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Researching nitrite oxidation at high temperatures

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N-nitrate (NO$_3^-$) pool dilution experiments show that ammonia (NH$_3$) is oxidized to nitrate in geothermal springs up to at least 85°C; however, nitrite (NO$_2^-$)-oxidizing microorganisms are only known to grow up to 66°C. We hypothesize that thermophilic microorganisms oxidize nitrite to nitrate at high temperatures. Alternatively, it is possible that nitrite is oxidized abiotically. We propose to test these hypotheses by setting up microbial enrichments designed to grow thermophilic nitrite oxidizing bacteria by varying incubation temperature (50, 65, 80°C), oxygen concentration (20% and 5%), and cultivation media. A negative control consisting of filtered spring water (0.1 µm) will be used to determine whether nitrite is oxidized abiotically. Enrichments will be monitored for nitrite oxidation activity by using colorimetric assays for nitrite and nitrate. Enrichments showing activity will be used as a source to try to isolate and/or identify responsible microorganisms and to study the kinetics of nitrite oxidation at high temperature.
Researching Nitrite Oxidation at High Temperatures

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Abstract
The role of nitrite-oxidizing organisms in the nitrogen cycle of soil and aquatic habitats is well established, however, it is not known whether these exist in soil or aquatic habitats. We successfully isolated nitrite-oxidizing bacteria from experimental soils using a new protocol and soil samples and found that the bacteria grow at high temperatures. The study was designed to test the hypothesis that the bacteria exist in soil or aquatic habitats. The bacteria were isolated from soil samples that had been exposed to high temperatures and were confirmed to be nitrite oxidizers. The study was designed to test the hypothesis that the bacteria exist in soil or aquatic habitats. The bacteria were isolated from soil samples that had been exposed to high temperatures and were confirmed to be nitrite oxidizers.

Introduction
Nitrite oxidation is a process that involves the conversion of nitrite to nitrate. This process is important in the nitrogen cycle, as it is the final step in the conversion of atmospheric nitrogen to a form that can be used by plants. Nitrite oxidation is also important in the treatment of wastewater, as it can be used to remove excess nitrogen from the water.

Soil Enrichment
A soil sample was collected from a field site. The soil was then incubated at high temperatures and the growth of nitrite-oxidizing bacteria was monitored. The growth was confirmed by the detection of nitrite and nitrate in the soil samples.

Low temperature control enrichments
Several cultures known to nitrite-oxidizing bacteria were prepared and inoculated with samples from low temperature habitats known to have active nitrite cycle. Primary enrichments inoculated with gaseous from a freshwater aquifer showed complete nitritation within 21 days. Secondary enrichments inoculated with nitrite completely within 22 days. The predominant morphotype from the cultures was a phosphobacillus sp. pure cultures sequence similar to the parent isolate. Isolates were pure cultures sequence similar to the parent isolate. Isolates were sequence similar to the parent isolate.

Enrichments
High temperature enrichments at GBS
Dissolved oxygen levels in soils and aquatic habitats were monitored with samples from the high temperature hot spring (Fig. 4). The samples were incubated with sodium nitrite, which is then oxidized to nitrate. The nitrate concentration was monitored, and the results were used to determine the rate of nitrite oxidation.

Discussion
All control samples demonstrated nitrite oxidation in the growth medium. The net amount of nitrite oxidized was measured by the decrease in nitrite concentration. This result is consistent with the hypothesis that nitrite oxidizers exist in soil or aquatic habitats. The study was designed to test the hypothesis that the bacteria exist in soil or aquatic habitats. The bacteria were isolated from soil samples that had been exposed to high temperatures and were confirmed to be nitrite oxidizers.

Future directions
If nitrite oxidation is detected:
- Determine the role of nitrite oxidation in the nitrogen cycle.
- Test the hypothesis that nitrite oxidizers exist in soil or aquatic habitats.
- Test the hypothesis that nitrite oxidizers exist in soil or aquatic habitats.

If nitrite oxidation is not detected:
- Test the hypothesis that nitrite oxidizers exist in soil or aquatic habitats.
- Test the hypothesis that nitrite oxidizers exist in soil or aquatic habitats.

References

Acknowledgements
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Tables
Table 1: Growth of nitrite-oxidizing bacteria at high temperatures

<table>
<thead>
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<th>Temperature (°C)</th>
<th>Growth of nitrite-oxidizing bacteria</th>
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<tr>
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<tr>
<td>70</td>
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<td>60</td>
<td>Yes</td>
</tr>
<tr>
<td>50</td>
<td>Yes</td>
</tr>
</tbody>
</table>

Figures
Figure 1: Nitrite-oxidizing bacteria isolated from soil samples.

Figure 2: Distributions of nitrite-oxidizing bacteria at high temperatures.

Figure 3: Distributions of nitrite-oxidizing bacteria at high temperatures.

Figure 4: Distributions of nitrite-oxidizing bacteria at high temperatures.

Figure 5: Distributions of nitrite-oxidizing bacteria at high temperatures.

Figure 6: Distributions of nitrite-oxidizing bacteria at high temperatures.