

THE USE OF AQUA AMMONIA FOR THE CONTROL OF *SCLEROTIUM ROLFSII*, AND ITS RESIDUAL VALUE, IN ISRAEL SOILS

By

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INTRODUCTION

The fungus *Sclerotium rolfii* has for some time caused considerable but sporadic damage to agricultural crops in Israel. But with the increase of intensive farming under irrigation, and especially with the increase in the area planted to sugar beets, the fungus has spread into previously uninfested fields, and the problem of its control has become serious.

No efficient and economical means of control has yet been found. Various fumigants, such as formalin, chloropicrin, chlorobromopropene, and edictol (1,2,5,6,12), have been tried and found to be more or less toxic to *Sclerotium rolfii*. Their disadvantages, however, are that they leave a toxic residue in the soil, are relatively expensive, and do not provide nutrients for plant growth. The possible advantages of ammonia, on the other hand, are that besides having fungicidal properties against certain microorganisms, it also provides nitrogen for plant growth and leaves no deleterious residue in the soil.

The toxicity of ammonia and some of its salts to certain soil fungi (such as *Rhizoctonia tuliparum*, *Phymatotrichum omnivorum*, and *Sclerotium delphinii*), and to nematodes, was proved by several workers (4,9,10). In laboratory experiments performed by Leach and Davey (7), a 50 ppm ammonia solution controlled the mycelium of *Sclerotium rolfii* within 24 hours, while a concentration of 250 ppm was needed to control the more highly resistant sclerotia within the same period. In another experiment by the same workers (8), anhydrous ammonia given at the rates of 22.4, 44.8 and 89.6 kg N per acre, decreased the rotting damage to sugar beet caused by *S. rolfii* by 28, 54 and 65 percent, respectively, in comparison with the untreated control lots. On the other hand, it has been shown (8) that large amounts of aqua ammonia or other nitrogen compounds may fail to kill, or even decrease the viability of, the fungi in the soil.

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In this country, Perlberger (11) concluded from preliminary laboratory trials that aqua ammonia increases the alkalinity of the soil and thus possesses an advantage over other materials for the control of *S. rolfssii*.

The effect of ammonia on the soil has been investigated in laboratory trials by Birecka *et al.* (3). In these trials, ammonia in the amounts used did not show a sterilizing effect on the bacterial microflora of the soil, but it did decrease greatly the fungal population. In the close vicinity of the point of injection, the ammonia was found to undergo rapid nitrification.

The use of ammonia as a nitrogenous fertilizer is spreading fast throughout the world, thanks to its favorable economical, physical and chemical properties in comparison with the solid fertilizers. It is therefore of primary economical importance to determine its efficiency in the control of *S. rolfssii*.

The preliminary trials reported below were concerned with the efficiency of aqua ammonia in controlling sclerotia of *S. rolfssii*, with the effect of ammonia sorption by the soil after treatment, and with the residual effect of ammonia on the growth of maize.

MATERIALS AND METHODS

Barrels, 50 cm in diameter, were filled with soil. Three soil types, described in Table 1, were used.

TABLE 1
THE COMPOSITION OF THE SOIL TYPES USED

Soil texture	Mechanical analysis in %			% Lime (CaCO ₃)	pH
	clay	silt	sand		
Highly sandy	2.8	4.7	92.5	none	7.1
Loam (Negev loess)	18.5	29.3	52.2	16.3	8.2
Red clay-loam	30.2	20.5	49.3	2.4	7.8

Sclerotia of *S. rolfssii*, taken from a culture of diseased sugar beets and grown on a nutrient medium of sterilized oats, were placed in the soil at a depth of 15 cm, and the inoculated spots were marked.

A 25% ammonia solution was then applied to the soil, at rates of 100 and 400 cc per barrel, which are equivalent to about 185 and 740 kg N per acre, respectively. In the case of the larger amount, two applications of 200 cc per barrel, with an eight-week interval in between, were also tried.

Two methods of applying the ammonia were compared: furrowing and flooding. In the first method, two narrow furrows were opened, 20 cm apart, to a depth of 12 cm; immediately after the ammonia injection the furrows were closed and packed with a roller. In the second method, the ammonia was diluted in 10 liters of irrigation water, and the soil surface in the barrels was flooded.

Since ammonia is highly volatile and may easily escape from the soil, an additional treatment, consisting of surface packing the soil after the ammonia application and then covering the surface with paper, for the purpose of preserving the ammonia in the soil, was tried. In the case of the two-time application, tilling the soil between applications, for the purpose of ensuring better mixing and closer contact between the ammonia and the fungi in the upper soil layer, was also tried.

In order to determine the viability of the fungus, soil samples were taken for examination a month after the ammonia application. This time was chosen after daily samplings had shown that the pH, which at first rose greatly in the treated soils, had already returned to the original level. Where the ammonia had been applied by the furrowing method, the samples were taken from between the furrows, in the marked spots, by means of a metal tube. The soil sample, containing the sclerotia which had been placed in the soil prior to treatment, was separated into three layers (0-5, 5-10 and 10-15 cm) in order to evaluate the possible relation between the distribution of ammonia in the soil and its effect on the fungus at different depths. From each soil sample, between 60 and 100 sclerotia were separated, placed in Petri dishes on moist filter paper, and incubated for 48 hours or more at the constant temperature of 30°C. The germinated sclerotia were counted as viable.

Soon after injection, and also two months later, separate soil samples were taken and analyzed, according to the A.O.A.C. methods for pH, total nitrogen, sorbed ammoniacal nitrogen, and nitrate content. The purpose of the pH determinations was to find out when the free ammonia disappears from the soil, and thus when the viability tests should be started. The purpose of the nitrogen determination was to investigate the changes in soil nitrogen following the ammonia injection, and to differentiate the fertilizing effect of the ammonia from its fungicidal effect.

To test the residual effect of the nitrogen, introduced into the soil by the ammonia injection on plant growth, maize (Laguna variety) was planted in the barrels approximately ten weeks after treatment, and harvested after 48 days of growth. Total yield, number of leaves, and the height and weight of the plants were recorded.

RESULTS AND DISCUSSION

1. THE EFFICIENCY OF AQUA AMMONIA IN CONTROLLING *SCLEROTIUM ROLFII*, *The effect of different treatments in red clay-loam*

The flooding and furrowing methods of application were carried out in one type of soil only. The results are presented in Table 2.

TABLE 2

THE EFFECT OF AQUA AMMONIA APPLICATIONS, IN RED CLAY-LOAM, ON THE VIABILITY OF *Sclerotium rolfsii*
(percent of viable sclerotia, means of 3 replicates)

Treatment	Ammonia applied (cc per barrel)	Soil layer (cm)			
		0-5	5-10	10-15	average
Control	—	97	98	100	98
Flooding	100	46	20	27	31
	400	17	22	21	20
	200+200	19	17	2	13
Flooding and tilling	200+200	2	0	0	1
	100	61	48	47	52
Furrowing	400	27	23	20	23
	200+200	7	1	1	3
	200+200	6	3	0	3
Furrowing and tilling	100	66	58	45	56
	400	22	8	3	11
Furrowing and packing and covering	100	57	42	45	48
	400	40	8	1	16

It appears that aqua ammonia is definitely toxic to *Sclerotium rolfsii*. All the treatments used, in all three soil layers examined, resulted in significant reductions in the viability of the sclerotia.

Applications of 400 cc aqua ammonia per barrel resulted in significantly lower viability than corresponding applications of 100 cc per barrel. The average viability percentages ranged between 11 and 23 for the large ammonia amount, against between 31 and 56 for the smaller amounts. In some of the replicates receiving the larger amounts, the viability was almost completely inhibited.

In general, the highest rate of viability was found in the upper (0-5 cm) soil layer.

Using 100 cc aqua ammonia per barrel, or 400 cc in two applications without tilling, the flooding method resulted in significantly better fungus control than the furrowing method. When 400 cc were used in a single application, no clear and consistent difference between the two methods was observed.

Surface packing (after applying the furrowing method) did not increase the effect of the ammonia when 100 cc per barrel were used, and only slightly when 400 cc per barrel were used.

Covering the soil surface with paper, after the surface packing, did not result in better fungus control. Similar negative results were obtained by Avizohar-Hershenson and Palti (1) in trials with CBP.

Dividing the high rate of application into two doses (200 + 200 cc per barrel) gave better results than giving the whole amount at once. This effect

was especially marked when the furrowing method was used, the double-dose treatment resulting in extremely low viability of the sclerotia.

Tilling the soil between the two 200 cc applications resulted in almost total fungus control when the flooding method was used. When the furrowing method was used, the tilling treatment did not improve on the good results obtained with the double-dose treatment without tilling

The effects of aqua ammonia in different soil types

The furrowing treatment with surface packing using only the largest amount of 400 cc aqua ammonia per barrel, was applied to the three soil types described in Table 1. The results of this special trial are presented in Table 3.

TABLE 3

THE EFFECT OF AQUA AMMONIA APPLICATIONS ON THE VIABILITY OF *Sclerotium rolfsii* IN 3 SOIL TYPES
(percent of viable sclerotia, means of 3 replicates)

Soil type	Treatment	Soil layer (cm)			
		0-5	5-10	10-15	average
Sandy	control	69	75	85	76
	ammonia	24	17	23	21
Loam (loess)	control	94	98	100	97
	ammonia	16	12	7	12
Clay-loam	control	96	100	100	99
	ammonia	22	8	3	11

From the data of the table there appears to be no difference, in the viability of sclerotia, between the loess and clay-loam soils. In both soils the viability decreased after treatment with 400 cc ammonia to a profile average of about 12% as against over 98% in the untreated control barrels. The application of aqua ammonia produced a highly significant beneficial effect in the sandy soil as well, but to a lesser degree than in the other two soil types. This fact can be attributed to the greater pores and smaller adsorbing capacity of the sand, which may cause an appreciable amount of the substance to escape into the air. The relatively low percentage of viable sclerotia in the untreated sandy soil, in comparison with the two other types, can perhaps be attributed to the greater warming and drying effect which takes place in the sandy soil and which probably kills some of the incorporated sclerotia.

2. THE SORPTION OF AMMONIA BY THE SOIL FOLLOWING TREATMENT

pH changes

The course of pH changes in red clay loam only, which was similar for all ammonia treatments, is exemplified by Fig. 1.

The soil pH, which was originally (prior to treatment) around 7.8 in the clay-loam soil rose, in general, within a day after treatment to a highly significant

100 cc aqua ammonia per barrel

400 cc aqua ammonia per barrel

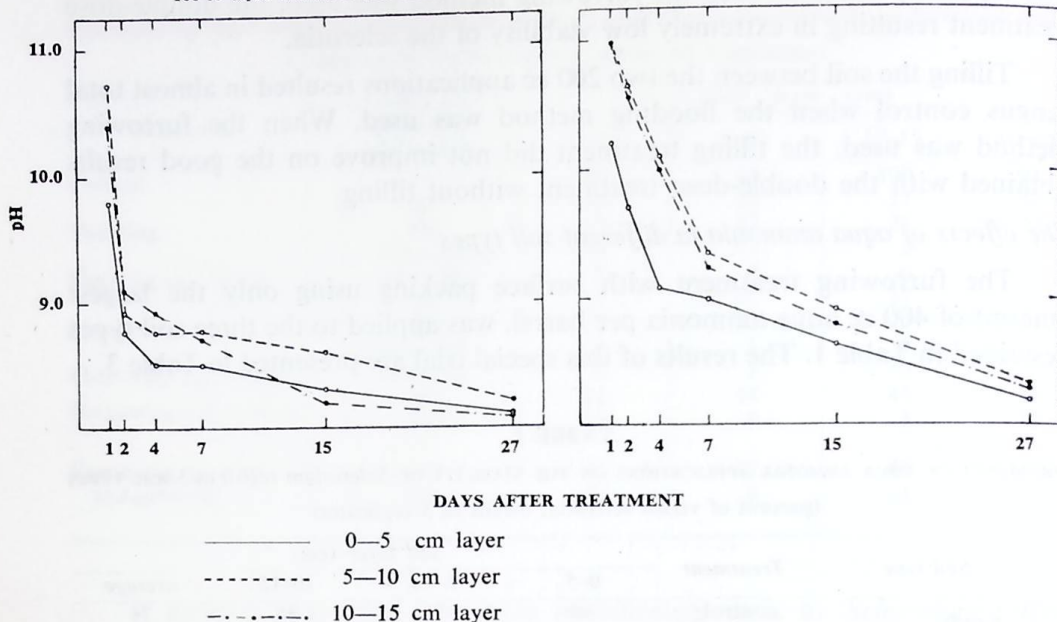


Fig. 1. pH changes in red clay-loam soil following the application of aqua ammonia in furrows and surface packing (means of 3 replicates)

and alkaline reaction (pH 10-11) in the 0-15 cm layer in which the sclerotia were placed. This took place regardless of the amount of ammonia applied and of the application method. On the other hand the rise in pH was greater when the larger amount of ammonia was applied, both in furrows and by flooding; this parallels the effect of the larger amount of ammonia in diminishing the viability of the sclerotia.

In most cases the soil pH attained its highest value 24 hours after treatment and then dropped steeply and returned, after about 5 weeks, to very near the same pH as existed originally (pre-treatment). The strong drop in soil pH can be attributed mainly to (a) the adsorption of free ammonia by the exchange complex, and fixation, and its eventual transformation in the process of nitrification, and (b) the partial volatilization of the free ammonia.

Covering the soil with paper, after the incorporation of ammonia in furrows, did not seem to affect the rise in soil pH. This also parallels the fact that covering the soil following treatment did not result in better control of the sclerotia.

The pH changes in the three soil types, following the application of 400 cc of aqua ammonia per barrel, in furrows and with surface packing, are presented in Table 4.

TABLE 4

pH CHANGE IN 3 SOIL TYPES FOLLOWING THE APPLICATION OF AQUA AMMONIA IN FURROWS AND SURFACE PACKING

(means of 3 replicates)

Days after treatment	Layer (cm)	Soil type		
		clay-loam	loam (loess)	sand
0	0-15	7.8	8.2	7.1
1	0-5	10.2	10.9	9.2
	5-10	11.0	11.4	9.1
	10-15	11.0	11.3	9.2
2	0-5	9.7	10.4	9.0
	5-10	10.7	11.0	8.9
	10-15	10.6	11.0	8.8
4	0-5	9.1	10.0	8.6
	5-10	10.1	10.5	8.8
	10-15	10.0	10.7	8.8
5	0-5	9.0	9.6	7.8
	5-10	9.3	10.4	7.5
	10-15	9.2	10.4	7.4
15	0-5	8.7	9.1	7.6
	5-10	8.9	10.0	7.3
	10-15	8.8	10.2	7.3
25	0-5	8.4	9.2	7.3
	5-10	8.5	9.2	7.2
	10-15	8.7	9.2	7.2
27	0-5	8.2	8.2	7.1
	5-10	8.3	8.6	7.1
	10-15	8.3	8.6	7.1

The ammonia treatment raised the pH in the 0-15 cm layer to highly alkaline values in all the soil types tested. The maximum pH levels attained and the subsequent drop in pH values, were similar in the clay-loam and loess soils. In the sandy soil, however, the pH did not reach the same maximal alkaline level and remained below 9.2 in all three layers. This is probably related to the typical properties of the sandy soil mentioned above. These facts may also explain the lower rate of sclerotia control obtained in the sandy soil in comparison with the other soils.

Nitrogen balance of the soil following treatments

The results of the analysis for nitrogen, which was carried out (in red clay-loam only) two months after treatment, are presented in Table 5.

Every one of the ammonia treatments used resulted in a considerable increase of the total soil nitrogen content. This effect was more pronounced when the larger amount of ammonia was applied.

It is of interest to note that from one third to over one half of the nitrogen added to the soil was in the adsorbed ammoniacal form. With the increase of the amount of ammonia applied, the adsorbed ammoniacal nitrogen content of the soil increased, but not in simple proportion. It can be assumed that in lighter or heavier soils different levels of adsorbed ammoniacal nitrogen, following the application of aqua ammonia, will be found.

TABLE 5

THE SORPTION OF NITROGEN BY RED CLAY-LOAM (0-15 cm LAYER) FOLLOWING TREATMENTS WITH AQUA AMMONIA
(means of 3 replicates)

Treatment	Ammonia applied (cc per barrel)	Total N		adsorbed ammonical N		NO ₃
		ppm	increment over control (%)	ppm	percent of total N	ppm
Control	—	210	—	3	1	4
Flooding	100	480	129	156	33	177
	400	830	295	526	63	40
	200+200	660	214	390	59	125
Flooding and tilling	200+200	690	229	366	53	210
Furrowing and packing	100	500	139	166	33	120
	400	770	267	330	43	40
Furrowing, packing and covering	100	540	157	250	46	117
	400	740	252	390	53	50

All the ammonia treatments produced great increases in the amount of nitrates in the soil. This leads to the conclusion, which was also reached by Birecka *et al.* (3), that the ammonia treatments do not destroy the nitrifying bacteria. On the other hand, it should be noted that treatments using the larger amount of ammonia resulted in smaller nitrate content increases than the corresponding treatments using the smaller amount, which indicates that an excess of ammonia may depress the activity of the nitrifying bacteria to some extent. This is also indicated by the large nitrate content produced by applying the larger ammonia amount (by flooding) in two doses, with tilling in between; the beneficial result of tillage may be attributable to its effect in improving the oxygen supply needed for the nitrification process.

3. THE RESIDUAL EFFECT OF AQUA AMMONIA ON THE SUBSEQUENT GROWTH OF MAIZE

The above results, concerning the accumulation of nitrogen in the soil two months after the ammonia treatments, explain the residual effects of these treatments on the subsequent growth of maize, which are presented in Table 6.

TABLE 6

THE RESIDUAL EFFECT OF AQUA AMMONIA ON THE SUBSEQUENT GROWTH OF MAIZE IN RED CLAY-LOAM
(means of 3 replicates)

Treatment	Ammonia applied (cc per barrel)	Total N in soil before planting (ppm)	Mean plant height (cm)	Mean number of leaves per plant	Mean plant weight (g)
Control	—	210	68	8.6	40
Furrowing and packing	100	500	120	11.0	133
	400	770	116	11.0	102
Flooding	100	480	102	11.6	182
	400	830	110	10.3	112

The table shows that the application of ammonia approximately 10 weeks before planting, by furrowing or by flooding, raised the yield of maize more than twofold. The beneficial results were evident in the external appearance of the plants, as well as in their height, number of leaves and total weight. No significant differences were found between the furrowing and flooding treatments. Plant weights were significantly greater, however, where the smaller amount of ammonia had been applied. It appears that an excess of adsorbed ammonia in the exchange complex may disrupt the nutritive balance of the plants and depress the yield somewhat; another reason for this effect could be that the uptake of nitrogen by maize is related more to the nitrate content rather than to the total nitrogen content.

SUMMARY

1. An experiment was carried out, in which sclerotia of *Sclerotium rolfsii* were placed in barrels filled with soil, and their viability following the application of aqua ammonia to the soil was determined. Three soil types, two amounts of ammonia, and two methods of application (furrowing and flooding) were compared.
2. Ammonia applied (by either method) at rates equivalent to about 185 and 740 kg N per acre was found to be toxic to *S. rolfsii*, in all soil depths down to 15 cm.
3. The fungicidal effect was found to increase with the amount of ammonia applied. When the larger amount was used some of the replications showed almost total sclerotia control.
4. The application of ammonia by flooding had an advantage over application in furrows, only when the smaller amount was used. Covering the soil surface following the furrowing treatment did not result in better preservation of the ammonia in the soil. Surface packing the soil slightly increased the toxic effect only when the larger ammonia amount was used.
5. The best results were obtained by applying the larger ammonia amount in furrows in two (equal) separate doses, with an eight-week interval in between. Tilling the soil during that interval further enhanced the fungicidal effect of the ammonia, leaving the soil practically free of viable sclerotia.
6. The application of aqua ammonia in both amounts used was found to cause an immediate rise in soil pH to highly alkaline values, followed by a rapid drop in pH which continued until the initial pre-treatment value was restored after about a month.
7. All the ammonia treatments tested raised the nitrogen content of the soil considerably, mainly in the adsorbed ammoniacal form. Large amounts of

nitrites were found in the soil two months after the treatments, indicating that ammonia in the amounts used do not prevent the activity of nitrifying bacteria.

8. A large residual effect of the ammonia treatments on the subsequent growth of maize was found, pointing to the efficacy of aqua ammonia as a fertilizer, in addition to its fungicidal action.

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