Sara additions/revisions

Do not mind at the wrong figure & table progressive numeration (due to text reorganisation), I will revise it on the final manuscript version.

Rock weathering creates oases of life in a High Arctic desert

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Running title: rock weathering creates fertility in a cold desert

Summary

During primary colonization of rock substrates by plants, mineral weathering is strongly accelerated under plant roots, but little is known on how it affects soil ecosystem development before plant establishment. Here we show that rock mineral weathering mediated by chemolithoautotrophic bacteria is associated to plant community formation in sites recently released by permanent glacier ice cover in the Midtre Lovénbreen glacier moraine (78°53'N), Svalbard. Increased soil fertility fosters growth of prokaryotes and plants at the boundary between sites of intense bacterial mediated chemolithotrophic iron-sulfur oxidation, and the common moraine substrate where carbon and nitrogen are fixed by cyanobacteria. Microbial iron oxidizing activity determines acidity and corresponding fertility gradients, where water retention, cation exchange capacity and nutrient availability are increased. This fertilization is enabled by abundant mineral nutrients and reduced forms of iron and sulfur in pyrite stocks within a conglomerate type of moraine rock. Such an interaction between microorganisms and moraine minerals determines a peculiar, not yet described model for soil genesis and crop formation with potential past and present analogues in other harsh environments with similar geochemical settings.

Introduction

The factors affecting initial steps of soil formation, ecosystem development and plant establishment in extreme arid environments like cold and hot deserts, where the availability of water and nutrients limits biomass growth (Sundareshwar *et al.*, 2003; Yoshitake *et al.* 2007) are poorly constrained. In arid systems, organismic responses result from overcoming a threshold in water availability, that is key limiting factor (Lopez *et al.*, 2008; Ogle and Reynolds, 2004; Schwinning *et al.*, 2004). Such responses may trigger dramatic changes and, after a cascade of events, finally manifest in comparably immense increases of biomass in soil in conjunction with increase in biological diversity and profound changes in the landscape. Such a behavior holds also true for microbial life which include true primary colonizers either autotrophic like photosynthetic cyanobacteria, or heterotrophic microorganisms (Hodkinson *et al.*, 2002). For instance in biological soil crusts, certain cyanobacteria quickly respond to water inputs and move actively toward the moist soil surface or refuge in the deeper soil layers when soil undergoes drying conditions (Garcia-Pichel and Pringault, 2005).

Mineral weathering in soil enhances availability of water and nutrients, leading to an increase of fertility (Yoshitake *et al.*, 2007; Anderson *et al.*, 2000; Konhauser, 2007; Bashan *et al.*, 2002; Puente *et al.*, 2004; Puente and Bashan, 2004). Weathering occurs in hot and cold deserts (Adams *et al.*, 2002; Ascaso *et al.*, 1990). During primary colonization of rock substrates, mineral weathering is strongly accelerated under plant roots (Bashan *et al.*,2002; Uroz *et al.*, 2007) and enhanced by the association with rhizoplane microbiota (Carrillo-Garcia *et al.*, 1999; Bashan *et al.*, 2000), but little is known on how weathering affects the soil ecosystem development before plant establishment. Arctic glacier moraines recently released from permanent ice cover are ideal models for the assessment of factors associated with the initial development of a mature soil ecosystem. Within the grey-colored moraine of the Midtre Lovénbreen glacier (78°53’N), Svalbard, between the Hodkinson proglacial chronosequence sites ML2 and ML3, which were released from the

permanent ice cover 23 and 44 years ago, respectively (Hodkinson *et al.*, 2003), we found several sites densely colonized by the mosses *Ditricum flexicaule* and *Bryum* sp. and the vascular plant *Saxifraga oppositifolia* (Hodkinson *et al.*, 2003) (Fig. 1A, B). Green strips of flourishing vegetation were adjacent to rusty iron oxide-leaching strips sloping along the moraine with the latter departing from yellowish-rusty-greenish stones, rock conglomerates rich in reduced iron, in the form of pyrite. In certain areas of the moraine we found several of these rusty and green strips that were exclusively associated with one particular stone type.

Here, we present evidence that under harsh conditions of recently deglaciated moraines of the High Arctic, enhanced rock weathering mediated by chemolithoautotrophic bacteria is associated to soil formation and increases of soil fertility that fosters the early establishment and growth of plant biocenosis, the ecological community of plants that sustains the ecosystem.

Results and Discussion

We initially focused on one of these sites, named ML-RS1, estimated to be released by ice about 27 years ago. We observed several sites like ML-RS1 in the south-east portion of the moraine. We counted at least four sites similar to ML-RS1 with stones of similar size. We detected tens of smaller sites with much smaller stones where signs of the plant flourishing observed in site ML-RS1 were present. We also found a larger site named ML-RC1 with the same but more extended phenomenon in place (Fig. S1). The observed phenomenon occurred at a relatively high frequency in certain parts of the moraine, while in other areas we did not observe any sites similar to the ML-RS1. Plant flourishing occurred where the type of conglomerate rock like that of site ML-RS1 emerged to open air from the subsurface. For instance, site ML-RC1 looked like a large conglomerate vein cropping out from the grey moraine.

A higher density of plants was apparent in the green strip of site ML-RS1 compared to the surrounding grayish moraine. Plant, and/or eukaryotic algal sterols, dominated by campesterol and sitosterol were 10 to 100 times more concentrated in the green strips than in the surrounding moraine and rusty area, respectively (Fig. 1C). Intact polar lipids (IPLs), representative of live biomass (Sturt *et al.*, 2004; Biddle *et al.*, 2006; Lipp *et al.*, 2008), were also more abundant in the green strips, but had similar absolute amounts in the rusty and moraine area. The composition of IPLs was diverse and represented a mixture of eukaryotic and bacterial biomass, largely related to phototrophic organisms. Abundant IPLs were betaine lipids linked to mono- and polyunsaturated fatty acids, glycolipids esterified to polyunsaturated fatty acids and phospholipids with saturated and monounsaturated fatty acids (Table S1). This combination of lipids is typical of lower plants and arctic mosses (Dembitsky and Rezanka, 1995), but also some degree of cyanobacteria (Rezanka *et al.*, 2003) and bacteria involved in plant symbiosis (Lopez-Lara *et al.*, 2003). Ornithine lipids and bacteriohopanepolyols, which are exclusive bacterial markers (Lopez-Lara *et al.*, 2003; Talbot *et al.*, 2008), were indeed detected in low abundance throughout the transect with highest amounts outside the rusty area. Notably, sterol profiles show pronounced peaks adjacent to the rusty strip but are almost absent in the rusty strip, suggestive that the latter milieu is hostile for plants and eukaryotic microorganisms. In general the elevated biomass content adjacent to the rusty strip underscores the link between increased fertility by rock weathering and the establishment of an oasis of life.

To examine micro-environmental conditions promoting plant growth, we established a grid pattern for a detailed sampling of site ML-RS1 (Fig. 1d) along mini-transects at increasing distance from the stone. Total organic carbon, nitrogen percentage, potential nitrogen fixation and respiratory activity were higher in the green than in the rusty strip but similar to those in the surrounding grayish moraine (Figure 1C, Table S2). Stable nitrogen isotope values ($\delta^{15}N$) slightly below 0‰ showed that in all transects bacterial nitrogen fixation is the main source of nitrogen (Hogberg, 1997). The presence of heterocyst glycolipids (e.g. Gambacorta *et al.*, 1998) throughout the

transect, highly specific markers for heterocystous nitrogen-fixing cyanobacteria (data not shown), is consistent with this interpretation. These data strongly suggest that nitrogen supply is similar in the green and the grayish soil crusts and is probably not the limiting factor for plant growth in the green strip.

We determined parameters indicative of soil fertility in the grid pattern of site ML-RS1 after removal of the plant cover (Table S2). Mean values of fine particles, cation exchange capacity (CEC), water holding capacity (WHC), exchangeable Mg, K and Fe and Fe^{2+} were significantly higher in the green than in the grayish crusts. Relative increments, for the whole soil crust (Table 1), were between 36.2 (K_{exch}) and 132% (Fe_{exch}), with CEC and WHC respectively increasing of 95.2 and 48.1%. Nutrient differences are particularly meaningful considering that in the green strip plants act as element scavengers and bioaccumulators that continuously extract nutrients from the soil crust. The increased WHC markedly supports more favorable conditions for plant growth in this desert ecosystem that receives an average precipitation of 190 mm $year^{-1}$ (range, 88-265 mm in the period 1988-2002; http://www.svalbard.com/weather.html). Such low values indicate that water is the primary limiting factor for development of plant biocenoses (Lopez *et al.*, 2008; Ogle and Reynolds, 2004).

X-ray diffractometry and scanning electron microscopy-energy dispersive X-ray analyses of thin sections of the stone at site ML-RS1 showed pyrite microcrystals (Fig. 3). The stone was a conglomerate consisting mainly of quartz, cemented by kaolinite, with additional contributions of pyrite, aluminium-silicates, calcium hydroxyapatite, and manganese oxides. On the surface, besides jarosite and gypsum, iron oxy-hydroxides and oxides could be detected as well as small amounts of organic carbon (0.3% w/w), supporting the presence of microbial biomass.

A low pH was measured (Figure 1C, Table S2) in the rusty strip (down to 3.5; mean value along the rusty vertical transect, 5.2±1.4, n=10) and presented a raising gradient in the green strips (7.1±0.3, n=25) to reach neutral-sub-alkaline values in the grayish moraine (7.6±0.4, n=9). pH is significantly correlated with both total (Fe_{tot}) and exchangeable (Fe_{exch}) iron contents ($r=-0.959$, $P<0.01$, n=15,

and r=-0.932, P<0.01, n=15, respectively). We hypothesized that ferrous iron, present in the form of pyrite, was dissolved and transported from the stone, and subsequently oxidized to form the acidic rusty strip. Fe^{2+} oxidation was corroborated by the lower Fe^{2+} content in the rusty than in the green strips, despite Fe_{tot} and Fe_{exch} contents were significantly higher (Table S2). The hypothesis is consistent with the presence of jarosite [$KFe_3(SO_4)_2(OH)_6$] on the stone surface and within the rusty strip as shown by DRIFT spectroscopy (Fig. S2A). Jarosite has been used as a water signature on Mars (Klingelhofer *et al.*, 2004), but is unstable and rare under oxic conditions on Earth (Darmody *et al.*, 2007). Jarosite can be formed by spontaneous or microbially-mediated oxidation of pyrite (Sasaki and Konno, 2000). The evidence at site ML-RS1 is consistent with such a scenario.

The observed physicochemical differences between the rusty, green and grayish strips are reflected in the structures of the respective bacterial communities. Bacterial diversity was higher in the green and the grayish than in the rusty strips that appeared as the least equitable as shown by Shannon-Wiener and Equitability indices of bacterial amplified ribosomal intergenic spacer analysis (ARISA) (Table S3). Detrended correspondence analysis (DCA) of ARISA profiles tended to cluster the samples according to the soil crust pH, indicating that different bacterial communities were present in the different soil crusts (Fig. S3). Soil crust pH was strongly correlated with bacterial diversity (ARISA DCA axis 1 vs. pH, r=0.835, P<0.001 SI Fig. S3). Soil pH has already been shown as a major driver of biogeographical repartition of bacteria in soil (Fierer and Jackson, 2006). To identify bacteria inhabiting the soil crusts, clone libraries of amplified 16S rRNA genes were generated from six soil crust samples within the grid pattern, two for each color (rusty, greenish and grayish strips) in two horizontal transects sampled at 5 and 23 cm from the stone originating the leaching strips. A total of 540 clones were sequenced (Fig. 2, Table S4). The rusty soil crusts were dominated by α-Proteobacteria of the genus *Acidiphilium* (order Rhodospirillales; 30-32% of the clones in the two libraries, respectively) and by Acidobacteria (37-39% of the clones, respectively), both frequently associated to acidic environments where oxidation of iron and sulfur occur (Gonzàlez-Toril, 2003; Diaby *et al.*, 2007). The grayish soil crusts were dominated by

Cyanobacteria (29-40% of the clones, respectively) followed by α-Proteobacteria mostly affiliated with orders other than Rhodospirillales. In contrast, green soil crusts showed a bacterial community typical of plant-inhabited soils. Hence, at the boundary between a cyanobacterial carpet typical of substrates recently released by permanent ice cover (Nemergut *et al.*, 2007), and iron weathering spots, the geochemical conditions selected a rhizosphere-like bacterial community able to support a plant biocenosis. We suggest that the major driver of such community transition was the pH gradient resulting from the activity of iron-oxidizing bacteria.

Ornithine lipids are consistent with the presence of bacteria involved in redox transformation of iron and sulfur (Gosh and Mishra, 1987) and have been recently observed in the highly acidic, iron-rich Rio Tinto river in the Iberian Pyrite Belt (Bühring et al., unpublished data). These lipids were suggested to play an important role in plant-bacteria symbiosis and acid tolerance (Rojas-Jiménez *et al.*, 2005). By inoculating an organic carbon-free medium added with reduced iron sulfate either with the stone surface or the rusty soil crust, we isolated rod shaped bacteria able to grow at 4°C and pH 2 and to produce jarosite (Fig. S2B). The isolates were identified as *Acidithiobacillus ferrooxidans* by 16S rRNA gene sequencing and contained ornithine lipids as a major lipid type, next to phosphatidyl-(N)-methylethanolamine and phosphatidylglycerol, all of which are present in the rusty strip and surrounding area. At a second site (ML-RC1) that resembles site ML-RS1 (Fig. S1), *A. ferrooxidans* counts decreased from 1.7×10^{4} cells g^{-1} close to the rock to 3.0×10^{0} at about 10 m distance from the leaching origin.

It can therefore be envisaged that an acidophilic iron-oxidizing bacterial community finds suitable conditions at specific sites within the Midtre Lovénbreen moraine, where stones rich in reduced iron and sulfur are present. Progressive stone fracturing and surface desegregation due to winter freezing, and chemical and biological weathering, and the water-mediated particle dragging maintained a constant pyrite supply over time on the substrate slope downstream the stone allowing the establishment of a chemolithoautotrophic bacterial community (Fig. S4). Stone leaching during summer snow melting created a gradient along the slope with decreasing iron concentration and

acidification, as shown by the increasing pH along the rusty strip (Table S2). Oxidation of reduced iron released from the stone caused jarosite and ferric oxy-hydroxide formation responsible for the increase of both soil crust specific surface area (SSA) and CEC, the latter determining a higher WHC and nutrient content in the green strip (Table 1; Fig. S4B). The positive and negative correlations between SSA and total and reduced iron contents, respectively $r=0.835$, $P<0.0001$ ($n=15$) and $r=-0.599$, $P<0.01$ ($n=15$), supported such a mechanism. Hence, with respect to the rest of the moraine, plant establishment was favored by the combination of an increased soil crust fertility (Fig. S4C) driven by the acidification and leaching activity of a chemolitotrophic community strongly influenced by iron oxydizers, and by cyanobacteria-mediated primary productivity.

The synergy between these two bacterial autotrophic processes in favoring microorganism and plant colonization is probably not restricted to the Midtre Lovénbreen glacier foreland, but should be explored in other environments where similar geochemical conditions exist (Anderson *et al.*, 2000; Darmody *et al.*, 2007; Skidmore *et al.*, 2005), such as natural pyritic belts (Fernàndez-Remolar *et al.*, 2005) and man-made pyritic mine tailings (Southam and Beveridge, 1992). Such a synergy could also have played a role in the primordial land colonization by plants (Rensing *et al.*, 2008) or in soil formation on extraterrestrial bodies where signatures of acidic aqueous systems with iron- and sulfur-based redox cycles have been found (Bibring *et al.*, 2007).

Experimental procedures

Soil crusts in the moraine of the Midtre Lovénbreen glacier, Ny Ålesund, Svalbard, were collected during three expeditions in August 2004 and 2005 and early September 2006 within the sampling grid designed on site ML-RS1 (Midtre Lovénbreen Rusty Site 1; 78°53.753'N, 12°05.115'E; 57.1 m a.s.l.) and along the slope of site ML-RC1. The top 5 mm of the crusts were separated from the

underlying mineral matter and the two fractions were separately processed. Recovered samples were stored at -20°C for chemical and nucleic acid-based studies, or at 4°C for microbiological enrichments or activity measurements.

Soil crust and stone chemical analysis

Soil parameters were determined as follows: Soil crusts pH in aqueous solution using a 1:2.5 sample/water ratio; fine (< 2 mm) and coarse (> 2 mm) particles by soil sieving after drying; total nitrogen by Kjeldahl method; available-P by using the Olsen and Bray and Kurtz methods depending on the pH; organic carbon by wet oxidation. For CEC and exchangeable Ca, Mg, K, Na determinations, samples were saturated with $BaCl_2$-Triethanolamine solution (pH 8.1) and exchangeable cations were determined by Inductively Coupled Plasma (ICP-MAS VARIAN, Liberty AX, Walnut Creek, CA). Total Ca, Mg, K, Na, Fe and Al were determined by samples digestion with HNO_3 (16 mol L^{-1}) in a microwave furnace (CEM Mars 5, Matthevs, North Caroline), and successive detection by Inductively Coupled Plasma. All the above methods were as described in (Pansu and Gautheyrou, 2006). WHC was determined by the Stackman box method (Klute, 1986). Exchangeable iron forms were extracted with EDTA or DTPA depending on crust pH (Lindsay and Norwell, 1969; Lakanen and Ervio, 1971) and determined with an Inductively Coupled Plasma. Reduced iron forms were extracted with the colored EDTA-BPDS solution and quantified spectrophotometrically at 535 nm (Wang and Peverly, 1998). SSA of soil crust samples were determined by N_2 adsorption (BET method) of dried samples by using a sorptometer apparatus (Quantachrome NovaWin2, NOVA Instruments, Boynton Beach, FL, USA). All analytical data were determined on the fine particle fraction (< 2mm) (Pansu and Gautheyrou, 2006). Parameters with statistically significant differences between the grayish and the green crusts (Table S2) were also referred to the whole sample (Table 1) by taking into account the fine and the coarse particle contents of the whole soil crust and their standard deviations recalculated according to statistical standards (Ciani *et al.*, 1990). All the analyses were made in triplicate and the results were analyzed

by ANOVA, considering the color strips as independent variables. Values of the means were separated by Duncan test, considering a significance level of $P<0.05$. All statistical analyses were performed using the SPSS 13.0 software package (SPSS International, Chicago, IL, USA).

Stable nitrogen isotopes were determined from 20-30 mg of homogenized soil on an elemental analyzer (ThermoQuest EA/NA 1110) coupled to a Thermo Finnigan Delta Plus isotope ratio mass spectrometer via a ConFlow II interface. Analyses were calibrated with a known standard (Wadden Sea soil) and were normalized to the international standard IAEA-CH-6, sucrose.

Shine thin sections of stone samples for ESEM-EDX (Environmental Scanning Electron Microscopy-Energy Dispersive X-ray) spectroscopy were visualized by a scanning electron microscope (FEI Quanta mod. 200, Endhoven Nl) at an acceleration voltage of 25 kV to determine elements in soil particles (triplicate) at 1.0 and 0.1 bar after 100 sec scanning. Mean concentrations and standards deviations of each element were calculated from three random determinations at different spots on the samples. The X-ray beam was 4 μm wide and penetrated to a depth of 2 μm.

X-ray diffractometry was performed on samples collected from the surface of the conglomerate on the basis of the different coating colors, reddish and yellowish, and from the internal part of the rock. Due to the rock nature was easy to break and separate the first 2-3 mm of the rock. Powdered samples were analyzed by a Philips PW 3830 X-ray diffractometer using CoKα radiation incident from 3 to 75°θ with a step size of 0.02°θ and a scan step time of 1s. Total C and N content were determined using a Carlo Erba (Milan, Italy) NA 1500 CHNS Analyzer. To distinguish organic from inorganic C was used a procedure reported in (Santi *et al.*, 2006). Two aliquots of each sample, in two replicates, were analyzed by dry combustion: the first without treatment, to assess the total C and N content and the other after treatment with excess HCl, for carbonate removal, to assess only the organic C content.

Rock sample and biologically made jarosite were analyzed by Diffuse Reflactance Infrared Fourier Transformed (DRIFT) spectroscopy using an Avatar 370 FT-IR from ThermoNicolet Instruments (Madison, WI, USA). Samples previously dried at 65°C for 48 h, and KBr (FT grade, Aldrich

Chemical Co, ST Louis, Missouri) were mixed in the 1:10 ratio (w/w) and finely ground for 10 minutes using an agate ball mill (Specamill-Greseby-Specac, Kent, UK). Instrument parameters used were: scanning 128, resolution 4 cm^{-1}, and frequency 400-4000 cm^{-1} gain 16. Peaks assignments were made according to (Sasaki and Konno, 2000).

Lipid biomarkers

Lipid analysis followed the procedures outlined in (Sturt *et al.*, 2004). In brief, 5-10 g of freeze-dried soil was extracted using a modified Bligh and Dyer protocol. Total lipid extracts (TLE) were analyzed on a ThermoFinnigan LCQ Deca XP Plus HPLC-MS system in positive and negative ionization mode. Quantification of IPLs was achieved using an internal standard (1-O-hexadecyl-2-acetoyl-sn.glycero-3-phosphocholine) and external calibration of this standard. For the analysis of BHPs, an aliquot of the TLE was acetylated with acetic anhydride/pyridine (1:1) for 1h at 50°C and subsequently measured on the same HPLC system using conditions described in (Talbot *et al.*, 2008). Sterols were analyzed in an alcohol fraction that was isolated by solid-phase extraction and derivatized according to (Hinrichs *et al.*, 2000). The fractions were measured on a ThermoFinnigan Trace GC-MS using instrument parameters described previously (Hinrichs *et al.*, 2000).

Microbial community studies

Total DNA was extracted with the FastDNA™ SPIN Kit for Soil (BIO 101 Systems Q-BIO gene; CA, USA) following manufacturer's instructions. ARISA fingerprinting was determined as described elsewhere (Cardinale *et al.*, 2004) and downstream multivariate statistical analyses were done with MVSP software (Kovach Computing Services, UK). 16S rRNA gene clone libraries from selected soil crust samples of site ML-RS1 were prepared as previously described (Van der Wielen *et al.*, 2005). 100 clones have been randomly chosen from each library. 600-800 bp long sequences, corresponding to the first half of the 16S rRNA gene, were aligned with ClustalX (Thompson *et al.*, 1997), and operational taxonomic unit (OTU) distribution was calculated with Vector NTI Advance

10 using a 97% identity threshold. Good coverage of the dominant OTUs within libraries was evaluated by rarefaction analysis with PAST software (Hammer *et al.*, 2001).

Isolation and cultivation of chemolithoautotrophs

One g of rusty soil crusts or stone surface were inoculated in carbon-free 9K medium supplemented with ferrous sulfate (Lizama *et al.*, 1988) and incubated at 4°C for 4 weeks until a rusty precipitate of iron oxides was visible, and bacterial cells were observable. Three subsequent steps of terminal dilution to extinction enabled obtaining pure cultures of rod-shaped bacteria. A 16S rRNA gene library from the culture was established, and the sequencing of several clones revealed identical sequences having 99% identity with the 16S rRNA gene of *A. ferrooxidans*. The isolated strains were cultured in modified 9K (Silvermann *et al.*, 1959) supplemented with either ferrous sulfate or powdered pyrite (0.5% W/V). At 28°C the strains produced jarosite after 3-days incubation. Most probable number counts of autotrophic iron oxidizing bacteria were performed on soil crusts from site ML-RC1. Soil crusts were inoculated (1% W/V) in decimal serial dilutions in modified 9K medium supplemented with reduced iron sulfate in 3 ml microtiter plates. After 4 weeks incubation at 15°C growth was positively scored when a rusty precipitate was observed in the wells.

Activity measurements

Potential nitrogen fixation activity of soil samples was measured as described elsewhere (Zielke *et al.*, 2002). Freshly collected one cm thick top layer soil crust samples (surface area, 64 cm^2) were conditioned at room temperature (24°C) in natural light and 0 water potential (water saturated) for 17 hours prior to incubation with 10% acetylene (v/v) for three hours in the same conditions. Soil crust respirations were measured by trapping with alkali the CO_2 produced during soil crust incubation at 20°C in the laboratory for 21 days (Bekku *et al.*, 1997). All the analyses were performed in triplicate.

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Table and figure legends

Table 1. Parameters indicative of soil fertility in the greyish and green soil crusts of site ML-RS1 and relative percentage increments in the green respect to the greyish crusts over the whole soil crust (fine plus coarse particle fractions).

Parameters[a]	Soil crust type[b]		Relative increment[c]
	greyish	green	(%)
CEC ($cmol^{(+)}$ kg^{-1} dm)	2.9±0.9	5.7±1.5	95.2
WHC (g 100 g^{-1} dm)	21.6±2.9	32.0±8.7	48.1
Fe_{exch} (mg kg^{-1} dm)	65.4±27.8	151.8±28.5	132.0
Fe^{2+} (mg kg^{-1} dm)	34.2±6.2	50.0±5.4	46.4
Mg_{exch} (mg kg^{-1} dm)	73.4±20.4	113.6±15.8	54.7
K_{exch} (mg kg^{-1} dm)	103.2±29.0	140.8±35.1	36.2

[a] CEC, Cation exchange capacity; WHC, Water holding capacity; exch, exchangeable.

[b] All means are significantly different ($P<0.05$) according to Duncan test.

[c] Relative percentage increments of parameters in the green crust respect to the greyish crust

Figure 1. The ML-RS1 site in the moraine of Midtre Lovénbreen glacier, Ny Ålesund, Svalbard. (*a*), A rusty leaching strip departs from a stone within the grey-colored moraine (white bar = 50 cm). White arrows indicate zones where dense plant biomass can be observed. A detail of an area densely-colonized by plants within the site is shown in (*b*) (white bar = 50 cm). (*c*), Relative and absolute abundance of different lipid biomarkers in soil crusts (s.c.) reflecting prokaryotic and eukaryotic biomass along transect A (*c*1) and B (*c*2). Shown are total intact polar lipids (IPLs), bacterially derived ornithine lipids (OL) and bacteriohopanepolyols (BHPs) consisting of bacteriohopane-32,33,34,35-tetrol (BHT) and 35-amino-bacteriohopane-32,33,34-triol (aminotriol) and plant sterols. Relative quantities of BHPs were normalized to the sample with maximum concentration. (*d*), map of the area showing the position of the sampling transects

Figure 2. Bacterial diversity in the soil crusts of site ML-RS1 in the moraine of Midtre Lovénbreen glacier, Ny Ålesund, Svalbard. Each graph shows the relative composition of the bacterial community inhabiting the soil crust. A4 and B4 were samples from the rusty soil crust, A6 and B6 from the green soil crusts and A9 and B9 from the grayish soil crusts.

Figure 3. Pyrite microcrystals and oxidation signatures on the surface of the stone upstream the rusty strip of site ML-RS1. (*a*) and (*c*), SEM-EDX back scattering electron micrographs of thin layer shiny sections of the stone, showing pyrite and apatite microcrystals (circled), respectively. Peaks attributable to the different elements of pyrite and apatite crystals, circled in (a) and (c), are shown in (*b*) and (*d*), respectively. (*e*), X-ray diffractograms of the surface and the internal part of the conglomerate. Diffractograms of the yellowish and rusty coatings and the internal part of the stone are shown in red, green and blue colors, respectively. Marked peaks are G, gypsum; J, jarosite; K, kaolinite; P, pyrite; Q, quartz.

Figure 4. A schematic of the overall process occurring in ML-RC1 site in the moraine of the Midtre Lovénbreen glacier, Svalbard.

Supporting information

Table S1. Fatty acid composition of IPL-glycerol derivatives. Major fatty acids found of IPL-glycerides along the two transects A and B as identified with HPLC-ESI-MS in positive and negative ion mode.

Table S2. Chemical, physical and microbiological parametersa in the greyish, green and rusty soil crusts after removal of plant cover.

Table S3. Diversity indices of bacterial communities in soil crusts of site ML-RS1. Shannon-Wiener and Equitability Indices deduced from ARISA profiles and 16S rRNA gene libraries of the bacterial community in different transects within the site ML-RS1. Means ± SD are reported.

Table S4. Results of the screening of 16S rRNA libraries from rusty soil crust (RUS), green soil crust (GRE) and greyish soil crust (GRY). The percentage of identified clones at phylum, class, order, and/or family and/or genus has been reported for each taxonomical rank as the average of samples of transect A and B.

Fig. S1. The ML-RC1 site in the moraine of the Midtre Lovénbreen glacier, Svalbard. This site is bigger than site ML-RS1 with 10 m-long rusty leaching strip departing from a rock amphitheater. The soil crust was sampled about every 2.5 meters from the origin close to the rock amphitheater to the end of the rusty color in the gray moraine (not visible). *Acidithiobacillus ferrooxidans* most probable number counts, determined on the samples, showed decreasing values from the origin to the end (respectively 2.3×104, 2.3×103, 6.8×102, 0.8×101 and 2.3×100 CFU g-1 wet soil).

Fig. S2. DRIFT spectra indicating the presence of the mineral jarosite. Relevant peaks used as signature for jarosite are indicated. a, DRIFT spectrum of the soil crust from the rusty strip of site ML-RS1. Several signature peaks of jarosite are indicated. b, DRIFT spectrum of jarosite formed by *Acidithiobacillus ferrooxidans* SB1 isolated from site ML-RS1, while growing on 9K medium containing ferrous sulfate.

Fig. S3. Bacterial community structure and diversity in the soil crusts of site ML-RS1. Detrended Correspondance Analysis (DCA) of the ARISA profiles obtained from all the samples withdrawn in four (A to D) minitransects of the site. Samples were labeled with different colors according to the pH of the soil crust. Circles indicate the soil crust samples, while triangles indicate the soil withdrawn 5 cm below the soil crusts. Letters and numbers accompanying each sample respectively indicate the transect and the sample within the transect. Soil crust samples showed a grouping trend according to the pH, while deeper samples did not. The inset shows the correlation curve between the ARISA-based DCA axis 1 diversity index and the pH of the samples.