Biogeosciences Discuss., 3, S517–S520, 2006
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Biogeosciences Discussions



# *Interactive comment on* "Culturable bacteria in Himalayan ice in response to atmospheric circulation" *by* S. Zhang et al.

**S. Zhang et al.**

Received and published: 10 September 2006

We thank referees #3 for his constructive comments. As we have indicated in response to referee #1, our initial results are reasonable but somewhat immature. However, this might arouse a wide interest in both the glaciology and microbiology fields. Below we address the specific comments in the review:

Question 1:

It is also quite difficult to assess whether these isolates are really from the ice core or are contaminants. The authors did an admirable job of decontamination (quite thorough), but no description of any controls for contamination is included. Were any plates left open in the laminar flow hood while sampling or plating was performed? Were any blank plates (sterile water or media) done? Was any sampling done of surfaces that



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may have contacted the plates?

Response:

Please refer to response to referee #1.

Question 2:

A better description of the significance of the data in Figure 2 relative to the different seasons needs to be verified. Why does 18O enrichment imply winter deposition?

Response:

In the Himalayan region, precipitation is mainly caused by the Indian Summer Monsoon air mass containing abundant moisture from Indian Ocean. When the moisture moves from Tropic Ocean to high latitude, the heavy isotopic content losses continuously with precipitation on the way, and delta-Oxygen-18 value in precipitation decreases dramatically when the water vapor falls in the Himalayan region (Zhang et al., 2005). Therefore, delta-Oxygen-18 depletion occurs during the summer monsoon season, and delta-Oxygen-18 enrichment during the winter dry season (Marinoni et al., 2001; Thompson et al., 2000; Kang et al., 2000). This can be used as an indicator of seasonality of the glacial snow/ice layers.

Question 3:

Why were the web sites for the specific sequences provided instead of the Genbank accession numbers?

Response:

Some isolates from Genbank have both title and journal, in which case the exact references were given, e.g. AJ298940. But others have only title, in which case the web sites for the specific sequences were given, e.g. AY745834.

Question 4:



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Presumably, they plated multiple times, as the number of different isolates is significantly higher than the number of CFU per ml-so why not include some actual statistics so that we can assess the significance of the results?

Response:

Samples of melt water (200ml) from the ice cores were filtered through polycarbonate filters to collect a 2.0ml sample in phosphate-buffered saline. Of this suspension, a total of 1.6 ml was used for inoculation of solidified medium R2A and PYGV, with 0.2ml suspension in each plate with duplicate repetition. Univariate Analysis of Variance factorial design 4 * 2 * 2 with 2 repetitions, with post hoc multiple comparisons between means conducted by Tamhane analysis was used at 0.05 significance level. Concentration of culturable bacteria recovered from Sample I was significantly higher than that from the other three samples ($p < 0.05$).

Question 5:

Is 15 isolates really different from 10?

Response:

Statistics of Chi-Square test suggests that the numbers of different ARDRA patterns reached statistical significance.

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Interactive comment on Biogeosciences Discuss., 3, 765, 2006.