Himalayan
JOURNAL OF SCIENCES

research paper

Microbial biomass and activity in high elevation (>5100 meters) soils from the Annapurna and Sagarmatha regions of the Nepalese Himalayas

Andrew J King, Debendra Karki, Laszlo Nagy, Adina Racoviteanu and Steven K Schmidt

Himalayan Journal of Sciences 6(8): 11–18, 2010
doi: 10.3126/hjs.v6i8.2303

Microbial biomass and activity in high elevation (>5100 meters) soils from the Annapurna and Sagarmatha regions of the Nepalese Himalayas

Andrew J King[1], Debendra Karki[2], Laszlo Nagy[3], Adina Racoviteanu[4] and Steven K Schmidt[1]*

[1] Department of Ecology and Evolutionary Biology, University of Colorado, Boulder, Colorado, 80309, USA
[2] College of Applied Science Nepal, Anamnagar, Kathmandu, NEPAL
[3] EcoScience Scotland, 2/1 27 Glencairn Drive, Glasgow, SCOTLAND
[4] Department of Geography, University of Colorado, Boulder, Colorado 80309, USA
* For correspondence, email: steve.schmidt@colorado.edu, phone: (01) 303-492-6248

High elevation subnival-zone soils are increasing in spatial extent in the Himalayas due to glacial retreat and grazing pressures. These seemingly barren soils actually harbor significant microbial diversity but have remained mostly unstudied in all of the major mountain ranges of the Earth. Here we describe a preliminary survey of subnival-zone soils and one vegetated high-elevation soil in the Annapurna and Sagarmatha regions of the Nepalese Himalayas. We examined microbial biomass and activity as well as key microclimatic and edaphic variables that may control microbial activity in these soils. Microbial biomass carbon levels were the lowest ever reported for any soil to date, whereas microbial nitrogen and soil enzyme activities were similar to levels measured in previous studies of subnival-zone soils of Peru and Colorado. Our initial studies also indicate that soil water availability is the primary limiting factor for life in these high-elevation soils.

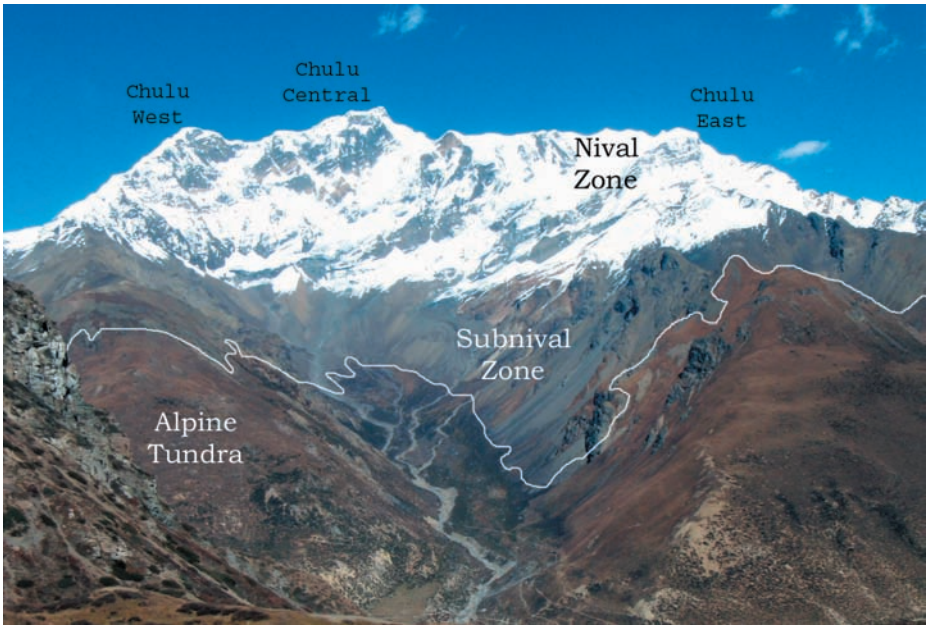
Key words: Microbial, soil, biogeochemistry, subnival, extracellular enzymes, microbial biomass

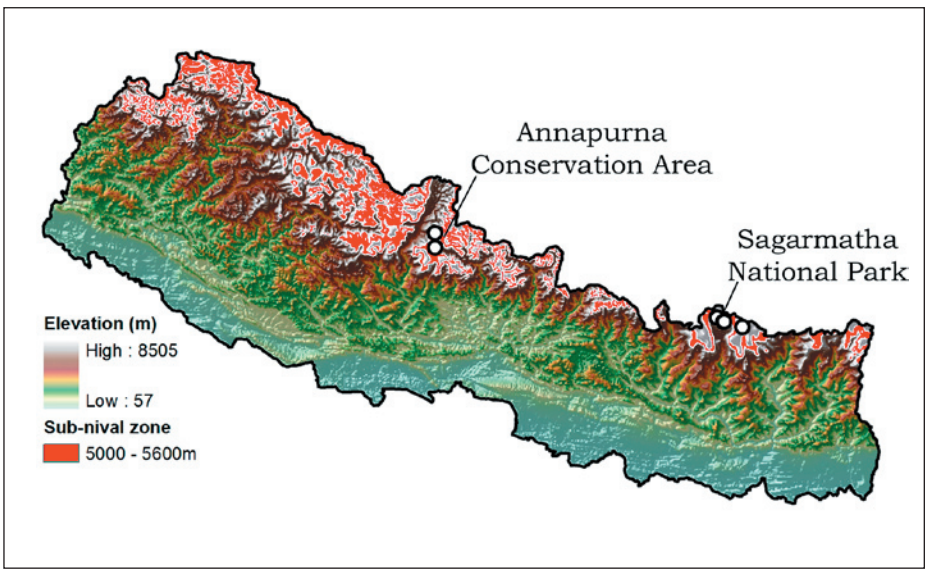
On the highest mountain ranges of the Earth, between the upper zone of year-round snow and ice (the nival zone) and the zone of continuous vegetation (the alpine zone), exists a stark expanse of seemingly bare rock and barren soils called the "subnival zone" or "mountain desert" (**Figure 1a**; Nagy and Grabherr 2009, Troll 1973). Yet, upon close inspection the subnival zone is revealed to be a landscape mosaic in which soil development and plant colonization are related to local variation in snow pack accumulation. This variation results in a patchy environment with barren soils underlying high snow areas, sparse plant communities in moderate accumulation areas, and further barren soils in wind-scoured locations. When not snow-covered, the soils of this highly exposed environment are subject to extreme fluctuations in temperature, solar radiation, and soil moisture. This effect is particularly pronounced during the fall season when daily soil temperatures can fluctuate over 40°C, with nighttime conditions below freezing and surprisingly hot daytime soil temperatures around 30°C (**Figure 2**). These factors create a harsh environment for subnival zone organisms and result in one of the most barren looking ecosystems on Earth. It is presently unclear if, given enough time, plants can colonize the upper-elevations of the subnival zone. However, extant subnival soils are dominated by abundant and surprisingly diverse microorganisms even in the highest elevation soils sampled to date (Costello et al. 2009, Schmidt et al. 2009).

Due to the dependence on snow accumulation, the area defined as the subnival zone occurs at much higher elevations in drier mountain ranges such as in the Andes and in the inner ranges of the Himalayas than in more humid mountain ranges such as the Alps. The subnival zone of the Bavarian Alps starts at about 2500 m above sea level (masl) compared to 4700–5000 m in the Andes of southern Perú and 5000–5600 m in the Himalayas (Chang 1981, Rawat and Pangtey 1987). Owing to their extremely high elevation and historical inaccessibility, much less is known about the subnival zone in ranges such as the Himalayas than in the comparatively well-studied Alps. Globally, the subnival zone is thought to have expanded downwards in recent years due to overgrazing in the upper alpine zone (Ahmad et al. 1990, Del Valle et al. 1998) and upwards, due to the retreat of glaciers and icepacks at high elevations (Byers 2007), and is predicted to increase over the next century (Zemp et al. 2006). In Nepal, the subnival zone currently occupies about 6% of the land area (**Figure 1b**); yet we know almost nothing about the organisms that inhabit these environments.

It is still unclear how life forms that survive in the subnival zone obtain the nutrients and energy needed to sustain life. Swan (1963, 1990, 1992) contended that life at these extreme elevations subsisted mainly upon aeolian-deposited

Himalayan Journal of Sciences 6(8): 11–18, 2010
doi: 10.3126/hjs.v6i8.2303; published online: Sept 27, 2010





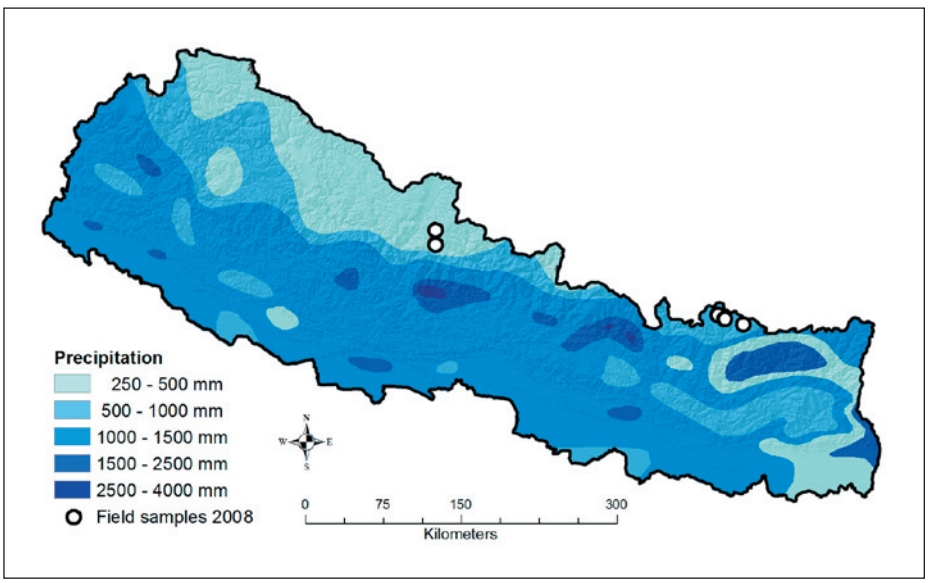


Figure 1. **a (top):** Photograph of the Chulu range taken (10/19/2008) from above Kangshar village during our fieldwork in the ACA. The labels indicate the distinct zonation of the alpine, subnival, and nival zones. The boundary between alpine and subnival zones is defined as the upper extent of continuous plant cover. The boundary between subnival and nival zones is defined as the lower extent of continuous ice and snow cover. **b (middle):** An elevational map of Nepal. The red area represents the extent of the subnival zone as estimated by locations having an elevation of 5000–5600 masl. Sampling locations are in the black circles. **c (bottom):** A map of average yearly rainfall for Nepal showing the sampling sites (data from Nepal Department of Hydrology and Meteorology, http://www.dhm.gov.np).

Table 1. Sampling sites used in this study

Site	UTM coordinates (zone)	Elevation (masl)	Rock type	pH
Zun Tal (ACA)	784700E, 3180600N (44)	5101 – 5289	shale	7.2
Kobresia sward/eroded (ACA)	786770E, 3176125N (44)	4824	soil	6.1
Thorong La (ACA)	787000E, 3188900N (44)	5482 – 5516	shale	7.3
Gokyo Lake 5 (SNP)	468350E, 3097500N (45)	5094 – 5111	granite	5.3
Chola Pass (SNP)	493650E, 3092850N (45)	5376 – 5387	granite	5.3
Island Peak (SNP)	493170E, 3089600N (45)	5272 – 5291	granite	5.3

ACA, Annapurna Conservation Area; SNP, Sagarmatha National Park

organic matter; that is, organic matter blown from lower elevations to higher elevations. However, recent studies of microbial communities at high elevations (Freeman et al. 2009, King et al. 2008, Schmidt et al. 2009) have caused us to reevaluate how life is sustained at these elevations. We have been studying microbial life in soils up to 6000 masl in the high Andes of South America and the southern Rocky Mountains of the United States for the past ten years. Although we have observed pockets of aeolian-supported life (Ley et al. 2004), we have found much larger areas of wind-swept lands that do not accumulate high amounts of organic debris from lower elevations but are nonetheless teeming with previously unreported microbial life (King et al. 2008, Schmidt et al. 2009). These studies have shown that in many subnival soils, microorganisms obtain their sustenance not from wind-blown organic matter but primarily from atmospheric gases through the processes of microbial photosynthesis and nitrogen fixation (Freeman et al. 2009, Schmidt et al. 2008, 2009) and that the buildup of microorganisms may be largely limited by soil water availability (King et al. 2008). Thus, although we have pushed our understanding of high-elevation life beyond the pioneering efforts of Swan (1963, 1990, 1992), the question remains as to whether these new discoveries made in the Andes and Rocky Mountains apply to even higher mountain ranges of the world.

The large area occupied by subnival soils in the Nepalese Himalayas (**Figure 1b**) makes it particularly important to understand the activity and abundance of high elevation microorganisms there. Here we examine microbial biomass and extracellular enzyme activity in subnival soils from the Annapurna and Sagarmatha (Everest) regions of the Nepalese Himalayas. Our study describes microbial life in four subnival soils: plant-covered, eroded previously vegetated, fine shale-derived gravel, and fine granite-derived gravel. This study is the first report of an ongoing research effort to characterize the microbial activity and diversity of the subnival soils of the Himalayas.

Methods

Study sites and sample collection Our sampling sites were in the Annapurna Conservation Area (ACA, Panthi et al. 2007, Shrestha et al. 2007) and the Sagarmatha National Park (SNP, Byers 2007) of the Himalayan Mountains in Nepal (**Figure 1, Table 1**). In each region, we sampled mineral soils from sites just above the highest plants and from sites as high as were attainable due to prevailing conditions (e.g. presence of ice and snow) at the time of sampling. Sampling was conducted in October and November of 2008 in order to take advantage of the seasonal lack of precipitation. Low precipitation seasons are ideal for this type of descriptive study because it minimizes short term variability due to individual precipitation events when comparing across sampling sites and allows access to the highest possible soils due to the relatively snow-free conditions. In addition to unvegetated soils of the ACA, we also sampled patches of soil dominated by the sedge *Kobresia* cf. *pygmaea* (C. B. Clarke) C. B. Clarke as well as soil from eroded areas adjacent to patches of *Kobresia*. These sites were located just below the lowest-elevation plant-free site. Soil samples were collected to a depth of 5 cm and placed in sterile zip-lock bags. Samples were frozen in the field by packing sample bags in a cooler along with snow and ice collected from the landscape and transported back to the laboratory over about a week. Samples were immediately extracted for K_2SO_4 dissolvable C and N (Weintraub et al., 2007) and then stored at –20°C for further analysis.

The low-elevation sites in the ACA are unvegetated, south-east facing slopes (28°43'N, 83°55'E) at 5122 masl and approximately 8 km east of the northern edge of Tilicho Lake. We also collected three samples from patches of *Kobresia* covered soil as well as three samples from eroded areas adjacent patches of *Kobresia* on a south-south-west facing slope (28°42'15"N; 83°56'05"E) at 4824 masl. The high-elevation sites in the ACA are unvegetated south-east facing slopes from 5503 to 5516 masl, 1 km north of Thorong Pass (28°48'N, 83°56'E). The Thorong Pass crosses the divide separating the Marsyangdi River to the east and the Kali Gandaki River to the west. All of the ACA sites are located on shale bedrock.

The sampling sites in the SNP receive significantly more precipitation than do the ACA sites (**Figure 1c**). We collected 4 samples from each of three unvegetated south-east facing slopes in this region, Chola Pass, Gokyo Lake 5, and Island Peak. All of the SNP sites were located on granite bedrock.

Soil temperature Soil temperature measurements were recorded using HOBO data loggers (Pendant temp/light, UA-002-64, Onset Computer Corp., Bourne, MA) from October 16th to 19th 2008 at the ACA *Kobresisa* dominated site. One data logger was placed flush with the soil surface and another at a depth of 4 cm.

Microbial biomass carbon and nitrogen Soil dissolved organic carbon (DOC), total dissolved nitrogen (TDN), and microbial biomass carbon (MBC) and nitrogen (MBN) were determined using the methods described in Weintraub et al. (2007). For soil DOC and TDN, 5 g of each soil sample was shaken with 25 ml of 0.5 M K_2SO_4 for 1 hour. For microbial biomass C and N, 5 g of soil was added to a 250 ml glass flask with 2 ml of chloroform, sealed and fumigated for 24 hours, and then vented for 1 hour; 25 ml of 0.5 M K_2SO_4 was added to each flask, and then they were shaken for 1 hour. Solutions were pre-filtered using a 1 µm Pall glass fiber filter (Pall Corporation, East Hills, NY). Solution C/N analysis was performed using a Shimadzu total organic carbon analyzer (TOC 5000) equipped with a total dissolved nitrogen (TDN) module (Shimadzu Scientific Instruments, Inc., Columbia, MD).

pH determination In order to determine soil pH, 2 g of each soil was placed to an individual 15 ml conical tube to which was added 2 mL of distilled water. Conical tubes were placed horizontally on a shaker table and shaken for 1 hr at 175 rpm. Soil pH was measured using a glass Fisher pH probe (Fisher Scientific, Pittsburgh, PA).

Extracellular enzyme activity Microbial extracellular enzyme activities were assayed using a modification by King et al. (2008) of the method of Weintraub et al. (2007). Enzymes assayed were: N-aceytalglucosaminase, cellulase (β-glucosidase), α-glucosidase, β-xylase, cellobiosidase, leucine aminopeptidase and organic phosphatase. For each sample, 2 g of soil was added to 150 ml of buffer at the average pH for the soil type (0.5M Acetate at pH 5 for granitic, 0.5M Acetate at pH 6 for eroded and vegetated, and 0.5M Bicarbonate at pH 7.3 for shale derived soils) and homogenized at 3000 rpm for 1 minute using a Ultra-Turrax homogenizer (IKA Works Inc., USA). Soil slurries were assayed using the same controls, fluorescent substrates, and solution volumes as in King et al. (2008). Soils were incubated with substrates for 20 hours at 14°C.

Water holding capacity Tubes for assaying water holding capacity (WHC) were constructed by cutting the bottom off of a 1 cm diameter 15 ml conical tube and covering the opening with 1-mm gauge plastic mesh. The mesh was wetted with deionized water prior to the addition of soil to the tube so that particles less than 1 mm in size would clump together at the bottom of the tube. For each sample we added ~4 g of soil to a tube and then added ~2 ml of H_2O. Wetted sample tubes were placed in 50 ml conical tubes, which were drained periodically. When an individual sample stopped dripping, the mass of the sample was recorded. Samples were then dried at 100°C for 24 hours. Water holding capacity is reported as the g H_2O at soil saturation divided by g dry soil.

Statistics and data analysis Tukey's honestly significant difference tests (Devore 2004) were performed in R (version 2.8.1, 12/22/2008, R Foundation for Statistical Computing http://www.r-project.org/index.html). A correlation test (Devore 2004) between microbial biomass C (MBC) and water holding capacity was also performed in R.

Results

Microbial biomass and extracellular enzyme activity were extremely low in the shale-derived soils from the ACA (**Figure 3, 4**). The granitic soils of the SNP had significantly higher biomass and activity than the shale-derived soils (**Figure 3, 4**). As expected, soils of the lower-elevation eroded area had higher microbial biomass than either of the two higher-

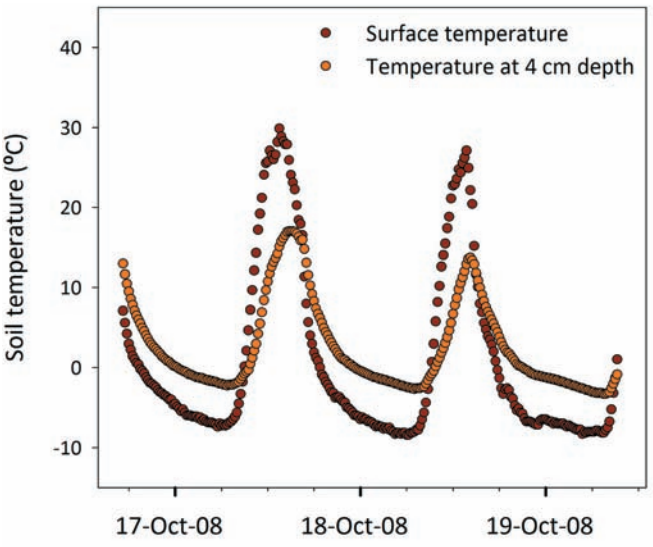


Figure 2. Soil temperature measurements from two days near the 4824 m elevation *Kobresia* dominated site in the ACA region.

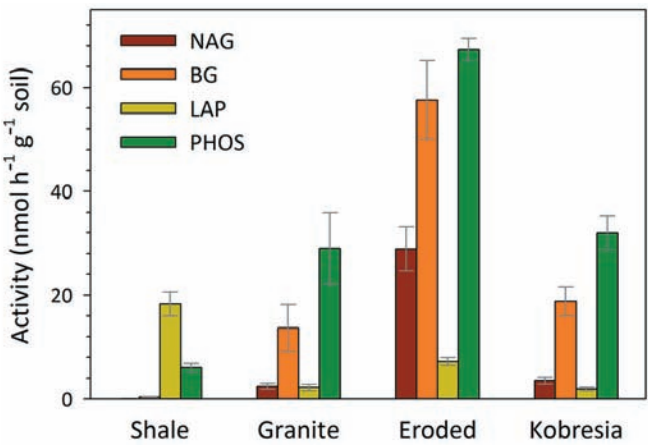


Figure 3. Extracellular enzyme activities for the four soil types. Enzymes activities shown are N-aceytalglucosaminase (NAG), cellulase (β-glucosidase) (BG), organic phosphatase (PHOS) and leucine amino peptidase (LAP). α-glucosidase and β-xylase activities were at similar levels to NAG and are not shown. Significant differences are designated by letters grouping similar levels of activity for an individual enzyme (Tukey's Test, $p < 0.05$). Error bars are standard error.

elevation mineral soils, and the *Kobresia* soils had the highest microbial biomass C and N (**Figure 4**). Surprisingly, while both the eroded soils and the *Kobresia* soils had higher enzyme activity than the mineral soils, the eroded soils had significantly higher extracellular enzyme activity than the vegetated soils (**Figure 4**). The biomass trends were mirrored by the water holding capacity measurements (**Figure 5**). Finally, there was a significant correlation between MBC and WHC for all samples (**Figure 6**, Pearson $r^2 = 0.426$, correlation test: $p < 0.001$). However, this trend was primarily driven by the increase of WHC and MBC with plant colonization.

Discussion

The subnival soils examined in this study are subject to some of the most extreme environmental conditions of any soils on Earth. Nevertheless, we found measurable amounts of microbial biomass and enzyme activity in even the most visually barren mineral soils (**Figure 3, 4**). Perhaps due to such extreme conditions, the biomass C numbers we observed were on average very low for both the mineral and the vegetated soils we sampled. Particularly low microbial biomass was found in the shale-derived mineral soils of the ACA. At an average of 20 µg C/g soil, these biomass levels

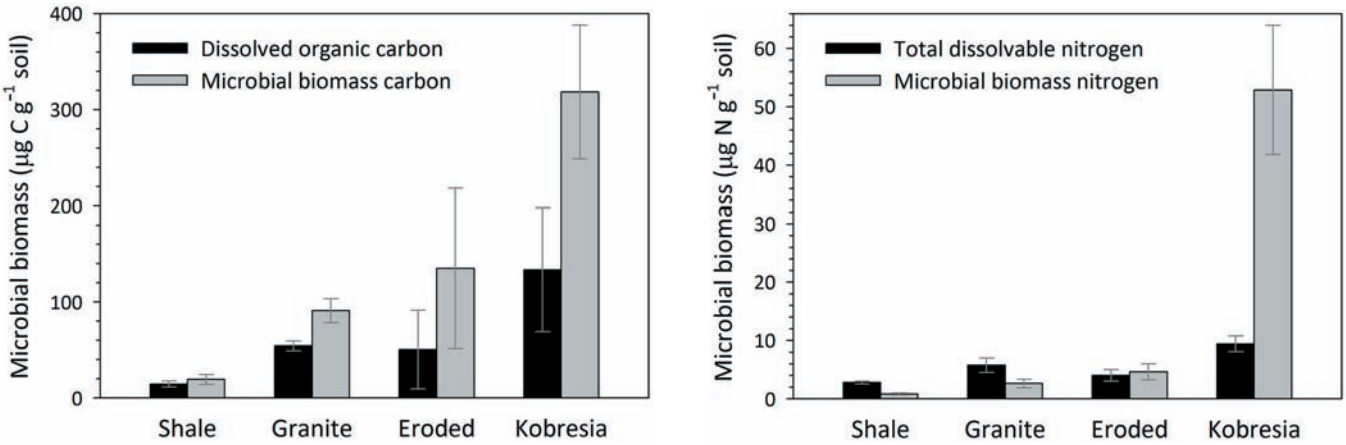


Figure 4. (a) Dissolved organic carbon (DOC) and microbial biomass carbon (MBC) for the four soil types. (b) Total dissolvable nitrogen (TDN) and microbial biomass nitrogen (MBN) for the four soil types. Significant differences are designated by letters grouping similar levels (Tukey's Test, $p < 0.05$). Error bars are standard error.

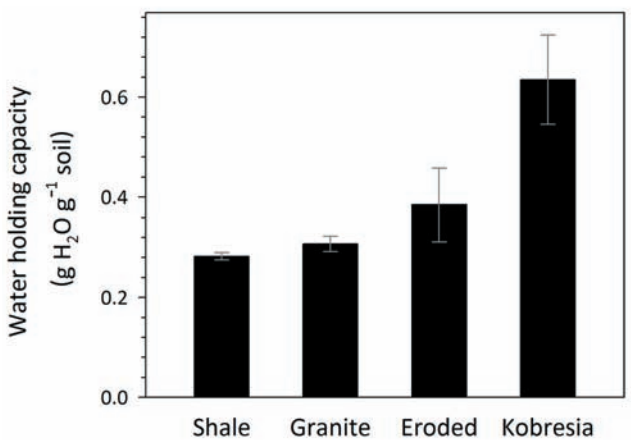


Figure 5. Water holding capacity (g H_2O/g dry wt of soil) for the four soil types. Significant differences are designated by letters grouping similar levels of activity for an individual enzyme (Tukey's Test, $p < 0.05$). Error bars are standard error.

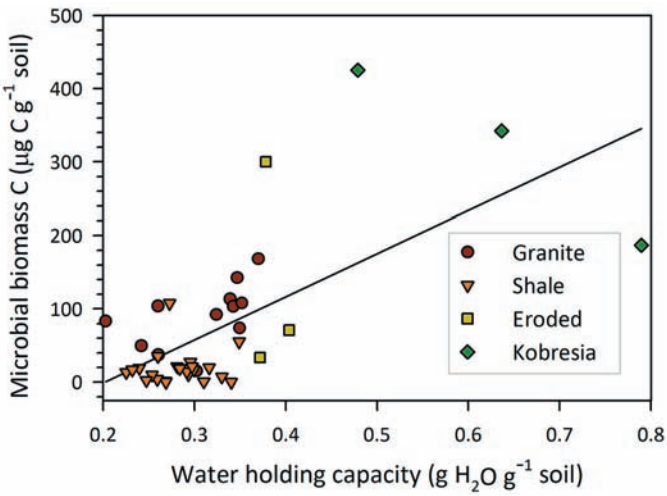


Figure 6. Microbial biomass C versus water holding capacity for all samples (Pearson $r^2 = 0.426$, correlation test: $p < 0.001$). It is apparent that this trend is driven by the increase in WHC and MBC with plant colonization. Taken individually the only sample group with a significant positive association between WHC and MBC was the granitic soils ($r^2=0.301$, $p<0.05$). Error bars are standard error.

are the lowest reported to date for subnival or recently deglaciated soils. Previously, the lowest microbial biomass numbers were reportedly found in alpine and Antarctic glacial moraines, which harbor 60 and 100 µg C/g soil, respectively (Tscherko et al. 2003a, b). Likewise, subnival-zone soils of Colorado and Peru contained 80 and 140 µg C/g soil, respectively (King et al. 2008). Thus, subnival soils may represent environments at the upper boundary of suitable conditions for sustaining microbial life. More work is needed to determine what component of the biomass is active at these sites and what component consists of dormant organisms blown in from lower elevations.

Our results support the hypothesis put forth by King et al. (2008) that the primary limiting factor determining microbial biomass levels in these extreme subnival soils is water availability (**Figure 6**). Indeed, the shale-derived plant-free soils from the ACA had the lowest water contents of any of the soils and the lowest measured levels of microbial biomass. These soils also had the lowest water holding capacities of any of the soils examined in this study (**Figure 5**). However, neither soil water content nor water holding capacity was significantly different among subnival soil sampling areas. Interestingly, the granitic soils of the SNP had similar microbial biomass levels to the granitic soils of the Rocky Mountains of the central United States that we have previously studied (King et al. 2008). In that same study we reported that shale-derived plant-free soils from the Andes in Perú had higher microbial biomass than the Rocky Mountain soils, however, the Himalayan soils display the reverse trend. This discrepancy suggests that rock type is not the main factor determining microbial biomass levels. Indeed, this trend is mirrored by the lower extracellular enzyme activities and DOC content of the shale-derived soils versus the granite-derived soils from the Nepalese Himalayas. These results point to possible differences in soil age and development between the barren subnival soils of the ACA and the SNP, differences that may originate from variation in slope stability due to degree of slope, bedrock hardness, or annual precipitation (Gabet 2004). As seen in **Figure 1c**, the SNP area receives greater precipitation than does the ACA, perhaps resulting in higher rates of weathering and soil formation in the SNP. However, once soil succession proceeds to the point of plant colonization, significant accumulation of microbial biomass does occur. Thus, while water holding capacity may only be a fair predictor of microbial biomass it appears to be a reasonable proxy for subnival soil development.

Although there is not a significant influence of soil bedrock on the overall microbial community biomass, we can see that the pH of the soils varies significantly depending on bedrock type and degree of plant colonization. Fierer and Jackson (2006) have shown that microbial community composition can vary significantly with changes in soil pH and Sinsabaugh et al. (2008) have demonstrated that this pH variation can result in different rates of extracellular enzyme activity. Indeed we see that cellulase (β-glucosidase) activity is very low in the shale barren soils (high pH) while it is one of the predominant enzymes in the granitic barren soils (low pH). The opposite trend was seen with protease (leucine aminopeptidase) activity, wherein activity was high in the barren shale soils and barely detectable in the granitic soils. Furthermore, as the soils became more *Kobreisia*-dominated there was an increase in extracellular enzyme activity in concert with a shift in soil pH. Therefore, as soils development proceeds, the differences in nutrient cycling between barren soil types may disappear. Moreover, the shift in patterns of snow accumulation and overall precipitation predicted to occur as the climate warms (Beniston 2003) may significantly influence rates of soil weathering and water holding capacity. Ultimately, said increases in soil weathering may alter the differences between bed rock types and result in an overall increase in productivity, biomass, and activity of these high altitude soils.

The microbial biomass C levels in the *Kobresia*-dominated soil patches of the ACA, with an average of 325 µg C/g soil, were also low for an alpine dry meadow community and suggestive of a low productivity system; *Kobresia myosuroides* dominated soils from the Rocky Mountains of Colorado, USA average 1260 µg C/g soil (King et al. unpublished data). Once again, the low precipitation of the ACA region may be the cause of the low microbial biomass by directly limiting plant photosynthesis and indirectly limiting the amount of carbon available to soil heterotrophs. However, relative to the soil C levels, the Nepal *Kobresia* soils had very high microbial N content (~54 µg N/g soil); levels very similar to *Kobresia* soils in Colorado (75 µg N/g soil, Fisk et al. 1998). This relationship results in a lower microbial C:N ratio for the Nepal sites (6.0) versus Colorado (16.8), further indicating increased microbial C-limitation for the Nepal soils.

High microbial N levels late in the fall may also be an adaptation that fosters N retention in this ecosystem over the winter. A similar retention mechanism occurs in other ecosystems via microbial growth on senescing plant material after the plant growing season, resulting in immobilization of N especially in seasonally cold (Jaeger et al. 1999, Zak et al. 1990) or seasonally dry (Singh et al. 1989, Vitousek and Matson, 1984) systems. This explanation is further supported by the low extractable nitrogen levels of the bulk soils from the *Kobresia* soil patches of the ACA (10 µg/g soil). Regardless of the mechanism for N storage, the data suggest that the *Kobresia* soil communities of the ACA have high N retention in the face of low productivity.

A final surprising result of our study is the finding that although *Kobresia* sward soils had the highest biomass of subnival Nepalese soils, the eroded soils adjacent to the *Kobresia* soils had the highest enzyme activity. This is likely a result of the eroded soils losing their structure, allowing the microbial community greater access to sequestered organic matter. This effect would be compounded by cessation of rhizodeposition of labile carbon sources, which could cause the microbial community to shift towards breaking down more recalcitrant organic matter, thus the increase in enzyme activity. In addition, the lower nitrogen availability in the eroded soils relative to the *Kobresia* sward may be responsible for the concurrent stimulation of N scavenging enzymes such as NAG and LAP. These effects have been theorized to explain the shift in dominance of extracellular enzyme production in

nutrient limited environments (Wallenstein and Weintraub 2008), but the majority of evidence for this phenomenon comes from studies of microorganisms in culture (Harder and Dijkhuizen 1983). Our results in the eroded soils support the long held hypothesis that high microbial enzyme activity should be higher in soils that have low amounts of labile nutrients relative to complex organic substrates.

Conclusions

The levels of microbial biomass and activity we observed in the subnival zone soils of the ACA and SNP regions of the Nepalese Himalayas are some of the lowest ever observed for this, or any, ecosystem type. It is likely that the subnival zone will further grow in size as a result of high-elevation glacial melt and in some areas by overuse of alpine grazing ranges, significantly increasing the already large area of Nepal that is occupied by this ecosystem type. The evidence from our work suggests that expanding areas of the subnival zone will be characterized by low levels of microbial biomass that increase gradually with water holding capacity and soil development. However, specific activity levels will be dependent on nutrient availability, soil pH, speed of soil development, and regional precipitation patterns. Our preliminary survey of microbial biomass and activity in the subnival zone of the Nepalese Himalayas reinforces the notion that the organisms that live in this precarious ecosystem are subjected to some of the most extreme environmental conditions on Earth. Further study of these areas has the potential to uncover novel microbial communities in these fascinating soils from the highest mountain range on Earth.

Acknowledgements

We are grateful to Bharat B. Shrestha, Ram P. Chaudhary, Sasha Reed, Mark Williams, Nima Sherpa, and Karma Gurung for helpful discussions and/or for assistance in the field. This work was supported by grants from the Microbial Observatories Program of the National Science Foundation of the USA (MCB-0455606) and the National Geographic Society Committee for Research and Exploration.

References

Ahmad A, JS Rawat and SC Rai. 1990. An analysis of the Himalayan environment and guidelines for its management and ecologically sustainable development. *The Environmentalist* **10**: 281–298

Barnett TP, JC Adam and DP Lettenmaier. 2005. Potential impacts of a warming climate on water availability in snow-dominated regions. *Nature* **438**: 303–309

Beniston M. 2003. Climatic change in mountain regions: A review of possible impacts. *Climatic Change* **59**: 5–31

Byers AC. 2007. An assessment of contemporary glacier fluctuations in Nepal's Khumbu Himal using repeat photography. *Himalayan Journal of Sciences* **4**: 21–26

Chang DHS. 1981. The vegetation zonation of the Tibetan Plateau. *Mountain Research and Development* **1**: 29–48

Costello EK, SRP Halloy, SC Reed, P Sowell and SK Schmidt. 2009. Fumarole-supported islands of biodiversity within a hyperarid, high-elevation landscape on Socompa Volcano, Puna de Atacama, Andes. *Applied and Environmental Microbiology* **75**: 735–747

del Valle HF, NO Elissalde, DA Gagliardini and J Milovich. 1998. Status of desertification in the Patagonian region: Assessment and mapping from satellite imagery. *Arid Soil Research and Rehabilitation* **12**: 95–121

Devore JL. 2004. *Probability and statistics for engineering and the sciences. Sixth Edition.* Belomont: Thomson, Brooks/Cole. 795 p

Fierer N and RB Jackson. 2006. The diversity and biogeography of soil bacterial communities. *PNAS* **103**: 626–631

Fisk MC, SK Schmidt and T Seastedt. 1998. Topographic patterns of above- and belowground production and nitrogen cycling in alpine tundra. *Ecology* **79**: 2253–2266

Freeman KR, MY Pescador, SC Reed, EK Costello, MS Robeson and SK Schmidt. 2009. Soil CO_2 flux and photoautotrophic community composition in high-elevation, "barren" soils. *Environmental Microbiology* **11**: 674–686

Gabet EJ. 2004. Climatic controls on hillslope angle and relief in the Himalayas. *Geology* 32, 629–632

Harder W and L Dijkhuizen. 1983. Physiological responses to nutrient limitation. *Annual Review of Microbiology* **37:** 1–23

Jaeger CH, RK Monson, MC Fisk and SK Schmidt. 1999. Seasonal partitioning of nitrogen by plants and soil microorganisms in an alpine ecosystem. *Ecology* **80**: 1883–1891

King AJ, AF Meyer and SK Schmidt. 2008. High levels of microbial biomass and activity in unvegetated tropical and temperate alpine soils. *Soil Biology and Biochemistry* **40**: 2605–2610

Ley RE, MW Williams and SK Schmidt. 2004. Microbial population dynamics in an extreme envrioment: controlling factors in talus soils at 3750 m in the Colorado Rocky Mountains. *Biogeochemistry* **68**: 313–335

Nagy L and G Grabherr. 2009. *The biology of alpine habitats*. New York: Oxford University Press. 436 p

Nemergut DR, SP Anderson, CC Cleveland, AP Martin and SK Schmidt. 2007. Microbial community succession in unvegetated, recently-deglaciated soils. *Microbial Ecology* **53**: 110–122

Panthi MP, RP Chaudhary and OR Vetaas. 2007. Plant species richness and composition in a trans-Himalayan inner valley of Manang district, central Nepal. *Himalayan Journal of Sciences* **4**: 57–64

Rawat GS and YPS Pangtey. 1987. Floristic structure of snowline vegetation in Central Himalaya, India. *Arctic and Alpine Research* **19**: 195–201

Schmidt SK, SC Reed, DR Nemergut, AS Grandy, CC Cleveland, EK Costello, MN Weintraub, AW Hill, AF Meyer, AP Martin and JC Neff. 2008. The earliest stages of ecosystem succession in high-elevation (5000 meters above sea level), recently deglaciated soils. *Proceedings of the Royal Society B* **275**: 2793–2802

Schmidt SK, DR Nemergut, AE Miller, KR Freeman, AJ King and A Seimon. 2009. Microbial activity and diversity during extreme freeze-thaw cycles in periglacial soils, 5400 m elevation, Cordillera Vilcanota, Perú. *Extremophiles* **13**: 807–816

Shrestha BB, PK Jha and MB Gewali. 2007. Ethnobotanical use and distribution of *Aconitum naviculare* in Upper Manang, Nepal. In: R.P. Chandhary TH, OR Aase, Vetaas and BP Subedi (eds.), *Local effects of global change in the Himalayas: Manang, Nepal*. Nepal: Tribhuvan University. 171–181 p

Singh JS, AS Raghubanshi, RS Singh and SC Srivastava. 1989. Microbial biomass acts as a source of plant nutrients in dry tropical forest and savanna. *Nature* **338**: 499–500

Singh P and L Bengtsson. 2004. Hydrological sensitivity of a large Himalayan basin to climate change. *Hydrological Processes* **18**: 2363–2385

Sinsabaugh, RL, CL Lauber, MN Weintraub, B Ahmed, SD Allison, C Crenshaw, AR Contosta, D Cusack, S Frey, ME Gallo, TB Gartner, SE Hobbie, K Holland, BL Keeler, JS Powers, M Stursova, C Takacs-Vesbach, MP Waldrop, MD Wallenstein, DR Zak, LH Zeglin. 2008. Stoichiometry of soil enzyme activity at global scale. *Ecology Letters* **11**: 1252–1264.

Swan LW. 1963. Aeolian zone. *Science* **140**: 77–78

Swan LW. 1990. The highest life. *Himalayan Journal* **46**: 125–133

Swan LW. 1992. The aeolian biome, ecosystems of the earth's extremes. *Bioscience* **42**: 262–270

Troll C. 1973. High mountain belts between the polar caps and the equator: their definition and lower limit. *Arctic and Alpine Research* **5**: 19–27

Tscherko D, M Bolter, L Beyer, J Chen, J Elster, E Kandeler, D Kuhn and HP Blume. 2003a. Biomass and enzyme activity of two soil transects at King George Island, Maritime Antarctica. *Arctic*

Antarctic and Alpine Research **35**: 34–47
Tscherko D, J Rustemeirer, A Richter, W Wanek and E Kandeler. 2003b. Functional diversity of the soil microflora in primary succession across two glacier forelands in the Central Alps. *European Journal of Soil Science* **54**: 685–696
Vitousek PM and PA Matson. 1984. Mechanisms of nitrogen retention in forest ecosystems: A field experiment. *Science* **225**: 51–52
Wallenstein MD and MN Weintraub 2008. Emerging tools for measuring and modeling the *in situ* activity of soil extracellular enzymes. *Soil Biology and Biochemistry* **40**: 2098–2106
Weintraub MN, LE Scott-Denton, SK Schmidt and RK Monson. 2007. The effects of tree rhizodeposition on soil exoenzyme activity, dissolved organic carbon, and nutrient availability in a subalpine forest ecosystem. *Oecologia* **154**: 327–338
Zak DR, PM Groffman, KS Pregitzer, S Christensen and JM Tiedje. 1990. The vernal dam: plant-microbe competition for nitrogen in northern hardwood forests. *Ecology* **71**: 651–656
Zemp M, W Haeberli, M Hoelzle and F Paul. 2006. Alpine glaciers to disappear within decades? *Geophysical Research Letters* 33: L13504, doi:10.1029/2006GL026319