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Bulletin No. 26: Recycling Mycelium - A Fermentation Byproduct Becomes an Organic Resource

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Recycling Mycelium

a fermentation byproduct becomes an organic resource

50th ANNIVERSARY

THE CONNECTICUT ARBORETUM
CONNECTICUT COLLEGE, NEW LONDON, CONNECTICUT

BULLETIN NO. 26

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Front cover: Close-up view of mycelium, an organic soil amendment, as spread on experimental fields at the Connecticut Arboretum.

Recycling Mycelium

a fermentation byproduct becomes an organic resource

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THE CONNECTICUT ARBORETUM

Bulletin No. 26 August, 1981

Foreword

Over the past two decades the Connecticut Arboretum has promoted an environmental awareness on a variety of fronts. In the 1960's we published two bulletins concerned with the sound use of herbicides in vegetation management along town roadsides and cross-country rights-of-way. At about the same time the need for tidal marsh preservation in the State was highlighted by *Connecticut's Coastal Marshes: A Vanishing Resource*. Soon thereafter protective marsh legislation was enacted. In 1975 our interest in naturalistic landscaping led to another effort, *Energy Conservation on the Home Grounds: The Role of Naturalistic Landscaping* which emphasized how using natural vegetation and native plants can reduce lawn size and save fossil fuel energy.

Now we turn to another important idea, recycling — a basic ecological process operative in all natural systems. Our constant goal in an industrial society must be to mimic this efficient recycling process that has evolved in natural ecosystems. At the human ecosystem level industries are producing useful products, but byproducts also result. Pfizer is no exception. The daily production of large tonnages of mycelial residues as a result of fungal activities in producing antibiotics and other products and their disposal on land initially posed some serious problems in the early 70's. However, they have been satisfactorily solved. This bulletin presents a series of articles which document how an industrial byproduct has been successfully turned into an asset. It is useful as a fertilizer for ornamental woody plants; it can aid in land rehabilitation; and it can increase agricultural productivity.

The Arboretum staff is proud to have been involved in this venture and we warmly acknowledge Pfizer's contribution in making this publication possible. The dedicated efforts of Gilbert C. Wagner and Anthony A. Biesada have been invaluable in the preparation of this publication.

Our environmental goal must be one of closing ecological cycles wherever possible. Our future survival on this planet may well depend upon our ability to harmonize with such basic ecological principles.

William A. Niering

William A. Niering
Director

A Question of "Waste"

EDWARD R. RICCIUTI
Journalist and Naturalist

The use of microbial techniques to make products as varied as fuels, foodstuffs and pharmaceuticals from natural raw materials such as grains has acquired the name "biotechnology." So bright are the prospects for biotechnology that talk mounts of its revolutionizing the chemical industry, just as miniaturization triggered the boom in electronics.

Perhaps so but, in a sense, biotechnology is hardly revolutionary. People have used one of the basic techniques of biotechnology for thousands of years; it is fermentation. When primitive hunters first aged meat, they unwittingly employed fermentation. The brewing and baking arts depend on it, and always have, although to be sure, people did not understand the nature of fermentation until a century ago.

The idea that fermentation was caused by microbes — living entities — awaited discovery until 1857, when the concept of the process as biological was affirmed by Louis Pasteur.

Of all the achievements based on microbial skills, none surpassed the development of antibiotics to combat infection. The evolution of technology to grow microbes in numbers sufficient for commercial harvest made the life-saving antibiotics possible.

Penicillin was the first mass-produced antibiotic. It accompanied the allied troops onto the Normandy beaches in 1944 and was administered to civilians on a wide scale after World War II ended. Discovery and distribution of many other antibiotics followed.

In a way, the fermentation industry can be viewed as a form of agriculture, with microscopic plants — yeasts, molds and bacteria — as crops. Traditionally, microbes are grown within a large tank. It contains both a broth, the medium that promotes sufficient microbial growth to yield the desired product, and the substrate, the source of the carbon building blocks that the microbes rearrange.

A variety of materials can make up the broth. Common are corn, sugar, animal and fish protein, and starches. The carbon source is sometimes starch, but usually sugar, from molasses or corn. The starch or sugar may provide carbon for building blocks as well as growth.

Once the microbes have built up to production level they may generate a product for several days. Fermentation ends when the nutrients in the broth are exhausted.

Although a fermentation vat may contain thousands of gallons of fluid, the harvest usually is quite small. In some cases it may be less than one-ten-thousandth of a percent of the contents of the vat.

The product is recovered from the mixture by a number of methods, including precipitation and adsorption. Remaining in the fermentor is the spent broth, plus a mass of microbial cell matter (Fig. 1). Much of the byproduct of fermentation, the basic process of biotechnology, is this fungal mixture called mycelium. As a mixture in its original form it is often an unappealing brown sludge. It is, nevertheless, essentially vegetable matter which has proven and potential value as a nourishing soil builder and fertilizer.

The Mycelium Story

Mycelium is familiar to botanists. It is the vegetative body of the fungi, usually consisting of fine threads, spreading from the plant like a tightly-woven network. In a mushroom, for instance, the mycelium extends underground below the stalk. The mycelium of a fungus parasite on a tree permeates the wood from which it obtains nutrients.

As microbes grow in a fermentor they produce mycelia, just as a mushroom does in the ground or a fungus in wood. The growth of mycelia in a fermentor has been likened to the way a tree produces leaves which, when their function is completed, die. Similarly, mycelium dies when fermentation, and thus cellular growth and chemical production, is over.

When a fermentor is cleaned the broth is filtered from the mycelium. When concentrated, it is sometimes sold as a protein supplement for animal feed. The broth can also be treated biologically as wastewater and discharged in accordance with standards governing industrial effluent. The disposal of mycelium is another matter.

Seventy percent water, the mycelial residue is now a dark, mucky sludge, smelling a bit like dried mushrooms and composed largely of carbohydrates such as cellulose and starch, fats, protein and plant nutrients, including traces of zinc and phosphorus.

An industrial plant making antibiotics, citric acid and similar fermentation products generates vast amounts of mycelial residue, 200 tons daily in the case of one large facility. Over the years, the fermentation industry has had to cope with this major waste disposal problem. At times it has sparked controversy, especially as public concern has risen over the effects of all industrial wastes on the environment.

Mycelium is not toxic, no more so than corn husks. Nevertheless, in purely physical terms, finding suitable sites in which to deposit the mountains of mycelium is a Herculean task that is sure to grow as the fermentation industry responds to the emphasis on biotechnology. The odds are good, in fact, that if the benefits of biotechnology are to be realized, highly efficient means of managing mycelium waste must be found.

One promising approach is the application of mycelium as a fertilizer and soil builder. Its potential has been proven by several scientific studies and by practical experiments conducted at the Groton, Connecticut plant of Pfizer Inc.

For some two decades, the Groton plant had disposed of mycelium by barging it to an approved site in Long Island Sound. Although authorized to do so by the United States Army Corps of Engineers, the practice was ordered stopped in 1972 by the Environmental Protection Agency, then newly-established as a regulatory arm of the federal government. The reason given was that ocean dumping violated New York State standards covering disposal of solid industrial wastes in its waters, which included the site near Little Gull Island.

Fig. 1. Spent mycelia are separated from fermentation broths in production process.

There was no evidence that mycelium degraded water quality nor that it harmed marine life. On the contrary, mycelium, an organic byproduct, appears compatible with the marine environment. It is somewhat similar to algae and detritus, eaten by some fish and other marine organisms. In fact, mycelium has been successfully tested as a diet for shellfish. Oysters thrive on it.

Be that as it may, Pfizer was faced with the problem of disposing of more than 50,000 tons of mycelium annually. Burning offered one solution, widely used in the fermentation industry. Landfilling was another. Pfizer opted for a method of handling non-toxic biological wastes that, while not brand new, was nevertheless a path scarcely trodden in the industry. The idea, explains Gilbert C. Wagner, Production Manager of the Groton facility, was to exploit the waste by converting it into a useful product, rather than wasting time, money and energy getting rid of it (Fig. 2).

Not without difficulty, Pfizer and a handful of other pharmaceutical companies have promoted the agricultural use of mycelial residues. Worked into the soil, mycelium improves its physical characteristics and ability to hold water. The organic matter enriches plants with nutrients and provides a nitrogen source that is released slowly for optimal systemic uptake.

Fig. 2. Potato field treated with five per cent mycelium at the Pfizer Demonstration Farm.

Southeastern Connecticut farmers receive mycelium at a cost which reflects their distance from the plant. The contractor, who hauls it from the plant, adjusts the price on a distance basis.

Spread over farm land, mycelium resembles soil and has an earthy odor. Only if allowed to remain unworked for a protracted period will the material generate anaerobes that produce an objectionable odor.

Since Pfizer began agricultural application of mycelium, several universities and research institutions have confirmed its value in the production of a variety of cash crops. Studies by Dr. Henry C. De Roo of the Connecticut Agricultural Experiment Station at New Haven have shown that corn fields to which 100 tons of fresh mycelium were applied over a two-year period produced significantly higher yields than those treated with a commercial fertilizer.

Similarly, corn grown with mycelium at the Arboretum of Connecticut College doubled in productivity over untreated control plots (Fig. 3).

Fig. 3. Root box demonstrates that corn roots thrive in mycelium-treated soil.

Tomato plants grown in mycelial compost also produced more fruit than untreated controls at the Arboretum.

"The use of industrial by-products, such as mycelium, may provide an important supplement for sustaining productivity in home gardens and in commercial operations," concluded Dr. William A. Niering, Professor of Botany at Connecticut College and Director of the Connecticut Arboretum who has worked with Pfizer since 1974 on a mycelium recycling program.

Another experiment involving

tomatoes, this one at Pfizer's 80-acre demonstration farm in North Stonington, Connecticut, showed that plants grown in 40-to-60 tons of raw mycelium per acre as the only source of nitrogen yielded a crop 20 percent greater than those cultivated in a well-known commercial fertilizer, or in urea, a key provider of the element.

Importantly, repeated examination of plant tissues grown in mycelium demonstrates that levels of heavy metals, such as zinc, are well below established tolerances. Pfizer scientists say that trace metals pose no problem to the use of mycelium in agriculture.

Moreover, with phosphorus and potassium supplements, mycelium has been blended into a prepared fertilizer at Groton where it is used experimentally in various plants and grasses grown in Pfizer's greenhouse. Although the mixture is popular with local garden clubs, production economics do not yet make it commercially practical. By the same token, the prohibitive cost of drying, a necessary first step for hauling mycelium significant distances, also has limited its wide-scale use in farming.

The point is, however, that land application of mycelium is demonstrably worthwhile. It is an especially attractive alternative to the wasteful disposal techniques sometimes prescribed. Incineration, for example, not only requires excessive energy but also destroys the ecological benefits latent within this "industrial waste."

Mycelium's beneficial impact goes beyond promoting better food crops. Experiments indicate that, under proper conditions, grass cover and certain native shrubs respond favorably to the fermentation byproducts. During experiments by the Connecticut Arboretum, red twig dogwood, silky dogwood, chokeberry, and mountain laurel grew observably more lush when mycelium was mixed with the soil, as compared with untreated control shrubs of the same species.

Growth of turf grasses which had received mycelium was "dramatic," according to Dr. William R. Wright of the University of Rhode Island Plant and Soil Science Department. Dr. Wright headed a group which tested the effects of mycelial residues on the physical and chemical properties of soils and their influence on plant growth.

The URI research team planted several types of grasses in an abandoned gravel pit composted with mycelium. And, even though the summer was exceptionally dry, the mycelium had a "definite impact on plant growth," the report stated. The experiment holds out hope that mycelium could be used to improve the chances of reclaiming landscape that has been marred by sand, gravel and surface mining operations.

The use of mycelium to reclaim degraded terrain fits in well with the practice of "landfarming," a technique defined as the method of treating industrial waste in the upper level of the soil. Specifically, a byproduct such as mycelium is spread in topsoil so it can decompose. Developed a quarter century ago — but only now drawing real attention — landfarming allows nature to provide its own waste treatment facility.

Mycelium appears to be ideally suited for landfarming because it lacks all but traces of heavy metals, is non-toxic and similar to organic matter already in the soil. Ironically, its decomposition is carried out by the same sort of microorganisms that made the vegetative waste in the first place.

Teeming in the upper layer of the soil, microbes continually recycle both organic and inorganic materials by breaking them down into components which are transformed into matter usable by higher organisms. Mycelium decomposes in the soil just as fallen leaves do.

The primary object of landfarming is waste disposal. But when the technique employs mycelium, a secondary benefit derived is the improvement of the soil itself. This fact, together with the success achieved in rehabilitating gravel pits, suggests that a truly ambitious use of mycelial residues for the reclamation of strip mines may be within the realm of possibility.

Restoration of strip-mined land has been carried out for many years using lime and commercial fertilizer. More recently, however, researchers have been experimenting with municipal sewage sludge in such reclamation projects. Mycelium might serve as well, perhaps better.

The Promise of Mycelium

To transmute process wastes into an environmental good — for many manufacturers it remains a dream. For the fermentation industry, what has already been accomplished with mycelium makes it a near reality — on a small scale, to be sure, but with immense promise.

The notion that mycelium may one day be used for the revitalization of barren land raises even more exciting possibilities, some farfetched, perhaps, but all conceptually down the road along which biotechnology beckons. Might the fermentation industry, for example, ease its waste load by providing byproduct residues to improve the impoverished soils that curse much of the Middle East, vast reaches of India, Africa and other non-productive regions of the Third World? Is it possible that mycelium might be processed into feed for marine animals, thus advancing aquaculture as a means of generating additional protein for humans? Given the scope of the global hunger crisis, such speculation merits serious thought.

Fighting human hunger and solving a waste disposal problem in the process may sound utopian. And yet, at the heart of this idea is a concept whose viability has been confirmed by Pfizer's extensive experience with mycelium thus far. Because fermentation is an inherently natural process, it has immense recycling potential.

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The Role of Mycelial Residues in Old Field Vegetation Development

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In an attempt to analyze the impact of mycelial residues on naturally occurring old field vegetation, a series of experimental plots was established in the Connecticut Arboretum at Connecticut College in the spring of 1974 (Figs. 1, 2). Vegetation development was studied throughout the summer and fall of 1974 and 1975 (Niering *et al.* 1974, 1976). Observations six years after treatment are also noted. Effects on soil moisture, temperature, and leaf chlorophyll content are reported, and observations on soil microfauna are briefly considered.

METHODS

Experimental Area I.

Study Area I was located on level, sandy loam soil south of Benham Avenue and adjacent to the Central Vermont Railroad tracks. Here "aged" mycelium (mycelium which had been stored in open air from twelve to eighteen months) was applied at varying rates (see Table I) to three contiguous plowed plots (Plots A, C, and D), each approximately 12 by 20 meters. A fourth plowed but untreated plot (Plot B) served as the control. These plots had been recently abandoned from vegetable garden use. After application of the mycelium the treated plots were again rototilled. In the heaviest applications complete incorporation of the mycelium was difficult. Eight contiguous plots to the west and south (Plots As through Hs), abandoned from agriculture for a decade or more, were established for surface application of aged mycelium. Here post-agricultural tree and shrub growth occurred within a matrix of perennial grasses and forbs, primarily goldenrod. The woody species were cut to the ground and the tree stumps treated with herbicide prior to the surface application of mycelium. Mycelium was spread at the rates of 75–100 tons/acre, 15–200 tons/acre, and 300 tons/acre.

Fig. 1. Connecticut College Arboretum staff spread fresh mycelium over an experimental plot (1974).

Three permanent transect lines three meters apart were established across each of the four plowed and eight surface-treated plots. In August, 1974, 0.5 by 2 meter quadrats alternating along both sides of these lines were surveyed. Percent coverage of the leafy foliage was recorded by species. Height of the dominant species was measured in every fifth quadrat. All plots were also divided into four large quadrats, where percent cover by species was estimated.

Experimental Area II.

On a second site, north of Benham Avenue and adjacent to the Central Vermont Railroad tracks, a section of perennial grassland, abandoned from light grazing more than a decade ago, was dominated by little bluestem (*Andropogon scoparius*) grassland. Here three treated plots and two controls were established. Following rototilling, two plots (Plots L and M, each measuring 10 by 15 meters) were treated in early June with fresh mycelium at the rate of 500–550 tons/acre; a third plot (Plot I) received 400 tons/acre. Every effort was made to incorporate the fresh mycelium with a large tractor, but the high rate of application and the slippery nature of the wet mycelium made complete incorporation difficult. Plots J and K, each 10 by 10 meters, were established as controls, one merely rototilled and untreated, and the other left in the natural little bluestem grass cover.

RESULTS

Incorporated Aged Mycelium.

Inspection of Experimental Area I plots in mid-August, 1974, approximately three months after treatment, revealed that aged mycelial residues rototilled into the soil did not greatly affect species diversity (Table I). Thirty different herbaceous and woody species were observed in the control plot, whereas the three experimental plots exhibited 24 species (75–100 tons/acre), 27 species (150–200 tons/acre), and 28 species (300 tons/acre). However, plant cover decreased with increasing amounts of mycelium applied (Table I). It was essentially continuous in the control plot (99%) but of low stature, poor vigor, and chlorotic compared to treated areas. Cover was 95% in the plot receiving the lightest application, 82% in that plot receiving the equivalent of 150–200 tons/acre, and only 74% in that plot receiving 300 tons/acre. Absence of plant cover was correlated with the presence of a mycelial crust where incorporation of the mycelium into the soil was incomplete. The dominant species in both treated and control plots were annuals such as crabgrass (*Digitaria ischaemum, D. sanguinalis*) and ragweed (*Ambrosia artemisifolia*) (Fig. 3). The most important perennial associate was lance-leaved goldenrod (*Solidago*

Fig. 2. Composted mycelium plots following preparation as they appeared in 1974.

graminifolia). Plants were taller in treated compared to untreated plots, average heights being: crabgrass 40 vs. 28 centimeters; ragweed 95 vs 71 centimeters; goldenrod 89 vs. 83 centimeters. Increased vigor was exhibited in plants within experimental plots compared to the control, and foliage was a more intense green in color.

During the second growing season there was a dramatic change in plant coverage and floristic composition (Table I). Whereas crabgrass had been the most important species recorded in 1974, ragweed was the dominant annual in 1975, and crabgrass had decreased to less than 1% on all plots except the control. This dramatic decline may be related to allelopathic properties associated with crabgrass, in which it tends to produce properties unfavorable to its re-establishment (Parenti and Rice 1969).

The height and vigor of the ragweed on treated plots were strikingly different from the control. With mycelium the height averaged 1.3 meters (maximum 1.5 meters), whereas on the control ragweed averaged only 0.7–0.8 meters in height. It formed such dense, dark green, tall stands in all treated plots that other species were unable to thrive beneath its continuous cover. This was not true in the control, where the chlorotic, open ragweed stand contained scattered other species.

Although there had been little difference in species diversity in treated and control plots during the initial season, during the second season species diversity appeared to be correlated with the rate of mycelial application (Table I). The control exhibited the largest number of species (46), with decreasing diversity with intensity of treatment (30, 25, and 24 species respectively for the three intensities). Note that only about one-half the number of species (24 vs. 46) occurred on the heaviest treatment compared to the control.

Another interesting contrast between the treated and control plots in the second growing season was the paucity of woody growth, especially big-toothed and trembling aspen (*Populus grandidentata* and *P. tremuloides*) suckers on the treated plots. The initial herbicide stump treatment which followed the clearing of woody growth on the contiguous surface-treatment control plot to the west (Plot Bs) failed to root-kill the aspen. In fact, it was markedly stimulated and its advance into control Plot B (18 suckers or clumps), compared to the three treated plots (3 suckers), suggests that the presence of mycelium had somewhat arrested the vigorous root-suckering habit of the aspen.

Although annual cover was generally dominant in all plots after two years, the perennial quackgrass (*Agropyron repens*) had increased markedly. Vetch (*Vicia* sp.) was more abundant in the untreated area than in the treated plots on either side. The increased vigor of the grass in treated sites may have tended to exclude the vetch, or the relatively high nitrogen levels resulting from the mycelium may have arrested this nitrogen-fixer. Lance-leaved goldenrod was again a conspicuous perennial exhibiting increased vigor in treated vs. control plots.

Total plant cover on treated plots had substantially increased during the second growing season (Table I), especially on the plot receiving the heaviest treatment, where it provided 85–90% cover, whereas one quarter of the plot had been unvegetated in 1974. However, bare spots persisted in all treated areas, ranging from 10–15% in the heaviest to less than 3% in the lighter treatments.

In 1980 field observations of these treated and control plots revealed that quackgrass (Fig. 4) formed a lush, continuous cover (95%) on all treated plots, with only a few ragweed-dominated areas (15%) in the plot that had received the heaviest treatment. These annual-dominated areas, where mycelium incorporation was incomplete, still exhibited a whitish salt layer on the surface soil. Within the dense quackgrass, associated forbs were lance-leaved goldenrod, rough goldenrod (*S. rugosa*), and milkweed (*Asclepias syriaca*), which together contributed only 5% cover. Among the woody species, the aggressive introduced vine, oriental bittersweet (*Celastrus orbiculatus*), already established in treated and control plots, was vigorous and expanding rapidly.

In the control high species diversity still prevailed, with quackgrass, rough goldenrod, and redtop (*Agrostis alba*) among the dominant species.

The clonal invasion of trembling and big-toothed aspen into control Plot B from the adjacent surface-treatment control Plot Bs, initially observed in 1975, had continued, and aspen covered one-third of the control plot in 1980. The trees were 20–25 feet in height, with bright green, vigorous foliage. This was in contrast with the contiguous treated plots, where aspen invasion was meager. On one treated plot only two small, chlorotic stems of aspen had advanced only two meters in the past six years. In fact, this invasion had occurred within the first three years following treatment, with no further advance since. (It is interesting to note that two trembling aspen seedlings had become established in the heaviest fresh mycelium experimental plot to be discussed later). It would appear that some inhibitor or concentration of salts may be present which discourages vegetative clonal invasion of aspen. Competition from the dense grass cover may also be a factor. This phenomenon was the most striking aspect in terms of effect on vegetation of treated vs. control plots.

Surface Application.

During 1974, the dominant aspect in surface-treated plots in Experimental Area I was the pronounced vigor of goldenrod. This trend continued into the second growing season. Heights ranged up to 1.5–1.8 meters for the goldenrod in treated plots compared to 1.0–1.4 meters in the control.

During the first growing season plant cover varied widely between plots, ranging from 50–60% on two treated plots to over 90% on three other treated plots. By 1975 most of the treated plots, regardless of the intensity of application, exhibited little open space or unvegetated bare surface, with 90% coverage or more on all treated plots. In 1974 plant growth had been greatly suppressed in the plot having the heaviest surface application (300 tons/acre), where a heavy mycelial crust had formed; by 1975 this area was colonized by dewberry (*Rubus flagellaris*) and Virginia creeper (*Parthenocissus quinquefolia*), which were creeping over the surface.

In 1975 other woody growth had increased on the surface-treated plots. Winged sumac (*Rhus copallina*) showed a striking response to mycelium, with plants up to 2 meters in height, leaves very shiny, and plants fruiting profusely. In contrast, no fruiting was observed in the control. More suckering of aspen appeared in the control (Plot Bs) than in the contiguous treated plots (As, Cs), but considerable suckering was evident on one treated plot (Es).

Species diversity was apparently not modified by the mycelium in surface-treated plots. In fact, the largest number of species (44) was found in lightly treated plot Gs (75–100 tons/acre), whereas the highest number of species found in any control plot was 35.

After two growing seasons the positive aspects of surface treatment of mycelium on post-agricultural fields were increased density and vigor of perennials, especially goldenrod, and certain woody species such as winged sumac. By 1980 a shrub-early forest community was evident. Increased development of winged sumac, oriental bittersweet, and aspen had reduced the former importance of goldenrod and other herbs. The surface application of mycelium had served as a marked stimulus to woody growth. However, aspen still showed a marked chlorotic and stunted appearance on mycelium treated plots.

Incorporated Fresh Mycelium.

By the end of the initial growing season in 1974, all plots treated with incorporated fresh mycelium in Experimental Area II were essentially devoid of plant cover, whereas the plowed, untreated control was covered with an almost continuous mat of the annual, carpetweed (*Mollugo verticillata*). In 1975, a year later, plant cover had increased dramatically to 70–80% on all plots. As in the incorporated aged mycelium plots, annuals were conspicuous pioneers, the dominant ones being ragweed and horseweed (*Erigeron canadensis*). The most important perennial was quackgrass, especially in those sections of the plots where mycelial incorporation was completed (Fig. 5). In the control, carpetweed was replaced in 1975 by redtop. Perennial clovers (*Trifolium* spp.) were present in all plots but were especially abundant in Plot M. Also present were scattered specimens of the annual sunflower (*Helianthus annuus*) and millet (*Setaria italica*), both probably dropped by birds from nearby winter feeders.

Preliminary data on species diversity suggested that the largest number of species (47), many of which were annuals, was found in the lighter treatment, decreasing to 23 species on one of the plots receiving the maximum rate of application (Table I). Although woody growth was 1% or less in all treatments, up to 12 species of trees and shrubs, mostly seedlings, were recorded, including oak (*Quercus alba, Q. velutina*) and flowering dogwood (*Cornus florida*). This emphasizes the very early natural establishment of forest tree seedlings, or the concept of initial vegetation floristics in old field development, in contrast to the concept of relay vegetation floristics (Egler 1954, Niering and Goodwin 1974). The most abundant seedling was oriental bittersweet. These seedlings, some up to nearly 0.5 meters in height, were especially abundant on Plot M.

The aggressive nature of species

Fig. 3. Connecticut College students analyze experimental plots treated with composted mycelium in the initial growing season, summer 1974. Dominant plant cover is ragweed. This plot received the heaviest application.

Fig. 4. Six years after composted mycelium treatment, a dense quackgrass has become established and aspen tree invasion has been arrested. In contrast, one third of control plot in background is covered by aspen.

colonization of these sites suggests that fresh mycelium, a year after incorporation into the soil, is not a deterrent to plant establishment and growth, even at tremendously high rates of application. A considerable number of herbaceous perennials was already present one year after treatment.

By 1980, all incorporated fresh mycelium plots exhibited a lush dominance of quackgrass (90%), as in the aged mycelium plots. Grass flowering stalks reaching a maximum of 1.4 meters in height were recorded. All associated species, including pokeweed (*Phytolacca americana*), bittersweet nightshade (*Solanum dulcamara*), common St. Johnswort (*Hypericum perforatum*), butter-and-eggs (*Linaria vulgaris*), exhibited vigorous growth as well as flowering and fruiting. Collectively they represented only 5–10% of the plant cover. The aggressive nature of the dominant quackgrass apparently contributed to limiting species diversity.

Woody growth by 1980 was confined primarily to those species, such as oriental bittersweet, trembling aspen, and Tartarian honeysuckle (*Lonicera tatarica*), which had become established in the first year or two. Bittersweet appeared to be the most aggressive species. Total woody cover was only 5%.

In the plowed but untreated plot two grasses, redtop (70%) and little bluestem (30%), were most conspicuous in 1980. The future trend appeared to be toward an increase in little bluestem. Woody growth, primarily winged sumac, provided less than 1% cover.

In the unplowed control, little bluestem contributed 95% coverage, with rough goldenrod and hairgrass (*Deschampsia cespitosa*) providing less than 5% cover. Woody cover, primarily winged sumac, was less than 1%. This plot was exhibiting remarkable stability as a grassland type of vegetation.

Fig. 5. Fresh mycelium plot two years after treatment. Note dark green quackgrass surrounding the tan control area in center (May, 1976).

TABLE I
Species diversity, plant cover and dominant vegetation in mycelium-treated and control plots.

	Number of Spp.		Plant Cover %		
Plot	1974	1975	1974	1975	Dominant Vegetation (% Cover)
Area I Aged mycelium rototilled (tons/acre)					
A (300 t/a)	28	24	74	88	1974 crabgrass (60); ragweed (40); quackgrass (15) 1975 ragweed (70); quackgrass (15)
C (150–200 t/a)	27	25	82	98	1974 quackgrass (40); crabgrass (30); ragweed (25); goldenrod (10) 1975 quackgrass (73); goldenrod (20); ragweed (15)
D (75–100 t/a)	24	30	95	97	1974 crabgrass (85); purslane (35); ragweed (8) 1975 ragweed (65); quackgrass (23); goldenrod (13)
B (control)	30	46	99	93	1974 crabgrass (90); ragweed (20) 1975 ragweed (65); quackgrass (18); goldenrod (18); aspen (18); vetch (3)
Area II Fresh mycelium rototilled					
I (400 t/a)	4	47	<5	80	1974 essentially devoid of plant cover 1975 quackgrass (28); redtop (18); ragweed (8); horseweed (8)
L (500–550 t/a)	6	38	<5	73	1974 essentially devoid of plant cover 1975 quackgrass (28); ragweed (18); pokeweed (5); panic grass (5); foxtail grass (5)
M (500–550 t/a)	4	23	<3	88	1974 essentially devoid of plant cover 1975 alsike clover (28); ragweed (18); quackgrass present
J (control, rototilled)	13	14	95	100	1974 carpetweed (95) 1975 redtop (65); sheep sorrel (45)
K (control, unrototilled)	6	17	95	95	1974 little bluestem (80) 1975 little bluestem (75)

Soil Moisture and Temperature.

Soil moisture was measured in the treated plots to which aged mycelium was added and rototilled into the soil, as well as in the rototilled control plot. Gypsum soil moisture blocks were placed at 5, 15, and 35 centimeters below the surface. Readings were taken with a Bouyoucos moisture meter, model BN-2B, which can be read directly as percent available moisture in the soil. Results are shown in Table II.

On July 3, 1974, soil temperature readings were taken in plots which were treated with incorporated aged mycelium and those treated with incorporated fresh mycelium, as well as in two untreated control plots. Readings were made with a Weston soil thermometer at 10 centimeters below the soil surface.

Temperatures in plots that received aged mycelium were only slightly higher than those in the controls (Plot A, 30°C; Plot C, 26°C; controls 20°C and 27°C).

TABLE II
Percent available soil moisture at three different depths during initial season in two aged mycelium-treated plots and contiguous control.

	DEPTH								
	5 cm			15 cm			35 cm		
DATE	T1*	T2*	C*	T1	T2	C	T1	T2	C
7/10/74	100	100	69	100	100	90	100	100	97
7/30/74	100	100	23	100	100	28	100	100	43
8/7/74	100	100	24	100	100	38	100	100	59
8/23/74	90	52	0	70	36	0	74	28	0
8/28/74	80	47	0	49	32	0	57	21	0
9/20/74	100	100	59	100	94	57	100	100	62

*T1 — Plot A, 300 tons of aged mycelium per acre
*T2 — Plot D, 75–100 tons of aged mycelium per acre
*C — Plot B, control (no mycelium)

Note that soil moisture was consistently higher in treated plots than in the control. During a three week period without precipitation in August, moisture readings in the control plot were zero, whereas moisture levels never fell below 21%, and averaged 53%, within aged mycelium plots. During this dry spell moisture readings in the plot receiving the largest amount of mycelium were consistently higher than those in the plot receiving the lightest application. In plots treated with fresh mycelium, available moisture remained at 100% during the entire season.

However, temperatures in the plots which were treated with fresh mycelium were nearly double those in the controls (Plot I, 37°C; Plot M, 38°C). Thus it appears that considerable heat of fermentation was being released into the soil in the fresh mycelium plots.

Leaf Chlorophyll Content.

In aged mycelium Plot A and control Plot B chlorophyll content, expressed in milligrams of chlorophyll per gram of plant tissue, was analyzed for two species of goldenrod, *Solidago graminifolia* and *S. rugosa*. Results are shown in Table III.

TABLE III

Leaf chlorophyll content for two species of goldenrod from mycelia-treated and control plots.

Plant	Chlorophyll Content mg chlorophyll/gm tissue	
	Chlorophyll a	Chlorophyll b
S. graminifolia		
treated (aged mycelium, incorporated)	2.344	1.856
control	1.298	.996
△ %	80.58	86.34
S. rugosa		
treated (aged mycelium)	2.769	2.382
control	1.479	1.262
△ %	87.22	88.74

Note that in all cases there is more chlorophyll a and chlorophyll b in treated than in untreated leaves. This was borne out by the more intense green of the leaves of treated goldenrod.

The Effect of Mycelial Residues on Soil Microfauna.

In an effort to determine the effect of mycelial residues on the soil microfauna, soil samples were obtained from one experimental plot which had received incorporated aged mycelium, one plot which had received incorporated fresh mycelium, and two control plots. Samples were obtained two months and four months after treatment in 1974, and three times during the summer of 1975. Soil was processed through modified Tullgren funnels and animals collected in jars containing 70% ethanol.

Application of mycelial residues did not affect the diversity of arthropods or their numbers, with the exception of mites. Very few of the latter were found in the untreated control plots in 1974 (.017 and .034 mites per gram of soil), but mites were found in large numbers in that year in the aged mycelium-treated plot (17.9 mites per gram of soil) and in moderately high numbers in the fresh mycelium-treated plot (.493 mites per gram of soil). The majority of the mites were hypopial nymphs of the genus *Caloglyphus* (family Acaridae), which are fungus and carrion-eating mites.

In 1975 the average number of mites per gram of soil in the aged mycelium plot was .034; in the fresh mycelium plot the average was .103; in the two controls the averages were .038 and .061. The difference in the numbers of mites found in 1974 and 1975 appears to indicate that the factor responsible for the large numbers of mites found soon after the application of both aged and fresh mycelium was no longer present one year after application.

Summary

Mycelial residues have a positive impact in increasing the vigor and growth of old field species. With the addition of mycelium there is a tendency to favor monocultures of certain species. After six years quackgrass is the dominant plant cover on all mycelial-treated areas in contrast to a mixed perennial grass forb cover on contiguous control plots. Species diversity tended to be highest in control plots. The lush green nature of the vegetation on treated plots is reflected by the higher levels of chlorophyll. The incorporation of mycelium also results in higher available soil moisture during the growing season compared to control plots. Among the soil microfauna, mite populations increased dramatically with mycelial applications.

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The Pfizer Demonstration Farm

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Pfizer Inc. expanded its own laboratory and greenhouse work on mycelium by establishing a demonstration farm on Taugwonk Road in Stonington, Connecticut in 1975. The staff of the Connecticut Arboretum at Connecticut College was invited to initiate a research program on agricultural crops at the newly purchased facility.

Experiments at the Pfizer Farm during the period 1975–77 centered on determining whether mycelium had a viable role in agriculture as a soil amendment for a wide variety of vegetable crops, silage corn and hay. Beginning in 1978, a slightly different approach was taken, with the focus turning to the production of silage corn and hay only, the two crops on which mycelium is currently used.

Demonstration farm established by Pfizer in 1975 at Stonington, Connecticut.

1975 GROWING SEASON

In 1975, experimental plots were established on fields originally under perennial grass cover. Plots were plowed and treated with 1) fresh mycelium only (40 tons/acre), 2) fresh mycelium (20 tons/acre) and fertilizer, 3) fertilizer only, and 4) untreated, to serve as controls. Harrowing and seeding were done shortly after the application of mycelium. Lime was applied to fertilizer and control plots for soybeans and corn. Approximately 2½ acres were in vegetable crops, soybeans, and ornamental woody plants and 2½ acres under silage and sweet corn. Hay fields were subjected to surface applications of fertilizer and fresh mycelium. A wet spring, poor drainage in some parts of the fields, and late application of mycelium may have affected plant response in some areas.

Vegetables, Soybeans, Corn & Hay.

The use of fresh mycelium had beneficial effects on soybeans and bush crops (green beans, yellow wax beans, peppers, and tomatoes). Productivity of tomatoes grown with fresh mycelium treatment was high. Fruits totalling 10,339 grams were harvested from treated plots, compared to a total of 6,509 grams in the fertilizer-treated plots. The reason for the dramatic response of soybeans to fresh mycelium treatment compared to fertilizer plots is not clear. However, the high levels of ammonia N and nitrate N in the mycelium may be involved. Cucurbits were affected positively by the addition of mycelium, showing greater production of cucumber and zucchini. Leafy crops (lettuce, cabbages) were stunted in plots treated with fresh mycelium only; best results occurred with mycelium and fertilizer or fertilizer only. Planting of most root crops (beets, onions, radishes and turnips) immediately following the application of fresh mycelium resulted in poor germination and initial stunting of growth; second plantings responded more favorably at the end of this first growing season. It was recommended that fresh mycelium be applied and incorporated several months prior to planting; however, experience in the subsequent growing years has shown that no lag time is necessary even for sensitive crops.

Silage corn showed the highest productivity where fertilizer alone was used. Production of sweet corn was lower in plots containing mycelium alone than in plots treated with mycelium plus fertilizer or fertilizer alone. Increased levels of mycelial application are recommended for growing corn since it is a high nitrogen-requiring crop. Hay fields treated with fresh mycelium showed a 42% increase in productivity over untreated portions of the fields. Fertilizer-treated areas showed evidence of leaf burn but responded slightly better than the control.

1976 GROWING SEASON

Although the first year's findings were mixed, due in part to drainage, weather and site preparation difficulties, mycelia use on cropland showed enough promise to continue experiments at the farm in 1976, under the direction of Pfizer personnel.

The 1976 growing season showed an improvement in both soil conditioning techniques and crop production. Experimental conditions were altered to specifically test mycelium's ability as a nitrogen fertilizer when compared to urea, by adding phosphorous and potassium to all experimental plots. The plots for 1976 consisted of: 1) mixed mycelium at 60 wet tons/acre, 2) citric mycelium at 40 wet tons/acre, 3) 180 pounds/acre nitrogen (urea), 4) no nitrogen. The crops included a majority of the common garden vegetables as well as sweet and silage corn and hay.

Vegetables, Soybeans, Corn & Hay.

Approximately 80% of the vegetable crops grown showed increased productivity over commercial nitrogen under at least one of the mycelium treatments. "Early Girl" and "Better Boy" tomatoes averaged 30% greater yields in the mycelium treatments compared to commercial nitrogen. Soybeans treated with mycelium averaged approximately 10% greater yields. Leaf crops showed improved yields over 1975 with the mycelium treatments. It was also noted that no inhibition of germination occurred in crops that had received mycelium just prior to planting.

Regular spaced and high density silage corn grown in the mycelium plots had an average 20% greater net weight than those treated with commercial nitrogen fertilizer. Two varieties of sweet corn, "Butter and Sugar" and "Golden Bantam" averaged a 20% greater yield of ears in the mycelium treatment. In the fall of 1975 one-half of a three-acre hay field was spread with mycelium equivalent to 40 tons/acre. The other half was treated with 300 lbs./acre of 10-10-10 fertilizer. The entire field was spread with limestone at the rate of one ton/acre. The production of hay bales was approximately 50% greater on the mycelium side of the field, with the majority of the difference occurring in the second cutting. Higher yields in the second cutting are believed to be due to the slow release long-lasting organic nitrogen contained in the mycelium.

1977 GROWING SEASON

In 1977 the major aim of the work at the Pfizer Farm was to determine an optimum application rate for mycelium. Mycelium was applied at rates of 15, 45, and 135 wet tons/acre. No additional phosphorous or potassium was added to the lots in 1977. Crops at the farm included silage corn, sweet corn, a variety of garden vegetables, hay, and potatoes.

Vegetables, Corn & Hay.

Vegetable production was clearly enhanced by the presence of mycelium in the fields in 1977. Crops such as tomatoes, peppers, cabbage, summer squash and beets produced their highest yields under one of the mycelium treatments. Overall, the 135-ton and 45-ton/acre applications of mycelium produced the highest yields on vegetables. Tomatoes again showed especially impressive yields with the 45-ton/acre plants averaging over 50 pounds of fruit production per plant.

Silage corn which received 45 tons/acre of mycelium produced the second highest overall yield of all silage corn treatments (15% higher dry wt. than the "no-nitrogen" controls). In 1977 no new treatments were applied to the hay field in order to test the residual effects of the previous year's treatment. Results showed a drop in overall productivity of approximately 50%, indicating a lack of fertilizer for both treatments. There was, however, no significant difference between the mycelium and the commercial fertilizer treatment.

1978 GROWING SEASON

In 1978 the focus of the mycelium experiments was shifted to emphasize the production of silage corn and hay, dropping the extensive testing of vegetable crops. The mycelium was applied to permanently established plots on the basis of two criteria; one that the crops's nitrogen requirement should be supplied by the mycelium, and two that the application should not impose any heavy metals stress on the soil. Calculations based on the metals content of the mycelium showed that it could be applied to land at agricultural rates for over 50 years before any theoretical metals limitations would ever be reached. The nitrogen content of the mycelium and the soil test recommendation made an application rate of approximately 40 tons/acre optimum for the silage corn crop. Commercial grade phosphorous and potassium (0-25-25) were added according to the soil recommendation.

Silage Corn & Hay.

Corn grown in the 40-ton/acre mycelium treatment produced over 50% greater dry weight than the controls which had received no nitrogen. Results in the hay field showed yields on the mycelium side to decline by 20% in the second year of no-treatment following the original fertilization in 1976.

A second hay field was prepared in the fall of 1977. After plowing and harrowing, one side of the field was treated with 40 tons/acre of mycelium and 400 pounds/acre of 0-25-25. The other side was treated with 480 pounds/acre of 10-10-10. The field was seeded with oats and timothy. Tabulations at the end of the 1978 growing season showed the hay field on the mycelium side was approximately 40% greater than on the commercial fertilizer side in the newly planted field.

1979 GROWING SEASON

In 1979 the tests were continued in the same manner as in 1978; however, commercial fertilizer plots were added to compare the nitrogen-feeding capability of mycelium with that of commercial fertilizer on corn. The amount of nitrogen contained within the mycelium was calculated and this was matched in the commercial fertilizer plots (150 lbs. N/acre). Phosphorous and potassium were also added to all plots in equal amounts.

Silage Corn & Hay.

Although silage corn yields were down overall in 1979 due to a wet spring and a

Silage corn is chopped and loaded at Lewis Farm in North Stonington.

Production Manager Gilbert C. Wagner of Pfizer is dwarfed by stand of mycelium-grown silage corn.

Field of rye in mycelium-dressed field at Salem, Connecticut.

summer drought, the mycelium plots again produced good yields. Dry weight (or plant production) in the mycelium-treated plots was 18% greater than the commercial fertilizer plots and 60% greater than the no-nitrogen controls. The hay experiments were altered in 1979 to specifically test two things; first, to compare mycelium and commercial fertilizer when applied on an equal nitrogen basis, and second to compare the two on an equal cost basis. In the fall of 1978, one-half of one hay field received the soil test recommended amount of nitrogen in the form of 10-10-10 fertilizer, while the other half received that same amount of nitrogen in the form of mycelium. A second field was divided in half with one half receiving the recommended commercial nitrogen application and the other half receiving an equal dollar amount of nitrogen in the form of mycelium (valued at $1.00/ton). First year results showed mycelium not to fare as well as in previous years, producing 8% fewer bales in both the equal nitrogen and equal cost comparison.

1980 GROWING SEASON

In 1980 mycelium plots were again established at the Pfizer Farm and the tests were continued on the same basis as the previous two years. The application rate for mycelium was reduced somewhat according to the nitrogen application formula which accounts for residual nitrogen in the previous years' application of organic residue.

Silage Corn & Hay.

The results for the 1980 crop yields proved mycelium to be an excellent nitrogen source for silage corn. Dry weight yields for mycelium in 1980 were 12% greater than commercial fertilizer and 37% greater than the no-nitrogen controls. The results of the past three years yields with mycelium on silage corn are summarized in Table I.

TABLE I
DRY WEIGHT YIELDS OF
SILAGE CORN 1978–1980

Treatment	Corn Yield Dry Tons/Acre		
	1978	1979	1980
Mycelium	5.5	4.4	7.4
Commercial Fertilizer	—	3.7	6.6
No Nitrogen	3.5	2.7	5.4

Hay yields continued to decline under mycelium treatment during 1980 on both an equal nitrogen and equal cost basis. The reason for this may be mycelium's inability to release its nitrogen quickly enough in the early spring just after application to meet the demands of the growing grass. The readily soluble commercial fertilizers apparently are more effective than a spring top-dressing of mycelium on the established field. Visual observations in early autumn showed the mycelium side to maintain a deeper green color, indicating the slow release of its nitrogen, despite having produced fewer bales of hay.

SUMMARY

In conclusion, it appears that the continued application of mycelium to agricultural land has maintained excellent soil fertility and produced yields comparable to, or greater than, commercial fertilizers on silage corn and a variety of garden vegetables. Results on hay fields have been mixed, but may indicate an over-wintering breakdown period is necessary for mycelium to compete with commercial fertilizer. Mycelium is currently trucked daily from the Pfizer plant in Groton to local farms, where it is used primarily as a nitrogen source for silage corn and hay.

Recycled Industrial Wastes Aid Revegetation

Applying mycelium, an industrial fermentation residue, to post-agricultural land resulted in significant plant growth.

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Since 1974 the Connecticut Arboretum at Connecticut College has worked with Pfizer Inc. in a program aimed at recycling mycelium, the residue from fermentation processes for organic acids and antibiotics. Approximately 200 tons of this byproduct are produced daily by the Groton production plant. For 20 years prior to federal legislation against offshore dumping, this material was deposited in Long Island Sound. This research arose out of the need for an alternative solution for disposal or use of this organic material.

A number of different mycelial products are produced by Pfizer, depending upon the fermentation process. Citric acid production results in a flaky oat meal-like material, while other processes yield a byproduct with the consistency of a thick pudding. Both kinds of material have a water content of approximately 64 percent. They can be used directly in the fresh state, or they can be composted. Filter aids, mostly gypsum and diatomaceous earth (silica), used in the production process account for up to 50 percent of the dry weight of some of the residues. The mycelium, dry basis, contains from 0.5–6.0 percent N, 0.03–0.11 percent P, and 0.02–0.06 percent K. Trace elements expressed in parts per million include Zn 50–400, Cu 5–10, Mg. 30–120, and Fe 40–400 (DeRoo, 1975).

Our initial research evaluating the effect of mycelium on the natural revegetation of post-agricultural fields and on the growth of native shrubs showed highly positive results (Niering et al., 1976). Studies using turf grasses, potatoes, row crops and container grown plants at the University of Rhode Island, the Connecticut Agricultural Experiment Station and the Pfizer Research Farm indicate that the mycelial residues have considerable value as a source of fertilizer and soil amendments (Niering et al., 1974; Wright, 1974, 1975, 1976; DeRoo, 1975; Taylor et al., 1976). The aim of this paper is to evaluate the potential value of this industrial byproduct in rehabilitating an unused gravel pit.

The study area is a former gravel pit, located in Ledyard, Connecticut, and owned by Pfizer Inc. Over a decade ago, the steep slopes were graded to 30–40 degrees and left to natural plant colonization. Although considerable plant establishment has occurred, it is highly variable and extensive areas are bare. The slopes, which rise about 33m from the lowest excavated surface, are composed of a mixture of sand and gravel and 7–12 cm stones (cobble). Where cobble dominates, plant cover is absent. Elsewhere on the slope scattered black birch (*Betula lenta*), aspen (*Populus* spp.), gray birch (*Betula populifolia*) and bayberry (*Myrica pensylvanica*) have become established. The black birch saplings can form relatively dense stands in isolated communities. Herbaceous growth is highly scattered.

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Fig. 1. Application of fresh mycelium-soil mixture on experimental plot, north-facing slope of gravel pit (May, 1976).

Fig. 2. Same slope as above three months later. Herbaceous plants, primarily annuals, cover area treated with mycelium-soil mixture. Note plant cover in topsoil treated plot to left. Scattered black birch saplings appear on the upper slope to right of center. Tapes outline treated and control plots (August, 1976).

Fig. 3. Fresh citric acid mycelium treatment plot (center) on lower slopes showing taller, more vigorous black birch saplings after two growing seasons, compared to untreated control at left margin (Fall, 1977).

METHODS

In May 1976 we established four plots on the lower, north-facing slope near its base which received the following experimental treatments: 1) fresh citric acid mycelium; 2) composted mycelium; 3) fresh mycelium mixed with topsoil; 4) sandy loam topsoil only; and 5) contiguous untreated control plots, (Table I). The mycelium and mycelium-topsoil mixture were placed on each area by crane, then spread by hand to a depth of 5–8 cm over the slope, covering any existing herbaceous plants (Fig. 1). The complications of lifting this material by crane from the dumping area onto the slopes created problems with depositing the same amounts per m^2 on each experimental plot. But the results have been spectacular enough to warrant this report.

Along a section of the uppermost slope near the forest edge we established two 50 m^2 plots where fresh citric acid mycelium was spread by hand. Here the sandy gravel substrate was barely covered. These plots were dominated by black birch saplings 1–2 m high.

Fifty bare-root plants of bristly locust (*Robinia hispida*) ½ m high were planted in the plots treated with topsoil, fresh mycelium and topsoil, and composted mycelium. An additional 50 plants were placed in the control areas.

Analyses were made of the major mineral nutrients in treated and control plots using the Morgan Soil Testing system (Lunt, 1958). Random collection of soil samples from depths of 6 inches were combined, and the pH determined for composite samples from each area.

At the end of the first two growing seasons (1976 and 1977) we estimated the percent of the experimental plot covered by plant growth and compared it to the untreated plots. All plots were thoroughly

Table I. Applications of Mycelium Residues to Experimental Plots

Treatment	Plot Area m²	Tons Applied	Equivalent Dry Tons Per Acre	Depth, cm
(1) Fresh mycelium	A) 250 B) 250	10 10	58 58	5 5
(2) Mixture		4	23 (a)	8
Fresh mycelium 10%		35	568	
Topsoil, 90%	250	39	591	8
(3) Composted Mycelium	150	11 (b)	154 (c)	8
(4) Topsoil	150	10 (d)	281	4
(5) Controls	550			

Note: (a) Based on 64% moisture
(b) 15 cu yd at density of 0.90 g/gm³
(c) Based on 50% moisture
(d) 7 cu yd at density of 1.76 g/gm³

searched for individual plant species, and the percent foliage cover contributed by each estimated, based on the area of the whole plot. Total woody and herbaceous cover was also estimated.

EFFECTS OF SURFACE MYCELIUM APPLICATION

Plant cover, primarily of annual species, increased dramatically in all areas treated with mycelial residues, compared to topsoil-treated and control areas (Fig. 2, Table II). However, where fresh mycelium completely covered the surface, no plant colonization was observed the first year, a pattern observed in our earlier research. Existing woody growth was noticeably stimulated. Black birch saplings up to 2 m high exhibited both increased growth and a strikingly darker green foliage compared to saplings in untreated plots (Fig. 3). Treated birch showed an average annual growth rate of 1 m during the first growing season as compared to 0.5 m in the untreated plots. Black birch saplings on the upper slope plots with a light surface application of fresh mycelium also increased growth two-fold.

The greatest increase in plant cover occurred with composted mycelium. An estimated 90 percent of the ground surface was covered by newly established plant growth by the end of the first two growing seasons (Fig. 2). A somewhat similar pattern occurred where the mycelium-soil mixture was applied on the unvegetated cobble. Here plant growth covered 85 percent of the plot. Where topsoil alone was applied, plant cover increased from 40 percent at the end of the first growing season to 70 percent in 1977. In contrast, untreated control plots were only 20–25 percent covered by plant growth after two years.

The floristic diversity increased dramatically from the first to the second growing season within the treated areas. The most striking increase occurred in the soil-mycelium mixture, where the number of species increased from 34 the first year to 58 the second year. In contrast, only 29 species were recorded in the untreated control areas by the end of the second year. There were 35 herbaceous perennial

Table II. Dominant Plant Cover in Percent Cover Following Treatment With Mycelial Residues on Mineral Soils. Lower North Slope, Pfizer Gravel Pit.

	Fresh mycelium		Fresh mycelium Topsoil		Composted mycelium		Topsoil		Control no treatment	
	'76	'77	'76	'77	'76	'77	'76	'77	'76	'77
HERBS										
Ragweed	—	1	35	30	25	10	18	15	—	—
Annual grasses	—	—	30	20	50	50	3	5	—	—
Goldenrod	3	5	<1	5	<1	<1	—	—	5	5
Quackgrass	<1	<1	—	20	0	20	—	15	—	—
Other species	7	24 *	15	5	15	10	9	15	5	5
Total Herb Cover	10	30	80	80	90	90	30	50	10	10
TREES										
Black birch	6	12	—	—	—	—	5	5	5	5
Poplar	—	2	<1	5	—	—	5	5	5	5
Other species	4	6	—	5	1	2	—	—	2	2
Total Woody Cover	10	20	5	10	1	2	12	20	12	12
TOTAL PLANT COVER	20	50	85	90	90	92	42	70	22	22

*In fresh mycelium plot knapweed and horsetail were dominant species in 1976–1977.

species recorded on the various treatments, and 30 species of annuals by the end of the first growing season. At the end of the second growing season, perennials had increased to 47 and annuals to 31. The significance of this finding is that permanent cover has been established, with the attendant stability of the root systems afforded by the perennial plants.

At the end of the first growing season, the mineral nutrients of the experimental plots were retested. Nitrate and ammonia nitrogen levels in the fresh mycelium plots increased from low to medium high, and remained at low levels in the control plots. Phosphorus levels increased from low to medium levels in all treated areas. The treated areas showed an increase in calcium levels, which might well be expected because of the presence of gypsum in the mycelium. The average change in pH in treated areas rose from 5.1 to 6.7.

Observations four years after treatment reveal that herbaceous perennial cover, especially quackgrass and mixed perennial forbs, dominate all treated plots except for the fresh mycelium area where the two species, knapweed and horsetail, present prior to treatment, have been vigorously stimulated. The planted bristly locust has also increased markedly in all treatments. One of the most dramatic changes evident over the past four years has been the development of vigorous growth of moss in openings not dominated by higher plant cover. This was especially pronounced on the fresh mycelium plot where a nearly continuous cover of moss has stabilized the exposed mycelial surface.

A significant finding was that no detectable erosion occurred in mycelium treated plots whereas topsoil washed downslope readily. This indicates that mycelial residues are especially suited to the stabilization of steep slopes as along newly constructed highway embankments and other such denuded sites in need of vegetation.

A significant finding was that no detectable erosion occurred in mycelium treated plots whereas topsoil washed downslope readily.

CONCLUSIONS

The use of mycelial residues in the revegetation of sparsely or unvegetated, well-drained, nutrient poor gravelly sites appears to be a sound land use practice. Both composted and mycelium-soil mixtures induce new plant establishment and stimulate existing woody plant growth. Fresh mycelium also markedly stimulated the growth and vigor of existing tree saplings. Complete coverage of the gravel surface with mycelial residues results in a lush plant cover; however, existing low perennial plants may be smothered. Therefore, lighter applications are recommended where considerable perennial cover already exists. Mycelial residues are highly suited for slope rehabilitation because they do not wash downslope as readily as does topsoil. The application of mycelial residues hastens the natural process of revegetation.

Both composted and mycelium-soil mixtures induce new plant establishment and stimulate existing woody plant growth.

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The Effect of Mycelial Residues on Selected Woody Plants[1]

SALLY L. TAYLOR

Fig. 1. Cotoneaster grown in control plot after six weeks (July, 1975).

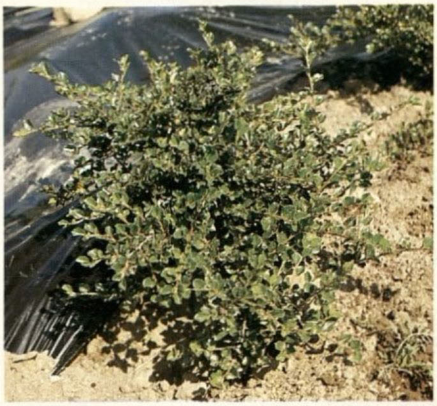

Fig. 2. Cotoneaster grown in mycelium after six weeks (July, 1975).

In 1974 and 1975 the Connecticut Arboretum tested the use of mycelial residues on the growth of trees and shrubs useful for roadside and right-of-way plantings and revegetation of gravel pits. During the first season seventeen different species of shrubs were set out in an area of the Arboretum south of Benham Avenue adjacent to the Central Vermont Railroad. One half bushel of composted mycelium (mycelium stored for twelve to eighteen months and mixed with an approximately equal amount of sand) was added to each planting hole, and an additional quarter bushel was added to the surface around each treated plant. No commercial fertilizer was used. A row of treated plants alternated with a row of untreated plants, and all rows were mulched with black plastic.

The results after the second growing season showed that six species had responded favorably to this treatment. Red twig dogwood (*Cornus stolonifera*), chokeberry (*Aronia arbutifolia*), mountain laurel (*Kalmia latifolia*), beach plum (*Prunus maritima*), fragrant sumac (*Rhus aromatica*), and nannyberry (*Viburnum lentago*) had larger, darker green leaves, as well as compact and sturdy shoot growth. Treated common juniper (*Juniperus communis*) exhibited shorter shoot length, but the more compact plant was especially vigorous. Treated sweetfern (*Comptonia peregrina*) and sheep laurel (*Kalmia angustifolia*) died during the first season, after initial bud break; untreated plants thrived. There was no significant improvement in growth, and no serious damage, to coralberry (*Symphoricarpos chenaultii*), blueberry (*Vaccinium corymbosum*), witherod (*Viburnum cassinoides*), shadbush (*Amelanchier alnifolia* and *A. canadensis*), silky dogwood (*Cornus amomum*), and bayberry (*Myrica pensylvanica*).

Fig. 3. Potted shrubs with mycelium added are transplanted on the east-facing slope of gravel pit (May, 1976).

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[1] Reprinted in part from: ORGANIC FERMENTATION RESIDUES AS A NITROGEN SOURCE AND SOIL AMENDMENTS IN WOODY PLANTINGS, by Sally L. Taylor. Wildland Shrubs, Proceedings, Shrub Sciences Laboratory, Provo, Utah. 1975.

In 1975 fresh mycelium was used on tree and shrub plantings at the Pfizer Demonstration Farm in Stonington, Conn. The mycelial residue was harrowed into the plowed surface soil of three experimental plots at the rate of 40 wet tons per acre or 20 wet tons per acre plus 100 lb. of 5-10-10 fertilizer per acre. An untreated control plot was also established.

In mycelium-treated plots growth was enhanced in alder (*Alnus rugosa*), black chokeberry (*Aronia melanocarpa*), cotoneaster (*Cotonester adpressa* and *C. dammeri*), (Figs. 1, 2) autumn olive (*Eleagnus umbellata*), Japanese holly (*Ilex crenata convexa*), privet (*Ligustrum amurense*), bayberry, beach plum, scarlet oak (*Quercus coccinea*), beach rose (*Rosa rugosa*), coralberry, purple osier willow (*Salix purpurea*), and cranberry viburnum (*Viburnum trilobum*). Vigor and growth were average in shadbush (*Amelanchier canadensis* and *A. laevis*), red chokeberry, sweet pepperbush (*Clethra alnifolia*), silky twig dogwood, gray twig dogwood (*C. racemosa*), red twig dogwood, (*Ilex decidua*), longstalk holly (*I. pedunculosa*), Bar Harbor juniper (*Juniperus horizontalis*), common juniper, Hall's honeysuckle (*Lonicera japońica halliańa*), carmine crabapple (*Malus atrosanguinea*), Hopa crabapple (*M. hopa*), swamp azalea (*Rhododendron viscosum*), elderberry (*Sambucus canadensis*), yew (*Taxus media wardi*), and lowbush blueberry (*Vaccinium pensylvanicum*). Fresh mycelium appeared to have an adverse effect on sweetfern, sheep laurel, mugho pine (*Pinus mugho*), witherod, mollis viburnum (*V. mollis*), and black haw (*V. prunifolium*).

The effect of mycelial treatment on the growth of several potted shrub species was also evaluated on the open gravelly east-facing slope of the abandoned gravel pit owned by Pfizer in Ledyard, Conn. In the spring of 1976 four contiguous plots 10 meters in width were laid out extending from the base to the top of the slope. A total of 160 container-grown specimens 1-3 feet in height were planted in the plots — three treated and one control plot. At the time of planting a soil-mycelium mixture comprising 9 parts of top soil and one part of mycelium* in different concentrations — 25 and 50 lb wet weight was used.

Periodic evaluations made since 1976 indicate that treated plants of red twig dogwood, coral berry (*Symphoricarpus orbiculatus*), Hall's honeysuckle, and Bar Harbor juniper showed a marked increase in growth and vigor. The red twig dogwood responded best to the lower concentration of mycelium.

Bayberry, beach plum, and common juniper showed little difference between treated and control areas. Beach rose was the most erratic in its response. One-third of the plants died, but those that survived had larger and greener leaves and more fruit than untreated plants. Those treated with the least mycelium, or untreated, appear to be surviving best. It is interesting to note that at least three of these species, which were indifferent to the treatment, are characteristically adapted to low-nutrient sandy sites, especially coastal dunes.

These results indicate that treatment with aged and fresh mycelium improves the vigor and growth of many woody ornamental plants. It would appear that mycelium incorporated into sandy or sandy loam soil at the time of planting or six weeks in advance of planting provides a valuable fertilizer substitute and soil amendment in land reclamation.

*Terramycin®, itaconic acid, 31A citric mycelium and C200 citric mycelium mixture.

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Back cover: Nutrient effect of composted mycelium is evident in (top to bottom) mountain laurel, fragrant sumac and winterberry grown at the Connecticut Arboretum.

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