produces annually only about 4.4 per cent of the world total.

On a state level, Michigan, producing around 19,000,000 bushels per year, ranks sixth and Maine ranks first by producing around 54,000,000 bushels annually. There is presented in table I, a production comparison of the fifteen leading potato states according to a recent 12 year average from the United States Department of Agriculture publication, Crops and Markets (1).

Of the fifteen leading potato states there were several which, during the 12 year period 1937-1948, produced 115 bushels or less an acre. They are in order of decreasing yield per acre; Chio, North Carolina, Michigan, Minnesota, and Wisconsin.

TABLE I

ACREAGE AND PRODUCTION IN THE 15 LEADING POTATO GROWING STATES

		Production	Yield
~	Acreage	(Thousand	Per Acre
State	(Thousands)	Bushels)	(Bushels)
	Average 1937-48	Average 1937-48	Average 1937-48
	1307-10	1501-40	1007-40
Maine	213	54,220	300
Idaho	148	35,204	238
New York	186	31,073	388
NOW TOTA	100	01,010	000
California	89	29,661	325
D	3.53	30 634	170
Pennsylvania	151	19,614	132
Michigan	182	19,231	109
•	_		_
Minnesota	190	18,881	104
North Dakota	139	17,512	121
2			
Colorado	80	15,933	200
Wisconsin	153	13,290	91
1,15 COLIS III	100	10,290	31
New Jersey	61	11,073	184
		10.000	3.45
Nebraska	71	10,229	143
North Carolina	83	9,250	113
		•	
Virginia	72	9,245	128
Ohio	76	8,445	115
OHIO	70	0,440	110

The relatively low yield per acre for Michigan, prior to 1948, is due to several limiting factors, each of which must be taken into account in any attempt to increase the yield. Some factors to consider are as follows: climate, soil, varieties, fertilizer, pest

control, and disease free "seed" stock. Although for the period 1937-48 the average yield in Michigan was 109 bushels per acre; for 1948 and 1949 it had risen to 150 and 165 bushels per acre respectively. This increase in yield per acre is due to several factors. They are: increased use of irrigation, increased use of high quality fertilizer, and the federal farm price support plan.

Potato Diseases of Michigan

The potato, being an established crop in Michigan and grown under a wide range of soil and climatic conditions, is subject to a wide variety of diseases. Some of these diseases are of only slight economic importance while others assume serious proportions under certain conditions and at other times caused very little loss to the crop. Some, such as scab and Rhizoctonia, cause serious reductions in yield and quality every year.

Each one of the diseases present in Michigan is not in itself a limiting factor in potato production in this state, but all of them together are responsible for annual losses amounting to many thousands of bushels.

Almost all of the diseases common to the potato are present in Michigan. Therefore, many are of considerable importance; scab, Rhizoctonia, wilts, late and early blight, and various virus diseases. They are discussed fully in various technical publications and bulletins of the Michigan Agricultural Experiment Station and Extension

Service. All of the above diseases are discussed in detail by Muncie (20) in one of these publications which should be very useful to all potato growers in this state.

History of the Causal Fungus of the Rhizoctonia Disease

Root destroying fungi of the gemus Rhizoctonia were first discovered by De Candolle in 1815 (9). He named two species: Rhizoctonia medicaginis, occurring on Medicago, Trifolium, and related hosts; and Rhizoctonia crocorum, destructive to crocus bulbs. Little important work was published on Rhizoctonia until 1851, when the Tulasne brothers (9) gave a comprehensive summary on all the described species of the genus and the various host plants. They concluded that all these species were not to be regarded as distinct from each other but rather as one species which they called Rhizoctonia violacea. However, in 1858 (9) Kühn discussed more at length certain forms of economic importance and described a new species which occurred on the potato, Solamum tuberosum L., and which he named Rhizoctonia solani.

Many attempts have been made to connect the sterile fungus

Rhizoctonia solani Kühn with a perfect stage of various fungi. Several

fungi have been suggested as being the perfect stage through their

frequent association with the sterile stage but evidence of genetic

relationship was always lacking. However, Rolfs in 1903 (22), while

working with the Rhizoctonia disease of potatoes in Colorado, con
stantly found a basidiomycete on the stems associated with the presence

of Rhizoctonia solani Kühn on the roots, stolons, and tubers.

Dr. E. A. Burt identified the fungus as Corticium vagum B. & C. var. solani but later dropped var. solani because he believed that there is no varietal distinction between Corticium vagum B. & C. and the fungus occurring on potatoes. Rolfs was able to trace the connection between the two fungal forms and finally completed his evidence when he obtained cultures of Rhizoctonia solani Kühn from single spores of Corticium vagum B. & C. For certain mycological reasons, Rogers (25) in 1943 proposed that the name Corticium vagum B. & C., be changed to Pellicularia filamentosa (Pat) comb. nov. Since the name Pellicularia filamentosa (Pat) Rogers has been used in the recent literature to denote the sexual stage of the fungus, it is evident that the name has become accepted and valid.

Taxonomy and Morphology of Rhizoctonia Solani Kühn

Taxonomy

Rhizoctonia solani Kühn is the imperfect or sterile phase of the basidiomycete, Pellicularia filamentosa (Pat.) Rogers, of the family Thelephoraceae, order Agaricales. As Rhizoctonia solani Kühn, the fungus is a member of the form genus Rhizoctonia, order Mycelia Sterilia of the Deuteromycetes (Fungi Imperfecti).

Peltier (22) states that Duggar has given the following synonomy under Rhizoctonia solani Kühn:

Rhizoctonia napaeae West (1846)

Rhizoctonia rapae West (1852)

Rhizoctonia solani Kühn (1858)

Rhizoctonia betae Eidam (non Kühn) (1887)

The following names are now synonyms for Pellicularia filamentosa (Pat.) Rogers:

Hypochnus solani Prill. & Del. (1891)

Corticium vagum B. & C. (1903)

Hypochnus solani Prill. & Del. has been accepted by the Europeans in general as the perfect stage of the fungus. However, Peltier (22) states that "in his monograph on the Thelephoraceae, Burt limits

Hypochnus to resupinate species with colored, echinulate spores, while under Corticium he includes species always resupinate, which have colorless spores and lack cystidia. According to Burts' classification, Hypochnus solani Prill. & Del. becomes a synonym under Corticium vagum B. & C.."

Both are now synonyms for Pellicularia filamentosa (Pat.) Rogers.

Morphology

The morphological characters of Rhizoctonia solani Kühn vary with the age of the mycelium. The young hyphae branch from the parent hypha at an acute angle and then grow parallel with it. A constriction is present at the union of a hyphal branch with the parent hypha and a septum is usually laid down a few microns from the point of union. The young hyphae are frequently strongly vacuolate and are light in color. With age the hyphal branches tend to form at right angles to the parent hypha and are darker in color and less strongly vacuolate than the young hyphae.

The formation of sclerotia in nature is common, but they vary considerably in size and shape. On potatoes they are generally flat and measure from about one to ten millimeters in width on the average. Occasionally giant flat sclerotia are formed. Sclerotia are formed by the grouping together of many hyphal filaments to form a compact mass of brown hyphae which soon turn black and become very resistant. Apparently they form in response to moist conditions in the environment. They appear on potato tubers in the fall, especially if the tubers have been left in the ground long beyond the normal harvesting time.

Less frequently, during the summer the Pellicularia (Corticium) stage is formed. This stage is formed by the dark-brown hyphae of the sterile stage gathering about the base of a plant and giving rise to an ashy gray mycelium which forms a loosely attached fine network about the stem. The outer hyphae of the ashy gray mycelium bear

club-shaped basidia with four sterigmata and basidiospores. The basidiospores are colorless, oval to ovate, and have pointed bases.

The usual basidiospore measurement varies from 9 to 14 µ by 6 to 8 µ.

Early History of the Disease in America

Although a Rhizoctonia disease of potato has long been common in Europe, especially in Germany, where it is known as "Grind" and "Pockenkrankheit", there had been no report on Rhizoctonia in America until Pammel (9) in 1891 reported that a beet rot in Iowa was caused by Rhizoctonia betae Eidam (R. solani Kuhn). The first record of R. solani Kühn occurring on potatoes in America as reported by Duggar and Stewart (9) was made by Sirrine and Rolfs who found it on potato stems collected on Long Island, New York in 1898. Later surveys in 1899, by Duggar and Stewart, demonstrated that the fungus could be found on the stems and tubers of potatoes in many areas of the state of New York. Specimens of tubers and stems bearing sclerotia of Rhizoctonia solani Kühn were received by Duggar and Stewart during 1899 and 1900 from many states including Pennsylvania, Ohio, Michigan, Alabama, Colorado, Iowa and Maryland. They suggested that the fungus is probably very generally distributed. Time has proven their assumption to be correct, because Rhizoctonia solani Kühn is now found wherever potatoes are grown in North America.

Economic Importance of the Disease

The Rhizoctonia disease of potatoes is very widespread; i.e., being present in all of the continents of the world and the West Indies, Japan, and New Zealand. Apparently all commercial variaties of potatoes are susceptible to the disease.

There is presented in table II a comparison of the reduction of yield of potatoes due to Rhizoctonia solani Kühn based on estimated figures from certain issues of the Plant Disease Reporter (3).

REDUCTION IN YIELD OF POTATOES DUE TO LOSSES RESULTING FROM THE RHIZOCTONIA DISEASE

Category	Per cent Reduction in Yield						
	131	132	133	134	135	136	137
All Diseases (U.S.)	18.8	22.3	16.6		21.0	15.7	18.3
All Diseases (Mich.)	24.7	12.0	12.3	18.6	10.3	18.0	31.8
Rhizoc. Disease	2.3	2.5	2.7		3.5	2.2	1.5
Rhizoc. Disease (Mich.)	0.5	0.5	1.0	1.0	1.0	5.0	2.5

For the seven year period covered in table II, Michigan lost on the average about 1.6% of the annual potato yield for the state.

During the same period the average yearly loss for the United States as a whole was about 2.5%.

Symptoms of the Disease

The Rhizoctonia disease of the potato, Solanum tuberosum L., caused by Rhizoctonia solani Kühn is very common and widespread; being found anywhere in the world where potatoes are grown to any extent. It is present, and severe under some conditions, in all the Northern potato growing states of the United States.

The disease not only affects the potato, but also many vegetables, field crops, ornamental plants, and certain weeds. In the case of potatoes, losses resulting from this disease are of two types. First, there is an actual loss due to poor stands, low yields, and a decrease in marketable tubers. Secondly, there is an indirect loss due to rejection for certification for "seed" stock and also lowered quality for table stock.

The disease is usually more severe on potatoes in the presence of abundant soil moisture in all types of soil; especially those of an acid nature, even though the fungus is frequently prevalent in neutral or slightly alkaline soils. The fungus generally thrives under those conditions favorable to the growth of the potato plant itself.

The Rhizoctonia disease of the potato has been given a wide variety of common names, each usually according to an expression of the disease on some particular portion of the plant. According to Heald (14) the disease has been given such names as black speck, russet scab, black pox, black scurf, and little potatoes, because of the effect on the tubers; brown stems, stem rot, stem canker, and collar rot due to the nature of the stem attacks; and rosette, leaf roll,

and aerial potato because of accompanying physiological disturbances. Because it is impossible to use a name which will describe the symptoms of all the different stages, the generic name of the fungus, Rhizoctonia, has been applied to the disease as well and apparently is the generally accepted common name for the disease now. In the past, the terms rhizoctonosis and rhizoctoniose have been used by some writers.

Top Symptoms

A severely injured root system or stem will result in certain physiological responses in the aerial portion, of the plant.

A leaf roll similar to the effect of drought or to early symptoms of the leaf roll virus disease is frequently the result of extensive root injury. The leaflets at the top of the shoots roll upward and may become yellow but the lower leaves do not become rolled, harsh, and brittle later as occurs when true leaf roll is present. Since true leaf roll is present in most potato seed stocks, the presence of Rhizoctonia can not be positively verified by this symptom alone.

A rosette of yellow or green leaves often occurs if the base of the stem is severely attacked after the plant has grown considerably. This is due to a shortening of the new petioles of the stem so that the top leaves are massed together in a rosette fashion. Since the canker sometimes prevents the translocation of elaborated sugars from the leaves to the tubers, the excess food causes the vine to become unusually large. These large plants will set a large number of small

knobby and russeted tubers which will never increase in size and are consequently unmarketable.

Leafy aerial tubers sometimes develop in the axils of the leaves when the stem has been injured to the extent that translocation can not occur. Aerial tubers can also result from injuries due to other causes and are, therefore, not a positive evidence of the Rhizoctonia disease.

Symptoms on Stems

According to Dana (8) injury to the stem may take several forms: stem cankers or lesions, growing point injury, corroded stems, or the stem may be covered by the mycelium of the fungus which does no noticeable damage but which can serve as a sign by which the disease may be identified.

The stem lesions are of varying size, and in extreme cases may even girdle the stem. These lesions develop most commonly on shoots from very early or volunteer plants and as the season advances these lesions may be largely outgrown.

Growing point injury is caused by lesions forming at the shoot tips where the tissues apparently are very susceptible to attack by the fungus. This type of injury is most serious in early planted fields because of the low temperatures and abundant moisture at that time. Every emerging shoot may be killed in this manner until the seed piece is exhausted and produces no more shoots, resulting in a missing hill.

Corroded stems are found on the older infected plants. This corrosion by the fungus may even girdle the stem at its base and give rise to the various physiological symptoms previously discussed. Root and stolon development may be hindered or even prevented from this area. Frequently a cluster of malformed tubers is formed about the stems very close to the surface of the ground. Loss from this type of injury will depend upon the extent to which the nourishment of the plant and the development of the tuber is disturbed.

Symptoms on Roots

Injury by Rhizoctonia solani Kühn occurs to roots of all sizes, but is most destructive to the feeding roots. The larger roots may show cankers while the largest roots are affected to a serious extent only when the base of the stem is seriously corroded in which case they may be prevented from developing properly.

Injury to the feeding roots is one of the most serious effects of the disease; because if the small rootlets are killed, the absorbing capacity of the plant is materially reduced. This is particularly harmful to the plant during dry situations and often results in poor or abnormal top growth even when sufficient moisture is present.

Careful observations tend to show that feeding root injury by

R. solani Kühn is common wherever potatoes are grown and constitutes
a serious phase of the disease.

Symptoms on Stolons

The stolons, or underground stems on which the tubers are borne may be attacked by Rhizoctonia solani Kühn in much the same way as are the stems. Cankers on the stolons will interfere with the nourishment of the young tubers and in severe cases may cut off the stolons and prevent the formation of tubers. This type of injury occurs most frequently in wet poorly drained soils and often results in considerable loss to the potato grower.

Symptoms on Tubers

Rhizoctonia solani Kühn in the resting or dormant stage is common on tubers as dark brown or black bodies (sclerotia) which vary in size from mere specks to giant sclerotia an inch or more in diameter. Since these sclerotia are often hard to wash off they have been called by some housewives as "the dirt that won't wash off." There may be only one or two sclerotia on a tuber, or the tuber may be almost completely covered by them. These sclerotia are sometimes called black scurfs.

The attack of the fungus on the tuber may take the form of a russeting or rotting. This russeting, or roughening of the skin of the tuber, resembles an infestation of the potato scab organism in many cases. It may cover a part or nearly all of the tuber and render it unattractive or subject to diseases in storage.

Rhizoctonia solani Kuhn seldom induces rotting in potato tubers, but at times it, together with bacteria and certain fungi, notably Fusarium spp., causes a so-called stem end jelly rot.

Under certain conditions R. solani Kühn invades the lenticels and produces very small dry pits that can be lifted from the tuber with the point of a knife. This type of infection has been designated sand scurf by Ramsey, Wiant and Smith (24).

REVIEW OF THE LITERATURE

Black scurf, or Rhizoctonia disease of the potato, caused by the fungus Rhizoctonia solani Kühn, is widespread in Michigan, and other states, and sometimes causes considerable loss to the potato grower. There is a definite need for control of this disease and since it is impractical to treat large areas of soil, fungicidal treatments of the infected "seed" potatoes has come into common usage. These treatments may reduce both the damage by Rhizoctonia and seed piece decay. Both results can increase the yield.

Seed treatments for the control of certain potato diseases was first proposed by Bolley (5) in 1891 when he suggested the use of disinfectants such as are used in medical practice. He recommended the use of mercuric chloride, 2 oz. in 15 gallons of water. This standard treatment for control of potato scab called for soaking whole potatoes in HgCl₂(1-1000) for one and one-half hours. As a result of the research by Arthur in 1895 (5), the use of formalin (1-240) almost completely replaced the use of corrossive sublimate solution in general practice in various states.

However, about this time the various workers investigating the relative killing power of the two chemicals on the sclerotia of Rhizoctonia solani Kühn began to fall into two schools of practice. One group advocated the use of corrosive sublimate solutions while the other group backed the use of formalin solution, either hot or cold as the case may be.

The literature on the use of corrosive sublimate solutions and formalin solutions is voluminous and will not be extensively reviewed here. However, excellent summaries on the use of corrosive sublimate solutions are given by Gloyer (11), Coons (5), and Goss and Werner (12); and on the use of formalin solutions by Melhus and Gilman (17), Gilman and Melhus (10), and Melhus, Gilman, and Kendrick (18). The use of acidulated corrosive sublimate was first proposed by Cunningham (16), modified by Leach, Johnson, and Parson (16), and comparative field trials were run by Plummer and Bonde (23), Sanford (26), and Wheeler and Moore (28) in comparison with the standard fungicides.

In reviewing the literature covering the use of these "old" standard fungicides, several general conclusions stand out:

- (a) Hot formalin solutions are more effective than the cold solutions but are more phytotoxic, resulting in low or retarded emergence of shoots from treated tubers if temperatures are not strictly regulated. The use of the hot solutions requires special equipment and sensitive temperature controls which add considerably to the cost of treatments.
- (b) If the treatment is applied for killing the tuber borne scab organism alsone, either the standard corrosive sublimate solution or the standard formalin solution may be used; but when both Rhizoctonia sclerotia and scab are present the corrosive sublimate solution has been found to be the more effective fungicide. Apparently the formalin penetrates the sclerotia with difficulty and sometimes doesn't even

kill certain fungi hand bacteria associated with the sclerotia.

- (c) The use of corrosive sublimate is a health hazard and it is also phytotoxic. If the tubers are not dried rapidly after treatment they may be injured to such an extent that no or few sprouts will emerge after planting.
- (d) Acidulated corrosive sublimate is more effective than non-acidulated corrosive sublimate in killing the sclerotia of Rhizoctonia solani Kühn. Treated tubers must be dried after treatment to avoid injury to the sprout primordia.

Within the past twenty years various organic mercury compounds have been developed and used to some extent for tuber treatments.

Conflicting data with the use of the organic mercurials in the control of Rhizoctonia have been presented by various workers.

Goss and Werner (12), state that White in summarizing experiments reported from various states and Canada in 1926, concludes that "In general, although by no means consistently so, the Semesan Bel compounds used as a 10 per cent dip have given satisfactory control of stem lesions, have resulted in at least 20 per cent increases in yield and have yielded a crop as free of sclerotia as the standard corrosive sublimate treatment." Clayton (4) has stated that the organic mercurials controlled potato scab as well as corrosive sublimate, but were less effective against black scurf (tuber borne Rhizoctonia). Miles (12) in 1929 stated that in no case was there any effective field control of Rhizoctonia, as measured by the sclerotia present on tubers

at harvest, with either Semesan Bel or Bayer Dip Dust, while mercuric chloride gave good control. Michigan tests conducted in 1928 and 1929 by Moore and Wheeler (19) indicated also that mercuric chloride is more effective than the organic mercurials. Wheeler and Moore (28) in 1933 found that Semesan Bel, Bayer Dip Dust, and Sano seed were less effective than corrosive sublimate in controlling scab and the Rhizoctonia disease.

The tendency of the work on the effectiveness of the organic mercurials is to consider them as less effective against the Rhizoctonia sclerotia than against scab. Cunningham (6,7) reports that treatments with yellow oxide of mercury and The New Improved Semesan Bel caused a delay of shoot emergence and decreased yield when used on certain varieties of potato tubers.

From the foregoing review of the literature it is readily seen that there is a need for new materials in the treatment of Rhizoctonia infected potato tubers. The author will present his results with laboratory experiments with certain "new" fungicides manufactured by the Dow Chemical Company, Midland, Michigan.

CURRENT RESEARCH

Objective

The objective of this investigation was to study the comparative action of certain fungicides for use in potato tuber treatments which will meet the following desired specifications for the control of black scurf of potato tubers:

- (a) The material must be as effective as the present standard treatment in Michigan; i.e., soaking whole tubers in HgCl₂ (1-1000) for 30 minutes.
- (b) The material must be effective in the same, or less, time than the standard 30 minute soaking period.
- (c) The material must be effective when the solution is cold so that the treating equipment and operating procedures will be simplified.
- (d) The cost of the material should be much lower than the cost of mercuric chloride.
- (e) The material should be non-corrosive so that metal containers can be used if desired.
- (f) The material should be other than the mercurials (calomel, corrosive sublimate, acidulated corrosive sublimate, yellow oxide of mercury, ethyl mercury iodide, and the organic mercury compounds such as Semesan Bel, etc.). Ken Knight (15) has shown that where these compounds are used in Michigan, and some other states, there is an increase of potato scab

- due to the presence of mercury tolerant parasitic actinomycetes in the soil. In addition, the use of mercurials presents a health hazard wherever they are used.
- it for a reasonable time. If the solution maintains its strength then there is no need to frequently renew its strength or to discard the solution and make up a new one. Christophy, to discard a solution after only a few lots of tubers have been treated or to renew its strength frequently is very expensive. Because of these difficulties, many farmers either ignore tuber disinfection or use easier, but less effective treatments.

From the foregoing statements it is readily seen that there is a real need for a tuber disinfectant that will meet the above specifications.

Materials and Methods

For the laboratory experiments potato tubers of the Katahdin, Chippewa, Pontiac, and Russel Rural varieties were obtained from the Lake City Experiment Station, Lake City, Michigan. Each tuber was selected for the presence of abundant sclerotia and an effort was made to obtain sclerotia of all sizes. These tubers were kept in storage until selections were made for the various fungicide trials.

Various standard and new materials were employed in a study of the comparative fungicidal effect in killing the sclerotia of Rhizoctonia solani Kühn at certain dilutions and exposure times.

These fungicides are: corrosive sublimate (HgCl₂), acidulated corrosive sublimate, New Improved Semesan Bel, SCD 765 (Dow Chemical Company), a number of proprietary organic compounds called Dowicides.

The New Improved Semesan Bel is an organic mercurial fungicide manufactured by the Dupont Semesan Company (Inc.). It is composed of 12 per cent hydroxymercurinitrophenol, 3.2 per cent hydroxymercuri-chlorophenol, and 84.8 per cent inert ingredients. The mercury equivalent of this compound is 9.5 per cent.

S.C.D. 765 is a proprietary compound produced by the Dow Chemical Company. The chemical components of this fungicide cannot be revealed at the present time.

The Dowicides are manufactured by the Dowicide Division, Dow Chemical Company, Midland, Michigan. They are for the most part, sodium salts of various chlorinated phenols. Some are sodium salts of phenyl phenol compounds. They are differentiated by the addition of

one of the capital letters of the alphabet immediately following the term Dowicide. Their known fungicidal properties and uses are discussed in a publication (2) of the Dow Chemical Company. They are as follows:

Dowicide A. Sodium Salt of Orthophenylphenol.

Dowicide B. Sodium Salt of 2,4,5-Trichlorophenol.

Dowicide C. Sodium Salt of 2 Chloro-Orthophenylphenol.

Dowicide D. Sodium Salt of 2 Chloro-4-Phenylphenol.

Dowicide E. Sodium Salt of 2 Bromo-4-Phenylphenol.

Dowicide F. Sodium Salt of 2,3,4,6-Tetrachlorophenol.

Dowicide G. Sodium Salt of Pentachlorophenol.

As a consequence of the results obtained by the early workers, both in the laboratory and the field, laboratory methods were devised by which the field value of a given tuber treatment could be determined without the necessity of field trials. A review of the earlier work showed that the results of the laboratory methods were confirmed by subsequent field trials.

Various investigators have made contributions to the methods of aseptically growing Rhizoctonia from sclerotia on agar in Petri dishes. Gloyer (11) states that Duggar and Stewart have noted that Rhizoctonia grows well on acidulated agar without interference from bacteria. In his work, Gloyer aseptically placed sclerotia on potato agar of one per cent acidity, but it was found that the plates were soon overrun by the potato bacillus, Bacillus vulgatus Trevisan, when untreated sclerotia or those treated with formaldehyde were used. To keep down

these bacteria, he added two drops of 50 per cent lactic acid to each 10 cc. of the medium. Gloyer generally placed five sclerotia in each Petri dish, allowed them to set at room temperature, and after three to five days the results from the untreated sclerotia could be determined. In the case of the treated sclerotia, the cultures were frequently examined and after ten days they were examined with the low power of the microscope to determine if hyphae of Rhizoctonia were growing from the sclerotia. Melhus, Gilman, and Kendrick (18) used essentially the same method in their laboratory work involving the fungicidal action of formaldehyde. In 1933, Sanford and Marritt (27), in a laboratory study of viability of sclerotia after certain treatments, aseptically placed treated sclerotia on potato dextrose agar which was acidified, while it was cooling after sterilization, by adding 3 per cent by volume of 5 per cent lactic acid.

The method as used by Sanford and Marritt was employed in the present investigations. It has proved very satisfactory in prevention of contamination, especially by bacteria. Usually ten or more sclerotia were placed in each Petri dish. The Petri dishes were stored at room temperatures, examined macroscopically every few days, and then examined with a low power binocular microscope after four to six days to determine the number of sclerotia which lived through the treatments and produced Rhizoctonia hyphae.

It has been observed by various investigators that the size and density of the sclerotia play an important part in any tuber treatment test either in the field or in the laboratory.

The first study on the relation of the size of the sclerotia to the required time necessary to kill them with various fungicides was published by Sanford and Marritt (27) in 1933. They found, in counting the sclerotia from representative lots of infected potato tubers, that from a total of 4,732 sclerotia, 76.3 per cent were small, 17.6 per cent were of medium size, and 6 per cent large. From these figures they suggested that the proportion of each size occurring in nature was a ratio of 13:3:1. Extra small and extra large sclerotia were not counted. They showed that all of the small and medium sclerotia could be killed in 15 minutes in acidulated mercuric chloride (1-500) and only 20 per cent of the large ones survived. In reference to the very small number of large sclerotia occurring in nature, this percentage is negligible. It can be assumed that any treatment which will kill the small, medium, and most of the large sclerotia is an effective one from both field and laboratory standpoints.

Coons (5), after treating infected tubers one half hour in corrosive sublimate (1-1000), without any regard to the sizes of sclerotia, obtained a 98 per cent stand of plants with commercial control of the disease.

In certain laboratory experiments by Muncie (21) in 1934 it was necessary to determine the thickness of the sclerotia in order to test the efficiency of certain tuber treatment fungicides. For this purpose, he devised a very simple measuring instrument. This consisted of a glass tube, 7.5 millimeters in internal diameter and about four inches in length, within which was inserted a glass rod somewhat

longer than the tube. A millimeter scale was marked on a label and the label in turn was glued to the glass rod so that the top line of the graduation was aligned with the top of the tube. In measuring the thickness of a sclerotium, the tube was set over the sclerotium and the rod lowered until it rested on the sclerotium. The thickness of the sclerotium was then determined by noting the distance between the top of the tube and the top mark on the scale on the rod. In practice the procedure followed was as follows: The thickness of each sclerotium on a tuber was measured and the figure recorded on the tuber with India ink near the sclerotium. The tubers were then treated, rinsed with sterile tap water, and the sclerotia of each size range were removed with a flamed scalpel and plated together on potato dextrose agar containing 3 per cent by volume of 5 per cent lactic acid as used by Sanford and Marritt (27). Table III presents unpublished data by Yuncie (21) in which is shown the relative efficiencies of certain tuber treatments in regard to the thickness of the sclerotia.

Table III tends to show that any treatment which would kill both the small and medium sized sclerotia would generally kill the large sclerotia also.

From the foregoing investigations it seems that the size of the sclerotia within narrow limits is not always the limiting factor in any tuber treatment trials either in the laboratory or in the field. Therefore, in the work reported by the writer, he considered the size of the sclerotia as a negligible factor in Michigan and performed his experiments using representative sclerotia from all size ranges. The same culture techniques were followed as used by the investigators previously mentioned.

TABLE III

EFFECT OF CERTAIN FUNGICIDES ON VARIOUS SIZES OF SCLEROTIA OF RHIZOCTONIA SOLAMI KÜHN

Material	Time: Minutes	Thickness of Sclerotia	Number Sclerotia Treated	Number Sclerotia Killed	Per Cent Sclerotia Killed
HgCl ₂ 1-1000	30	0.5 mm 1.0 1.5	264 46 16	264 46 16	100 100 100
HgCl ₂ 1-1000	60	0.5 1.0 1.5	435 40	429 39	98.6 97.5
HgCl ₂ 1-1900	90	0.5 1.0 1.5	264 40 8	264 40 8	100 100 100
Acid HgCl ₂	10	0.5 1.0 1.5	504 50	504 50	100 100
HgCl ₂ 1-834	10	0.5 1.0 1.5	392 31	392 31	100 100
Semesan Bel 1-125	10	0.5 1.0 1.5	264 24 11	264 24 11	100 100 100
Check	No treat- ment	0.5 1.0 1.5	152 8 8	103 1 0	68 12.5 0

Results and Conclusions

It was thought that the results of some of the laboratory experiments of the earlier workers should be confirmed and substantiated by laboratory experiments.

Table IV presents the results obtained by the author in determining the killing efficiency of corrosive sublimate, acidulated corrosive sublimate, and the New Improved Semesan Bel on the sclerotia of Rhizoctonia solani Kühn. The data presented here represents the summarized results of five separate tests.

TABLE IV

COMPARATIVE EFFECTS OF CERTAIN MERCURIAL FUNGICIDES ON THE SCLEROTIA

OF RHIZOCTONIA SOLANI KÜIN

<u> </u>				
Material	Time: Min- utes	Number Sclerotia	Number Sclerotia Killed	Per Cent Sclerotia Killed
Semesan Bel 1-125	10	1242	1155	92.99
HgCl ₂ 1-1000	30	21 98	1686	76.71
HgCl ₂ 1-1000	60	1214	1127	92.83
HgCl ₂ 1-1000	90	1008	942	93.45
Acid HgCl ₂ 1-1000	10	2379	2187	91.93
HgCl ₂ 1-834	10	423	423	100.00

Table IV shows that a 100 per cent kill of Rhizoctonia sclerotia was obtained by soaking the infected tubers in HgCl_2 (1-834) for ten minutes. Because the number of sclerotia was rather low in this case, this percentage may not be valid. The use of more sclerotia in this test would have given figures which would be more indicative of its efficiency. The data also shows that 30 minute treatment in HgCl_2 1-1000 was not as effective as treatments of 60 and 90 minutes.

On the average, the percentages are high enough to consider these mercurials as excellent fungicides from a laboratory standpoint. However, when the mercurials are used in actual field trials they may prove to be phytotoxic if the tubers are not dried rapidly in the shade after the treatments. Ken Knight (15) has shown also that there is an increase of potato scab after using the mercurials because of the presence of certain mercury tolerant Actinomycetes in the soils of several states, including Michigan. In addition, the mercurials present a definite health hazard and are also corrosive in nature.

It can be concluded that although the mercurials are excellent fungicides they do not meet all the desired specifications because of possible phytotoxicity, high cost, and the health hazard involved.

To overcome these objections certain non-mercurial fungicides were used in later tests.

Tables V, VI, and VII present the results of tests on the toxicity values of certain new fungicides on the sclerotia of Rhizoctonia solani Kuhn. The data presented here represent the summarized results of eight separate tests.

TABLE V

COMPARATIVE TOXICITY OF CERTAIN DOWNCIDES AT A 1-500 DILUTION FOR A
30 MINUTE PERIOD ON SCLEROTIA OF RHIZOCTONIA SOLANI KÜHN

Material	Number Sclerotia	Number Sclerotia Killed	Per Cent Sclerotia Killed
Dowicide A	93	93 .	100.00
Dowicide B	277	276	99.64
Dowicide C	367	367	100.00
Dowicide D	161	129	80.12
Dowicide E	154	99	64.29
Dowicide F	50	50	100.00
Dowicide G	2 95	269	91.19
NaOC1 1-2 Check	70	0	0.00

From the results shown in table V it can be readily seen that of the Dowicides used in this test the large majority of them are highly toxic to the sclerotia of R. solani Kühn.

The fungicidal properties of these compounds were not fully known at the time of the initial tests. Therefore, the 1-500 dilution was arbitrarily selected. Dowicides D and E are snown to be definitely inferior, from a fungicidal standpoint, at this dilution. The sclerotia

exposed to Dowicides A, C, and F were killed in all cases. Dowicides B and G showed over 90 per cent kill and therefore might, at this concentration, be considered as effective fungicides.

Under the conditions of the experiment, Dowicides A, B, C, and F might be considered as acceptable fungicides for tuber treatments.

From the foregoing results, it was decided that a series of tests, involving a higher dilution, would be made. The results are presented in table VI. The data presented represent the summarized results of eight separate tests.

TABLE VI

COMPARATIVE TOXICITY OF CERTAIN DOWICIDES AT 1-1000 DILUTION FOR A

30 MINUTE PERIOD ON SCLEROTIA OF RHIZOCTOMIA SOLANI KÜHN

Material	Number Sclerotia	Number Sclerotia Killed	Per Cent Sclerotia Killed
Dowicide A	984	815	82.82
Dowicide B	510	487	95.49
Dowicide C	183	180	98.36
Dowicide D	183	137	74.86
Dowicide E	119	98	82.35
Dowicide F	496	4 56	91.94
Dowicide G	496	474	95.57
NaOCl 1-2 Check	90	5	5.55

The results of the tests presented in table VI show that at a dilution of 1-1000, Dowicides B, C, F, and G are more effective in killing Rhizoctonia sclerotia than Dowicides A, D, and E. At a dilution of 1-1000 Dowicide A was not as effective as it was at a dilution of 1-500. However, Dowicides B, C, F, and G proved to be almost as highly toxic at a 1-1000 dilution as at a 1-500 dilution. Of course, Dowicides D and E still showed a low toxicity value at a 1-1000 dilution.

Dowicides B, C, F, and G are apparently excellent fungicides but Dowicides D and E are inferior fungicides under the conditions of the experiments. It is indicated that Dowicide A is an excellent fungicide at a 1-500 dilution but is a rather poor one at 1-1000.

While working with the Dowicide compounds it was observed that Dowicide F gave high toxicity values, was highly soluble, and also relatively safe to handle. It was thought that it would prove interesting to determine its toxicity values at various dilutions. At the same time, tests were made with an instant dip compound designed to prevent bacterial soft rot of potato tubers in storage. This compound is known as S.C.D. 765. It is composed of one of the Dowicides and a special detergent.

The results of these tests are presented in table VII. The data presented represent the summarized results of a number of separate trials.

TABLE VII

COMPARATIVE TOXICITY OF CERTAIN DOMICIDES AT VARIOUS DILUTIONS AND DIFFERENT LENGTHS OF TIME ON SCLEROTIA OF RHIZOCTONIA SOLANI KUHN

Material	Time: Min- utes	Number Sclerotia	Number Sclerotia Killed	Per Cent Sclerotia Killed
Dowicide F l-1000	30	910	900	98.90
Dowicide F 1-2000	30	436	433	99.31
Dowicide F 1-3000	30	126	126	100,00
Dowicide F 1-4000	30	170	168	98.82
SCD 765 1-500	15	500	416	83.20
SCD 765 1-500	30	50	35	70.00
NaOCl 1-2 Check	5	210	6	2.86

The results of the tests presented in table VII show that Dowicide F has a very high toxicity value from a dilution of 1-1000 to as high a dilution as 1-4000. This indicates that this fungicide is highly efficient and should prove to be an excellent compound for treating potato tubers in field practice.

Despite the bactericidal value of SCD 765 its fungicidal value is apparently inferior to those of the Dowicides.

During this investigation it was thought that the addition of a detergent to an experimental fungicide would lower the surface tension of the solution and possibly achieve the same, or improved, fungicidal

efficiency in a shorter soaking period. Vel, a neutral sulfated monoglyceride detergent, was used in this experiment at a concentration of .05 grams per liter of solution.

The results of this investigation are presented in table VIII.

The data presented represents the summarized results of three separate trials.

From the results shown in table VIII it can be concluded that in the case of the Dowicides compounds the addition of Vel actually lowered the fungicidal efficiency of each to a great extent; that is, about 20 per cent in the case of Dowicides F and G and about seven per cent for Dowicide C. However, the Dowicides have been shown to be excellent fungicides without the addition of Vel.

In the case of the mercurials used in this experiment the addition of Vel at the rate of .05 grams per liter increased the fungicidal value of Semesan Bel and acidulated corrosive sublimate only about one per cent but appeared to lower the standard corrosive sublimate about 4 per cent.

It is apparent that the value of the addition of a detergent to a fungicide depends upon the chemistry of the compounds used.

In an investigation of this type, the conditions of the laboratory experiments must simulate the conditions and procedures in the field as closely as possible if the results are to prove valid and applicable in future field practice.

Therefore it was deemed that because the common field practice is to place the sacks or crates of treated potato tubers in a pile and

TABLE VIII

TOXICITY OF CERTAIN FUNCICIDES, USING VEL AS A DETERGENT, TO SCLEROTIA

OF RHIZOCTONIA SOLANI KÜHN

Material	Time: Min- utes	Number Sclerotia	Number Sclerotia Killed	Per Cent Sclerotia Killed
NaOCl 1:2	5	190	0	0
HgCl ₂ 1-1000 *	30	55	51	92.72
HgCl ₂ 1-1000	30	155	149	96.13
Acid HgCl ₂ 1-1000 *	10	95	95	100.00
Acid HgCl ₂ 1-1000	10	250	243	97.20
Semesan Bel 1-1000 *	10	60	60	100.00
Semesan Bel 1-1000	10	3 30	327	99.1
Dowicide C 1-1000 *	30	100	91	91.00
Dowicide C 1-1000	30	183	180	98.36
Dowicide F 1-1000 *	15	75	55	73.33
Dowicide F 1-1000	15	4 96	456	91.94
Dowicide F 1-1000 *	30	80	63	79.75
Dowicide F 1-1000	30	910	900	98.90
Dowicide G 1-1000 *	30	190	142	74.74
Dowicide G 1-1000	30	4 96	474	95.57
* .05	g. Vel/L	added		

allow them to set there until they were cut and planted, that an experiment simulating this practice would be made in the laboratory. In the field these treated tubers sometimes remain in a pile from one half hour to three or four hours. It was thought that in the laboratory a two hour setting period would represent the typical time in the field. Therefore in tests performed in the laboratory the tubers were treated for the desired exposure period, the fungicide was then drained off and the tubers allowed to set for two hours in covered containers.

The results of this investigation are presented in table IX.

From the results presented in table IX, it can be concluded that the toxicity values of the mercurials, SCD 765, and Dowicides C and G are decreased as a result of the common field practice in the method of handling treated tubers after removal from the treatment baths. Since this conclusion is based on a very low number of trials and sclerotia used, it can be assumed that the conclusion may not be valid. Further work is required along this line.

TABLE IX

COMPARATIVE STUDY OF COVERING TREATED POTATO TUBERS IN RELATION TO THE TOXIC EFFECT ON SCLEROTIA OF RHIZOCTONIA SOLAMI KÜHN

Material	Time: Min- utes	Number Sclerotia	Number Sclerotia Killed	Per Cent Sclerotia Killed
HgCl ₂ 1-1000 *	3 0	194	92	47.4
HgCl ₂ 1-1000	30	323	313	96.9
Semesan Bel 1-125	10	2 99	299	100.0
Semesan Bel 1-125	15	315	290	92.4
Acid HgCl ₂ 1-1000 *	30	554	554	100.0
Acid HgCl ₂ 1-1000	15	282	2 68	94.7
SCD 765 1-500 *	3 0	50	3 5	70.0
SCD 765 1-500	15	100	64	64.0
Dowicide C 1-1000 *	30	183	180	98.4
Dowicide C 1-1000	15	140	125	89 .2
Dowicide G 1-1000 *	30	4 96	474	95.6
Dowicide G 1-1000	15	60	53	88.3
NaOCl 1:2 Check	5	80	30	37.5
* N	ot cover	ed		

Summary

Certain desired tuber treatment fungicide specifications have been presented. These fungicides should be non-phytotoxic, non-mercurial, low in cost, non corrosive, present no or very little health hazard, and be as effective as the standard corrosive sublimate.

Sclerotia of R. solani Kühn from treated tubers were planted aseptically on acidulated agar plates according to the method of Sanford and Marritt (27) to prevent bacterial contamination.

Sanford and Marritt have suggested that the numbers of small, medium, and large sclerotia occur in nature in an approximate ratio of 13:3:1. In tuber treatment experiments it has been indicated that size of sclerotia is not always in itself a limiting factor in tuber treatments.

The mercurials are excellent fungicides but may be phytotoxic, tend to increase potato scab, are corrosive, and present a health hazard.

Some of the new Dowicides were used in experiments by the author.

Laboratory results indicate that they are excellent fungicides at high dilutions and do not have the undesirable characteristics of the mercurials.

The addition of a detergent, Vel, to the experimental fungicides, decreased the effectiveness of the Dowicides but had little effect on the mercurials.

It is concluded from the data presented that Dowicides B, C, F, and G are excellent fungicides for control of Rhizoctonia on potato

tubers in the laboratory. Also in the absence of soil infestation they should give satisfactory control in the field.

Of these four materials, Dowicides F (sodium tetrachlorophenol) appears to be one of the better fungicides for potato tuber treatment.

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PLATE I

Sclerotia of Rhizoctonia solani Kuhn on Potato tubers.

Note the Variability of Size of the Sclerotia.



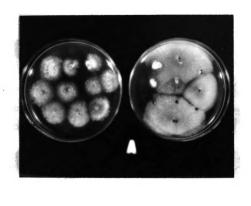




PLATE II

Effect of treatment on growth of sclerotia of Rhizoctonia solani Kühn.

- A. Left: Check (MaOCl 5 minutes).
 - Right: SCD 765 (1-500) 15 minutes
- B. Left: Check (NaOCl 5 minutes).
 - Right: Dowicide G (1-1000) 15 minutes.



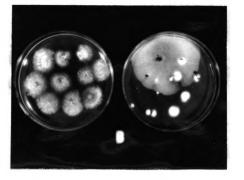


PLATE III

Effect of treatment on growth of sclerotia of Rhizoctonia solani Kühn.

- C. Left: Check (NaCCl 5 minutes).
 - Right: Dowicide C (1-1000) 15 minutes.
- D. Left: Check (MaOCl 5 minutes).
 - Right: Dowicide D (1-1000) 2 hours.

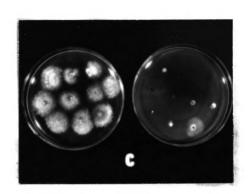


PLATE IV

Effect of treatment on growth of sclerotia of Rhizoctonia solani Kuhn.

E. Left: Check (NaCCl - 5 minutes).

Right: $HgCl_2$ (1-1000) - 30 minutes.

ROOM USE ONLY



