# TREATMENT EFFECTS ON PERFORMANCE OF N-FIXING LICHENS IN DISTURBED SOIL CRUSTS OF THE COLORADO PLATEAU

Diane W. Davidson,[1,3] Matthew Bowker,[1,2,4] Dylan George,[1,5] Susan L. Phillips,[2] and Jayne Belnap[2,4]

[1]*Department of Biology, University of Utah, 257 South, 1400 East, Salt Lake City, Utah 84112-0840 USA*
[2]*USGS Forest and Rangeland Ecosystem Science Center, 2290 S. West Resource Boulevard, Moab, Utah 84532 USA*

*Abstract.* Biological soil crusts arrest soil erosion and supply nitrogen to arid ecosystems. To understand their recovery from disturbance, we studied performances of *Collema* spp. lichens relative to four experimental treatments plus microtopography of soil pedicels, oriented north-northwest to south-southeast in crusts. At sites in Needles (NDLS) and Island in the Sky (ISKY) districts of Canyonlands National Park, lichens were transplanted to NNW, SSE, ENE, WSW, and TOP pedicel faces and exposed to a full-factorial, randomized block experiment with four treatments: nutrient addition (P and K), soil stabilization with polyacrylamide resin (PAM), added cyanobacterial fiber, and biweekly watering.

After 14.5 mo (NDLS) and 24 mo (ISKY), both visual rankings of lichen condition and measures of chlorophyll fluorescence were generally higher at ISKY than on more fertile but less stable soils at NDLS. On ENE and NNW pedicel faces, both these values and nitrogenase activity (NDLS only) exceeded corresponding values on WSW and SSE faces. Treatment effects were site specific and largely negative at NDLS; both nutrient and cyanobacterial addition led to poorer lichen condition, and added nutrients led to reduced fluorescence. Responses to nutrients may have been mediated partly by disturbance of unstable soils and by competition with cyanobacteria.

In a separate experiment investigating recruitment responses to adding fungal spores or *Nostoc* cells, rates of *Collema* establishment responded significantly to the former but not the latter. Low establishment overall suggests that natural recruitment may occur mainly from iscidia or thallus fragments, not spores.

Measured simultaneously on artificial soil pedicels at NDLS and ISKY, both deposition and erosion declined at NDLS across the four pedicel microaspects as: WSW > SSE > NNW > ENE (or ENE > NNW), during fall and spring trials. Patterns were similar at ISKY, but WSW ≈ SSE for spring deposition, and deposition did not differ by microaspect in fall. Greater deposition at ISKY, despite higher abundance of cyanobacteria, may be explained by stronger wind velocities.

Together, microtopographic differences in erosion, microclimate, and nutrient regimes help explain variable lichen performance, but microtopography influenced lichen performance more consistently than did any treatment. Demonstrated effects of pedicel development in crust recovery concur with prior surveys showing greatest microbial biomass and/or cover on ENE and NNW exposures at various spatial scales.

*Key words: biological soil crusts;* Collema; *Colorado Plateau; cryptobiotic crusts; cyanobacteria; lichen; microbiotic crusts;* Microcoleus vaginatus; *microtopography; nutrients; Utah.*

## Introduction

In semiarid rangelands of the southwestern United States, a collection of cyanobacteria, algae, lichens, and mosses form biological soil crusts, also known as cryptobiotic, cryptogamic, or microbiotic crusts. In pristine form, these crusts account for at least as much soil surface cover as do vascular plants, and provide a num-

ber of ecosystem services (reviewed in Harper and Marble 1988, West 1990, Johansen 1993, Belnap and Lange 2001). In cold, arid ecosystems, nitrogen fixation by microorganisms in soil crusts can be the dominant source of nitrogen (Evans and Ehleringer 1993). The higher soil moisture and fertility associated with soil crusts can translate into greater nutrient levels and higher seedling survivorship in vascular plants (Belnap 1995, Belnap and Harper 1995), and perhaps benefit herbivores and other consumers (Harper and Pendleton 1993). Soil crusts can also contribute to soil stability (Belnap and Gillette 1997, 1998), and can have mixed effects on water infiltration and retention by soils (George 2000).

Growing recognition of the importance of biological soil crusts to ecosystem processes has led to increased

concern about the impact of anthropogenic disturbance by both non-native grazers and various recreational users. Estimates of time for unaided recovery of disturbed biotic crusts range widely from a few years to >1000 yr (Harper and Marble 1988, Belnap and Warren 1998). Calculations based on visual surveys of cover and pedicellation of frost-heaved soils can underestimate recovery times by ignoring superficially unapparent but persistent effects of disturbance on cyanobacterial biomass and on physiological functions such as nitrogenase activity (Belnap 1993, Belnap et al. 1994).

Despite recognition of the importance of biological soil crusts, and concerns about degradation, we know little about processes limiting their growth and recovery after disturbance, i.e., about ecological succession. Delays in recovery are clearly due in part to intrinsically low growth rates of constituent species. However, as shown by inoculant experiments (Lewin 1977, Tidemann et al. 1980, Ashley and Rushforth 1984, St. Clair et al. 1986, Belnap 1993), recovery can also be slowed by poor propagule dispersal. Additionally, colonization by late-arriving species could depend on significant site alteration by early colonists, though virtually nothing is known about this aspect of the reestablishment of healthy crusts. Knowledge of the mechanisms limiting colonization, establishment, and growth rates of crustal species is essential for sound management practices. Access to such information could also greatly improve estimates of recovery times, which have necessarily been based on the possibly misleading assumption of linear recovery through time.

### Succession in crusts of the Colorado Plateau

The composition of biological soil crusts changes through time after disturbance in a manner analogous to transitions occurring during primary succession in terrestrial plant communities. However, rather than nitrogen-fixing species arriving immediately after disturbance, such species colonize only after an extended period of soil stabilization. Our study focuses on biological soil crusts typical of calcareous soils in relatively high, cold and arid lands of the Colorado Plateau, encompassing much of southern Utah, northern Arizona, western Colorado, and northwestern New Mexico. Here, the filamentous cyanobacterium *Microcoleus vaginatus* dominates this initial stage of soil stabilization (Belnap and Gardner 1993). Colonizing within just days of soil disturbance (Johansen et al. 1981, 1984), *Microcoleus* is highly mobile (Campbell 1979) and can thus tolerate some burial by sediment. Accounting for up to 95% of the biomass of crusts on this plateau (Belnap and Gardner 1993), *M. vaginatus* secretes a thick, extracellular gelatinous sheath that surrounds live filaments and adheres to soil particles, aggregating them into larger, less erodible clumps (Harper and Marble 1988, Belnap and Gardner 1993). As the organism grows, sheath material is left behind as fiber, which protects against soil erosion and can alter nu-

trient and moisture regimes (George 2000, Belnap and Lange 2001).

*Microcoleus*, which lacks heterocysts, fixes little or no nitrogen; the weak nitrogenase activity consistently associated with it is apparently attributable to free-living heterotrophic bacteria in the sheath matrix (Steppe et al. 1996). Consequently, recovery of the high nitrogen-fixing ability of the soil crust depends on colonization of cyanolichens, especially *Collema tenax* and *Collema coccophorum* (with the photobiont *Nostoc*), which generally arrive years after the cyanobacteria establish (Belnap 1993). These and later arriving lichens and mosses contribute more stability to soil surfaces than do cyanobacteria alone (Belnap and Gillette 1998). However, lichens and mosses grow more slowly than does *Microcoleus*, and they are more susceptible to burial by wind- or water-borne sediments. In one long-term experiment, *Collema* accounted for just 0–0.5% of crust cover 9 yr after the establishment of plots from which cyanobacteria had been ''scalped,'' and despite an abundant propagule source just 0.25 m away (Belnap 1993). The poor colonization rate of *Collema* on scalped plots suggests that the environment for *Collema* might somehow be more favorable with abundant cyanobacteria and their fibers present.

As crusts develop, they assume a rugose microtopography with parallel ridges consisting of small pedicels (mounds), typically 5–15 cm long, up to 7 cm tall, and aligned in a NNW–SSE direction (Fig. 1 and George et al. 2000). These mounds likely form due to a combination of abiotic processes, such as soil upheaval during winter freezing and differential downward erosion by water, and biotic processes, such as differential growth of cyanobacteria and *Collema* lichen on the different mound faces (George et al. 2000). *Collema* spp. and *Catapyrenium squamulosum* are generally the first lichens to establish, followed by a diverse group of later successional, non-N-fixing lichens. Some of these species also perform differentially well on particular microaspects of the mounds (George et al. 2000; M. Bowker, *unpublished data*).

### Mechanisms of succession in crusts

Better understanding of the mechanisms determining ecological succession in crusts may suggest ways of assisting natural regeneration of crusts after disturbance. We therefore designed experiments to detect mechanisms that may alter rates of succession to *Collema* spp. on *Microcoleus*-dominated mounds. Treatments addressed ways in which the presence and abundance of cyanobacteria and their sheaths may affect the performance of *Collema* lichen. In many vascular plant communities, successional changes appear to track resource ratios, usually nitrogen to light, with plant growth being limited at first by nitrogen and later by light (e.g., Tilman 1982). Since *Microcoleus* is unlikely to alter either the light or nitrogen regimes experienced by *Collema* (see *Succession in crusts of the Colorado*

FIG. 1. Rugose microtopography of microbiotic crusts on the Colorado Plateau. Soil ridges are composed of small mounds aligned in a NNW–SSE direction.

*plateau*), and because *Collema* itself fixes nitrogen, other effects are likely involved here. We conjectured that *Microcoleus* could facilitate *Collema* recruitment by stabilizing soils, improving soil moisture status (George 2000), helping to produce the pedicellation that provides microhabitats favorable to *Collema* (George et al. 2000), or altering levels or ratios of nutrients other than nitrogen in ways favorable to the lichens.

As a first step toward exploring successional mechanisms, we investigated the effects of four different treatments on the success of transplanted *Collema* lichen (a mixture of *C. tenax* and *C. coccophorum*): (1) Cyanobacteria—addition of *Microcoleus* fibers; (2) PAM—application of polyacrylamide spray, commonly used as a soil stabilizer in reclamation; (3) Water—biweekly water supplements; and (4) Nutrients—biweekly supplements of nutrients in water solution. Not unexpectedly, since *Collema* spp. fix nitrogen, preliminary experiments in an outdoor laboratory indicated that nutrient consumption vectors (see Tilman 1982) were steeper for phosphorus (P), and (somewhat less so) for potassium (K), than for nitrogen (J. Belnap, *unpublished data*). Our nutrient amendments therefore focused on P and K. In addition to these treatments, because earlier work had suggested that mound microtopography significantly affected lichen distribution (George et al. 2000), we compared the performances of lichen plugs on five different microaspects of soil mounds.

## METHODS

### Study sites

In late March 1997, we established two permanent study areas in Canyonlands National Park: in the Nee-dles District (NDLS, 109°48′ W, 38°10′ N), 20 miles southwest of Moab, Utah, and in the Island in the Sky District (ISKY, 109°44′ W, 38°28′ N), 20 miles northwest of Moab. Mean annual precipitation at the two sites is identical (21.7 cm, Western Regional Climate Center 1965–1999), and during the course of our study, was slightly wetter than normal at both sites (by 4.1 cm at NDLS and 4.6 cm at ISKY for 1997–1999). Soils at both study areas are shallow, fine sandy loams (2 cm to ~1 m deep), and sparse herbs and a pinyon–juniper association (*Pinus edulis* and *Juniperus osteosperma*) comprise most of the woody vegetation. NDLS soils were more sandy and fragile than ISKY soils, with less cyanobacterial cover (Table 1) by *M. vaginatus* (microscopically the predominant cyanobacterial species).

Seasonally, mid-October through mid-May coincides with the highest average ratios of precipitation : evaporation at both sites. Growth of microorganisms should be greatest in October–November and February–March, when warm daytime temperatures combine with relatively high soil moisture to create conditions favoring metabolic activity. Winds blew predominantly from the south and southwest throughout the year (1999) at NDLS, and from the southeast (summer) or the northwest and east-southeast (remaining months) at ISKY (NPS 1993, 1998, 1999).

### Design of long-term field experiment

At each study site, we established irregularly shaped plots including multiple patches of soil crusts on shallow soils surrounded by exposed sandstone and containing numerous linear mounds with cyanobacterial but no *Collema* spp. cover. Within plots, we marked

TABLE 1. Characteristics of soils at the Needles and Island in the Sky sites.

| Soil characteristic | Needles | Isky |
|---|---|---|
| Chlorophyll content ($\mu$g chlorophyll $a$/g soil) | 3.97 | 9.24 |
| Percentage sand | 76 | 80 |
| Percentage clay | 9.3 | 5.3 |
| Percentage silt | 14.7 | 14.7 |
| Percentage $CaCO_3$ | 6.1 | 1.7 |
| Percentage organic matter | 3.75 | 2.65 |
| Cation exchange capacity | 6.52 | 3.26 |
| P (ppm) | 6.0 | 4.3 |
| Ca-exchangeable (ppm) | 3170 | 1314 |
| Mg-exchangeable (ppm) | 94.4 | 31.3 |
| K-exchangeable (ppm) | 72.2 | 48.2 |
| Na-exchangeable (ppm) | 47.2 | 50.3 |
| K-available (ppm) | 48.0 | 28.8 |
| Zn (ppm) | 0.35 | 0.34 |
| Fe (ppm) | 2.98 | 2.04 |
| Mn (ppm) | 3.9 | 5.6 |
| Cu (ppm) | 0.43 | 0.21 |
| P : $CaCO_3$ | 0.98 | 2.53 |

*Notes:* Soils at both sites are Rizno fine sandy loam of the Lithic Ustic Torriorthents. Twenty or more samples, to 10 cm depth and spread over the study area, were combined to form a single composite sample at each site. Chlorophyll *a* determinations are for the top 33 mm of soil.

160 such mounds and characterized their sizes as small (surface area $\approx$ 40–50 cm$^2$), medium (51–70 cm$^2$), or large ($\geq$70 cm$^2$). Mounds within 0.75 m radius of shrubs were excluded due to possible effects of shading, nutrient inputs, and allelopathy (Rychert and Skujins 1974). Four treatments were applied to the mounds in a full-factorial, randomized block design, with blocks corresponding to the two study sites, and randomization within each block. This design resulted in 16 distinct mound categories, each assigned to a different, randomly determined color of marker flags already in place. We then mapped the (thoroughly intermixed) mounds in different treatment categories.

Treatments were applied to the mounds on the following day. Half of all mounds within each treatment received applications of dry *Microcoleus* spp. (mainly *M. vaginatus*) sheath material obtained by sieving *Microcoleus*-dominated topsoils collected in March 1997 outside the Park boundary at ISKY. Sheath material, ~10% of it containing live organisms, was separated by a number 30 (600-$\mu$m) soil sieve and stored dry in coin envelopes. We added just enough sheath to thinly cover mounds: 3 g, 4.5 g, and 6 g for small, medium, and large mounds, respectively. Spray-wetting of mounds with deionized (DI) water (15, 30, and 45 mL, respectively) before and after additions sealed fiber to mounds to prevent its erosion. Two of three remaining treatments consisted of spraying fixed amounts of DI water with or without nutrients. Prior to nutrient addition, we applied 15, 30, or 45 mL of DI water (depending on mound size) to mounds assigned to the No Water treatment, to compensate for watering during establishment of other treatments. To mounds in the biweekly Water treatment, we applied 30, 60, or 120 mL of DI water. After applications of cyanobacteria and water, half of all mounds in each treatment category received nutrient applications (15, 30, or 45 mL) containing phosphorus at 7 $\mu$g/g and potassium at 150 $\mu$g/g, from $K_2HPO_4$ and $KHCO_3$, respectively. Nutrient application followed that of water, to avoid eroding nutrients from the mounds. Lastly, half the mounds in each treatment were sprayed with polyacrylamide (PAM, Soil Master WR, Environmental Soil Systems, Granada Hills, California), a synthetic resin emulsion and co-polymer of methacrylates and acrylates/acetates (tripolycate). PAM, at concentrations of 5.5 mL to 1 L of DI water, was sprayed to yield ~10 mL per mound. PAM was also applied to all footpaths to prevent erosion of disturbed sand onto experimental mounds.

Following treatment applications, 5 mm diameter, circular plugs of *Collema* lichen from outside the Park near ISKY were added to the mounds. Plugs were assigned haphazardly to mounds and to positions on their NNW, ENE, SSE, WSW, and TOP exposures, where they were gently inserted into pre-excavated holes of the same diameter.

Experiments were established on 22–23 March 1997 at ISKY, and on 25–26 March at NDLS. Except after significant natural precipitation, we returned at 2-wk intervals through 14 March 1998, to renew water treatments and at 3-mo intervals to reapply nutrients (in each case, 30 mL on small and medium mounds, and 45 mL on large mounds). PAM was reapplied at ~6-mo intervals, but cyanobacterial additions were not repeated. During hot seasons, treatments were applied in early morning or late in the day. After 1 yr, we halted watering and nutrient treatments due to erosion of soils around lichen plugs, especially on fragile NDLS soils. Finally, on 18 June (ISKY) and 20 June (NDLS) 1997, we replaced plugs missing at sites initially overlooked or crushed by native animals.

*Monitoring of long-term field experiment*

In March of both 1998 and 1999 (ISKY only), we evaluated conditions of lichen plugs by scoring them visually as good, fair, and poor, corresponding approximately to $\leq 30\%$ loss, $30\% < \text{loss} < 70\%$, and $\geq 70\%$ loss, respectively. (Positive growth was not noted during this period.) Missing plugs were excluded from analysis, since most or all of these were lost to stochastic events, especially trampling by native animals.

We used log-linear models (SYSTAT Version 9, SPSS 1999) to evaluate treatment effects on plug conditions in the context of the factorial design. We explored only models with plug Conditions (our response variable) contained in all higher order effects, and examined only those effects for significance. For each effect, the program provides a hierarchical chi-square ($\chi^2$) statistic to test the significance of deleting that term. If this test is not significant, then neither the focal term nor higher order effects containing that term are significant, and both terms can be deleted. Iteration of this procedure identifies the most parsimonious model, i.e., that including all significant terms but excluding nonessential terms, except the first-order variables (treatments and Condition) required for higher order models. We began with models containing all possible three-variable effects and then deleted nonsignificant two- and three-variable terms to increase the fit to the whole model. (High $P$ values in a likelihood ratio $\chi^2$ statistic show that data fit the model.) We do not report $\chi^2$ values from the hierarchical test, but rather those testing the significance of removing individual (nonhierarchical) terms from the final model. Finally, our first round of analyses included Site as a variable to distinguish results at ISKY and NDLS. After finding marked site differences, we analyzed data for each site independently.

*Physiological conditions of plugs*

At the end of our experiments, we harvested subsets of plugs (ENE- and WSW-facing plugs at NDLS) or all surviving plugs (ISKY) and evaluated their physiological states by quantifying chlorophyll fluorescence and nitrogen (N) fixation rates. Harvests took place on 28 and 31 May 1998 at NDLS and on 18–19 June 1999 at ISKY. Plugs were moistened, removed, placed flush atop essentially sterile sand in 2 cm diameter, 3.3 cm deep, nalgene vials and then air dried. Associated cyanobacteria were removed. At the USGS Moab laboratory, the lichens were covered with tissue paper and stored for 24 h in the dark before uncovering and hydrating them with DI water in light, to compensate for any environmental differences experienced by plugs before harvesting.

Chlorophyll fluorescence and N fixation (ISKY only) were quantified on 1–3 June 1998 (NDLS plugs) and 21–24 June 1999 (ISKY plugs). Fluorescence was remeasured on 2–4 August for ISKY plugs after loss of original data due to computer failure. Prior to remeasurement, plugs were stored dry for 6 wk under warm, dark conditions, then preconditioned by exposure to wet/dry cycles simulating typical summer conditions. Approximately 150 samples from each mound aspect were placed on trays oriented toward those respective directions at $45°$ angles from horizontal, or on horizontal platforms (plugs from mound TOPs). We then subjected them to two 5-d wet/dry cycles by administering 5 mm and 3 mm of DI water during the first 3 d, and 2-d dry periods immediately thereafter.

*Chlorophyll fluorescence.*—To measure the physiological states of the photosynthetic apparatus in the lichens, we determined chlorophyll fluorescence activity on a pulse-amplitude-modulated fluorometer (Model-2000, WALZ, Effeltrich, Germany) and related Fv/Fm (optimal quantum yield, linearly related to the quantum efficiency of photosynthesis) to mound microaspects and treatments. The fluorometer's fiberoptic head was placed $\sim 3$ mm from the lichen.

*Nitrogenase activity.*—We estimated nitrogenase activity (NI) by acetylene reduction (Stewart et al. 1967) and related results to mound microaspects and experimental treatments. Sample tubes were corked at both ends with rubber stoppers, one fitted with a self-healing septum, and injected with acetylene to bring the internal concentration to $>10\%$. Immediately after injection, samples were placed in an incubator at $25°C$ for 4 h, then removed and shaken lightly to homogenize gases. Gas samples were extracted and run through the Shimadzu GC 14-A gas chromatograph (Columbia, Maryland), to analyze for acetylene and ethylene concentrations. Gas chromatograph units were converted to nanomoles of $C_2H_4$ per square centimeter per hour. Plug surface areas were categorized as small, medium, and large, corresponding to condition designations (good, fair, and poor) of plugs in field censuses. For a representative 10 plugs in each size class, we estimated rectangular surface areas as the products of maximum plug diameters and perpendicular diameters, measured to an accuracy of 0.01 mm with a Mitutoyo digital caliper (Rochor Centre, Singapore). Means for each relative size category were assigned to all plugs in those categories.

*Data analysis*

At both sites, untransformed Fv/Fm ratios were normally distributed with equal variances, both across microaspects and over treatment categories for every treatment ($P > 0.05$ in Levene's and Bartlett's tests). We used a full-factorial ANOVA in JMP-IN (SAS Institute, Version 3.2.6, 1996) to analyze each data set for treatment effects and plug microaspect. Both unequal variances and nonnormal distributions characterized NI data from NDLS, even after transformation. Thus, we used log-linear models to analyze data ranked into three categories: 0 for $NI = 0$; 1 for $0 < NI < 0.4$, and 2 for $NI > 0.4$. Many zero cells precluded

analysis of both the full factorial model and submodels with more than three treatment variables, including Microaspect. We began with all possible three-variable models and eliminated terms as justified by hierarchical test statistics.

### Lichen establishment from fungal spores and cyanobacterial cells

We sought to determine whether a lack of *Collema* spores and/or cyanobacterial (*Nostoc*) cells might limit recolonization of disturbed crusts. At NDLS, we selected, marked, and numbered 60 mounds lacking lichen, moss, and dark cyanobacterial cover. On ENE and WSW bases of each mound, we marked the centers of 2 cm diameter, circular monitoring sites with short wires. A quarter of the 60 mounds were then assigned haphazardly to each of the following treatments: (1) +Spores, −*Nostoc*; (2) −Spores, +*Nostoc*; (3) +Spores, +*Nostoc*, and (4) −Spores, −*Nostoc*. Apothecia-bearing thallus material of *Collema* spp., from outside the Park's Needles District, was mounted upright on plastic disks with glue. Disks were inverted directly over monitoring sites on soil mounds. Wire "legs" on the disks held lichens ~2 cm from the soil surface. In both treatments without spore addition, disks were supplied with infertile thalli as a control for the *Collema* recruitment from fallen isidia or thallus fragments. A mixed culture of *Nostoc*, isolated from *C. tenax* and *C. coccophorum* by J. Johansen, was added in amounts ranging from 10–20 flakes (=clusters of 2–5 cells).

We established these experiments on 30 September 1999, and applied DI water to both thallus material and mounds periodically beginning on 3 October. Frequency of watering (1 mL and 2 mL, respectively, on thalli and monitoring sites) varied inversely with natural precipitation and simulated an above-average rainfall year. We watered seven times before 13 December, when mounds were covered by snow. *Nostoc* was re-inoculated on 2 November, on all +*Nostoc* mounds. Thalli were repaired or replaced when necessary, and disks and lichens were removed from the field on 24 January to repair damage caused by snow. Repaired disks were watered and set out again on 27 January, when they were also rotated among mounds in the same treatment categories to control for differential spore production by lichens on different disks.

The experiment was terminated on 6 April 2000, when 1.9 cm diameter circular cores were made at the monitoring sites of surviving mounds. (Trampling by native animals reduced final sample size to 55.) Cores were transported to the USGS laboratory in Moab, Utah, and examined beneath a stereoscope for presence of *Nostoc* colonies and for counts of *Collema* thalli. Excluded from counts were thallus material suspected of falling from disks: isidia, large fertile thalli, unanchored fragments, and *Collema* with glue or foreign soil particles.

Due to the many zeroes and nonnormal distributions of counts of lichen thalli, we converted thallus counts to binary data (+/−) and analyzed these data with nonparametric log-linear models. We then tested for significant interactions between additions of *Nostoc* and fungal spores and the establishment of lichen (presence of thalli) in cores. In a second model, we examined the effects of fertile thalli and *Nostoc* presence in the final cores (an indicator of efficiency of *Nostoc* addition) on lichen recruitment. The largest models included order-2 but not order-3 terms, because larger models failed to converge on solutions.

### Deposition and erosion in relation to microaspect on soil mounds

To help account for disparities in *Collema* performance across both sites and mound microaspects, we assessed erosion–deposition on four exposures of artificial mounds at both study sites, beginning on 10 March 2000. An 18 cm diameter, 1.5-L funnel was filled with local soil moistened with DI water and inverted to deposit soil in a loose conical mound on the ground. Four ~5 cm diameter impressions were made at about one-third mound height on, respectively, NNW, ENE, SSE, and WSW mound faces. Four 6-m Lautosampler vials (Glass Incorporated, Vineland, New Jersey; 22 mm diameter, 38 mm deep) were inserted into the holes with lips protruding 1–2 mm from mound surfaces. Each of 20 mounds at each site was then firmed and rounded into uniform domes. About 1 cm above each of the vials, a 12 cm plastic straw was inserted, with 3 cm exposed.

Experiments were dismantled on 25 May (ISKY) and 26 May (NDLS) 2000, after recording exposed straw lengths. Labeled vials were cleaned of external soil, and oven-dried with their contents for at least 24 h at 59°C before weighing on a Fisher microbalance (Fisher Scientific, Houston, Texas). Dry mass of deposited soil was calculated as the difference between these values and the masses of vials alone (emptied, washed, and dried again). Soil mass deposited per 1.76 cm$^2$ (the area of the vial at its neck) was converted to values of grams per square centimeter and log-transformed for ANOVA. Differences in exposed straw length at the start and end of the experiment estimated "net erosion" from the mound surfaces. Though reflecting a mixture of erosion and deposition, net change was negative in <4% of cases for pooled data in both spring and fall.

We repeated funnel mound trials at both sites using identical procedures, beginning on 13 October and ending on 20 December 2000. Neither the fall deposition nor the net erosion data (both trials) met assumptions of parametric statistics. Net erosion was analyzed by ordinal logistic regression, with site and microaspect as grouping variables, and the effects of these variables on deposition were analyzed independently by Wilcoxon summed-rank tests.
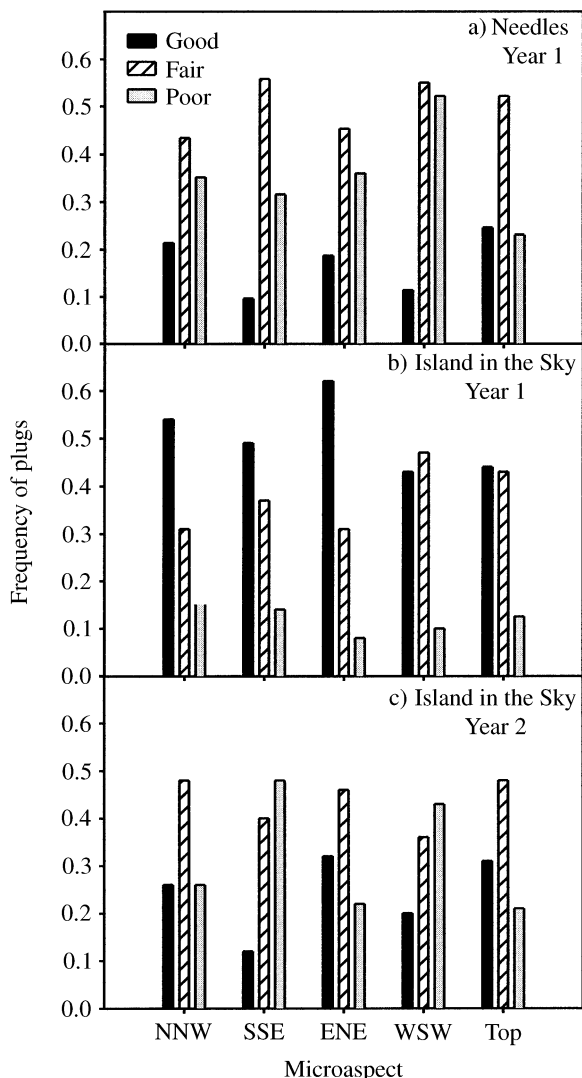
FIG. 2. Conditions of lichen plugs differed across Microaspects at (a) Needles ($\chi^2 = 23.20$, df = 8, $P = 0.0031$), and at (b) Island in the Sky ($\chi^2 = 19.61$, df = 8, $P = 0.0119$) after 1 yr, and (c) at ISKY ($\chi^2 = 60.99$, df = 8, $P = 0.0000$) after 2 yr. At the end of 1 yr, lichen plugs were in better condition at ISKY than at NDLS ($\chi^2 = 208.88$, df = 8, $P = 0.0000$).

## RESULTS

### Plug conditions in long-term field experiment

After 1 yr, field assessments of plug conditions pooled over both sites included a total of 1423 surviving plugs ($N = 694$ at NDLS and 729 at ISKY). The most parsimonious log-linear model with Condition included in all higher-order interactions, contained second-order effects of just Site and Microaspect (LR $\chi^2 = 344.73$, df = 446, $P = 0.9998$, indicating that the data fit the model). Lichen plugs at NDLS were in poorer condition than were those at ISKY (Fig. 2a, b). Combined for the two sites, plug conditions differed

significantly by Microaspect ($\chi^2 = 24.25$, df = 8, $P = 0.0021$), but the interactive effect of Site and Microaspect with plug Condition bordered on significance ($\chi^2 = 14.96$, df = 8, $P = 0.0599$). Significant three-variable terms in the model included the interactive effects of plug Condition and Site with two other treatment variables, Cyanobacteria treatment ($\chi^2 = 11.62$, df = 2, $P = 0.0030$) and Nutrient treatment ($\chi^2 = 11.20$, df = 2, $P = 0.0037$). Neither the PAM nor the Water treatments produced a significant effect in any model ($P > 0.05$ for all higher order effects containing these terms and plug Condition as the response variable). After noting the overwhelmingly important effect of Site in this model, we analyzed the data subsequently for each site independently, and we present the results separately below.

At termination of the NDLS experiment after 1 yr, the most parsimonious log-linear model for this site included just three second-order effects involving plug Condition (LR $\chi^2 = 72.53$, df = 9.8, $P = 0.9748$). Condition interacted significantly with Microaspect, Cyanobacteria addition, and Nutrient addition. Based on frequencies of surviving plugs in good condition, performance was ordered as mound TOPS, followed successively by NNW > ENE > WSW > SSE exposures (Fig. 2a). Plugs in poor condition occurred at lowest frequency on mound TOPS, followed successively by SSE ≤ NNE ≈ ENE < WSW. Under additions of cyanobacteria and of nutrients, slightly lower frequencies of plugs were in good and fair condition, and higher frequencies were in poor condition, compared with plug frequencies under no such additions (Figs. 3a and 4a, respectively). There were no other significant higher-order effects.

The most parsimonious log-linear model for ISKY data after 1 yr (LR $\chi^2 = 189.83$, df = 221, $P = 0.93655$) was that containing plug Condition × Microaspect as the only significant second-order effect. The frequency
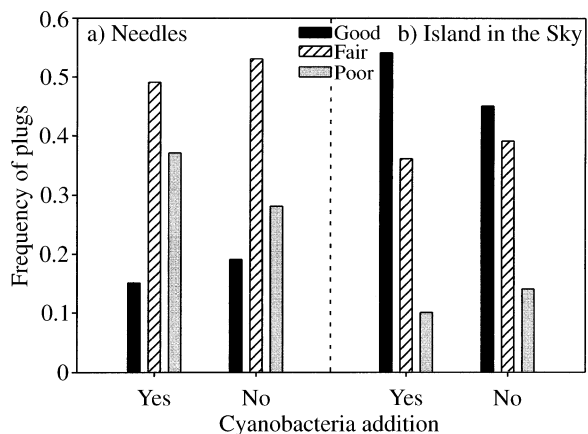


FIG. 3. One year into the experiment, addition of cyanobacteria had significantly reduced the conditions of transplanted *Collema* lichen plugs at (a) Needles ($\chi^2 = 7.02$, df = 2, $P = 0.0299$) but not at (b) Island in the Sky ($P > 0.05$).
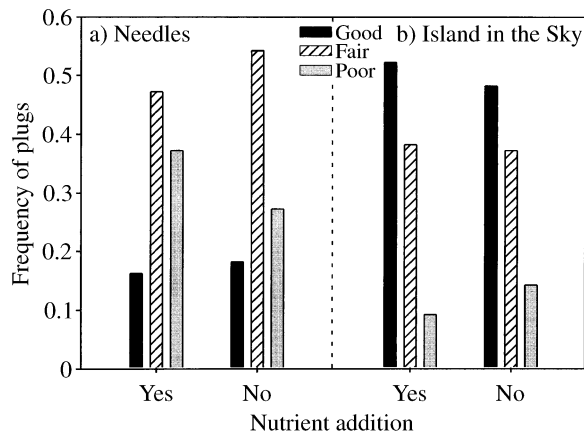
FIG. 4. One year into the experiments, the addition of nutrients (P and K) had significantly reduced the Conditions of transplanted *Collema* lichen plugs at (a) Needles ($\chi^2$ = 8.05, df = 2, $P$ = 0.0179) but not at (b) Island in the Sky ($P$ > 0.05).

of plugs in good condition was greatest on ENE exposures, followed by NNW > SSE > TOP ≈ WSW (Fig. 2b). Those in poor condition were least frequent on ENE exposures, followed by WSW < TOP < SSE < NNW. Trends for higher frequencies of plugs in good condition, as well as fewer in poor condition, on both Cyanobacteria and Nutrient supplements were not significant (Figs. 3b and 4b, respectively).

At termination of the ISKY experiment after 2 yr, the most parsimonious model (LR $\chi^2$ = 487.14, df = 172, $P$ = 0.99482) was that containing Condition × Microaspect as the only significant second-order effect. The frequency of plugs in good condition was greatest on the ENE exposure, followed by TOP > NNW > WSW > SSW (Fig. 2c). Those in poor condition were least frequent on mound TOPS, followed by ENE < NNW < WSW < SSW. There were no other significant higher-order effects.

### Physiological conditions of plugs

*Chlorophyll fluorescence.*—Because *Collema* is heavily pigmented lichen (appearing black to the unaided eye), it absorbs considerable light. Therefore, fluorescence measurements are much lower than are generally recorded for less pigmented lichens, or for vascular plants. After 14.5 mo at NDLS, the five-variable, full-factorial ANOVA was significant ($F_{31, 184}$ = 1.74; $P$ = 0.0138, adjusted $r^2$ = 0.0962, $N$ = 216 plugs). Within this model, significant effects included those of Microaspect alone and Nutrients alone, as well as one second-order and one third-order effect. Plugs on the ENE mound exposures fared better on average than did those on the WSW microaspect ($P$ = 0.0001, least squares means: ENE = 0.21; WSW = 0.15). Fluorescence was significantly depressed on NDLS mounds receiving P and K Nutrient supplements ($P$ = 0.0075, least squares means: + Nutrients = 0.16; − Nutrients

= 0.20). In contrast, the Water treatment enhanced plug performance, but only in the absence of cyanobacteria (Fig. 5). Finally, Cyanobacteria produced a negative effect on fluorescence on fertilized mounds sprayed with polyacrylamide, but a positive effect in the absence of both nutrient addition and soil stablization by PAM (Fig. 6).

For ISKY, where we measured fluorescence for plugs on all five microaspects of mounds, we could not analyze the full-factorial, five-variable model (four treatments and microaspect) due to lost degrees of freedom, since cells for certain combinations of independent variables held no observations. Therefore, we reduced the ANOVA model so as to include up to just fourth-order interactions involving the four treatments and plug microaspect on mounds. This model was significant ($F_{75, 303}$ = 2.87; $P$ < 0.0001, adjusted $r^2$ = 0.2701, $N$ = 379 plugs) and included significant effects of Microaspect, as well as two second-order interactions. In a significant one-way ANOVA of fluorescence vs. microaspect ($F_{4, 374}$ = 36.22; $P$ = 0.0001), a Tukey-Kramer test revealed the ENE and WSW mound faces as the only exposures for which fluorescence levels were statistically indistinguishable (Fig. 7a, $P$ < 0.05). Thus, fluorescence was greatest on the NNW > ENE ≈ WSW > TOP ≫ SSE. The Nutrient × Cyanobacteria interaction effect was significant, with nutrient additions enhancing fluorescence in the absence of cyanobacteria but reducing yield in their presence (Fig. 8a). Watering slightly improved fluorescence in the absence of PAM but led to a decline in yield under its addition (Fig. 8b). Finally, the interaction of Polyacrylamide × Microaspect bordered on significance (Fig. 7b). Fluorescence was greater on ENE exposures than on WSW faces under the PAM addition, but the reverse was true in the absence of PAM.

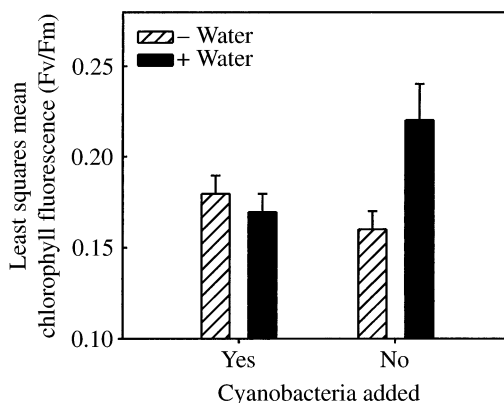*Nitrogenase activity.*—The most parsimonious log-linear model for the nitrogenase data (LR $\chi^2$ = 0.0272,



FIG. 5. One year after the start of the experiment at Needles, chlorophyll fluorescence ($\log_{10}$ Fv/Fm) was enhanced by water in the absence of polyacrylamide, but reduced where this resin emulsion had been applied ($P$ = 0.0425 for interaction term). Error bars represent +1 SE.
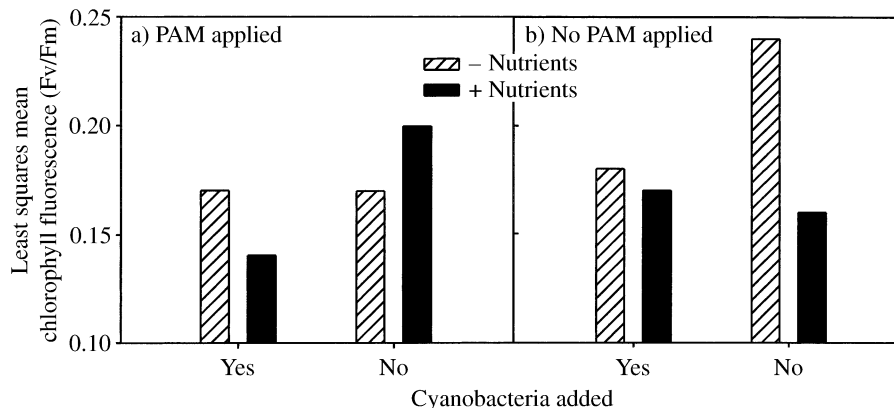
FIG. 6. Almost 15 mo after the start of the NDLS experiment, cyanobacteria had produced a negative effect on fluorescence on fertilized mounds sprayed with polyacrylamide, but a positive effect in the absence of both nutrient addition and soil stabilization by PAM ($P = 0.0315$ for interaction term).

df = 3, P = 0.86909) contained the effects of just two treatment variables, Cyanobacteria and Microaspect. Nitrogenase activity was greater on ENE than on WSW exposures (Fig. 9a), and greater in the absence of Cyanobacteria than in their presence (Fig. 9b).

### Lichen establishment from fungal spores and algal cells

The log-linear analysis relating application of fertile vs. sterile lichen fragments and/or *Nostoc* cells to num-



FIG. 7. After 2 yr at Island in the Sky, chlorophyll fluorescence ($\log_{10}$ Fv/Fm): (a) differed significantly across microaspects ($P < 0.0001$), and (b) the interaction of Polyacrylamide × Microaspect was marginally significant ($P = 0.06$). Error bars represent +1 SE.

bers of cores with lichen thalli yielded a significant lack-of-fit test for the model as a whole ($\chi^2 = 8.228$, df = 1, $P = 0.016$). Therefore, we used independent $2 \times 2$ chi-square tests to probe separately the significance of adding *Nostoc* and fertile vs. sterile *Collema* thalli. Only the latter effect was significant (Fig. 10a, b). When *Nostoc* status in the final cores (an indicator of existing plus applied *Nostoc*) was substituted for *Nostoc* addition in the log-linear analysis, the most parsimonious model was that in which the only higher order interaction term was thallus status (fertile vs. sterile) × *Collema* presence (+ or −). The lack-of-fit test was not significant ($\chi^2 = 2.77$, df = 3, $P = 0.4279$), indicating a fit to the model in which application of fertile thalli was associated with greater lichen recruitment ($\chi^2 = 8.08$, df = 1, $P = 0.0045$). The ubiquity of *Nostoc*, present in all but two cores, undoubtedly contributed to the lack of significance of this treatment. Overall, our results support the hypothesis that a shortage of fungal spores, but not *Nostoc* cells, inhibited recruitment of *Collema* lichen on disturbed soil crusts.

### Deposition and erosion vs. mound microaspect

For the spring data, ANOVA revealed significant effects of Site ($P < 0.0001$), Microaspect ($P < 0.0001$), and the Site × Microaspect interaction ($P < 0.0001$) on substrate deposition, and this model accounted for 91.2% of the variation in the data ($F_{7, 152} = 236.07$; $P < 0.0001$, Fig. 11a). Based on Tukey tests, run independently on data from the two sites, deposition at NDLS was greatest on WSW mound faces, followed by SSE > NNW > ENE. The pattern was similar at ISKY, except that deposition did not differ significantly between the WSW and SSE exposures. Deposition was also much greater at ISKY than at NDLS.

Net erosion in spring differed significantly in relation to Microaspect, and the Site × Microaspect interaction was also significant, though this model accounted for just 15.3% of the variation in the data (Fig. 11b, Whole-Model Test of Ordinal Logistic Regression: $\chi^2 = 94.26$,
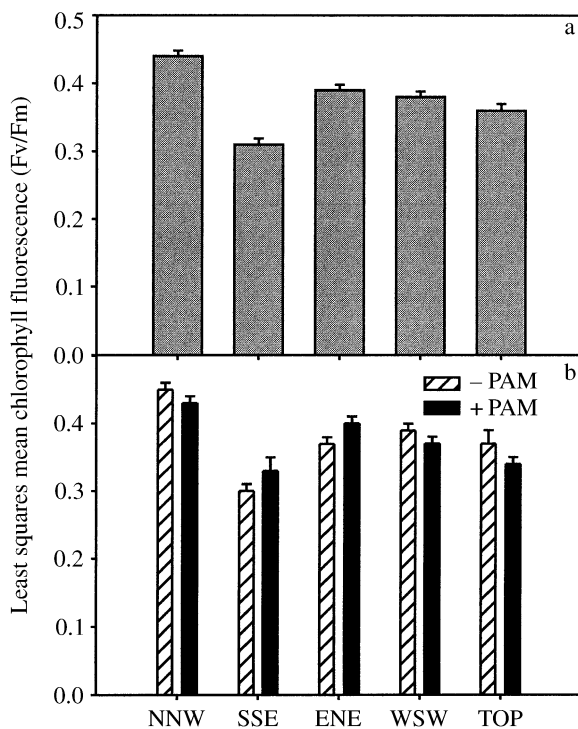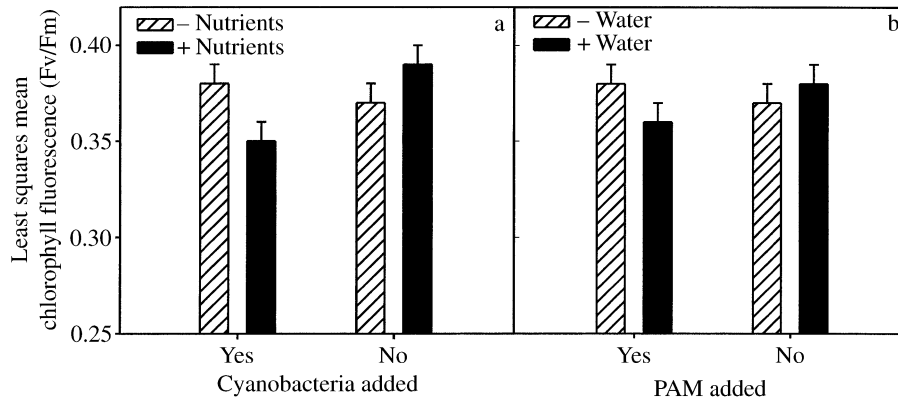
FIG. 8. Twenty-seven months after the start of the experiment at Island in the Sky: (a) chlorophyll fluorescence ($\log_{10}$ Fv/Fm) was enhanced by nutrients in the absence of cyanobacteria, but depressed in their presence ($P = 0.0071$ for interaction term), and (b) watering boosted yield in the absence of polyacrylamide but reduced it where this resin emulsion had been applied ($P = 0.0425$ for interaction effect). Error bars represent +1 SE.

df = 7, $P < 0.001$). In contrast to deposition, net erosion did not differ significantly between sites ($P \gg 0.05$). Net erosion was greatest on WSW exposures at both sites, intermediate on SSE exposures, and lowest on either NNW faces (ISKY) or ENE exposures (NDLS).

In fall, with all microaspects pooled, deposition was again far greater at ISKY than at NDLS (Wilcoxon rank sums: 8464.5 and 3163.5, respectively; $\chi^2 = 95.41$, df = 1, $P < 0.0001$). At NDLS, deposition differed significantly by microaspect, with WSW (rank sum = 1055.5) $\gg$ SSE (743.5) > NNW (590.5) $\geq$ ENE (536.5) ($\chi^2 = 17.45$, df = 3, $P = 0.0006$ in Wilcoxon test). At ISKY, deposition declined in the sequence: ENE (rank sum = 888.5) > WSW (642.5) > SSE (816) > NNW (582), but differences were not significant ($\chi^2 = 4.40$, df = 3, $P = 0.2216$). Net erosion did not differ

either across microaspects (with or without sites pooled, $P > 0.50$) or between sites ($P = 0.49$).

## DISCUSSION

### Lichen performance in relation to microaspect

Of all the treatments included in our study, microtopography exerted the greatest effect on *Collema* performance. First, as evaluated by the frequencies of plugs in good condition, lichens on ENE and NNW microaspects of soil mounds exhibited better overall conditions than did those on WSW and SSE exposures, 1 yr (NDLS and ISKY) and 2 yr (ISKY) after transplantation in field experiments (Fig. 2a, b, and c, respectively). By this same criterion, plugs on the Tops of mounds also did relatively well, outperforming those from all other mound exposures at NDLS, and all but
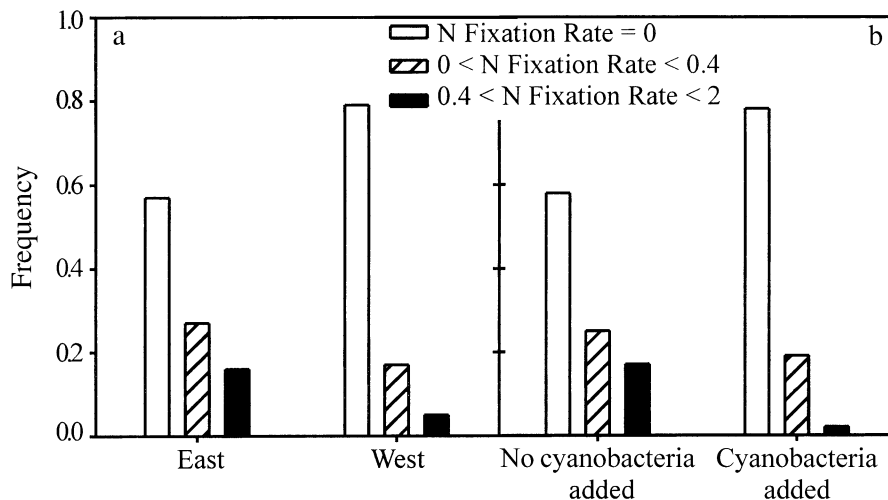


FIG. 9. Nitrogenase activity of *Collema* lichen plugs after 14.5 mo at NDLS was (a) greater on the ENE than on the WSW sides of soil mounds ($\chi^2 = 18.62$, df = 2, $P = 0.0001$), and (b) greater in the absence of cyanobacteria than in their presence ($\chi^2 = 13.95$, df = 2, $P = 0.0001$).
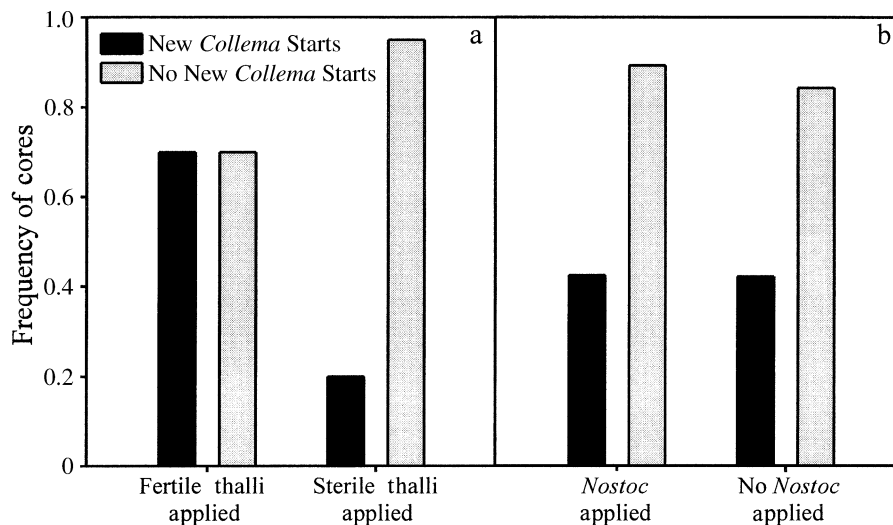
FIG. 10. Numbers of cores with *Collema* thalli present at end of apothecia experiment, as a function of: (a) the application of fertile (with apothecia) vs. infertile (no apothecia) lichens during the course of the experiment and (b) the application (or not) of *Nostoc* cells. In separate two-way chi-square tests, the application of fertile lichens produced greater lichen recruitment in sample cores ($\chi^2 = 7.729$, df $= 1$, $P = 0.005$), but application of *Nostoc* did not ($\chi^2 = 0.009$, df $= 1$, $P = 0.925$).

ENE plugs by the end of the second year at ISKY. Second, lichen plugs on ENE exposures performed better than did those on WSW exposures after 14.5 mo, as measured by chlorophyll fluorescence at NDLS. Such a difference was not significant at ISKY after 27 mo, though plugs from NNW aspects again performed well, significantly exceeding all other plug categories in dark quantum yield (Fig. 7a). Third, in the absence of cyanobacterial addition at NDLS, nitrogen fixation by *Collema* lichens on ENE exposures significantly exceeded that on WSW exposures (Fig. 9), as the only significant effect of microaspect in the model. These observations accord well with data from recent surveys of crustal species growing naturally in areas near our study sites. George et al. (2000) found greater cyanobacterial biomass, *Collema* lichen cover, and colonization frequencies of other lichen species on ENE than on WSW mound exposures. In a similar study (M. Bowker, *unpublished data*), *Collema* cover on ENE and TOP microaspects exceeded that on WSW and SSE exposures, and was intermediate on NNW faces. Similar patterns are also observed on a landscape scale. Macrotopographic surveys (M. Bowker, *unpublished data*) indicate that lichen and moss cover in soil crusts is highest on northfacing slopes on the Colorado Plateau, and differential distributions of this kind are apparently a common feature of soil crusts in other desert areas as well (Nash et al. 1977, Kidron 1999).

Likely contributors to the patterns in lichen performance and abundance include differences in microclimate, nutrient regimes, and soil surface stability across microtopographic and topographic features of the landscape. First, reduced insolation leads to lower temperatures and reduced moisture losses on northern and eastern slopes, compared to southern and western slopes, on both the landscape and much smaller scales (Larcher 1980), and is apt to do so as well on the microscale of the pedicels comprising microbiotic crusts. Preliminary data from TDR microprobes (Time Domain Reflectometry) support this contention. Thus, over the annual cycle, ENE and NNW mound surfaces retain moisture longer than do SSE and WSW microaspects during hours when PAR (photosynthetically active radiation) is sufficient for photosynthesis (J. Belnap, *unpublished data*). In cooler parts of the year, the sun's rays strike most directly on southerly mound exposures. Although western and eastern mound exposures should receive the same amount of insolation, temperatures are likely cooler in the mornings than in late afternoons and evenings, leading to less evaporation on eastern than on western exposures. During summer, high air temperatures generally promote rapid drying of the crust microorganisms after rainfall events. Because most summer storms occur during the afternoon (Tigges 1999), organisms on southern and especially western aspects experience greater solar radiation immediately after rain than do their counterparts on eastern and northern aspects, and therefore dry more rapidly. In temperature and moisture retention, mound TOPS resemble ENE and NNW exposures in winter, and SSE and WSW microaspects in summer (J. Belnap, *unpublished data*).

As a consequence of these differential microclimates, summer rainfall may frequently result in carbon deficits, especially on southern and western aspects, where the organisms often cannot stay wet long enough to achieve net carbon gain (Jeffries et al. 1993). For most cyanobacteria and lichens of desert soils, carbon deficits may be further exacerbated during warmer sea-
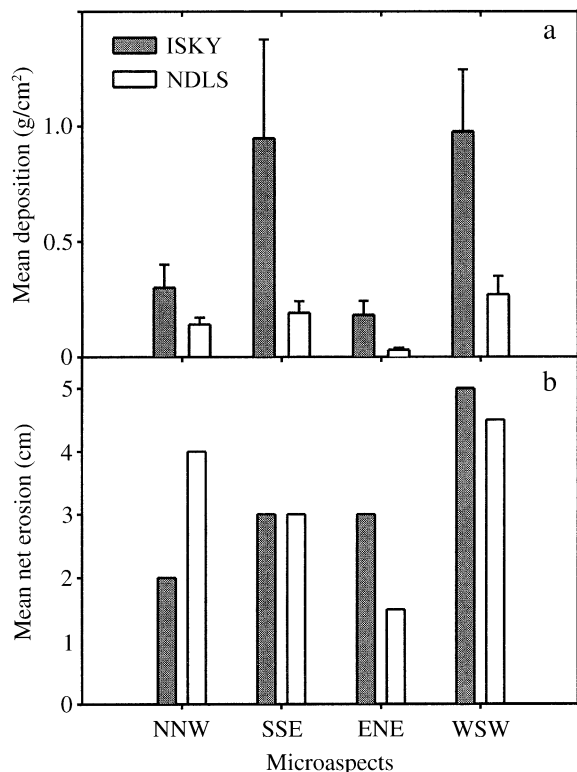
FIG. 11. (a) Mean substrate deposition differed between ISKY and NDLS ($F = 689.70$, df $= 1, 3$; $P \ll 0.0001$) and across four microaspects of artificial soil mounds ($F = 300.55$, df $= 3$; $P < 0.0001$), over 11 weeks in spring of 2000. The Site $\times$ Microaspect interaction term was also significant ($P < 0.0001$). Within sites, deposition data in each column differed significantly from those in each other column, with the exception of SSE and WSW at ISKY (Tukey tests, $\alpha = 0.05$). (b) Mean net erosion (erosion $-$ deposition) also differed across microaspects at ISKY and NDLS in spring of 2000 (Wald $\chi^2 = 66.54$, df $= 3$, $P < 0.0000$), and the Site $\times$ Microaspect interaction was also significant (Wald $\chi^2 = 21.21$, df $= 3$, $P < 0.0001$).

sons by thermal limits to carbon, and also nitrogen, fixation (at $\sim 25°–30°C$, and $30°C$, respectively [Belnap and Lange 2001]). Thus, even with sufficient moisture available, thallus temperatures commonly reached during summer days may preclude both carbon and nitrogen fixation. Across all seasons, the balance among moisture, PAR, and temperature on different aspects of the mounds comprising soil crusts should have a profound influence on lichen growth and performance, and this seasonal variation is currently under investigation (J. Belnap, *unpublished data*).

Second, nutrient levels can also differ significantly on the different microaspects of mounds. Preliminary studies have found that levels of available K and Zn, as well as K/Mg ratios, are higher on ENE mound faces than on WSW (J. Belnap, *unpublished data*), regardless of whether *Collema* spp. are present. The mechanism for this is not known, although variation in nutrient levels with slope aspect has also been seen on larger

spatial scales, with north-facing slopes exhibiting higher fertility than do south-facing slopes in the Negev Desert (Danin and Garty 1983). The greater mass of cyanobacterial sheath material on ENE and NNW microaspects, compared to WSW and SSE exposures (George et al. 2000), may result in greater dust capture (Verrecchia et al. 1995) and soil nutrient availability (Reynolds et al. 2001).

Third, soil stability is also very important to slow-growing lichens, as these microorganisms cannot respond quickly to burial by sediment, and the effects of both wind and water erosion and soil stabilization by microorganisms may vary by microaspect. Measured simultaneously on artificially constructed soil mounds at NDLS and ISKY, both deposition and erosion declined across the four mound microaspects at NDLS as: WSW $>$ SSE $>$ NNW $>$ ENE (or ENE $>$ NNW), during both fall and spring trials. Patterns were similar at ISKY, except that WSW $\approx$ SSE for spring deposition, and that deposition did not differ by microaspect in the fall of 2000. Overall, the observed patterns of deposition and erosion (Fig. 11) could have contributed to the relatively poor performances of *Collema* lichen plugs on WSW and SSE faces of soil pedicels. However, they cannot be the entire explanation, since across the two sites, lichens performed better at ISKY, where deposition was higher than at NDLS. Higher mean wind velocities at ISKY (National Park Service 1993, 1998, 1999 vs. CLIM-MET 1999 [Southwest Climate Impact Meteorological Stations][6] for NDLS) may explain the greater deposition rates there.

The Colorado Plateau lies in the $30°–60°N$ latitudinal belt characterized by prevailing southwesterly winds (e.g., Pianka 1988), and such winds might be expected to contribute to the consistency of both mound orientations and organismal distributions on mounds throughout the Colorado Plateau (e.g., George et al. 2000; M. Bowker, *unpublished data*). In general, our deposition data are consistent with greater sediment deposition on the upwind sides of mounds. Although both the predominant and the highest velocity winds at NDLS tend to blow from the south and southwest throughout the year, the wind pattern at ISKY is more complicated due to its location on an exposed ridge (National Park Service 1993, 1998, and 1999, CLIM-MET 1999[6]). There, the highest velocity winds blow from the northwest or west, especially in fall and winter but not in summer, and from the south or WSW in spring but not fall. Although more frequent, winds from the east to southeast, especially in summer and fall, are low velocity and not apt to carry much sediment. Despite differences in wind patterns at ISKY and NDLS, the microtopography of crusts and the distributions of component organisms are similar at the two sites, as well as in other topographically diverse sites that may have still different wind patterns (D. W. Davidson, *per-*

[6] URL: ⟨http://climweb.cr.usgs.gov/info/sw/clim-met/comm⟩

*sonal observation*). It therefore appears unlikely that the latter patterns are explained entirely by the effect of winds on deposition or erosion.

We suggest that the influence of microtopography on lichen performance is due to a combination of deposition and erosion from relatively high-velocity winds, together with disparities in microclimate and nutrient regimes across mound exposures. More favorable microclimates and nutrient regimes on NNW and ENE mound faces (or, e.g., at the more fertile ISKY site) favor strong performance. Better performance should lead, in turn, to increased cyanobacterial and lichen biomass (George et al. 2000; M. Bowker, *unpublished data*) (see Table 1 for site differences), which should be associated with lower net erosion. By this line of reasoning, the often scoured appearances of WSW mound faces are likely due to the effects of erosion in microhabitats where poorly performing components of crusts offer little resistance to erosion. In contrast, since deposited soils are, by definition, from off site rather than the mound itself, the greater deposition on WSW and SSE exposures is probably due entirely to relatively high-velocity winds.

### Lichen performance in relation to four experimental treatments

Experimental treatments produced some unanticipated negative effects, especially at NDLS. The additions of *Microcoleus* and of nutrients were the only two treatments producing significant individual effects on lichen performance, and each of these supplements harmed lichens at NDLS but not at ISKY, where slight but nonsignificant positive effects were actually apparent after 1 yr (Figs. 3 and 4). Moreover, nutrient supplements were associated with diminished yield in chlorophyll fluorescence at NDLS, though they enhanced yield at ISKY, except under cyanobacterial addition. *Microcoleus* addition also reduced the positive effect of Water on chlorophyll fluorescence at NDLS (Fig. 5). Finally, nitrogenase activity also declined with *Microcoleus* addition at NDLS, though just on ENE mound exposures (Fig. 8b).

Given the extremely fragile soils at NDLS, it is likely that some of the negative treatment effects there were due to disturbance. For example, applications of water, nutrients, or both eventually led to some transport of soils and lichen plugs down the sides of mounds and probably accounts in part for why plugs on the tops of mounds performed well in relation to plugs at other mound positions there (Fig. 2a). However, such disturbance cannot explain all of the results: for example, why chlorophyll fluorescence responded negatively to nutrients but positively to water (absent cyanobacterial addition). We can only suggest that the nutrient component of these additions might somehow have accentuated competition from existing or supplemental cyanobacteria. *Microcoleus* has been shown to compete with vascular plants for phosphorus (Belnap and Harper

1995; J. Belnap, *unpublished data*), and the various negative impacts of *Microcoleus* on *Collema* lichens (at both sites, see above) might be taken as evidence that *Microcoleus* competes with *Collema*, at least at the levels of abundance included in our experiment. Such putative competition would have been for nutrients or water, rather than for light, since *Microcoleus* fiber was added adjacent to lichen plugs and did not cover them. Phosphorus (P) is required for both growth and nitrogen fixation, and our laboratory studies indicate that *Collema* performance is probably P limited in these soils. However, the negative effects of nutrient addition occurred at the NDLS site, where cyanobacteria were much less abundant than at ISKY, perhaps due to the adsorption of available phosphorus by high levels of $CaCO_3$ in NDLS soils (Table 1). Such adsorption of phosphorus as insoluble compounds is typical at the high soil pH levels at our sites (Lajtha and Bloomer 1988).

### The role of propagule availability in the recovery of microbiotic crusts

Our results indicate that the availability of fungal spores, rather than of the photobiont *Nostoc*, limits the establishment of *Collema* from these independent stages. This is somewhat expected, given the rarity of apothecia-bearing *Collema* on the Colorado Plateau (Anderson and Rushforth 1976; L. St. Clair, *personal communication*). It would appear that inoculation of disturbed crusts with *Collema*-isolated *Nostoc* is unlikely to enhance *Collema* establishment via spore germination. Inoculation with spores should probably be investigated further. Because spore production is limiting and naturally rare (due to rarity of apothecia), it is likely that *Collema* relies primarily on vegetative propagules such as isidia or thallus fragments for dispersal. Our experiments did not focus on this form of establishment.

### Conclusion

Aside from site differences likely associated with soil properties, the most consistent and easily interpreted effects on lichen performance were those of microtopography, i.e., of slope exposure on the soil pedicels. The strong effects of microaspect on the performances of *Collema* spp. here, and of other lichens and mosses on the Colorado Plateau (George et al. 2000; M. Bowker, *unpublished data*), suggest that the gradual establishment of soil microtopography favorable to lichen recruitment is a significant factor limiting the rate of recovery of crusts after disturbance. Therefore, one of the most important means of assisting the natural regeneration of crusts is protection of the developing microtopography. It is clear also that measurements of recovery rates of disturbed crusts should include some characterization of substrate structure and microtopography, as well as of biotic components and features of the crusts. Thus, George et al. (2000) have suggested

that subtle differences in mound orientation and organismal distribution may be useful in comparing the disturbance histories of crusts retrospectively.

Our studies suggest other potentially fruitful avenues to be investigated. Application of fertile *Collema* thalli may help in hastening the recovery of soil crusts, especially if treatments are focused on appropriate mound aspects. Second, the effects of enhancing K, K/Mg, and P imply that *Microcoleus* and *Collema* may compete for nutrients, and that, despite the importance of *Microcoleus* for soil stabilization, better understanding of this relationship could lead to other ways of enhancing *Collema* establishment and performance.

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