ACETIC ACID AND PYROLIGNEOUS ACID IN COMPARISON WITH FORMALDEHYDE AS SOIL DISINFECTANTS¹

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INTRODUCTION

Formaldehyde is one of the most commonly used of soil disinfectants. Its efficacy against fungi in the soil is well established, but it has at least two faults. The cost of the treatment is relatively high and the time which must be allowed to elapse between treatment and seeding, if the treatment is not to injure germination, is sometimes objectionable. The principal object of the work here described was to find a soil disinfectant as effective against fungi, lower in cost, and less injurious to seeds than is formaldehyde.

In earlier experiments, the results of which have been described by the writer,² 1.0 to 1.2 per cent acetic acid used as a soil disinfectant was found to protect seedlings from damping off. Similar results with acetic acid as a soil disinfectant have since been secured by other investigators.^{3 4}

The evidence presented in this paper verifies the conclusions previously reached in regard to acetic acid and serves as a basis for comparing acetic acid, pyroligneous acid, and formaldehyde as soil disinfectants. In the present paper various dilutions and rates of application of acetic acid, pyroligneous acid, and formaldehyde are compared with each other as regards prevention of damping off (caused by species of Pythium and Rhizoctonia), effect on seed germination, and effect on growth (dry weight) of plants.

METHODS

The soil used is a water-deposited fine sandy loam. In all cases (except in a forest nursery, to which reference is made below) manure was composted with this soil as in ordinary greenhouse practice. The soil prior to the application of the treatments contained water to the extent of 60 per cent of its water-holding capacity (except as otherwise indicated).

For each series of experiments, all seeds were sown the same day, in order that the effects of the treatments on growth of plants might be Treatments were in triplicate and 900 seeds of beet, compared. cucumber, and lettuce were used for each. Seeds were sown at the rate of 50 per linear foot. After germination was completed and damping off had ceased, seedlings were thinned so as to leave the same number per linear foot, in order that competition between plants should not interfere with the effects of soil treatment on growth.

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 ANDERSON, P. J., SWANBACK, T. R., and STRELT, O. E., and others. DAMPING OFF OF YOUNG TOBACCO SEEDLINGS. Conn. Agr. Expt. Sta. Bul. 311: 269-270, illus. 1930.
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Three weeks later the plants were pulled, washed, and dried to constant weight.

The dilutions of pyroligneous acid, acetic acid, and formaldehyde which were applied to soil are recorded in Table 1, as are also the intervals of time which elapsed after soil treatment and before seeding. Except as is otherwise indicated, the diluted chemicals were applied to soil at the rate of $2\frac{1}{2}$ quarts per square foot.

Undistilled pyroligneous acid was used. As described by the manufacturers, it was made by the destructive distillation of hardwood (beech, birch, and maple) in sealed retorts. Pyroligneous acid was found to have certain advantages over either acetic acid or formaldehyde as a soil disinfectant, and the results are accordingly presented. Further work will, however, be necessary before the observed effects of pyroligneous acid on fungi in the soil can be traced to each of its several constituents, since, according to Hawley,⁵ pyroligneous acid is not a chemical compound but contains a number of constituents, including acetic acid, methyl alcohol, formaldehyde, and furfural.

 TABLE 1.—Effects of formaldehyde, acetic acid, and pyroligneous acid on seed germination, damping off of seedlings, and dry weights of plants

Soil treatment	Time inter- val be- tween soil treat- ment and seeding	Germination of seeds of different plants			Plant	seedlings lamped o	Dry weights of 100 plants		
		Beet «	Cucum- ber	Let- tuce	Beet	Cucum- ber	Let- tuce	Beet	Cu- cum- ber
	Days	Number	Per cent	Per cent	Per cent	Per cent	Per cent	Grams	Grams
Check		83	60	45	40	61	45	16.61	30, 50
Formaldehyde 1:50	10	129	84	45	0	0	0	20.78	94.85
Acetic acid 1.19 per cent	5	129	100	72	0	0	0		111.27
Do	13	138	85	60	0	0	0	20.79	
Acetic acid, 1.78 per cent	13	40			0			20.58	
Pyroligneous acid, 1:100	12				1	29	-	35.00	
Pyroligneous acid, 2:100	{ 1	120	92	56	0	0	0		
D0	2	136	89	63	0	0	0		113. 50
D0	3	107	80	80	0	. 16	0		
D0	5	86	93	75	17	4	7		
D0	13	149			6			23.69	
Pyroligneous acid, 3:100	1	128	72	49	0	0	0		
Do	2	83	93	57	2	0	0		118.93
D0	3	108	81	72	0	0	0		
Do	6	92	80	53	0	0	0		
D0	13	139			3			22.66	
Pyroligneous acid, 4:100	1	128	97	60	0	0	0		
D0	2	108	73	56	0	0	0		125.28
D0	3	155	97	68	0	0	0		
D0	5	124	72		0	0	0		
Do	7	124	83	45	0	0	0		
D0	13	141			0			17.26	
Pyroligneous acid, 5.100	13				0	0			38.73
Pyroligneous acid, 10:100	13				0	0		27.40	

" Number of seedlings which came up for each 100 beet seed balls sown.

⁵ HAWLEY, L. F. WOOD DISTILLATION. 141 p., illus. New York. 1923.

EFFECTS OF SOIL DISINFECTANTS ON DAMPING OFF OF SEEDLINGS

The average percentages of seeds which germinated, seedlings which damped off, and dry weight of plants are recorded in Table 1. Damping off was severe in untreated soil, for in it 40 per cent of the beet seedlings, 61 per cent of the cucumber seedlings, and 45 per cent of the lettuce seedlings damped off. There was no damping off in soil to which formaldehyde 1:50 (1 gallon of formaldehyde with 49 gallons of water) had been applied 10 days before seeding.

All damping off of the seedlings of beet, cucumber, and lettuce was also prevented by acetic acid 1.19 per cent applied to soil 5 or 13 days before seeding and at the rate of $2\frac{1}{2}$ quarts per square foot. In other experiments conducted by the writer damping off has been prevented equally well by acetic acid 1.19 per cent applied to soil at the rate of 2 quarts per square foot. Thus used, the cost of the acetic-acid treatment, per unit area of soil, is about three-fourths of the cost of soil treatment with formaldehyde.

In connection with this use of acetic acid, experiments with vinegar as a soil disinfectant were undertaken, for vinegar is, of course, readily available to every farmer and gardener. The user of vinegar is relatively certain of obtaining a standardized product as regards content of acetic acid, for, according to the standard adopted in enforcing the Federal food and drug act,⁶ cider or apple vinegar (also grape or wine vinegar, malt vinegar, sugar vinegar, glucose vinegar, and spirit vinegar) must contain not less than 4 g of acetic acid per 100 cc. Such vinegar when diluted by the addition of 21/2 parts of water to 1 part of vinegar (by volume) will, therefore, contain in this dilution about 1.16 per cent acetic acid. Vinegar thus diluted was applied to soil (at the rate of 2 quarts per square foot), and 10 days later tobacco seeds were sown in the treated soil and in soil not treated. Seeds germinated well (equally well in both cases), and as may be seen by reference to Figure 1, there was no damping off in soil to which vinegar had been applied, although the disease was severe in the untreated soil.

In earlier experiments,⁷ seedlings of white spruce were protected against damping off by 1.12 per cent acetic acid applied to soil at the rate of 1.64 quarts per square foot seven days before seeds were sown. With the object of improving upon this method for use in forest nurseries, acetic acid (0.47 to 0.80 per cent) was applied to seed beds, at the rate of three-fourths of a quart per square foot, immediately after the seeds of red or Norway pine (Pinus resinosa Sol.) were sown. Damping off was severe in the untreated soil. Living seedlings (in 4 square feet of each seed bed) were counted three months after the date of seeding. The results are recorded in Table 2. Seed germination was improved, damping off was controlled, and the number of seedlings living three months after seeding was increased most (more than 700 per cent) by 0.8 per cent acetic acid (4.2 pounds of 80 per cent acetic acid diluted with water to 50 gallons) applied to the soil at the rate of three-fourths quart per square foot immediately The merits of this treatment for the prevention of after seeding. damping off in a forest nursery are the successful control of damping off, the harmlessness to seeds (of red pine), the relatively small amount of water, and therefore of labor, needed, and the avoidance of delay between soil treatment and seeding.

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⁶ UNITED STATES DEPARTMENT AGRICULTURE, FOOD AND DRUG ADMINISTRATION. DEFINITIONS AND STANDARDS FOR FOOD PRODUCTS. U. S. Dept. Agr., Food and Drug Admin. Ser. and Regulat. Announc., Food and Drug No. 2 (second revision), 19 p. 1931. ⁷ DORAN, W. L. Op. cit. (See footnote 2.)



FIGURE 1.—Tohacco seedlings protected against damping off by soil treatment with pyroligneous acid and with vinegar: A, Pyroligneous acid, 2:100, applied to soil at the rate of 2 quarts per square foot (7 days before seeding); B, vinegar 1 part with water 2½ parts (by volume) applied to soil at the rate of 2 quarts per square foot (10 days before seeding); C, check, no disinfectant usod

Apr. 1, 1932 Acetic and Pyroligneous Acids as Soil Disinfectants 575

TABLE	2Effects	of	soil	treatment	with	various	strengths	of	acetic	acid	on	the
	-	Ū.	damp	ng off of	the se	edlings o	f red pine					

Percentage of acetic acid applied to soil	Living seedlings per square foot (3 months after seeding)	Increase in seed- lings per unit area as compared with check	Percentage of acetic acid applied to soil	Living seedlings per square foot (3 months after seeding)	Increase in seed- lings per unit area as compared with check	
0 (check) 0. 47 0. 67	Number 37 163 265	Per cent 340 616	0. 33 0. 80	Number 236 301	Per cent 538 713	

All damping off of seedlings listed in Table 1 was prevented by pryoligneous acid 10:100, 5:100, and $4:100.^8$ Pyroligneous acid 3:100 prevented all damping off of cucumber and lettuce seedlings, but there was a little damping off of beet seedlings, 2 to 3 per cent, in soil to which pyroligneous acid 3:100 had been applied. In these experiments, pyroligneous acid 4:100 was as effective in preventing all damping off as was formaldehyde 1:50 or acetic acid 1.19 per cent. In other experiments, damping off of seedlings was controlled equally well by pyroligneous acid $3\frac{1}{2}:100$, applied to soil at the rate of 2 quarts per square foot. Thus used, the cost of soil treatment with pyroligneous acid $3\frac{1}{2}:100$ was about 58 per cent of the cost, per unit area, of soil treatment with formaldehyde 1:50.

In the experiments recorded in Table 1 pyroligneous acid 1:100 or 2:100 did not prevent all damping off of the seedlings of beet, cucumber, and lettuce. These concentrations are considered too dilute to be dependable, although in some cases pyroligneous acid 2:100 has given adequate protection. When pyroligneous acid 2:100 was applied to soil at the rate of 2 quarts per square foot seven days before tobacco seeds were sown there was, as may be seen by reference to Figure 1, no damping off of tobacco seedlings grown in soil so treated, although the disease was severe in untreated soil.

EFFECTS OF SOIL DISINFECTANTS ON SEED GERMINATION

In the untreated soil 60 per cent of the cucumber seeds and 45 per cent of the lettuce seeds germinated and 83 beet seedlings came up for each 100 beet seed balls sown. Much of this poor germination was due to the decay of seeds resulting from the attack of dampingoff fungi in the soil.

The germination of the seeds of beet and cucumber was improved, and the germination of the seeds of lettuce was unaffected by formaldehyde 1:50 applied to the soil 10 days before seeding.

The germination of the seeds of beets was injured by 1.78 per cent acetic acid, and on the basis of these and other experiments it is not considered necessary to use a greater concentration of acetic acid than 1.2 per cent for soil disinfection.

The germination of these seeds was improved as much, or more, by 1.19 per cent acetic acid as by formaldehyde, and this was the case in the experiments represented in Table 1, whether acetic acid

⁸ Dilutions of pyroligneous acid to which reference is made in the text and in Table 1 are indicated as number of parts (by volume) of pyroligneous acid in 100 parts of water.

was applied to the soil 5 days or 13 days before seeding. In other experiments by the writer seed germination has sometimes been injured, however, if seeds were planted in less than 10 days after the application of acetic acid to soil, and this is considered the minimum time interval before seeding which is usually safe after soil treatment with either acetic acid or formaldehyde.

In the case of tobacco seed beds the delay which must follow the application of these soil treatments in the spring is sometimes objectionable. This is of course avoidable by applying such treatments in the fall; and, as observed by the writer, best results have been secured in tobacco seed beds when the soil was disinfected with acetic acid in the fall rather than in the spring.

Chemical soil disinfection of tobacco seed beds in the spring may have another disadvantage, for if the soil is very wet, as it often is at that season, neither acetic acid nor formaldehyde as ordinarily applied always prevents all damping off. Earlier investigators⁹ have suggested applying formaldehyde to tobacco seed beds in the fall rather than in the spring if the soil is likely to be very wet. Their conclusions are supported by the results of experiments by the writer, in which formaldehyde 1:50 (2 quarts per square foot) was less effective in preventing damping off when applied to watersaturated soil than it was when applied to soil which, previous to treatment, contained water to the extent of 50 per cent of its waterholding capacity.

As may be seen by reference to Table 1, the germination of the seeds of beet, cucumber, and lettuce was in most cases improved and in no case injured by soil disinfection with pyroligneous acid 2:100, 3:100, and 4:100. Seeds were uninjured even though the interval between soil treatment with pyroligneous acid 3:100 or 4:100 and seeding was reduced to one or two days. In other experiments it was, however, found unsafe to shorten this interval to less than one day or to apply pyroligneous acid 2:100, 3:100, or 4:100 to living plants. When these treatments were applied, at the rate of 1 quart per square foot, to seedlings of beet, cucumber, and lettuce which had begun to damp off, the plants were severely injured. When pyroligneous acid 3:100 was applied to soil at the rate of 2 quarts per square foot, at the same time that the seeds of pepper, lettuce, cucumber, and tomato were sown, the germination of the seeds of pepper and lettuce was injured.

These observations lead to the conclusion that pyroligneous acid like acetic acid or formaldehyde should be applied to the soil before sowing most seeds. But in the experiments above described and with the seeds used it was safe to apply pyroligneous acid to the soil as late as one day before seeding, and this is a matter of convenience which is sometimes important in practice.

EFFECTS OF SOIL DISINFECTANTS ON DRY WEIGHT OF PLANTS

By reference to Table 1 the dry weight of beet and cucumber seedlings in each of the several soil treatments may be compared with the dry weight of plants in untreated soil. There was considerable increase in dry weight of plants following most treatments, and it was greater with cucumber than with beet. The dry weight of plants was increased by the application of formaldehyde to the soil 10 days before seeding. Acetic acid 1.19 per cent was no less beneficial, for in soil to which it had been applied the dry weight of cucumber seedlings was increased more and the dry weight of beet seedlings was increased as much as by formaldehyde. In these, as in other experiments, the application of acetic acid 1.19 per cent, at the rate of 2 or $2\frac{1}{2}$ quarts per square foot of soil, was followed by an improved growth of plants, and the improvement in growth was ordinarily as great as that associated with the use of formaldehyde. Even the use of acetic acid 1.78 per cent, a concentration which may injure seed germination, was without any injurious effect on the growth of beets in soil to which this treatment had been applied 13 days before seeding.

There was no retarding of growth of plants in soil to which pyroligneous acid 1:100, 2:100, 3:100, 4:100, 5:100, and 10:100 had been applied, even though the treatments with pyroligneous acid 2:100, 3:100, and 4:100 were applied to soil only one or two days before seeding. Dry weight of cucumber seedlings was increased more by pyroligneous acid 3:100 or 4:100 applied to soil two days before seeding than by formaldehyde 1:50 applied to soil 10 days before seeding than by formaldehyde 1:50 applied to soil 10 days before seeding. The dry weight of beet seedlings was also increased by these treatments, and the increase, as compared with that which followed the use of formaldehyde, was greater with pyroligneous acid 3:100 and less with pyroligneous acid 4:100. In these and in other experiments by the writer the beneficial effect of soil treatment with pyroligneous acid on growth of plants was no less than with formaldhyde.

SUMMARY

Acetic acid was as safe and as effective a soil disinfectant as formaldehyde, and the cost of soil disinfection with acetic acid was less than with formaldehyde. Damping off of seedlings (of beet, cucumber, and lettuce) was prevented without injury to seed germination and with benefit to growth of plants by soil treatment with 1.19 per cent acetic acid (1 gallon of 56 per cent acetic acid or 2¾ quarts of 80 per cent acetic acid with water to total 50 gallons), applied at the rate of 2 to 2½ quarts per square foot of soil. An application of 2 quarts per square foot was usually enough.

Best results with acetic acid against soil-borne fungi in tobacco seed beds have been secured when the soil was treated in the fall rather than in the spring.

With acetic acid, as with formaldehyde, it was necessary that there be some interval of time, usually 10 days, between soil treatment and seeding; otherwise seed germination was injured.

Damping off of seedlings (of tobacco) was prevented with no injury to germination by vinegar 1 part diluted with water 2½ parts (by volume), applied to soil at the rate of 2 quarts per square foot 10 days before seeding.

Seedlings of red or Norway pine were protected against damping off, and germination was not injured by acetic acid 0.8 per cent (equal to 6 pounds of 56 per cent acetic acid or 4.2 pounds of 80 per cent acetic acid with water to total 50 gallons), applied to soil at the rate of three-fourths of a quart per square foot at the time of seeding. Pyroligneous acid 3:100 to 4:100 applied to soil at the rate of two quarts per square foot protected seedlings from damping off, and this treatment did not injure the germination of the seeds of beet, cucumber, and lettuce even when it was applied to the soil as late as one day before seeding. Soil treatment with pyroligneous acid resulted in an increase in the dry weight of plants. Per unit area of soil treated, the cost with pyroligneous acid was less than with either formaldehyde or acetic acid. Pyroligneous acid was as effective a soil disinfectant as formaldehyde or acetic acid, and safer and cheaper than either.