A Contemporary Microbiially Maintained Subglacial Ferrous “Ocean”

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An active microbial assemblage cycles sulfur in a sulfate-rich, ancient marine brine beneath Taylor Glacier, an outlet glacier of the East Antarctic Ice Sheet, with Fe(III) serving as the terminal electron acceptor. Isotopic measurements of sulfate, water, carbonate, and ferrous iron and functional gene analyses of adenosine 5′-phosphosulfate reductase imply that a microbial consortium facilitates a catalytic sulfur cycle. These metabolic pathways result from a limited organic carbon supply because of the absence of contemporary photosynthesis, yielding a subglacial ferrous brine that is anoxic but not sulfidic. Coupled biogeochemical processes below the glacier enable subglacial microbes to grow in extended isolation, demonstrating how analogous organic-starved systems, such as Neoproterozoic oceans, accumulated Fe(II) despite the presence of an active sulfur cycle.

Subglacial environments represent a largely unexplored component of Earth’s biosphere (1). In the McMurdo Dry Valleys, Antarctica, an iron-rich subglacial outflow (Blood Falls) flows from the Taylor Glacier (Fig. 1A), providing unique access to a subglacial ecosystem. The likely fluid source to Blood Falls is a pool of marine brine of unknown depth trapped underneath the glacier ~4 km from the glacier snout where the overlying ice is ~400 m thick (2). Pliocene surface uplift of the Taylor Valley floor, and the associated recession of the Ross Sea Embayment, isolated this pocket of brine (3). Before isolation from direct contact with the atmosphere, the brine was cryoconcentrated (~5, resulting in hypersalinity (~1375 mM Cl). This brine has been isolated for at least 1.5 million years (My), when the Taylor Glacier last advanced over the area (6). Although the brine at

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Materials and Methods
Figs. S1 to S11
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or dissimilatory (APS) metabolisms, although APS also has been identified in sulfur oxidizers and certain organisms that only assimilate sulfate (18). The majority of APS genes detected in Blood Falls brine (Fig. 2) clustered with APS sequences of known dissimilatory and sulfur-disproportionating species (group 1). Sequences closely related to the APS gene from Desulfocapsa sulfexigens are consistent with the presence of a relative of this species among the 16S ribosomal RNA clones (7). Clone APS_20 (group 2) shares relations with sulfur-oxidizing isolates and Thermacetogenium phaeum, a syntrophic acetate-oxidizing bacterium that can also reduce sulfate (19). We cannot eliminate the possibility of trace production of H₂S because one sequence (clone B_11) showed distant relation to groups known to mediate complete sulfate reduction (e.g., Desulfovibrio spp.). However, we were unable to detect dissimilatory sulfite reductase (dsrA) genes across several methods and attempts, and the sulfur isotope data are inconsistent with measurable quantities of sulfide formation.

The presence of metabolically active cells requires a small supply of assimilated sulfur because sulfur composes ~0.1% of cell biomass (15). We estimated a doubling time for heterotrophs of ~300 days (Table 1) (10) equaling ~10⁶ generations over 1.5 My of isolation. If all cellular organic sulfur requirements were derived each generation by de novo assimilatory reduction, an upper limit of ~35% of the SO₄²⁻ pool would have been used. This result is consistent with our estimate of 30 to 40% SO₄²⁻ turnover but is unlikely to provide the full explanation for sulfur cycling in the brine. Initially the system would have included reduced organic nutrients (N, S, and P), including S-containing amino acids, in stoichiometric proportion to the initial supply of organic matter. Availability of this additional pool of biological metabolites could decrease the effective assimilatory SO₄²⁻ demand substantially. Additionally, the presence of diverse sulfur-cycling microbes (7) and several groups of APS genes indicate the presence of metabolic processes beyond assimilation strictly for biomass.

How then is the additional sulfate cycled? The unchanged values of δ¹⁸OSO₄ indicate that there is insignificant loss of sulfur to sedimentary pyrite. This implies that the reduction of SO₄²⁻ to

**Table 1.** Biogeochemical parameters of Blood Falls outflow during a brine discharge event. Outflow samples collected in December 2004. nd indicates none detected. The first six parameters and sulfate and chloride concentrations are from (33); total iron is from (7).

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Temperature</td>
<td>-5.2°C</td>
</tr>
<tr>
<td>pH</td>
<td>6.2</td>
</tr>
<tr>
<td>Dissolved oxygen</td>
<td>nd</td>
</tr>
<tr>
<td>DIC</td>
<td>55 mM</td>
</tr>
<tr>
<td>Dissolved organic carbon (DOC)</td>
<td>420 μM</td>
</tr>
<tr>
<td>Dissolved inorganic nitrogen (DIN)</td>
<td>94 μM (100% as NH₄⁺)</td>
</tr>
<tr>
<td>Total iron</td>
<td>3.45 mM (&gt;97% as Fe(II))</td>
</tr>
<tr>
<td>Sulfate</td>
<td>50 mM</td>
</tr>
<tr>
<td>Chloride</td>
<td>1375 mM</td>
</tr>
<tr>
<td>Total cells</td>
<td>6 × 10⁴ ml⁻¹</td>
</tr>
<tr>
<td>Thymidine (Tdr) incorporation rate</td>
<td>1.9 × 10⁻⁴ nM Tdr day⁻¹ (±2 × 10⁻⁵)</td>
</tr>
<tr>
<td>Δ¹⁵N_Cic</td>
<td>-93 ± 1‰</td>
</tr>
<tr>
<td>Δ³⁴S_Sulfate</td>
<td>21.0 ± 0.4‰</td>
</tr>
<tr>
<td>Δ³³S_Sulfate</td>
<td>0.08‰ (range = 0.06 to 0.09‰)</td>
</tr>
<tr>
<td>Δ¹⁸O_Sulfate</td>
<td>3.3 ± 0.8‰</td>
</tr>
<tr>
<td>Δ¹⁸O_H₂O</td>
<td>-39.5 ± 0.1‰</td>
</tr>
<tr>
<td>Δ⁶Fe</td>
<td>-2.60 ± 0.5‰</td>
</tr>
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SO$_3^{2-}$ does not proceed all the way to H$_2$S, which is consistent with the observation of ferruginous and not euxinic conditions (no detectable H$_2$S). To achieve these isotopic values, the net metabolism in the brine requires two remarkable properties: (i) SO$_4^{2-}$ must recycle through sulfur intermediates, and (ii) these sulfur intermediates must be quantitatively reoxidized to SO$_3^{2-}$.

We advocate that sulfur is catalytically cycled to facilitate the oxidation of organic matter in a system in which Fe(III) is the terminal acceptor. Both assimilatory and dissimilatory reduction of SO$_4^{2-}$ proceed enzymatically through intermediate species (15). To date, no cultured dissimilatory organism has been described in which SO$_4^{2-}$ reduction does not ultimately terminate in H$_2$S. The initial steps of SO$_4^{2-}$→SO$_3^{2-}$→S$_2$O$_3^{2-}$ are endergonic, but progression to SO$_4^{2-}$ when coupled to common electron donors [e.g., acetate and lactate (10)] does yield free energy (table S2). Coupling of any of the subsequent reoxidation reactions to the reduction of Fe(III) would also yield free energy (table S2). Additionally, numerous examples suggest that microbes use carrier molecules, including sulfur compounds, as electron shuttles between cells and iron oxides (20–22). Cycling sulfur as an electron shuttle to catalyze iron reduction would also be a feasible route to reduced sulfur species in this system.

Such a catalytic cycle would preserve the total dissolved sulfur concentration (Fig. 1, inset) while progressively accumulating Fe(II) to a level controlled by the solubility product constant of siderite. Although the mechanism by which recycling of sulfur species mediates Fe(III) reduction is not fully understood (23), in contemporary marine pore waters at depths below O$_2$ penetration values of $\delta^{34}S$SO$_4$ show SO$_3^{2-}$ regeneration perhaps in association with Fe and Mn oxides (24). The net thermodynamics of such a system are favorable and could be described as syntrophic.

Ferric iron likely is mobilized by the scouring action of the glacier over the basement complex of metamorphic rocks intruded by granodioritic sheets of the Taylor Glacier (69). The action of the glacier over the basement complex must be quantitatively reoxidized to SO$_4^{2-}$ to reconstitute the oxidized reservoir because it can only lose oxygen to SO$_3^{2-}$ (30–31). During periods of diminished net photosynthesis, such as Neoproterozoic Snowball Earth episodes (30, 31), a decrease in organic flux from reduced photosynthetic production would drive the ocean away from SO$_4^{2-}$ reduction, analogous to the Blood Falls system. When organic matter became sufficiently limited, euxinia would cease and Fe(II) would accumulate. This respiratory source of Fe(II) would add to the flux of hydrothermal Fe to the deep ocean (13), and both could lead to an episodically ferruginous ocean (32). Importantly, our model is independent of the size of the marine SO$_3^{2-}$ reservoir because it depends only on the ratio of Fe(III) to labile organic matter. The brine below the Taylor Glacier provides a contemporary, natural example of an active, catalytic sulfur cycle, and it uniquely allows for the study of the long-term persistence of life and associated bioenergetics under ice.

References and Notes

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10. Material and methods are available as supporting material on Science Online.

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Recursive Processes in Self-Affirmation: Intervening to Close the Minority Achievement Gap

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A 2-year follow-up of a randomized field experiment previously reported in Science is presented. A subtle intervention to lessen minority students’ psychological threat related to being negatively stereotyped in school was tested in an experiment conducted three times with three independent cohorts (N = 133, 149, and 134). The intervention, a series of brief but structured writing assignments focusing students on a self-affirming value, reduced the racial achievement gap. Over 2 years, the grade point average (GPA) of African Americans was, on average, raised by 0.24 grade points. Low-achieving African Americans were particularly benefited. Their GPA improved, on average, 0.41 grade point average (GPA) of African Americans was, on average, raised by 0.24 grade points. Focusing students on a self-affirming value, reduced the racial achievement gap. Over 2 years, the GPA improved, on average, 0.41 grade points. Low-achieving African Americans were particularly benefited. Their GPA improved, on average, 0.41 grade points, and their rate of remediation or grade repetition was less (5% versus 18%). Additionally, treated students’ self-perceptions showed long-term benefits. Findings suggest that because initial psychological states and performance determine later outcomes by providing a baseline and initial trajectory for a recursive process, apparently small but early alterations in trajectory can have long-term effects. Implications for psychological theory and educational practice are discussed.

Whether and how psychological interventions produce lasting positive consequences are critical questions for scientists and policy-makers. This report presents evidence of how interventional, even brief or subtle, can produce lasting benefit when targeted at important psychological processes. It does so by focusing on the long-term impact of a psychological intervention designed to reduce the racial achievement gap through the lessening of academic underperformance.

The achievement gap between academically at-risk minority students and European American students has long concerned the educational community. In a society where economic success depends heavily on scholastic accomplishment, even partial remediation of this gap would be consequential. This is especially true for low-achieving students, given the societal, institutional, and personal costs of academic failure.

Research shows the importance of psychological factors in intellectual achievement. Situations where one could be judged or treated in light of a negative stereotype can be stressful and thus undermine performance. For African Americans in school, the concern that they or another African American could be seen as confirming a negative stereotype about their group’s intelligence can give rise to stress and depress performance.

Findings of two randomized field experiments addressing this psychological threat in the classroom were reported in Science. These tested a values-affirmation intervention. Beginning early in seventh grade, students reflected on an important personal value, such as relationships with friends and family or musical interests, in a series of structured writing assignments. Such self-affirmations reduce psychological threat and stress and can thus improve performance. The intervention should benefit students from groups subjected to threat pervasive enough to undermine their average performance—in this case, negatively stereotyped minority students. As predicted, relative both to a control group and to historical norms, one or two administrations of the intervention improved the fall-term grades of African Americans and lowered the psychological availability of the stereotype. European Americans were unaffected.

A 2-year follow-up is now reported. We assess whether the affirmation buffers minority students from the effects of psychological threat over the long term, leading to academic benefits beyond the short-term ones of a single academic term previously found. Generally, psychological processes and their consequences are examined for relatively brief periods, often in experimental studies lasting 30 min or an hour. By contrast, because the present study spans 2 years, its findings speak to how an apparently brief psychological intervention triggers processes that affect performance and psychological outcomes over considerable periods of time. Given the multitude of factors that could mute the effects of such processes in the classroom, the findings address the longevity and real-world significance of these processes. This is particularly important given that the effects of interventions and psychological manipulations often decay and may even reverse over time for reasons that are little understood.

Because chronic evaluation is a key aspect of school and work environments, performance in these settings can be self-reinforcing. A recursive cycle, where psychological threat lowers performance, increasing threat and lowering performance further, in a repeating process, can magnify early performance differences among students. Early outcomes set the starting point and initial trajectory of a recursive cycle and so can have disproportional influence. For instance, the low self-confidence of students who experience early failure, even by chance, is surprisingly difficult to undo. A well-timed intervention could provide appreciable long-term performance benefits through early interruption of a recursive cycle.

Results encompass the original two student cohorts and a third cohort run after the original two experiments. The cohorts were observed for a period running from the first term of seventh grade to the end of eighth grade, typically covering ages 12 to 14. Although the period involves the last 2 years of middle school, for clarity these will henceforth be referred to as Year 1 and 2, respectively. Individual students were randomly assigned to the affirmation condition or the control condition. The former completed affirmation exercises, the latter neutral exercises. The treatment consisted of variations on the original affirmation exercise in which students wrote about the personal importance of a self-defining value. The control exercises consisted of variations on the original control exercise in which students wrote about an unimportant value or a similarly