

Could Fe-metabolizing microbes weather sub-surface minerals in a semi-arid climate?

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Mineral weathering shapes Earth's surface by transforming bedrock to soil in the 'critical zone'. Among these transformation processes, microbial weathering plays an important role, as it contributes to all stages of rock-soil transformation such as primary rock colonization, rock breakdown, saprolite formation, and element cycling. Fe-metabolizing microorganisms, i.e. **Fe(II)-oxidizing and Fe(III)-reducing microorganisms**, are key players in weathering as they can directly attack minerals via their metabolism. However, most direct evidence for the role of these microbes in critical zone processes comes from shallow and humid tropical soils and saprolite, or from transects across corestones. Much less is understood about the direct role of these microorganisms in critical zone processes in more arid climates.

In this study we have obtained drill cores from the critical zone of a semi-arid region of the Chilean Coastal Cordillera (**Santa Gracia Reserve**). Despite receiving only 66 mm of rain per year, the weathering profile is very deep (>80 m). The rock material of the drill core is a Cretaceous quartz monzodiorite rich in hornblende, biotite and chlorite with ca. **1-2 wt.-% Fe(III) oxyhydroxides and very low TOC content**. Using cultivation-based methods we found **microaerophilic Fe(II)-oxidizing bacteria in zones of weathered saprolite** (up to ca. 25 m depth) **and at the weathering front** (70-76 m), while Fe(III)-reducing bacteria, grown either with dihydrogen or organic carbon, were successfully enriched from samples across the whole 87 m profile. A robust contamination control confirmed that cultivated microbes were from the *in-situ* community and not related to drill fluid contamination.

These findings suggest there is potential for Fe-metabolizing microbes to contribute to mineral-weathering processes even in deep weathering profiles in semi-arid environments. The occurrence of cultivatable Fe(II)-oxidizing bacteria is controlled by the presence of highly fractured zones functioning as fluid and oxygen transport pathways. It is notable that despite the fact that much of the silicate minerals contain Fe(II), Fe(III)-reducing bacteria are more common. The co-occurrence of Fe(II)-oxidizing and Fe(III)-reducing bacteria in some isolated parts of the profile could represent a self-sustaining cycle of iron redox reactions.



Chile – Earthshape field sites and their climate

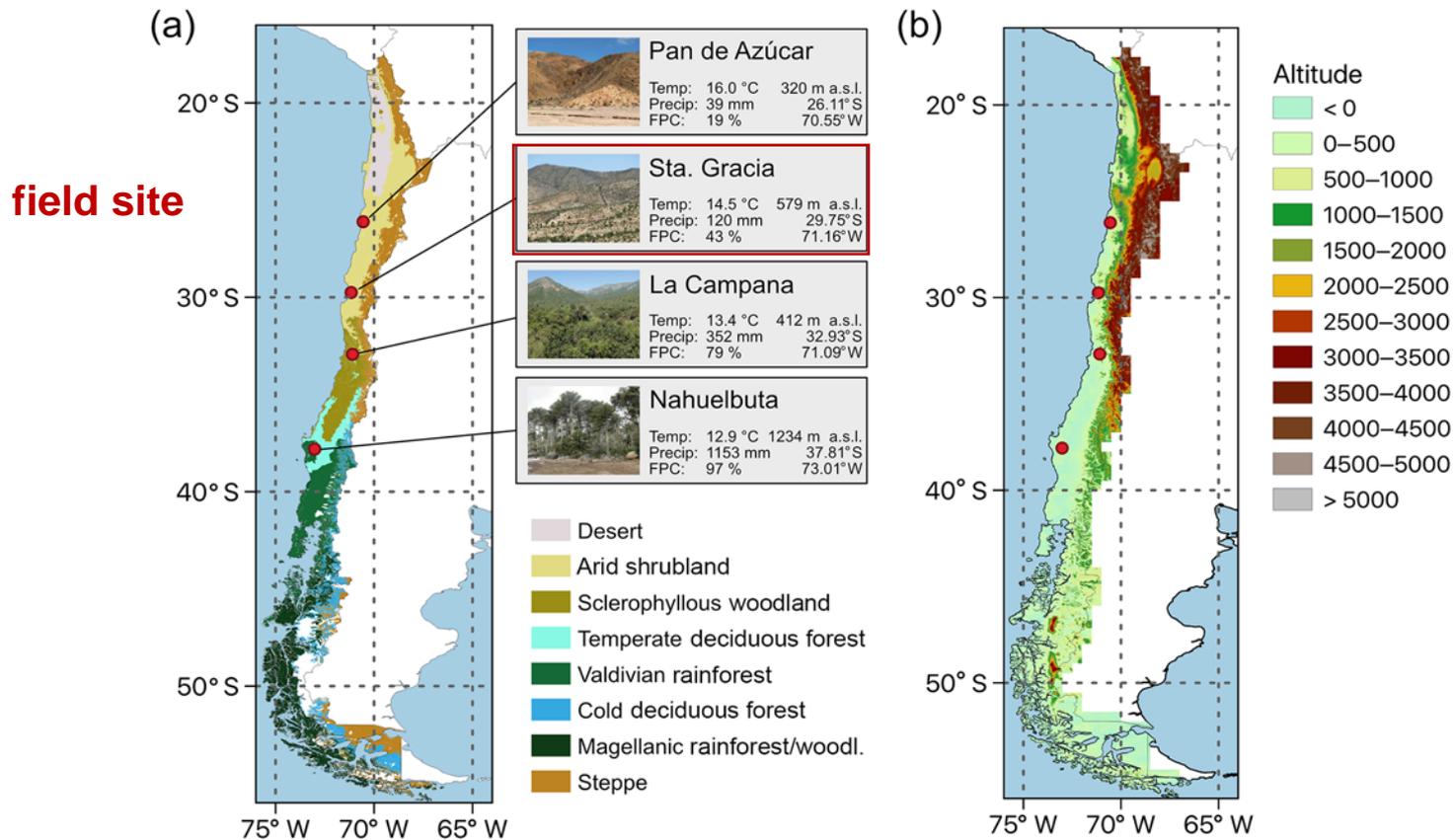


Figure 1. (a) Distribution of major vegetation zones in Chile and the location of the four EarthShape SPP focus sites: Pan de Azúcar, Sta. Gracia, La Campana, and Nahuelbuta (“Temp” represents average annual temperature, “Precip” represents average annual precipitation – data: ERA-Interim 1960–1989, and “FPC” represents foliage projected cover (MODIS VCF v6, total vegetation cover, 2001–2016 average, Dimiceli et al., 2015). Vegetation zones are based on Luebert and Pliscoff (2017). (b) Elevation of the model domain. (from Werner et al., 2018)

How do microbes contribute to rock weathering?

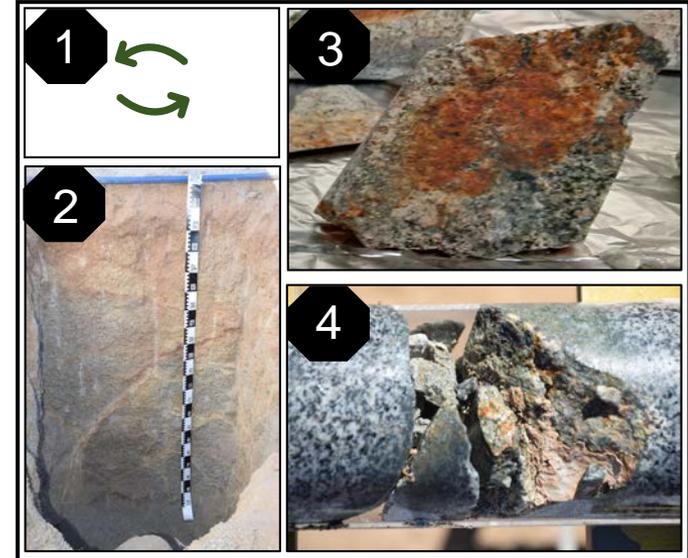
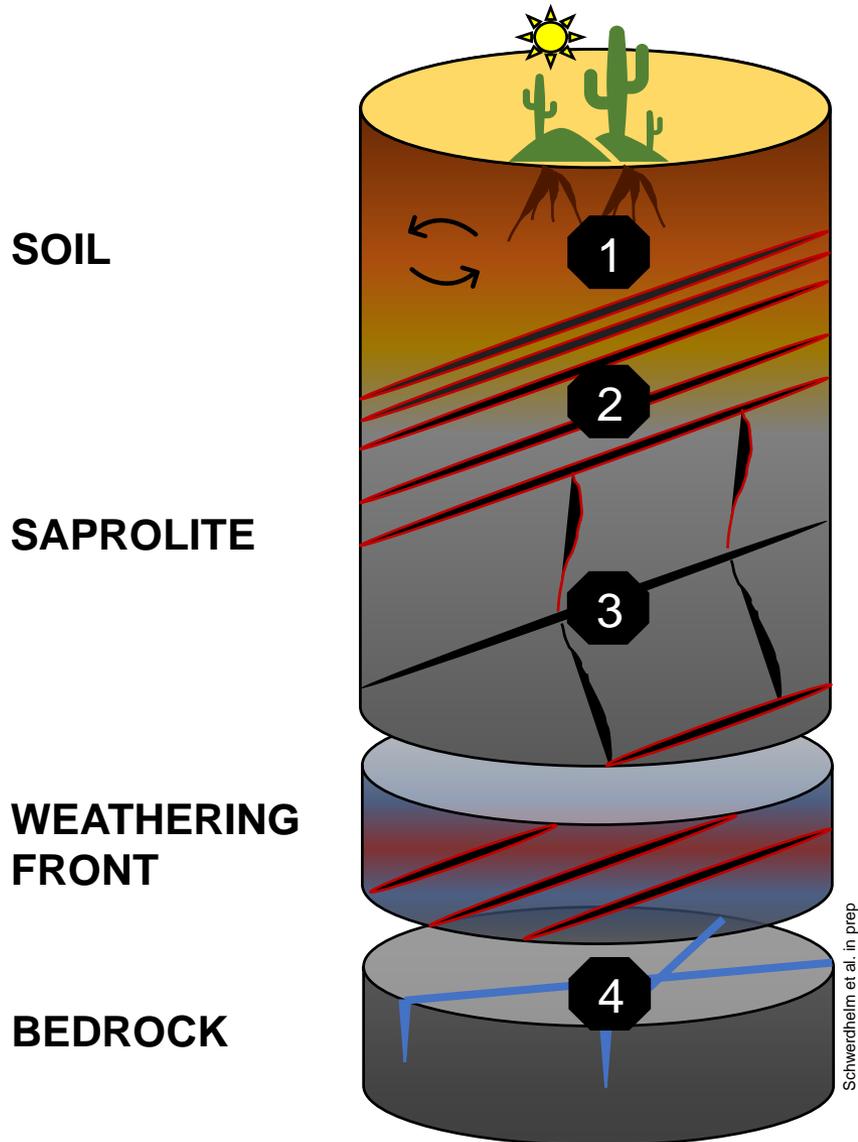


Figure 2. Representative pictures (1) - (4) for microbial mechanisms of rock weathering, as indicated in box below (Schwerdheim et al. in prep).

Microbial mechanisms

1. Element cycling in soils
2. Saprolite / soil formation
3. Rock breakdown
4. Primary rock colonization

Microbes can directly weather rocks and minerals

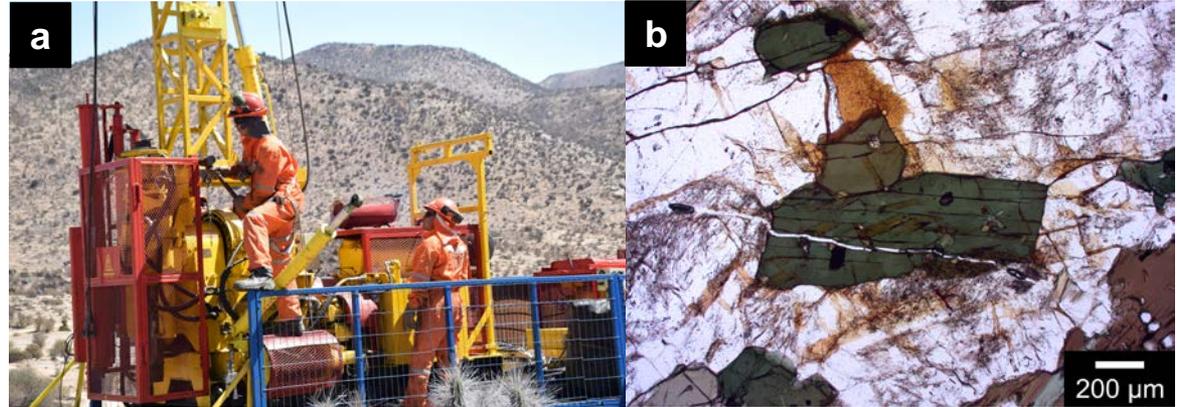
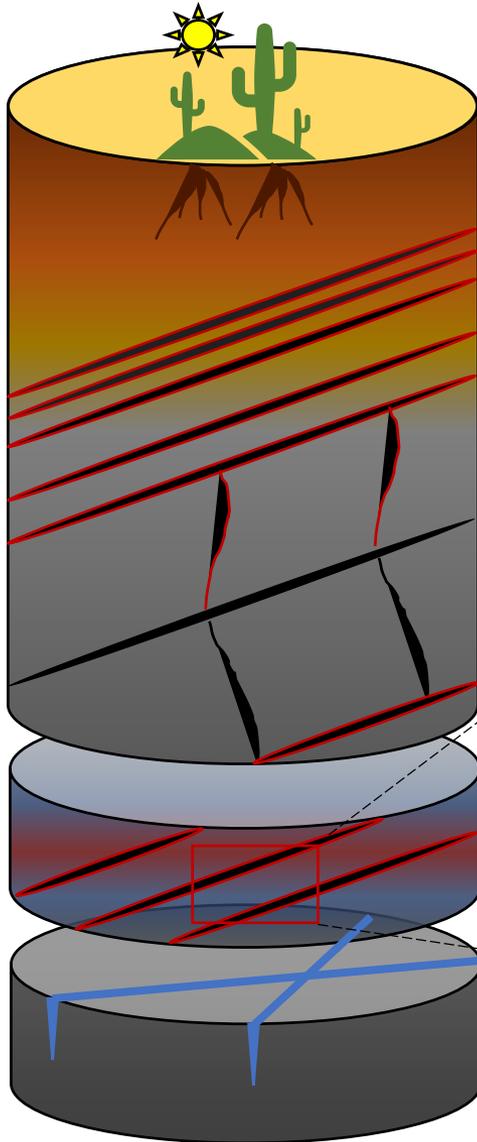
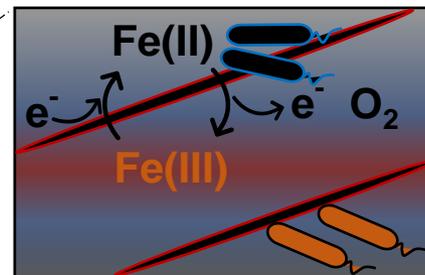


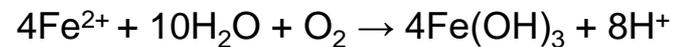
Figure 3. (a) Drill rig in action during Deep Earthshape drilling campaign in Santa Gracia (Chile) in March and April 2019. Final drill depth in Cretaceous quartz monzodioritic bedrock was 87.2 m. (b) Thin section of weathered hornblende from 11.8 m depth in transmitted light, a potential energy source for iron-metabolizing bacteria (photo: Ferdinand Hampl, TU Berlin).



= mFeOx ^{*1}
 = FeRed ^{*2}

Microaerophilic Fe(II)-oxidation

- direct attack via redox reaction -



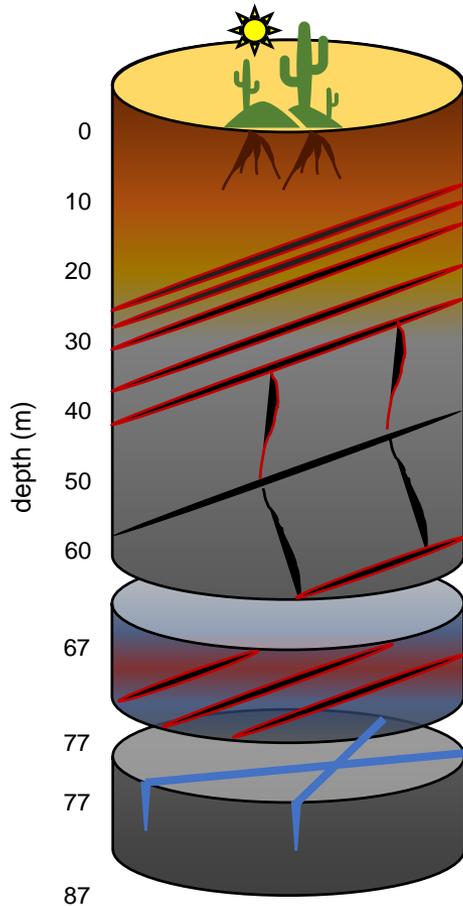
$$\Delta G_r = -16.9 \text{ kJ mol}^{-1} \text{ (pH 7, } I = 0.5 \text{ M)}$$

^{*1} mFeOx = microaerophilic Fe(II)-oxidizing bacteria

^{*2} FeRed = Fe(III)-reducing bacteria



Site characterization I - mineralogy



Major iron and iron-bearing silicate minerals found in Santa Gracia
 hornblende > biotite > chlorite > magnetite > hematite (Hampl et al. in prep)

Overall mineralogical data of drill core samples

Total iron concentration (wt. %): 3.6 - 5.5

TOC content (wt. %): < detection limit (d.l.); d.l. = 0.02 wt. %

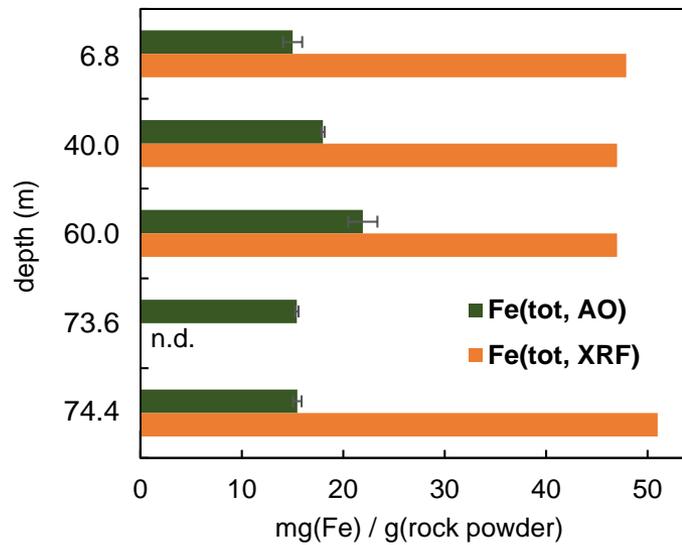


Figure 4. Total iron content of drill core samples from Santa Gracia (Chile) extracted with 0.2 M ammonium oxalate (AO) (mg Fe / g rock powder) (protocol modified from Schwertmann, 1964) and from XRF measurements normalized on Fe (XRF data: Laura Krone, GFZ Potsdam). Data points and error bars denote the mean and range of duplicate extractions. n.d. means no data due to no measurement.

Site characterization II - microbiology

Table 1. Characteristics of screening for Fe-metabolizing bacteria.

Parameter	Fe(II)-oxidizing bacteria (mFeOx) ●	Fe(III)-reducing bacteria (FeRed) ●
pH	7	7
replicates (n)	3	6
cultivation vessel	gradient tubes 	deep well plates 
cultivation time	1 week	6 weeks
electron acceptor	5 - 20 $\mu\text{M O}_2$	5 mM ferrihydrite
electron donor	1 mM Fe(II) *	5 mM $\text{C}_{\text{org}} / \text{H}_2$

* 1 mM Fe²⁺ is originating from the FeS bottom plug (see photo).

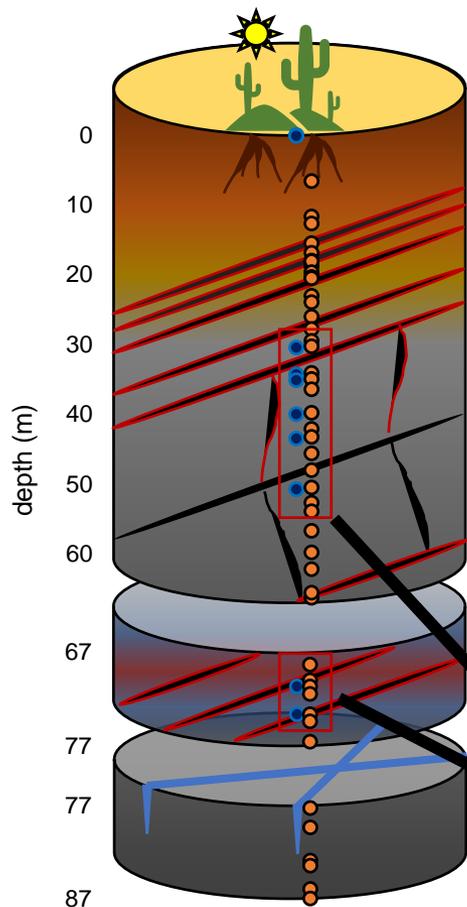


Figure 5. Depth profile of enriched Fe(II)-oxidizing and Fe(III)-reducing bacteria (blue and orange circles respectively) from Santa Gracia drill core samples.

OBSERVATION I

Fe(II)-metabolizing bacteria are associated with fracture zones.

EXPLANATION I

Fe(II)-metabolizing bacteria could be enriched, because they successfully compete with abiotic Fe(II) oxidation. Fracture zones function hereby as fluid and oxygen pathways.

OBSERVATION II

There is a co-occurrence of Fe(II)-oxidizing and Fe(III)-reducing bacteria in some isolated parts of the profile.

EXPLANATION II

The co-occurrence of Fe(II)-oxidizing and Fe(III)-reducing bacteria in some isolated parts of the profile could represent a self-sustaining cycle of iron redox reactions.

Results at a glance

1. **Poorly crystalline ammonium oxalate extractable iron** represents about one third of total iron present in drill cores of Santa Gracia. Silicate minerals and magnetite are a major source of Fe(II).
2. **Fe(III)-reducing bacteria** could be cultivated from the whole depth profile range in contrast to **Fe(II)-oxidizing bacteria**, of which the latter are associated with highly fractured zones functioning as fluid and oxygen pathways.
3. The **co-occurrence of Fe(II)-oxidizing and Fe(III)-reducing bacteria** in some isolated parts of the profile could represent a self-sustaining cycle of iron redox reactions.

These findings suggest there is potential for Fe-metabolizing microbes to contribute to mineral-weathering processes even in deep weathering profiles in semi-arid environments.

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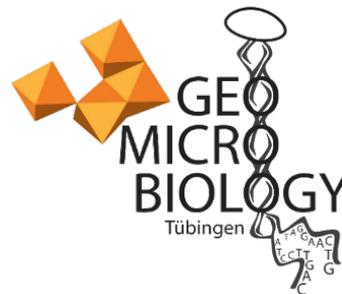
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Werner, C., et al. (2018). "Effect of changing vegetation and precipitation on denudation – Part 1: Predicted vegetation composition and cover over the last 21 thousand years along the Coastal Cordillera of Chile." Earth Surf. Dynam. **6**(4): 829-858.