BULLETIN No. 370.

THE EFFICIENCY OF FORMALDEHYDE IN THE TREATMENT OF SEED POTATOES FOR RHIZOCTONIA.

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SUMMARY.

The object of this investigation was to determine the relative efficiency of the standard scab treatments in the disinfection of seed potatoes affected with Rhizoctonia. Experiments were made with formaldehyde gas, formaldehyde solution, and corrosive sublimate solution.

It was found that neither formaldehyde gas nor formaldehyde solution can be depended upon to kill all of the Rhizoctonia sclerotia. The principal reason for this appears to be the inability of the formaldehyde to penetrate readily to the center of the larger and more compact sclerotia. Also, the efficiency of the gas treatment depends to a considerable extent upon the quantity of tubers per cubic foot of space in the disinfection chamber. Other things being equal, the smaller the quantity of potatoes the greater the efficiency. Neither temperature nor humidity are factors of much importance.

The standard corrosive sublimate treatment, on the contrary, is thoroughly efficient. Even with a 1-to-2000 solution (half standard strength) all Rhizoctonia sclerotia are killed.

The conclusion reached is, that when it is desired to treat seed potatoes for Rhizoctonia the corrosive sublimate treatment should be used.

INTRODUCTION.

The numerous fumigation experiments made in the course of the investigation reported in Bulletin No. 369 of this Station offered an excellent opportunity for observing the effect of formaldehyde gas on the Rhizoctonia adhering to potato tubers. Potatoes in various quantities and under various conditions of temperature and moisture were subjected to the formaldehyde gas treatment, and the effect on Rhizoctonia noted. In other experiments Rhizoctonia-infested tubers were dipped in solutions of formaldehyde and corrosive sublimate as

recommended for the prevention of scab. The effect of the treatment was determined by comparing the growth of the Rhizoctonia in artificial cultures made before and after treatment. In this respect, our methods differ from those used by previous investigators. Heretofore, the efficiency of the treatment has been determined, in most cases, by means of field experiments and the results have been more or less unsatisfactory. They have been affected by outside factors such as climatic conditions, texture and drainage of the soil, insects and other fungus diseases. By the culture method such factors are eliminated.

THE DISEASE.

The disease under consideration is that caused by *Rhizoctonia solani* Kühn which is the sterile form of *Corticium vagum* B. & C. var. *solani* Burt.[1] It has been known in Europe for many years. Its occurrence in America was first reported by Duggar and Stewart [2] in 1901, and it is now found in all portions of this country where the potato is extensively grown. It is recognized on the surface of the tubers by the masses of resting mycelium or sclerotia. These sclerotia-like bodies are often overlooked, for they may be mistaken for soil clinging to the potato. By immersing the tubers in water or washing them free from all soil, the sclerotia can readily be distinguished as dark brown bodies which cling tenaciously to the surface of the tuber. Plate I shows a tuber with the sclerotial bodies upon it. When the fungus is in this form it is of little importance as the tubers are not injured by it, but affected tubers may disseminate the disease, and in very bad cases the market value of the tubers may be lowered.

To this fungus is attributed, to a great degree, the so-called "skips" in the planted row. Rolfs,[3] in 1902 and 1904, held it responsible for the failure of potato crops in Colorado. The fungus may live on the organic matter in the soil or it may live as a parasite attacking the stems, stolons, or roots of the plants. At the point of attack, cankers are formed which in time may increase so

[1] Probably the same as *Hypochnus solani* Prill. & Delacr. (Clinton, Conn. Sta. Rpt. for 1904 : 326; Riehm, *Mitt. K. Biol. Anst. f. Land u. Forstw*, 11 : 23. 1911).

[2] Duggar, B. M., and Stewart, F. C. The Sterile Fungus Rhizoctonia. Cornell Sta. Bul. 186. 1901; also N. Y. (Geneva) Sta. Bul. 186. 1901.

[3] Rolfs, F. M. Potato Failures. Colo. Sta. Bul. 70. 1902; and Bul. 91. 1904.

as to girdle the plant or separate portions from the main stem. That a wound appears to be necessary for the fungus to enter the tissue is suggested by Clinton's [4] observations. He has found the "inconspicuous grayish mealy growth" of the Corticium or fruiting stage on the stalks of the potatoes near the surface of the ground, and no injury was done to the stem at that point. He visited several fields and found that 15 to 20 per ct. of the plants showed this condition. To this same fungus Selby [5] attributes the rosette of the potato, and Rolfs [6] reports that the occurrence of aerial tubers may be explained partially by its presence. However there is no doubt that other factors may bring about similar effects.

PREVIOUS LITERATURE.

Since Loew's discovery [7] of the germicidal action of formaldehyde, in 1888, a great deal has been written on the subject of fumigation and disinfection for bacteria and fungi. The disinfection of seed potatoes was first taken up by Bolley [8] who has recommended the use of corrosive sublimate (mercury bichloride) solution for potato scab. Later, Arthur [9] found that a formaldehyde solution gave just as good results and was safer to handle. These experiments have been repeated and the conclusions confirmed by numerous other experimenters. The formaldehyde gas treatment for seed potatoes orginated with Jones and Morse as described in Bulletin No. 369, page 386.

It is generally taken for granted that the treatment made for scab is also effective in killing Rhizoctonia. Rolfs [10] conducted extensive field tests of the dip method of treating Rhizoctonia, and concluded that the mercury bichloride solution was effective in controlling it; also that formaldehyde solution gave less favorable results. Selby,[11] in his first report on the potato rosette (which

[4] Clinton, G. P. Report of Botanist. Conn. Sta. Rpt. for 1904:325.
[5] Selby, A. D. A Rosette Disease of Potatoes. Ohio Sta. Buls. 139 and 145. 1903.
[6] *Loc. cit.* Bul. 70.
[7] Loew, O. Physiologische Notizen über Formaldehyd. Ber. Gesell. Morph. u. Phys. zu Munchen. 1888.
[8] Bolley, H. L. Potato Scab and Possibilities of Prevention. N. Dak. Sta. Bul. 4. 1891.
[9] Arthur, J. C. Formaldehyde for Prevention of Potato Scab. Ind. Sta. Bul. 65. 1897.
[10] *Loc. cit.* Bul. 91.
[11] *Loc. cit.* Bul. 139:58.

he attributed to Rhizoctonia), believes the formalin more effective than the corrosive sublimate bath. He has noted the yields of the treated and untreated tubers and used the appearance of the foliage as a criterion in determining the effectiveness of the treatment. He states, "the warranted conclusion, drawn from tests of two seasons (1901 and 1902) with formalin and the extended work of many seasons at the Station with corrosive sublimate appears to be that corrosive sublimate seed treatment does not prevent the Rhizoctonia disease to any appreciable extent, while the formalin seed treatment, as shown conspicuously by study of the growing plants and usually in the yields of tubers, does prevent the disease to a very marked extent." In Selby's second report [12] of his investigations on the treatment of Rhizoctonia, there are recorded experiments in which treated and untreated tubers were planted in infected soil as well as in soil in which the disease was not supposed to be present. The Carman variety is the only one that received treatment with the standard substances and was planted in infected soil and grown apparently under similar conditions. He used formalin at the rate of one pound (pint) to 30 gallons of water, immersing the tubers for two hours. It is uncertain what strength of mercury bichloride solution was used for he states, "the usual strength of one ounce to 16 gallons of water was employed," and in this solution the tubers remained for one hour. However, this is one-half the strength used by Bolley [13] who recommends one ounce to $7\frac{1}{2}$ gallons of water, and an immersion for $1\frac{1}{2}$ hours. The average yields from the check, formalin, and corrosive sublimate treated plots are calculated at 139.9, 131.4, and 127.1 bushels per acre, and the percentages of harvested tubers showing Rhizoctonia are 62, 46, and 29.5 per ct., respectively. The yields from the treated tubers are slightly smaller than those from the untreated, but the least percentage of Rhizoctonia is found on the product of tubers treated with mercury bichloride solution.

Selby also treated Rhizoctonia-infested tubers with formaldehyde solution for various periods of time. The Early Trumbull variety was immersed in standard formalin solution for 2, $3\frac{1}{2}$, and $4\frac{1}{2}$ hours and planted in infected soil under apparently similar conditions. The check plots showed 94 per ct. of the harvested tubers infected with Rhizoctonia, while the tubers treated for 2, $3\frac{1}{2}$, and $4\frac{1}{2}$ hours

[12] *Loc. cit.*
[13] *Loc. cit.*

showed 87, 45, and 48 per ct. of the tubers attacked by it. Though no comment was made on these results it appears that a longer immersion than two hours is more effective in killing the fungus.

Güssow [14] conducted field experiments in which lots of seed tubers attacked by Rhizoctonia were immersed for 3 hours in a solution of one part mercury bichloride to 2000 parts of water (equal to 1 ounce to 15 gallons of water), and, also in a bath of formalin made up of one half pint to 15 gallons of water. At the harvest it was found that the tubers treated with formalin were almost as badly covered with Rhizoctonia as were the original potatoes, while those immersed in mercury bichloride solution were practically free.

PRESENT TESTS.

METHODS.

To ascertain the efficiency of the formaldehyde gas, three or more Rhizoctonia-infected potatoes were placed in the fumigator along with the various quantities of tubers that were used in the experiments made for the determination of factors involved in tuber injury. The chemicals used, the fumigator, the quantity of tubers, and the conditions of moisture and temperature under which these experiments were conducted are described in Bulletin No. 369.

For comparison with the results obtained by the gas method of treating seed potatoes, experiments were also conducted in which the standard immersion or dip methods were employed. Rhizoctonia-diseased tubers were immersed in solutions of formaldehyde and mercury bichloride for 2 and 1½ hours respectively. The standard strength of formalin was used unless otherwise noted, namely, one pint of formalin (a 37 per ct. solution of formaldehyde) to 30 gallons of water. Of mercury bichloride, two different strengths were used, namely, one part of the salt to 1000 and 2000 parts of water. The latter strength is one-half that usually employed in practical disinfection. After immersion in these solutions the potatoes were permitted to drain and dry under sterile conditions.

Before and after each experiment cultures were made of the untreated and treated sclerotia to determine the efficiency of the treatment. In making these cultures the usual precautions were used to maintain sterile conditions in handling instruments, petri

[14] Güssow, H. T. Canada Expt. Farms Rpts. 1912:200.

dishes, potatoes, etc. At the outset, sclerotia were placed on potato agar of one per ct. acidity, but it was found that the plates were soon overrun by the potato bacillus. This was generally the case when sclerotia from untreated or formaldehyde-treated tubers were taken. The spores of this bacillus (*Bacillus vulgatus* Trevisan) are very resistant, and are invariably found on potatoes. An idea of their resistance can be secured from the fact that the usual method of obtaining this organism is by boiling potatoes for one-half hour, halving them, and incubating in a sterile moist chamber.[15] These bacteria were kept down by adding two drops of 50 per ct. lactic acid to each tube of about 10 cubic centimeters of the medium. By omitting the lactic acid, the effect of the treatment on the bacteria that are found in the sclerotia and on the surface of the tuber was determined.

That Rhizoctonia on acidulated agar grows well without interference from bacteria has been noted by Duggar and Stewart.[16] However, Duggar later states that Rhizoctonia is apparently not readily affected by weak alkalies or acids.[17] F. C. Stewart, in some of his unpublished work on the carnation Rhizoctonia, has shown that the growth of the fungus is more rapid on agar which is slightly acid than on neutral or slightly alkaline media, while on more strongly acidulated agar the growth again becomes less rapid. In the writer's work it was found that when two drops of 50 per ct. lactic acid were added to a tube containing about 10 cubic centimeters of one-per-ct. acid potato agar, the growth of potato Rhizoctonia was about one-half as fast as on the same medium without the lactic acid. In Tables I–III these two kinds of agar are designated as "strongly acid agar" and "agar, one per ct. acid."

Generally, five sclerotia were placed in each petri dish and allowed to develop at room temperature. After three to five days the nature of the results from the untreated sclerotia could be ascertained. In case of the treated sclerotia, the cultures were examined from time to time and kept ten days before they were discarded. At the end of ten days the cultures were examined by means of the low power of the microscope.

[15] Frost. Laboratory Bacteriology, p. 95. 1909.
[16] *Loc. cit.* N. Y. (Geneva) Sta. Bul. 186 : 7.
[17] Duggar, B. M. Fungous Diseases of Plants, p. 453. 1909.

Table I shows the results of the cultures made from sclerotia taken from untreated tubers and tubers subjected to the formaldehyde gas treatment. Unless otherwise stated, the quantities of the chemicals used in the fumigator are 14.25 grams of potassium permanganate and 30 cubic centimeters of formalin, which is equal to 23 ounces of the salt and 3 pints of the liquid for every 1000 cubic feet of space. The exposure was usually for 24 hours and any change in exposure is noted. Where the quantity is expressed in pounds, it is to indicate the number of pounds of tubers per cubic foot of space in the fumigator; otherwise, the number given shows the total number of tubers exposed at one charge. As each experiment is numbered, the condition of the potatoes before they went into the fumigator and the effect of the gas upon them can be readily learned by referring to the corresponding experiments in Tables I and II, Bulletin No. 369. The data in regard to the efficiency of the gas treatment are summarized in Table II of the present bulletin; while Table III shows the results of experiments in which the liquid treatment was used.

TABLE I.—EFFECT OF FORMALDEHYDE GAS ON THE SCLEROTIA OF RHIZOCTONIA.

QUANTITY OF POTATOES.	TREATED.												UNTREATED.					
	Number of experiment.	HUMIDITY.			TEMPERATURE.			NUMBER OF CULTURES ON STRONGLY ACID AGAR.*					AGAR, 1 PER CT. ACID.		NUMBER OF CULTURES ON STRONGLY ACID AGAR.			
		Initial.	Maximum.	Closing.	Initial.	Maximum.	Minimum.	Sterile.	Showing growth of Rhizoctonia.	Other fungi.	Total.	Percentage Rhizoctonia.	Bacterial colonies.	Sterile.	Sterile.	Showing growth of Rhizoctonia.	Other fungi.	Total.	Percentage Rhizoctonia.
Tubers.		*Per ct.*	*Per ct.*	*Per ct.*	*Deg. F.*	*Deg. F.*	*Deg. F.*					*Per ct.*							*Per ct.*
36	59	45	—	77	50	58	—	13	2	0	15	13	4	1	1	12	2	15	80
36	65	48	88	79	63	66	57	15	0	0	15	0	0	5	2	2	6	10	20
23	72	71	96	83	53	61	53	15	0	0	15	0	—	—	0	9	6	15	60
23	73	50	89	82	68	71	60	15	0	0	15	0	—	—	1	6	3	10	60
Lbs. per cu. ft.																			
1.5	50	66	85	78	55	63	55	15	0	0	15	0	3	2	0	8	7	15	53
1.5	67	46	78	78	69	69	58	11	4	1	15	26	1	4	0	4	11	15	26
1.5	76	79	91	86	54	56	53	13	2	0	15	13	—	—	1	10	4	15	66
1.5	80	66	82	70	50	51	49	15	0	0	15	0	—	—	0	14	1	15	93
1.5	85	69	91	77	54	54	51	15	0	0	15	0	—	—	0	10	5	15	66
2	45	53	84	85	53	57	49	13	1	1	15	7	5	0	0	4	11	15	26
2	52	85	85	73	40	42	40	11	4	0	15	26	5	0	0	10	5	15	66
2	56	70	92	86	43	46	43	13	1	1	15	7	—	—	0	13	2	15	86
2	79	62	81	74	49	50	48	5	4	6	15	26	—	—	0	15	0	15	100
2	83	89	94	90	50	51	49	15	0	0	15	0	—	—	0	9	6	15	60
3	43	54	84	78	54	60	54	20	0	0	20	0	0	5	2	6	7	15	40
3	51	60	77	77	45	46	40	15	0	0	15	0	5	0	0	9	1	10	90
3	57	77	89	85	46	50	46	14	0	1	15	0	5	0	0	12	3	15	80
3	61	58	89	87	48	48	37	10	4	1	15	27	4	1	0	13	2	15	86
3	63	58	80	78	47	55	47	7	5	3	15	33	2	3	0	15	0	15	100
3	64	64	86	83	65	65	55	15	0	0	15	0	5	0	0	13	2	15	86
3	66	51	80	78	69	69	61	7	2	1	10	20	5	0	0	7	3	10	70
4	42	61	87	76	54	58	53	15	0	0	15	0	5	0	0	7	8	15	47
4	47	66	77	76	62	68	62	15	0	0	15	0	5	0	0	7	8	15	47
4	58	62	91	91	49	50	45	9	5	1	15	33	5	0	0	15	0	15	100
4	70	59	88	88	47	51	47	12	3	0	15	20	—	—	0	10	5	15	66
4	77	80	89	83	53	55	52	12	3	0	15	20	—	—	0	14	1	15	93
4	84	90	96	91	50	52	50	13	1	1	15	7	—	—	1	12	2	15	80

*The acidulation of media is described on page 422.

5	41	60	90	89	54	57	54	15	0	0	15	0	5	0	2	4	9	15	27
6	29	40	74	73	64	67	57	20	0	0	20	0	——	——	6	3	6	15	33
8	27	62	73	73	53	55	52	24	1	0	25	4	5	0	4	10	7	21	47
8	53	83	87	81	40	42	40	11	3	1	15	20	5	0	1	6	8	15	40
8	60	49	94	94	59	59	49	9	6	0	15	40	5	0	0	14	1	15	93
8	74	47	94	94	78	87	70	34	5	1	40	12.5	3	0	0	14	1	15	93
10	25	75	92	92	46	48	44	25	3	5	33	9	5	0	0	8	12	20	40
12	22	80	90	89	51	54	51	29	0	1	30	0	5	0	3	7	10	20	35
12	54	88	93	93	42	42	42	14	7	4	25	28	5	0	1	9	15	25	38
12	62	44	79	79	41	44	41	10	3	2	15	20	5	0	0	12	3	15	80
12	75	67	96	96	65	65	53	22	16	2	40	40	5	0	0	15	0	15	100
14	19	89	98	96	52	53	50	13	2	5	20	10	——	——	1	4	8	13	31
16	13	65	78	78	46	53	46	8	11	1	20	55	5	0	2	3	10	15	20
16	16	51	79	79	51	52	49	21	4	0	25	18	10	0	0	11	4	15	73
16	55	93	94	93	43	43	43	15	3	2	20	15	5	0	0	15	.....	15	100
16	86	51	104	*104	59	59	54	22	12	11	45	26	——	——	0	8	7	15	53
Exposed for three hours.																			
23 tubers	68	88	95	95	51	52	51	10	9	1	20	45	5	0	0	15	0	15	100
23 "	69	95	100	100	52	54	52	20	0	0	20	0	0	5	0	15	0	15	100
23 "	71	52	93	93	48	50	48	12	3	0	15	20	——	——	0	12	3	15	80
Lbs. per cu. ft. — *Exposed for 24 hours, but double quantity of chemicals were used.*																			
4	78	75	93	89	52	54	50	18	1	1	20	5	——	——	0	15	0	15	100
3.7 Dry	89	——	——	——	46	——	45	25	9	6	40	22.5	——	——	2	5	18	25	20
3.7 Wet	89	——	——	——	46	——	45	47	1	2	50	2	——	——					

*No doubt there was a supersaturated atmosphere in the fumigator for when it was opened (fumigator was out in the rain) the relative humidity fell to 100 per ct.

TABLE II.—EFFECT OF FORMALDEHYDE GAS ON RHIZOCTONIA; SUMMARY.

QUANTITY OF POTATOES.	Number of experiments.	TREATED.		UNTREATED.	
		Total number of cultures.	Average percentage showing growth of Rhizoctonia.	Total number of cultures.	Average percentage showing growth of Rhizoctonia.
			Per ct.		*Per ct.*
Exposed for 24 hours: Standard charge.					
23 to 36 tubers	4	60	3.3	50	55
1.5 lbs. per cu. ft.	5	75	7.8	75	60.8
2 " "	5	75	15.2	75	67.4
3 " "	7	105	11.4	95	78.8
4 " "	6	90	13.3	90	72.1
5 " "	1	15	0	15	27
6 " "	1	20	0	15	33
8 " "	4	95	19.3	66	68.2
10 " "	1	33	9	20	40
12 " "	4	110	22	75	63.2
14 " "	1	20	10	13	31
16 " "	4	110	28.5	60	54
Exposed for 3 hours.					
23 tubers	3	55	21.6	45	93.3
Exposed for 24 hours, but double quantity of chemicals.					
4 lbs. per cu. ft.	1	20	5	15	100
3.7 lbs. per cu. ft., dry	1	40	22.5	25	20
3.7 lbs. per cu. ft., wet	1	50	2		

DISCUSSION OF RESULTS.

The data given in Table I and summarized in Table II, show that the formaldehyde gas treatment is not reliable; that is, it is not efficient in killing Rhizoctonia and some other fungi and bacteria which occur on the surface of the seed potatoes. Humidity and temperature do not appear to be responsible for the irregularities in the efficiency of the treatment. Where several experiments were conducted with equal quantities of tubers, the average results of such experiments show that there is a lesser degree of efficiency when there is a large quantity of tubers in the fumigator. However,

in individual cases where small quantities of tubers are used, the percentage of Rhizoctonia not killed may be almost as high as the average percentage for large quantities of tubers.

TABLE III.— EFFECT OF FORMALDEHYDE AND CORROSIVE SUBLIMATE SOLUTIONS ON RHIZOCTONIA.

Number of experiment.	Proportions.	Time of immersion.	Temperature C.	Number of cultures on strongly acid agar.					Agar, one per cent acid.	
				Sterile.	Showing growth of Rhizoctonia.	Other fungi.	Total.	Percentage showing growth of Rhizoctonia.	Bacterial colonies.	Sterile.
	FORMALIN SOLUTION.									
1....	1-240	2 hrs.	17.5	12	0	3	15	0	15	0
2....	1-240	2 "	19	73	22	17	112	20	30	0
3....	1-240	2 "	19	16	10	34	60	17	10	0
4....	1-240	2 "	19	6	8	16	30	21	10	0
5....	1-240	2 "	21	23	8	4	35	23	5	0
6....	1-240	2 "	19.5	1	16	8	25	64	——	——
7....	1-240	2 "	22.5	6	4	15	25	18	——	——
8....	1-240	2 "	20	9	10	6	25	40	——	——
Total and average.....			19.6	146	78	103	327	23.85	70	0
1....	1-240	24 "	17.5	5	0	0	5	0	5	0
5....	1-240	24 "	21	18	0	2	20	0	3	2
6....	1-240	6.75"	19.5	8	0	2	10	0	5	0
9....	1-60	2 "	21	49	3	8	60	5	7	3
10....	1-20	2 "	18	15	0	0	15	0	1	4
	MERCURY BICHLORIDE (CORROSIVE SUBLIMATE) SOLUTION.									
3....	1-1000	1.5 "	19	60	0	0	60	0	0	10
4....	1-1000	1.5 "	19	40	0	0	40	0	0	10
9....	1-1000	1.75"	21	70	0	0	70	0	0	10
5....	1-2000	1.5 "	21	40	0	0	40	0	0	5
6....	1-2000	1.5 "	19.5	27	0	0	27	0	2	3
7....	1-2000	1.5 "	22.5	20	0	0	20	0	——	——
8....	1-2000	1.5 "	20	40	0	0	40	0	——	——
Total and average.....			20	297	0	0	297	0	2	38

In Experiment 89 (Table I), there is compared the efficiency of formaldehyde gas on the sclerotia having different moisture content.

In this experiment, the tubers designated as dry were cleaned by dusting off the excess dirt, while the tubers designated as wet were washed with water. Just previous to placing these potatoes in the fumigator, the washed tubers were immersed in distilled water for 45 minutes, and then all excess water was wiped from the surface. The 80 pounds (equal to 3.7 pounds per cubic foot) of potatoes were subjected for 24 hours to the gas evolved from a double quantity of chemicals. Sclerotia of about the same size and texture were used in making the cultures. As there was a slight leakage of the gas from the fumigator these results are only comparative. Of the 40 sclerotia taken from the dry tubers, $22\frac{1}{2}$ per ct. developed Rhizoctonia, while from the 50 moistened sclerotia but 2 per ct. showed growth. This experiment indicates that the state of dessication of the sclerotia has an important bearing on the results of fumigation. One would be led to believe that the humidity of the atmosphere ought to have some effect on the efficiency of this gas, but the data in Table I show no correlation between humidity and efficiency.

The data in Table III show that by the formalin dip method as high as 64 per ct. of the sclerotia may still be alive after the treatment, and an average of 8 tests shows that 24 per ct. of the sclerotia were not killed. A longer period of immersion of the tubers gave more satisfactory results than did a two-hour bath, but the deficiency of the treatment can not be overcome by using a stronger solution.

That the formaldehyde, when used in any form, did not kill resistant spores of the potato bacillus when they were inside the sclerotia or particles of soil, is shown where "agar, one per ct. acid" was used as a culture medium. In fact, so prevalent was this organism that it was impossible to make cultures unless additional acid was added to the medium. That other fungi are not killed by formaldehyde treatments when these organisms are inside of the sclerotia is noted in Table I–III under the heading "other fungi." Most of these fungi were species of Penicillium, but several were uncommon or produced no spores, and no effort was made to identify them.

Where a solution of mercury bichloride of a strength of one part of the salt to 1000 parts of water was used, there was no indication of the growth of Rhizoctonia or of any other fungus or bacterium. Even where half of the above amount of salt was used to a like

amount of water the Rhizoctonia was entirely killed. With a total of 297 cultures the efficiency of mercury bichloride in killing the potato Rhizoctonia was 100 per ct.

The inefficiency of formaldehyde can not be attributed to any single cause, for there are several factors which, when combined, may produce variable results. While making cultures of the sclerotia taken from tubers immersed in formaldehyde solution, it was found that the larger sclerotia were often not moistened throughout. Such sclerotia showed a compact, dry mass of mycelium at the center from which the fungus readily grew. The outer layers, however, were spongy and readily crumbled. In some cases the compact small sclerotia showed the same conditions generally found in the larger ones. The weak penetrative power of formaldehyde may be explained, perhaps, by assuming that the formaldehyde unites chemically with the outer layers of sclerotial tissue forming some substance that retards or inhibits further penetration. This is substantiated by the fact that when tubers are immersed in the ordinary formaldehyde solution for 24 hours the sclerotia are all killed, and it is found that the liquid has penetrated them throughout. Dienes,[18] working on the penetration of formaldehyde gas, has shown that after 14 hours bacteria were killed by the gas penetrating through a porous porcelain plate 20 mm. thick. The structure and composition of the sclerotia, however, is not analogous to that of a porous plate for in the former the compact mass of resting mycelium is more or less in a state of dessication, which affects the rate of penetration of this chemical. Mercury bichloride, unlike the formaldehyde, penetrates all sclerotia, regardless of size or compactness, in $1\frac{1}{2}$ hours. Such sclerotia are all killed, and readily crumble into small pieces when crushed. In the formaldehyde gas treatment, the temperature and humidity, as usually encountered, do not appear to be factors that influence the efficiency. McClintic,[19] in disinfecting passenger cars with formaldehyde gas, states that humidity is an important factor in killing bacteria. He maintains that before starting the fumigation the relative humidity should not be lower than 60–65 per ct. In his earlier work

[18] Dienes, L. Uber Tiefwirkung des Formaldehyds. *Ztschr. Hyg. u. Infectionskrank.* 37:43. 1912.

[19] McClintic, T. B. Pub. Health and Marine Hosp. Ser. U. S. Hyg. Lab. Bul. 27. 1906.

on the treatment of the potato scab, Morse [20] states that a temperature of 60–65 degrees Fahr. is more effective than a lower one, and that in moist air the gas is more efficient. Later [21] he states that a temperature above 80 degrees is most effective. However, under ordinary farm conditions, unless specially equipped, it is almost impossible to obtain a temperature of 80 degrees when it is most desirable to treat tubers. In the preceding bulletin, No. 369, temperature does not prove a factor in determining the cause of the injury to the tubers, but humidity shows some evidence of being a causal agent. The data in Table I of the present bulletin, however, indicate that temperature and humidity are less important in the treatment for Rhizoctonia than is usually believed, for under almost similar conditions widely variable results were obtained. One of the most important factors concerned in the inefficiency is the quantity of tubers in the fumigator. Where several experiments were conducted with the same quantity of tubers the averages show least efficiency where potatoes at the rate of 16 pounds (equal to 167 bushels per 1000 cubic feet) per cubic foot were used. In Bulletin No. 369 it is shown that the factor of adsorption is dependent upon the quantity of tubers in the fumigator. The writer believes that if a sufficient number of experiments were conducted the data would show that there is a gradual decrease in the efficiency of the gas as the quantity of tubers is increased.

The conclusion drawn from laboratory experiments that the formaldehyde gas and liquid treatments are inefficient in killing the Rhizoctonia is partially substantiated by other writers. Rolfs [22] and Güssow [23] conclude, from the results of field experiments, that the mercury bichloride gives better results than the formaldehyde solution. Selby [24] came to an opposite conclusion, namely, that the formalin dip method was better than the corrosive sublimate treatment. However, so far as the control of Rhizoctonia is concerned this conclusion appears unwarranted. The plats in his experiments were very small, mostly on infected soil and the yields irregular. Moreover, the causal relation of Rhizoctonia to rosette is uncertain.

[20] Morse, W. J. Potato Diseases in 1907. Me. Sta. Bul. 149:316. 1907.
[21] Morse, W. J. Blackleg: A Bacterial Disease of Irish Potato. Me. Sta. Bul. 174:325. 1909.
[22] Rolfs, F. M. Colo. Sta. Buls. 70 and 91.
[23] Güssow, H. T. Canada Expt. Farms Rpts. 1912:200.
[24] Selby, A. D. Ohio Sta. Buls. 139 and 145. 1903.

The field experiments of many writers indicate that formaldehyde is as efficient as the mercury bichloride solution in killing potato scab, but there are certain indications that make it appear doubtful if such is actually the case. The difficulty with which formaldehyde penetrates Rhizoctonia sclerotia, and its inability to kill certain other fungi and the potato bacillus suggest that it may, sometimes, be unable to reach and kill the scab organism which, according to Lutman,[25] may be imbedded in the corky tissue of the scab lesions. Also, since adsorption plays such an important role in the formaldehyde gas treatment the efficiency would be least where a large quantity of tubers was fumigated. Unfortunately, the culture method can not be used successfully in determining the efficiency of the treatment on scab. Although it is not difficult to obtain pure cultures of the scab organism it does not grow on culture media readily enough to make this method practicable.

The results of this investigation, considered in connection with those recorded in Bulletin No. 369, seem to warrant the following conclusions: The disinfection of seed potatoes by means of formaldehyde gas is not to be recommended except in cases in which it is impracticable to use either of the liquid treatments. When the treatment is applied for scab alone either corrosive sublimate solution or formaldehyde may be used; but when both Rhizoctonia and scab are involved the corrosive sublimate solution is to be preferred.

[25] Lutman, B. F. The Pathological Anatomy of Potato Scab. *Phytopathology* 3:261. 1913.