

APPLICATION OF THE PRINCIPLES OF  
MICROBIAL ECOLOGY TO THE ASSESSMENT  
OF SURFACE MINE RECLAMATION<sup>1</sup>

by

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**Abstract** Soil micro-organisms fulfill a number of vital functions in the below ground ecosystem, including mediating many plant nutrient transformations and contributing to soil structure formation and maintenance. It has been demonstrated that the disruption to the soil during surface mining operations results in changes in the size, composition and activity of the soil microbial community. Any decrease in the size and diversity of microbial populations may have serious consequences for the development of self-sustaining ecosystems on disturbed areas. However, these changes also offer an opportunity to assess and monitor the state of restored ecosystems by using the soil microbial community as an indicator of the success of differing restoration strategies. This study reports on an investigation into areas restored after opencast coal mining in Britain. Differences in soil microbial activity with depth have been used as such an indicator and significant differences in profile development have been demonstrated, with both time after restoration and management practices. Rates of dehydrogenase activity in areas restored up to 10 years prior to sampling were determined. The rates of activity in recently restored areas were only 2.5% as large as those of adjacent undisturbed semi-natural areas, and in some samples microbial activity was undetectable. Organic carbon data was less able to reveal differences between sites and treatments. Comparison with undisturbed arable sites was also made.

Additional key words: Dehydrogenase activity, soil activity profiles, opencast mine restoration.

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### Introduction

Conventionally, the assessment of the success of restoration after mineral extraction has been based on the use of chemical techniques and plant growth performance (Bradshaw and Chadwick 1980). However, chemical analyses are unable to provide a summary of the dynamics of the changes occurring within any restoration scheme, unless sampled repeatedly, and plant growth measured in situ may take a number of years to manifest themselves. By that stage, vital time and opportunity may have been lost. One aspect of the ecosystem which has been largely neglected and may prove extremely useful in this regard is the soil microbial community. The micro-organisms forming this community are intimately associated with the cycling of nutrients and the formation and maintenance of a stable and open soil structure, both essential features of a successful restoration. Molope et al. (1987) have shown that the microbial community is important in maintaining aggregate stability in soils, with fungi enmeshing soil particles in hyphae and bacteria secreting polysaccharides. Clough and Sutton (1978) indicated that a mycorrhizal fungus stabilized aggregates in sand dunes, and Rothwell (1984) has shown that vesicular-arbuscular mycorrhizae (VAM) had a role in producing aggregates in a surface minesoil.

Cundell (1977) was among the first authors to stress the importance of micro-organisms in the reestablishment of a functioning ecosystem in strip

mined land. He reviewed the limited work done in this area and drew heavily on material from undisturbed ecosystems. Cundell reported on a study of spent shale wastes in western Colorado and lignite overburden in North Dakota. He outlined the important functions of the soil microbial community, and indicated how treatments such as fertilization, mulching, seeding, and inoculation with nitrogen fixing heterotrophs affected the establishment of a self-sustaining system. The major conclusion was, however, that a more detailed knowledge of the microbial processes involved in soil formation and plant growth on new substrates was required. This review appears to have stimulated other work from the number of subsequent citations.

Other workers have reported on the microbial communities found in restored or abandoned mineral extraction operations. Miller and Cameron (1978) and Miller and May (1981) reported on the state of the microflora of two abandoned mines in the midwestern United States. They found that the substrate, bare spoil in many cases, was unfavourable to microbial colonisation and growth and required some amendment to support plant life. There were fewer micro-organisms in the minesoil than some adjacent undisturbed areas, and there was a lower species diversity in the minesoils. This tended to produce a 'zymogenous' microflora capable of short term rapid growth in response to addition of fertilizers, and was deficient in the 'autochthonous' species capable of long-term degradation of organic matter typical of a

self sustaining system. They recommended the addition of organic amendments to spoil, a conclusion supported by subsequent workers in the field (Fresquez and Lindemann 1982; Visser 1985). Stroo and Jencks (1982) indicated that using inorganic fertilizers allowed the zymogenous population of micro-organisms to attack any organic matter which did exist, leading to regression of the system within ten years of revegetation treatments. Several workers have shown that certain aspects of civil engineering operations have deleterious effects on the soil microbial community (Abdul-Kareem and McRae 1984; Harris et al, 1989).

Recent work in Britain has indicated the importance of micro-organisms in re-establishing ecosystem function, and has suggested a link between microbial activity and soil structure (Harris and Birch 1989). There is increasing interest in restoration to conservation systems which, coupled with the traditional pasture and wild flower systems in the British Isles, has made restoration to meadow particularly attractive. There is little doubt that systems can be moved, and that suitable existing stable natural systems can be managed toward meadow. However, re-creation of such habitats after the disturbance of mining is little understood, with measures of restoration success relying on traditional methods.

#### Aims

The objective was to investigate the biological activity in restorations up to

10 years old and to compare them with adjacent similar areas (used as controls) and, in particular, to investigate the potential for using soil microbial activity profiles to discriminate between treatments.

#### Materials and Methods

##### Site Details

Soil cores were collected from the following Open Cast Coal Sites (OCCS) and areas within them:

1. Shilo: An undisturbed site with a variety of species-rich meadows; two were sampled, one dry and one wet area. National Grid Ref SK 460460.

2. South Shilo: Restored for 5 years; two fields were sampled, one grazed and one ungrazed (cut for silage). National Grid Ref SK 475435

3. Old Shilo: Restored for 10 years; one grazed and one ungrazed restoration were sampled. National Grid Ref SK 470450

These areas were adjacent to one another, and comprised of soils of the same soil series and textural class.

##### Sampling

Five 30 cm deep cores were taken by means of sterilised 4 cm gouge augers from each area and placed in sterile plastic bags. They were transported back to the laboratory in insulated bags containing ice packs. In the laboratory the cores were cut into 5 cm segments in order of increasing depth, representing depths of 0-5, 5-10, 10-15, 15-20, 20-25, and 25-30 cm. The core segments were chopped, using aseptic technique, and kept separate

for subsequent analysis. Fresh soil was used for moisture content and dehydrogenase activity assay, a separate subsample was air-dried at 30°C and ground to pass through a 0.5 mm sieve for carbon analysis.

#### Analysis

Dehydrogenase activity was determined by the method of Tabatabai (1982). A subsample of moist soil was incubated with a triphenyl tetrazolium chloride solution for 24 h at 37°C, and the formazan produced extracted with methanol, and the absorbency measured at 485 nm. Carbon was determined using a wet-oxidation technique (Nelson and Sommers 1982).

#### Results and Discussion

Steady decreases in microbial activity with increasing depth were found in the case of both profiles from the undisturbed areas, with high activities in the top 0-5 cm segments (Fig 1, bars represent standard deviation). This finding is typical of profiles to be expected in a soil ecosystem that has developed over many years and has remained essentially undisturbed. Over time, organic matter incorporation into the surface layers and oxygen depletion in the deeper layers has led to microbial activity in the surface as compared to the deeper layers.

In the five year old grazed restoration at South Shilo very poor microbial profile development has occurred, the total activity is only 2.5% of the value obtained for the undisturbed dry meadow area. There was no detectable

microbial activity in the two deepest core segments at 20-25 and 25-30 cm. The ungrazed restoration has a little higher activity, and is consistent throughout the profile, albeit with some low values. The overall activity was 8.3% of that of the dry meadow control (Table 1).

The 10 year old restoration (Old Shilo) had significantly higher activity (at  $p < 0.001$ ) down the profile as compared to the five year old site, with the grazed and ungrazed profiles having activities of 19.6% and 32.4% of the control, respectively. There is marked development of a profile, especially in the ungrazed site. Fairley (1985) demonstrated that grazing led to a marked reduction in the quantity of roots produced on areas re-instated after opencast coal mining in the UK. This is an effect of the constant defoliation leading to a reduction in the above-ground photosynthetic area. This in turn would lead to less root exudate and turnover in the grazed areas as compared to the ungrazed areas.

This data suggests that there is a strong influence of both age of restoration and type of management regime on the development of activity profile. The carbon data shows a poor discrimination between the different areas (Fig. 2). Although the differences between the unworked and worked areas are clear, the differences between age of restoration and management practice are far less clear. The overall levels of carbon in the worked areas (6.13 to 8.04% profile average) are very similar to those in the dry meadow at Shilo (7.81%),

Fig. 1. Microbial Activity  
 (Formazan formation  $\mu\text{g g}^{-1}$  soil  $24\text{h}^{-1}$ )

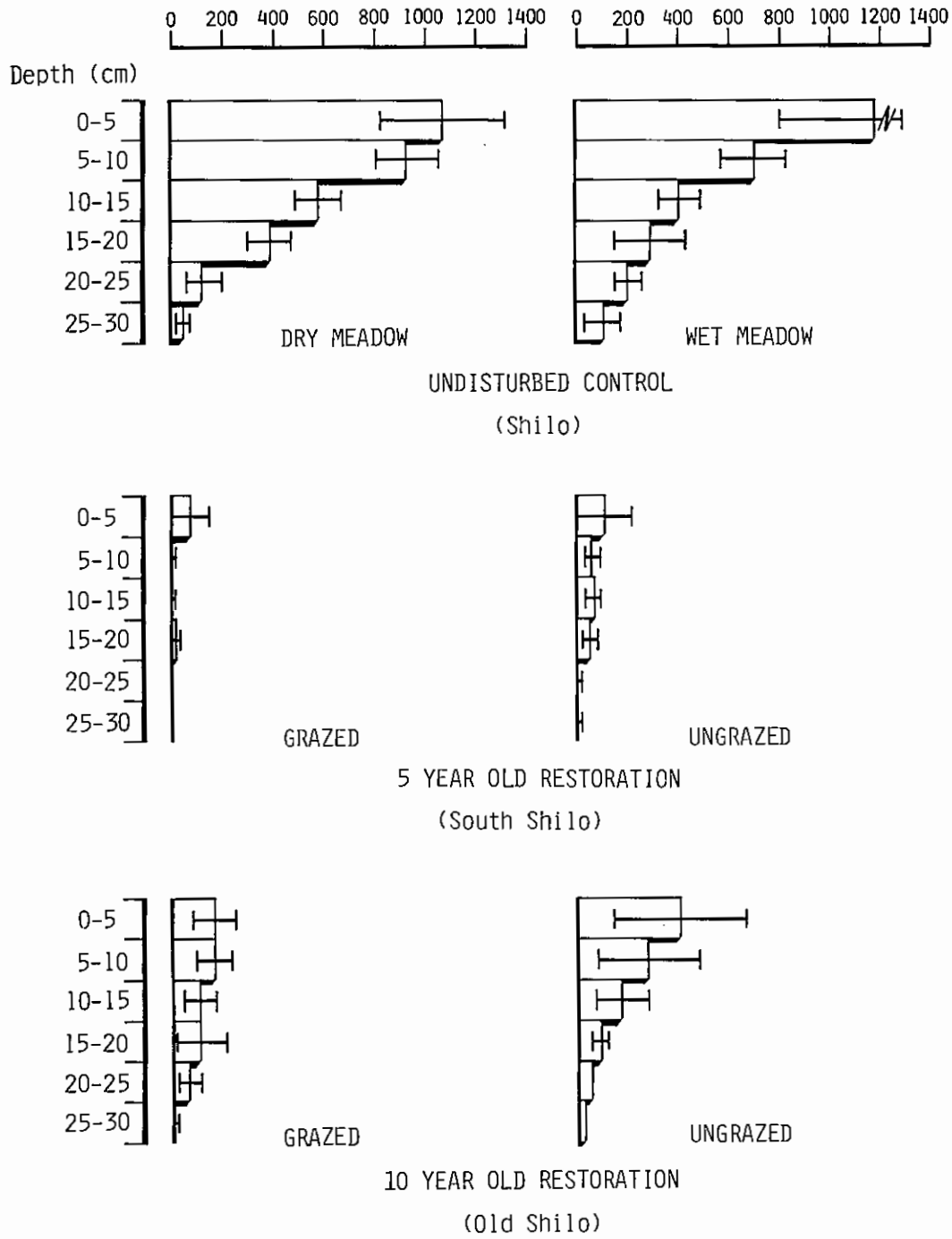


Fig. 2. Total Carbon (%)

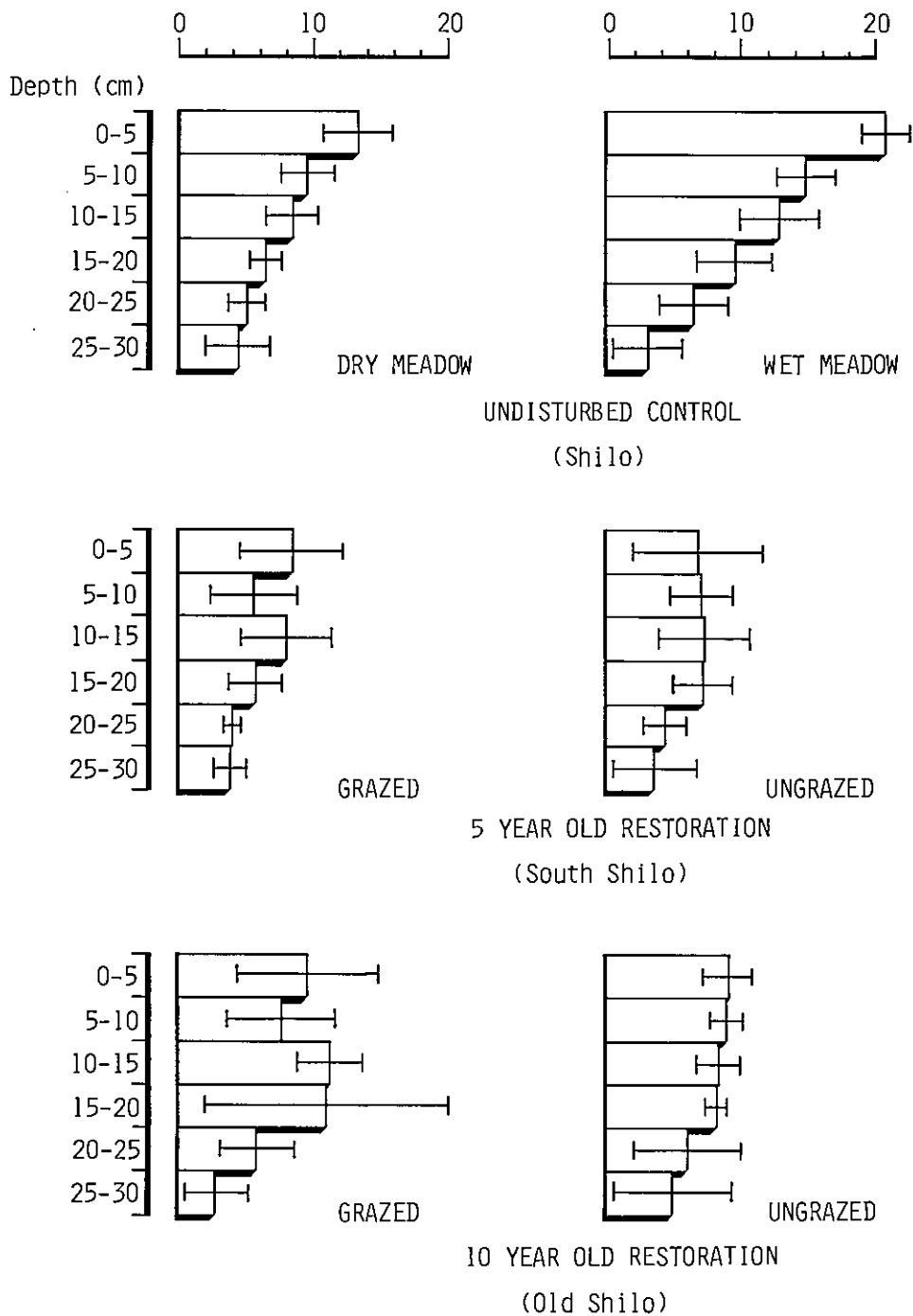


Table 1. Comparison of whole profile (0-30 cm) activity averages

Area	Activity ( $\mu\text{g g}^{-1} 24\text{h}^{-1}$ ).	% of dry meadow control
Shilo dry meadow	515	100 a
Shilo wet meadow	478	93 a
S.Shilo grazed	13	3 d
S.Shilo ungrazed	43	8 cd
O.Shilo grazed	101	20 bcd
O.Shilo ungrazed	167	32 b
Kirk cereal field	140	27 bc
Kirk pasture	210	41 b
Buckhead pasture	580	113 a

Letters following percentages indicate homogenous groups, as determined by multiple range test following ANOVA, at 95% confidence and  $p < 0.001$

indicating that the total amount of organic matter has returned to its former levels in the 10 year old restorations, but the profile is rather uneven with no clear depth trends as seen in the microbial activity values (Fig.1).

Table 1 allows a comparison of the data from the various Shilo sites with the 0-30 cm profile averages from three undisturbed areas sampled previously, which consisted of of a pastures and a cereal field at Kirk and Buckhead, (Harris and Birch 1989). Very similar activity was observed in the undisturbed Shilo areas as compared to the undisturbed pasture at Buckhead, which are all considerably more active than the mined areas at Shilo. However, the Old Shilo restorations have similar levels of (not significantly different at  $p < 0.001$ ) activity to the undisturbed areas at Kirk (Table 1). If this is the desired end point then the ungrazed area at Old Shilo

would appear to be very close to achieving it. From this small data set, it would appear that some 25-30 years may be required for the 'full' meadow profile to reappear, at these rates of increase. However, more detailed and intensive sampling at both areas would be required to confirm this. A database of basic ecosystem types containing information on the size, composition and activity of the soil microbial community, ensuring accountability of seasonality and spatial variation. A survey of early, mid and late successional types could be carried out to provide a computer based ordination, perhaps using principal components and factor analysis in conjunction with classification procedures such as cluster analysis. This could then be used to assess the state of restored ecosystems in terms of undisturbed systems. Further, it could give early indications as to which direction the system needed to be 'pushed' in

order to achieve the desired restoration end point.

### Conclusions

This study would appear to suggest that microbial activity profiles are potentially useful tools for assessing restoration success and discriminating between different management practices, and that further work is required in this area of research.

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