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AGRICULTURAL UTILIZATION OF A MYCELIA FILTER CAKE

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AGRICULTURAL UTILIZATION OF A MYCELIA FILTER CAKE

By

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ABSTRACT

AGRICULTURAL UTILIZATION OF A MYCELIA FILTER CAKE

BY

BOBBY JOE HOLDER

A study was conducted at the agricultural experiment farm of Upjohn Pharmaceutical Co., Kalamazoo, Michigan to evaluate agricultural land application of mycelia filter cake derived from lincomycin production. Lincomycin mycelia filter cake was applied to a Kalamazoo sandy loam soil (fine-loamy, mixed, mesic Typic Hapludalf) in the spring of 1980 and 1981 at rates of 0, 7.5, 16.0 and 30.5 dry metric tons/ha. The lincomycin filter cake proved toxic to corn in 1980 so winter wheat was planted to half of the field to check residual toxic effects. A change in the lincomycin production process and a period of 34 days between application of the filter cake and planting of corn resulted in no toxic effects to corn in 1981. The winter wheat showed no residual toxicity from the 1980 application of lincomycin and samples of wheat grain, corn grain and corn leaf tissue, all from the 1981 crops, showed no uptake of lincomycin. In 1981 the yield of wheat was 15.8, 26.5, 31.2, and 41.7 hl/ha for 0, 7.5, 16.0, and 30.5 dry metric tons/ha of filter cake, respectively. The corn yields were 49, 67, 84, and 90 hl/ha for the same rates respectively. The yields for the high rates compared well with commercially fertilized fields of corn and wheat on similar soil types.

Data from suction cup lysimeters and from soil samples showed that NO_3 and soluble-salts were moving downward through the soil profile and that the conversion from NH_4 to NO_3 was slowed down by the lincomycin wastes.

The results from this experiment indicate that application of lincomycin mycelia filter cake on agricultural land at the rate of 16.0 dry metric tons/ha is environmentally safe with regard to NO_3 pollution of groundwater. When added to the soil, this antibiotic waste material is degraded by soil organisms into useful nutrients. These nutrients are readily utilized by crops and the antibiotic is not translocated into the plant.

DEDICATION

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INTRODUCTION

Many investigators have suggested that land application may be a satisfactory ultimate disposal method for antibiotic wastes. Not only has it been found that many of these wastes contain valuable nutrients which could be used by agricutural crops, but land application also alleviates many problems encountered by attempts to dispose of these wastes in sanitary land-fills, or by incineration, composting or ocean dumping. Other benefits include defrayed fertilizer costs, and use of mycelia as a good soil amendment.

Of great importance in considering the application of antibiotic wastes to agricultural land is a management system which will allow maximum crop production without degrading the environment and at the same time applying the maximum possible amounts of these wastes.

The major concerns associated with the use of antibiotic wastes on agricultural land are NO_3 pollution of the groundwater, inhibited crop growth due to the build-up of soluble-salts, and addition of some toxic material to the soil.

Thus, the major objective of this experiment was to develop a management system to utilize mycelia filter cake waste from the lincomycin production process in increasing agricultural production.

Specific objectives were to: 1) evaluate land application of lincomycin mycelia filter cake on agricultural land and to determine the optimum application rates to obtain satisfactory yields of corn and

winter wheat; 2) determine if lincomycin is absorbed by corn or wheatand if it is decomposed in the soil; and 3) determine the maximum application rate of filter cake that may be made without leaching NO_3.

LITERATURE REVIEW

One of the many challenges facing agriculturalists is to develop uses for the enormous quantities of municipal and industrial wastes generated by man (De Roo, 1975). Many of these wastes can be returned to the soil where plant nutrients may be recycled and soil physical properties improved instead of disposing of them in dumps and landfills or by incineration.

Land application has become an attractive ultimate disposal method for sewage sludge and certain types of organic wastes. There are very good reasons to use the soil-plant system for organic waste disposal: a) human pathogenic agents present in the waste materials do not readily survive in the hostile soil environment; b) soils can detoxify hazardous organics by adsorption followed by microbial decomposition; c) the nutrients present in the waste materials are recycled to crops; and d) the soil microbial population has the ability to rapidly degrade organic materials to unoffensive products and to synthesize humus from the intermediates of degradation. Also, land application of organic wastes is far cheaper than alternative treatment methods (Nelson and Sommers, 1979).

The period in which the majority of antibiotics were discovered was from 1945 to 1960 (Bewick, 1977). During this period of time most of the initial investigations on the effects of antibiotics in soils, uptake of antibiotics by plants, and the effects of antibiotics on plant

pathology were carried out. There was little emphasis on the recycling of resources and the possible pollution effects of the fermentation wastes, and therefore, the waste materials were simply regarded as a material to be discarded. Towards the end of this period pharmaceutical companies began to look at the field of animal feedstuffs and some companies began to market the fermentation waste as an animal feed, with additional benefits that the residual antibiotics stimulated growth, increased appetite, increased efficiency of food conversion and reduced vitamin requirements. In the late 1960's there was a change in attitudes toward feedstuff additives and fermentation waste could no longer be sold as feedstuff. More concern was shown for the environment and it became more difficult for the pharmaceutical companies to dump their wastes. Along with this, in the early 1970's there was a great increase in the price of raw materials, including fertilizers, and closer examination has had to be made of the limits of our natural resources.

The application of industrial wastes and sewage sludges on agricultural soils is today receiving greater and greater emphasis because of the increasing energy requirements for maximum crop yields and costs associated with the alternative disposal methods.

The amounts of these wastes being produced has increased each year as our society became more technologically oriented and as the demand for current antibiotics increased and new antibiotics were brought into production. Also, the increased costs of N fertilizers have provided economic incentives for the use of industrial and municipal wastes on agricultural land.

These wastes contain considerable N, as well as other nutrients

which are valuable to the agricultural industry. Recent reports (Sommers et al., 1979; Nelson and Sommers, 1979; De Roo, 1975; Wright, 1975 and 1978) have indicated active research programs aimed directly at the use of certain industrial wastes such as mycelia for agriculture.

Many studies have shown that pure antibiotics fed to swine, ruminants, turkeys and chicks stimulate growth (Bewick, 1977). Optimum levels range between 2 and 50 ppm of the antibiotics. The stimulated growth, increased appetite, and increased efficiency of food conversion appeared to be due to the action of the antibiotics on the microflora of the animals as germ-free animals showed no such response (Bewick, 1977; Hungate et al., 1955; Klopfenstein et al., 1964).

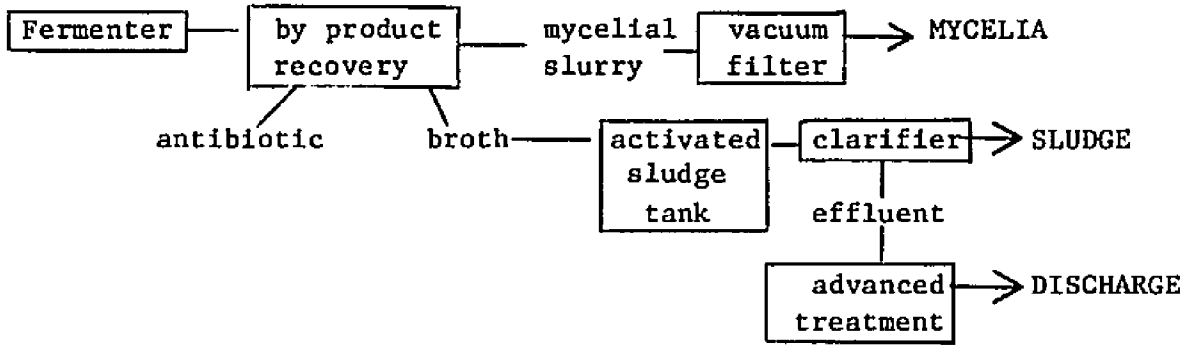
Antibiotics such as chlortetracycline and oxytetracycline are routinely fed to livestock and poultry to increase weight gain and feed efficiency and to maintain animal health (Addis et al., 1976, Brown et al., 1975; Gilliam and Martin, 1975). Antibiotics ingested in feed have been shown to be excreted in significant quantities in feces and urine (Elmond et al., 1971; Morrison et al., 1969; Tietjen, 1975; Warman et al., 1977; Webb and Fontenot, 1975). Morrison et al., (1969) and Elmond et al., (1971) reported that 75% of the antibiotic chlortetracycline fed to cattle was recovered from manure and that the excreted antibiotic had a half-life of less than 20 days.

Two types of organic wastes are by-products of antibiotic production (Nelson and Sommers, 1979):

1) mycelia mat (microbial cells and filter aid) from filtrates of solids from the broth used as a growth media for antibiotic-producing organisms.
2) the sludge from aerobic digestion (activated treatment) of

the fermentation broth after separation of the product.

This is shown diagramatically as follows:



Barker et al. (1958) examined the possibilities of speeding up the breakdown of antibiotic wastes using compressed air in a sewage tank and achieved 80-90% reduction in B.O.D. Aeration of lagoons with pure oxygen was also found to produce 90% reduction in B.O.D. (Lederman et al., 1975). Hilgart (1950), Hurwitz et al. (1952), Cushman and Hayes (1956), and Tompkins (1957) developed effective methods for treating antibiotic waste produced by the Upjohn Company, Michigan, U.S.A., and B.O.D., reduction of 97% was achieved. Several other pharmaceutical companies also have biological treatment plants to reduce the B.O.D. of fermentation wastes (Reimers et al. (1954); Edmondson, 1954; Liontas, 1954; Howe and Paradisio, 1956; Horne and Rinaca, 1960).

The production of organic wastes, both municipal and industrial, has reached prodigious quantities in recent years. Many past practices of disposal such as burning, burial, ocean dumping, and stream discharging have been banned or severely restricted. Even the more acceptable methods of disposal, such as incineration and landfill, are under attack because of the impact on the environment.

These disposal problems, along with the concept that many of the materials are valuable as sources of plant nutrients or as soil

conditioners, have resulted in an increase in land applications of organic wastes.

Several studies have concluded that moderate application of sewage sludges and other wastes can increase crop production (Cunningham et al., 1975; Dolar et al., 1972; Hinesly et al., 1972; King and Morris, 1972; Sabey et al., 1975). Most of these investigations have related increased yields to increased levels of N from the organic wastes.

In terms of the behavior of antibiotics in soils, they can be classified into three groups.

1) Basic antibiotics (i.e. streptomycin, neomycin and erythromycin) are adsorbed rapidly by clay particles and organic matter.

2) Acidic antibiotics, such as penicillin and chloramphenical, although not adsorbed, are inactivated rapidly by micro-organisms in the soil.

3) Amphoteric antibiotics (i.e. aureomycin, terramycin and bacitracin) can act either as acids or bases, but as the pH of the soil is usually below the isoelectric point of the antibiotic they behave as bases and are adsorbed onto clay minerals.

The adsorption of antibiotics by soil has been shown by many workers (Waksman and Woodruff, 1942; Pramer and Starkey, 1951; Gregory et al., 1952; Hessayon, 1953) and studied extensively be Siminoff and Gottlieb (1951), Gottlieb, Siminoff and Martin (1952), Gottlieb and Siminoff (1952), Jefferys (1952), Martin and Gottlieb (1952, 1955), Pinck, Holton and Allison (1961), Pinck and Allison (1961, 1962), Pramer and Starky (1962), Soulides (1969), and Ghosal and Mukherji (1970). Basic antibiotics eg. streptomycin, dihydrostreptomycin, neomycin, kanamycin,

and erythromycin are adsorbed by the clay minerals smectite (montmorillonite), hydrous mica (illite and bentonite), kaolinite and vermiculite, although different clays adsorb different amounts of the same antibiotic (Pinck et al., 1962). Adsorption of antibiotics is not always permanent and some antibiotics can be released from the clay complexes by phosphate and citrate buffers. This release is more prevalent with amphoteric than with basic antibiotics (Soulides et al., 1961, Pinck, Soulides and Allison (1961). Ghosal and Mukherji (1970) found that different cations can produce different rates of release of streptomycin from clays. In the case of streptomycin, Siminoff and Gottlieb (1951) and Jefferys (1952) found it to be inactive when adsorbed on soil and montmorillonite; whereas, Pramer and Starkey (1951, 1953) reported it to be bactericidal. Jefferys (1952) and Pramer and Starkey (1951) reported that it undergoes microbial decomposition.

Bewick (1977) found that the levels of tylosin released by phosphate buffers from clays was greatest in kaolinite and illite (25 and 30 per cent, respectively). Bentonite and montmorillonite clays, with a higher cation-exchange capacity, showed tylosin release of 7.6 and 7.4 per cent, respectively. He also, points out that soil temperature had little effect on the amount of tylosin released. At all temperatures higher concentrations of fermentation waste released larger amounts of tylosin.

Many workers have shown that illite and kaolinite have a low adsorption capacity for most of the basic antibiotics (Ghosal and Mukherji, 1970; Pinck, Holton, and Allison, 1961; Pinck, Soulides and Allison, 1961; Siminoff and Gottlieb, 1951). This is because of the non-expanding lattice in these clays with resulting restriction of

cation-exchange to the outer surfaces of the clay particles. Marshall (1964) has shown that cation-exchange is higher in illite because of the presence of hydrated layers interlaced with the mica units of the clay.

Before the residue from any antibiotic fermentation could be used as a fertilizer it would have to be determined whether or not low levels of the antibiotic that may be present would be taken up by plants and what the effects on growth, if any, would be. Numerous studies have been conducted on the uptake of antibiotics by plants. This subject was reviewed by Brian (1957) and Goodman (1962). These studies have been concerned mainly with the uptake of streptomycin and its effect on plant growth (Pramer, 1953; Rosen, 1954; Gray, 1955; Dye, 1956; Bajaj and Durbin, 1961; Drury and Khan, 1969; Ellis and Sinclair, 1974; Mukherji and Bag, 1974; Mukherji, Bag and Paul, 1975). Other antibiotics have also been investigated: griseofulvin (Brian et al., 1951; Crowdy et al., 1955, 1956; Srivastra, 1966), chloramphenical (Pramer, 1953, Crowdy et al., 1955), and aureomycin, neomycin and terramycin (Pramer, 1953). Streptomycin, at concentrations greater than 100 ug/ml was found to inhibit root and shoot development in many plants although this effect could be reversed by additions of Mn (Rosen, 1954; Gray, 1955). Griseofulvin and chloramphenical were also found to be taken up by plants, while aureomycin, neomycin and terramycin did not appear to be taken up by cucumber seedlings (Pramer, 1953). There have been no reports in the literature to date concerning the uptake of low levels of antibiotics contained in fermentation waste.

Attempts to set guidelines on rates of application of organic wastes have frequently been based on the N content of the wastes and the N demand of the agronomic crop (Powers et al., 1975). Studies have shown

that applications of organic wastes to agricultural soils in large quantities can cause groundwater pollution since NO_3 can be readily leached through the soil (Chang et al., 1973; King and Morris, 1972). Therefore, rates must be carefully assessed to obtain the desired crop benefits from added N and yet maintain a quality environment. It is generally recommended that the amount of plant available N supplied by organic waste be limited to less than twice the N requirement of the crop grown. Thus, the mineralization rate of the added waste must be known in order to calculate proper levels of land application.

In the work done by Wright (1978) it was noted that the N mineralization rate on mycelia filter waste was very fast. In his laboratory experiment he noted that as much as 95% of the organic N added was mineralized within 24 weeks of incubation. However, he also stated that the mineralization rate of the fermentation wastes was not as rapid in his field trials with the highest levels of inorganic N occurring within 8 weeks of application.

MATERIALS AND METHODS

Field Studies

Mycelia filter cake was applied to a Kalamazoo sandy loam (fine-loamy, mixed, mesic Typic Hapludalf) by use of a flail spreader at rates of 0, 7.5, 16.0 and 30.5 dry metric tons/ha and immediately disked to a depth of 25 cm. The study was originally designed to be a two year study comparing three rates of application of mycelia filter cake either annually or applied once. The eight 15.2 by 30.4 meter plots were replicated four times and the experimental design was a randomized block design. The applications were designed as follows:

Treatment 1 - no filter cake applied 1980 or 1981

Treatment 2 - no filter cake applied 1980 or 1981

Treatment 3 - 7.5 dry metric tons/ha applied 1980 and 1981

Treatment 4 - 7.5 dry metric tons/ha applied 1980 and 1981

Treatment 5 - 16.0 dry metric tons/ha applied 1980 and 1981

Treatment 6 - 16.0 dry metric tons/ha applied 1980 and 1981

Treatment 7 - 30.5 dry metric tons/ha applied 1980 and 1981

The rates of mycelia filter cake used were based on the nitrogen content of the filter cake assuming that 80 percent of this nitrogen would be available during the growing season. Table 1 gives the actual quantities of mycelia cake applied in the field.

Soil samples were obtained prior to the application of mycelia filter cake by collecting 20 cores from each plot to a depth of 90 cm. Each core was divided by depth into the following increments: 0-15,

15-30, 30-45, 45-60, 60-90 cm. Each four weeks during the growing season soil samples were collected to a depth of 45 cm (0-15, 15-30, 30-45 cm). After the corn was harvested, soil samples were again collected to a depth of 90 cm. The analyses performed are shown in table 2.

Table 1. Rates of application of mycelia filter cake in 1980.

Treatment	Wet Weight Applied		Dry Weight Applied		Nitrogen Applied	
	tons/acre	metric tons/ha	tons/acre	metric tons/ha	lbs/acre	kg/ha
Control	none		none		none	
Low	9.06	20.4	3.35	7.5	114	128
Medium	19.20	43.0	7.13	16.0	242	272
High	33.70	75.0	13.58	30.5	462	518

Corn was planted on all plots in 1980 immediately following the application of mycelia filter cake. Since the material proved toxic to corn, the original experiment was modified in the following way: after harvest of the corn at the end of the growing season all plots were planted to winter wheat. In 1981 the western half of the total acreage was left to winter wheat in order that residual effect of the mycelia filter cake might be estimated while the eastern half was disked and the same rates of mycelia filter cake applied as in 1980 giving four replications of control, low, medium and high applications of mycelia filter cake. In 1981 corn was not planted until 34 days following the application of the filter cake.

Six plots were randomly selected in which porous cup lysimeters were installed so that contamination of groundwater, if any, might be

Table 2. Analysis performed on soil samples.

Parameter	Sampling period		
	1st	Each 4th week*	Last
Kjeldahl N	X		X
NH_4	X	X	X
NO_3	X	X	X
Avail. P	X		X
Exchangeable Ca	X		X
" Mg	X		X
" K	X		X
DTPA Extractable Zn	X		X
" " Cu	X		X
" " Mn	X		X
" " Fe	X		X
" " Ni	X		X
" " Cd	X		X
pH	X		X
Electrical Conductivity	X	X	X

*Samples were collected each fourth week during the growing season.

detected. The porous cup lysimeters were buried to a depth of 152 cm(60.8 inches) and sampled monthly during the growing season.

Yields of corn grain and wheat grain were estimated by hand harvesting two rows of corn and wheat, each 25 feet in length, from each plot. The corn was weighed and corrected for moisture to obtain an estimate of the final yield. The wheat was thrashed in a head thrasher and the grain was weighed for yield estimate.

Samples of the corn tissue were obtained during the growing season by collecting the ear leaf from ten plants per plot at the time the corn tassels and samples of the grain were collected at the time of harvest.

Tissue samples of corn were analyzed for total N, P, Ca, Mg, and K. Samples were also sent to Upjohn laboratories and analyzed for lincomycin. Samples of wheat grain were also analyzed for lincomycin.

Laboratory Methods

Kjeldahl N - Standard methods were used which do not include nitrate nitrogen.

Ammonium - Steam distillation.

Nitrate - Technicon standard method. Reduction of nitrate to nitrite by Cd-Cu reduction column and subsequent colorimetric determination of nitrate. The nitrite present in the sample is subtracted.

Available P - Bray P1. Soil was extracted with 0.025 N HCl + 0.03 N NH_4F with a soil to solution ratio of 1:10.

Exchangeable ions - Soil was extracted with 1 N NH_4OAc at pH 7.0 and Ca and Mg measured by atomic absorption spectrophotometry and K by flame photometry.

DTPA Extractable metals - Soil was extracted with DTPA by the method of Lindsay and Norvell (1978). All metals are determined in the extract by plasma emission spectroscopy.

Soil Reaction- The pH of a 1:1 soil to distilled water mixture was measured with an Orion 801 pH meter with a glass electrode.

Electrical Conductivity - The conductance of a 1:2 soil to distilled water suspension was measured and the values corrected to 30 percent moisture.

RESULTS AND DISCUSSION

In 1980, lincomycin filter cake proved toxic to corn at all levels of application. The corn damage is reflected in the reduced yield at the two higher application rates (Table 3). The control plots yielded 51 hectoliters per hectare and the maximum yield was at the low application rate which yielded only 58 hectoliters per hectare. The low treatment showed some damage to the corn from applied mycelia filter cake. The high and medium treatment plots suffered more severe damage with the high treatment yielding only 20 hectoliters per hectare.

Table 3. Effect of lincomycin mycelia filter cake on growth and yield of corn in 1980.

Treatment	Estimated Stand	Yield*	
	%	bushels/acre	hl/ha
Control	80	58	51
Low**	55#	67#	58#
Medium**	45	61	53
High**	15	23	20

*Yield is corrected to 15.5% moisture on a wet weight basis.

**Low is 7.5 dry metric tons/ha, Medium is 16.0 dry metic tons/ha, and High is 30.5 dry metric tons/ha applied immediately before planting.

#Mean of 6 replications; other values are means of 8 replications.

Observation of the corn damage were made 29 days after seeding (Table 3). The root system of the corn plants showed little damage indicating the probability that a phytotoxic material had been absorbed through the roots and translocated into the plant. Rosen, (1954) and Gray, (1955) found that streptomycin at concentrations greater than 100 ug/ml inhibited root and shoot development which was later reversed by additions of Mn. However, streptomycin is a pure antibiotic and should be more likely to be absorbed into the plant than is an antibiotic waste which usually only shows traces of the pure antibiotic.

In light of the toxic effects on corn in 1980, two very important questions arise: 1) can the lincomycin mycelia filter cake be managed to be non-toxic to an agricultural crop, and 2) will the mycelia filter cake have residual toxic effects?

Management of N in waste material is of critical importance in both agricultural production and environmental quality. The 1980 data suggest that application of a mycelia filter cake waste immediately before planting is too late from the N management standpoint, in addition to the toxic effects to the corn. The corn had relatively little N available to it during the first four to eight weeks after application of the mycelia filter cake waste (Table 8). Corn requires the largest amount of N during the month of July, during which time the N should be well distributed throughout the root zone. Our data show greater movement of N to the lower depths occurred in the medium and high rates of mycelia filter cake during the months of July and August. The increased downward movement of NO_3 through the soil profile was probably due to the sparse stand of corn which removed less NO_3 from the soil in the higher treatment plots. Weed growth in late August and

September then removed large quantities of NO_3 from the soil.

The winter wheat is considered a residual study. However, as already noted, the crop was not removed from the field at the end of the growing season, but was disked back into the soil. There would probably be less residual N carry-over from a field that had a good corn crop removed.

The yield of corn and wheat in 1981 are comparable to well fertilized crops on similar type soils (Tables 4 and 5).

Table 4. Yield of wheat from residual mycelia filter cake.

Treatment	Weight Applied	Yield of Wheat	
	metric tons/ha	Bu/Acre	hl/ha
Control	none	18.2	15.8
Low	7.5	30.4	26.5
Medium	16.0	35.8	31.2
High	30.5	47.4	41.2

*Dry weigh of Lincomycin mycelia filter cake applied in 1980 yields are for the 1981 season.

Table 5. Yield of corn as affected by rate of mycelia filter cake applied in 1981

Treatment	Yield of Corn	
	bu/acre*	hl/ha
Control	56	49
Low	77	67
Medium	96	84
High	103	90

*Corrected to 15.5% moisture on a wet weight basis.

The corn for 1981 showed no damage from the lincomycin filter cake. There are two probable reasons for this: 1) in 1981, the corn was seeded 34 days after the mycelia filter cake was applied to the soil; and 2) a change in processing removed more of the lincomycin from the filter cake used in 1981 compared to the 1980 processing.

The corn in both 1980 and 1981 showed some N deficiency (Table 6). In 1980, these symptoms were readily observable in all plots; whereas, in 1981 they were seen in the control and low treatment plots only.

Table 6. Total nitrogen in ear leaf of corn as affected by application of mycelia filter cake.

Date	Treatment			
	Control	Low	Medium	High
	ppm*			
1980	20,800	25,900	30,000	30,300
1981	20,600	22,000	27,100	28,000

*Each value is an average of 8 replications.

The data from the wheat yield (Table 4) indicates that there is no residual toxic effect from the lincomycin mycelia filter cake applied in 1980.

The samples of wheat grain, corn grain and corn leaf tissue, all from the 1981 crops, were at or below detectable limits for lincomycin.

Table 7 shows the total P, Ca, Mg, and K contents of the corn leaf tissue for 1980 and 1981.

Phosphorus in the leaf tissue increased as the rate of mycelia filter cake application increased, with the highest P content in the medium rate of filter cake for both 1980 and 1981.

Calcium in the leaf tissue increased greatly with the increase of

filter cake. Magnesium showed slight increase, with the greatest increase at the medium rate in 1980, and the greatest increase for 1981 is seen at the low rates of filter cake. The only explanation offered for this is that the mycelia filter cake has extremely high gypsum content which adds greatly to the Ca in the soil at higher application rates. It may also be that the Mg did not increase much because of competition with Ca on the exchange site of the soil particles.

Table 7. Nutrient content of corn ear leaf as affected by mycelia filter cake application.

Year	Treatment	Total P	Total Ca	Total Mg	Total K
		ppm			
1980	Control	3,858	4,626	2,157	21,200
1980	Low	4,066	5,213	2,063	19,000
1980	Medium	4,418	5,970	2,383	19,600
1980	High	4,258	6,218	2,262	18,800
1981	Control	4,069	4,318	2,008	20,180
1981	Low	4,129	5,353	2,318	18,920
1981	Medium	4,617	5,862	2,188	18,600
1981	High	4,452	6,087	2,186	18,600

The K concentrations in the leaf tissue showed a general overall decrease for both 1980 and 1981, with the highest K levels in the control plots for both years. A likely reason for this is, as with Mg, that the K ion may be in competition with the Ca ion for the exchange sites leading to some loss of K.

Relatively little increase in NO_3 was seen in the soil one month after application of mycelia filter cake in either 1980 or 1981 (Tables

Table 8. Nitrate content of soil as affected by the application of lincomycin mycelia filter cake in 1980.

Treatment	Depth	Date					
		5/80	6/80	7/80	8/80	9/80	10/80
	cm	ppm N as NO_3*					
Control	0-15	4.58	4.97	11.1	2.68	1.76	1.62
Low**	0-15	3.05	5.74	11.9	4.51	3.73	2.83
Medium**	0-15	3.53	7.42	19.7	4.60	4.61	3.95
High**	0-15	2.89	6.97	25.3	11.2	6.88	3.07
Control	15-30	3.07	4.63	6.07	1.74	1.53	1.95
Low	15-30	2.42	5.10	7.66	4.58	2.24	3.38
Medium	15-30	2.45	6.24	9.34	5.00	5.20	3.48
High	15-30	2.34	6.00	13.9	14.4	5.33	8.03
Control	30-45	2.34	3.13	4.67	1.34	1.42	1.44
Low	30-45	1.72	3.00	5.07	3.36	2.06	2.51
Medium	30-45	1.81	3.77	6.26	4.32	4.60	3.07
High	30-45	1.74	3.98	7.21	14.0	6.59	7.09

*Each value reported is a mean of eight replications.

**Low is 7.5 dry metric tons/ha, Medium is 16.0 dry metric tons/ha, and High is 30.5 dry metric tons/ha.

Table 9. Ammonium content of soil as affected by the application of lincomycin mycelia filter cake in 1980.

Treatment	Depth	Date					
		5/80	6/80	7/80	8/80	9/80	10/80
	cm	ppm N as NH_4*					
Control	0-15	0.58	0.63	0.99	0.87	0.17	0.65
Low**	0-15	0.39	0.70	0.81	1.11	0.15	0.30
Medium**	0-15	0.39	0.98	0.71	0.96	0.14	0.30
High**	0-15	0.50	1.13	0.73	0.96	0.14	0.58
Control	15-30	0.36	0.57	0.98	0.82	0.19	0.36
Low	15-30	0.09	0.49	0.64	0.83	0.18	0.34
Medium	15-30	0.23	0.58	0.79	0.70	0.18	0.27
High	15-30	0.34	0.86	0.67	0.78	0.17	0.35
Control	30-45	0.28	0.46	0.78	0.67	0.29	0.25
Low	30-45	0.33	0.45	0.08	0.63	0.22	0.23
Medium	30-45	0.49	0.42	0.83	0.65	0.25	0.26
High	30-45	0.31	0.52	0.55	0.65	0.49	1.76

*Each value reported is a mean of eight replications.

**Low is 7.5 dry metric tons/ha, Medium is 16.0 dry metric tons/ha, and High is 30.5 dry metric tons/ha.

Table 10. Nitrate content of soil as affected by the application of lincomycin mycelia filter cake 1981.

Treatment	Depth	Date					
		3/81	5/81	6/81	7/81	8/81	11/81
	cm	ppm N as NO_3*					
Control	0-15	0.99	2.47	6.58	4.81	2.15	1.18
Low**	0-15	1.18	4.29	10.50	5.38	3.00	1.45
Medium**	0-15	1.54	7.63	15.90	4.29	1.89	2.54
High**	0-15	2.06	6.99	10.70	5.30	2.51	6.51
Control	15-30	1.03	1.90	5.68	4.67	1.10	1.36
Low	15-30	1.21	2.55	8.20	4.17	2.98	1.71
Medium	15-30	1.25	3.13	12.30	2.72	1.00	2.50
High	15-30	1.46	4.88	9.17	3.72	2.17	5.82
Control	30-45	1.00	1.66	4.85	3.57	1.02	1.39
Low	30-45	1.25	1.66	5.09	3.46	1.19	1.62
Medium	30-45	1.25	2.37	6.70	2.29	1.98	2.25
High	30-45	2.39	3.08	6.51	3.18	4.09	5.19

*Each value reported is a mean of four replications.

**Low is 7.5 dry metric tons/ha, Medium is 16.0 dry metric tons/ha, and High is 30.5 dry metric tons/ha in 1980 and again in 1981.

Table 11. Ammonium content of soil as affected by the application of lincomycin mycelia filter cake in 1981.

Treatment	Depth	Date					
		3/81	5/81	6/81	7/81	8/81	11/81
	cm	ppm N as NH_4*					
Control	0-15	1.07	0.76	0.81	0.79	0.67	0.38
Low**	0-15	0.92	0.39	0.59	0.90	1.19	0.37
Medium**	0-15	0.91	0.49	0.74	1.10	1.21	0.39
High**	0-15	0.97	0.36	0.61	0.88	0.99	0.34
Control	15-30	0.80	0.46	0.68	0.74	0.74	0.19
Low	15-30	1.06	0.34	0.46	0.85	0.96	0.26
Medium	15-30	0.82	0.35	0.35	0.73	0.62	0.25
High	15-30	0.83	0.25	0.42	0.80	0.78	0.36
Control	30-45	0.77	0.17	0.18	1.31	1.20	0.16
Low	30-45	0.82	0.23	0.35	0.63	0.72	0.23
Medium	30-45	0.62	0.34	0.32	0.52	0.49	0.25
High	30-45	0.73	0.16	0.28	0.47	0.62	0.62

*Each value reported is a mean of four replications.

**Low is 7.5 dry metric tons/ha, Medium is 16.0 dry metric tons/ha, and High is 30.5 dry metric tons/ha applied in 1980 and again in 1981.

8 and 9). However, in both years, by the end of the second month levels of NO_3 in the soil were considerably higher as the result of applications of filter cake. In 1980, the higher levels in the surface soil persisted through the September sampling. In 1981, these higher levels of NO_3 were seen through the November sampling. The data in Table 10 indicate that the earlier application of mycelia filter cake and the delay of 34 days before planting in 1981 resulted in the greatest amount of available N in June rather than in July when the corn plant most needs it. There was also considerably less available N in the soil in the early growing season in 1981 as compared to 1980.

There is some indication that the lincomycin waste slows the conversion from NH_4 to NO_3 during the early stages of breakdown.

The principal forms of N in mycelia wastes are organic, insoluble but slowly available, and readily available NO_3 and NH_4. When the mycelia filter cake is disked into the soil, NO_3 is immobilized very rapidly for about two weeks, depending on the temperature, after which time the NO_3 would be expected to accumulate readily as a result of increasing nitrifying activity in the soil. Our data, however, show this nitrifying activity occurring at a much slower rate (Tables 9 and 11).

There appears to be considerable increase in the NH_4 content in the surface soil one month after application of mycelia filter cake in 1980 (Table 9). The increase in NH_4 was much slower in 1981 (Table 11) showing a marked decrease in NH_4 from the March sampling to the May sampling. As the weather became warmer there was an increase in NH_4 content in the soil and a more rapid conversion from NH_4 to NO_3 occurred (Tables 9 and 11).

A major objective of this study was to follow the movement of N and soluble-salts through the soil profile. To help obtain data for this movement, suction cup lysimeters were installed at a depth of 152 cm. These suction cup lysimeters produced samples only intermittently because the soils at this depth were usually below field capacity and, thus, rarely was there sufficient liquid available to sample. It was impossible to obtain any samples from lysimeters during much of the summer when we experienced drought periods. Although the data that were collected are neither complete nor conclusive and some dates and samples are missing, these data are included to illustrate some of the trends which occurred with the application of the mycelia filter cake.

Samples that were collected correlated well with the NO_3 and soluble-salt content of the soil samples obtained from corresponding plots except for the first sample obtained after installation of the lysimeters (Tables 12 and 13). This sampling is assumed to have shown contamination from the installation of the lysimeters. Higher NO_3 levels are found in the soil-water at 152 cm on the plots which received the high rate of mycelia filter cake. It is also evident that the soluble-salts are being translocated downward through the soil profile (Table 12 and 13). Although neither the NO_3 levels nor the soluble-salts concentration appear to be high enough to cause crop damage or to add much contamination to the groundwater, additional time and a more complete sampling program are needed to make more positive assumptions. The time required would depend on the application rates and the type of soil; sandier soils being leached at a more rapid rate.

The application of mycelia filter cake increased the soluble-salts concentration in the soil in 1980 and 1981, but in neither year did these levels become sufficiently high to cause possible damage to corn or wheat (Tables 14 and 15). DeRoo, (1975) points out that one of the limiting factors in the use of mycelia wastes as a N fertilizer appears to be its total soluble-salts concentration. The soluble-salts in the mycelia filter cake are caused by the calcium sulfate in the wastes. The salts not only suppress nitrification in the soil and possible the uptake of N by the roots, but it also appears to disrupt the normal metabolic pathways within the plant. As a result, crop response to mycelia waste may be largely controlled by the salt tolerance of the plants. Both wheat and corn appear to be relatively salt tolerant, thus, having no difficulty coping with the levels of soluble-salts encountered in this experiment.

It appears that the application of mycelia filter cake had little effect on the concentrations of DTPA extractable metals in the soil (Tables A-1 to A-14). Iron, Mg, Pb and Zn were found in highest concentrations, as would be expected, but were not related to treatment and not found at any time of the year to be near toxic levels. Nickel, Cu and Cd were found in small amounts causing no problems to corn or wheat.

Table 12. Nitrate and soluble salt content of soil water extracted by suction lysimeters in 1980.*

Treatment	Date		
	7/80	8/80	9/80
	ppm N as NO_3**		
Control	79	7.8	8.4
High	137	13.1	14.5
	E.C. mmhos/cm		
Control	0.68	0.66	0.28
High	0.74	0.88	0.62

*The porous cups were at the five foot depth.

**Control plots are an average of two replications and the High Treatment is an average of three replications.

Table 13. Nitrate and soluble salt content of soil water extracted by suction lysimeters in 1981.*

Treatment	Date		
	6/81	7/81	8/81
	ppm N as NO_3**		
Control	13.4	12.1	6.4
High	--	9.7	12.1
	E.C mmhos/cm		
Control	0.48	0.62	0.41
High	0.51	0.58	0.76

*The porous cups were at the five foot depth.

**Control plots are an average of two replications and the High Treatment is an average of three replications.

The available P in the soil showed a slight decrease with the increased application rate of mycelia filter cake in 1980 and 1981. In most cases, the highest available P was in the control plots of this experiment. It is possible that high applications of Ca contained in the filter cake have reduced the extractable P.

Table 14. Specific conductance of soil as affected by the application of lincomycin mycelia filter cake in 1980.

Treatment	Depth	Date				
		5/80	6/80	7/80	8/80	9/80
	cm	mmhos/cm*				
Control	0-15	0.60	0.74	0.86	1.13	0.65
Low**	0-15	0.52	0.83	0.91	1.01	0.63
Medium**	0-15	0.51	0.97	1.29	1.19	0.75
High**	0-15	0.52	1.20	1.46	1.17	0.87
Control	15-30	0.50	0.63	0.62	0.75	0.55
Low	15-30	0.46	0.70	0.72	0.83	0.57
Medium	15-30	0.43	0.79	0.88	0.94	0.64
High	15-30	0.48	1.00	0.98	0.79	0.73
Control	30-45	0.40	0.52	0.63	0.56	0.49
Low	30-45	0.40	0.53	0.59	0.67	0.50
Medium	30-45	0.38	0.58	0.63	0.70	0.55
High	30-45	0.42	0.63	0.66	0.76	0.62

*Each value reported is a mean of eight replications.

**Low is 7.5 dry metric tons/ha, Medium is 16.0 dry metric tons/ha, and High is 30.5 dry metric tons/ha.

Table 15. Specific conductance of soil as affected by the application of lincomycin mycelia filter cake in 1981.

Treatment	Depth	Date					
		3/81	5/81	6/81	7/81	8/81	11/81
	cm	mmhos/cm*					
Control	0-15	0.59	0.92	0.82	0.73	0.66	0.62
Low**	0-15	0.47	0.89	1.05	0.96	0.90	0.76
Medium**	0-15	0.52	0.78	1.05	1.44	1.10	1.00
High**	0-15	0.56	1.03	1.64	1.34	0.80	0.62
Control	15-30	0.54	0.85	0.70	0.68	0.72	0.97
Low	15-30	0.47	0.85	0.94	0.83	0.66	0.67
Medium	15-30	0.45	0.69	0.94	1.07	0.99	0.95
High	15-30	0.49	0.91	1.30	1.09	0.82	0.71
Control	30-45	0.49	0.68	0.53	0.60	0.72	0.90
Low	30-45	0.46	0.58	0.75	0.67	0.62	0.63
Medium	30-45	0.44	0.68	0.84	0.84	0.86	0.96
High	30-45	0.48	0.83	0.91	0.70	0.71	0.67

*Each value reported is a mean of four replications.

**Low is 7.5 dry metric tons/ha, Medium is 16.0 dry metric tons/ha, and High is 30.5 dry metric tons/ha.

SUMMARY AND CONCLUSIONS

Mycelia filter cake from the lincomycin production process was applied to a Kalamazoo sandy loam soil in 1980 and 1981 at rates of 0, 7.5, 16.0 and 30.5 dry metric tons/ha and immediately disked into the soil. In 1980 corn was planted immediately after application of filter cake and the material proved toxic to corn at all rates of application. Thus, winter wheat was planted on one half of the study area as a residual study to determine if the filter cake had residual toxic effects.

In 1981, filter cake was again applied to the other half of the study area at the same rates as in 1980, but the corn was not planted until 34 days after the application of filter cake.

Soil samples were taken prior to application of mycelia filter cake to a depth of 90 cm. Each four weeks during the growing season soil samples were taken to a depth of 45 cm and after corn was harvested soil samples were again collected to a depth of 90 cm. In addition, suction cup lysimeters were installed in control plots and high treatment plots to a depth of 152 cm to study movement of nitrates and salts out of the rooting zone.

In 1980, lincomycin filter cake proved toxic to corn at all rates of application. This damage was reflected in reduced yields of corn at the two higher application rates. In 1981, a change in the lincomycin

production process and the period of 34 days between application of filter cake and planting of corn resulted in no toxic effects to corn. Also, winter wheat showed no residual toxic effects from the 1980 application of mycelia filter cake.

From the yield and soil NO_3 data it is concluded that an application of 16.0 dry metric tons/ha of mycelia filter cake is environmentally safe and gives satisfactory yields. It is also concluded that a period of at least 30 days between application time and planting of corn is necessary to prevent toxicity from lincomycin filter cake.

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APPENDIX

Table A-1. DTPA extractable zinc from soils receiving filter cake.

Date	Depth	Treatment			
		Control	Low	Medium	High
	cm	mg/kg soil			
5/80	0 -15	1.54	1.66	0.70	1.80
	15 -30	0.64	0.57	0.90	0.85
	30 -45	0.61	0.45	0.27	0.65
	45 -60	1.33	0.32	0.33	0.29
	60 -90	0.33	0.85	0.20	0.26
10/80	0 -15	1.39	2.17	1.74	1.54
	15 -30	0.90	0.82	0.98	0.96
	30 -45	0.39	0.34	0.63	0.41
	45 -60	0.31	0.33	0.48	0.37
	60 -90	0.15	0.37	0.15	0.35

Table A-2. DTPA extractable zinc from soils receiving filter cake.

Date	Depth	Treatment			
		Control	Low	Medium	High
	cm	mg/kg soil			
3/81	0 -15	1.62	1.48	0.50	1.00
	15 -30	0.81	0.62	0.20	0.81
	30 -45	0.54	0.32	0.18	0.61
	45 -60	1.28	0.28	0.30	0.61
	60 -90	0.30	0.65	0.31	0.72
11/81	0 -15	1.30	2.89	0.20	0.44
	15 -30	0.82	0.47	0.09	0.66
	30 -45	0.30	0.30	0.34	0.29
	45 -60	0.20	0.29	0.32	0.23
	60 -90	0.08	0.18	0.20	0.48

Table A-3. DTPA extractable iron from soils receiving filter cake.

Date	Depth	Treatment			
		Control	Low	Medium	High
	cm	mg/kg soil			
5/80	0 -15	30.6	50.8	17.5	27.6
	15 -30	25.0	27.8	32.1	32.8
	30 -45	31.4	46.4	24.7	29.0
	45 -60	33.1	45.6	33.7	33.2
	60 -90	21.7	26.5	28.3	24.6
10/80	0 -15	38.1	36.2	46.8	36.4
	15 -30	42.6	30.4	37.6	19.0
	30 -45	25.5	24.4	28.0	29.8
	45 -60	26.2	24.5	27.8	25.3
	60 -90	21.8	27.6	24.0	24.8

Table A-4. DTPA extractable iron from soils receiving filter cake.

Date	Depth	Treatment			
		Control	Low	Medium	High
	cm	mg/kg soil			
3/81	0 -15	29.6	47.8	17.6	31.2
	15 -30	24.0	30.6	20.1	29.6
	30 -45	30.0	29.9	18.2	28.4
	45 -60	26.1	40.8	19.2	36.2
	60 -90	1.1	29.8	21.2	40.9
11/81	0 -15	35.2	16.9	30.0	20.1
	15 -30	44.6	37.8	22.0	18.2
	30 -45	20.6	29.9	27.9	16.2
	45 -60	22.6	22.3	22.8	21.2
	60 -90	21.8	18.9	19.7	27.9

Table A-5. DTPA extractable cadmium from soils receiving filter cake.

Date	Depth	Treatment			
		Control	Low	Medium	High
	cm	mg/kg soil			
5/80	0 -15	0.38	0.10	0.10	0.10
	15 -30	0.07	0.05	0.10	0.07
	30 -45	0.05	0.03	0.03	0.03
	45 -60	0.03	0.08	0.03	0.02
	60 -90	0.03	0.02	0.03	0.02
10/80	0 -15	0.11	0.12	0.13	0.20
	15 -30	0.08	0.09	0.07	0.23
	30 -45	0.06	0.06	0.06	0.06
	45 -60	0.43	0.16	0.45	0.53
	60 -90	0.03	0.03	0.03	0.07

Table A-6. DTPA extractable cadmium from soils receiving filter cake.

Date	Depth	Treatment			
		Control	Low	Medium	High
	cm	mg/kg soil			
3/81	0 -15	0.03	0.03	0.10	0.10
	15 -30	0.02	0.01	0.01	0.04
	30 -45	0.01	0.01	0.09	0.03
	45 -60	0.01	0.01	0.03	0.01
	60 -90	0.01	0.01	0.03	0.01
11/81	0 -15	0.04	0.03	0.04	0.04
	15 -30	0.02	0.02	0.06	1.12
	30 -45	0.02	0.08	0.01	0.08
	45 -60	0.01	0.17	0.01	0.06
	60 -90	0.01	0.03	0.01	0.03

Table A-7. DTPA extractable manganese from soils receiving filter cake.

Date	Depth	Treatment			
		Control	Low	Medium	High
	cm	mg/kg soil			
5/80	0 -15	16.7	19.9	24.2	19.7
	15 -30	21.3	14.0	21.2	19.0
	30 -45	9.9	11.6	11.8	11.0
	45 -60	10.9	18.0	13.1	13.1
	60 -90	12.9	12.5	8.6	12.4
10/80	0 -15	16.6	20.5	21.2	18.3
	15 -30	11.7	18.1	17.3	14.5
	30 -45	10.1	5.58	8.48	9.09
	45 -60	7.42	7.43	8.97	7.43
	60 -90	4.20	7.53	4.56	5.00

Table A-8. DTPA extractable manganese from soils receiving filter cake.

Date	Depth	Treatment			
		Control	Low	Medium	High
	cm	mg/kg soil			
3/81	0 -15	12.9	21.2	10.1	17.4
	15 -30	18.7	16.7	12.1	18.6
	30 -45	9.00	8.97	18.9	17.4
	45 -60	12.8	15.7	16.8	12.9
	60 -90	13.9	12.5	7.99	8.69
11/81	0 -15	18.8	27.2	20.1	7.43
	15 -30	10.7	16.8	17.6	10.1
	30 -45	11.1	4.89	18.6	9.81
	45 -60	7.98	8.82	12.5	10.1
	60 -90	3.87	7.20	17.4	5.00

Table A-9. DTPA extractable copper from soils receiving filter cake.

Date	Depth	Treatment			
		Control	Low	Medium	High
	cm	mg/kg soil			
5/80	0 -15	1.05	0.45	0.18	0.47
	15 -30	0.31	0.27	0.39	0.35
	30 -45	0.36	0.41	0.28	0.39
	45 -60	0.34	0.36	0.33	0.22
	60 -90	0.45	0.29	0.26	0.29
10/80	0 -15	0.42	0.55	0.46	0.47
	15 -30	0.28	2.61	0.39	0.38
	30 -45	0.37	0.26	0.34	0.36
	45 -60	0.29	0.27	0.34	0.29
	60 -90	0.23	0.23	0.23	0.27

Table A-10. DTPA extractable copper from soils receiving Filter Cake.

Date	Depth	Treatment			
		Control	Low	Medium	High
	cm	mg/kg soil			
3/81	0 -15	0.89	0.29	0.11	0.38
	15 -30	0.20	0.27	0.18	0.45
	30 -45	0.30	0.16	0.20	0.46
	45 -60	0.34	0.19	0.30	0.31
	60 -90	0.51	0.20	0.77	0.72
11/81	0 -15	0.41	0.29	1.11	0.81
	15 -30	0.20	0.39	0.89	0.17
	30 -45	0.30	0.21	0.91	0.47
	45 -60	0.21	0.24	0.67	0.29
	60 -90	0.19	0.17	0.45	0.38

Table A-11. DTPA extractable nickel from soils receiving filter cake.

Date	Depth	Treatment			
		Control	Low	Medium	High
	cm	mg/kg soil			
5/80	0 -15	0.80	0.45	0.10	0.43
	15 -30	0.57	0.41	0.26	0.46
	30 -45	0.81	0.69	0.32	0.50
	45 -60	0.80	0.74	0.69	0.62
	60 -90	0.90	0.77	0.74	0.91
10/80	0 -15	0.91	1.14	1.36	0.97
	15 -30	0.58	0.79	1.16	0.87
	30 -45	0.66	0.93	0.73	0.74
	45 -60	0.79	0.83	0.99	0.80
	60 -90	0.64	0.78	0.58	0.65

Table A-12. DTPA extractable nickel from soils receiving filter cake.

Date	Depth	Treatment			
		Control	Low	Medium	High
	cm	mg/kg soil			
3/81	0 -15	0.70	0.40	0.18	0.27
	15 -30	0.67	0.61	0.11	0.40
	30 -45	0.60	0.52	0.69	0.38
	45 -60	0.80	0.40	1.12	0.18
	60 -90	0.79	0.27	0.99	0.36
11/81	0 -15	0.81	0.18	1.00	0.42
	15 -30	0.47	0.67	1.24	0.29
	30 -45	0.62	0.49	0.86	0.17
	45 -60	0.18	0.39	0.17	0.19
	60 -90	0.69	0.17	0.29	0.24

Table A-13. DTPA extractable lead from soils receiving filter cake.

Date	Depth	Treatment			
		Control	Low	Medium	High
	cm	mg/kg soil			
5/80	0 -15	1.93	2.05	1.00	1.63
	15 -30	10.69	1.43	1.59	1.46
	30 -45	1.62	1.93	1.20	1.65
	45 -60	1.39	1.69	1.51	1.28
	60 -90	0.86	1.34	1.40	1.28
10/80	0 -15	1.95	2.74	2.27	1.83
	15 -30	1.12	1.78	1.52	1.63
	30 -45	1.48	1.46	1.44	1.49
	45 -60	1.48	1.25	1.45	1.51
	60 -90	0.98	1.19	1.11	1.12

Table A-14. DTPA extractable lead from soils receiving filter cake.

Date	Depth	Treatment			
		Control	Low	Medium	High
	cm	mg/kg soil			
3/81	0 -15	1.80	1.97	1.51	1.11
	15 -30	12.91	1.19	1.26	1.05
	30 -45	1.42	2.16	1.22	1.29
	45 -60	1.20	2.11	1.61	1.28
	60 -90	0.80	1.86	1.99	1.57
11/81	0 -15	1.88	1.60	2.11	1.66
	15 -30	1.00	1.54	1.58	1.89
	30 -45	1.61	1.20	1.54	1.66
	45 -60	1.20	1.30	2.19	1.27
	60 -90	0.88	1.11	2.01	1.81

Table A-15. Available phosphorus in soils as affected by additions of mycelia filter cake.

Depth	Treatment	date			
		5/80	10/80	3/81	11/81
		ppm			
0 - 15	Control	88	73	68	57
	Low	84	60	58	48
	Medium	83	61	60	51
	High	85	59	60	50
15 - 30	Control	79	55	52	35
	Low	64	44	47	37
	Medium	65	50	48	43
	High	75	52	51	42
30 - 45	Control	38	27	28	22
	Low	24	16	16	13
	Medium	23	18	18	14
	High	30	24	24	19

Table A-16. Total NO_3-N and NH_4-N in top 45 cm as affected by application of mycelia filter cake.

Treatment	date					
	5/80	6/80	7/80	8/80	9/80	10/80
	kg/ha NO_3-N in 1980					
Control	21.6	29.1	47.1	12.0	10.5	10.9
Low	15.6	30.4	53.3	27.4	17.3	19.2
Medium	16.1	35.6	67.9	30.7	31.7	22.0
High	15.9	36.9	87.6	91.8	41.5	58.0
	kg/ha NH_4-N in 1980					
Control	2.6	3.5	6.0	5.1	1.4	2.7
Low	1.7	3.6	4.9	5.6	1.2	1.9
Medium	2.4	4.3	5.2	5.0	1.3	1.7
High	2.3	5.4	4.3	5.2	1.2	2.8

Table A-17. Total NO_3-N and NH_4-N in top 45 cm as affected by application of mycelia filter cake.

Treatment	date					
	3/81	5/81	6/81	7/81	9/81	11/81
	kg/ha NO_3-N in 1980					
Control	9.4	13.1	37.3	28.6	11.7	8.7
Low	15.7	18.3	48.3	28.3	14.6	9.2
Medium	16.0	27.9	70.8	21.3	12.5	16.0
High	28.6	32.3	54.2	26.5	13.1	38.3
	kg/ha NH_4-N in 1980					
Control	4.8	3.5	2.9	3.8	4.0	1.6
Low	3.6	2.2	3.0	5.2	3.0	1.9
Medium	3.8	2.5	3.0	5.1	3.2	1.8
High	2.7	1.6	2.8	4.6	3.3	2.0