BULLETIN 750

Agricultural and Horticultural

Utilization of Fermentation Residues

Henry C. De Roo

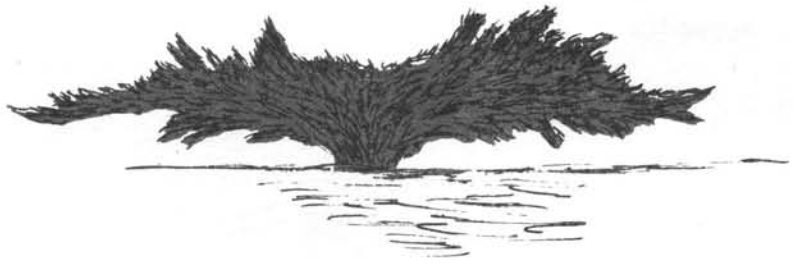


Table of Contents

Published March 1975

AGRICULTURAL AND HORTICULTURAL UTILIZATION OF FERMENTATION RESIDUES

Henry C. De Roo

One of the many challenges facing agriculturists is to develop uses for the enormous quantities of municipal and industrial wastes generated by our affluent society. Many of these wastes could be returned to the soil where plant nutrients would be recycled and soil physical properties improved, instead of carting them to dumps, landfills and incinerators.

Recent increases in the cost of nitrogenous fertilizers may provide economic incentives for use of such materials with many agricultural crops. In addition, the expanding horticultural industry utilizes many artificial potting mixtures and requires a continual supply of materials to stay in business.

Previous studies at this Station have examined such diverse soil amendments as peat and swamp muck (27), wood ashes (20), wood chips (21), sewage sludge (22) and green manures (14). Nitrogenous organic fertilizers have been studied extensively (11), as have potting mixtures for container grown plants (12,13).

Wastes produced by the pharmaceutical industry in Connecticut amount to about 52,000 tons per annum. These wastes contain about 350 tons of nitrogen, an amount adequate to fertilize a substantial acreage of crops. In addition, these wastes pose a substantial disposal problem, being equivalent to the sewage sludge generated by a city of about 300,000 people.

Hence, we undertook the study reported here to determine if these wastes could be used in agriculture and horticulture. First, we describe the chemical and physical properties of the wastes including some of the difficulties of handling, stockpiling and composting the large quantities involved. Next we report the results of experiments with container-grown chrysanthemums and junipers, and the results of greenhouse studies with corn, tomatoes and oats. Finally, we report the results of 2 years of field experiments with tobacco and corn.

CHEMICAL AND PHYSICAL PROPERTIES

The fermentation residues result from the commercial production of various organic acids and antibiotics by microorganisms. The precise details of the fermentation processes are trade secrets. In general, the materials are produced by fermentation in large vats, using molasses or other carbon sources for growth of the organisms. The pH of the reaction mixture is frequently controlled by the addition of $CaCO_3$. In some cases, Zn is added to control growth rates. When fermentation is complete, H_2SO_4 is added to

lower the pH to approximately 2-3. This also causes the precipitation of $CaSO_4$. At this point the desired fermentation product is removed. The residue consists largely of spent fungal mycelial tissue, $CaSO_4$, and inert filter aids such as perlite. Before leaving the factory, wastes from the various vats are combined and mixed with approximately 1% (w/w) unslaked lime which helps to raise the pH to about 7.5-8.0. The pH of unlimed mycelial wastes will increase slowly with time but will not rise to the same levels as the limed wastes. Under anaerobic conditions little change in pH is evident.

Since the production of these wastes varies from day to day, we analyzed each of the wastes from the six major fermentation processes before they were mixed and limed, and then calculated the composition of an average waste based on approximate annual production figures supplied by Chas Pfizer Co. The results of these analyses are shown in Table 1. To determine whether

TABLE 1. Chemical composition of fermentation residues.

Name	Citric 31A	Citric 31B	Itaconic	Ketoglu-taric	Terra-mycin	Tetra-cycline	Calc. Av.	Obs. Av.
Annual production, %	24	18	20	12	21	5	100	
				Chemical Analyses				
pH	2.99	2.55	2.55	2.12	2.30	2.45	2.55	8.0
Moisture, %	80.1	67.9	60.6	52.9	54.7	62.0	64.50	64.4
Loss on ignition, %	37.4	35.1	11.7	25.6	82.5	50.1	40.54	46.0
Sulfur, %	0.003	0.26	13.74	11.47	1.96	0.33	4.60	7.1
Fiber, %	6.59	3.63	0.98	0.17	6.34	0.55	3.81	N.A.
Fat, %	0.36	0.43	1.20	4.41	7.04	5.58	2.69	N.A.
Calcium, %	0.26	2.35	3.90	4.50	1.22	1.60	2.58	8.2
Nitrogen, %	4.36	1.21	0.57	0.37	4.39	3.02	2.50	1.90
Zinc, %	0.040	2.52	0.027	0.0075	0.0075	0.014	0.472	0.18
Phosphorus, %	0.028	0.028	0.028	0.082	0.090	0.110	0.052	0.11
Sodium, %	0.020	0.045	0.044	0.044	0.030	0.026	0.034	0.34
Potassium, %	0.024	0.035	0.025	0.028	0.015	0.012	0.024	0.06
Iron, %	0.004	0.026	0.024	0.018	0.042	0.037	0.023	0.21
Magnesium, %	0.003	0.011	0.004	0.004	0.012	0.004	0.007	N.A.
Manganese, ppm	N.D.	10	8	8	30	60	14	N.A.
Copper, ppm	5.5	9.5	8.5	4.8	6.0	7.5	7	N.A.
Nickel, ppm	2	2	1	1	1	1	1	N.A.
Lead	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	-	N.A.
Cadmium	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	-	N.A.
Chromium	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	-	N.A.

N.D. = none detected. N.A. = not analyzed.

the variability encountered from day to day would seriously affect the outcome of biological experiments with the material, we obtained a considerable number of analyses of samples collected at random after the wastes had been mixed and limed. The results in Table 1 show that the observed composition does not differ greatly from the calculated composition. The range of values observed varied somewhat as might be expected, but the variability did not appear sufficient to interfere with experimentation.

Samples were also analyzed for available plant nutrients by Morgan's methods (24). In general, a good supply of plant nutrients was indicated. The salinity of the wastes, an important property of any soil amendment, was relatively high, with an average electrical conductance (EC_e) of 5-7 mmhos/cm at 25°C as determined by the saturated paste method (4,35).

Some physical properties of the mycelial residues are shown in Table 2. The water contents were determined after saturation on porous tension plates at suctions of 0, 15, 50, and 100 cm of water.

TABLE 2. Physical properties of fermentation residues.

Bulk density (dry wt) g/cc	0.45
(dry wt) lbs/cu yd	682
Particle density (dry wt) g/cc	1.78
Particle sizes (dry wt) %	
coarse 1.0-0.5 mm	2.4
medium 0.5-0.25	3.3
fine 0.25-0.10	16.6
very fine 0.10-0.05	11.4
silty 0.05-0.002	31.3
water soluble salts, following digestion with H_2O_2 (mainly $CaSO_4$)	35.0
Porosity vol %	78
Water relations	
Water content, vol %	
0 cm tension	79.7
15 cm tension	70.0
50 cm tension	67.5
100 cm tension	65.6
Percolation rate cc/min	0.15
in./hr	0.08
Swelling at saturation, vol %	3
Shrinkage after oven drying, vol %	24

The volume of water at 0 cm tension or saturation is an approximation of the total pore space of the mycelial residues and ideally should equal the porosity. The agreement between both values (Table 1) of 79.1 and 78 vol % respectively, is good. The porosity was derived from the solid particle density (S.P.D.) of the wastes and their bulk densities (B.D.) or volume weight, using the formula

$$\left(1 - \frac{\text{B.D.}}{\text{S.P.D.}}\right) \times 100.$$

Measurements of water retention at tensions of 15, 50, and 100 cm were based on their value for later studies, in which mycelial residues were used as an ingredient for growing mixtures in pots. The water contents at these tensions show that mycelial wastes have a great capacity and that they hold their water with relatively great tenacity: their moisture retention characteristics are comparable to those of a clay loam (8).

The high water-holding capacity of the mycelial residues prompted further investigation, since under some circumstances such a wet material might be undesirable. Microscopic examination of the residues after a few days of field spreading revealed few if any intact cell membranes in the fungal tissue; hence the water is apparently adsorbed by the dead cellular material. Freezing or air drying thus altered these properties very little. Samples of screened and air-dried residues (7.4% water by volume) were placed on porous tension plates at a tension of 2.5 cm and allowed to rewet. Within 3 days the moisture content reached 45% and after 8 days the samples were practically as wet as the fresh material.

The percolation rates indicate a poor water permeability, caused by the relatively fine texture and nature of the wastes and their tendency to swell when wet. The considerable shrinkage on drying is indicative of the high content of organic matter.

To summarize, fresh mycelial wastes have the consistency of a heavy sludge, containing about 2/3 water on a wet weight basis. About half of the dry weight of the silty-textured solids is organic or volatile matter. The high moisture content and relatively small amounts of plant nutrients N, P, and K are typical of any biological tissue. The differences are that Zn has been added to the wastes and that they also contain substantial amounts of $CaSO_4$ and filter aids. Hence, we might anticipate certain problems due to salinity, Zn, high water-retention capacity, and poor permeability.

COMPOSTING AND STOCKPILING

Composting of organic, municipal, and industrial wastes in windrows has been used to decompose, stabilize, and reduce the weight and volume of such wastes. For effective composting the material should have a coarse, open structure to allow adequate air circulation, a carbon (C) to nitrogen (N) ratio near 30:1, and a moisture content of about 50% (28). Poincelot and Day (29) found that leaves in windrows compost readily when mixed with mycelial wastes in an approximate dry weight ratio of 6:1.

We conducted some laboratory studies of composting of mycelial waste using equipment described by Vesilind (36). The mycelial residues are rich in chitin, a polysaccharide which forms a major component of fungal cell walls. Their nitrogen content averages 1.9% (Table 1). Carbon was estimated by dividing the percent loss on ignition by the factor 1.8 (28); thus the mycelial waste has an average C/N ratio of about 13:1. Various coarse materials such as forest litter, sawdust, and charcoal bits were added to the wastes to provide a more open structure, and water and N, if necessary, were added to create optimal conditions for composting. The results were disappointing; the microbiological generation of heat did not increase temperatures in the middle of the composting mixtures above 105°F (40°C). Thus, the thermophilic stage was not reached, and as a result little organic matter was decomposed.

Concurrent with our investigation, composting and stockpiling of these mycelial residues was conducted during the winter of 1972-73 at a site in eastern Connecticut.

The available site was a small, uneven clearing in the woods. After arrival, the wastes were piled for a few days and subsequently spread and stirred with a disc-harrow. Thereafter, the partially dewatered material was placed in stockpiles and turned frequently -- even daily -- for further drying. Unfortunately, the stockpiles did not dewater readily and aerobic conditions could not be maintained. Hence, they produced odors characteristic of anaerobic decay processes. Observed temperatures within the piles never exceeded 95°F (35°C), thus the thermophilic decomposition characteristic of composting (28) did not occur. Some stabilization of the stockpiles was obtained by mixing with bark, wood chips, or sand, but the sheer volume of wastes arriving daily made such operations difficult. Subsequent experience indicated that spreading the wastes thinly on land and allowing them to dry was perhaps the best alternative. This led to studies of direct agricultural uses of the wastes.

In any area where plant nutrients accumulate, be it a lawn, a corn field or a compost yard, some nutrients may escape to nearby waterways (15). The Connecticut Department of Environmental Protection requested our assistance in evaluating possible nutrient losses from this site to aid them in the preparation of an environmental impact statement. We report the results of these investigations below.

We installed four test wells at the compost yard in February 1973 to determine possible losses of nutrients. The wells were placed in carefully selected locations, while two other sampling stations were located on small brooks draining either end of the compost yard. Further details are given in Appendix I.

Water samples were collected at weekly or biweekly intervals for a year and analyzed by Morgan's methods (24). Since phosphorus is bound so tightly to soils that little is lost by leaching, we focused our attention on nitrate and ammonium nitrogen. Since the wastes contain large amounts of $CaSO_4$, we also analyzed for sulfate.

We found mycelial residues can be composted when mixed with sufficient material to maintain aerobic decay. Stockpiled residues do not dewater readily and frequently become anaerobic with attendant characteristic odors. Losses of plant nutrients from stockpiled residues appear to be small, but some sulfates may be lost. Contrary to expectations, losses of nitrogen were generally very low.

CONTAINER STUDIES WITH CHRYSANTHEMUMS AND JUNIPERS

Mycelial residues were tested as a component of artificial root media for growing chrysanthemums and junipers. These tests were part of our continuing studies to develop effective and economical soilless growing mixtures for container-grown plants.

Methods

In this report the results with mixes including mycelium are compared with those obtained with mixes composed of standard ingredients such as sphagnum moss peat, sand, perlite, vermiculite, and bark. Besides mycelial wastes, some of the mixes also contained charcoal bits, a waste product of a local distillery (12), digested sewage sludge, originally obtained from the treatment plant in Hartford, and now piled at a local nursery (13), or waste cellulosic fibers from the production of paper towels and tissues.[1/] The porous, pea-sized charcoal bits provide a coarse ingredient, which should help create an open-structured, well aerated medium. The bits, made up of hardwood (98% cherry, maple, beech; 2% ash, birch) were denatured at the distillery by drenching about 100 pounds of charcoal with 1 gallon of 4% ammonium solution. The sewage sludge was hauled and shredded in the early spring of 1972 and kept in a pile outdoors. The properties and uses of various digested sewage sludges were investigated and reported by Lunt (22,23). The cellulosic fiber waste is fine textured, and contains 46% carbon and 0.53% nitrogen with a C/N ratio of 90:1. The basic ingredient in all mixes was a coarse, washed concrete sand, which provided sufficient weight to keep the container and plant upright. The mixes tested, their components, preplanting amendments and fertilizations are all summarized in Table 3.

The liming and fertilization treatments varied with the chemical properties of the individual ingredients. These amendments, based on soil tests, were intended to create optimum pH and fertility for either chrysanthemums or junipers. Table 3 shows that the main differences were the amounts of limestone added; mixes containing alkaline mycelial residues (pH 8) did not receive limestone, while those containing bark received less limestone than the rest. Mixes with sewage sludge received a smaller amount of 20% superphosphate than did the control and other mixes, since sludge contains more available phosphorus. Another difference was the addition of ureaformaldehyde to mixes 1, 2, 6 and 7. This served as a comparison for the organic nitrogen provided by mycelium and by sewage sludge. Finally, some nitrogen as ammonium nitrate was added to all mixes except 3 and 5, since soil tests indicated these mixes contained adequate nitrate and ammonium nitrogen from mycelium.

1/ Supplied by Kimberly-Clark Corp., New Milford, Connecticut.

TABLE 3. The composition and fertilization of growing mixtures used in 1-gallon containers for the production of chrysanthemums and junipers.

	Treatment No.									
	Chrysanthemums					Junipers				
	1	2	3	4	5	6	7	8	9	10
Materials, % by volume										
Sand	45	25	25	33	33	40	33	25	25	25
Peat moss	45	25					33			
Perlite	10	25								
Vermiculite		25								
Bark - hardwood						60	33	25	25	25
Sewage sludge				33				25		
Charcoal bits			25						25	
Wood fiber waste			25	33	33			25		25
Mycelial residues			25		33				25	25
Fertilizers, lbs/cu yd										
Limestone, dol.	12	6.5		6		2.5	4	4		
Superphosphate, 20%	2.5	2.5	2.5	1.75	2.5	2.5	2.5	2.0	2.5	2.5
Potassium sulfate, 50% K_2O	.5	.5	.5	.5	.5	.5	.5	.5	.5	.5
Urea-form, 38% N	1.5	1.5				1.5	1.5			
Ammonia nitrate, 33% N	.5	.5		.5		.75	.75	.75	.75	.75
ozs/cu yd										
F.T.E. 519*	2	2	2	2	2	2	2	2	2	2
Iron chelate	1	1	1	1	1	1	1	1	1	1

* Fritted potash (29% K_2O) and trace elements.

More nitrogen was added to the juniper mixes than to the chrysanthemum mixes since the latter mixes amended with hardwood bark are known to require more nitrogen to avoid nitrogen deficiency.

Rooted cuttings of Chrysanthemum moriflorum cv. Best Regards and of Juniperus horizontalis plumosa compacta cv. Andorra Juniper were planted on June 30 and June 15, 1972, respectively. Eight chrysanthemums and nine juniper plants, one in each 1-gallon container, were used per treatment and each treatment was replicated four times in a randomized block. The potted chrysanthemums were managed according to standard practices. Pinching of the terminal buds to induce branching was started about 2 weeks after planting. The junipers, on the other hand, were not pruned as is customary since we wished to measure total shoot growth in response to the differential treatments.

After planting, liquid fertilizations were based on soil tests made at regular intervals of 10 to 15 days unless the appearance of the plants warranted more frequent testing. A complete liquid fertilizer (23-8-22) was prepared from urea, monoammonium phosphate and potassium nitrate mixed at a ratio of 1-2-3 by weight and dissolved in water. Liquid fertilizers supplying either N or K were applied as ammonium nitrate or potassium sulfate. All chrysanthemum mixes were fertilized with the complete fertilizer at a rate of 200 lbs/acre on day 12, followed on day 21 by an additional application of 66 lbs N/acre to mixes 3, 4, and 5. From then on, the fast growing and well watered plants in all mixes received complete fertilizer at the rate of 250 lbs/acre every 10 to 15 days. The slow growing junipers received the complete fertilizer on day 29 at the rate of 300 lbs/acre. On day 41 mix 8 was supplemented with 134 lbs N/acre. On day 56 mixes 7, 8, 9, and 10 received K at the rate of 125 lbs/acre, while mix 6 received the complete fertilizer at the rate of 250 lbs/acre. These applications were followed by a complete liquid fertilization on day 76, and a final fertilization before winter storage in a plastic house on day 148. At this time, dry fertilizer (13-7-12) was supplied to each mix at the rate of 700 lbs/acre. Its N was all urea-form which becomes slowly available over a long period of time.

The effect of the various media and treatments on the plants was evaluated throughout the growing season by commercial criteria, i.e. quality and quantity of foliage, and number and size of flowers. As soon as the plants were salable a final evaluation was made by two commercial growers.

At the end of the growing season the physical properties of the various media in containers prepared at the beginning of the growing season were examined. These pots were not planted; instead a brass cylinder of 250 cc capacity (2-7/8 in. ID x 2-5/16 in. long) was buried in the center of each pot and filled with the mix in the same manner as in the planted containers. Since the containers were not planted, the effect of root growth on the structural condition of the mixes was not measured. However, the high root concentrations in small pots makes it difficult to take undisturbed core samples. Thus, we chose to compare each mix unplanted, but after it had undergone all crop management practices throughout the growing season and had stabilized to its natural compression.

The water contents of the mixes were determined at suctions of 0 cm (saturation), 15 cm (average height of medium in 1-gallon containers), 50 cm and 100 cm of water. Water retentions were expressed on a volume basis because of the variable bulk densities of the media. Furthermore, since some of the mixes shrunk on drying, the water contents were expressed on a wet volume basis using the volume of the mixes at container capacity or 15 cm suction (37). The water holding values at 15, 50, and 100 cm were determined on porous tension plates; tensions higher than 100 cm were not determined because such moisture stresses seriously retard growth of potted plants (10). Total pore space (T.P.S.) was calculated after determining the solid particle density (S.P.D.) of the mixes and their bulk densities (B.D.), using the formula

$$\% \text{ T.P.S.} = \left(1 - \frac{\text{B.D.}}{\text{S.P.D.}}\right) \times 100.$$

The total pore space is a measure of the volume of water at saturation (or 0 cm tension). The % air space or aeration at container capacity (15 cm tension) is the difference between total pore space or vol % water at saturation and the vol % water at 15 cm. This is considered equivalent to the volume of air in the upper portion of a 1-gallon container after irrigation and subsequent drainage. The amount of water held between 15 cm tension or container capacity and 50 cm tension is considered readily available water (R.A.W.), while that held at tensions between 50 and 100 cm is less available and forms a water buffer capacity (W.B.C.) against extreme evaporative demands of the atmosphere on the plants (10).

The vol % of shrinkage after oven drying is used to characterize the relative shrinkage of the various media under practical irrigation management, since excessive shrinkage of the media on drying is undesirable.

Results with Chrysanthemum moriflorum cv. Best Regards

The development of the chrysanthemums in the different growing mixtures is summarized in Table 4. As early as day 12, the plants in mix 5 containing 33% (v/v) mycelium were stunted, narrow-leaved, yellow and seriously wilted; all indicators of a high concentration of soluble salts in this medium. The plants growing in mix 3 containing 25% mycelium were medium sized and light green to yellowish, while the plants in mix 4 containing no mycelium were medium to good sized and still light green. The plants in standard mixes 1 and 2 were good sized, broad-leaved and green. During the rating on day 19 (Table 4) a change was noted: the plants in mix 4 were now yellow and in worse condition than on day 12; the color of the plants in mixes 3 and 5, on the other hand, had improved. Evaluations continuing throughout the season showed that the plants in mix 3 or particularly those in mix 5, kept on improving and eventually were commercially rated good to excellent. The plants in standard mixes 1 and 2 had more blooms than any of

TABLE 4. Effect of various growing mixtures on growth and flowering of chrysanthemum plants.

		Treatment No.					L.S.D.
		1	2	3	4	5	(0.05)
Day 19 plant rating*		VG	E	VP	P	P-VP	
Day 46 plant height	cm	20.9	23.6	9.8	7.1	10.4	3.5
width	cm	22.8	25.1	13.2	9.3	13.0	3.1
Day 86 plant height	cm	37.2	37.6	27.1	20.8	27.8	2.2
width	cm	27.8	30.9	21.8	20.2	22.2	5.6
Blooms	No.	9.2	11	7.3	3.8	7.5	1.4
Day 90-Commercial rating*		VG-E	E	G	P-VP	G-E	

* E = Excellent; G = Good; P = Poor; V = Very
L.S.D.(0.05) = least significant difference at the 5% level.

the other plants, but the plants in mix 3 and mix 5 were rated highly because of their relatively compact, bushy shape, which when covered with blooms, contributes markedly to the market quality.

Since soil salinity is known to reduce stem length of chrysanthemums (31), it appears that the development of the plants in mixes 3 and 5 was largely controlled by salinity. Rutland (31) found that the market quality of chrysanthemums grown with variable water supply from manual irrigation was less adversely affected at EC_e 1.8 mmhos/cm than at EC_e 3.6 mmhos/cm. On day 90 the salinities in mixes 1, 2, 3, 4, and 5 were EC_e 0.50, 0.50, 2.30, 0.60, and 2.30 mmhos/cm, respectively. Thus, the highly saline mixes 3 and 5 probably suppressed early growth by inducing water stress and decreasing N utilization (3,18). After leaching had lowered the salinity, these effects were less severe and the appearance of the chrysanthemums, tolerant of moderate salinity, improved greatly.

We attempted by adding nitrogen to correct the stunting and yellowing of the plants in mix 4 which contained no mycelium. This was done for two reasons. First, we may have underestimated the utilization of nitrogen by the wood fibers. Second, an analysis of the sewage sludge which had been exposed to leaching since early spring showed that it had lost most of its readily or potentially available N. The addition of nitrogen to mix 4, however, did not improve growth so we suspected phytotoxic substances in the wood fiber waste. However, water extracts of the wastes did not harm tomato plants in a simple bioassay. This leads us to conclude that in a mix containing 33% (v/v) sewage sludge, the high concentration of heavy metals may have caused iron deficiency (chlorosis) and decreased growth (22). This agrees with previous observations with chrysanthemums (13), in which 20% (v/v) sewage sludge in a sand-peat moss mix slightly retarded growth and produced incipient iron deficiency.

The physical properties of mixes 4 and 5 (Table 5) are quite similar. Mycelium added to a sand-wood fiber mixture formed a more open-structured medium than sewage sludge. Sewage sludge is known to improve the physical properties of soil (22,23) and artificial media (13). Both mixes had a desirable air capacity of about 20% (v/v), although their total pore space (T.P.S.) was relatively low for a container mix. As a result, the readily available water (R.A.W.) in both mixes was much smaller than that in a standard mix. This mix, however, lacks the favorable air capacity found in mixes 4 and 5. In all 3 mixes (1, 4 and 5) the water buffer capacity (W.B.C.) was low, indicating that special care is needed to provide a regular water supply.

Results with *Juniperus horizontalis plumosa compacta* cv. Andorra Juniper

The development of this relatively slow growing wood ornamental was without marked defect. The differences in growth among plants due to treatment were not large (Table 6). Unlike the chrysanthemums, the early growth of the junipers in the mixes containing mycelium, with or without wood fiber, did not show any discoloration or retardation. Bernstein (5) reports that spreading juniper (*Juniperus chinensis*) tolerates salinities up to 8 mmhos/cm.

TABLE 5. Physical characterisitcs of various growing mixtures.

Treatment No.	B.D. g/cc	T.P.S.	Air	R.A.W. vol %	W.B.C.	Shrink.
1 - Sand-peat-vermiculite, 45-45-10	.87	66.9	6.0	17.2	1.3	2.2
4 - Sand-wood fiber-sewage sludge	.99	61.3	20.5	7.5	1.1	3.3
5 - Sand-wood fiber-mycelium	.88	63.6	22.2	7.5	1.1	9.2
6 - Sand-bark, 40-60	.84	64.6	24.2	13.3	2.1	5.4
7 - Sand-bark-peat	.72	70.0	16.7	21.2	3.8	5.4
8 - Sand-bark-wood fiber-sewage sludge	.76	67.6	29.7	9.1	2.0	4.2
9 - Sand-bark-charcoal-mycelium	.62	72.6	33.1	4.8	2.1	2.5
10 - Sand-bark-wood fiber-mycelium	.67	71.2	34.7	6.7	1.7	3.3

B.D. = Bulk density or volume weight (x 62.42 = dry wt in lbs/cu ft).
T.P.S. = Total pore space or water content at saturation (SAT.).
Air = Air space at 15 cm water suction or container capacity.
R.A.W. = Readily available water, released between 15-50 cm suction.
W.B.C. = Water buffer capacity, water released between 50-100 cm suction.
Shrink. = Shrinkage after oven drying.

TABLE 6. Effect of various growing mixtures on growth of juniper plants.

	Treatment No. 6	7	8	9	10	L.S.D. (0.05)
Day 60 plant height cm	16.3	17.4	14.7	14.7	16.6	.94
width cm	10.3	12.3	8.3	10.1	12.2	1.3
Day 130 plant height cm	20.5	20.6	17.8	18.2	20.8	1.1
width cm	15.1	15.9	13.1	16.4	17.4	1.7
Day 360 plant height cm	18.2	18.2	16.8	14.8	18.2	1.2
width cm	20.0	19.8	18.2	23.2	25.8	2.3
color rating*	1.5	1.25	2	3	2.75	

* 1 = yellowish; 2 = light green; 3 = green

The highest salinity observed in our experiment was 4.3 mmhos/cm, measured in mix 9 on day 5. The salinity (EC_e) of mixes 6, 7, 8, 9, and 10, measured throughout the season, decreased as shown in Table 7. Thus, it is not surprising that we did not observe salt injury on junipers.

TABLE 7. Electrical conductivities (EC_e in mmhos/cm) in various growing mixtures under container-grown junipers.

Days after transplanting	Treatment No. 6	7	8	9	10
5	2.6	2.7	2.3	4.3	3.0
52	.8	.8	.6	2.4	2.7
140	.3	.2	.2	1.8	1.7

The poorest early growth occurred in mix 8 containing sewage sludge. The factors responsible were probably the same as in the experiments with chrysanthemums, namely the low nitrogen content of the sewage sludge and its high content of heavy metals.

About a year after planting, junipers in mixes 9 and 10 containing mycelium had the largest spread and the best color. During the long period of winter storage in the plastic house, the mycelial residues in these mixes apparently released enough N to keep the foliage of the plants green, while the standard mix 6 and the improved mix 7 did not, in spite of an application of mixed fertilizers with organic N in late fall.

The physical properties of the juniper mixes 6 through 10 are presented in Table 5. Again, the mixes containing mycelial residues showed a much higher air capacity and much lower R.A.W. than mixes 6 and 7 containing only standard ingredients. Comparing mix 8 and 10, sewage sludge was less effective in increasing the air capacity and lowering R.A.W. than mycelium; both materials had similar effects in the chrysanthemum studies.

In summary, growing mixtures made up with 25 vol % or 33 vol % mycelium and wood fiber waste eventually produced chrysanthemums of good to excellent market quality. The early growth, however, showed stunting and yellowing.

Mixes with 25 vol % mycelium with or without wood fiber waste did not produce harmful effects at any stage of development of junipers and eventually produced just as good or slightly better growth than the standard mixes. The slow release of nitrogen by the mycelial residues apparently produced junipers that remained greener throughout winter storage than did the control plants.

On the basis of these results, we conclude that fresh mycelial residues supply nitrogen that is only slowly available. The rate of application of mycelial residues is apparently limited more by their high soluble-salt content than by the amount of nitrogen or organic matter. Furthermore, the kind of plant to be grown in the container mix is an important consideration, since salt-tolerant species such as junipers will suffer the least from fairly heavy applications.

Observations of physical properties of the various mixes show that mycelial residues, while enlarging the total pore space somewhat, greatly increase the air capacity of growing mixtures. However, this increase seems to be at the expense of the readily available water (R.A.W.) in the medium. The mycelial residues, consisting largely of organic matter and gypsum, are apparently conducive to aggregation and formation of an open-structured medium with large pores. This could play an important role in the production of potted plants or containerized nursery stock because of the usually heavy watering regimes common in commercial practice.

GREENHOUSE EXPERIMENTS WITH TOMATOES AND CORN, FOLLOWED BY OATS

The preceding studies with container-grown plants indicated that mycelial residues could serve as a nitrogenous fertilizer for certain horticultural uses. However, both the high salinity and the slow rate of mineralization of nitrogen obviously needed further examination. We report here a series of greenhouse experiments with tomatoes and corn, followed by oats, which were designed to further evaluate the usefulness of mycelial residues as a fertilizer for agricultural use.

Methods

Experiments with Tomatoes and Oats. Two experiments with tomatoes were conducted to study the release of ammonium and nitrate nitrogen from mycelial waste incorporated in soil at various rates. In the first, the rate of release was compared with that from standard organic and inorganic nitrogenous fertilizers. In the second, an attempt was made to maintain a constant level of nitrate N in each of the different treatments based on frequent soil testing and fertilization. In this second experiment the mycelial waste treatments were compared with one non-mycelial treatment, fertilized and top-dressed with nitrate N in the same manner as the mycelial treatments.

The soil was a coarse-textured, well-aerated, but infertile Windsor loamy sand. This soil represents the type of marginal land which one would expect to benefit most from an addition of nitrogenous organic materials such as mycelial waste.

As shown in Table 8, the release of N by the mycelial waste in the loamy sand (treatments 1, 2, and 3) was compared with that supplied by a synthetic organic N source (ureaform, treatment 4), a natural nitrogenous organic source (cottonseed meal, treatment 5), and an inorganic, standard fertilizer 10-10-10 (treatment 6). Treatment 6 formed the control: after amendment with the required amounts of dolomitic limestone, 20% superphosphate and potassium sulfate, its nitrogen fertilization was based on the commercial practice of supplying about 80 lbs N/acre before transplanting, followed later in the season, if required, by a side-dressing with 40 to 50 lbs N/acre. All treatments received ammonium nitrate as a starter at the rate of 30 lbs N/acre.

The amendments with limestone, 20% phosphate, and potassium sulfate were based on soil tests and were intended to create optimum pH and fertility

TABLE 8. Amendments and fertilizations of Windsor loamy sand for growing tomatoes and corn in pots in the greenhouse.

	Treatment No.						
	1	2	3	4	5	6	7
Tomatoes							
Mycelial residues, tons/acre (wet wt)	12	36	108	0	0	0	
Fertilizations, lbs/acre							
Organic nitrogen							
Mycelial residues, 2% N (dry wt)	160	480	1440				
Urea-form, 38% N				160			
Cottonseed meal, 6.5% N					80		
Inorganic nitrogen							
10-10-10 grade						80	
Limestone, dolomitic, tons/acre	1.25	.5	0	2	2	2	2
Superphosphate, 20%	450	450	450	450	450	300	450
Potassium sulfate, 50% K_2O	450	450	450	675	675	120	675
Ammonium nitrate, 33% N	90	90	90	90	90	90	90
Corn							
Mycelial residues, tons/acre (wet wt)	12	36	108	0	0	0	
Fertilizations, lbs/acre							
Organic nitrogen							
Mycelial residues, 2% N (dry wt)	160	480	1440				
Urea-form, 38% N				320			
Cottonseed meal, 6.5% N					160		
Inorganic nitrogen							
10-10-10 grade						160	
Limestone, dolomitic, tons/acre	1.25	.5	0	2	2	2	
Superphosphate, 20%	450	450	450	450	450		
Potassium sulfate, 50% K_2O	450	450	450	675	675		
Ammonium nitrate, 33% N	90	90	90	90	90	90	

for the growth of tomatoes. The main differences in amendments were in the amounts of limestone (Table 8); the greater the amount of alkaline mycelial waste added to the soil, the smaller the limestone application.

The soils and amendments for each of the six treatments were thoroughly mixed in a cement mixer. Each treatment mixture supplied enough material to fill 16 1-gallon containers, which were subsequently placed in four randomized blocks on a bench in the greenhouse.

The following day, May 7, 1973 (day 0), three pots of each treatment in each replicate were planted with uniformly sized (about 9 inches tall) tomato transplants (Lycopersicum esculatum, var. Rutgers); one transplant in

each 1-gallon container. A 250 cc brass cylinder was placed in the fourth container to study the physical properties of the soils. During the growing season, the containers with the cylinders, although not planted, were treated in the same manner as those with the tomato plants.

Soil tests were performed weekly, both to study the release of ammonium and nitrate N from the mycelium treatments and also to determine when and how much liquid fertilizer to add to ensure a satisfactory nutrient supply. On day 28 all treatments received 54 lbs N/acre in the form of ammonium nitrate except treatment 3 (108 tons mycelial waste/acre). On day 35 all treatments received complete fertilization with 23-8-22 fertilizer at the rate of 250 lbs/acre. This fertilization was repeated at the rate of 300 lbs/acre on day 43, one day before harvesting the tomato plants, but 15 days before seeding the oats.

The effects of the various treatments were evaluated by noting the quality and quantity of foliage, by measurements of plant height and, on day 44, by harvesting and weighing the plants exclusive of roots. On day 37 two leaves adjacent to the second inflorescence were picked, washed in a dilute solution of a mild detergent, rinsed with distilled water, dried on filter paper and ground for an analysis for heavy metals (17).

Two weeks after harvesting the tomatoes, all pots were seeded to oats (Avena sativa Clinton; 15 grains were planted in each pot with the germ end down) to study the residual effect of the various N treatments. The oats were grown to maturity without any additional fertilization, after which the number of plants and the total plant weight exclusive of roots were determined.

After harvesting the oats the physical properties of the 3 mycelial soil mixtures, treatments 1, 2, and 3, were determined and compared with those of the check, treatment 6. We followed the procedures described in our studies of potting mixtures and added a measurement of the oxygen diffusion rates (O.D.R.) with the platinum microelectrode (19). The O.D.R. measurements were made at a depth of 3 in. in all pots after the oats were harvested.

The second greenhouse experiment with tomatoes was initiated about 4 weeks after the first experiment when we found that ammonium and nitrate N release from the mycelial waste had to be observed more frequently than once a week. The mycelial treatments 1, 2, and 3 were applied exactly as in the first experiment, but no comparison was made with other organic nitrogenous sources. Treatment 7, without organic nitrogen, served as the control, since the design of the experiment required maintenance of adequate available nitrate N concentrations by topdressing based on soil testing every 3 days. These additions were made as water soluble fertilizers with different nitrate N and ammonium N ratios from ammonium, calcium, or sodium nitrate. Nearly all of the additions were made at the rate of 64 or 66 lbs N/acre.

Experiments with Corn and Oats. This greenhouse experiment with corn and oats was conducted simultaneously with the tomato experiments to determine the effectiveness of mycelial residues for corn production.

The materials and methods were similar to those used with the tomatoes, except that additions of fertilizer were adjusted to meet the requirements of field corn (Table 8). The treatment mixtures, contained in 12 2-gallon plastic pots, were placed on a bench in the greenhouse and randomized in four replicates of three pots each.

The following day, May 4, 1973 (day 0) the pots were planted with corn (Zea mays L., var. 'Pa. 602a); each pot received three seeds with the germ end down to facilitate sprouting. After germination, on day 11, the seedlings were thinned leaving only the most vigorous one in each pot.

Soil tests of the six treatments were again conducted. No topdressing was applied, although on day 42, treatment 1, the lowest rate of mycelial application, showed only a trace of nitrate N.

The early response of the corn was evaluated by observing leaf growth, quality and plant height. On day 48 the plants were cut off at the soil surface, oven dried and weighed. Two weeks later, 30 oat grains were planted in the stubble in each pot.

Further, we made visual observations of the development of corn roots in soil amended with mycelial residues. Three planter boxes 10 x 12 x 18 in., with a slanting (30°) glass panel on one side were used. Half of the box was filled with Windsor loamy sand, fertilized according to treatment 6; the other half with one of the three Windsor loamy sand and mycelial residues mixtures, treatments 1-3 of Table 8. A single corn plant was grown at the interface of both media, which was perpendicular to the glass panel. By removing the cover on the glass panel, the development of the roots in both media behind the glass panel was clearly visible and could be readily studied and photographed.

Results with Tomatoes and Oats

The growth of the tomatoes in the first experiment is presented in Fig. 1. Development of the plants can be divided into two groups: i.e., those treated with mycelial waste (treatments 1-3) that were shorter than those treated with other N fertilizers (treatments 4-6). However, it was only during the first 15 days after transplanting that the growth rate of the group treated with mycelium was significantly less than that of those treated with other N fertilizers. After day 15, the growth rate of both groups of plants was about the same. On day 45, it was obvious that the shorter plants of treatment 3 were more branched and bushy than the plants of the other treatments. This observation of greater growth was confirmed when the plants were harvested: The dry weights of the plants of treatments 1-6 are presented in Table 9.

The foliage of the plants showed some effects of the differential treatments. On day 9 the lower leaves of the plants in mycelial treatments 1-3 were yellowed in a mottled pattern. Furthermore, the top leaflets of the plants treated with mycelium were narrower and smaller than those of the other plants. In the early growth stages the worst injuries were found on plants in treatment 1 and particularly in treatment 2, while treatment 3

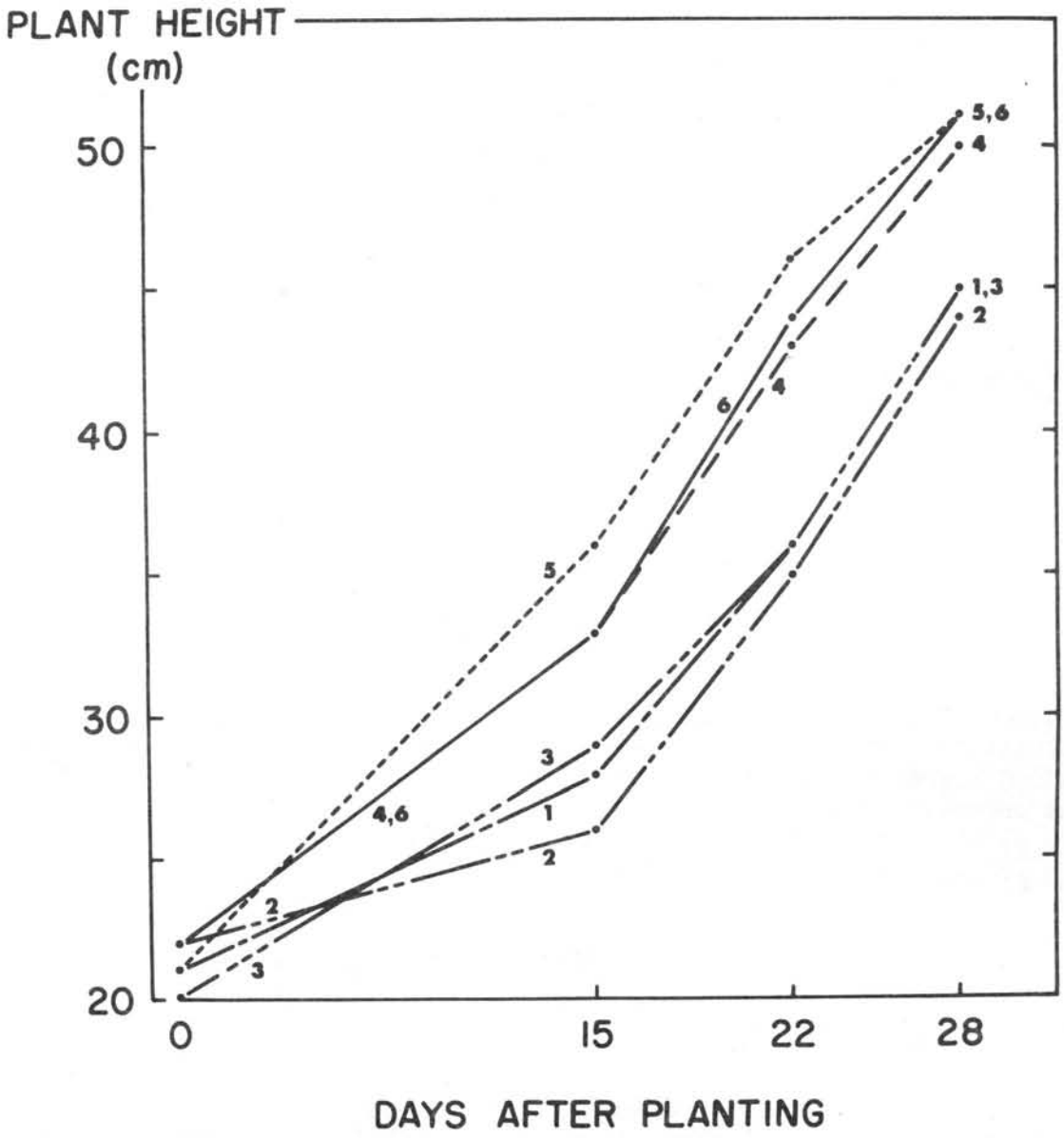


FIGURE 1. Height growth of potted tomato plants in Windsor loamy sand as affected by mycelial amendments (treatments 1-3) and by organic and inorganic N fertilizers (treatments 4-6). Treatments are described in detail in Table 8.

showed less damage. We have no explanation of this as yet. Later in the season these symptoms increased with increasing rates of mycelial application, and the yellow mottled-leaf pattern was also found higher on the plants. The yellowing of the leaves suggested that the response of the plants to N was affected by the high salinity, since N nutrition and soil salinity are reportedly related (3,18).

TABLE 9. Yield of tops in g/plant, dry wt.

Treatment No.	Tomato*	Oats**	Corn*	Oats***
1	10.2	.69	10.4	.33
2	11.6	.88	9.0	.70
3	14.0	1.00	6.6	.90
4	13.2	.69	13.7	.71
5	13.8	.63	10.1	.50
6	14.1	.54	11.2	.69
L.S.D. (0.05)	1.2	.12	3.0	-

* Average of 12 tomato plants, 44 days after transplanting or 12 corn plants, 48 days after seeding.

** Oats following tomatoes, average of about 40 plants, replicated four times.

*** Oats following corn, average of about 80 plants.

Our weekly testing of the various treatments allowed us to examine this relationship. The release of nitrate N by the various treatments is shown in Fig. 2. During the first week the mycelial treatments 1-3 actually immobilized nitrate N and subsequently, during the second week, kept nitrate concentrations very low. The N fertilizer treatments 4-6, on the other hand, maintained higher nitrate N concentrations in the loamy sand with only a gradual decrease in supply. After day 14, treatment 3 with mycelial waste applied at the highest rate of 108 tons/acre showed a sharp increase in nitrate N supply, which peaked on day 28. On that day the remaining treatments (1, 2, 4, 5, and 6) received their first supplemental fertilization with ammonium nitrate at the rate of 54 lbs N/acre. As shown by soil tests on day 35, the fertilization on day 28 had little effect. Two more additions of fertilizer at the rate of about 60 lbs N/acre on day 35 and on day 43 were required to increase the nitrate N in all treatments to the satisfactory concentrations tested on day 50.

The generally moderate concentrations of ammonium N showed less variation. During the first 14 days, treatments 5 and 6 showed slightly higher ammonium N concentrations than the mycelial treatments, while after day 14 treatments 2 and 3 were highest in ammonium. Additions of ammonium nitrate on days 28, 35, and 43 did not increase the concentrations of ammonium N, indicating that nitrifying bacteria were plentiful in all substrates.

The conductivities of saturated extracts of the various substrates were as follows: Salt concentrations in mycelial treatments 1-3 were generally 2 to 3.5 times as high as in the N fertilizer treatments 4-6, and all decreased slightly with time. Within these two groups of treatments the differences in soil salinity were negligible. The nutrient-salt concentrations of the N fertilizer treatments dropped more sharply than those of the mycelial treatments, approaching levels at the end of the experiment considered less than optimal for tomatoes. The salinities of the mycelial treatments changed little with time, remaining slightly above 2 mmhos/cm. This is understandable,

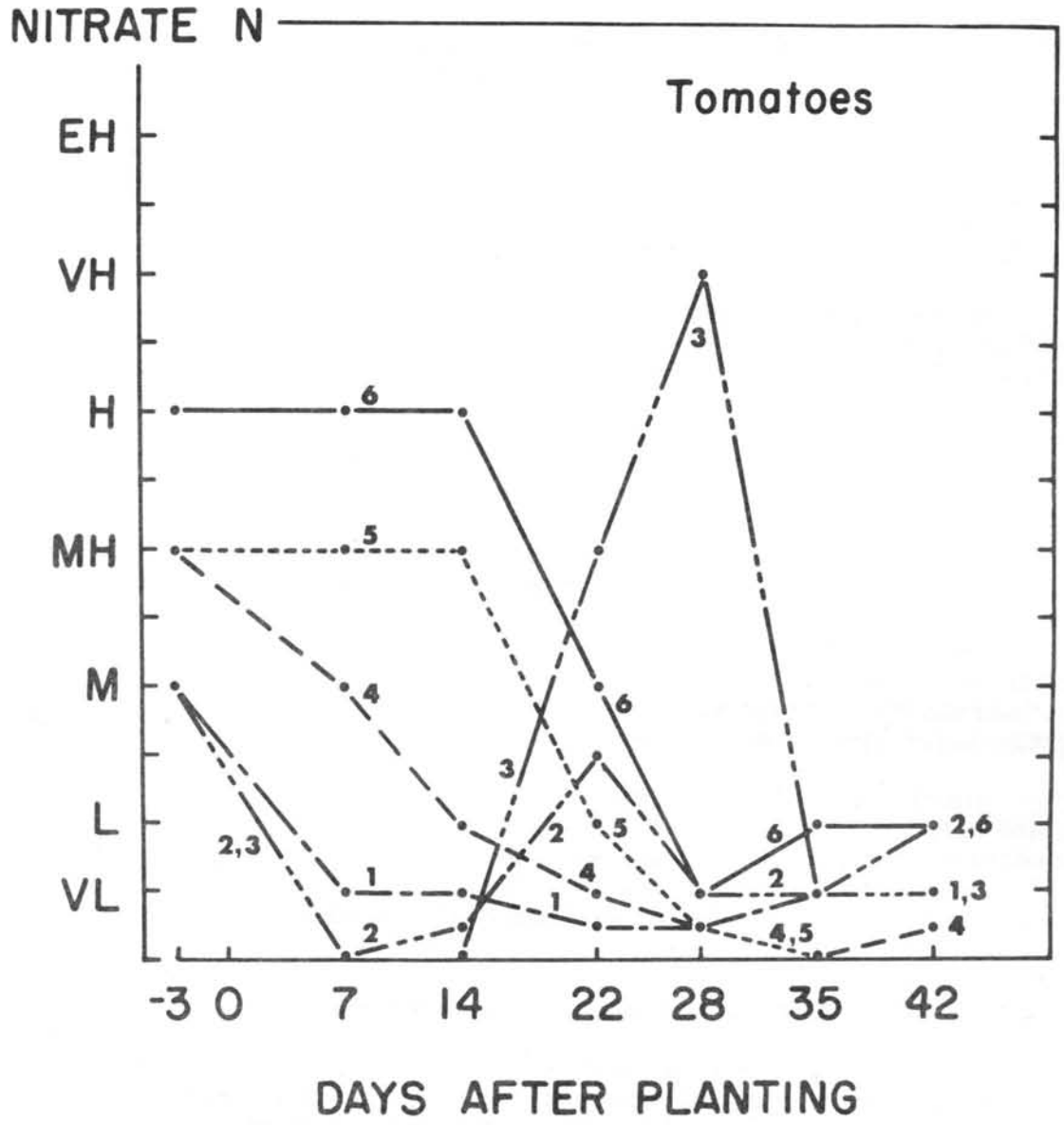


FIGURE 2. Changes in nitrate nitrogen concentrations with time in Windsor loamy sand planted with tomatoes and amended and fertilized according to treatments 1-6 (Table 8). Morgan soil test (24) L = low; M = medium; H = high; V = very; E = extremely.

since the conductivity of a saturated solution of gypsum is 2.20 mmhos/cm (35), and these wastes contain substantial gypsum.

Although the salinity measurements were started 5 days after the lower leaves of the mycelium treated plants were injured, they do not offer

a clear explanation for the injury. Tomato is considered moderately tolerant of salinity (5). In experiments in Israel, the yield of tomatoes was reduced by 10% for every 1.5 mmhos/cm increase in EC_e above 2.0 mmhos/cm; the yield of tops (stems and leaves), however, was not affected by salinity (32). We estimate that the salinities of treatments 1-3 were initially about 3 mmhos/cm. Thus it is not clear whether the moderately saline conditions of treatments 1-3 injured the tomatoes. Although the slight stunting of growth is indicative of excessive salt, the N deficiency of the lower leaves could well result from a lack of available N. Indeed, the nitrate N concentrations in treatments 1-3 during days 0 to 14 were diminishing rapidly to very low levels.

The second experiment with tomatoes was undertaken to determine whether a constant supply of nitrate and ammonium N to treatments 1-3 would improve the growth of tomatoes. Treatment 7 (Table 8) served as the control. When soil tests indicated low nitrate N in a treatment, nitrogen was added at the rate of about 66 lbs/acre. Even with this intensive management considerable N was immobilized in the mycelial treatments 1-3 during the first 14 days. Treatments 2 and 3 had to be supplemented with N three times in 13 days, while the original modest supply of N in the check, treatment 7, was maintained for 17 days. After 17 days, however, treatment 7 needed more nitrate and ammonium N than did treatments 1-3 to maintain satisfactory nitrate N levels in the soils. Apparently mineralization of the mycelial residues became more effective at this time. The readily available nitrate N supply in the experimental substrates is shown in Fig. 3. For simplification the nitrate N concentrations in treatment 2 are not shown, since they were generally only slightly lower than those of treatment 3.

Ammonium N in the different treatments changed much less with time than did nitrate N. In control treatment 7, nitrification rapidly diminished the moderate ammonium content to zero; the addition of ammonium nitrate on days 25 and 34 briefly raised the ammonium N level on day 38. The mycelial treatments, on the other hand, contained medium levels of ammonium N most of the time. On day 20, after a week of no N addition, the ammonium N levels in treatments 2 and 3 increased to medium high levels and then declined reflecting increased nitrification in the mycelial treatments.

Thus, these nitrate and ammonium tests (Figs. 2 and 3) show that two processes occur when mycelial residues are added to sandy soil in pots. Ammonification of the mycelial residues apparently occurs as soon as they are incorporated into the soil. Ammonium N increased for about 3 weeks, although not to excessive levels, and then gradually disappeared during the following 3 weeks. During the first 2 weeks nitrate N was readily immobilized or not formed, after which nitrification was discernable. This apparent inhibition of nitrification may have been caused by one or more factors, the most likely one being that salts from the mycelial waste produced an osmotic concentration too high for optimal activity of nitrifying bacteria.

In spite of repeated fertilization the response of the tomato plants was not much different from that observed in the first experiment. On day 3 the lower leaves in treatment 2 were yellow and mottled. On day 9 all plants in the mycelium treatments were yellow and mottled, with those in treatment 2 showing the most severe symptoms. Again, it is not quite clear why the plants

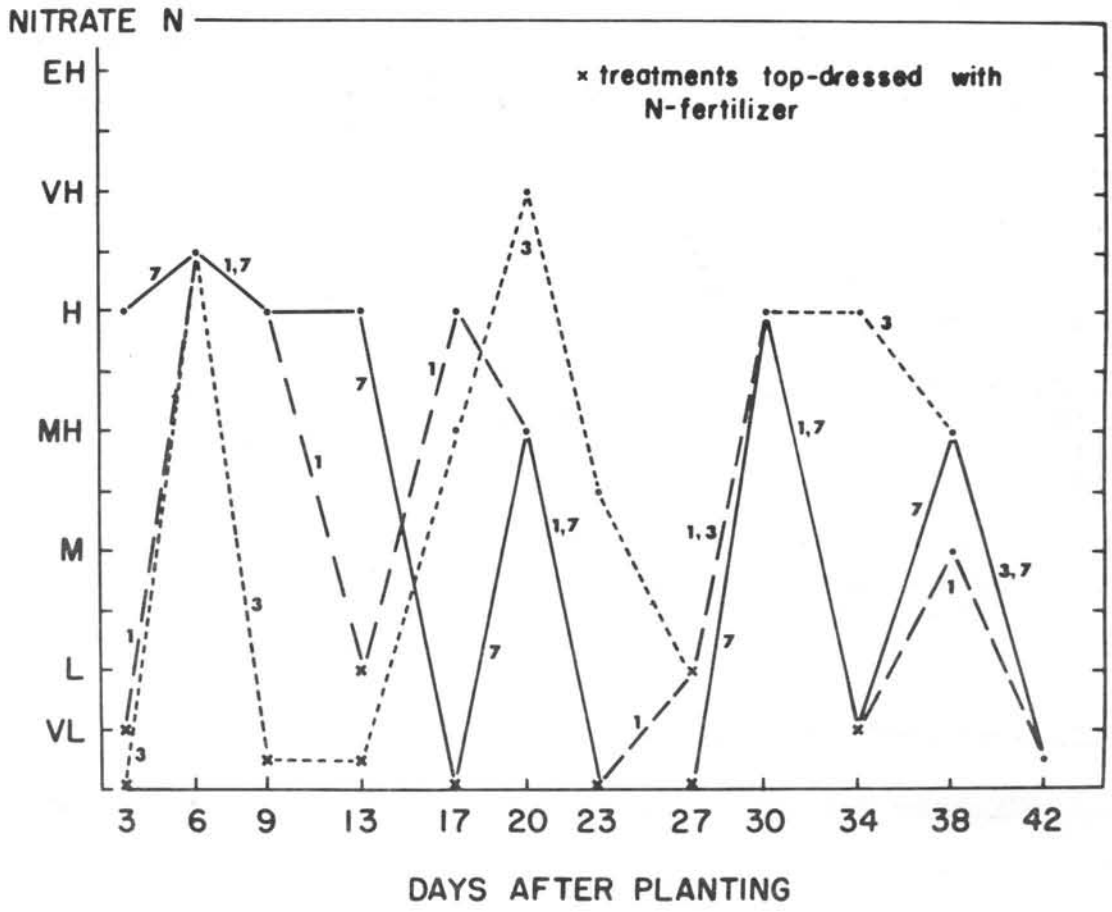


FIGURE 3. The effect of nitrate and ammonium top-dressings on nitrate concentrations in Windsor loamy sand planted with tomatoes and amended and fertilized according to treatments 1, 3, and 7 (Table 8).

in treatment 2 (36 tons mycelium/acre) were injured more than the plants in treatment 3 (118 tons/acre). The soil salinities of the three treatments (Table 10) did not increase with increasing rates of mycelium application which at least explains why the plants in treatment 3 were no worse than the plants in treatment 2.

TABLE 10. Electrical conductivities (EC_e in mmhos/cm) in Windsor loamy sand in pots planted with tomatoes.

Days after transplanting	Treatment No. 1	2	3	7
3	2.4	2.4	2.1	1.7
23	2.1	2.4	2.2	.6
45	2.4	2.4	1.9	.8

The results of the soil tests, fertilizations, and growth measurements of the tomatoes in the second experiment generally confirmed those obtained in the first. In both, the mycelial residues incorporated into soil immobilized nitrate N for about 2 weeks. Although the yellowing of the bottom leaves resembled that caused by N deficiency, the injury occurred too readily to be explained solely by an insufficient supply of nitrate N. The rapid yellow mottling of the lower leaves after transplanting and the stunted or compact growth indicates an effect of salinity. The salinity of the mycelial wastes apparently not only suppressed nitrification in the substrates and possibly the uptake of N by the plants, but also disrupted the normal N metabolism within the plant (18).

There remains the possibility that the damage to the plants was caused by heavy metals in the mycelial wastes. To investigate this, selected leaves of the tomato plants were analyzed (17) with the results shown in Table 11. None of the metals had concentrations in the leaves considered sufficient to produce toxic symptoms (9). The Cu and particularly the Zn contents of the plants grown in the mycelial substrates were higher than those of the plants grown in the fertilized soil substrates. The increased Zn concentrations in the tomato leaves of treatments 1-3 undoubtedly reflect the relatively high Zn content of mycelial wastes (Table 1). But even the highest Zn concentrations did not approach those known to be toxic.

TABLE 11. Concentrations of metals in dry tomato leaf samples (ppm).

Treatment No.	Zn	Cu	Cd	Ni	Pb
1	87	19	.7	3.1	5.0
2	112	28	.5	3.0	3.5
3	151	24	.4	2.8	5.0
4	40	14	.6	2.8	-
5	36	14	.6	5.0	8.0
6	32	15	.8	2.5	5.0

It is also possible that the high sulfate-sulfur concentrations of the gypsiferous mycelial wastes are toxic to plants. In the few studies of excessive sulfur accumulation from substrates in plants, the leaf symptoms were interveinal yellowing and marked reduction in size (9). Analyses of the tomato tissue for total sulfur showed that the plants in mycelial treatments 1-3 contained about 1.8 times as much sulfur as did the plants in treatments 4-6. The literature is not clear, however, whether the concentrations we observed should be considered toxic to tomatoes (9). Therefore, we conclude that the salinity of the mycelial wastes is the principal cause of the yellow mottling of the lower leaves of the tomato plants and of the suppression of growth in the early stages of development. Whether the reaction of the tomato plants is merely an osmotic effect or a more specific ion effect is an open question.

The growth of oats gave us a measure of the residual fertility of the amended and fertilized Windsor loamy sand. The total weights of the mature oat plants exclusive of roots are presented in Table 9 where it is evident that yield increased with increasing application of mycelial waste. The

smallest addition of mycelial waste, 12 tons/acre (wet wt), matched the residual nitrogen fertility of the slow release urea-form of treatment 4. Thus, although their N content is low, mycelial residues have a rather lasting effect as a nitrogenous fertilizer.

The physical properties of the different substrates used for growing the tomatoes and oats are given in Table 12. As noted earlier with container grown stock, mycelial wastes added to loamy sand improve the air capacity of the soil, increasing it from 3% to 6-9% (v/v). The oxygen diffusion rates (O.D.R.) show the same effect. The O.D.R. of the fertilized soil (treatments 4-6) was significantly lower than those of the mycelial treatments. However, an O.D.R. of 30 x 10^{-8} g cm^{-2} min^{-1} or higher is considered satisfactory for crop growth (19). The percolation rate of saturated samples was drastically lowered by the highest rate of mycelial waste application. This decrease in hydraulic conductivity was likely caused by the swelling of the mycelium. However, a permeability rate of 4 in./hr is considered satisfactory and should not cause waterlogging or aeration problems in containers.

TABLE 12. Physical characteristics of Windsor loamy sand amended with mycelial waste in 1-gallon containers.

Treatment No.	Mycelium added tons/acre (wet wt)	Percol. in./hr	B.D. g/cc	T.P.S.	Air	R.A.W.	W.B.C.	O.D.R.
				vol %				
1	12	19	1.35	48	6	15	6	51
2	36	21	1.40	47	6	15	5	75
3	108	4	1.26	52	9	14	6	82
4	0	11	1.43	46	3	14	8	36
5	0	-	-	-	-	-	-	41
6	0	-	-	-	-	-	-	35

Percol. = Percolation rates in in./hr.
B.D. = Bulk density or volume weight (x 62.42 = dry wt in lbs/cu ft).
T.P.S. = Total pore space or water content at saturation.
Air = Air space at 15 cm water suction or container capacity.
R.A.W. = Readily available water, released between 15-50 cm suction.
W.B.C. = Water buffer capacity, released between 50-100 cm suction.
O.D.R. = Oxygen diffusion rate value in g x 10^{-8}/sq cm/min, average for 10 measurements in containers following cropping with tomatoes and oats.

Thus, these observations on the effect of mycelial waste on the physical properties of sandy soils suggest the following: At moderate application rates (about 100 tons/acre, wet wt) the amendment appeared to improve the porosity and aeration of the soil; however, the amounts of readily available water retained by the soil were hardly increased by the mycelium additions. Furthermore, heavy applications of mycelial wastes might decrease structural stability, impairing the flow of water through the soil.

Results with Corn and Oats

The early development of the corn plants is presented in Table 13. During the first 5 weeks, the height of the plants showed a consistant gradation: the greater the addition of mycelial residues, the shorter the corn plants. However, the lowest rates (treatments 1 and 2, 12 and 36 tons/acre of mycelial residues) had heights comparable to those of control treatment 6. The highest rate of mycelium (treatment 3, 108 tons/acre of mycelial waste) significantly depressed the height as measured on days 21, 28, and 35. On day 45, however, this suppression of height compared to the control was not statistically significant. On day 48, after harvesting and drying the plants, the yield of dry matter in treatment 3 was significantly less than the yield of the control (Table 9).

TABLE 13. Height growth (cm) of corn plants in Windsor loamy sand as affected by mycelial amendments (treatments 1-3) and by organic and inorganic N-fertilizers (treatments 4-6). Treatments are described in detail in Table 8.

	Day				
Treatment No.	11	21	28	35	45
1	7	28	48	80	119
2	6	26	45	74	120
3	6	22	36	61	114
4	8	32	54	88	123
5	6	24	44	74	116
6	7	28	46	77	117
L.S.D. (0.05)	1	5	6	8	N.S.

N.S. = Not significant.

Weekly soil testing did not reveal the exact cause of the stunting in treatment 3. But, as with the tomatoes, two factors may have played a role: (i) the relatively high concentration of salt in treatment 3 throughout the observation period (Table 14), and (ii) the complete immobilization of nitrate N during the second week of the growing period (Fig. 4). By comparison, the relatively low salinity of treatment 4 in combination with a sufficient supply

TABLE 14. Electrical conductivity of saturation extracts (EC_e in mmhos/cm) of the substrates of treatments 1-6 (see Table 8) and its change with time after seeding with corn.

	Treatment No.					
Days after transplanting	1	2	3	4	5	6
14	3.0	3.0	2.6	1.8	1.9	2.3
28	2.5	3.0	3.0	2.4	2.5	2.8
42	2.0	2.4	2.6	1.2	2.0	2.3
53	1.7	2.0	2.5	0.9	1.2	1.4

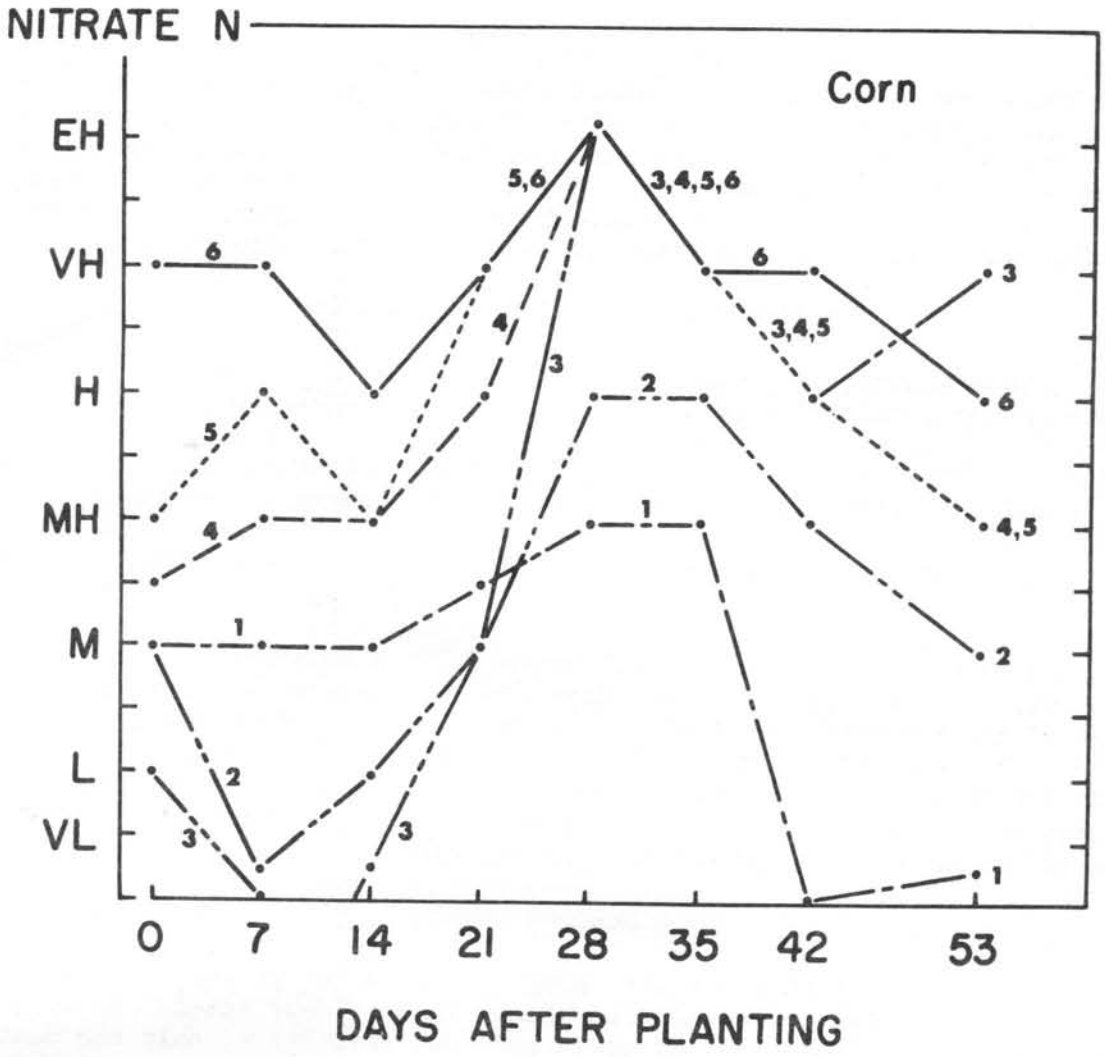


FIGURE 4. Changes in nitrate concentrations with time in Windsor loamy sand planted with corn and amended and fertilized according to treatments 1-6 (Table 8).

of nitrate and ammonium N produced plants that were significantly taller on days 29 and 35 than those in treatment 6 (Table 13). Treatment 1, the lowest rate of mycelial residues, had a relatively low and consistently decreasing soil salinity and no apparent immobilization of nitrate N. However, its nitrate N supply was the first to be depleted and, on day 48, the corn plants of treatment 1 showed definite signs of N deficiency; the top leaves were light green and the bottom leaves were yellowing.

The oats seeded in the corn stubble again responded well to the residual effect of the heavy application of mycelial waste (Table 9). Yields were generally less than those following tomatoes since the corn plants were not topdressed with inorganic fertilizer, while the tomato plants received several topdressings, the last one just before they were harvested.

In conclusion, it appears that corn, considered only moderately salt tolerant, did not show any foliar abnormalities as a result of the additions of mycelial waste to Windsor loamy sand. Except for a retardation of the early top growth, no other signs of phytotoxicity were observed. Furthermore, observations of root growth did not indicate any inhibitory effect of the mycelial residues on the growth and distribution of roots in the planter boxes with glass panels. Root development in Windsor loamy sand amended with mycelial waste was just as profuse as that in the unamended soil.

GERMINATION STUDIES WITH CORN AND RYE

In a separate experiment we studied the effect of additions of mycelium on the germination of corn and rye. The rye was included because field observations in eastern Connecticut suggested that some stands of rye were affected by uneven application of mycelial residues. The reasons for the germination study of corn were twofold: (i) emergence of the corn in the containers of our greenhouse experiment was far from uniform, although it had been seeded carefully with the germ end down; (ii) the emergence of weeds, mainly crabgrass, was significantly lower in the treatment 3 and 5 containers. The Windsor loamy sand used in our pot experiments had been fallow for some years and was heavily infested with crabgrass. A nematocidal effect of cottonseed meal has been known for some time (25) but we are not aware of any knowledge of its apparent phytotoxicity to germinating seeds or at least crabgrass seeds. This observation may form an interesting problem with practical aspects for further investigation. For the present, however, we limited our studies to the effect of mycelial residues at various rates on the germination and young seedling growth of corn and rye.

Methods

The germination media were prepared by adding pulverized, air-dried mycelial waste to air-dried Merrimac sandy loam at four rates: 0, 108, 216 and 324 tons/acre (wet wt). No media received fertilizer; only the control (treatment 1) was limed to raise the pH to that of the mycelial media (pH 7.0 to 8.0). Each medium was placed in a small germination tray, well watered, and seeded with corn or rye. Each tray received 18 corn or 30 rye seeds; the seeds were individually planted with the germ end down. Each treatment was replicated twice.

Each tray was wrapped in a polyethylene bag to maintain high humidity and kept at about 78°F. When the young seedlings touched the polyethylene cover it was removed, and the trays were placed in a well lighted room at 65°F.

Each day we counted the number of seeds germinated, while 3 days after seeding we measured the length of the young shoots. On this day the control trays (treatment 1) had to be moved to the 65°F temperature room. From day 6 until day 11 all trays were kept in a greenhouse (minimum night temperature about 60°F), after which the final germination percentage and young shoot growth were determined. The shoots were cut off at the soil line and weighed. After harvesting, the salinity of the various media was determined.

Since rye is normally seeded in cool weather, 78°F may have been too warm for good germination. Thus, in a second experiment with rye, the trays were kept at temperatures of 63° to 67°F for 6 days and then moved to the greenhouse.

Results

Table 15 shows that the percentage germination of corn was not greatly affected by the mycelial waste. However, the rate of germination and especially the shoot growth of the young seedlings were apparently suppressed by the mycelial additions.

TABLE 15. Germination studies with corn and rye.

Treatment No.	Medium*	Germination and seedling shoot growth: After 3 days at 78°F — Germ. %	Shoot growth range in mm	Exposure to 78°F days	On day 11** — Germ. %	Avg. weight g/plant
			Corn***			
1	0	97	20-72	3	97	.55
2	108	94	11-43	3	100	.34
3	216	75	4-20	3.5	97	.35
4	324	97	2-15	4	100	.46
			Rye#			
1	0	78 (72)##	10-45 (6-44)	3	85 (87)	.10 (.08)
2	108	54 (62)	4-18 (2-27)	5	68 (85)	.07 (.05)
3	216	35 (25)	3-11 (1-18)	6	78 (85)	.06 (.04)
4	324	8 (2)	1-6 (1-13)	6	62 (85)	.04 (.03)

* Merrimac sandy loam mixed with mycelium residues at the rate of 0, 108, 216 and 324 tons/acre, respectively.

** After terminating exposure to ambient temperatures of about 78°F, trays were kept in a well lighted room at 65°F until day 6, after which all trays were placed in a greenhouse (minimum night temperatures around 60°F).

*** Agway 595S, 1973 Hybrid.

\# Stanfords Balbo

Results in parentheses were obtained when trays were kept at about 65°F for the first 6 days and then moved to the greenhouse.

The young shoot growth on day 3 demonstrates the suppressive effect of the additions of mycelium. Compared with the control, the growth in treatments 2, 3, and 4 was increasingly suppressed by increasing rates of mycelial waste additions. On day 11, this effect on the fresh weight of the shoots was less evident. The growth of the seedlings in treatments 3 and 4 was apparently stimulated by the additional exposure to the favorable conditions of high temperature and humidity for another 1/2 and 1 day, respectively.

We conclude that applications of mycelial waste at relatively high rates did not affect the percentage germination of corn, but slightly retarded the rate of germination and suppressed the growth of young seedlings. This effect is apparently due to the high salinity of the mycelial wastes. On day 11 the salt concentrations in the mycelial media were about 2.5 times as high as in the control media; the conductivities averaged 2.55 and 1.0 mmhos/cm, respectively. For seedlings, conductivities less than 1.0 mmhos/cm are generally preferred.

Table 15 shows that the mycelial wastes had a more pronounced effect on the germination of rye. Not only the rate but the percentage of germination, and the young shoot growth, were suppressed by increasing additions of mycelial waste to Merrimac sandy loam. By day 11, the percent germination had increased but shoot growth was still affected, in spite of the additional exposure of the mycelial treatments 2, 3, and 4 to the favorable polyethylene bag conditions of higher temperature and humidity.

The salinity of the germination media of treatments 1, 2, 3, and 4 was 1.2, 2.5, 2.8, and 2.8 mmhos/cm, respectively. The high concentrations of soluble salt are attributed to the calcium sulfate in the waste. Morgan's soil test of the germination media on day 11 showed that fertility levels were low to medium and did not contribute to the high salinity. The fertility levels in the rye germination media, however, were slightly higher than those in the corn media which may account for the slightly higher EC_e values in the rye media.

The results of the second test with rye (see Table 15 data in parentheses) confirmed most of the conclusions obtained with the first experiment. The main exception being that the eventual germination of the rye seeds, as counted on day 11, was generally satisfactory and not affected by the various rates of the mycelial additions.

In summary, the relatively high application of mycelial waste (108 to 324 tons/acre, wet wt) to a sandy loam soil did not affect the eventual germination of corn seed, although the rate of germination and especially the subsequent growth of the young seedlings were retarded and stunted.

The rye seeds responded in a similar, but more pronounced, manner. Particularly under the rather high ambient temperature used to germinate the corn (78°F), the mycelial residues were detrimental to the percentage and rate of germination. Furthermore, a definite, increasingly suppressive effect was observed with increasing application rates, particularly on the shoot growth of the young seedlings. The inhibitive effect of the mycelial waste additions appeared to be due to their soluble salt content. Under cooler ambient temperatures (65°F) during germination and emergence, the eventual germinations on day 11 were satisfactory and not affected by the mycelial residue applications.

FIELD EXPERIMENTS WITH TOBACCO

Field experiments with tobacco had two objectives. First, we wished to determine if the mycelial residues could substitute for the conventional

but scarce and expensive organic nitrogen fertilizers such as cottonseed meal, castor pomace and others (11) used in tobacco culture. Second, it has been shown that some organic materials incorporated in soil can reduce populations of plant parasitic nematodes as well as lessening the severity of some soil-borne diseases (25,26). Hence, we wished to determine whether mycelial residues would reduce damage from the tobacco cyst nematode under field conditions. Two years of experiments with shade-grown tobacco are reported here.

Methods

Experiments were conducted in 1972 and 1973 at the Valley Laboratory in Windsor on a typical tobacco soil, a Merrimac sandy loam (33). The four treatments of both experiments (Table 16) were replicated and randomized in three blocks. Each block covered one 33 ft x 33 ft bent of the shade tent or

TABLE 16. Amendments and fertilizations of Merrimac sandy loam for shade-grown wrapper tobacco in 1972 and 1973.

	Analyses $N-P_2O_5-K_2O$	Treatment No. 1	2	3	4
1972					
Materials, lbs/acre (wet wt)					
Mycelial waste, 66% moisture	2.5-0-0	8720	17,440	17,440	
Wood fiber waste, 76% moisture	.5-0-0			38,544	
Fertilizers, lbs/acre					
Cottonseed meal	6.5-3-2	1280			2520
Triple superphosphate	0-45-0	250	330	330	210
Potassium nitrate	13-0-44	200	200	200	200
Potassium sulfate	0-0-50	150	200	200	100
1973					
Materials, lbs/acre					
Mycelial waste '73, 64% moisture	1.98-0-0	23,000			
Mycelial waste '74, 44% moisture	1.68-0-0		17,400		
Fertilizers, lbs/acre					
Tobacco fertilizer mix	6-3-6				3760
Urea-form	38-0-0			430	
Triple superphosphate	0-45-0	330	330	330	
Potassium nitrate	13-0-44	200	200	200	
Potassium sulfate	0-0-50	200	200	200	

1/40 of an acre; thus each plot, measuring 1/160 of an acre, had five rows of 11 cigar-wrapper tobacco plants. These blocks were located in a part of the shade-field heavily infested with the tobacco cyst nematode Heterodera tabacum, Lownsbery & Lownsbery.

The control for both experiments, treatment 4, is the conventional fertilization for commercial production of cigar tobacco (2,11). It provides 190 lbs/acre of N with about 164 lbs from cottonseed meal and 26 lbs from nitrate N, as well as 150 lbs/acre of P_2O_5 and 190 lbs/acre of K_2O.

In 1972 mycelium added to treatments 1 and 2 replaced half or all of the organic N of control treatment 4. The wood fiber waste was added (treatment 3) for two reasons: first, for its nematocidal effect, since cyst nematodes are one of the principal pests in Connecticut tobacco soils. Second, when this experiment was begun, the rate of release of inorganic N from mycelial residues was not known. If most of the nitrogen in the mycelial residues was readily released, the wood fibers would induce microbial fixation of this inorganic N preventing the possible loss of nitrate N by leaching. Nitrogen from mycelium was expected to be utilized at the rate of 1 lb of N for each 100 lbs of carbonaceous material. The amount of cellulosic waste added was based on the utilization and later mineralization of half the amount of organic N supplied by treatment 4, i.e., 82 lbs/acre of N. We expected this nitrogen to be released at a rate comparable to that of other organic nitrogenous fertilizer materials from animal and plant sources (11).

In 1973 the primary objective was to retest the mycelial treatments for their effect on the cyst nematode. The results of the 1972 experiment were inconclusive and warranted further examination. The location was adjacent to that of the previous test in the same shade tent. A count of the nematode population 8 days before the application of the experimental treatments confirmed that the area was still heavily infested (Table 20).

The 1973 experimental design and procedure was similar to the 1972 experiment. The soil was fertilized and amended as shown in Table 16. The organic N of treatments 1 and 2 was derived from mycelial residues. For treatment 1 a raw mycelial waste was used, freshly received from the pharmaceutical plant, and containing 1.98% N. In treatment 2 we used mycelial residues remaining from those used in 1972, which contained 1.68% N. This provided two treatments in which to observe nematocidal effects and also provided an opportunity to determine whether a year of "aging" of the residues had any effect on the response of tobacco or nematodes.

These wastes had been stored outdoors since 1972 in a 2 to 3 foot high pile, subject to wetting, drying, and freezing. This exposure to the elements apparently did not impoverish the mycelial waste. Its salinity (EC_e) in June 1973 was slightly higher than that of the fresh mycelial waste (8.50 and 7.25 mmhos/cm, respectively). Readily available plant nutrients assayed on June 15, 1973 by Morgan's method (24), were also higher in the 1972 mycelial waste than in the raw 1973 waste. That the mycelial wastes were not leached is closely related to their peculiar moisture characteristics. The mycelial residues hold their moisture tightly and are almost impervious to water, as is indicated by the data in Table 2. As a result, the leaching of mycelial waste by percolating water is understandably poor.

The organic N of treatment 3 was urea-formaldehyde, more or less water insoluble and slowly transformable into ammonium N. Treatment 3 made it possible to test this N source against the natural organic N from cottonseed meal in treatment 4. Furthermore, these treatments allowed us to observe the nematocidal effect of the decomposing cottonseed meal in treatment 4 (25).

In 1972 as well as in 1973 the organic N supplied by treatments 1, 2, and 3 was supplemented with standard inorganic fertilizer ingredients to match the composition of treatment 4 (Table 16); in doing this, the phosphorus and potassium levels of the mycelial and cellulosic wastes were disregarded.

Following transplanting, sidedressings with fertilizer were based on visual observations of the quality and quantity of foliage and on soil tests (24). In 1972, 19 days after transplanting the tobacco seedlings, treatment 3 was sidedressed with ammonium nitrate at the rate of 40 lbs/acre of N. On day 29 all treatments received a standard tobacco sidedressing of 250 lbs/acre of sodium nitrate and 200 lbs/acre of 6-3-6 (cottonseed meal base). This supplied N at the rate of 52 lbs/acre to replace nitrogen lost by leaching during heavy early summer rains. In 1973 we increased the frequency of testing the soils in the various plots; they were analyzed every 3 or 4 days so that sufficient N could be maintained at all times in all plots. All treatments were sidedressed with sodium nitrate at the rate of 32 lbs/acre of N 32 days after transplanting; this was repeated on day 42 and on day 54.

The shade-grown tobacco was a commercial strain (L) developed from "Connecticut 49", a wrapper-type cigar tobacco (Nicotiana tabacum L.). The crop was grown according to standard practices (2); in 1972 the seedlings were transplanted 6 days after application of the treatments and, in 1973, 1 day after application. The effect of the various treatments was evaluated on the basis of the size and color of the leaves and by measurements of plant height and stem diameter. The stem diameter of flue-cured tobacco (measured 3 in. above the soil surface) is a good index of total leaf area (34); here it was used simply to measure overall growth. Instead of normal, weekly primings, the leaves were harvested only once: the first five bottom leaves were dropped and the following four (1972) or three (1973) mature leaves on the plant were picked. These leaves were taken to a commercial grower for further processing and final evaluation of yield and quality. Growth and yield measurements were averaged for the three inner rows of plants in each plot.

The cyst nematode infestations in the soil were investigated using the sugar flotation method on soil cores (25). In 1972 the first count was made a few days before the treatments were applied, while the second count was made 28 days after transplanting the tobacco. In 1973 the cyst nematode populations were investigated more frequently, namely 8 days before transplanting and subsequently on days 7, 14, 27, and 43. The roots themselves were investigated on day 43 (25).

Results

In 1972 the mycelial and wood fiber waste treatments produced less growth than did control treatment 4 at 36 days, but differences in yield and

quality after day 36 were not statistically significant (Table 17). A visual inspection on day 12 revealed that plants in treatment 3 were yellowish and smaller than the plants in the other treatments. Plants in treatment 2 showed less pronounced but similar symptoms. Although a soil test on day 6 showed the nitrogen in all plots to be more than sufficient, the clearly stunted and yellow plants of treatment 3 were sidedressed with nitrogen on day 19. The N was added since it was possible that the very fine wood fibers had immobilized more N than was anticipated. On day 86, the salinity of the plots was determined. The salt concentrations in treatments 1, 2, and 3 were several times as high as in control treatment 4; the conductivities averaged 1.0, 1.8, 1.8, and 0.4 mmhos/cm, respectively. Although these concentrations of soluble salts are only moderately high, they likely were harmful to the young shade-grown tobacco plants. The salt tolerance of young wrapper tobacco is not well known, but salts affect many other plant species by depressing the uptake of nitrogen and by disturbing metabolic pathways within the plants (3,18). The severe stunting and yellowing of the plants in treatment 3 apparently was due to a combination of effects of soil salinity and of N-utilization by the wood fibers. As the growing season advanced, these effects diminished, as indicated by the non-significant differences in growth and yield among treatments (Table 17).

TABLE 17. The effect of mycelial residues, wood fiber waste, and nitrogenous fertilizers on growth, yield and quality of wrapper tobacco.

		Treatment No.				L.S.D.
		1	2	3	4	(0.05)
		1972				
Day 36 plant height	cm	51.0	42.2	22.2	49.0	14.6
	CV,%	15.6	16.3	25.5	20.5	N.S.
Day 63 leaf yield*	lbs/acre	183	168	177	187	N.S.
leaf quality	grade index	270	203	282	364	N.S.
Day 104 stem diameter	mm	24.7	24.6	24.9	26.2	N.S.
		1973				
Day 26 ranked on growth**		3	3	2	1	-
Day 33 ranked on wilting***		3	4	1	2	-
Day 46 plant height	cm	92	88	105	112	18
Day 60 leaf yield#	lbs/acre	142	136	158	148	N.S.
leaf quality	grade index	291	250	303	296	-

* For four-leaf priming.
** 1 = best, 4 = poorest growth and leaf color.
*** 1 = none or slight wilting, 4 = worst, mostly severe wilting.
# For three-leaf priming.
CV,% = coefficient of variability, percent.

In 1973 the tobacco plants were evaluated visually on day 26 (Table 17). Based on the general appearance of the plants, treatment 4 ranked first and both mycelial treatments (1 and 2) ranked last. The plants in mycelial treatments 1 and 2 had a yellow cast, which was less evident in treatment 3, and not seen in treatment 4. The yellowing of the plants in treatments 1 and 2 could not be explained by an insufficient supply of N, since the nitrate N supply was kept at adequate levels throughout the growing season. Again, the high soluble salt concentrations seem to offer a better explanation of the detrimental effect of the mycelial treatments (Table 18). Salinities of 3.0 and 3.3 mmhos/cm are indicative of excess concentrations of soluble salts for tobacco. This conclusion is confirmed by the ranking according to wilting on day 33 (Table 17). This ranking is consistent with the increasing salt concentrations (Table 18) which suggests that the increase in osmotic pressure of the soil solution decreased the availability of water to the plants. The final commercial evaluation of the harvested, cured, fermented, and sorted leaves ranked the treatments from high to low as follows: 3, 4, 1 and 2 (Table 17).

TABLE 18. Electrical conductivity (EC_e in mmhos/cm) in Merrimac sandy loam under field-grown shade tobacco.

Days after transplanting	Treatment No.			
	1	2	3	4
4	3.0	3.3	1.6	1.4
42	1.5	2.4	0.6	0.7

These 1973 observations on the development of the tobacco plants are generally in agreement with the results obtained in 1972. The salt concentrations in the soil treated with mycelial waste are relatively high and injurious to the young shade-grown tobacco plants.

The nematocidal effects of the mycelial and wood fiber wastes in 1972 were evaluated by counting nematodes and by observing plant growth. Populations of nematodes, including Heterodera spp. have been shown to decrease when soils are amended with crop residues, compost, sawdust, paper, manure and organic fertilizers (25). Miller, et al. (26) found that populations of plant-parasitic "meadow" nematodes were reduced by soil amendments of mycelial residues, cellulosic wastes, and chitin. Thus, prior to treatment and on day 28, counts were made in all plots of larvae and eggs of the cyst nematode. The results are presented in Table 19. As is characteristic for most plant-parasitic nematode infestations, the population varies greatly from location to location. In spite of this variability it appears that in most plots on day 28 the cyst nematode populations were reduced. The cellulosic wastes of treatment 3, and to a lesser extent the mycelial residues of treatment 2, seemed most effective in reducing the population of nematodes.

The second evaluation of nematocidal effects was based on the growth of the tobacco. The above-ground symptoms of nematode infestations are stunting, slow growth and wilting which results in a very uneven stand. Thus, a more even stand might be expected in some treatments. A statistical analysis of the measurements of plant height in the various plots on day 36 showed

TABLE 19. The effect of mycelial residues, wood fibers, and cottonseed meal on populations of the tobacco cyst nematode Heterodera tabacum, Lownsbery & Lownsbery in Merrimac sandy loam under field-grown shade tobacco in 1972.* Treatments are described in Table 16.

	Replicate					
	I		II		III	
	Larvae	Eggs	Larvae	Eggs	Larvae	Eggs
Pretreatment	1795	111	103	762	45	320
Treatment 1	13	0	42	402	7	2
2	3	0	13	15	9	8
3	2	6	1	1	1	1
4	28	96	1	1	53	196

* Numbers of larvae and eggs in 100 g soil samples.

that although mean plant height and coefficient of variability differed, the latter was not significant (Table 17). From these observations of nematocidal effects, we conclude that the mycelial residues and cellulosic fibers were at least as effective as cottonseed meal in suppressing nematode populations in the soil.

The results of the more intensive investigations in 1973 are shown in Table 20. The cyst nematode population in the soil was not significantly affected by the addition of mycelial wastes. Cottonseed meal seemed to have a slight depressive effect, at least during the first week after its application, although the difference is not statistically significant. Inside the tobacco roots, on the other hand, the cyst nematode population on day 43 was significantly increased by the fresh mycelial wastes (treatment 1) and seemingly suppressed by the aged mycelial residues (treatment 2). We have no explanation for this difference between the two mycelial wastes, but generally speaking we conclude that the nematocidal properties of these wastes, if any, were only modest.

TABLE 20. Effect of mycelial residues, fertilizer, and cottonseed meal on populations of the tobacco cyst nematode Heterodera tabacum in Merrimac sandy loam under field-grown shade tobacco in 1973.

	Treatment No.								L.S.D. (0.05)	
	1		2		3		4			
Time*	Soil**	Root***	Soil	Root	Soil	Root	Soil	Root	Soil	Root
Day -8	29		44		38		53		N.S.	
7	34		45		38		14		N.S.	
14	70		55		80		61		N.S.	
27	63		54		71		49		N.S.	
43	117	110•	107	56	156	84	255	73	N.S.	31

* Days before or after applying mycelial waste and fertilizers.
** Number of cyst nematodes/5-oz cup of soil.
*** Number of cyst nematodes/20 cm of tobacco root.

Thus, in 2 years of experiments, mycelial wastes used as a natural organic nitrogenous fertilizer material caused stunting and yellowing of the early growth of field-grown wrapper tobacco. Quality of the cured leaves was impaired. Such treatments did not suppress the tobacco cyst nematode population in the soil.

FIELD EXPERIMENTS WITH CORN

Experiments with field corn were undertaken to determine whether corn could be grown using mycelial residues as the sole fertilizer. About 50,000 acres of silage corn are grown in Connecticut, and prices of many nitrogenous fertilizers have doubled or tripled in the last few years. Hence, if the mycelial residues proved satisfactory for corn, dairy farmers might find them useful. Two years of field experiments with corn are reported here.

Methods

Silage corn was grown in 1973 and 1974 at Lockwood Farm in Mt. Carmel on Cheshire fine sandy loam. In 1973, growth of corn receiving 100 tons/acre (wet wt) of mycelial residues was compared with that produced from conventional fertilization with 1000 lbs/acre of 15-10-10. In 1974, the experiment was repeated, with additional treatments including measurements of the residual nitrogen supplied by one application in 1973, and the effects of applying 100 tons/acre in 2 successive years.

In 1973, 30 ft x 30 ft plots were treated either with 100 tons/acre of fresh mycelial waste or with 1000 lbs/acre of 15-10-10. The treatments were replicated three times in a randomized block. The treatments were applied on May 2 and harrowed into the soil. Traction in the plots treated with mycelial residues was poor but improved as the material was incorporated into the soil. The plots were harrowed again on May 8 and planted with corn (Zea mays L., var. Agway 595S) on May 14 in 30 in. rows with seeds spaced 5 in. apart. Granular Diazinon was applied for cutworm control at 25 lbs/acre and Aatrex 4L plus Lasso were applied, each at the rate of 1/2 gallon/acre, for pre-emergence weed control.

Germination and emergence appeared normal, with no differential effect of treatment. Weed control appeared somewhat poorer in plots treated with mycelium, probably due to a partial inactivation of herbicide by organic matter in a manner similar to that reported for charcoal (1). Weather conditions during the growing season were generally favorable for corn production.

On August 16, 1973 samples of the ear leaves were taken for heavy metal analysis. After 120 days, on September 10, 1973, 25 plants were harvested from the five center rows of each plot and fresh weights determined. Subsamples were taken for determination of moisture content and total N.

Results

The heavy metal content of the leaves is shown in Table 21. Plants grown on mycelial residues contained more zinc than did those receiving

TABLE 21. Concentrations of heavy metals in corn silage, 1973.

Treatment	Zn	Cu	Ni	Cd	Pb
	ppm				
100 tons/acre mycelium	45	17	3	0.2	6
1000 lbs/acre 15-10-10	19	18	4	0.2	7

conventional fertilizer, but the concentrations are much lower than those considered toxic to plants (9). It should be noted, however, that continued heavy applications of mycelial residues containing zinc could increase the concentrations in the soil to levels toxic to some plants. Indeed, analyses of mature corn tissue in 1974 showed a modest increase in Zn content where mycelial residues had been applied to the soil in 2 successive years.

At harvest, the fresh weights of the plants in the two treatments were not significantly different. However, those grown on mycelial residues had a higher dry matter content. The final yields, expressed on a 70% moisture basis, were: mycelium, 37.0 tons/acre; and conventional fertilizer, 32.3 tons/acre. This difference was statistically significant at the 5% level. The nitrogen content of the corn in the two treatments was not different and averaged 1.28% N or 8.0% protein.

Thus, we concluded that satisfactory yields of corn silage could be obtained with an application of 100 tons/acre of mycelial residues; indeed, in this experiment the yield was about 15% greater than with conventional fertilizer. The yields obtained in these trials are much higher than are generally achieved in practice and derive in part from extrapolating results from small plots to yields in tons/acre. Hence, a relative comparison is perhaps more useful.

Further trials were conducted in 1974. Six additional 30 ft x 30 ft plots were added to repeat the experiment conducted in 1973. Also, both plots were split in order to study additional effects including residual N from the treatment in 1973, the effect of two applications of 100 tons/acre in successive years, and also to determine if supplements of P and K in the presence and absence of nitrogenous fertilizer affected the growth of corn. Details of planting, weed and grub control, and harvesting were essentially identical to those in 1973. Contrary to 1973, there was no differential effect of herbicides evident during the growing season which was marked by a cold, wet spring and a dry summer.

The yields obtained with seven different treatments are shown in Table 22. The yields in the absence of nitrogen fertilizer (T1 and T2) were low. The mycelium added in 1973 (T3), when supplemented with P and K increased yields over the controls (T1 and T2) but yields were less than those obtained with conventional fertilizer (T4). Two successive treatments in 1973 and 1974 (T5) produced yields equivalent to that from a single application (T6), while supplementing the fresh mycelium with P and K produced the highest yield (T7). Yields were less than those in 1973, due apparently to poorer weather and about an 8% reduction in number of plants/acre.

TABLE 22. Corn silage yields, Lockwood Farm, 1974.

Treatment No.	Mycelium tons/acre	Fertilizer Grade at 1000 lbs/acre	Yield at 70% Moisture tons/acre*
T1	0	0-0-0	15.2[a]
T2	0	0-10-10	16.9[a]
T3	100 (1973)	0-10-10	20.9[b]
T4	0	15-10-10	23.5[c]
T5	100 (1973) 100 (1974)	0-0-0	26.6[d]
T6	100 (1974)	0-0-0	28.1[d]
T7	100 (1974)	0-10-10	31.2[e]

* Means sharing the same superscript letter are not significantly different at the 5% level.

Thus, in 2 years of experiments, 100 tons/acre of fresh mycelium wastes produced yields of corn silage 15-30% greater than obtained with 1000 lbs/acre of 15-10-10. Nitrogen content of the tissue in 1973 was not affected by the treatment. The Zn concentration in corn tissue grown on mycelial residues was higher than in corn grown on conventional fertilizer but did not appear to be toxic to the plant.

SUMMARY

Fermentation residues from the pharmaceutical industry in Connecticut were examined to determine their usefulness in agriculture and horticulture. Their chemical and physical properties are typical of most biological tissue, namely high moisture content and relatively small amounts of the plant nutrients N, P and K. The wastes also contain zinc salts and substantial amounts of $CaSO_4$ and filter aids.

The mycelial residues can be composted when mixed with sufficient material to maintain aerobic decay processes. Stockpiled residues do not dewater readily and rapidly become anaerobic with attendant characteristic odors. Losses of plant nutrients and $CaSO_4$ from stockpiled residues were found to be small.

In studies of container-grown plants, mixtures containing 25 to 33 vol % mycelium and wood fiber waste eventually produced chrysanthemums of good to excellent market quality. The early growth, however, showed some stunting and yellowing.

Mixes with 25 vol % mycelium with or without wood fiber waste did not produce harmful effects at any stage of development of junipers, and eventually produced as good or slightly better growth than the standard mixes. The slow release of nitrogen by the decomposition of mycelial residues produced junipers that remained greener throughout winter storage than did the control plants.

Tomatoes grown in the greenhouse in Windsor loamy sand, amended and fertilized with 12, 36, and 108 tons/acre of mycelial waste, wet wt, responded as follows: At all three application rates early growth was retarded, while the older leaves were yellow with a mottled-leaf pattern. At harvest, about 6 weeks after transplanting, the somewhat stunted, bushy and leafy plants grown at the 108 tons/acre rate were as heavy as those grown with commercial fertilizers. The plants receiving lower rates weighed less, indicating an insufficient supply of nitrogen. The zinc concentration in the tomato foliage increased with increasing application of mycelial waste, but even the highest rate did not produce phytotoxic zinc levels.

Oats planted in the pots with the tomato stubble showed increasing growth with increasing application rates of the mycelial waste originally applied to the tomatoes. The mature oat plants were as heavy or heavier than those grown with insoluble urea-form fertilizer, indicating a response by the oats to the slow mineralization of the nitrogenous organic residues.

Corn grown in the greenhouse in Windsor loamy sand, amended and fertilized with 12, 36, and 108 tons/acre of mycelial waste (wet wt) responded without any foliar toxicity symptoms. The early growth was not yellow-mottled and only at the highest application rate of mycelial waste was a retardation and stunting of growth observed.

This stunting was also observed in germination studies with corn and rye. High applications of mycelial wastes (108 to 324 tons/acre, wet wt) to a sandy loam soil did not significantly affect the eventual germination of the seeds, although the rate of germination, and especially the subsequent growth of the young seedlings, particularly of the rye, was retarded and stunted.

In 2 years of experiments, shade-grown wrapper tobacco did not benefit from mycelial residues applied at rates equal to the organic nitrogen in a standard commercial tobacco fertilizer mix. Indeed, the residues were somewhat detrimental, causing yellowing and stunting of the early growth. Repeated counts of the tobacco cyst nematode in the soil did not show any nematocidal effect of the mycelial waste applications.

In field experiments at Mount Carmel the over-all response of corn to an addition of 100 tons/acre of mycelial residues (wet wt) was favorable. In this fertile Cheshire fine sandy loam, the nitrogen of the mycelial residues produced as good or better forage as did the standard inorganic fertilizer.

The results of these experiments lead to the following conclusions about the suitability of raw mycelial waste as a nitrogen fertilizer and as an organic soil amendment:

The nitrogen in raw mycelial waste, slightly less than 2% on a dry wt basis, is present in several forms. The principal forms are organic, insoluble but slowly available, while the remainder is the readily available nitrate and ammonium nitrogen. When incorporated into soil nitrate N is rapidly immobilized for about 2 weeks. After this period nitrate accumulates readily, apparently as a result of increasing nitrifying activity in the soil.

The limiting factor in the use of raw mycelial waste as a nitrogen fertilizer appears to be its total soluble-salt concentration. The salt not only suppresses nitrification in the substrate and possibly the uptake of nitrogen by the roots, but it also appears to disrupt the normal nitrogen metabolism within the plant. As a result, crop response to raw mycelial waste is largely controlled by the salt tolerance of the plants. Salts are generally more toxic to young than to old plants as shown here by depressed early growth and foliar symptoms. Field corn and grain crops (oats and rye) responded favorably to fairly heavy applications (100 tons/acre, wet wt), which only slightly delayed seed germination and stunted early growth. Tobacco, tomato, and chrysanthemum showed a yellow mottled-leaf pattern on the lower leaves, and stunted or compacted top growth. Toxic effects caused by the relatively large amounts of zinc were not observed.

The soil-amendment benefits provided by relatively heavy applications of raw mycelial waste are twofold: (1) it neutralizes soil acidity and thus serves as a liming material, and (2) it improves some physical conditions of the soil or growing mixtures. The high calcium (8%, dry wt) and organic matter content (40 to 50%, dry wt) appear to improve the aggregation of the soil and as a result help its aeration. Increases in water-holding capacity are rather modest and are mainly in the range of water held at tensions above 100 cm.

In conclusion, raw fermentation wastes can provide adequate nitrogen for crop production; about 100 tons/acre increased yields of silage corn. Salinity and slowness to nitrify may inhibit the use of this waste on salt intolerant species. The zinc content of soils may be increased to levels toxic to plants if these wastes are applied repeatedly at high rates to the same fields. Although bulky and costly to handle, the recent increases in the cost of nitrogen fertilizers may provide an economic incentive for use of such wastes.

ACKNOWLEDGEMENTS

The author thanks his colleagues at the Experiment Station for their willing assistance in many of the studies reported here. Special thanks is due Drs. C. R. Frink and B. L. Sawhney for growing the silage corn at Lockwood Farm and to Dr. G. S. Taylor for assisting with the tobacco harvesting and nematode studies. The Chas Pfizer Co. supplied the various mycelial residues and Mr. Harry Hansen of Landsco, Inc. cooperated in the composting studies.

LITERATURE CITED

1. Ahrens, J.F. 1964. Antidote for herbicides. Activated carbon "blots them up" when unwanted. Frontiers of Plant Science, Fall Issue 1964.
2. Anderson, P.J. 1953. Growing tobacco in Connecticut. Conn. Agr. Exp. Sta. Bull. 564, 110p.
3. Arnold Bik, R. 1970. Nitrogen, salinity, substrates and growth of gloxinia and chrysanthemum. Agr. Res. Repts 739. Centre Agr. Publ. & Doc., Wageningen.
4. Baker, K.F., ed. 1958. The U. C. system of producing healthy container-grown plants. Calif. Agr. Exp. Sta. Manual 23.
5. Bernstein, L. 1964. Salt tolerance of plants. Int. Bull. U.S. Dept. Agr. No. 293. pp. 23.
6. Bernstein, L. 1964. Reducing salt injury to ornamental shrubs in the West. Home & Garden Bull. 95. U.S.D.A., Washington, D.C.
7. Bernstein, L., L.E. Francois, and R.A. Clark. 1972. Salt tolerance of ornamental shrubs and ground covers. J. Amer. Soc. Hort. Sci. 97(4): 550-556.
8. Buckman, H.A. and N.C. Brady. 1969. The nature and properties of soils (7th ed.), The Macmillan Co., New York. 653p.
9. Chapman, H.D., ed. 1966. Diagnostic criteria for plants and soils. Univ. of Calif. Div. Agr. Sci., Riverside, California, U.S.A.
10. DeBoodt, M. and N. DeWaele. 1968. Study on the physical properties of artificial soils and the growth of ornamental plants. Pedologie XVIII, 3:275-300.
11. De Roo, H.C. 1958. Fertilizing Connecticut tobacco. Conn. Agr. Exp. Sta. Bull. 613, 37p.
12. De Roo, H.C. 1967. Artificial root media and fertilizations for container-grown woody ornamental plants. Conn. Agr. Exp. Sta. Bull. Soils 26. 12p.
13. De Roo, H.C. 1972. Artificial root media and fertilizations for container-grown chrysanthemums. Conn. Agr. Exp. Sta. Circ. 245, 11p.
14. Downs, D.B., H.G.M. Jacobson and P.E. Waggoner. 1962. Rotations, organic matter and vegetables. Conn. Agr. Exp. Sta. Circ. 220, 16p.
15. Frink, C.R. 1971. Plant nutrients and water quality. Agr. Sci. Review 9:11-25.
16. Fuller, W.H. 1963. Reactions of nitrogenous fertilizers in calcareous soils. Agr. Food Chem. 11(3):188-193.
17. Jones, J. Benton, Jr. 1972. Plant tissue analyses for micronutrients. In Micronutrients in Agriculture. Ed. J.J. Mortvedt, P.M. Giordano, W.L. Lindsay. Soil Sci. Soc. of America. Madison, Wisconsin, U.S.A.
18. Langdale, G.W., J.R. Thomas, and T.G. Littleton. 1973. Nitrogen metabolism of Stargrass as affected by nitrogen and soil salinity. Agron. J. 65:468-470.
19. Letey, J. and L.H. Stolney. 1964. Measurement of oxygen diffusion rates with platinum microelectrode. Hilgardia 35(20):545-576.
20. Lunt, H.A. 1945. Wood ashes as a fertilizer. Connecticut Woodlands, May 1945.
21. Lunt, H.A. 1955. The use of woodchips and other wood fragments as soil amendments. Conn. Agr. Exp. Sta. Bull. 593, 46p.

22. Lunt, H.A. 1959. Digested sewage sludge for soil improvement. Conn. Agr. Exp. Sta. Bull. 622, 30p.
23. Lunt, H.A. 1961. Improving nursery soil by addition of organic matter. Conn. Agr. Exp. Sta. Circ. 219, 8p.
24. Lunt, H.A., H.G.M. Jacobson, and C.L.W. Swanson. 1950. The Morgan soil testing system. Conn. Agr. Exp. Sta. Bull. 541, 60p.
25. Miller, P.M., G.S. Taylor, and S.E. Wihrheim. 1968. Effects of cellulosic soil amendments and fertilizers on Heterodera tabacum. Plant Dis. Reptr. 52(6):441-445.
26. Miller, P.M., D.C. Sands, and S. Rich. 1973. Effect of industrial mycelial residues, wood fiber wastes, and chitin on plant parasitic nematodes and some soil-borne diseases. Plant Dis. Reptr. 57:438-442.
27. Morgan, M.F. 1940. Peat and swamp muck for soil improvement in Connecticut. Conn. Agr. Exp. Sta. Circ. 142, 12p.
28. Poincelot, R.P. 1972. The biochemistry and methodology of composting. Conn. Agr. Exp. Sta. Bull. 727, 38p.
29. Poincelot, R.P. and P.R. Day. 1973. Rates of cellulose decomposition during the composting of leaves combined with several municipal and industrial wastes and other additives. Compost Sci. 14(3):23-25.
30. Richards, S.J., et al. 1964. Physical properties of soil mixes. Soil Sci. 98:129-132.
31. Rutland, R.B. 1972. Salt induced water stress as a determinant of flower quality and longevity in chrysanthemums. Hort. Science. 7(1):57-59.
32. Shalhevet, J. and B. Yaron. 1973. Effect of soil and water salinity on tomato growth. Plant and Soil 39:285-292.
33. Shearin, A.E., and D.E. Hill. 1962. Soil survey of Hartford County, Connecticut. U.S. Dept. of Agr., Soil Conservation Service, Series 1958, No. 14, 126p., illus.
34. Splinter, W.E., and J.F. Beeman. 1968. The relationship between plant stem diameter and total leaf area for certain plants exhibiting apical dominance. Tobacco Sci. XII:139-143.
35. U.S. Salinity Laboratory Staff. 1954. Diagnosis and improvement of saline and alkali soils. U.S. Dept. Agr., Agr. Handbook 60. 160p.
36. Vesilind, P.A. 1973. A laboratory exercise in composting. Compost Sci. 14(5):14-15.
37. White, John W., and J. W. Mastalerz. 1966. Soil moisture as related to "container capacity". Proc. Amer. Soc. Hort. Sci. 89:758-765.

APPENDIX

Analyses of test well and surface water samples taken at the composting site in Griswold, Connecticut, 1973-1974.

Originally, the composting site consisted of a roughly rectangular plot of land, about 8 acres in size, with its long axis oriented approximately north-south. At both ends small brooks, which ran along the western side of the yard and drained the adjacent composting area, provided an opportunity to obtain surface water samples during most of the year. The area that drained southward was relatively small; most of the site drained northward into the small brook and a swamp beyond. At the low, northwest corner of the compost yard a small dike of soil had been built to contain surface water, since in November 1972 heavy rains had caused some surface runoff in this area. During the year of water sampling, the filling and diking of the site was improved at various times. On both brooks, at the periphery of the composting site, we located a sampling station, identified in Tables 1a, b, and c as BN (Brook North end of yard, beyond dike) and BS (Brook South end of yard).

The wells to intercept subsurface water leaving the site were located as follows: At the north end of the site three wells were located across the slope east of the brook and just north or beyond the small dike. We installed two test wells identified as 1 and 2, while a big concrete well about 12 feet deep was installed by Landsco, Inc. and is identified as BW. Our test wells of perforated aluminum pipe reached a depth of 6 to 7 feet below the soil surface. Test well 1 was placed in the drainage channel or spillway just beyond the dike. Test well 3 was installed on the slope east of the south brook, not far from Station BS. Test well 4 at the north end of the site served as a control. It was placed at the foot of the hillside west of the brook and thus did not receive any surface runoff or groundwater from the compost area.

Besides the identification of the wells, Tables 1a, b, and c show the dates on which samples of ground and brook water were collected and analyzed. The analyses focused on the soluble plant nutrients nitrate and ammonium nitrogen (N), and on sulfate sulfur (S), since the wastes contain gypsum in substantial amounts. For a rapid and economical test we used Morgan's soil testing system (24); if desired, approximate quantitative concentrations can be inferred from the information in Table 2.

The analyses of the groundwater samples taken from the wells show that during the first 6 months of monitoring (2/14 to 8/17/73; Tables 1a and b) no significant movement occurred of nitrate, ammonium or sulfate from the compost yard into the groundwater of the adjacent area. This observation is particularly interesting since our water sampling began about 6 months after the composting operations at the yard had started. Nitrate concentrations in the wells were essentially zero during this period. The ammonium concentrations during these 6 months were also essentially zero, with the exception of well 1, where at least very low (VL) concentrations were consistently found. Water samples drawn from well 1 also contained more sulfate than the other wells. These wells generally showed at least traces of sulfate, as did control well 4.

These results suggest that very little leaching of the mycelial wastes occurred. Although composted and stockpiled for almost a year, and exposed to the percolating action of rain, the losses of soluble nitrogen and sulfate compounds were negligible. Water taken from well 1 probably was contaminated by the earlier spill of mycelial residues prior to construction of the dike, since nearby wells BW and 2, on slightly higher ground, did not show any sign of contamination.

The analyses of the surface water samples taken from both brooks during this first half of the monitoring period also did not show any significant concentration of nitrogen or sulfate. Heavy rains causing runoff apparently did not occur during this time.

This situation, however, changed somewhat during the second part of our water-sampling period. A downpour occurred in mid-August and on 8/17 the water taken from the brooks contained medium (M) concentrations of ammonium and low (L) to medium (M) concentrations of sulfate. In the area of well 1, some spillage of mycelial residues over the earthen dike occurred. As a result concentrations in well 1 remained high during most of the remainder of the sampling period.

During the period from November 1973 through January 1974, however, well 2 and both brooks contained medium (M) to high (H) concentrations of nitrogen and sulfate. This was apparently caused not only by heavy rains but also by increased leveling and grading at the compost site. Bulldozers stirred and moved large stockpiles of mycelial residues and soil mixed with mycelial waste. The site was enlarged to the west of the small brooks, which became filled up to the boundary of the site. In spite of these activities, the water drawn from wells 3 and 4 stayed clean, as did the water from well BW, although soil mixed with mycelial wastes had been graded up to and around this well. A possible explanation is that the groundwater that fed deeper well BW may have been derived from deeper soil layers, that is, from layers beneath a hard, impervious pan. This pan, called a fragipan, is a normal characteristic of the soil type occurring at the site. The shallower test wells 1, 2, 3, and 4 probably sampled water running over this impervious fragipan.

By March 1974, however, concentrations of dissolved constituents were clearly declining and, at the last sampling period in April, most samples contained little dissolved nitrogen, and sulfate was present only in wells 1 and 2.

Thus the year-long sampling of ground and surface water around the periphery of the compost site showed that, contrary to expectation, the losses of nitrogen and sulfate from the site were not large. Mycelial wastes with their high water-holding capacity were not readily leached, and when surface runoff was sufficiently contained by dikes, pollution of adjacent areas was minimal. The subsurface waters that eventually left the intensely worked compost site contained only small amounts of nutrients which presumably were readily absorbed during flow through the swamp below the composting area.

TABLE 1a. Ground and brook-water analyses, mycelium compost yard - Groton.

			Sampling Dates 1973									
			Feb	Mar					Apr			
	Location	Test	14	5	9	16	20	30	5	13	19	27
Well 1	N-end yard, below dike	Nitrate-N	0	0	0	0	0	0	0	0	0	0
		Ammonium-N	0	M	L	VL	VL	VL	VL	L	VL	VL
		Sulfate-S					L	L	L	L	L	L
		pH				5.9		5.7	5.8	6.1	6.0	6.1
BW	N-end yard, below dike	Nitrate-N										0
		Ammonium-N										0
		Sulfate-S										0
		pH										7.2
2	NW-corner yard, below dike	Nitrate-N	L^+	0		0	0	0	0	0	0	0
		Ammonium-N	L	T		0	0	0	0	0	0	0
		Sulfate-S					T	T	T^+	T	T	T
		pH				6.0		5.7	6.0	6.0	5.9	5.9
3	S-end yard	Nitrate-N				0		T	0	0	0	0
		Ammonium-N				0		0	T	0	0	0
		Sulfate-S						T	T	0	0	0
		pH				5.9		6.0	5.7	6.0	5.7	5.4
4	Bottom of hill on W-side yard - Control	Nitrate-N								VL	T	0
		Ammonium-N								0	0	0
		Sulfate-S								T	0	0
		pH								5.9	6.1	6.1
Brook BN	N-end yard, below dike	Nitrate-N	0			0	0	0	0	0	0	0
		Ammonium-N	0			0	0	T	VL^+	T	0	T^+
		Sulfate-S					T	0	T	0	0	0
		pH				6.0		6.2	6.1	6.3	6.1	6.1
BS	S-end yard	Nitrate-N						T	-	0	0	0
		Ammonium-N						0	T	0	VL	T
		Sulfate-S						0	T	0	T	0
		pH						6.1	5.8	6.2	5.8	6.3

Morgan Soil Testing System: 0 = zero; T = trace; V = very; L = low; M = medium; H = high; E = extremely. See Table 2 for quantitative indications of concentrations.

TABLE 1b. Ground and brook-water analyses, mycelium compost yard - Groton.

	Location	Test	Sampling Dates 1973 May 4	May 11	May 25	Jun 8	Jun 22	Jul 6	Jul 20	Aug 3	Aug 17*	Sep 28
Well												
1	N-end yard, below dike	Nitrate-N	0	T	-	**		VL	**		EH	VH
		Ammonium-N	VL	VL	L			L			L	H
		Sulfate-S	L	VL	VL			L			M	VH
		pH	6.3	6.3	6.3			5.9			6.0	6.3
BW	N-end yard, below dike	Nitrate-N	0	0	0	0	0	0	T	0	VL	0
		Ammonium-N	0	0	0	0	0	0	0	0	0	0
		Sulfate-S	0	0	0	0	0	0	0	0	0	0
		pH	7.4	7.8	8.0	8.4	8.0	8.2	8.2	8.0	7.4	6.7
2	NW-corner yard, below dike	Nitrate-N	0	0	0	0	0	0	0	**	L	VL
		Ammonium-N	0	0	0	0	0	0	T		T	0
		Sulfate-S	VL	T	T	T	T	VL	T		L	T
		pH	5.9	6.0	6.0	6.2	6.2	5.9	6.4		6.3	6.2
3	S-end yard	Nitrate-N	0	0	0	0	0	0	0	0	0	0
		Ammonium-N	T	0	T	0	0	0	T	0	T	0
		Sulfate-S	T	0	0	T	0	0	T	T	0	0
		pH	5.8	5.8	5.7	5.7	5.8	5.1	5.7	5.6	5.5	5.3
4	Bottom of hill on W-side yard - Control	Nitrate-N	0	0	0	0	0	0	T	T	VL	VL
		Ammonium-N	0	0	T	0	T	T	T	VL	0	0
		Sulfate-S	T	T	T	VL	T	T	T	T	0	0
		pH	6.1	6.5	6.0	6.6	6.2	6.3	6.3	6.3	6.0	5.8
Brook												
BN	N-end yard, below dike	Nitrate-N	0	0	0	0	0	0	0	**	0	**
		Ammonium-N	0	0	0	0	VL	L	L^+		M	
		Sulfate-S	0	0	0	0	0	T	T		M	
		pH	6.4	6.4	6.2	6.4	6.2	6.2	6.5		5.6	
BS	S-end yard	Nitrate-N	0	0	0	0	0	0	0	0	0	0
		Ammonium-N	0	T	0	0	0	VL	0	T	M^+	M
		Sulfate-S	T	0	0	0	0	T	0	0	L	T
		pH	6.3	6.2	6.1	6.4	6.3	6.4	6.4	6.3	6.1	6.4

Morgan Soil Testing System: 0 = zero; T = trace; V = very; L = low; M = medium; H = high; E = extremely. See Table 2 for quantitative indications of concentrations.

* Sampled 2 to 3 days after heavy rains. Well 1 in spillway, heavily silted up; brooks were running again.

** Wells and brooks dry.

TABLE 1c. Ground and brook-water analyses, mycelium compost yard - Groton.

			Sampling Dates 1973/74									
			Oct		Nov		Dec		Jan	Feb	Mar	Apr
	Location	Test	12	26	9	30	14***	28	25	14	8	3
Well 1	N-end yard, below dike	Nitrate-N	**			EH	VH	O	VH		L	T
		Ammonium-N				VH	VH	H	H		M	M^+
		Sulfate-S				VH	VH	VH	VH		H	H
		pH				6.4	6.8	8.1	6.6		6.8	6.8
BW	N-end yard, below dike	Nitrate-N	O	O	O	O	O	O	O	O	T	L^+
		Ammonium-N	O	O	O	O	O	O	O	O	O	O
		Sulfate-S	O	O	T	O	O	O	T	O	O	O
		pH	7.1	7.3	7.1	6.8	6.7	7.9	6.8	6.5	6.7	6.6
2	NW-corner yard, below dike	Nitrate-N	**		H	VH	VH	VH	H		VH	O
		Ammonium-N			O	O	T	O	O		O	O
		Sulfate-S			L	M	MH	H	H		H	H
		pH			6.0	5.9	6.2	7.6	6.2		6.2	6.1
3	S-end yard	Nitrate-N	O	O	O	O	O	O	O			O
		Ammonium-N	O	O	O	O	O	O	O			O
		Sulfate-S	O	O	O	O	O	O	T			O
		pH	5.9	6.4	5.6	5.6	5.6	5.4	5.2			6.3
4	Bottom of hill on W-side yard - Control	Nitrate-N	T	T	T	O	O	O	T		T	O
		Ammonium-N	O	O	O	O	O	O	O		O	O
		Sulfate-S	T	T	T	O	O	O	T		O	O
		pH	6.6	6.4	6.2	6.0	5.9	5.6	6.1		6.6	6.3
Brook BN	N-end yard, below dike	Nitrate-N				H	H	O	O	O	**	O
		Ammonium-N				M^+	H	M	L^+	L		L
		Sulfate-S				M	H	M	M	T		T
		pH				5.8	6.6	6.9	6.4	6.2		6.8
BS	S-end yard	Nitrate-N	O	O	O	T	VH	O	O	O	O	O
		Ammonium-N	O	O	L	H	VH	MH	L^+	M	L	L
		Sulfate-S	O	O	T	H	VH	M	L	T	VL	O
		pH	6.0	6.2	6.3	6.4	7.0	6.9	6.4	6.2	6.5	6.6

Morgan Soil Testing System: O = zero; T = trace; V = very; L = low; M = medium; H = high; E = extremely. See Table 2 for quantitative indications of concentrations.

** Wells and brooks dry.

*** During a period of heavy rains, runoff along brooks and spillway.

TABLE 2. Calibration of Morgan soil testing system in ppm of dissolved constituents.

Symbol	Index No.	NO_3	NH_4	SO_4
		ppm		
0	0	0	0	0
T	<1	<0.3	<1	<50
VL	1	0.3	1	50
L	2	0.9	3	100
L^+	3	1.4	4	150
M	4	1.8	5	200
M^+	5	2.4	7.5	250
MH	6	3.0	10.0	300
MH^+	7	4.5	15.0	350
H	8	6.0	20.0	400
VH	9	9.0	30.0	500
EH	10	12.0	40.0	600

THE CONNECTICUT
AGRICULTURAL EXPERIMENT STATION
NEW HAVEN, CONNECTICUT 06504

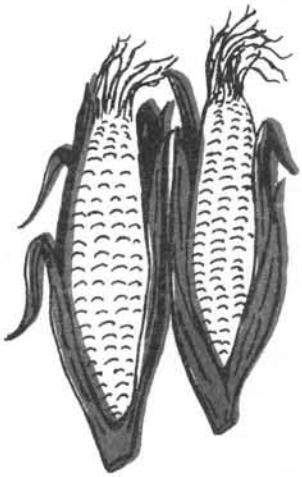
Paul E. Waggoner
Director

POSTAGE AND FEES PAID
U.S. DEPARTMENT OF
AGRICULTURE
AGR 101

U.S.MAIL

PUBLICATION

PENALTY FOR PRIVATE USE, $300



STATE AGRICULTURAL EXPERIMENT STATIONS
Serving 1875 Mankind 1975

The Connecticut Agricultural Experiment Station