Soil Microbial Processes and Dynamics: Their Importance to Effective Reclamation

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Abstract

The dynamics and activity of the soil microflora are regulated by complex and synergistic interactions between carbon and nutrient availabilities, microfaunal grazing, and abiotic constraints. Evaluation of reclamation success must focus on measuring the occurrence and strengths of the interactions among these factors at various ecological levels. Microbial species abundance distributions and the annual variability parameter are appropriate methods for examining short and long term recovery rates and community stability. The ratio of microbial respiration (R) to microbial biomass (B), termed the metabolic quotient, links microbial activity and organic matter production, and should provide the best approximation of overall system recovery and stability.

Introduction

Disturbance events are now recognized as important factors affecting community structure, ecosystem functioning, and landscape patterns (Pickett and White 1985a; Pickett et al. 1989). In natural systems disturbances occur at various spatial and temporal scales with the effects dependent upon parameters such as intensity, frequency of occurrence, size of the event, and and lifespans of the component organisms (Pickett and White 1985b). What is common to all disturbances are the alterations of resource availability and community structure. Differences in system response to disturbance are a consequence of the degree to which the before mentioned parameters are altered. Since disturbances can affect all levels of ecological organization, which parameters are useful in assessing the responses and recovery of the biotic component will depend upon the hierarchical level at which the investigator is examining the response (Table 1). Depending upon whether one is interested in population versus ecosystem level response to disturbance will dictate which components are measured and the ability of the chosen parameter to examine short and long term perturbation effects. Pickett et al. (1989) have argued that an understanding of disturbance effects can only be revealed through an examination of the "minimal structure" of the ecological level that is being studied. By "minimal structure" they are referring to: (1) organization of entities into the observed units (e.g., individuals, populations, communities); (2) coordination of the interactions between units; (3) regulating function of the unit; (4) information flow between units (i.e., nutrients and energy); and (5) interactions between levels. With this approach, disturbance becomes a change in the minimal structure. Events which result in changes in species composition of a community but have no effect on the functioning of the observed unit are considered as configurational and do not constitute a change in the minimal structure of the unit.

Table 1. — Disturbance effects at hierarchical levels of ecological organization

Level	Component	Response
Individual	Biomass	Growth
Population	Density	Extinction
Community	Abundance distributions	Structure
Ecosystem	Functional groups	Resilience
Landscape	Fluxes of organisms	Stability

From Pickett et al. (1989).

Depending on the scale and severity of the disturbance, Odum (1985) has indicated that in most instances there is an initial negative change in the structure and composition of the component communities. At the ecosystem level nutrient cycling and efficient resource utilization is decreased, and this may result in changes in succession trajectories. The latter is more likely to occur when the disturbance is large or out of the realm of naturally occurring disturbances (e.g., surface mining). Although there is an initial decline in the relative abundances of component species, small scale disturbance usually increases system heterogeneity. Moreover, new patches for colonization are created thus increasing the overall diversity and species richness of the habitat (Denslow 1985). Species which are characterized as "r" or ruderal strategists (Grime 1979) are more likely to colonize these

open patches and would not occur in a specific habitat without a disturbance regime since they are generally poor competitors (Tilman 1982). Disturbances which are viewed as catastrophic events (e.g., surface mining) impose a functional homogeneity on the system. From a surface mine reclamation perspective, the questions become: (1) at which level should the effort be made to reestablish the minimal structure of the functioning unit; (2) how does one go about reestablishing the necessary structure and associated functions; and (3) what parameters should be measured to monitor success. This paper will focus specifically on the impact of surface mining and subsequent reclamation efforts on the microbial component of the system, and will examine alternative approaches to measuring reclamation success.

Ecosystem Functioning and Microbial Activity

The processes of decomposition and subsequent nutrient cycling are intimately associated with the activity of the soil microflora (bacteria and fungi) and the microfauna (protozoa, nematodes, and microarthropods). Through their acquisition of carbon and nutrients for growth, fungal and bacterial biomass in soils constitutes an important sink and source of essential plant nutrients. The mechanisms by which nutrients immobilized in microbial biomass become available to plants are not clearly understood; however, increasing evidence indicates that microbial grazing by the microfauna is important in mineralization (e.g., Moore et al. 1988). These interactions are particularly important during litter decomposition (Coleman et al. 1988), an essential aspect of soil formation, and in the root region of plants (Ingham 1986 a,b). Several studies have shown that microbial grazing in the root region can increase plant growth (e.g., Coleman et al. 1984). These increases in plant growth are attributed to greater mineralization rates of nitrogen as a consequence of microfaunal grazing. Microfaunal grazing affects mineralization rates by: (1) altering the composition of the microbial community; (2) affecting microbial growth; and (3) altering nutrient availability (Newell 1984; Moore 1988).

The importance of microorganisms in the reclamation process has been previously stressed by Cundell (1977), Jurgensen (1978), and Parkinson (1979). Because of their ability to fix atmospheric nitrogen and their involvement in converting soil nitrogen into various forms, soil bacteria are crucial components of the nitrogen cycle (Paul and Clark 1989). Because fungi constitute the majority of the microbial biomass in terrestrial systems and are the major decomposing organisms, most studies on the effects of surface mining andd reclamation have focused only on the the fungal component of the system. However, both bacteria and fungi constitute important pools of carbon and nutrients in soilsq, and recent models have emphasized the need to include bacteria, fungi, and the mircofauna in below ground food webs (e.g., Hunt et al. 1987; Moore 1988). With respect to reclamation efforts of highly disturbed soils, simply elucidating the microbial component of the system (i.e., generating species lists) following the application of amendments over time will not provide the information necessary to evaluate the success of the effort. Studies of reclamation efforts will have to focus on the structure and functioning of the microbial component of the system as modified by the activity of the soil microfauna. The importance of microfloral-microfaunal interactions in a reclamation effort has not previously been addressed. However, evidence from undisturbed systems would suggest that these linkages are critical to system functioning and development.

Microbial Community Structure

The choice of a microbial community descriptor to examine the short and long term effects of stripmine reclamation efforts is not a trivial matter. At the community level various characteristics and parameters can be obtained and calculated that will provide the investigator the ability to examine changes in the microbial assemblages with time and with reclamation effort (Table 2). Which parameters are most useful in describing changes in community organization and structure is open for discussion. As stated previously the majority of the research concerning microbial community structure in reclaimed sites has concentrated on the fungal component. Analysis of bacterial assemblage data at the community level has been descriptive due to the difficulties and time constraints of current bacterial taxonomy. Most studies discuss the densities of bacterial colony forming units of various bacterial groups or key genera as affected by the reclamation protocol (e.g., Fresquez and Sabey 1989).

Table 2. — Parameters used to access redevelopment of microbial activity in reclaimed surface mine spoils

Level	Parameter
Population	Number of propagules Species frequencies
Community	Species composition Spatial heterogeneity Species diversity Species abundances Annual variability
Ecosystem and landscape	Decomposition rates Respiration Enzyme activity Microbial biomass carbon/ total organic carbon Metabolic coefficient (qCO_2)

Once a surface mined site is recontoured, previously stockpiled topsoil or another source of organic matter is usually applied to the site (see Visser 1985 for a review). Even under the best of conditions, the microbial richness and inherent system heterogeneity has been destroyed.

Allen and MacMahon (1985) suggested that the loss of spatial heterogeneity between the soil fungi, organic matter, and nutrient pools was a key factor affecting reclamation success on a strip-mined site in Wyoming. Fungal genera on the reclaimed site were broadly dispersed across the landscape, an indication of site homogeneity, while on the undisturbed area fungal taxa were organized around shrubs (Table 3). The loss of spatial heterogeneity within the soil and plant community affected both the species composition and structure of these fungal assemblages. Allen and MacMahon (1985) suggest that the loss of fungal spatial heterogeneity and community structure could be used to measure the success of a reclamation effort.

Table 3. — Spatial heterogeneity of fungal species in topsoiled spoil

	Sorensen's Index of Similarity		
Pot size	**4 cm²**	**40cm²**	**4m**
		Undisturbed	
4cm²	1	0.25	0.30
40cm²		1	0.22
		Topsoil Amended	
4cm²	1	0.43	0.50
40cm²		1	0.50

Data from Allen and MacMahon (1985); values represent the percent of similarity in fungal species composition between the various size sampling areas.

Stanton et al. (1981) proposed that changes in microbial species composition during reclamation may be as important as changes in microbial densities. This statement implies that shifts in microbial species composition will affect the structure and function of the observed microbial assemblages. Compositional changes alone may not affect either structure or function if the changes are compensatory. Fluctuations in the composition of microbial assemblages and their respective frequencies of occurrence have been used by numerous investigators to examine the outcome of a reclamation effort (e.g., Visser et al. 1983; Fresquez and Sabey 1989). For fungi the number of fungal propagules and the number of fungal taxa are reduced following mining. The microbial community may increase after the addition of organic matter and as the vegetation cover increases (e.g., Visser 1984). While some tentative conclusions concerning reclamation success can be obtained by comparing species lists from reclaimed vs. undisturbed sites, this approach does not allow the investigator to address the central issue of microbial community structure. Elucidating the structure of microbial assemblages provides a means for determining the relationships among component species as they may affect key processes such as decomposition and nutrient cycling. By examining the structure of microbial assemblages, one hopes to be able to relate change in structure with change in function, which is a more difficult parameter to measure and understand.

Measures of diversity, which include species abundance measures and diversity indices, have in general been used to examine community structure and to indicate the "well being" of ecological systems (Magurran 1988). These indices utilize both the number of species within a community and the distribution of individuals among species (evenness). By calculating a diversity index, large data sets of species frequencies can be condensed into one value for comparison between sampling times or treatments. Care should be exercised, however, when inferring changes in community stability from a diversity index since the parameters combine a number of variables that actually characterize community structure (Ludwig and Reynolds 1989). By first understanding the limitations of these indices, one can utilize a diversity index to examine large shifts in community structure. The investigator should be aware that two communities can have the same diversity index, but contain either different numbers of species or different evenness values. Although the communities are structurally different, the diversity indices are similar.

Dennis and Fresquez (1989) and Fresquez et al. (1986) have utilized the Shannon index of diversity and associated evenness parameter to examine the effects of either topsoil or sewage sludge on diversity indices of soil fungal assemblages. For a surface mine site in New Mexico, diversity of the soil fungal assemblage was higher in the undisturbed areas than in a site which did not have topsoil applied but which did receive fertilizer and 4.5 metric tons of hay/ha (Table 4). Even after 8 years, diversity and the evenness parameter were low in the nontopsoiled area as compared with the undisturbed site. A second and younger series of areas that received the same hay and fertilizer treatment plus a 20-30 cm layer of topsoil was examined over a 4-year period (Table 4). For the topsoil plots, fungal diversity and evenness increased compared to those sites not receiving topsoil. Based on the diversity indices, evenness, and species composition of the fungal assemblages, Fresquez et al. (1986) indicated that the microbial component may have begun to stabilize in the topsoil treatment by four years following application. With increasing application rates of sewage sludge applied to a degraded range land, Fresquez and Dennis (1990) found that fungal diversity declined by 2 months after application. (Table 5). Evenness declined in a similar manner. In the succeeding three years of the study, diversity was lowest in the plots receiving the highest sewage application with diversity similar in the other treated plots and the control. The results of Fresquez and Dennis (1990) indicate that initial application rates of sewage will have a long term impact on the resultant structure of the soil fungal community.

Table 4. — Diversity indices for fungal assemblages from topsoil amended surface mine soil

Treatment	Reclamation age (yrs)	Shannon (H) Diversity Index
Undisturbed		1.09
Nonamended	6	0.48
	7	0.65
	8	0.45
Topsoil amended	1	0.86
	2	0.88
	4	0.75

Data from Fresquez et al. (1986).

Table 5. — Shannon diversity indices for fungal assemblages in sewage amended soil

Sludge application (Mg/ha)	Time from reclamation			
	2 mon	1 yr	2 yr	3 yr
0	0.94	0.89	1.03	0.72
23	0.69	0.87	1.01	0.89
45	0.51	0.97	0.84	0.75
90	0.24	0.66	0.50	0.61

Data from Fresquez et al. (1986).

Species abundance distributions provide an alternative to diversity indices in describing community structure. Microbial data sets which contain information on the number of species and their relative abundances are used to generate these distributions. Magurran (1988) has emphasized that a species abundance distribution utilizes all the community information and is the most complete mathematical description of the data. Four models have been developed which describe species abundance distributions (Fig. 1). These distributions are characterized by specific biological parameters which account for the form of the distributions (May 1975, 1981) (Table 6). With species abundance distributions, changes in the structure of the microgbial community as a consequence of reclamation can be followed over time. Tilman (1982) observed marked changes in plant species abundance patterns in response to fertilizer addition of a permanent pasture.

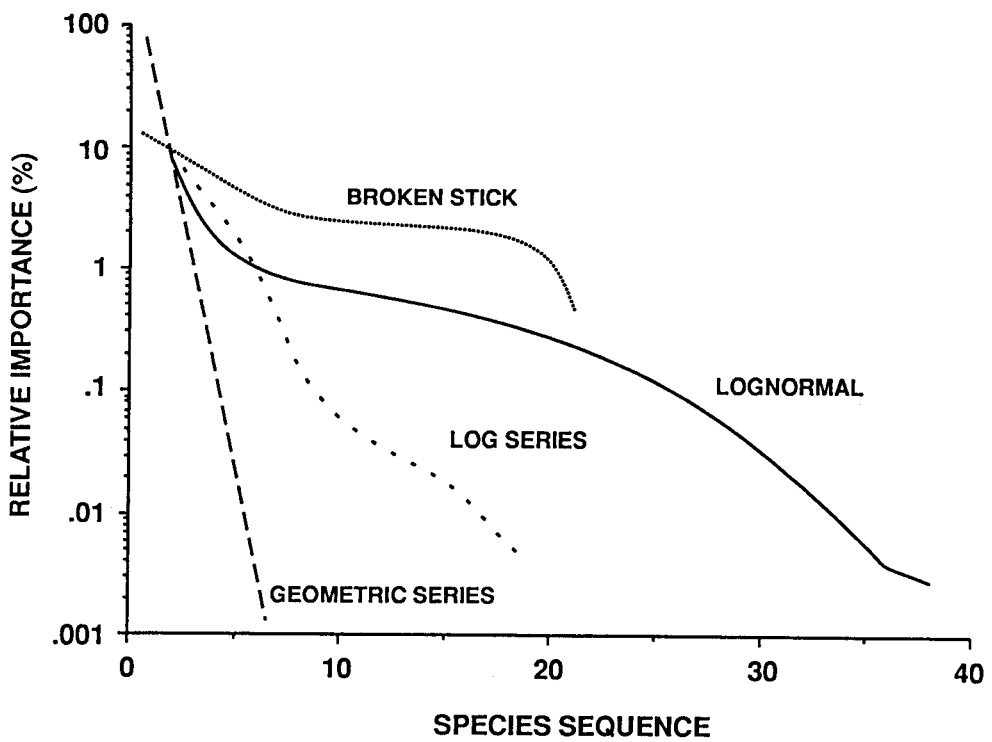

Figure 1. — Species abundance distribution models.

Table 6. — Parameters which characterize the four major species abundance models

Geometric	Log series	Lognormal	Broken stick
Dominant species preempt a portion of limiting resource	Small assemblage of species	Large and varied community	Subdivision of niche space is random and simultanous
Abundance is proportional to amount of resource utilized	Propagules arrive at random intervals	Abundances governed by many independent factors	Abundances governed by even division of major resource
Associated with species poor habitats	Small number of abundant species, large number of rare species	Resource utilization is multidimensional	Characterized solely by the number of species
Propagules arrive at regular intervals of time	Abundances governed by one or a few factors	Equilibrium community	
Nonequilibrium community	Nonequilibrium community		

Information from Magurran (1988) and May (1975, 1981).

Zak and Parkinson (1984) and Zak (1988) have used this approach to examine the effects of nutrient and organic matter application to an extracted oil sands spoil and a subalpine coal mine spoil, in Alberta, Canada, on the structure of root surface fungal assemblages of *Agropyron trachycaulum* (Link, Malte.), slender wheatgrass. Zak and Parkinson (1984) originally showed that the fungal species abundance distributions were not described by the truncated lognormal model. Reexamining the initial 2-year oil sands data and including a fourth year, Zak (1988) found that the forms of the distributions in the control, peat, and fertilizer amended plots were best described by a logarithmic function over the 4-year period. Assemblages from the sewage treated plots differed in that they were best described by an exponential function, and may approximate the geometric model (Zak 1990). The high nutrient concentrations in the sewage shifted the distributions from logseries to geometric (Zak 1990). Although fungal species composition and species numbers associated with the grass root surfaces were altered by the reclamation program (Zak and Parkinson 1984), except in the sewage plots, the structure of these assemblages was not modified by either organic matter addition (peat) or by fertilizer application. The fungi associated with the grass roots in the amended oil sands spoil, except in the sewage treated plots, represent stable assemblages that differ only in composition. The observed structure of these assemblages reflect the paucity of fungal species in the spoil and may be a consequence of the initial homogeneity of the system.

Utilizing the fungal data set from decomposing timothy litter on a subalpine mine spoil of differing reclamation ages (1 to 7 years), Zak (1990) observed that species abundance distributions from the original material were best described by the geometric model. Following field placement of 1, 2, and 4 months, the distributions were found not to change with decomposition age or time from reclamation. At all sampling times, the species abundance distributions were best described by either the geometric of logseries models. The forms of the timothy litter fungal species abundance distributions would indicate that disturbance effects were still evident 7 years after reclamation (Zak 1990).

Calculating species abundance distributions from the data set from Fresquez et al. (1986), we observed that the structure of these fungal assemblages did not change after the first year following topsoil application (Fig. 2A) (Table 7). While both the geometric and logseries models could describe the assemblage from the undisturbed area, the geometric was a better fit. The geometric series is characteristic of species - poor habitats where the abundance pattern is governed by one major factor. The logseries is similar to the geometric, but for which abundances may be governed by more than one factor. The change in the species abundance distribution after topsoil application may reflect the increased heterogeneity of the system. Although fungal diversity was found to change with reclamation age (Fresquez et al. 1986), the structure of these assemblages as determined by species abundance distributions did not differ with time. For those sites which did not receive topsoil, the structure of the fungal assemblages was also found not to change after 6, 7, and 8 years (Fig. 2B). The structure of the assemblages was best described by the logseries model at all sampling times (Table 7). The assemblage from the topsoil amended and control sites represent depauperate systems that are very stable. The addition of sewage sludge at 23, 45, and 90 mg/ha to a degraded range land in west-central New Mexico (Fresquez and Dennis 1990) also shifted the species abundance distribution for the fungal assemblage from this site from geometric to logseries (Fig. 3, Table 8). Although diversity indices and numbers of species declined with increasing sewage application, at the 2-month sampling period the structure of the fungal community was not altered. Except for the 2-month sample in the undistributed area, which was best described by the geometric model, all other distributions over the 3-year period were best described by the logseries (Table 8).

These results may imply that other factors besides nutrients and organic matter may be responsible for the structure of these assemblages. As with most arid systems, moisture availability may be the key regulator affecting structure and function (Whitford 1989).

Recent work by Woolhouse and Harmsen (1987) has suggested that the following community descriptor:

Annual variability (AV) = Variance (log N_i-log N_j)

where N_i = the density of species 1 at time i and N_j = the density of species 1 at time j, may prove to be useful in examining the stability of the microbial component following reclamation. The parameter focuses on the stability in the densities of component species over time. While the descriptor cannot shed light on the underlying mechanisms for or against stability, AV can be used to examine the relative stability of microbial assemblages. The validity for employing this parameter as a measure of stability is based on the assumptions that (1) the densities of component species will not fluctuate widely over time; and (2) although individual species densities will change over a year, the pattern is repeated from year to year. Calculating the AV parameter from the fungal decomposition study of Durall (1984) on a subalpine-coal mine spoil, Zak (1990) found that during decomposition of timothy litter over a 4-month period, the AV parameter for the fungal assemblages on the litter was lowest in the oldest reclaimed site (7 years). In the more recently reclaimed sites, the parameter increased with decomposition age.

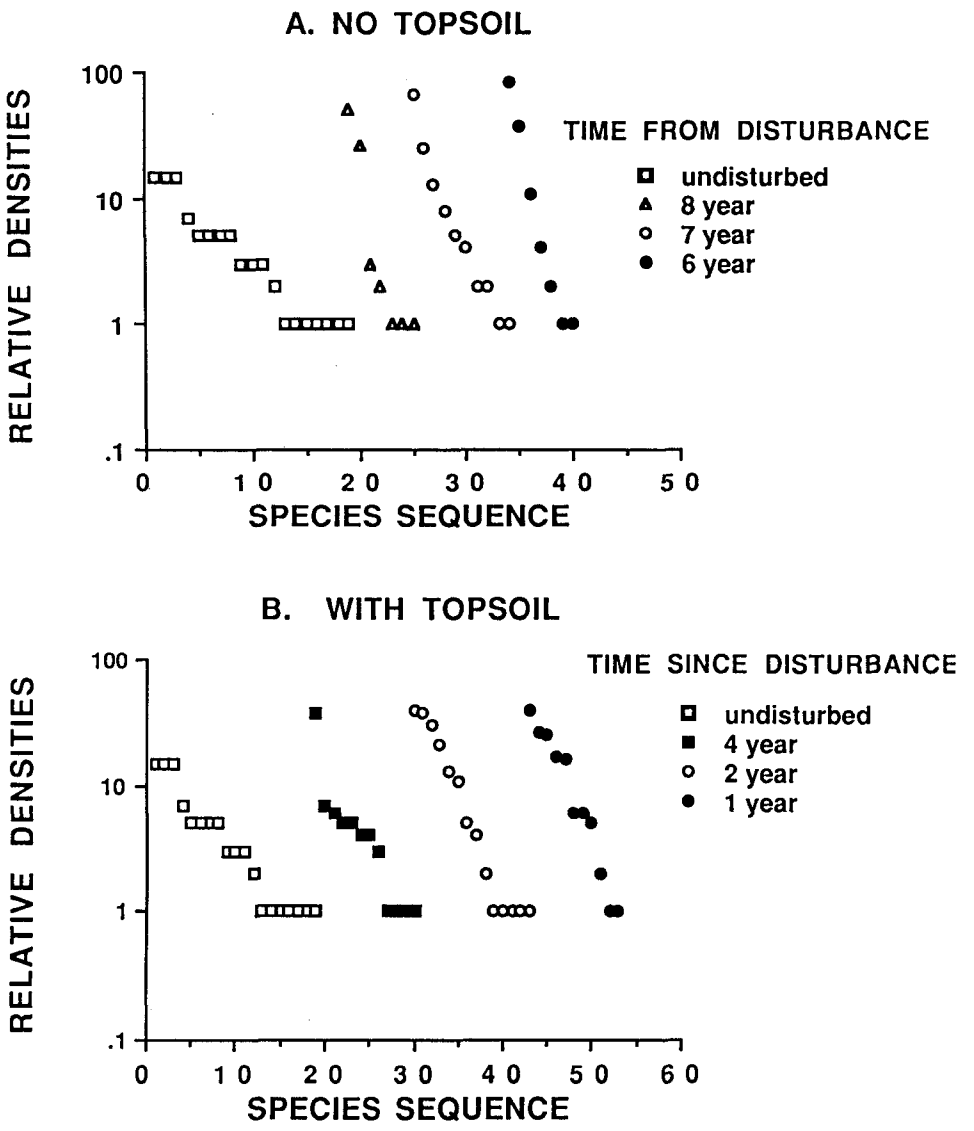

Figure 2. — Effects of topsoil application on fungal species abundance distributions from a surface mine spoil (original data from Fresquez et al. 1986).

Table 7. — Effects of topsoil addition on fungal species abundance distributions

Treatment (time from reclamation (yrs))	Model	P Values
Undisturbed	Geometric	>0.99
	Logseries	>0.30
Topsoil		
1	Geometric	>0.30
	Logseries	>0.50
2	Geometric	>0.80
	Logseries	>0.90
3	Geometric	>0.01
	Logseries	>0.20
Nonamended		
6	Geometric	>0.20
	Logseries	>0.50
7	Geometric	>0.05
	Logseries	>0.99
8	Geometric	>0.02
	Logseries	>0.30

P values indicate probability that Geometric or Logseries species abundance models fit observed data.

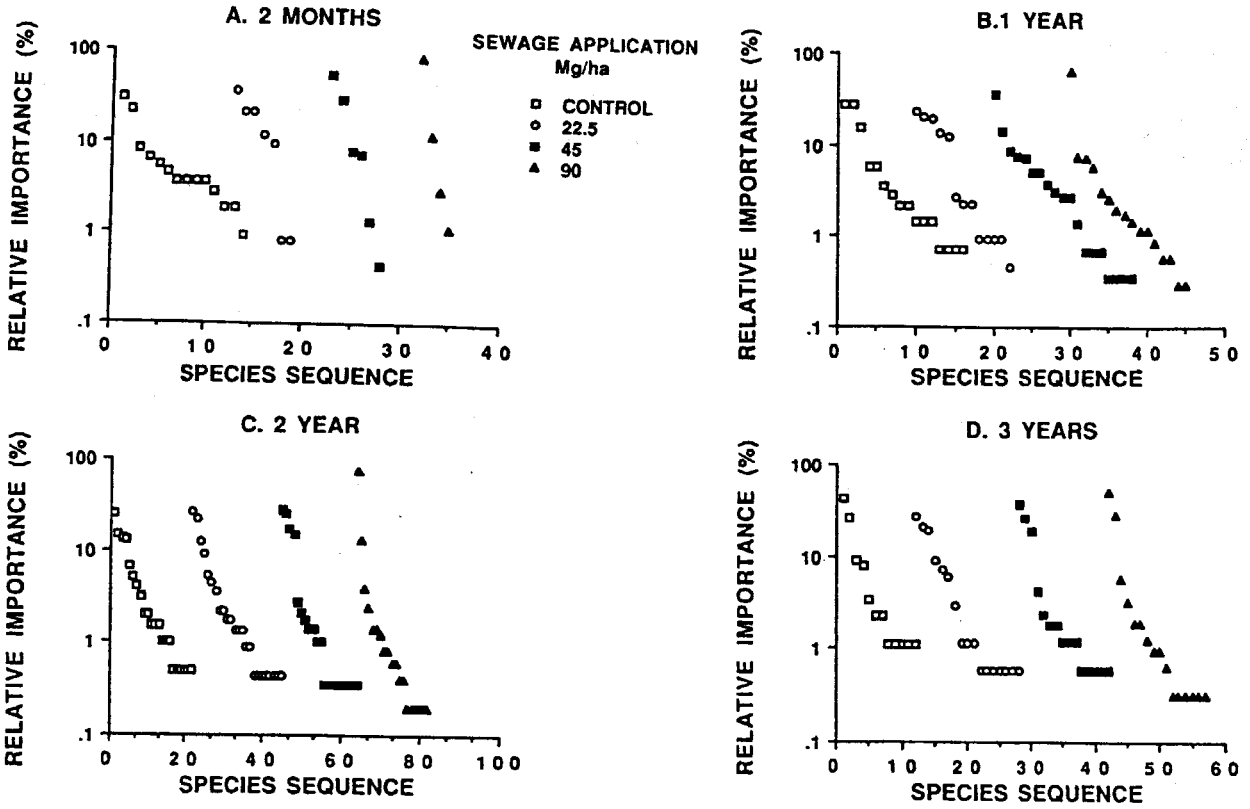

Figure 3. — Fungal species abundance distributions from a dergraded rangeland soil amended with varying application rates of sewage sludge (original data from Fresquez and Dennis 1990).

Table 8. — Effects of sewage sludge on fungal species abundance distributions

Treatment (Mg/ha)	Model	Time from reclamation 2 mon	1 yr	2 yr	3 yr
Control	Geometric	>0.50	>0.10	>0.20	>0.02
	Logseries	>0.20	>0.70	>0.80	>0.50
23	Geometric	>0.001	>0.02	>0.0001	>0.10
	Logseries	>0.01	>0.70	>0.90	>0.30
45	Geometric	>0.01	>0.01	>0.001	>0.001
	Logseries	>0.80	>0.80	>0.30	>0.30
90	Geometric	>0.02	>0.001	>0.001	>0.001
	Logseries	>0.70	>0.90	>0.50	>0.50

P values indicate probability that Geometric or Logseries species abundance models fit observed data.

The AV parameter for the mine site in New Mexico examined by Fresquez et al. (1986) did not change for 3 years after topsoil application (Fig. 4). By the end of the fourth year, the AV was lower compared with the previous years. Whether the decrease indicated the beginning of a stable system cannot be evaluated. For the area which did not receive topsoil, the AV value was lower 8 years after the initial reclamation effort than for the topsoil amended site (Fig. 4). These results indicate that while mine spoils may have a reduced number of fungal species, those that do function within the disturbed system represent a stable assemblage of species that is adapted to the abiotic constraints of the habitat. Moreover, unless nutrients and organic matter are added to most mine spoils, the soil fungal community that establishes after the disturbance will remain unchanged for long periods of time.

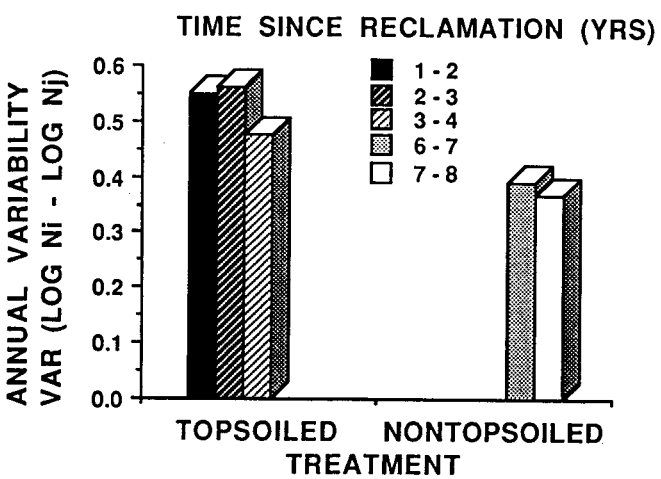

Figure 4. — Stability of the soil fungal community in topsoiled vs. nontopsoil amended surface mine spoil as measured by the annual variability parameter (original data from Fresquez et al. 1986).

Sewage sludge has been examined in several studies as a source of nutrients and organic matter for ameliorating the adverse effects of surface mine spoils and as a primer for initiating the activity of the soil microflora in these systems (Visser 1985). The rates at which sludge is applied are usually based on plant response without considering the effects on the soil microflora. The development of the soil microbial component of terrestrial systems is dependent on soil organic matter levels and inputs via plant litter in order to maintain the microbial biomass necessary for efficient decomposition and nutrient mineralization. The initial rates of sewage sludge addition become critical in maintaining the microbial component of the system prior to establishing sufficient inputs of carbon and nutrients via plant litter. The choice of sewage application rate can be based on the assessment of plant growth response coupled with the determination of the impact of the sewage application on the AV value for the soil fungal assemblage. Ideally, the amount of sewage applied to a spoil should significantly increase plant growth without causing large fluctuations in AV values. Excessive nutrient inputs to a system can lead to destablization (e.g., Tilman 1982).

Based on plant growth response (Fresquez et al. 1990 a, b) coupled to diversity indices of the soil fungal community, Fresquez and Dennis (1990) found that an application rate of 45 mg/ha as compared to 23 or 90 mg/ha maximized growth without significantly affecting microbial diversity over a 3-year period. All sewage application rates in the study of Fresquez and Dennis (1990) increased the AV value for the fungal assemblages between the first and second growing seasons, with the highest variance found in the 90 mg/ha treatment (Fig. 5). Comparing years one and two, the variance parameter declined in all treatments with the highest application rate being most similar to the value observed for the control area. Even in the control there was a considerable change in the AV value over the first two years of the study. Contrary to the original results of the study by Fresquez and Dennis (1990), the fungal assemblage in the highest sewage application treatment was as stable as that observed in the control plots by the fourth growing season. For the specific system studied by Fresquez and Dennis (1990), the lowest application rate decreased fungal stability while the highest rates resulted in

increased plant growth and the maintenance of fungal community stability. The species composition in the 90 mg/ha application (highest) was least similar to the control site over the 4-year period (Table 9). The high nutrient and subsequent litter inputs changed the interactions between fungal species and possible microfaunal regulators, allowing a new assemblage of fungal species to become dominant. Although fungal species composition in the high sewage plot differed from the control area, the structure of the two assemblages was similar.

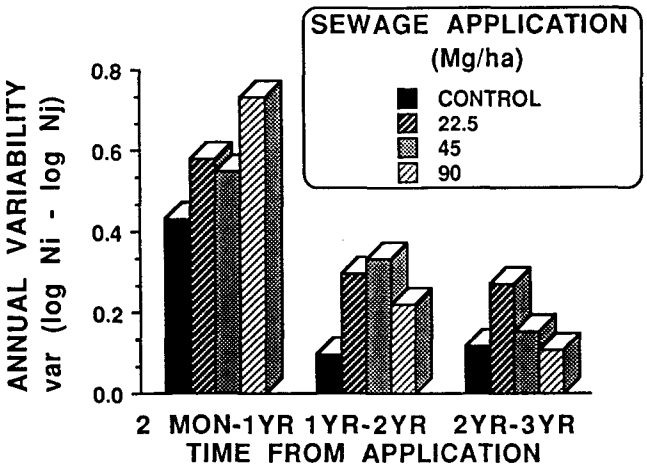

Figure 5. — Changes in fungal community stability following application of sewage sludge application to a degraded rangeland soil as measured by the annual variability parameter (original data from Fresquez and Dennis 1990).

Based on the analyses of the previous fungal data sets, the choice of a community descriptor will significantly affect the conclusion reached concerning the effects of reclamation on the soil microbial community. Comparisons of species lists, while important, do not convey any information concerning how these species are organized into a community or assemblage, and the potential relationship to function. Ultimately we are interested in determining the stability in the composition and structure of the microbial community in space and time as these two components affect function. The previous examples illustrate that at the level of the community, reclamation efforts may impose a stability on the microbial component that may be long term and that may lead to an undesirable recovery sequence. Based on the observed structure of the microbial community, we can infer the probability of reclamation success.

Microbial Processes and Activity

Through its role in decomposition and nutrient cycling, microbial activity is a key factor affecting the functioning of all terrestrial systems. Measurement of process rates governed by the soil microflora and the general metabolic

Table 9. — Sorensen's similarity coefficients for the fungal assemblages from nonamended and sewage amended soil

Treatment (Mg/ha)	2 months	4 years
	0 23 45 90	0 23 45 90
0	38 30 11	76 67 57
23	77 73	75 67
45	80	71

Data from Fresquez and Dennis (1990).

activities of these organisms can therefore be used to address the question of system "health and stability." As with examining reclamation success via microbial community dynamics, several approaches have been utilized to evaluate reclamation efforts, each with varying degrees of success (Table 2).

Decomposition potential is the simplest parameter with which to estimate microbial activity in surface mine spoils. Visser (1985) found that decomposition rates in two mine spoils (prairie grassland and subalpine coal mine) and associated undisturbed areas were dependent upon the material examined, filter paper vs. wood. Over the short term no differences in decomposition rates were detected between disturbed or undisturbed areas. After 33 months in the field, however, decay rates were greater in the disturbed areas than on the control sites (Table 10). Durall et al. (1985) also reported higher decomposition rates of timothy litter on a 1-year-old reclaimed subalpine coal mine site as compared to the 3- and 7-year-old reclaimed areas (Table 11). The low level of available nutrients in the 7-year-old site, resulting from a large amount of undecomposed litter, retarded decomposition rates on this reclaimed site with time. Durall et al. (1985) emphasized that management practices will need to stimulate decomposition of those reclaimed sites where grass litter is increasing so as not to retard long term decomposition potential. The decline in decomposition rates would suggest that the linkages between decomposition and primary production in this system were breaking down.

Table 10. — Percent mass loss of filter paper in a prairie and subalpine surface mine spoil

	Grassland		Subalpine	
Months (no.)	Undisturbed	Disturbed	Undisturbed	Disturbed
6	1	0	0	15
12	1	3	1	10
24	1	49	16	23
44	18	99	39	72

Table 11.—Percent mass loss of timothy litter on different aged, reclaimed subalpine coal mine spoil

Months (no.)	Reclamation age 1 yr	3 yr	7 yr
4	52	49	50
13	75	62	60
16	82	63	62

Soil or litter respiration rates (e.g., Durall 1984) and soil enzyme activity (e.g., Fresquez et al. 1987) have also been followed in mine spoils as an alternative approach to estimating microbial activity. Cundell (1977) originally advocated the use of respiration rates and enzyme activity to monitor reclamation efforts. Visser (1985) reported significantly lower CO_2 efflux from disturbed vs. undisturbed grassland and subalpine, surface mine spoils. She attributed the lower activity in the spoil to the loss of microbial biomass and soil organic matter. For surface mine spoils varying in age and vegetation in West Virginia, Stroo and Jencks (1982) observed that respiration rates were lowest for barren sites and highest on the youngest amended sites (3 to 5 yrs after reclamation) which were vegetated with grasses and legumes (Table 12). While microbial activity was high on the younger sites as a consequence of the mulch and fertilizer addition, respiration rates and organic matter content generally declined over time. Only in one 17-year-old locust site were respiration rates not significantly different from values detected in the native soils. The decline in respiration rates suggests that the necessary linkages between microbial acitvity, plant growth, and decomposition were not present in the majority of these sites. Stroo and Jencks (1982) emphasized that reclamation practices at the time would not generate stable systems over the long term.

Table 12.—Spoil respiration rates (μO_2/g/h) for vegetated and nonvegetated surface mine spoils of varying ages

Site characteristic	Age (years)	Respiration Rate
Barren	17	0.58
Black locust	11	0.84a
	20	1.96b
Grass and legume	3	5.17a
	17	2.07b
Native site		2.77

For each site, letters differ at P=0.05; data from Stroo and Jencks (1982).

Enzymatic activity of the associated soil microflora was followed by Stroo and Jencks (1982) in the same mine spoils as described for the respiration studies to examine the potential of the microbial component to carry out decomposition and mineralization. Amalyase activity (carbon utilization) was lowest in the barren sites and highest on the youngest amended sites (Table 13). With increasing time since reclamation, activity declined in both the grass-legume and locust plots. Phosphatase activity was significantly higher in the the native soils compared to any reclaimed site. Stroo and Jencks (1982) indicate that the cycling of phosphorous may be repressed in these systems. The activity level of microbial enzymes in the mine spoils was found to be positively correlated with available carbon and nitrogen. Fresquez et al. (1987) measured microbial enzyme activity in an unamended and topsoil amended mine spoil in the arid Southwest and found that in the nontopsoiled area enzyme activity was low, particularly in the 8-year-old site (Table 14). Three months after topsoil application the activity of most enzymes was not significantly different from the undisturbed area. Microbial activity increased subsequently and peaked 1 to 2 years following the initial reclamation effort. By 4 years after reclamation, enzyme activity was similar to that observed in the undisturbed site.

Table 13.—Enzyme characteristics of vegetated and nonvegetated surface mine spoils of varying age

Site characteristic	Age (yrs)	Amalyase	Phosphatase
Barren	17	0.6	1.2
Black locust	11	1.1a	0.6
	20	3.5b	4.8
Grass and legumes	3	6.5a	2.9
	17	3.6b	2.8
Native soils		4.6	8.6

Amalyase = µm of reducing sugar/100g soil; phasphatase = µm of PNP/g soil/hr. For each site, letters indicate significant differences at P=0.05; data from Stroo and Jencks (1982).

Table 14. — Soil enzyme activities in surface mined spoils with and without topsoil

Treatment (Time from reclamation (yrs))	Nitrogenase (nm C2H4)	Phosphatase (µg PNP)	Cellulase (µg glucose)
Undisturbed	204a	368a	52bc
No topsoil			
6	809bd	1561c	32cd
7	21c	885b	4e
8	9c	698b	11de
Topsoil			
1	367a	379a	161a
2	1774d	1018bc	91ab
4	208a	325a	35bcd

Data from Fresquez et al. (1987). Within a column, values followed by the same letter do not differ at P=0.05.

Some organic matter and nitrogen amendment should be added to most mine spoils to prime the microbial component. Lindemann et al. (1989) examined the effects of various topsoil and spoil mixtures with or without sewage sludge or sorgum straw on nitrogen mineralization rates (Table 15). For unamended soil and spoil mixtures, nitrogen mineralization declined as the proportion of spoil increased. These results emphasize the importance of the microbial biomass in the immobilization of nutrients. For the sewage amended mixtures, mineralization was highest at the intermediate levels of soil and spoil. The increased nutrients may have compensated for the lower microbial biomass in these soil:spoil mixtures.

Table 15.—Nitrogen mineralization in mixtures of topsoil and spoil material amended with sewage sludge

Treatment	Net organic N mineralization
	Percent
Topsoil only	8.2
Soil:spoil (80:20)	7.1
Soil:spoil (60:40)	6.1
Soil:spoil (20:80)	4.2
Topsoil + sludge	9.8
Soil:spoil + sludge (80:20)	9.4
Soil:spoil + sludge (60:40)	10.1
Soil:spoil + sludge (20:80)	8.4

In some cases microbial biomass, respiration rates and enzyme activity on vegetated, reclaimed sites may recover to near predisturbance levels within a short period of time following reclamation. Insam and Domsch (1988) have argued that the problem with these single microbial parameters as descriptors of reclamation success or system development is that while the level at which they are measured may achieve some stability, they cannot assess the stability and development of the linkages between levels that are necessary for total system stability. This argument supports the minimal structure hypothesis of Pickett et al. (1989) which emphasizes the importance of linkages between levels in examining the impact of disturbance. For measuring system stability, descriptors should link carbon and nutrient flow through the microbial component and organic matter accumulation. Insam and Domsch (1988) proposed that for a steady state to be achieved, there should be a equilibrium level for the ratio of:

$$\frac{\text{microbial biomass carbon } (C_{micro})}{\text{total soil organic carbon } (C_{organic})}$$

If the equilibrium constant is known, then deviations from this value should provide a measure of system stabiity and whether the soil is gaining or losing carbon. Examining a chronosequence of reclaimed agriculture and forest soils in the German Rhineland mining district, Insam and Domsch (1988) reported that annual carbon accumulation was 0.03% during the first 5 years after reclamation and 0.01% for the subsequent 40 years. They calculated an equilibration time of 90 years to achieve a C organic level of 1% in the agricultural site. For the forest site, annual carbon accumulation was 0.59% during the first five years, and 9.05% over the next 40 years. Equilibration was not reached within 46 years. The ratio of C_{micro}/C_{org} declined with time for both sites. While soil organic carbon and microbial biomass taken separately had indicated a steady state by 15 years following reclamation, the ratio of C_{micro}/C_{org} had not reached an equilibrium value even after 50 years (Insam and Domsch 1988). Recently Insam and Haselwandter (1989) have proposed that the metabolic quotient, which is much easier to measure than C_{micro}/C_{org}, is an appropriate integrating parameter with which to measure reclamation success:

$$qCO_2 = \text{Microbial respiration (R)/Microbial biomass (B)}$$

The metabolic quotient is based on the work of Odum (1971), who showed that as long as production is greater than respiration, organic matter and biomass will accumulate in the system and R/B will decrease with time. Two reclaimed sites revegetated with a mixture of grasses and legumes were sampled over a 5-year period to estimated the qCO_2 parameter (Table 16). The qCO2 values for both sites reached a peak 2 years after reclamation and decreased subsequently (Table 16).

Table 16.—Metabolic quotients ($qCO_2 \cdot 10^3$) of two reclaimed sites

Site	Initial	Time from reclamation			
		1 yr	2 yr	3 yr	4yr
I	3.0	3.8	4.2	4.0	1.8
II	4.2	6.1	6.5	6.2	2.5

Data from Insam and Haselwandter (1989).

Summary

Evaluation of reclamation success must focus not only on plant growth response but, as importantly, on determining the dynamics and functional status of the soil microbial community. While at the outset this may seem to be a formidable task, the approaches recently presented by Picket and White (1985 a, b) and Pickett et al. (1989) concerning patch dynamics and disturbances provide a framework within which to address reclamation success from a microbial perspective. However, we should not lose sight of the fact that the microbial component of the soil is intimately linked to primary production through decomposition and nutrient cycling. Ultimately, it is the efficiency of these linkages that we are trying to access. The problem we are confronted with is the difficulty in measuring the linkage strengths between decomposition and primary production and the stability or instability of the interactions. Since we are not able to directly access these system characteristics, methods have been developed at

the community or ecosystem level. Surface mining can impose a functional homogeneity on the system which results in severely altered structure and organization. Thus, this paper addressed two questions: (1) at which ecological level should we be concerned in trying to reestablish a functional unit, and (2) what system parameters should we utilize to measure our success in reestablishing the critical microbial-plant linkages? We also propose parameters that would allow the investigator to determine changes in microbial dynamics and function as they affect system stability.

Management Implications

Based on the minimal structure concept of Pickett et al. (1989), microbial species lists and similarity indices among treatment and times by themselves will not provide the means to address changes in community structure, implied function, and reclamation success. At the community level, parameters which estimate the structure (i.e., numbers of species and their relative abundances) of the microbial assemblages are most appropriate. Of the several approaches that can be used, species abundance distributions (see Magurran 1988 for the procedures to generate these distributions) and the annual variability (AV) parameter (this text) will provide the most complete assessment of community structure and stability. The difficulty with utilizing community descriptors is that they require a considerable investment in time to obtain the species frequency and density data (specifically fungal) required for the calculations. While they provide a theoretical basis for studying reclamation success, the time involved in data collection limits their usefulness as quick methods for evaluation. However, if facilities and expertise are available these parameters should not be overlooked. The paper by Zak and Parkinson (1984) describes the methods to determine fungal frequencies from roots and can be used as a guide for collecting fungal community data. These same approaches can be utilized to examine the plant components of the system.

At the functional level, single parameter measurements (e.g., respiration rates, enzyme activity, decomposition) cannot estimate the long term stability aspects of the system. Variables such as C_{micro}/C_{org} (Insam and Domsch 1987) and qCO_2 (Insam and Haselwandter 1989) will be the most useful since they integrate microbial function, primary production, and decomposition at the ecosystem level. The ratio C_{micro}/C_{org} is predicted to increase over time to some equilibrium level for the system. The direction and rate of change will provide an indication as to the stability of the system. The metabolic quotient qCO_2 may be the easiest functional parameter with which to measure system stability and potential reclamation success. Like the previously described carbon ratio, the qCO_2 value will decrease with time as systems accumulate carbon. Microbial respiration measurements can be assessed by either placing soil in Mason jars with beakers of 1.0 N NaOH solution to trap the CO_2, and titration of the alkali solution to obtain amounts of CO_2 or utilizing automatic CO_2 measuring devises (e.g., infrared gas analyzer). Details of the various procedures can be found in Anderson (1982) and the papers cited in this chapter. Microbial biomass is best determined by the substrate-induced respiration procedure of Anderson and Domasch (1878). Respiration is expressed as CO_2 /h/g dry soil, and converted into microbial biomass (μg C micr/g dry soil) by applying the formula y= 40.04x+0.37 (Insam and Haselwandter 1989), where x is the glucose induced respiration. Once the equipment is obtained and protocols established, this method should allow for effective short and long term monitoring of reclamation success.

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Mycorrhizae and Reclamation Success: Importance and Measurement

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Introduction

A mycorrhiza is a mutualistic association between plants and fungi that affects all terrestrial and some aquatic communities. By affecting the success of individual plants, the association may play a role in the success of reclamation efforts by their presence (improving the growth and fitness of desirable species) or in failure by their absence (enhancing the competitive ability of species with little positive response to the association such as many weedy annuals). Thus, any effort to gauge reclamation success must measure the structure and functioning of the mycorrhizal symbiosis.

The study of mycorrhizae began at the ecosystem level when it was recognized that mycorrhizae aid in the acquisition of critical nutrients for individual plants. Efforts to establish forests in the Caribbean, Australia, and Rhodesia, and shelterbelt plantings in midwestern North America and the Ukrainian region of the Soviet Union demonstrated the importance of ectomycorrhizae to tree survival (e.g., Hacskaylo 1967). More recent work demonstrating that mycorrhizal fungi are sensitive to human perturbations initiated several studies into the demise of mycorrhizae with such anthropogenic disturbances as surface mining, tillage agriculture, and road building (M. Allen 1989). Studies demonstrating that inoculation by mycorrhizae could improve growth and survival of outplanted individuals onto recontoured mined soils (e.g., Aldon 1975; Marx 1975; Carpenter and Allen 1988) clearly indicated a need to understand better when and how to measure adequately mycorrhizal associations in reclaimed sites.

Types of Mycorrhizae and Reclamation Goals

One difficulty in measuring mycorrhizae in natural or human-disturbed communities is differentiating the types of mycorrhizae and developing techniques to characterize them. There are 5 major types of concern to most reclamation efforts. Vesicular-arbuscular mycorrhizae (VA, often referred to as endo-, phycomycetoid, or coenocytic mycorrhizae) are the most prevalent, associating with fungi in the Glomeales (Zygomycete) and forming associations with all grasses, most herbs, and many trees. In this association, the fungus penetrates the cell walls of the root cortex and forms a hyphal matrix radiating out into the surrounding soil. The second most common mycorrhizae type is the ectomycorrhizae. This type is common to many trees important to reclamation, such as pines and oaks. The hyphae of the mycorrhizal fungi (in this case, members of the Ascomycets and Basidiomycetes in addition to members of the genus *Endogone,* a Zygomycete) are found only externally to the root cortical cells but completely encase the short roots in a hyphal mantle. There are three other predominant mycorrhizal types that form a gradient in structure between these two. These include the ericoid mycorrhizae in which the fungus penetrates the cortical cells of ericaceous plants. They form only minimal external hyphae but have a vast array of enzymes enabling them to utilize organic nutrients. They are found in highly organic (peaty) soils, or in soils with high concentrations of heavy metals. The arbutoid mycorrhizae also occasionally penetrate the cells of ericaceous plants but primarily form a hyphal mantle as in the ectomycorrhizae, but with a less extensive covering. Ectendomycorrhizae are formed when the mycorrhizae of pines and other ectomycorrhizal plants form associations with a specialized group fo mycorrhizal fungi that penetrate cell walls and form only a partial mantle.

Other mycorrhizal types of a less general nature include orchid mycorrhizae and monotropoid mycorrhizae. More detail can be found on all types in other books (e.g., Harley and Smith 1983; Allen 1990). These less dominant mycorrhizal types should be considered only if rare and endangered plants forming these types of associations are of concern to the success of reclamation on a given site.

The above descriptions refer to the types of mycorrhizae. Recalling that a mycorrhiza is composed of both a plant and a fungus, that means that both groups of organisms are of concern. As an aid, most plants only form one specific type of mycorrhiza. Thus, knowing the plants of interest can tell the land manager which mycorrhizal types to be concerned within any given community. Relatively comprehensive lists are continually being updated but most plants and their mycorrhizal associates can be found in Trappe (1987) or Harley and Harley (1987). Unfortunately,

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the paper by Harley and Harley (1987) is still the only list to the generic level and it is compounded for Britain. A similar listing of North American plants is needed.

As a generalization, we can suggest that virtually all North American terrestrial plants form mycorrhizae except for annuals in the Chenopodiaceae, Brassicaceae, and Amaranthaceae. Reports of nonmycorrhizal perennials are probably inaccurate because of the difficulties in adequately sampling for mycorrhizae, especially in highly variable habitats (e.g., Allen 1983; E. Allen and M. Allen 1990).

Reclamation Success: A Function of Mycorrhizae?

Do mycorrhizae hasten the rates of reestablishment of desirable plant communities? To test this hypothesis, several research groups have inoculated small plots or individual plants to assess the role of mycorrhizae in reclaiming surface mines following the replacement of the plant growth medium, be that medium spoil (unconsolidated subsoils) or respread topsoil.

Since ectomycorrhizal fungi can be mass produced in culture, they have been used to enhance reclamation of ecosystems where conifers and oaks predominate for several decades. Importantly, these plants approach obligate dependence on mycorrhizae. Without natural or artificial inoculation, these plants simply will not reestablish. Artificial inoculation with selected species found to tolerate the harsh conditions of most reclamation sites enhances the regrowth of many forest systems dramatically. Greater detail on these systems can be found in Marx (1977) and Molina and Trappe (1982). These studies have demonstrated the importance of artificial inoculation of individuals in a reclamation site. In many large scale efforts, such procedures can become very expensive. However, these inoculated forests have value as sources of wood and this may recover much of the reclamation costs. In such systems, considerable efforts have been made to develop mechanical means to plant inoculated trees in easily harvestable rows.

In many habitats, the predominant mycorrhizae are the VA type which cannot be cultured without an actively growing host. Thus, artificial inoculation can only proceed in small patches created by planting inoculated individuals. Also, many of these lands have minimal agricultural value and are primarily reclaimed for wildlife and grazing. In the first study of reclamation of a western arid system that utilized artificial inoculation, Aldon (1975) demonstrated that inoculation with VA mycorrhizae could enhance the survival of *Atriplex canescens.*

In other studies, the potential role of mycorrhizae on the reclamation of VA mycorrhizal dominated disturbed sites was expanded in an important direction. Nicolson (1960) noted that the early successional species of dunes in Scotland, *Salsola kali* (Russian Thistle), was nonmycotrophic. That is, this species does not form mycorrhizae while the later successional species were mycorrhizal. Reeves et al. (1979) and Miller (1979) found similar patterns in disturbed versus undisturbed arid lands in surface mined areas of Colorado and Wyoming. E. Allen and M. Allen (1980) noted that when topsoil containing inoculum was replaced immediately following stripping, mycorrhizal associations were rapidly reestablished with the planted grasses. Alternatively, where topsoil was not replaced, no mycorrhizae were found and the nonmycotrophic weeds persisted for up to 10 years following replanting with no reestablishment of the seeded grasses and shrubs.

E. Allen and M. Allen (1984) hypothesized that mycorrhizae should enhance the succession rates on distrubed areas such as mined lands by enhancing the competitive ability of mycotrophic grasses and shrubs in the presence of nonmycotrophic weeds that naturally colonize these sites. Details of these studies can be found elsewhere and will not be repeated here (see E. Allen 1989; E. Allen and M. Allen 1990; Carpenter and Allen 1988). To summarize, the responses of the dominant plants representing the successional stages can be arranged in continuum (Fig. 1). The species response to mycorrhizae ranged from a negative response of the early successional weeds *Salsola kali* and *Atriplex rosea* to the highly positive responses of *Artiplex canescens* and *Artemisia tridentata.* The important N-fixers, *Purshia tridentata* and *Hedysarum borealis,* depended on both the bacterial and the mycorrhizal fungal symbionts to survive and reproduce. While competitive and facilitative interactions can complicate the successional patterns created by mycorrhizae (E. Allen and M. Allen 1988; E. Allen 1989), in general, the fungal associations are essential to the recovery of these sites.

Measuring Mycorrhizae in Disturbed and Undisturbed Lands

Identifying most mycorrhizae is relatively easy with a minimum of experience. Determining the presence or absence of any given mycorrhizal type is within the capabilities of land managers. The only requirement is a microscope and a lab bench to process samples. Sterile technique is not necessary. Mycorrhizae invade cortical cells and form specific structures that can be readily identified following staining of root segments. Arbuscules, peletons and vesicles are the predominant structures. External mycorrhizae (ecto-, ectendo-, or arbutoid mycorrhizae) are distinguished by the hyphal mantle and hartig net that forms between the cortical cells within a root. These features are well illustrated in other books (e.g., Schenck 1982; Harley and Smith 1983; Allen 1990). In addition, most land grant universities currently have faculty capable of teaching land managers how to identify the symbiosis.

Perhaps the greatest challenge to the study of mycorrhizae is to quantify the association to determine whether the mycorrhiza is present in adequate quantities to be effective in improving the growth and survival of the desired plants. Estimating the activity of mycorrhizae or even the functional

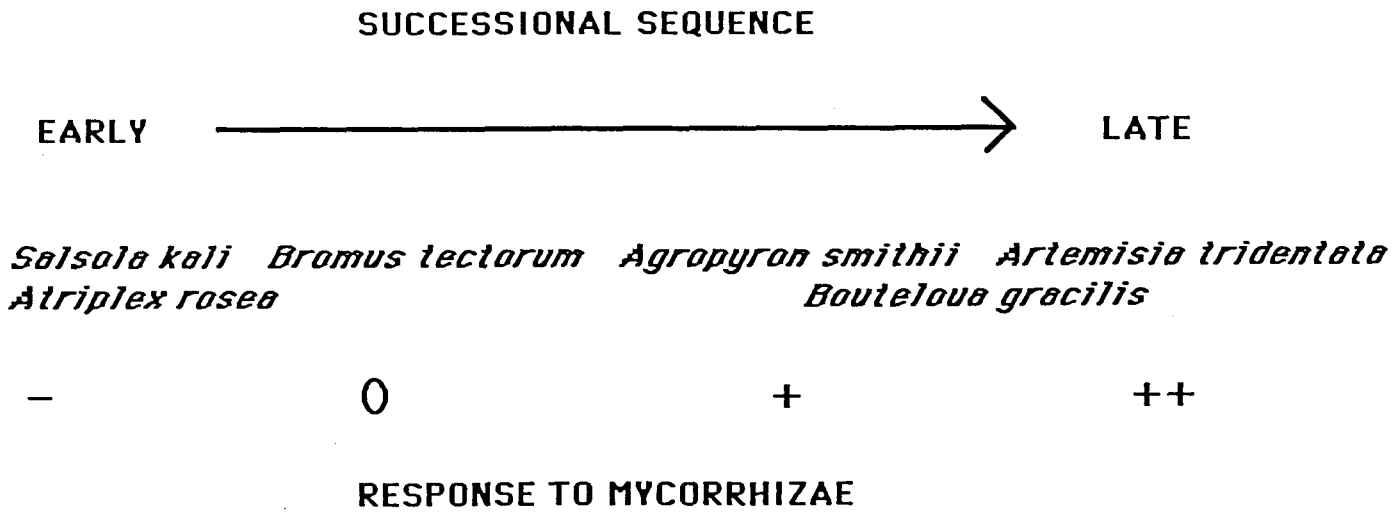

Figure 1.—The relationship between successional sequence of some plant species from mineland shrub-steppe ecosystems and their response to mycorrhizae.

significance of the association within a community is difficult and controversial. Nevertheless, the presence of mycorrhizal associations and of a diversity of fungal species is important and some indications of the presence and the fungal species may be critical to predicting reclamation success.

Techniques for Assessing Mycorrhizal Activity

Several methods currently are used to assess mycorrhizal activity. These include both direct and indirect methods, some simple and some requiring considerable training and time to complete. Also of consideration are the sampling scheme and the types of samples that should be analyzed. Specifically, what sampling regimes are important to the reclamation officer as opposed to researchers currently involved in attempting to understand the specific roles of mycorrhizae in ecosystems? At what scales should samples be taken to assess reclamation success as opposed to determining the fine structure of mycorrhizal dynamics within a single plant or across the entire landscape?

Bioassays of soils for mycorrhizal fungi have been commonly used for a long time. For example, White (1941) determined that a lack of natural ectomycorrhizal inoculum in prairie soils inhibited the establishment of ectomycorrhizal trees. More recently, Reeves et al. (1979) and Schoenberger and Perry (1982) surveyed differing disturbances for their effects on mycorrhizal inoculum. In these techniques, soils to be tested are simply seeded with plants that readily form mycorrhizae and the plants are grown for a given time period in the greenhouse. They are then harvested and the roots stained and assessed for the presence of mycorrhizae by looking for the structures listed above.

There are two indirect techniques for quantifying mycorrhizal activity based on bioassays for mycorrhizal fungi. These have primarily been used to test soils prior to planting to estimate the potential for recovery of mycorrhizae. Mycorrhizal inoculum potential (MIP) was proposed by Moorman and Reeves (1979) as a means to determine the potential for mycorrhizae to reestablish following a disturbance. A similar procedure is called the most probable numbers estimate (MPN) of mycorrhizal fungal densities. This procedure is outlined by Wilson and Trinick (1982) in greater detail. In both of these procedures, a known amount of test soil is mixed into a standard, sterile soil and seeded with a given mycorrhizal plant. After a known period, the plant is harvested and the number of propagules (MPN) or mycorrhizal inoculum potential (MIP) estimated by the percentage of root length infected by the mycorrhizal fungi (for VA mycorrhizae) or by the proportion of root tips infected (ectomycorrhizae).

Several methods are available for direct estimation of mycorrhizal activity. Primarily, this involves determining the percent of the root length containing VA mycorrhizae or the percent of root tips that are ectomycorrhizal using plants collected from the field at differing times following the replacement of the growth medium. Generally, soil cores are taken and the results extrapolated to the surface 10-20 cm of the soil using a known soil volume. To gain a greater understanding of the activity, root length can also be estimated using the same cores. Deeper cores can be used to estimate total mycorrhizal root length per plant or per community. These data give the actual mycorrhizal activity and can best be used to understand the recovery of mycorrhizae of given plants at specific times following reclamation. Examples of these studies giving greater detail of the methods can be found in E. Allen and M. Allen (1980), Allen et al. (1989) and Schenck (1982).

To determine the reproductive potential of the mycorrhizal fungi, direct counts of the reproductive propagules also can be made for many mycorrhizae. VA mycorrhizal fungal

reproduction can be assessed using asexual spores isolated directly from field soils. Although several procedures are available in the literature, we have concluded that differential water/sucrose centrifugation obtains a more reliable estimate of the total spore numbers (Ianson and Allen 1986). However, if the reclamation officer does not have access to a centrifuge, simple sieving of soils can separate many spore types in several soils (Ianson and Allen 1986). The fungi forming ectomycorrhizae often form mushrooms fruiting above the soil surface or just under the litter layer (hypogeous fungi). Upon fruiting, these fungi can be surveyed using sample techniques also used for plants or by simple density estimates. Both of these assessments also indicate the species composition of those fungi actively reproducing.

Existing Sampling Strategies for Assessing Mycorrhizal Activity

Several strategies for sampling and statistical analyses have been used to assess mycorrhizal activity. This is especailly important as most research efforts at understanding the distribution and ecology of mycorrhizae have concentrated on small-scale studies or theoretical assessments of their distributions. Few studies have assessed the large-scale practical effects of land management practices. A complicating factor is that mycorrhizal fungi appear to have differing dispersion patterns in different regions and ecosystems. Moreover, since the association is of plant and fungus, the distribution is highly dependent upon the dispersion patterns of the host.

Many studies have stated clearly that one or two strategies must be followed when sampling and assessing the mycorrhizae of a site or of a research effort. Most efforts have concentrated on using a regular (e.g., grid or line intercept) or a random sampling procedure. St. John and Hunt (1983) stated that mycorrhizal infection is clumped and that a negative binomial distribution for assessing percent mycorrhizal infection should be used. St. John and Koske (1988) also stipulated that spores are distributed according to a negative binomial model. What this model states is simply that the infection units or spores are aggregated into discrete patches in a sample area, i.e., the data are skewed with most values being low and a few high.

Alternatively, the negative binomial distribution does not always describe either infection or spore count data. In evenly planted grasslands, mycorrhizae can often be distributed evenly (if a later-successional site) or randomly (for a newly planted site). At the other extreme, in arid lands where plants are widely spaced, both infection and spores can be extremely skewed if both interspace and plant are considered or a value approaching a normal distribution can be obtained if only under-plant samples are considered. More importantly, all of these models only considered the distribution of data from within community sampling as opposed to assessing the influence of a management technique across a hillside or group of communities.

When comparing several studies that looked at the distribution of spore counts and percent VA mycorrhizal infection across geographical regions, the statistical distribution of points and sizes of patches varies considerably across biomes. For example, Anderson et al. (1983) reported that a patch was approximately 10cm^2 whereas Allen and MacMahon (1985) found that even a 2 cm^2 sample grid may have not been small enough to identify the patch size. Even within a site, the statistical distribution of values varies considerably depending on the size of area of interest. Moreover, this variation is not linear with area size or plant diversity (Fig. 2). As can be seen in Fig. 2, our data indicate that the degree of skewness can change across sample scales even on the same site.

Finally, mycorrhizal fungi are sensitive to both seasonal (Allen 1983) and annual (Allen et al. 1987) variations. Simply assessing the percent mycorrhizal infection, sporocarps, or spores at one time will not always give an investigator an accurate assessment of activity. Importantly, simple assessments of surrounding undisturbed (reference) areas may not give an accurate assessment of the density or diversity that one should expect of a reclaimed site. We have found the curvilinear relationship between propagule size and number (Chaung and Ko 1981) to provide a good estimate of the maximum propagule density by which to gauge both the reference and the reclaimed area (Fig. 3).

Responses of Mycorrhizal Fungi to Reclamation Efforts: A Case Study

We conducted a study to determine how differing reclamation strategies affect the outcome of both plant establishment and mycorrhizal redevelopment. We assessed two major factors that could affect reclamation success: soil management and initial plant density. Our indicator of success are spore counts of VA mycorrhizal fungi. We believe that this parameter gives the best indicator of recovery for the following reason: mycorrhizal root infection is exttremely variable in time and the spores represent an indication of the reproductive potential of the fungus and, thus, its fitness on the site. If the fungus is actively reproducing, then it is actively associating with a plant (M. Allen and E. Allen 1990).

Three soil treatments were tested: (1) zero topsoil (using the spoil material only), (2) stored topsoil (wherein the topsoil was removed, stored for 8 years, then replaced back over the plots at a depth of 10 cm), and (3) nonstored topsoil (where fresh topsoil was scraped and directly respread over test plots at a depth of 10 cm). These treatments were subjected to further treatments. The stored topsoil was subdivided onto three subtreatments: unplanted, planted with a moderate density of shrubs, grasses and forbs, and the normal management treatment. The nonstored topsoil treatment was subdivided into two subtreatments, unplanted and planted, also using a moderate density of shrubs, forbs and grasses. A moderate

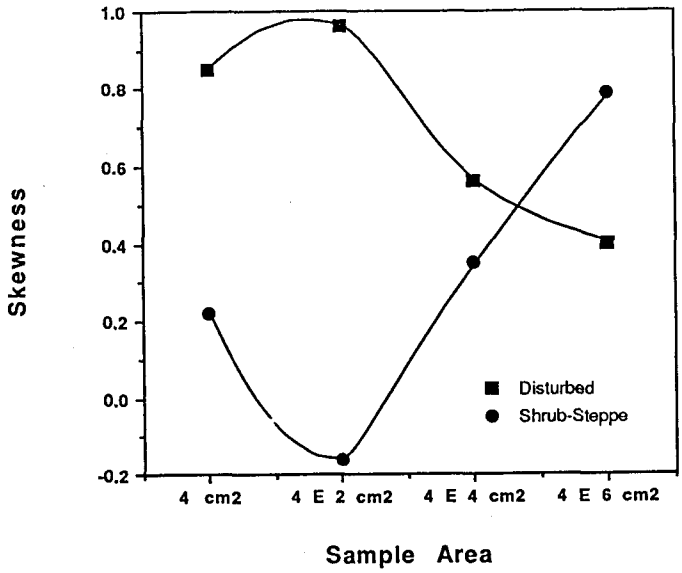

Figure 2.—Skewness using a constant sample size as the area samples increases. Data are from the 1981 Kemmerer site (Allen and MacMahon 1985). All samples were taken using a grid over the sample area indicated (E=10ⁿ).

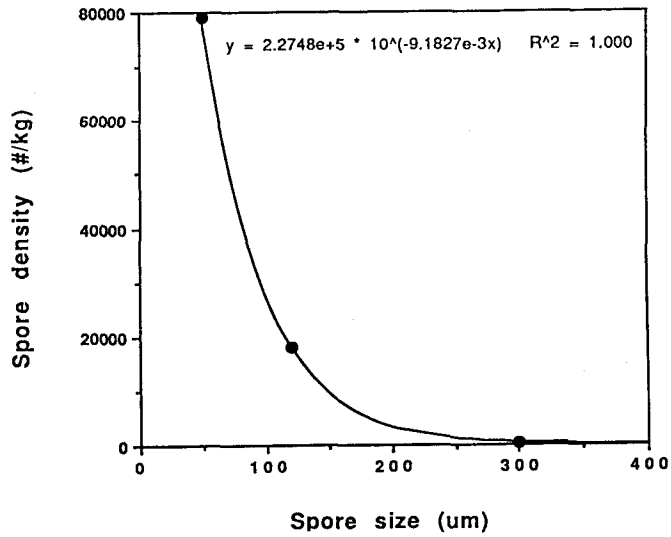

Figure 3.—The relationship between VA mycorrhizal fungal spore density and size using the spore data from Ianson and Allen (1985).

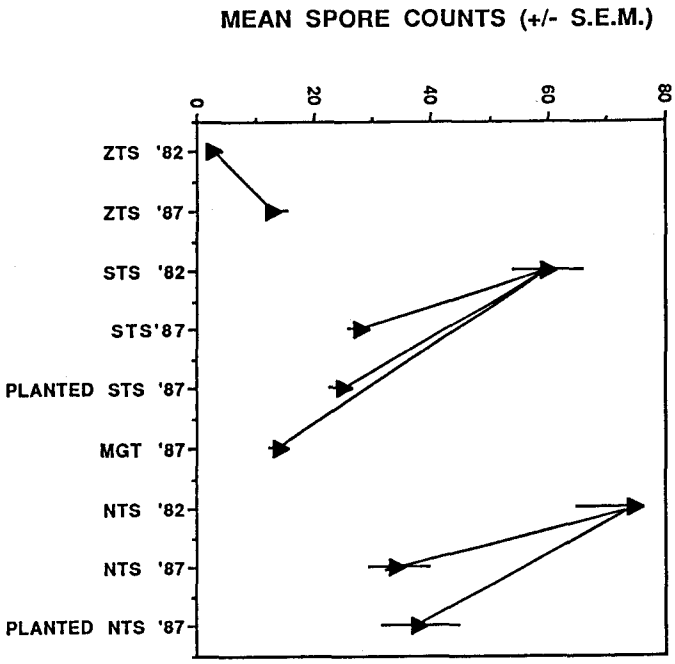

Figure 4.—Influence of initial soil treatment and subsequent management on VA mycorrhizal fungal spore counts in 1982 (the first growing season following the initialization of the site) and 1987. ZTS means no topsoil added, STS means stored topsoil added, NTS means non-stored topsoil added. Planted means that the plots were replanted with a moderate density of shrubs, grasses and legumes (see text for details). MGT means that the plots were managed using the normal company procedures (see text).

density planting meant that individual shrubs were planted at 0.6 x the density of shrubs in the adjacent undisturbed area. An identical number of patches of grasses and legumes were also planted. The normal management treatment consisted of planting barley for the first year after respreading of the stored topsoil, followed the second year by seeding a mixture of grasses, forbs and shrubs. All soil treatments were initiated during the autumn of 1981 and all plantings occurred in the spring of 1982. Each treatment was replicated using three 20 x 50 m plots. We took four soil samples from each plot evenly distributed downslope through the plot using 4 cm diameter by 10 cm deep cores. The data from 1982 (initial conditions) and 1987 (the final sample year) are shown here. Detailed spore, plant production and climate data associated with the El Nino years of 1983-84 and the drought of 1985 can be found in Allen et al. (1987).

Clear differences in the reproductive potential of the fungi were apparent at the whole-plot scale due to the initial conditions to which the plots were exposed. The initial soil treatments and the subsequent management practices had significant impacts on the reproductive activity of the VA mycorrhizal fungi. In both the stored and nonstored topsoil, the total spore density declined to levels that were not significantly different from each other. However, total fungal reproduction was significantly reduced in the management treatments to a level not significantly different from that of the plots that had no topsoil added (and no spores initially present, Fig. 4). Also, we found that the variance to mean ratio (an indicator of the development of patchiness, important to the redeveloment of "islands of fertility" in these semi-arid conditions) was also affected by initial soil treatment (Fig. 5). This ratio increased in the zero topsoil

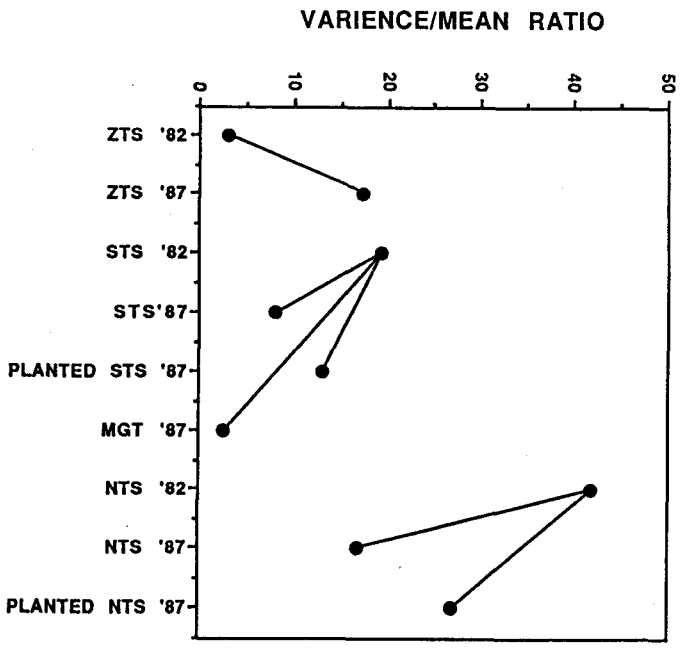

Figure 5.—Variance to mean ratio of the VA mycorrhizal fungal spore counts of different soil and management treatments in 1982 (the first growing season following the initialization of the site) and 1987. ZTS means no topsoil added, STS means stored topsoil added, NTS means non-stored topsoil added. Planted means that the plots were replanted with a moderate density of shrubs, grasses and legumes (see text for details). MGT means that the plots were managed using the normal company procedures (see text).

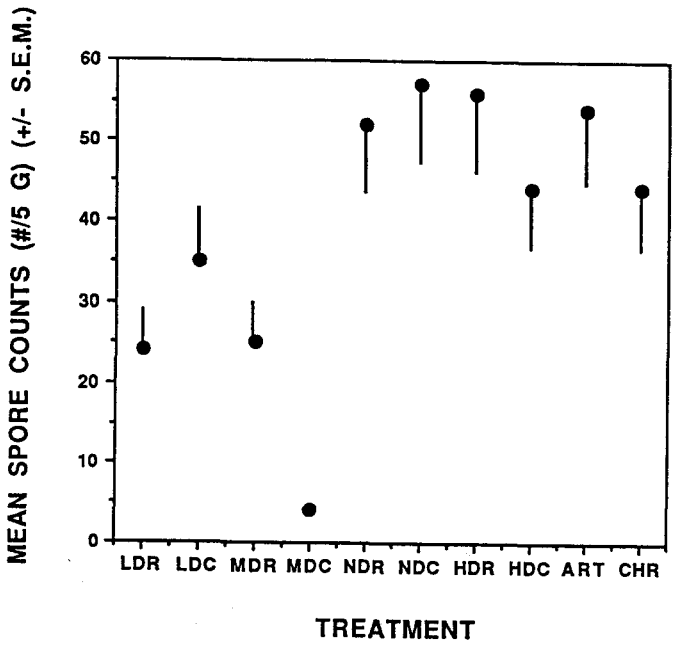

Figure 6.—Mean spore counts from samples taken at regular intervals (regardless of the plant location) in plots with the differing planting treatments (see text for treatment details). The data are from 1987 counts (S.E.M.=standard error of the mean).

and declined in all others. Importantly, it remained relatively high in the plots where the plants were arranged in patchiness but declined to the lowest levels in the management treatment. These data clearly indicate that, in arid lands where the reformation of spatial patches that serve as cues for animals, as windbreaks for snow and propagule deposition, the traditional covercrop/seeding management treatment resulted in a less desirable site for mycorrhizae.

We were also interested in how changing the initial planting density might affect the reestablishment of mycorrhizal associations. Using the same site described above, we altered the planting pattern as follows. We created low density (0.2 x normal), moderate density (0.6x), normal density (1.0x), and high density (1.5 x normal) plantings. A normal density planting was equal to the shrub density of the surrounding native areas (16,000 plants per ha). In addition, shrubs were planted in clumped or regular patterns to test for the development of patchiness. The higher density plantings resulted in greater total fungal reproduction than the lower density plantings (0.6x). However, when we tested for the differences in spore counts as a function of individual plant groups, no differences were detected between adjacant plots that contained differing numbers of plant shrubs (Fig. 7).

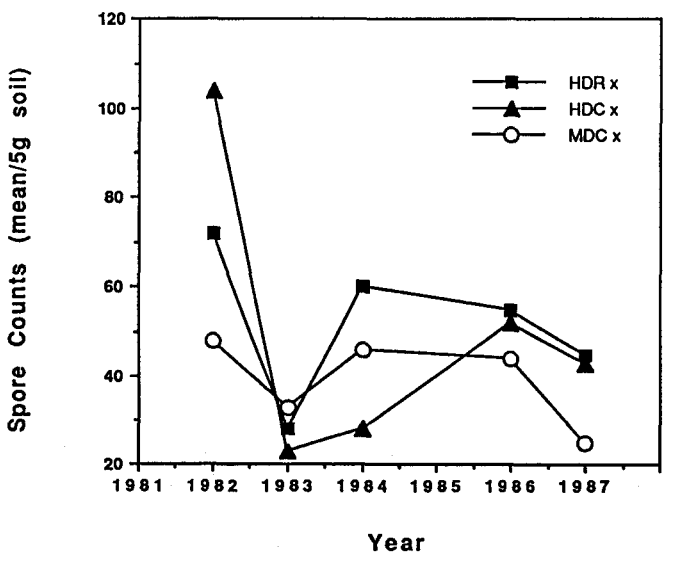

Figure 7.—Mean density of spores from adjacent plots with differing plant densities using samples taken only in association with individual plants. Shown are the mean spore counts from a plot with planting of a moderate density (clumped), a high density regular, and a high density clumped planting. These represent data from treatments showing the greatest extremes in mean spore counts (Fig. 6.).

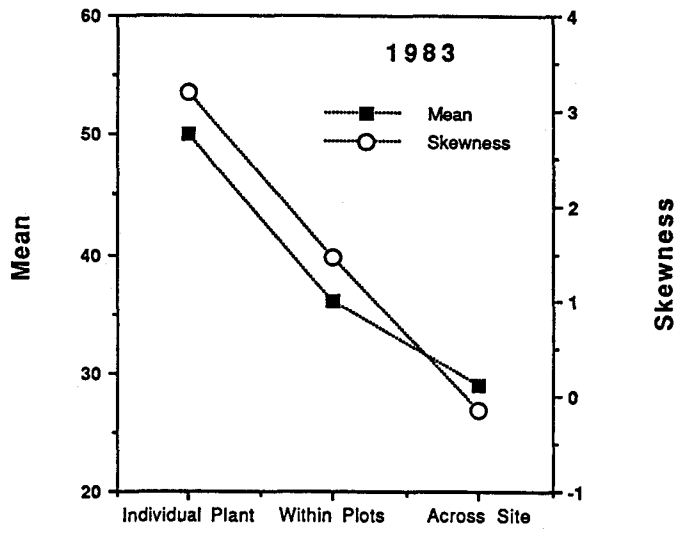

Figure 8.—Mean and skewness of VA mycorrhizal fungal spore counts in 1983 comparing samples taken among individual plants, within a single plot at regular intervals, and at regular intervals among plots with the same treatment across the site. All samples were taken using the medium density treatments (see text for details.

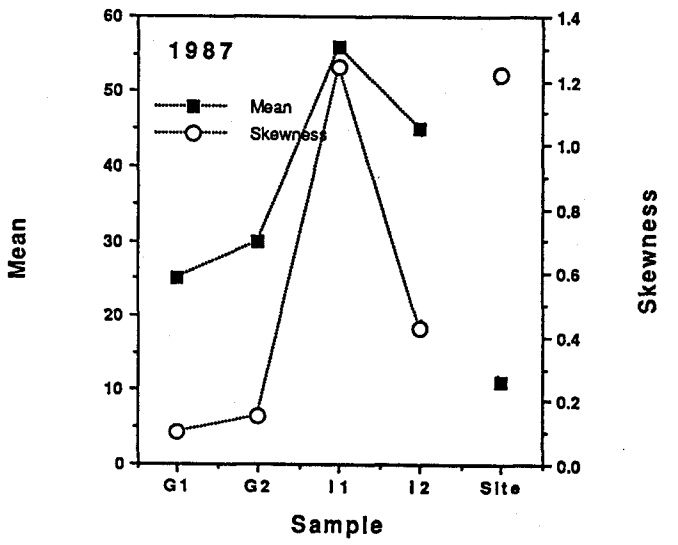

Figure 9.—Mean and skewness of VA mycorrhizal fungal spore counts from 1987 comparing samples taken among individual plants (G1, G2, I1, I2) and at regular intervals among the plots with the same treatment across the site. All sample were taken using the medium density treatments (see text for details).

These data suggest that the major factor affecting the fungal reproduction on a plot basis is the total density of plant patches placed on the plot.

No clear pattern of spatial variation versus planting treatment emerged either at the scale of the whole plot or at the scale of the individual plants. However, when we contrasted the spore count mean and distribution of individual plants with the larger units, an interesting pattern emerged. In 1983, the year following planting, both the means and the skewness (degree of clumping of values) were highest at the scale of the individual plant and lowest at the whole site scale (Fig. 8). By 1987, the mean values tended to be greatest at the individual plant scale but the skewness increased (Fig. 9). These data suggest that both the mean reproduction of the fungus and the distribution in space change with successional time. We also suggest that habitat heterogeneity may be important as the patch size appears to be localized around individual plants and different plants represent different habitats.

Finally, we were interested in the potential for changes in the fungal species composition with time. Is there any predictability to changes in species composition? Is the species composition simply a product of initial treatment or are the species richness and composition simply random? We hypothesized that island biogeography theory may provide a model for understanding aspects of the reestablishment of mycorrhizal fungi. To this end, we looked at changes in spore density and the VA mycorrhizal fungal species richness in 1987. Interestingly, both VA mycorrhizal fungal species richness and total spore numbers were related to the size of the shrub "islands" (Fig. 10).

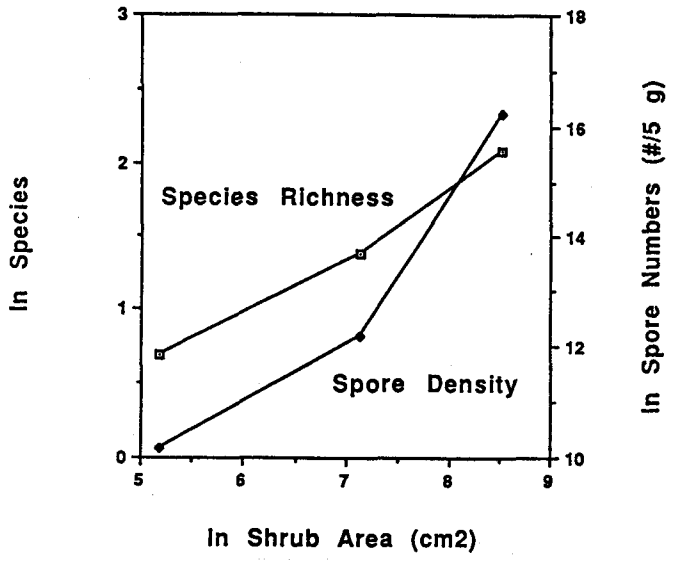

Figure 10.—Relationship between the size of shrub island and the density and richness of VA mycorrhizal fungal species using a natural log scale.

Recommendations for Assessment of Mycorrhizae by Land Managers

In conclusion, after wading through the research jargon, how should a land manager determine mycorrhizal activity in the context of assessing reclamation success? We suggest that a few simple observations can not only help the reclamation officer but can also contribute in a major way to our scientific data base. This will enable us to make better recommendations to individual reclamation officers

and to regulatory agencies. We suggest that the reclamation officers conduct simple surveys of mycorrhizal activity in conjunction with their assessments of plant parameters. This would include two parts. First, a survey of percent infected root segments of the dominant plants should be made. This would indicate whether or not mycorrhizal activity was present. This would entail no more than 5 plants of each of the dominant species from each major community on the reclamation site. Second, some measure of reproduction should be undertaken. This would require a survey of fruiting structures of the fungi themselves. Density estimates and diversity of sporocarps of ectomycorrhizal fungi (mushrooms) should be made as plants are assessed. Spores of VA mycorrhizal fungi should be extracted, identified (if the expertise is available) and counted. These surveys should be conducted at two scales, within a group of individual plants within each community type, and at regular intervals across the site. This would not necessarily need to be a large sample size, just enough to establish that the fungi are reproducing and that desired associations with the plants of interest are occurring. These data should be shared among reclamation researchers, officers, and regulators to understand better the dynamics of reestablishment of an association critical to the success of reclamation efforts. Continued cooperation among all concerned will better enable us to attain the common goal of reclaiming the structure and functioning of desired communities following the extraction of important resources.

Acknowledgments

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Nutrient Cycling: The Key to Reclamation Success?

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Abstract

The results of research into nutrient cycling on reclaimed land has enabled the control of nutrient input and species content during reclamation schemes to provide for faster nutrient cycling leading to improvements in ground cover, erosion control and appearance. However, the ecological value of abandoned mine lands where nutrient input is low and cycling very slow is often greater than that of many reclamation schemes. Research has indicated that a "capital" of nutrients is required in reclamation schemes for an ecosystem to be self sustaining and that intermittent inputs of nitrogen often lead to moribund vegetation with nutrients immobilized. Also small but frequent pulses of nutrients as observed in legume -supported swards or others where nutrient cycling is not obstructed can support effective reclamation schemes of some ecological value. These findings are considered in the context of the manipulation of nutrient cycling in order to enhance the ecological value of reclaimed sites.

Introduction

The supply of nutrients has been recognized as an important factor governing the establishment of vegetation on despoiled land since researchers began investigating vegetation establishment on mined-out areas in the early part of this century (McDougall 1918; Croxton 1928). Even before such research work began, enlightened mine owners were using techniques such as spreading used animal bedding on spoil as a mulch to encourage the establishment of nutrient cycling in some early reclamation attempts (Richardson 1984). It is not surprising therefore that much research into mined land reclamation during this century has concentrated on ways of establishing and accelerating nutrient cycling. Much of this research has concentrated on nitrogen supply with a fair body of work also on phosphate supply. Because of the scale of coal mining, probably more research has been carried out on reclamation after the mining of coal than on all other types of mining combined.

The primary emphasis of the reclamation of mined out land this century has been to put the land to beneficial use. Other important requirements have been the removal of visual scars, the prevention of erosion and the prevention of the contamination of water courses or land. The requirement to put the land to "beneficial use" has meant that much of the research carried out has been related to nutrient input and cycling to maximize herbage and tree production levels. Reclamation success has been measured in terms of productivity comparisons with similarly cropped unmined land. However in more recent times, measures such as the Surface Mining and Control and Reclamation Act (SMCRA) in the USA have done much to raise the level of importance of the consideration of ecological parameters in reclamation (Brenner 1984). In the United Kingdom such considerations have also become an important factor in land reclamation and rehabilitation.

This paper aims to summarize what has been learned from research into nutrient cycling in reclaimed land and to relate this to the concept of ecological success.

Types of Despoiled Land

There are a wide variety of ways in which land becomes despoiled and the characteristics of despoiled land vary considerably (Fig. 1). Many of the constraints on the development of vegetation on such land interact and on some types of despoiled land one factor, toxicity, for example, may dominate all other considerations. Nutrient deficiency is however a characteristic of most types of despoiled land and results from either a lack of one or more of the major nutrients or an impediment to the cycling and supply of one or more of those nutrients. In this paper three types of derelict land will be used as examples. These are colliery spoil, china clay waste, and spoil from metalliferous mining. The characteristics of these spoils are presented in Table 1.

Nutrients Cycling

A major limiting factor on despoiled land is lack of nitrogen and discussion here will be primarily concerned with nitrogen supply. A nitrogen cycle which could be applied to most types of reclaimed land is presented in Figure 2. In nitrogen deficient systems, cycling is at a low level because inputs of nitrogen are low and are also often impeded because nitrogen is bound up in compounds of high C:N

ratio or immobilized in microbial biomass. In such situations the C:N ratio in organic matter is too high for microbes to decompose the organic matter and any available nitrogen arising is quickly further taken up and immobilized by microbes. This interruption of cycling is also often assisted by factors affecting microbial action such as low pH. Mineralization and nitrification are the most common places of interruption of the nitrogen cycle on reclaimed land (Fig. 2). The effect of this interruption has been shown in the United Kingdom by Williams (1975) and in the United States by Reeder and Berg (1977). Some results of William's experiments are summarized in Figure 3 and illustrate well how substrate availability in terms of ammonium and fluctuations in pH as affected by lime additions influence nitrification, a microbially controlled process. The message is clear: for nitrogen to be supplied to vegetation there has to be a steady supply of ammonium to be nitrified and the pH has to be high enough for that nitrification to take place.

	Stability	Combustion	Slope Angle	Flooding. Stress	Toxicity	Compaction	Temperature	Wind Erosion	Nutrients	Stoniness	Uneven Surface	Erosion	Soil Fauna and Microbes
Colliery Spoil	•	•	•	•	•	•	•	•	•	•	•	•	•
Smelter Slag	•	•	•	•	•	—	•	•	•	•	•	•	•
Slate/Shale	•	—	•	•	—	—	•	•	•	•	•	•	•
Metal Wastes	•	—	•	—	•	•	•	•	•	•	•	•	•
Quarry Pits	—	—	•	•	—	•	•	•	•	•	•	•	•
Brick Pits	—	—	•	•	—	•	•	•	•	•	•	•	•
Peatland	—	•	—	•	•	—	•	•	•	—	—	•	•
China Clay	•	—	•	•	—	—	•	•	•	—	—	•	•
Ironstone	•	—	•	•	—	•	•	•	•	—	•	•	•
Chemical Waste	•	—	—	—	•	•	•	•	•	—	—	•	•
PFA	—	—	—	•	•	•	•	•	•	—	—	•	•
Sand and Gravel	—	—	•	•	—	•	•	•	•	—	•	•	•
Domestic Refuse	—	•	—	•	•	•	—	•	—	—	•	—	—

Figure 1.—Constraints on vegetation development on different wastes (after Kent 1982)

Table 1.—Characteristics of colliery spoil, china clay waste and metalliferous mine spoil

Item	Colliery spoil	China clay waste	Metalliferous mine waste
pH	<3 to >8	<4 to 6	<3 to 8
Physical properties	Weathering silt and mudstones, high clay fraction	Mica (silt) or sand waste, the latter free draining	Clay to gravel size, thixotropic to free draining
N	Nitrogen deficient	Nitrogen deficient	Nitrogen deficient
P	Phosphate deficient, fixes phosphate	Phosphate deficient, easily leached out	Phosphate deficient
K	Adequate supply	Deficient, easily leached out	Deficient
Potential toxicities	Al and Zn at low pH, salinity on some sites	None	Heavy metals
Moisture	Waterlogged in winter, potentially droughty in summer	Sand waste: low moisture-holding capacity; mica waste: less of a problem	Sands and gravels have low moisture-holding capacity; silts retain moisture
Erodibility	Easily eroded	Easily eroded	Easily eroded

The ammonium has to come from either organic matter or external sources such as fertilizer and there have been many studies to show that intermittent applications of nitrogen on nutrient deficient colliery spoil sites can encourage the accumulation of material of a wide C:N ratio which then acts as a nitrogen sink. In such situations nitrogen supply to vegetation is only adequate shortly after fertilizer application (Wieder et al. 1983; Palmer et al. 1986).

A way of boosting a supply of organic matter rich in nitrogen is by the encouragement of a legume with an active nitrogen symbiont. It has been suggested by work in the UK on the colonization and reclamation of china clay waste that a legume component is an important factor in successional stages leading to a "self sustaining ecosystem" (Dancer et al. 1977). Studies in natural succession after the retreat of glaciers at Glacier Bay Alaska have also reinforced the importance of a nitrogen-fixing component in succession (Crocker and Major 1955). On china clay waste the amount of nitrogen accumulated under vegetation of various types and ages was compared to that of natural ecosystems (Table 2) and it was concluded that a level of nitrogen in the soil of 700 kg/ha or in the soil and vegetation

Table 2.—Comparison of nitrogen content and compartmentation between major ecosystem pools in china clay wastes and semi-natural temperate ecosystems (from Marrs and Bradshaw 1982)

Ecosystem	Total N	Shoots (%)	Roots (%)	Litter (%)	Soil (%)	Sampling depth (cm)	Reference
	kg/ha					cm	
Naturally colonized china clay wastes							
Pioneer (Lupinus arboreus)	291	37	1	7	56	21	
Pioneer (Calluna vulgaris/Ulex europaeus)	823	13	5	3	79	21	
Intermediate (Salix atrocinerea)	981	8	18	6	68	21	Marrs et al. (1981)
Woodland (Betula pendula/ Rhododendron ponticum/Quercus robur)	1770	30	3	0	67	21	
Reclaimed china clay wastes							
Sand tips	211	11	59	nd	30	21	
Mica dam walls	441	8	61	nd	31	21	Roberts et al. (1981)
Semi-natural temperate ecosystems							
Heathland	5644	combined shoot root=3		1	96	20	Robertson and Davies (1965)
Grassland	10460	0.7	0.5	0.3	98.5	30	Perkins (1978)
Oak woodland	7940	combined shoot root=4.9		0.9	94.2	30	Duvigneaud and Denaeyer-de Smet (1973)

of 1,000 kg ha-1 was necessary for the ecosystem to be self sustaining (Roberts et al. 1981).

Work on colliery spoil has not been able to quantify the accumulation of nitrogen in relation to ecosystem development in the same way as on china clay waste. However, the importance of legumes, not only in accumulating nitrogen but also in providing a steady supply of nitrogen throughout the growing season, has been demonstrated on colliery spoil (Palmer and Chadwick 1985; Palmer et al. 1986). Despite these findings and the many records of naturally occurring legumes on colliery spoil, there is little evidence in the literature to suggest that legumes are at a competitive advantage or are contributing significantly to the nitrogen input to the soil in situations where legumes are naturally colonizing a despoiled site (Palmer 1984). It is on sown, reclaimed land that the real advantages of legumes have been found. This has undoubtedly been due to the fact that these sites have often received phosphate applications and have been limed. Adequate levels of phosphate are necessary of maximize nitrogen fixation and for the accumulation of nitrogen in legume-supported swards (Palmer and Iverson 1983).

Metalliferous mine sites are nitrogen deficient also but the overriding limitation on plant growth is due to toxic metals. Nevertheless, the principles governing nutrient cycling still apply. The metals affect not only higher plants but also the microbes responsible for N-cycling and vegetation establishment can be improved by the addition of fertilizer and organic matter (Winterhalder 1988). Two mechanisms seem to operate. One is that at higher nutrient levels there is an increased ability of the plant to cope with stress due to toxicity and the other is that the chelation of metals by organic matter reduces the availability of these metals for uptake by plants.

Ecological Considerations

It is not the purpose of this paper to discuss at length how ecological success in reclamation might be evaluated but it is worth pointing out how ecological considerations might be treated in land reclamation. There are perhaps two

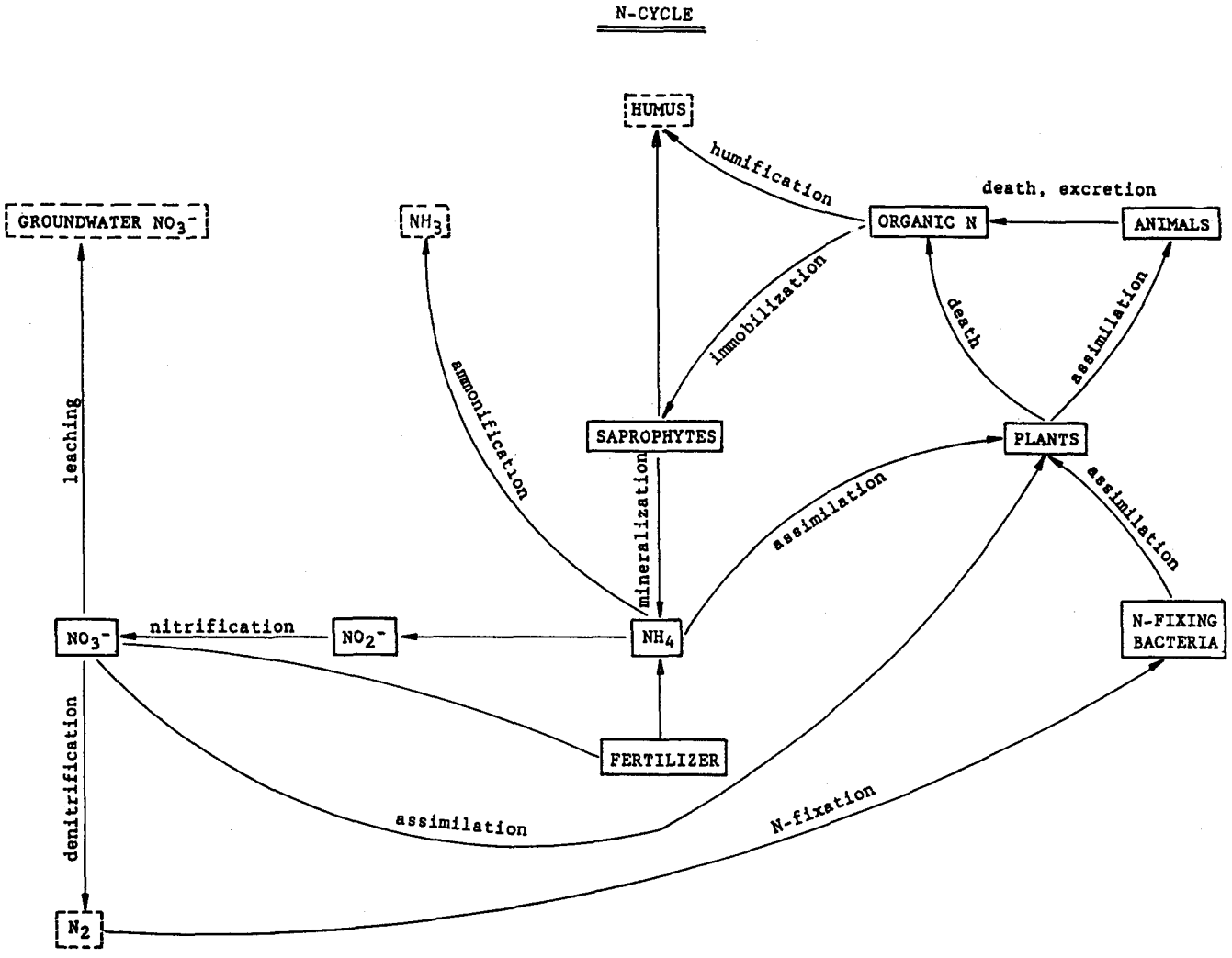

Figure 2.—A nitrogen cycle applicable to reclaimed land. Decomposition, mineralization and nitrification are processes likely to be impeded on reclaimed land.

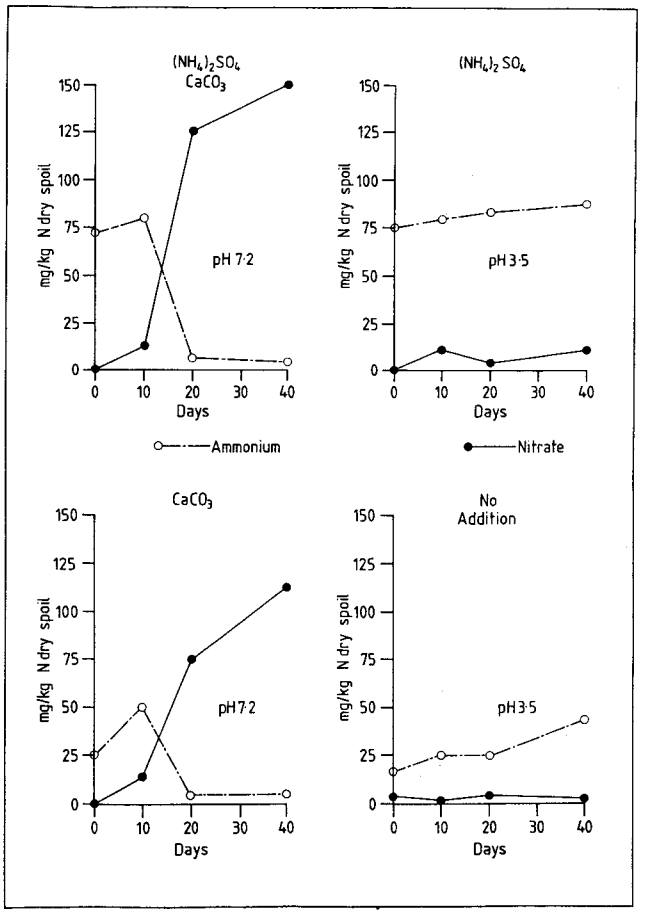

Figure 3.—Nitrogen transformations on reclaimed colliery spoil incubated at 25°C and 10% moisture for 40 days with additions of ammonium sulphate, calcium carbonate or no additions. (from Williams 1975).

distinct ways in which this may be done. The first is where land of some ecological merit is being destroyed as part of, say, a mineral extraction operation and land reclamation has to restore the land to the same ecological status and type. Here the requirements of land reclamation in terms of vegetation composition and type and even nutrient cycling can be quite well defined and success can be measured by assessing similarity with undisturbed land of a similar type to that disturbed. In the United Kingdom where land of some ecological value is to be destroyed as part of a mining or other activity, a technique becoming more common to ensure restoration of ecological value is to move the vegetation and soil in blocks to either a holding area to be replanted as part of the reclamation scheme later or to a different area of less ecological value. This has been successful for areas of limestone grassland (Park 1985) and is even being attempted for the ground flora of ancient woodland. In this type of reclamation, if effective, nutrient cycling processes are transferred with the soil, vegetation, and management.

The second way in which ecological considerations may be taken into account is where the opportunity is taken during reclamation to increase the "ecological value" of the site. For the purposes of assessing ecological value the factors listed in Table 3 have been considered to be important. The concept of diversity has of course in been embodied in United States reclamation legislation since SMCRA and is important in the United Kingdom too. However in the United Kingdom the concept of "naturalness" is currently being given more weight than previously. Although most of the vegetation of the United Kingdom is semi-natural and has been disturbed by man, the extent to which the vegetation conforms to recognized natural communities and the lack of recent gross disturbance are considered important. Even with grossly disturbed sites, such as mined-out land, lack of recent intervention by man allows colonization and ecosystem development to take place naturally. This development, in response to the disturbed environment, can lead both to communities which may fit into recognized natural community types and those which are modifications of natural types. Such communities are nevertheless the result of the natural processes of colonization and be considered important because of this.

In order to consider the influence of nutrient cycling where ecological value is a goal of reclamation it is instructive to consider what has happened on naturally vegetated mined out sites. It is a paradox facing the reclamation practitioner that in order to "reclaim" a site one is often faced with the

Table 3.—Criteria used in wildlife conservation evaluation in nine studies reviewed by Margules and Usher (1981) as adapted from Usher (1986)

Criterion	Frequency of use in nine published studies
Diversity (both of species and habitats)[a]	8
Naturalness[a]	7
Rarity (both of species and habitat)[a]	
Area (extent of habitat)[a]	6
Threat of human interference	
Amenity value	
Education value	3
Recorded history[a]	
Representatives	
Scientific value	2
Typicalness[a]	
Uniqueness	
Availability	
Ecological fragility[a]	
Management considerations	
Position in ecological/geographic unit[a]	1
Potential value[a]	
Replaceability	
Wildlife reservoir potential	

[a]Criteria used by the Nature Conservancy Council for assessing sites in the United Kingdom.

destruction of spoil heaps of some considerable wildlife value. Many sites abandoned to be colonized naturally are of some ecological interest. This often the case if the waste is alkaline or has a high heavy metal content. In the United Kingdom many of these sites are protected because of their ecological interest (Table 4). It is also the case with some sites which are low in nutrients such as colliery spoil sites where the naturalness of the sites is considered of value. Whether these sites have special characteristics (e.g., heavy metals and alkalinity) or not, they have all developed their vegetation and ecological value under very low inputs of nutrients.

In the United Kingdom the annual input of nitrogen from rainfall would be of the order of 9 to 10 kg/ha in industrialized areas. Phosphate levels would also be low, thus reducing the chance of large inputs of nitrogen fixed by legumes. Under all of these systems cycling of nutrients would have been at a low level and slow, limited by factors such as substrate nitrogen required to allow the sward to develop but at which the growth of productive species is not too great. Another important trigger is pH. Chadwick and Hardiman (1976) in the United Kingdom and Glenn-Lewin (1979) in North America have both shown that species number on colliery spoil heap is correlated with pH rather than age of the spoil. Lime applications to raise pH would also act as a trigger to facilitate colonization. Facilitating the natural development of vegetation in this way while at the same time meeting reclamation goals from other stand-

Table 4.—Examples of disused mineral extraction sites in Britain which have acquired conservation value status on botanical grounds (from Johnson 1978)

Location	Origin	Distinguished botanical features
Brickfields, Buckinghamshire[a]	Brick-clay extraction	Blackstonia perfoliata, Lotus tenuis, Ophrys apifera
Sanford, Berkshire[a]	Sand extraction	Dactylorhiza incarnata, Equisetum variegatum, Epipactis palustris, Potamogeton coloratus
Aston Clinton, Buckinghamshire[a]	Chalk ragstone quarrying	Hippocrepis comosa, Ophioglossum vulgatum, Polygala calcurea, various Orchidaceae
Hurley, Berkshire[a]	Chalk quarrying	Anacamptis pyramidalis, Gymnadenia conopsea, Ophrys apifera
Millers Dale, Derbyshire[b,c]	Limestone quarrying/ calcining	Botrychium lunaria, Daphne mezerem, Parnassia palustris, mezerum Saxifraga tridactylites, various Orchidaceae
Wingate, Durham[b]	Magnesian limestone quarrying	Botrychium lunaria, Crepis mollis, Gentinella amarella, Salix nigricans, various Orchidaceae
Honister Crag, Cumbria[d]	Slate quarrying	Alchemilla alpina, Saxifraga aizoides, Oxyria digyna, Sedum rosea

[a]Kelcey (1975).
[b]Davis (1976).
[c]Bradshaw (1977).
[d]Ratcliffe (1974).

points such as erosion control and visual appeal would seem to be a valid way of achieving ecological success. In the reestablishment of heathland, a vegetation type which normally receives only atmospheric nutrient inputs, the use of low levels of fertilizer to accelerate establishment has been found to be beneficial (Putwain et al. 1982). Alternatives to allowing development of ecologically valuable swards, such as wholesale transplantation of ecosystems ahead of mining, are valid but costly. The reclamation practitioner should not overlook the possibility of leaving some of the reclaimed area unsown and unfertilized in order to facilitate natural colonization.

This review has concentrated primarily on nutrient cycling in spoil materials. Where a very fertile topsoil is used in reclamation, other kg N/ha in the soil suggested by china clay reclamation research may be an achievable goal. At the same time, the rate of mineralization of nitrogen must be low enough to allow a diverse sward to develop, but not too low to prevent ingress of new species. Where this level should be is not clear from research so far but previously unpublished data on the ingress of unsown species into trial plots on colliery spoil provides some clues. These plots were established by the Derelict Land Reclamation Research Unit, University of York , at Thorne Colliery in the United Kingdom. The plots had received phosphate fertilizer but no nitrogen and had been sown with a legume. Nitrogen fixation was initially at a very low level and the level of mineral nitrogen in the soil remained extremely low for three years. The sward was largely moribund during this time with bare ground between plants. In the third year after sowing, nitrogen fixation was much higher, legumes grew vigorously, soil mineralizable nitrogen levels increased and the sward closed up. This was accompanied by an increase in the number of unsown species establishing in the plots (Fig. 4). The level of mineralizable nitrogen was between 10 and 40 mg/kg when the number of unsown species entering the plot increased (Fig. 5).

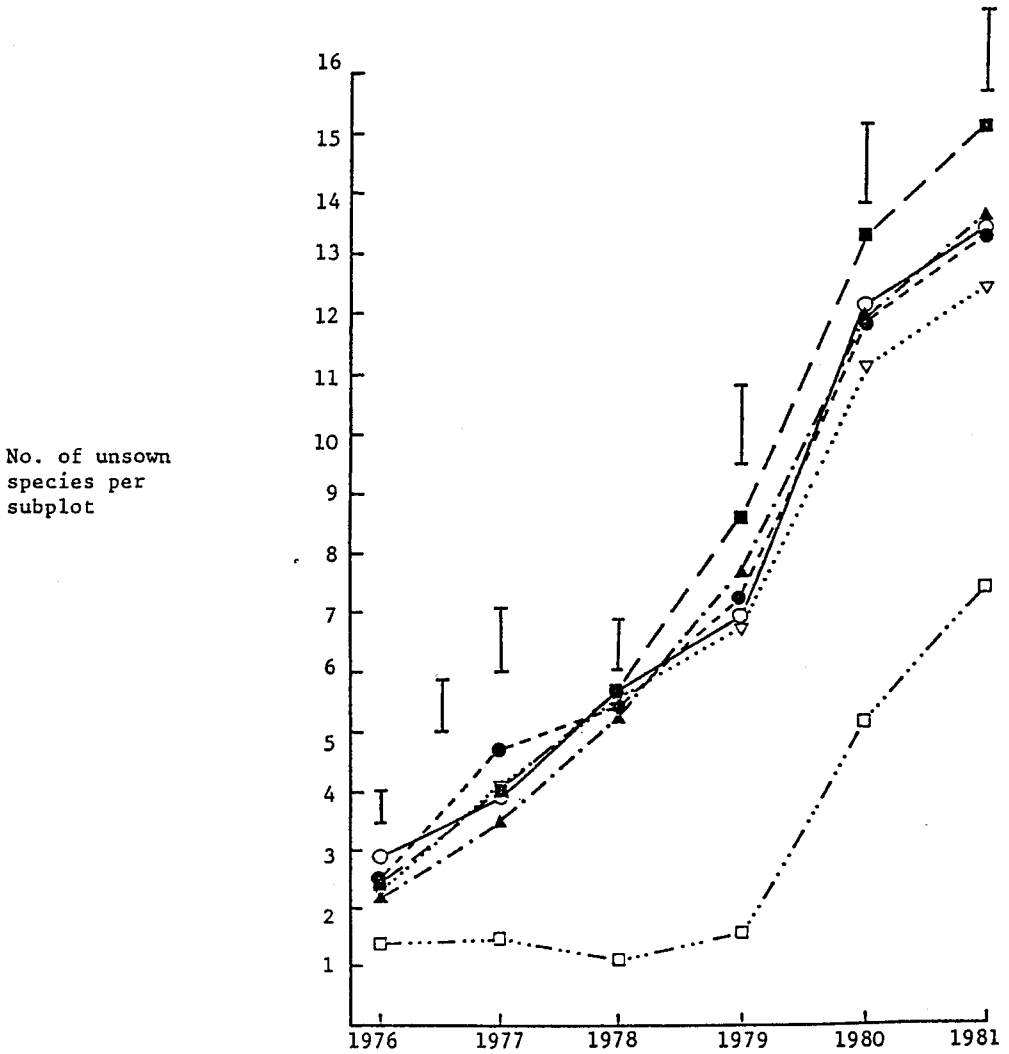

Figure 4.—The ingress of unsown species into legume and fertilizer plots at Thorne Colliery, South Yorkshire, UK between 1975 and 1981. Symbols: □ legume plots, ▲, ▼, ■, ○, ●, various forms of applied nitrogen fertilizer. Least significant differences (p 0.05) for each year are shown.

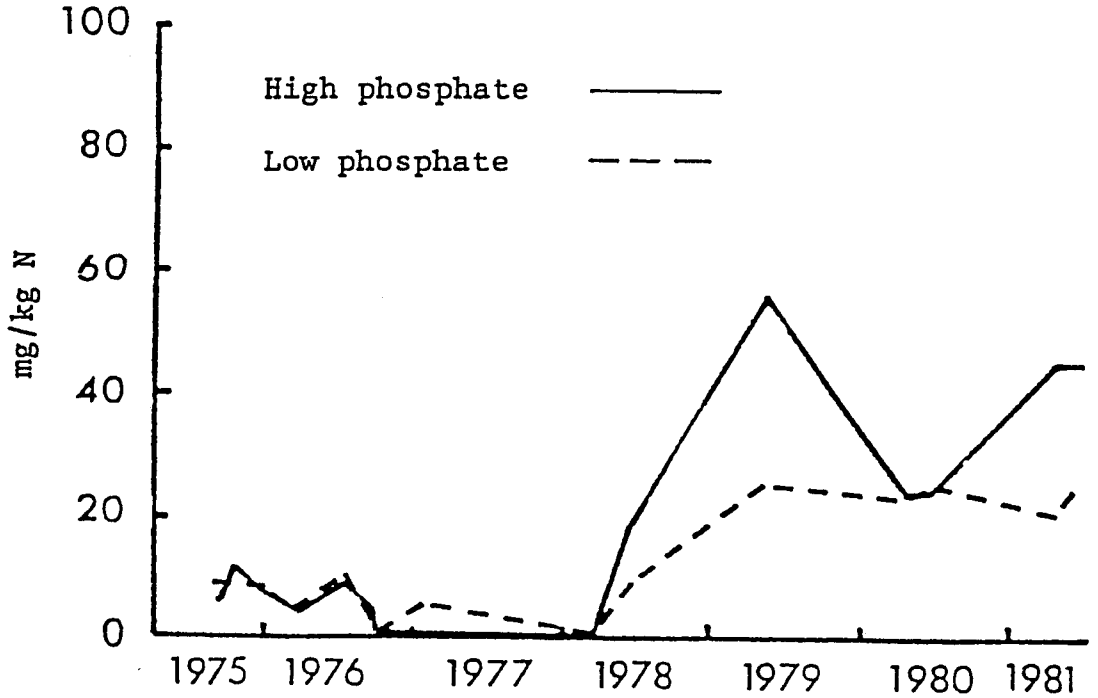

Figure 5.—Mineralizable nitrogen levels in legume plots at two different phosphate levels at Thorne Colliery, South Yorkshire, UK between 1975 and 1981.

Such data indicate that there is a "trigger level" of mineralizable availability and pH already discussed. Nutrients would accumulate slowly often bound up in organic matter with thin humic layers developed at the surface. At one site in Wales, for example, after 60 years a closed sheep-grazed sward had developed on colliery spoil. An organic layer of about 50 mm had developed above the spoil. In harsher environments, due to factors such as toxicity or climatic extremes, the development of such a layer would take much longer. Metalliferous mines in the same area of Wales had virtually no vegetation or organic layer development after similar periods of abandonment. They were still of ecological value however because of their lower plant flora. In this context the ecological value of bare slopes and open communities must also not be overlooked in reclamation scheme design.

Although ecologically valuable sites have developed with very small inputs of nutrients on abandoned mine lands, the reclamation practitioner cannot wait the length of time required for new sites to be colonized in this way.

On many sites a balance has to be struck between obtaining a vegetation cover which will be visually acceptable and performing an erosion control function but which will develop in an ecologically valuable way. For example, difficulties may be encountered in restoring land of high ecological value because the high nutrient status of replaced topsoil will encourage productive species in the sward at the expense of species diversity. In such situations a cropping regime may have to be practiced which will over a period of years remove nutrients from the system and allow a more diverse sward to develop. Experiments using this technique on grasslands which have begun to lose diversity due to changes in management have met with some success in the United Kingdom (Marrs 1985). The management problems associated with over-fertile soils and productive species can, however, be prevented by avoiding the use of these soils at the time of land reclamation. The successful management of nutrient cycling, whatever the nature of the substrate, must be based on a proper assessment of the subtrate and the tailoring of the land reclamation techniques to substrate characteristics and reclamation goals.

Conclusions

1. Mine spoils are often low in the major nutrients. Nitrogen is a major limiter of plant growth and the nitrogen cycle may be interrupted by microbial substrate availability, low pH, or toxicity.
2. Additions of high levels of fertilizer lead to the promotion of productive species in the sward. If these additions are intermittent, then moribund swards of high C:N ratio often result.
3. Legume-based swards can provide a steady but low level of nitrogen input which will give a continuous input of mineral nitrogen throughout the growing season and thus avoid accumulation of moribund productive grasses.
4. Work on china clay waste reclamation has suggested that a capital of 700 kg N/ha is needed in the soil before the ecosystem becomes self-sustaining.
5. Naturally colonized spoil can even with very low nutrient inputs result in ecologically valuable vegetation after many years.

6. On colliery spoil a trigger of a low level of mineralizable nitrogen is needed to increase species diversity by ingression of unsown species. Between 20 and 40 kg mineralizable N/ha appears to be the level at which this occurs.
7. Ecologically valuable swards are most likely to be achieved and sustained by providing a low level of mineralizable nitrogen throughout the season. This is best obtained by the use of a legume which in itself necessitates an input of phosphate.
8. Where the soil material is rich in nutrients care should be taken with species choice and management in order to achieve a diverse sward.
9. Opportunities should not be missed during mining and reclamation schemes to consider transplanting habitats and leaving areas unsown to colonize naturally.

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