ASPECTS OF MICROBIAL AND PROTOZOAN ABUNDANCES IN SIGNY ISLAND FELLFIELDS

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ABSTRACT. Determinations of the abundances of protozoan and microbial taxa (bacteria, fungi, yeasts, algae, flagellates and testate amoebae) were made in two fellfield sites on Signy Island. Samples for counts were taken along short transects located to include a variety of substrata: fines, coarse, *Usnea-*colonized stones and *Andreaea* cushions. All materials except *Andreaea* had low moisture and organic content. Bacteria and flagellates showed declining abundances along transect gradients from fines to coarse. Highest protozoan numbers were associated with *Andreaea* cushions, but microflora were lower in these than in unvegetated materials.

INTRODUCTION

The microbial communities of habitats dominated by peat-forming bryophytes on Signy Island in the maritime Antarctic were the subject of detailed studies between 1962 and 1982 of their species composition, population dynamics and eco-physiology. Syntheses and summaries of the results have been prepared by Wynn-Williams (1980) and Davis (1981). However, the microbiology of Signy Island's mineral substrata, many of which are sparsely vegetated by lichens and short-cushion mosses, has been studied comparatively little, although these are the most widespread and commonly occurring terrestrial ecosystem in the maritime Antarctic.

Preliminary observations between 1963 and 1969 on the microbiota of glacial debris indicated that, in midsummer, the uppermost 3 cm of such till contained an estimated 10³ filamentous fungi and 10⁵–10⁶ bacteria (Bailey and Wynn-Williams, 1982), and flagellate Protozoa (Smith, 1969) (colony-forming units g⁻¹ dry weight). Observations during 1972–74 on algae in the 0–1.5 cm horizon of mineral materials (Broady, 1979) gave counts of 10⁵ colony-forming units cm⁻². It was thus apparent that the unvegetated, or sparsely vegetated, skeletal soils on Signy Island supported an autotrophic and heterotrophic community, although the numbers recorded were, in general, one or two orders of magnitude lower than those for the same taxa inhabiting Signy Island moss peats and grass loam soil (Smith, 1973; Broady, 1979; Bailey and Wynn-Williams, 1982).

The Fellfield Ecology Research Programme (FERP) aims to test the hypotheses that the physical and chemical nature of fellfield substrata regulates the development of the ecosystem and that water availability, rather than temperature, is the direct limiting influence upon the biota. Maritime Antarctic fellfields are characterised by extensive areas of patterned ground in the form of sorted stone circles, polygons, nets and stripes of stones and fines (Chambers, 1966a). There is some colonization of the ground, the

degree varying with locality, by fruticose lichens and moss cushions, mostly *Usnea* spp. and *Andreaea* spp. (R. I. L. Smith, 1972). Fellfields are subject to diurnal fluctuations of temperature up to 30°C (Chambers, 1966b). They thus present an opportunity to investigate the ecology of microorganisms in unstable conditions were habitat colonization, survival and establishment depend upon adaptations to extremes of physical factors.

The aim of the present investigation was to provide an initial statement of the abundance of various protozoan and microbial taxa inhabiting the range of conditions existing in fellfields on Signy Island, prior to detailed investigations of their ecophysiology. Logistics constrained the number of counts that could be performed upon samples immediately after collection. Therefore, samples were taken along transects located so as to include as wide a variety of substrata as was conveniently possible, namely fellfield fines, fellfield coarse, *Usnea*-colonized stones and *Andreaea* cushions. Thus, the abundance of each microbial taxon could be compared between samples and inspected for association with various physical properties.

DESCRIPTION OF THE SITES AND SAMPLING PROCEDURE

Transects were located at two FERP sites on Signy Island: Factory Bluffs and Moraine Valley (British Antarctic Survey, 1982). The Factory Bluffs site was sampled on 24 January 1984. Weather during the preceeding 10 days was characterized by a small diurnal air temperature range (c. 3 deg. d^{-1} between -2.3 and $+6.9^{\circ}$ C), dense cloud cover, little sunshine (0.8 h d^{-1}), and little precipitation (0.3 mm d^{-1}). Daytime ground surface temperatures at the site varied between +6.4 and $+20.3^{\circ}$ C compared with a summer average of c. 5.0° C. On the sampling date, calm strong insolation conditions induced ground surface temperatures (recorded by D. W. H. Walton) up to 18 degrees above ambient screen temperatures. The fellfield materials at the time of sampling were thus in a drier and warmer state than average, but not outside the range commonly experienced. The transect at Factory Bluffs (Fig. 1) consisted of five sampling points at 12-cm intervals along a radius of a sorted circle on level ground. The central patch of fine clay and silt 40 cm in diameter was surrounded by concentric annuli of unvegetated coarse, coarse bearing *Usnea* sp., and colonies of *Andreaea* spp. coalesced to form more or less continuous stands of moss.

The Moraine Valley site was sampled on 1 February 1984. Since the Factory Bluffs sampling eight days previously, ambient temperatures had been cooler $(-1.1 \text{ to } +4.6^{\circ}\text{C})$, with dense cloud cover, negligible sunshine (0.09 h d^{-1}) , and daytime ground temperatures at the site between +1.4 and $+8.8^{\circ}\text{C}$. At sampling, temperatures at ground surface and at 5 cm depth were close to equilibrium with ambient conditions between -1.0 and 0.0°C . The substratum was thus in a state of incipient freezing, a condition that occurs frequently in the maritime Antarctic summer. The transect (Fig. 1) consisted of four sampling points at 15-cm intervals across a stone stripe on sloping ground. The 50-cm-wide stripe of fines was bordered on either side by 15-cm-wide bands of unvegetated coarse, then by *Andreaea* cushions.

Fines and moss in the 0-3 cm horizon were sampled with a sterilized stainless steel corer 1.5 cm in diameter and coarse removed with a sterilized stainless steel spoon. Samples were sealed in polythene bags and transported within one hour to laboratories at Signy Island station. About 40 g of continuous material were taken at each sampling point and subdivided for determinations of physical properties and counts for bacteria, yeasts, algae, flagellate Protozoa and testate rhizopods.

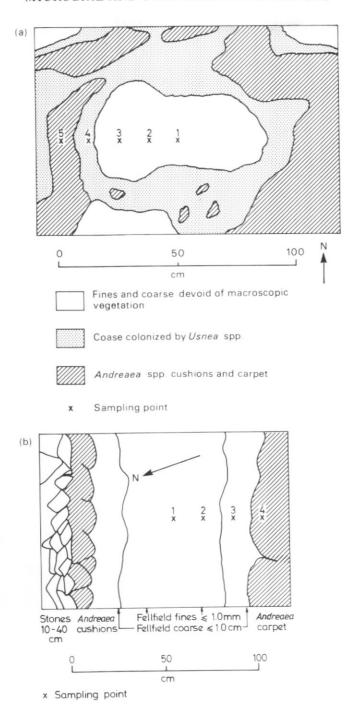


Fig. 1. Sketch map of the areas of the sampling transects at FERP Factory Bluffs (a) and Moraine Valley (b) sites.

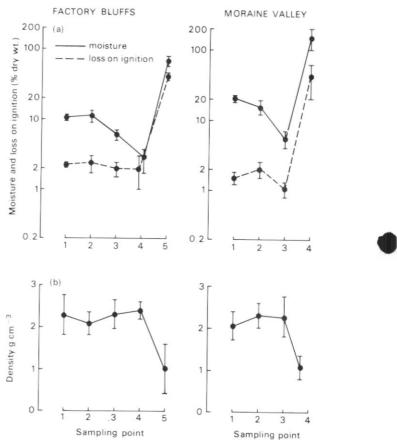


Fig. 2. Moisture content and loss on ignition (a), and density (b), of samples of fellfield materials from FERP sites.

LABORATORY METHODS

The density of each sample was determined by measuring the volume displacement of weighed sub-samples in a measuring cylinder, the moisture content by oven drying at 90°C for 24 h, and loss on ignition by incinerating in a muffle furnace at 500° for 6 h.

Viable counts (colony-forming units) of bacteria, fungi and yeasts were determined by the methods of Miles and Misra (1938). Ten-fold serial dilutions from 10^{-1} to 10^{-5} were made of fresh in 1/4 strength Ringer's solution with $2.5~{\rm g}\,{\rm l}^{-1}$ of sodium hexametaphosphate to act as surfactant. $20~\mu{\rm l}$ of each dilution were pipetted onto Petri plates in triplicates. Two different media were used: 1/10 strength casein peptone starch with 0.005 per cent actidione and Sabouraud dextrose agar with 0.005 per cent chloramphenicol. The plates were incubated at $15^{\circ}{\rm C}$ for up to fourteen days.

Direct counts of algae were performed by the method of Tchan (1952) on a suitable dilution from the series previously prepared. One-millilitre aliquots were passed through 0.45- μ m membrane filters. Filters bearing residue were mounted on slides of algal cells counted directly by epifluorescent microscopy utilizing the autofluorescent nature of chlorophyll pigment.

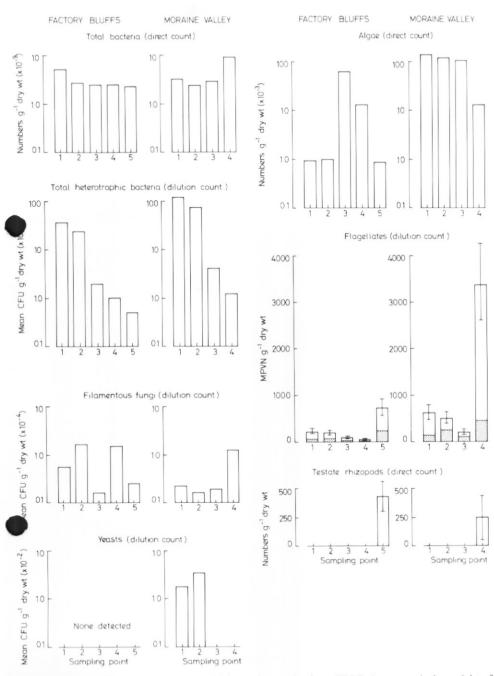


Fig. 3. Abundance of protozoan and microbial taxa in samples from FERP sites per unit dry weight of the 0-3 cm horizon. CFU denotes colony-forming units; MPVN denotes most probable viable numbers.

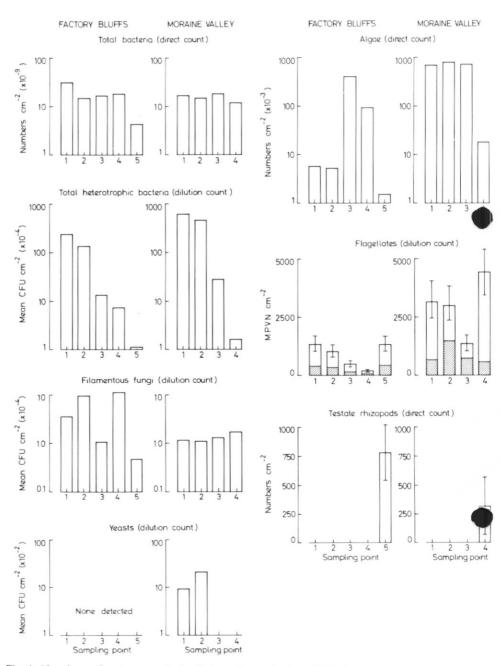


Fig. 4. Abundance of protozoan and microbial taxa in samples from FERP sites per unit area of the 0–3 cm horizon. CFU denotes colony-forming units; MPVN denotes most probable viable numbers.

Direct counts of bacteria were performed on the 10^{-3} dilution of the previously prepared series by the methods of Clarholm and Trolldenier (Wynn-Williams, 1979). Two 5- μ l aliquots were transferred to a PTFE-coated multispot slide (SM010 Hendley, Essex) and stored in the dark at 0°C. The bacteria were stained with FITC (Babiuk and Paul, 1970) and counted directly by epifluorescent microscopy using FITC excitation and BG23 red-cutting filters. Ten microscope fields per spot were counted.

Most probable viable numbers of flagellate Protozoa were estimated using the principle of the method of Singh (1955) as modified by Darbyshire (1973). Both total and cyst estimates were obtained by performing counts on two subsamples, one treated with 0.075 M HCl (Sneddon, 1983) for two hours and one untreated. Doubling serial dilutions from 1/5 to 1/10240 were prepared with 1.8-strength Ringer's solution. Cultures were established using $250-\mu l$; dilution and $50-\mu l$ suspension of Aerobacter aerogenes (NCIB 418) as food supply, with eight replicate cultures at each dilution. Cultures were incubated for fourteen days at 15° C.

Testate rhizopods were counted directly by phase-contrast microscopy as numbers beserved in $50-\mu l$ drops of the first six of the previously prepared serial dilutions.

RESULTS AND DISCUSSION

The physical properties of the samples are shown in Fig. 2. The results show low moisture and organic content for all materials except moss cushion. Higher values of loss on ignition were recorded for unvegetated fines and coarse at Factory Bluffs than at Moraine Valley, possibly owing to the latter's greater horizontal stability. However, the Moraine Valley unvegetated materials had a higher moisture content and also generally higher microbial counts; these may have been a consequence of the difference in meteorological conditions around the sampling dates, so it is possible that this contrast between the sites was atypical. Numbers of microbial taxa per gram dry weight are given in Fig. 3 and per cm2 in Fig. 4. The numbers of bacteria and yeasts from both sites were comparable to those recorded from sorted circle fines at Jane Col, Signy Island, during 1980-81. D. D. Wynn-Williams (personal communication) obtained values of 77.1 + 21.6 for total heterotrophic bacteria and 0.16 ± 0.06 for yeasts as colony-forming units × 10⁴ g⁻¹ dry weight. His counts of filamentous fungi $(0.5+0.7\times10^4)$, however, whilst similar to earlier counts from glacial moraines, are an order of magnitude lower than the present fungi counts; this may be a reflection of a significantly greater development of the heterotrophic microbial communities at the much older sites at Moraine Valley and Factory Bluffs.

Examining the microbial counts along the transects within each site, no consistent trend for all taxa is apparent. However, bacterial and flagellate Protozoa showed a declining trend along the transects, from fines to coarse, at both sites; this is associated with the gradient of declining moisture as particle size increases. A response to the higher moisture content of *Andreaea* moss-cushion was shown only by the Protozoa, microflora numbers being, in general, lower in the moss-cushion than in unvegetated materials. The observation that testate rhizopods occur only in the moss-cushion is consistent with previous records of Protozoan species distribution (Smith, 1978, 1984).

In conclusion, present data are consistent with the view that the FERP sites at Factory Bluffs and Moraine Valley show a more advanced stage of microbial community succession, associated with their greater age and development of vegetation, than does the high altitude exposed fellfield at Jane Col and the glacier moraines, and that moisture is an important influence on the numbers of viable microbes and Protozoa inhabiting such sites.

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