

**Influence of aeolian activities on the distribution of microbial abundance in glacier ice**

Y. Chen et al.

# Influence of aeolian activities on the distribution of microbial abundance in glacier ice

Y. Chen<sup>1</sup>, X.-K. Li<sup>1</sup>, J. Si<sup>2</sup>, G.-J. Wu<sup>3</sup>, L.-D. Tian<sup>3</sup>, and S.-R. Xiang<sup>1,3</sup>

<sup>1</sup>School of Life Science, Lanzhou University, Lanzhou, Gansu 730000, China

<sup>2</sup>Instituten of Modern Physics, Chinese Academy of Sciences, Lanzhou 730000, China

<sup>3</sup>Key Laboratory of Tibetan Environment Changes and Land Surface Processes, Institute of Tibetan Plateau Research, Chinese Academy of Sciences, Beijing 100085, China

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Correspondence to: Y. Chen (chenyong@lzu.edu.cn)

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[Title Page](#)

[Abstract](#)

[Introduction](#)

[Conclusions](#)

[References](#)

[Tables](#)

[Figures](#)

[⏪](#)

[⏩](#)

[◀](#)

[▶](#)

[Back](#)

[Close](#)

[Full Screen / Esc](#)

[Printer-friendly Version](#)

[Interactive Discussion](#)

## Abstract

Microorganisms are continuously blown onto the glacier snow, and thus the glacial depth profiles provide excellent archives of microbial communities and climatic and environmental changes. However, it is uncertain about how aeolian processes that cause climatic changes control the distribution of microorganisms in the glacier ice. In the present study, microbial density, stable isotopic ratios,  $^{18}\text{O} / ^{16}\text{O}$  in the precipitation, and mineral particle concentrations along the glacial depth profiles were collected from ice cores from the Muztag Ata glacier and the Dundee ice cap. The ice core data showed that microbial abundance was often, but not always associated with high concentrations of particles. Results also revealed clear seasonal patterning with high microbial abundance occurring in both the cooling autumn and warming spring-summer seasons. Microbial comparisons among the neighbouring glaciers display a heterogeneous spatial pattern, with the highest microbial cell density in the glaciers lying adjacent to the central Asian deserts and lowest microbial density in the southwestern margin of the Tibetan Plateau. In conclusion, microbial data of the glaciers indicates the aeolian deposits of microorganisms in the glacier ice and that the spatial patterns of microorganisms are related to differences in sources of microbial flux and intensity of aeolian activities in the current regions. The results strongly support our hypothesis of aeolian activities being the main agents controlling microbial load in the glacier ice.

## 1 Introduction

Microorganisms are continuously blown onto the glacier snow, and thus the glacial depth profiles provide good archives of microbial communities during the course of global climatic and environmental processes. Despite the importance of microorganisms in our knowledge of global climate and environment changes, it is uncertain how aeolian processes, that causes climatic changes Basin (Wake et al., 1993; Davis et al., 2005), control the distribution of microorganisms in the glacier ice.

### Influence of aeolian activities on the distribution of microbial abundance in glacier ice

Y. Chen et al.

Title Page

Abstract

Introduction

Conclusions

References

Tables

Figures

◀

▶

◀

▶

Back

Close

Full Screen / Esc

Printer-friendly Version

Interactive Discussion



## Influence of aeolian activities on the distribution of microbial abundance in glacier ice

Y. Chen et al.

[Title Page](#)

[Abstract](#)

[Introduction](#)

[Conclusions](#)

[References](#)

[Tables](#)

[Figures](#)

[⏪](#)

[⏩](#)

[◀](#)

[▶](#)

[Back](#)

[Close](#)

[Full Screen / Esc](#)

[Printer-friendly Version](#)

[Interactive Discussion](#)

Recently, microbiological data have been collected from several ice cores extracted from the geographically different glaciers such as the Vostok Station ice core, Antarctica (Abyzov et al., 1998; Christner et al., 2006; Priscu et al., 2008), the Malan Glacier (Yao et al., 2006) and the Guoqu Glacier in the Mount Geladaindong on the central Tibetan Plateau (Yao et al., 2008). All results of the ice cores have showed a high abundance of microbial abundance corresponds with a high concentration of particles, which suggests a strong effect of aeolian activities on the influx of microorganisms in the glacier ice.

There is a big challenge, however, in developing a better understanding of how the global climatic and environmental changes, especially, the aeolian processes influence the distribution of microorganisms in the deep glacier ice. This process reportedly involves two main coupled mechanisms, aeolian deposition and post-deposition (Xiang et al., 2009). The aeolian deposition mechanism suggests that microbial loads by aerosol, dust and precipitation determine the distribution of microorganisms in the ice, while the post-deposition mechanism, through microbial metabolic activities in the surface snow cause the community shift of microorganisms in the glacial ice. The results have indicated that the microbial distribution is mainly determined by the aeolian deposition mechanism. Furthermore, the post-deposition processes act to strengthen the deposition patterns of microorganisms with the selection for glacial-tolerant species on the glacier's surface in warm seasons. However, the behavior of aeolian activities and its influences on the microbial distribution in the deep ice is unclear.

Previous investigations showed an important role of climatic changes on the microbial distribution along the glacial depth profiles. The Antarctica ice from the cold period from 140 to 180 kyr BP had a cell density of  $8 \times 10^3$  cells mL<sup>-1</sup>. This is greater than the maximum cell density observed of  $2 \times 10^3$  cells mL<sup>-1</sup> during the warm period from 118 to 140 kyr BP. Abyzov et al. (1998) found that increased cell concentrations was correlated with elevated dust loads in the ice core. The similar result was also reported in the 102 m long Malan ice core (Yao et al., 2006). This core contained two phases with high microbial density during two cold periods (AD 1130 to 1540

and AD 1700 to 1893) and two stages with low microbial density during two warm periods (AD 1540 to 1700 and AD 1893 to 1999), respectively. Both Vostok Station and Malan ice cores suggested that there was a high influx of microbes and particles in Antarctic and the Kekexili Region of the Tibetan Plateau during the extreme global cold period. However, other ice core data showed a positive effect of temperature on the distribution of microorganisms in ice. Those data were from the shallow ice cores from the Sofiskiy Glacier in the South Chuyskiy range of the Russian Altai (Uetake et al., 2006) and the Geladaindong ice core from the Guoqu Glacier at the summit of Tanggula Mountains (Yao et al., 2008). Despite the discrepancy in microbial abundance and temperature changes seen between the shallow and deep ice cores, the results showed a consistent association of high microbial abundance with the presence of dust layers (Abyzov et al., 1998; Yao et al., 2006, 2008). This suggests that aeolian activities are the main processes controlling the distribution of microorganisms in ice. Furthermore, it implies that the microbial profiles of ice cores could be considered a bio-indicator of past climatic and environmental changes. It can therefore be predicted that the microbial profiles of ice cores could reflect the differences of aeolian activities in the geographically different regions.

## 2 Study area, data collection and methodology

Data discussed in this study were collected from the Muztagata Glacier (38°17' N, 75°04' E), and the Dunde ice cap (38°06' N, 96°24' E). As shown in the Fig. 1, the Muztagata Glacier is located in the most western margin of the Tibetan Plateau where precipitation is mainly derived from air masses originating in the arid, and semi-arid regions, including deserts Sary-Ishykotrau, Muyun Kum, Kyzyl Kum and Kara Kum, Taklimakan and Gurbantunnut (Wake et al., 1990; Li et al., 2003). The Dunde ice cap is located in the northern margin of the Qaidam Basin, and in the Qilian mountain region on the northeastern Tibetan Plateau, where the winter precipitation results from the incursion of westerly depressions along the southern slopes of the Himalayas

### Influence of aeolian activities on the distribution of microbial abundance in glacier ice

Y. Chen et al.

Title Page

Abstract

Introduction

Conclusions

References

Tables

Figures



Back

Close

Full Screen / Esc

Printer-friendly Version

Interactive Discussion



(Murakami, 1987; Davis et al., 2005); while the summer precipitation is derived from the monsoon circulation from the Bay of Bengal to the central Himalaya, and further to the Qaidam Basin and large depressions in Taklamakan Desert and Daidam Basin (Davis et al., 2005; Dregne, 1968; Chen and Bowler, 1986).

The first ice core Muzt (37 m-long) was extracted at 7010 m a.s.l. from the Muztagata Glacier in the summer of 2003 (Tian et al., 2006). The Dunde ice core (9.5 m-long) was extracted at 5325 m a.s.l. from the Dunde ice cap summit in October 2002 (Wu et al., 2009). The visible stratigraphic features were recorded immediately after ice core drilling. All ice cores were returned frozen to the freezer room (air temperature between  $-18$  to  $-24$  °C) at the Key Laboratory of the Ice Core and Cold Regions Environment of the Chinese Academy of Sciences. The ice core sections were split lengthwise into four portions and stored in a refrigerated room at  $-18$  °C to  $-24$  °C.

All ice core sections were cut into 37–156 samples in intervals of 15–30 cm using a band saw within walk-in freezers ( $-18$  to  $-24$  °C). The ice core samples were decontaminated, and slowly melted at 4 °C by following the protocols previously described by Yao et al. (2006). The freshly melted water (10 mL) from the Muzt and Dunde ice cores was diluted 10 fold. 100  $\mu$ L of diluted sample was added to the known concentration of fluorescent-dyed bead solution Trucount (Becton Dickinson) mixture with cell sorting marker carboxyfluorescein diacetate (cFDA) and propidium iodide (PI). The cFDA and PI staining was separately prepared by following the method of Amor et al. (2002), except for the cell suspensions which were incubated for 15 min in the dark at the room temperature (25 °C) for cell staining. The live and total cell numbers in the melt-water were determined with a FACSCalibur flow cytometer (Becton Dickinson Immunocytometry Systems, San Jose, California, USA) by following manufacturer's instruction.

A 10 mL aliquot of melt-water from the Muzt and Dunde ice cores was used for analysis of the mineral particle. Total microparticle concentrations were measured by using a Coulter counter Multisizer3 (Beckman).

**BGD**

11, 14531–14549, 2014

**Influence of aeolian activities on the distribution of microbial abundance in glacier ice**

Y. Chen et al.

Title Page

Abstract

Introduction

Conclusions

References

Tables

Figures

◀

▶

◀

▶

Back

Close

Full Screen / Esc

Printer-friendly Version

Interactive Discussion



A 10 mL aliquot of melt-water from the Dundee ice core was used for analysis of the stable isotopic ratios,  $^{18}\text{O} / ^{16}\text{O}$  ( $\delta^{18}\text{O}$ ) in the precipitation. A Finnegan MAT-252 mass-spectrometer was used to determine  $\delta^{18}\text{O}$  values within  $\pm 0.5\%$ . The Muzt ice core dating and  $\delta^{18}\text{O}$  data were previously described by Tian et al. (2006).

## 3 Results

### 3.1 Changes in physical–chemical and biological records in the Muztagata ice cores

There was a strong influence of aeolian activities on the physical–chemical and biological records along the ice core extracted at 7010 m a.s.l. of the Muztagata Glacier (Fig. 2). There was an apparent seasonal temperature change indicated by the proxy value of the stable isotopic ratios,  $^{18}\text{O} / ^{16}\text{O}$  ( $\delta^{18}\text{O}$ ) with a low value in winter and high value in summer (Fig. 2b). The live cell density was greatly variable at a range from  $6.5 \times 10^2$  to  $2.1 \times 10^4$  cells  $\text{mL}^{-1}$  during 1964 to 2000 (Fig. 2a), and the total cell density varied from  $4.4 \times 10^4$  to  $8.7 \times 10^5$  cells  $\text{mL}^{-1}$  (Fig. 2c). Peaks of live cell density were present in both summer seasons and winter-spring seasons (Fig. 2a). Several live cell density peaks were formed during the summer seasons in 1969, 1971, 1973, 1979, 1983, 1988, 1990 and 1993 for a total of 8 events, B1 to B7 (open triangles in Fig. 2a), respectively. However, cell density peaks were found during the winter seasons (filled triangles in Fig. 2a). This ice core also had an increased cell density in the summer seasons in 1978, 1988 and 1993 (open triangles B1, B2, and B3 in Fig. 2c). The abundance of microbial cell density significantly correlated with the peaks of mineral particle concentrations and possessed a high  $R^2$  value of 0.7 (Fig. 3).

### 3.2 Changes in physical–chemical and biological records in the Dundee ice core

Dundee ice core data showed the strong aeolian effects on the physical–chemical and biological components in precipitation in the Dundee Glacier (Fig. 4). Oxygen isotope

BGD

11, 14531–14549, 2014

## Influence of aeolian activities on the distribution of microbial abundance in glacier ice

Y. Chen et al.

Title Page

Abstract

Introduction

Conclusions

References

Tables

Figures

◀

▶

◀

▶

Back

Close

Full Screen / Esc

Printer-friendly Version

Interactive Discussion



## Influence of aeolian activities on the distribution of microbial abundance in glacier ice

Y. Chen et al.

[Title Page](#)[Abstract](#)[Introduction](#)[Conclusions](#)[References](#)[Tables](#)[Figures](#)[⏪](#)[⏩](#)[◀](#)[▶](#)[Back](#)[Close](#)[Full Screen / Esc](#)[Printer-friendly Version](#)[Interactive Discussion](#)

ratios of the melt-water samples from the Dunde ice core showed an apparent seasonal temperature change with a low value of in winter and high value in summer ranging from  $-10.78\text{‰}$  to  $-8.24\text{‰}$  (temperature proxy  $^{18}\text{O}/^{16}\text{O}$ , Fig. 4c), while microbial cell density varied from  $1.2 \times 10^3$  to  $9.1 \times 10^4$  cells mL $^{-1}$  (Fig. 4b) and  $1.3 \times 10^5$  to  $1.9 \times 10^6$  cells mL $^{-1}$  (Fig. 4d) for live and total cell density, respectively. Throughout the ice core, only one significant peak of live cell density appeared in Autumn of 2002 (Fig. 4b). On the other hand, a few peaks of the total cell density appeared in spring and correlated with peaks in the oxygen isotope ratios. During autumn and summer, the peaks of the total cell density were associated with minimum values of the oxygen isotope ratio, as shown in Fig. 4d. Abundance of microbial cell density occurred both at the dirty ice layers and clean ice layers (Fig. 4d and e).

## 4 Discussion

Previous reports have documented a coincidence of microbial abundance with the presence of dense particles in the glacier ice, and temperature effects on the microbial distribution through the variations in global aeolian activities (Yao et al., 2006; Abyzov et al., 1998). Similar phenomena have also been found in the glacier depth profiles from the Dunde ice core and Muztagata ice cores. However, our present data from two ice cores reveals that microbial abundance appears in both dirty and clean ice layers and warm and cold seasons This reinforces the concept of aeolian loading of microorganisms on the glacier surface, and supports our hypothesis of aeolian processes strongly determining the structure of microbial-deposits in the glacial ice.

### 4.1 Dust deposition and microbial distribution along the glacial depth profiles

Ice core data from the Muztagata and Dunde glacier showed a frequent association of microbial abundance with high concentrations of particles (Figs. 3, 4b, d and e), which was consistent with previous data from the Antarctic Glacier (Abyzov et al.,

1998; Priscu et al., 2008) the Malan Glacier (Yao et al., 2006), and the Guoqu Glacier on the Tibetan Plateau (Yao et al., 2008). This indicates that wind carries both dust and microbes in the atmosphere to the glacial surface, suggesting a strong effect of dust deposition on the distribution of microorganisms in the ice. However, our present data also showed a high microbial cell density at some clean ice layers (dashed lines in Fig. 4d and e). This indicates that microbial loading onto the glacier surface does not always associate with the dust deposits or “dirty” wind, may transport with “clean” wind, implying the importance of wind as an agent for microbial transport on the glacial surface.

## 4.2 Changes in glacial microbial density at variable temperatures

There is one common feature that both ice cores share, as well as many differences. All isotopic data from the Muztag Ata and Dunde glaciers showed a positive relationship between the isotopic ratios and air temperature (Tian et al., 2003, 2006), and revealed clear seasonal change patterns both in the temperature proxy (Figs. 2b and 4c) and microbial cell density (Figs. 2a, c, and 4c, d). The main difference is the cell density response patterns to the temperature changes. Early results showed a high load of microbial abundance onto the glacier during the strong cold-windy periods (Abyzov et al., 1998; Yao et al., 2006). The data collected during the last few decades from the Muztag Ata and Dunde glaciers also showed an appearance of high cell density in cooling, autumn to winter seasons (filled triangles in Figs. 2a and 4c). However, the ice core data taken during 1965 to 2000 from the Muztaga Ata Glacier showed that high cell density frequently occurred in the warming spring-summer seasons (open triangles in Fig. 2a). This is consistent with another independent microbial investigation on the Muztaga Ata Glacier (Liu et al., 2013). Uetake et al. (2006) also found that high microbial abundance was present in the warming spring-summer seasons in the Sofiyskiy Glacier in the south Chuyskiy range of the Russian Altai, so did Price and Bay (2013). The positive relationship between microbial abundance and temperature was very evident in the Guoqu Glacier in the Geladaindong mountain regions (Yao et al.,

### Influence of aeolian activities on the distribution of microbial abundance in glacier ice

Y. Chen et al.

Title Page

Abstract

Introduction

Conclusions

References

Tables

Figures

◀

▶

◀

▶

Back

Close

Full Screen / Esc

Printer-friendly Version

Interactive Discussion





2008). High microbial abundance in warming seasons is possibly attributed to the high microbial load by wind and microbial growth in the processes of post-deposition (Xiang et al., 2009c). The occurrence of microbial abundance during the climatic fluctuation periods and driven by the strong wind action strengthens the importance of aeolian activities of microbial loading on the glacial surface.

### 4.3 Geographically driven microbial density differences

Under the same project guideline for microorganisms in glacier ice and the relation to climatic and environmental changes, microbiological data have been collected from the ice core depth profiles from the geographically different glaciers in western China over the last decade. The same methodological system has been used for the investigation on the labeled glaciers in Fig. 5, which makes it possible to explore the geographical features of microorganisms in the glacier ice across western China. Results display a heterogeneous spatial pattern of microbial abundance in the geographically different glaciers (Fig. 5). Among the investigated glaciers in western China, the highest microbial cell density was found in the Kuytun 51 and Dunde glaciers with  $10^6$  cells mL<sup>-1</sup>. The spatial microbial abundance pattern could be attributed to the differences in sources of microbial flux and intensity of aeolian activities in the current regions. Both Kuytun 51 and Dunde glaciers are located in arid regions. The Kuytun 51 Glacier is located in the middle of several Asian deserts. The climatic dryness and the sparse plant coverage in these regions are favorable for the intensive aeolian processes (Peltier, 1950; Thornbury, 1954), and microbial transportation onto the glacial surface from the neighboring human settlements, farmland, and pasture. The long-range transportation is also important for microbial influx onto the glacier surface (Prospero et al., 2005). The Kuytun 51 and Muztag Ata glaciers are located in the north-west regions, and receive their precipitation from the westerly circulation (Wake et al., 1990, 1993; Li et al., 2003). The south-eastern part of the Tibetan Plateau (e.g., Geladaindong, Zadang, Qiang Yong, Palong and Rongbuk glaciers) is influenced mostly by the monsoonal air masses (Murakami, 1987). The northern and middle part

## Influence of aeolian activities on the distribution of microbial abundance in glacier ice

Y. Chen et al.

Title Page

Abstract

Introduction

Conclusions

References

Tables

Figures

⏪

⏩

◀

▶

Back

Close

Full Screen / Esc

Printer-friendly Version

Interactive Discussion



## Influence of aeolian activities on the distribution of microbial abundance in glacier ice

Y. Chen et al.

Title Page

Abstract

Introduction

Conclusions

References

Tables

Figures

◀

▶

◀

▶

Back

Close

Full Screen / Esc

Printer-friendly Version

Interactive Discussion

of the Tibetan Plateau (e.g., Dunde, and Puruogangri) is affected by the monsoonal moisture during the summer, westerly depressions during the winter (Murakami, 1987; Davis et al., 2005), and numerous large depressions in Takalamakan Desert and Daidam Basin as well (Dregne, 1968; Chen and Bowler, 1986). The lowest microbial cell density occurred in the Rongbuk Glacier which contained  $9 \times 10^6$  cells  $\text{mL}^{-1}$  (Fig. 5). This could be attributed to the “clean” moisture influx during the entry of monsoonal air masses into the Tibetan Plateau, and less influences by the human settlements, farmland, and pasture in the central Tibetan plateau. Other factors including local wind action, climatic, and physical–chemical conditions that influence the growth rate of microorganisms in the snow or ice also cause the variations in the spatial distribution of microorganisms in the glacial ice. More microbial data will be helpful for our better understanding of microbial deposit in the glacial ice and the relation to the global climatic and environmental changes.

The microbial cell density in the mountainous glaciers of western China ranges from  $10^4$  to  $10^6$  cells  $\text{mL}^{-1}$  in maximum values (Fig. 6). This is higher than the Antarctic glaciers which contain cell density ranging from  $5 \times 10^3$  cells  $\text{mL}^{-1}$  at the South Pole (Carpenter et al., 2000) to  $8 \times 10^3$  cells  $\text{mL}^{-1}$  at the Vostok station (Abyzov et al., 1998) and in the Arctic glacier, Kongsvegen, with  $2 \times 10^5$  cells  $\text{mL}^{-1}$  (Amato et al., 2007). This is consistent with previous results (Christner et al., 2000; Zhang et al., 2007; Liu et al., 2009). The relatively high values of maximum cell density in the mountainous glacier are frequently associated with dust events dust origination centers (Figs. 2, 4 and 5; Xiang et al., 2009b; Yao et al., 2008). The high microbial influx from the neighboring human settlements, farmland, and pasture may be another important reason for the high microbial cell density in the mountainous glaciers. By contrast, the relatively low cell density in the polar glaciers may be indicative of a decrease in the microbial input due to the longer transport distance from non-polar environments.

## 5 Conclusions

Physical–chemical and microbiological data from three ice cores presented here show microbial abundance occurred during the rapidly changing temperature phases, and are frequently associated with high concentration of particles. This suggests that aeolian activities are the dominant factor in controlling microbial loading in the glacial ice. Spatial patterns of microbial cell density across the geographically different glaciers appeared to possess higher cell density in the glaciers lying adjacent to the central Asian deserts and lowest microbial density in both the south-western margin of the Tibetan Plateau (Rongbuk Glacier) and polar glaciers. This suggests the importance of microbial flux sources and aeolian activities in the current regions. The results strongly support our hypothesis of aeolian activities being the main agents controlling microbial loading in the glacial ice.

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### Influence of aeolian activities on the distribution of microbial abundance in glacier ice

Y. Chen et al.

Title Page

Abstract

Introduction

Conclusions

References

Tables

Figures

◀

▶

◀

▶

Back

Close

Full Screen / Esc

Printer-friendly Version

Interactive Discussion



## Influence of aeolian activities on the distribution of microbial abundance in glacier ice

Y. Chen et al.

[Title Page](#)

[Abstract](#)

[Introduction](#)

[Conclusions](#)

[References](#)

[Tables](#)

[Figures](#)

[⏪](#)

[⏩](#)

[◀](#)

[▶](#)

[Back](#)

[Close](#)

[Full Screen / Esc](#)

[Printer-friendly Version](#)

[Interactive Discussion](#)

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## Influence of aeolian activities on the distribution of microbial abundance in glacier ice

Y. Chen et al.

[Title Page](#)

[Abstract](#)

[Introduction](#)

[Conclusions](#)

[References](#)

[Tables](#)

[Figures](#)

[⏪](#)

[⏩](#)

[◀](#)

[▶](#)

[Back](#)

[Close](#)

[Full Screen / Esc](#)

[Printer-friendly Version](#)

[Interactive Discussion](#)

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## Influence of aeolian activities on the distribution of microbial abundance in glacier ice

Y. Chen et al.

[Title Page](#)

[Abstract](#)

[Introduction](#)

[Conclusions](#)

[References](#)

[Tables](#)

[Figures](#)

[⏪](#)

[⏩](#)

[◀](#)

[▶](#)

[Back](#)

[Close](#)

[Full Screen / Esc](#)

[Printer-friendly Version](#)

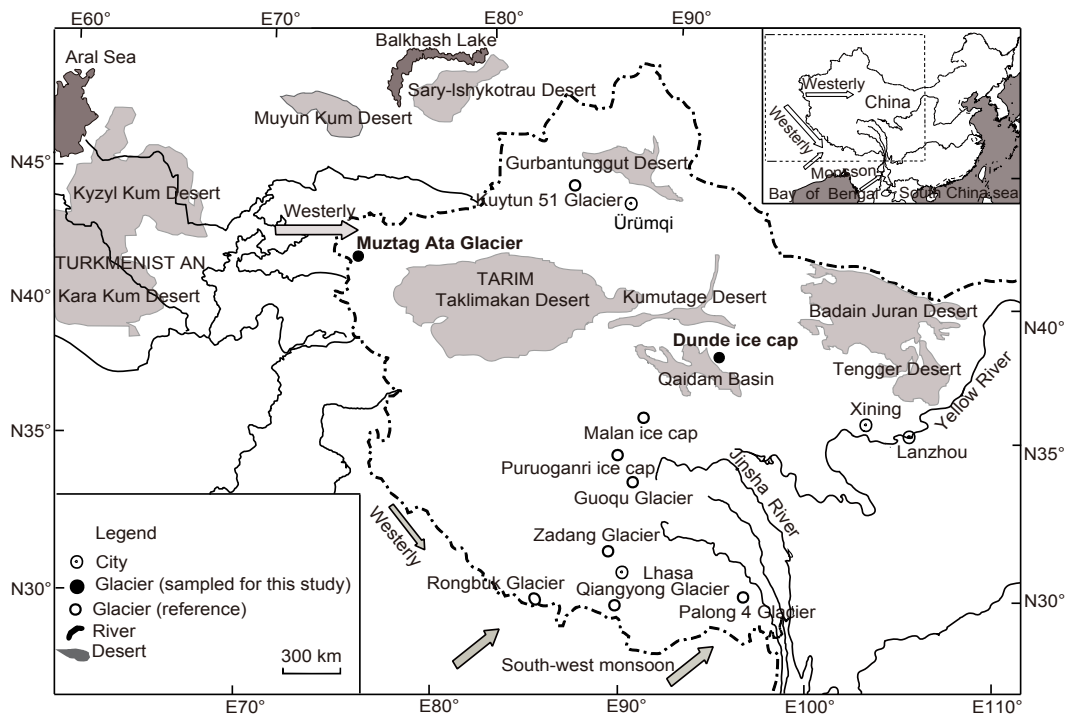
[Interactive Discussion](#)



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**Influence of aeolian activities on the distribution of microbial abundance in glacier ice**

Y. Chen et al.



**Figure 1.** Map illustrating the location of glaciers discussed in this study.

[Title Page](#)

[Abstract](#)

[Introduction](#)

[Conclusions](#)

[References](#)

[Tables](#)

[Figures](#)

◀

▶

◀

▶

[Back](#)

[Close](#)

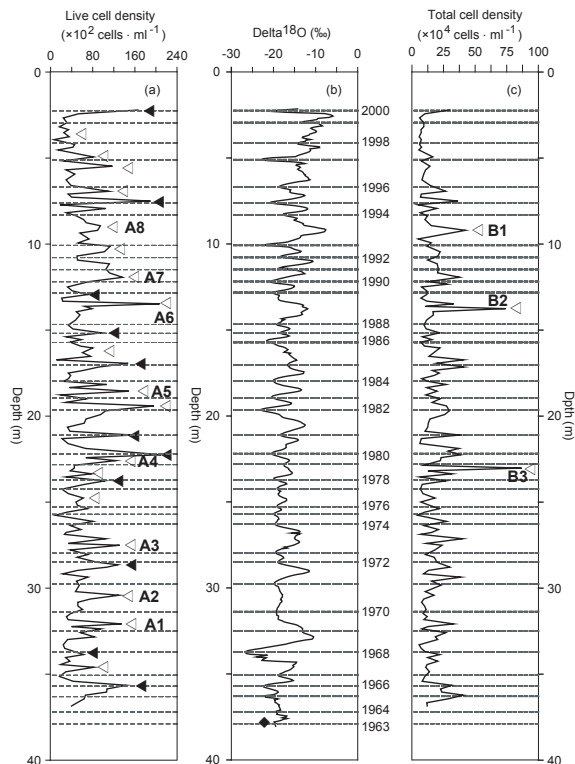
[Full Screen / Esc](#)

[Printer-friendly Version](#)

[Interactive Discussion](#)

## Influence of aeolian activities on the distribution of microbial abundance in glacier ice

Y. Chen et al.



**Figure 2.** Bacterial cell density, mineral particles and  $\delta^{18}\text{O}$  in the Muzt ice core. Muzt (43 m-long) was extracted at 7010 m a.s.l. from the Muztagata Glacier in the summer of 2003. **(a)** Live cell density in the ice. **(b)** The  $\delta^{18}\text{O}$  value was measured by Finnegan MAT-252 mass spectrometer (adapted from Tian et al., 2006). Ice core was annually dated by using seasonal  $\delta^{18}\text{O}$  variations and annual visible dust layers (Tian et al., 2006). **(c)** Total bacterial cell density estimated by using flow cytometer and cFDA/PI-stain, see the detailed in the materials and methods.

Title Page

Abstract

Introduction

Conclusions

References

Tables

Figures

◀

▶

◀

▶

Back

Close

Full Screen / Esc

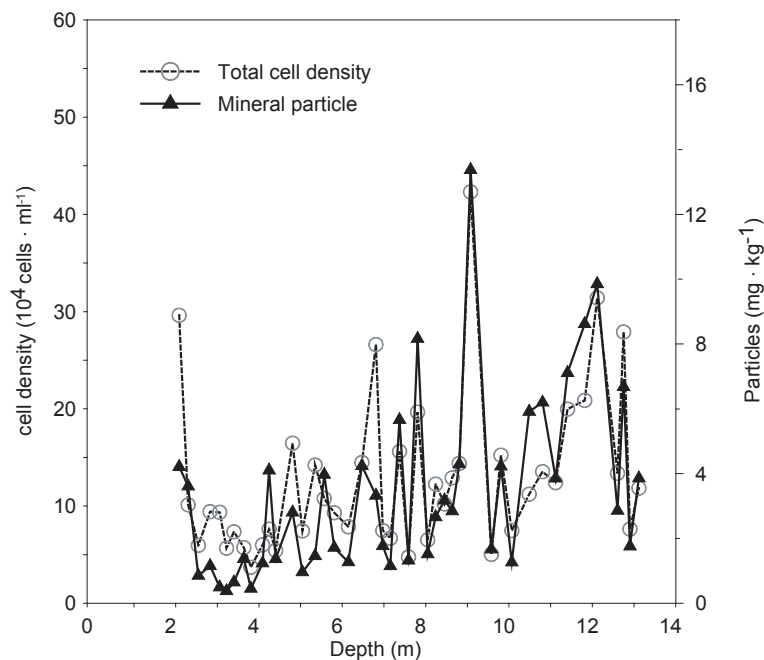
Printer-friendly Version

Interactive Discussion



## Influence of aeolian activities on the distribution of microbial abundance in glacier ice

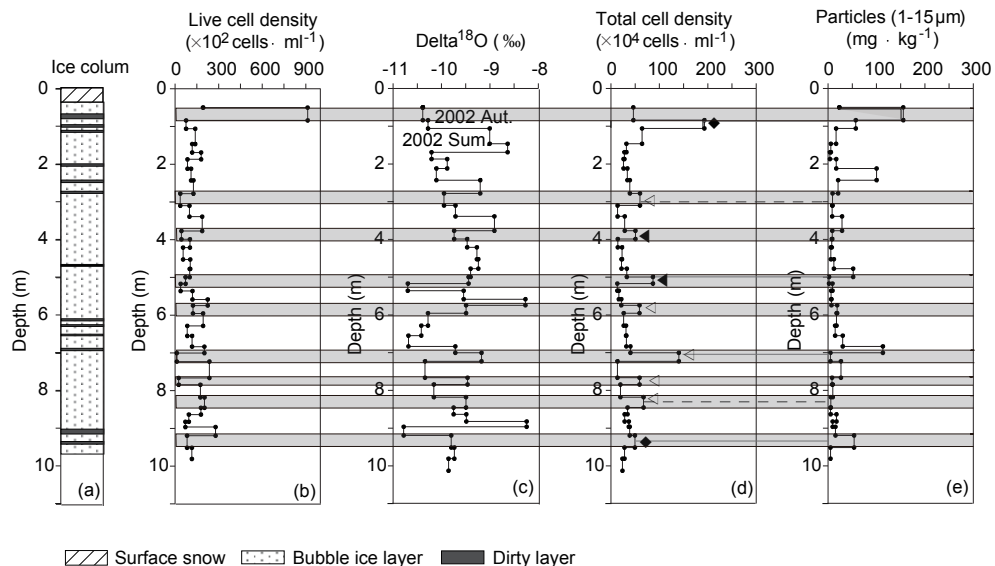
Y. Chen et al.



**Figure 3.** Correlation between mineral particle concentration and total cell density in the Muztag Ata Muzt ice core. Total microparticle concentrations were measured by using a Coulter counter Multisizer3 (Beckman).

## Influence of aeolian activities on the distribution of microbial abundance in glacier ice

Y. Chen et al.



**Figure 4.** Bacterial cell density, mineral particles and  $\delta^{18}\text{O}$  in the Dundee ice core. **(a)** Physical characteristics of ice layers along the depth profile. **(b)** Total live cell density. Total live cells were estimated by using flow cytometer and cFDA/PI-stain. **(c)**  $\delta^{18}\text{O}$  value. The  $\delta^{18}\text{O}$  value was measured by Finnegan MAT-252 gas stable isotope ratio mass-spectrometer. **(d)** Total bacterial cell density was estimated by using flow cytometer and cFDA/PI-stain. **(e)** Mineral particle concentration along the depth profile. Total microparticle concentrations were measured by using a Coulter counter Multisizer3 (Beckman).

Title Page

Abstract

Introduction

Conclusions

References

Tables

Figures

◀

▶

◀

▶

Back

Close

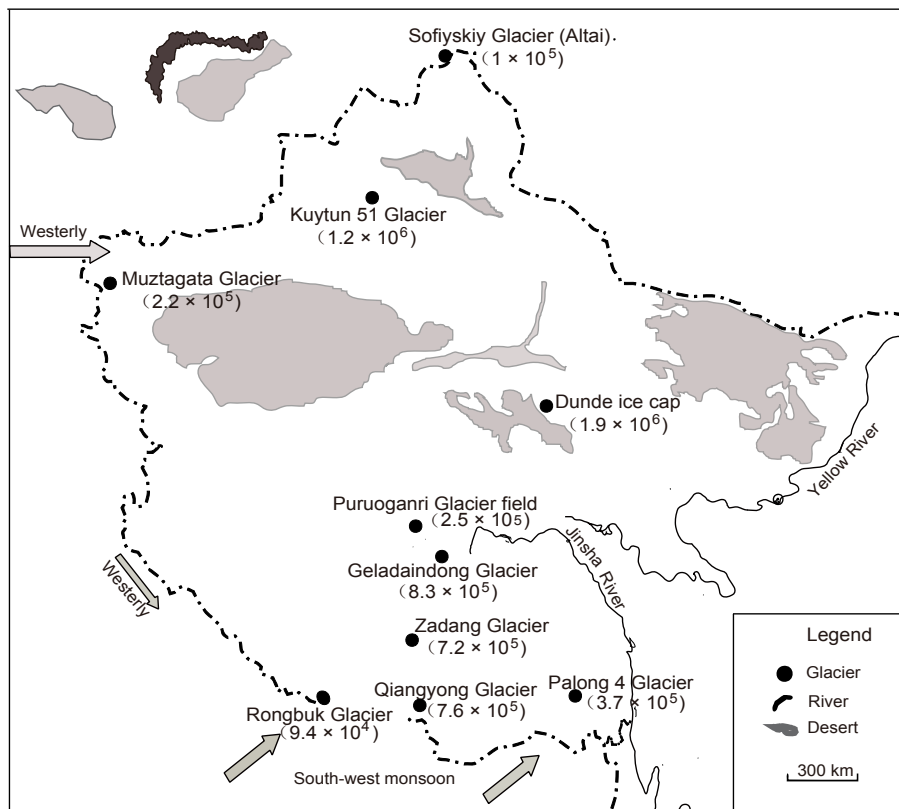
Full Screen / Esc

Printer-friendly Version

Interactive Discussion

## Influence of aeolian activities on the distribution of microbial abundance in glacier ice

Y. Chen et al.



**Figure 5.** Map showing the geographic orientation of climate zones and glaciers with associated microbial cell density (cells mL<sup>-1</sup>). Data was adapted from the glaciers Sofiyskiy (Altai, Uetake et al., 2006), Kuytun 51 (Xiang et al., 2009b), Muztag Ata (this study), Dunde (this study), Puruoganri (Zhang et al., 2006), Geladaidong (Yao et al., 2008), Zadang (Liu et al., 2009), Palong (Liu et al., 2009), Qiangyong (Xiang et al., 2010), Rongbuk (Liu et al., 2007).

[Title Page](#)
[Abstract](#)
[Introduction](#)
[Conclusions](#)
[References](#)
[Tables](#)
[Figures](#)
[Back](#)
[Close](#)
[Full Screen / Esc](#)
[Printer-friendly Version](#)
[Interactive Discussion](#)