Bulletin of Glaciological Research 27 (2009) *25–35*

Article

A snow algal community on the surface and in an ice core of Rikha-Samba Glacier in Western Nepali Himalayas

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(Received July 15, 2009; Revised manuscript accepted December 14, 2009)

Abstract

A snow algal community on Rikha-Samba Glacier in the western Nepali Himalayas was investigated in October, 1998. Examination of surface snow and ice on the glacier revealed that this community consisted of mainly seven taxa including green algae and cyanobacteria. The algal community showed an altitudinal distribution on the glacier: its biomass decreased with increasing altitude and community structure changed with altitude. Snow algae were also found in an ice core drilled at the top of the glacier. The ice core was 15 m deep and spanned 37 years (1962–98) based on the analyses of tritium, dust, and stratigraphy. Snow algae in the ice core included four taxa of snow algae, which were likely the same taxa observed on the glacial surface. The record showed that the biomass and species composition of the snow algae varied among layers in the ice core. Some layers contained algae that were observed only at a lower altitude of the glacier in the studied year. This implies that altitudinal distribution of a snow algal community could shift up or down on the glacial surface from year to year. Although the variation in this algal community in the ice core might be affected by spatial variation and diagenesis of algal cells in the glacial ice, the biomass increase in the 1990s and the variation in species composition are likely to reflect an algal community appearing on the glacial surface.

1. Introduction

Glaciers are inhabited by specialized organisms that have adapted to extremely cold environments, *e.g.,* snow algae, insects, ice worms, copepods, rotifers, tardigrada, and bacteria. These organisms spend their whole lives on snow and ice, and they are forming glacial ecosystems, which are simple in terms of their species diversity and sources of energy and nutrients (*e.g.,* Kohshima, 1987; Hoham and Duval, 2001). Recently, glacial changes, such as alterations in mass balance, snow chemistry, and/or dust concentration have been reported in many regions of the world (*e.g.,* Thompson *et al.,* 1993, 2000), which may affect a biological community on the glacial surface. However, little attention has been given to the total impact on glacier ecosystems. Also, the biological communities have a geophysically important effect: blooms of the microbes in snow and ice can reduce the surface albedo and then accelerate the melting of snow and ice (*e.g.,* Takeuchi *et al.,* 2001; Kohshima *et al.,* 1993). Although the degree of glacial shrinkage is now a matter of concern in response to recent climate change, such shrinkage also may possibly be accelerated from an increase in biological productivity. Thus, the temporal variations in that activity on glaciers are of great interest to the study not only of glacial biology but also of glacial fluctuations.

Ice core studies could provide a means to reconstruct the temporal variations in biological activity on the glacial surface. Spores of microbes are usually deposited on the glacial surface along with windblown dust in the dry winter season. The microbes photosynthetically grow later on the wet glacial surface during the summer thawing season, and then they are finally frozen and buried in new snow in the late summer. They can be preserved every year in the glacial ice as long as the surface is located in an accumulation area. Thus, an ice core should provide

a continuous record of microbes deposited and grown on the glacial surface (*e.g.,* Xiang *et al.,* 2005; Yoshimura *et al.,* 2000, 2006; Willerslev *et al.,* 1999). Furthermore, since growth of the organisms depends on physical or chemical conditions on the glacial surface, the biogenic material in ice cores can be expected to constitute a new indicator for interpreting past environments. Biological analysis of ice cores from areas of heavy melt could be particularly useful since biogenic materials appear to be less affected by meltwater percolation due to their larger size, even when stable isotopes or chemical signals are reduced or eliminated (Yoshimura *et al.,* 2000).

In this paper, we aim to describe the spatial and temporal variations in the snow algal community on the Rikha-Samba Glacier in the western Nepali Himalayas and to demonstrate possible factors affecting algal biomass and community structure on the glacier. The altitudinal distribution of a snow algal community was analyzed qualitatively. The temporal variation in snow algal community for the last several decades was reconstructed from a shallow ice core drilled at the top of the glacier. These variations were discussed with physical and chemical conditions on the glacier.

2. Study site and methods

2.1 Rikha-Samba Glacier

The research was carried out in October 1998 on the Rikha-Samba Glacier in the Himalayas. The glacier is located near Mt. Dhaulagiri (8167 m a.s.l.), which is a summit renowned for mountaineering in western Nepal (Fig. 1). The glacier flows southward from a snowy pass (5900 m a.s.l.) down to the terminus (5300 m a.s.l.). The equilibrium line was located at 5800 m a.s.l., which is based on the mass balance surveys conducted in 1998 and 1999 (Fujita *et al.,* 2001a). The length and area of the glacier are approximately 6.0 km and 4.8 km^2, respectively. The recent glaciological studies on this glacier have shown a significant retreat over the last 40 years (*e.g.,* Fujita *et al.,* 2001a).

2.2 Examination of the snow algal community on the glacier

To describe a snow algal community on this glacier, surface snow and ice were examined in accordance with previous studies on glaciers (*e.g.*, Yoshimura *et al.,* 1997). Collection of surface snow/ice including snow algae was carried out at the seven sites in the area between 5350 and 5880 m a.s.l. (S1-S7, Fig. 1). In the study period, the entire glacial surfaces were covered with new snow, which likely fell in the month prior to this study. The depth of new snow increased with elevation; it was 1 cm at site S1 and 50 cm at site S7. The sampling was done on the summer glacial surface below the new snow, on which snow algae usually grow. The condition of the summer surface was ice at sites S1-S5 and snow at sites S5-S7. Four samples were collected from randomly selected surface at each study site with a stainless-steel scoop (cylindrical shape, 66 mm in diameter × 215 mm length). The collected surface ice/snow was approximately 5 × 5 cm in area and 1–2 cm in depth. After the collection, the exact area was measured with a ruler to calculate the amount of the algal volume biomass per unit area. The samples were melted and preserved as a 3% formalin solution in clean 30mL polyethylene bottles within 3 hours at the base camp (located at the front of the glacial terminus). These samples were used for cell counting. Although the fine structure of the algae in the samples might be lost by formalin, the algae could still be counted with a microscope. For identification of algal taxa, other samples were collected at each study site and preserved with 5% glutaraldehyde solution. The identification of the taxa was done based on mainly Yoshimura *et al.* (1997), which is a report on snow algae on a Himalayan glacier. All samples were transported to the biological laboratory of Tokyo Institute of Technology in Japan for analysis. The algal biomass of each site was represented by the cell number per unit water volume and algal volume per unit area. Cell counts and estimations of cell volume were conducted with an optical microscope (Nikon E600). The samples were stained with 0.5% erythrosine (0.1 mL was added to 3 mL of the sample) and ultrasonicated for 5 minutes to loosen sedimented particles. The sample water was filtered through a hydrophilized PTFE membrane filter (pore size 0.5 μm, 13 mm diameter, Advantec, H020A013A), which became transparent with water, and the number of algae on the filter was estimated by counting the cells along a line crossing the filter center (counted area/entire filter area = 4.9 mm^2/78 mm^2). The volume of filtered water was adjusted between 50 and 1000 μL to obtain an optimal cell number for the counting (50–1000 cells in the total of taxa). The filtering and counting were repeated 3–6 times on the same sample until the standard error of the mean of this measurement became less than 10%. From the mean of the repeated measurements and the filtered volume, the cell concentration (cells mL^{-1}) of the sample was obtained. Mean cell volume was estimated by the nearest geometrical approximations using measurements of 50–100 cells for each taxon (Reynolds, 1984). The total algal biomass was estimated by summing algal volume biomass of each taxon obtained by multiplying cell concentration by the mean cell volume. This calculation was done for each taxon at each site. The total biomass was represented as cell volume per unit area of glacial surface ($\mu m^3\ m^{-2}$). Community structure was represented by the proportion of each taxon to the total algal volume.

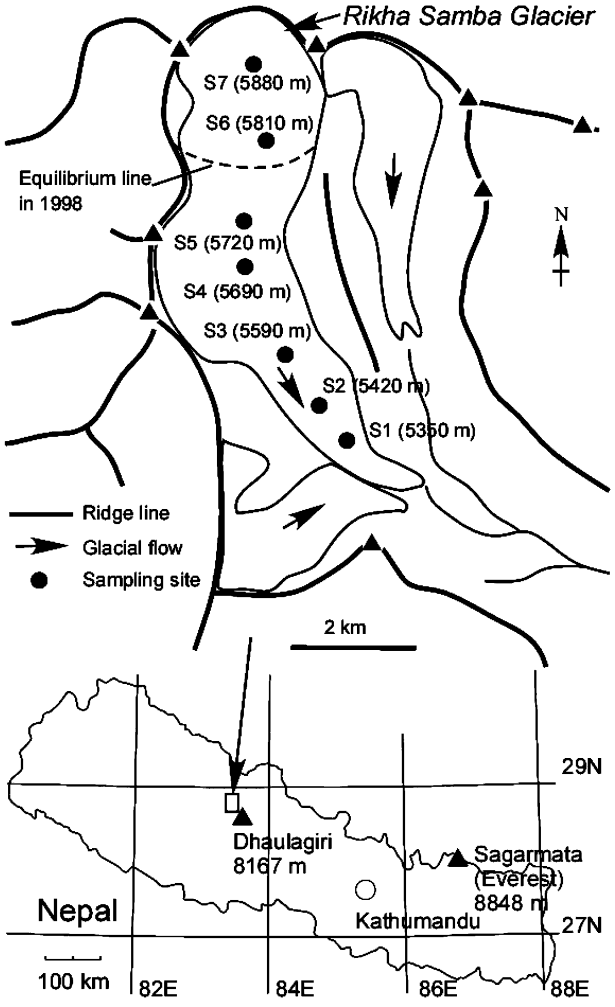

Fig. 1. Maps of Rikha-Samba Glacier in western Nepal showing sites of surface snow/ice sample collection and ice core drilling. Ice core drilling was carried out at site S7.

2.3 Ice core drilling and analysis

The ice core was drilled on a flat glacial surface at an elevation of 5880 m a.s.l. above the equilibrium line (site S7 in Fig. 1). The site was located in a superimposed ice zone, where percolated melt water continuously refreezes at the interface between snow layer and glacier ice. Superimposed ice appeared at 56 cm below the surface at the drilling site. A 14.90-m long ice core was recovered with a hand auger (Pico, U.S.A.). The drilling conditions have been described by Fujita *et al.* (2002). All of the ice core sections were vertically split in half in the field. One half of the core was horizontally cut every 5 cm (total 296 samples), melted in a clean plastic bag, and preserved in clean plastic bottles in the field, and then transported to a laboratory in Nagoya University, Japan. The other half was kept frozen and transported to a cold laboratory in the university. All of the processes in the field were conducted on a clean aluminum foil spread on a box with a disposal plastic glove to avoid contamination. Also, the surface of each piece of the ice was cleaned with a ceramic knife before the ice was put in the plastic bag.

The samples cut in the field were used for the analysis of tritium, snow algae, and dust concentration. The samples for analysis of snow algae were preserved as a 3% formalin solution in the field. The frozen core was used for a visual stratigraphy record and density measurement in the cold laboratory. After the measurements, the core was cut every 5 cm (total 296 samples) and used for hydrogen stable isotope (δD) and soluble chemical ions analyses. Tritium concentration was measured with a liquid scintillation counter. Hydrogen stable isotope was analyzed via gas-source mass spectrometry using chrome reduction (precision: $\pm 0.5‰$) in the University of Maine. The soluble chemical ions, including five cations (Na^+, NH_4^+, K^+, Mg^{2+}, and Ca^{2+}) and seven anions (CH_3COO^-, $HCOO^-$, Cl^-, NO_2^-, NO_3^-, SO_4^{2-}, and $C_2O_4^-$), were measured by an ion chromatography systems (Dionex DX500). In this paper, the results of NH_4^+, NO_3^-, Ca^{2+}, and $C_2O_4^-$ are shown.

The algal biomass in the core samples was represented by the cell volume per unit volume of water. Cell counts and estimations of cell volume were conducted with a florescence microscope (Nikon E600). 200 μL of the sample water was filtered with a hydrophilized PTFE membrane filter (the same as filter used for surface samples), and the number of autofluorescent algae on the filter was counted by the same manner as the algal examination for the glacial surface samples. From the mean results and filtered sample water, the cell concentration (cells mL^{-1}) of each sample was obtained. The theoretical detection limit of this analysis was 0.01×10^5 μm^3 mL^{-1}.

Dust concentration was also quantified by microscopy using the same filter. The surface of the filter observed microscopically was stored as a digital image file (TIFF format) on an 8-bit grey scale. After the image was converted to black and white with a threshold of the maximum level of background, the number of particles was automatically counted with an image-processing application (Image Pro Plus, ver. 4). Size of the dust particles by this method ranged from 5 to 200 μm in diameter. The dust concentration was obtained from the number of particles and volume of filtered water. The detection limit and error range in this procedure are 5.0×10 number mL^{-1} and 10%, respectively. Our method may underestimate the dust concentration because the image processing procedure could erase pale-colored dust particles and could not separate overlapped dust particles on the filters. The error was estimated as roughly -5%, based on comparison of the results from this method and direct counting with a microscope. However, the threshold level does not affect the relative variation in dust concentration of each sample.

3. Results

3.1 Snow algal community on the glacial surface

The snow algal community on the glacial surface consisted of mainly seven taxa (Fig. 2). Descriptions of the seven taxa are shown in Table 1.

Each taxon showed a different pattern of spatial distribution (Fig. 3). *C. brebissonii,* Nostocaceae cyanobacterium, and Oscillatriaceae cyanobacterium 2 were observed mainly in the lower part of the glacier (sites S1-S3). *M. berggrenii,* Chroococcaceae cyanobacterium, and Oscillatriaceae cyanobacterium 1 were observed on all of the sites from S1 to S7. The Unknown Alga was observed in mainly higher altitudes (sites S5-S7).

The total algal biomass generally decreased with increasing altitude (Fig. 4). The mean total algal biomass ranged from 4.7×10^{9} to $4.5\times10^{11}\mu m^{3}\ m^{-2}$ (mean: $1.3\times10^{11}\mu m^{3}\ m^{-2}$). The largest biomass ($4.5\times10^{11}\mu m^{3}\ m^{-2}$) occurred at site S2. The biomass generally decreased with altitude to sites in the snow area. The smallest biomass was found at site S6.

The community structure showed that dominant algae differed among the study sites (Fig. 5). In lower sites of the glacier, Nostocaceae cyanobacterium, Oscillatriaceae cyanobacterium 2 and *C. brébissonii* were dominant. Total biomass of the three taxa occupied from 70 to 80% of the total at the lower three sites (S

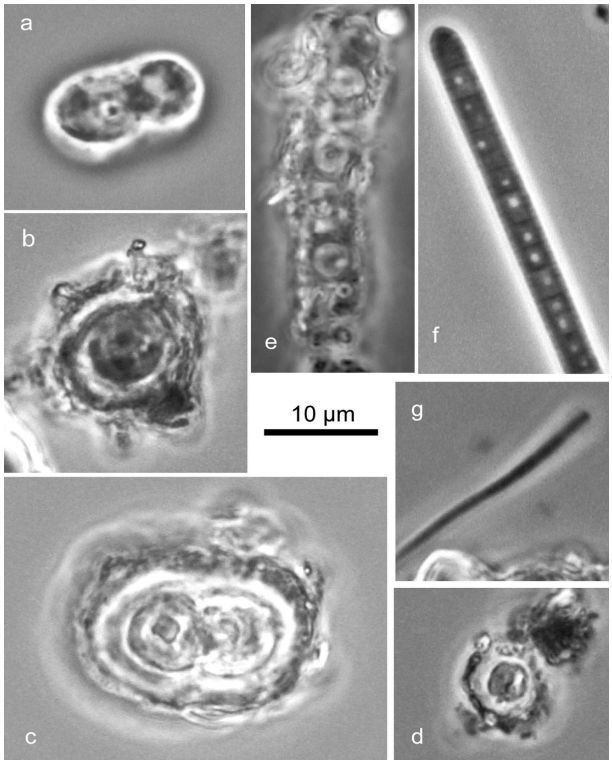

Fig. 2. Pictures of snow algae observed on Rikha-Samba Glacier. a. *Mesotaenium bregrenii*; b. Unknown Alga; c. *Cylindrocystis brébissonii*; d. Chroococcaceae cyanobacterium; e. Nostocaceae cyanobacterium; f. Oscillatoriaceae cyanobacterium 1; g. Oscillatoriaceae cyanobacterium 2. All pictures were taken with a phase contrast microscope.

Table 1. List of snow algae observed on Rikha-Samba Glacier and their description.

Taxa	Photo	Size and Morphology
Chlorophyta (Green algae)		
Mesotaenium berggrenii var. *alaskana* (Wittrock) Lagerheim	2a	Cells single or paired, Cylindrical with rounded apices. One to two chloroplasts, with one pyrenoid. 13.4 ± 3.0 μm in length, 9.0 ± 0.5 μm in width. Cell sap dark brown.
Cylindrocystis brébissonii (Ralfs) De Bary f. cryophila Kol	2c	Cells cylindric with rounded apices. Axial choloroplast usually two, with one pyrenoid. Cells 34.9 ± 6.1 μm in length, 23.8 ± 4.3 in width
Unknown Alga	2b	Cells round, a chloroplast without pyrenoid. Round cells 12.4 ± 5.7 μm (mean ± SD). This alga could be *Trochiscia* sp. or zygospore of *Chloromonas* sp., reported to be common species on snow surface of Himalayan glaciers.
Cyanobacteria (blue-green algae)		
Chroococcaceae cyanobacterium	2d	Cell spherical, single or paired, 4.8 ± 1.4 μm in diameter with mucilaginous envelopes.
Nostocaceae cyanobacterium	2e	Trichomes spherical, 5.7 ± 0.7 μm in width, 4.3 ± 1.2 μm in length, with mucilaginous sheath.
Oscillatriaceae cyanobacterium 1	2g	Trichomes 1.5 ± 0.28 μm in width. Cells about 1.5 times longer than width.
Oscillatriaceae cyanobacterium 2	2f	Trichomes 3.7 ± 0.4 μm in width, 2.7 ± 0.92 μm in length without sheath

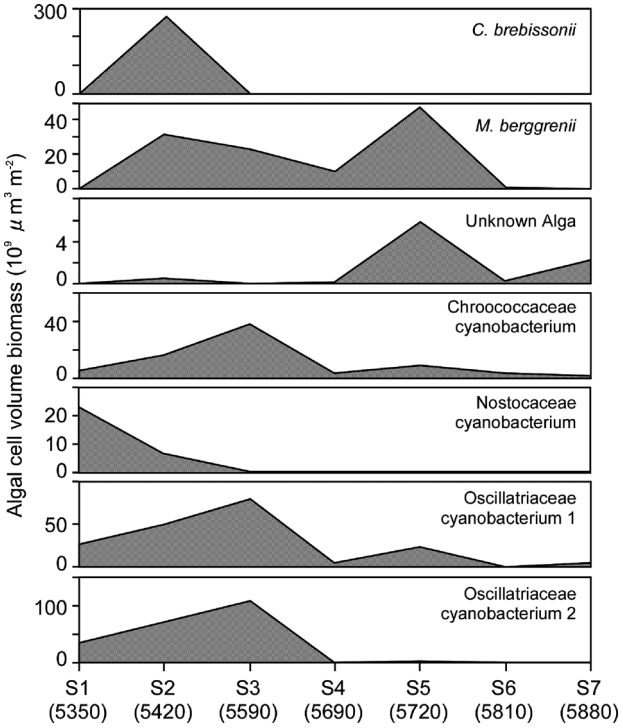

Fig. 3. Altitudinal distribution of cell volume biomass ($10^9 \mu m^3\ m^{-2}$) of each snow alga on Rikha-Samba Glacier. Numbers in brackets show altitude of each site (m a.s.l.).

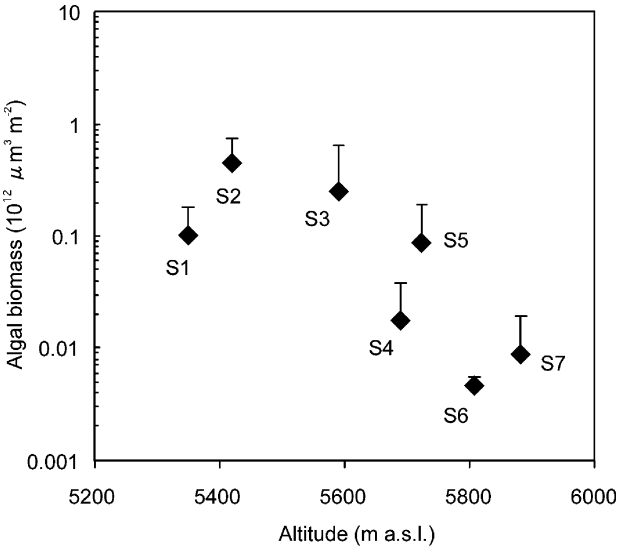

Fig. 4. Altitudinal change of total cell volume biomass on Rikha-Samba Glacier.

1-S3). *C. brébissonii* were particularly dominant at site S2 (more than 60% of total biomass). *M. berggrenii* was dominant in the middle area of the glacier (S4-S5), accounting for 50–60% of the total cell volume biomass at the sites. Chroococcaceae cyanobacterium and Oscillatriaceae cyanobacterium 1 were the second and third most dominant in the middle area, respectively. In the upper area (S6-S7), Chroococcaceae cyanobacterium and Oscillatriaceae cyanobacterium 1 were dominant, accounting for 70–80% of the total

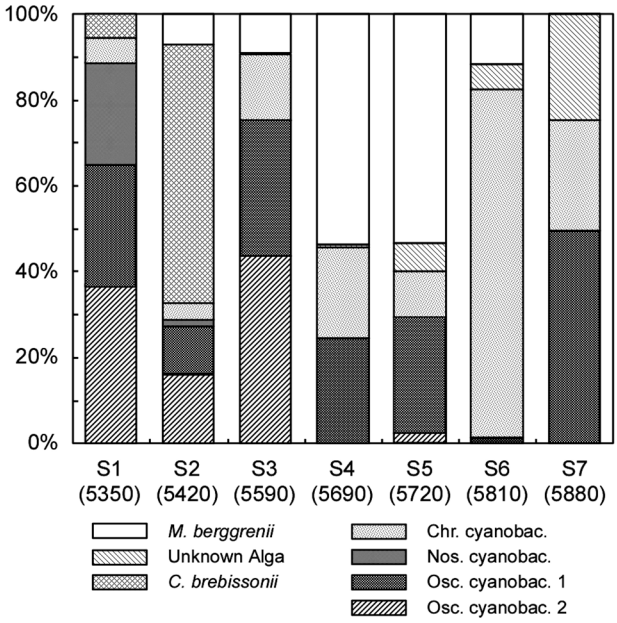

Fig. 5. Altitudinal change of community structure of snow algae (proportion of cell volume biomass) on Rikha-Samba Glacier. Numbers in brackets show altitude of each site (m a.s.l.).

dominance.

3.2 General characteristics of ice core and results of analyses of stable isotope, dust, and soluble ions

The density and stratigraphy of the ice core showed that three fourths of the core was melt and refrozen ice (Fig. 6). The density varied from 340 to 921 kg m^{-3} (mean 656 kg m^{-3}), and its variation in depth was different from those expected for dry-snow. Stratigraphy showed that 75.1% of the total core length was refrozen ice. The ice layers were a mix of ice formations with and without bubbles. Although some firn layers were observed, they accounted for only 21.2% of the total length. The remaining 3.7% was new snow at the glacial surface. Many visible dust layers were observed from the surface to the bottom. Tritium analysis showed that the level was lower than 30 TU between 0.0 and 13.0 m and there was a significant peak at 14.5 m in depth (101 TU, Fig. 6).

Hydrogen stable isotope varied from −146.2‰ to −86.2‰ (Mean: −120.3‰, Fig. 6). Seasonal fluctuation was not clear in the profile. Although some fluctuation was observed, for example, at a depth of between 5 and 8 m, most of the core had little fluctuation.

Microscopy revealed that the ice core contained significant amounts of insoluble dust particles. The dust consisted of both mineral and organic particles. The size of dust ranged up to 102 μm in diameter. The profile of the dust concentration showed many spikes from the surface to the bottom of the core (Fig. 6). The concentration ranged from 0 to 4.5×10^4 particles per mL. The spikes in the dust profile corre-

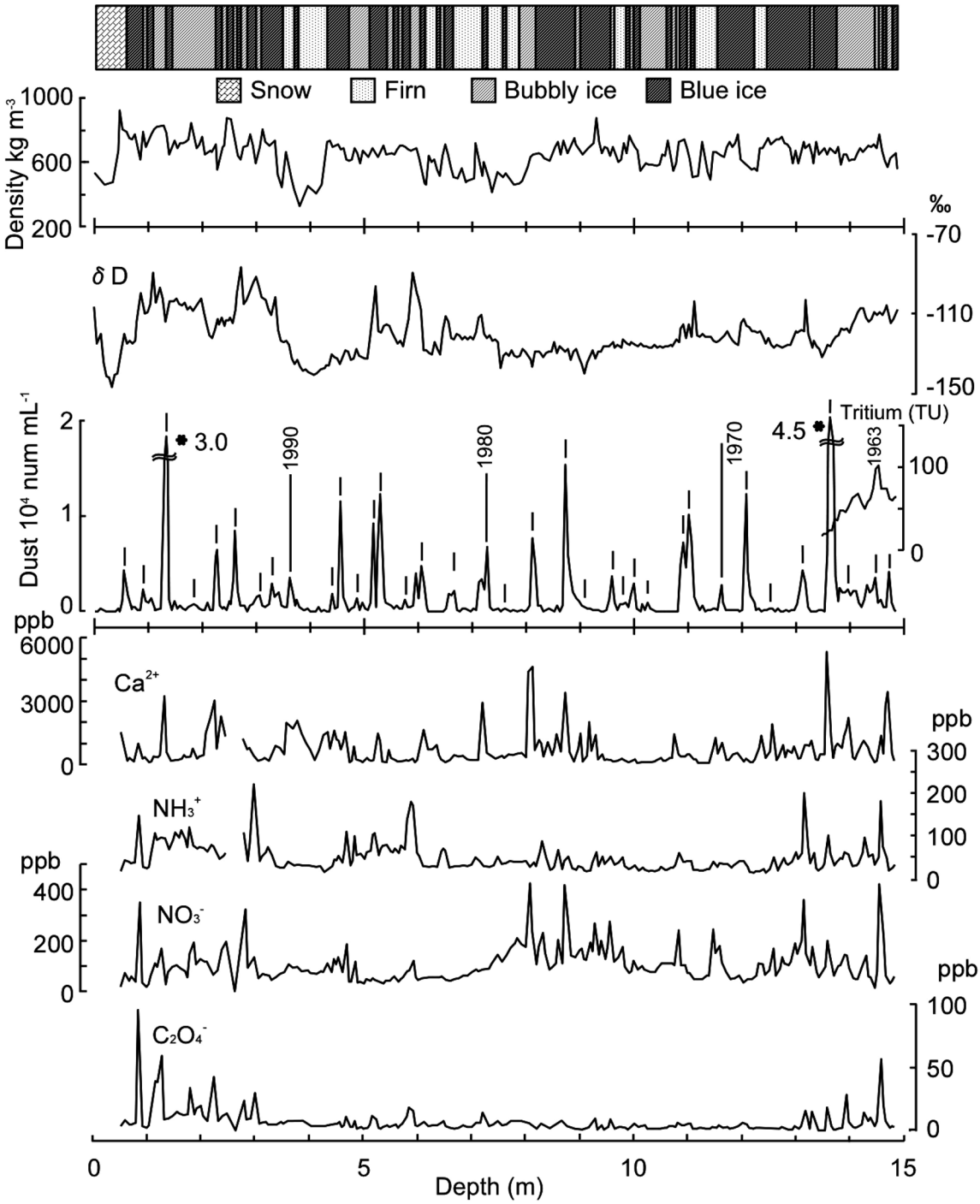

Fig. 6. Profiles of stratigraphy, density, hydrogen stable isotope (δD), dust particles, tritium, and soluble ions (Ca^{2+}, NH_3^+, NO_3^-, $C_2O_4^-$) of the ice core of Rikha-Samba Glacier. Annual boundary layers are indicated with the dust profile.

sponded well to those of visible dust layers.

Chemical analysis showed that the core contains various and abundant soluble ions, each of which showed a different depth profile (Fig. 6). The major soluble ions in this core were Ca^{2+} (85% in weight) and NH_4^+ (6%) for cations, and NO_3^- (37%), SO_4^{2-} (27%), and Cl^- (18%) for anions.

3.3 *Snow algae in the core*

Microscopy revealed algal cells in 101 out of 296 samples of the ice core. The algae included the four taxa (Fig. 7). Based on the cell size and morphology of each taxon, they corresponded with those observed on the surface of the glacier. The taxa observed in the ice core were Oscillatriaceae cyanobacterium 1 (cell size: 1.3±0.2μm in diameter, mean±SD), Oscillatriaceae cyanobacterium 2 (3.3±0.3μm in diameter), Chroococcaceae cyanobacterium (4.1±1.5μm in diameter), and the Unknown Alga (12.3±3.4μm in diameter).

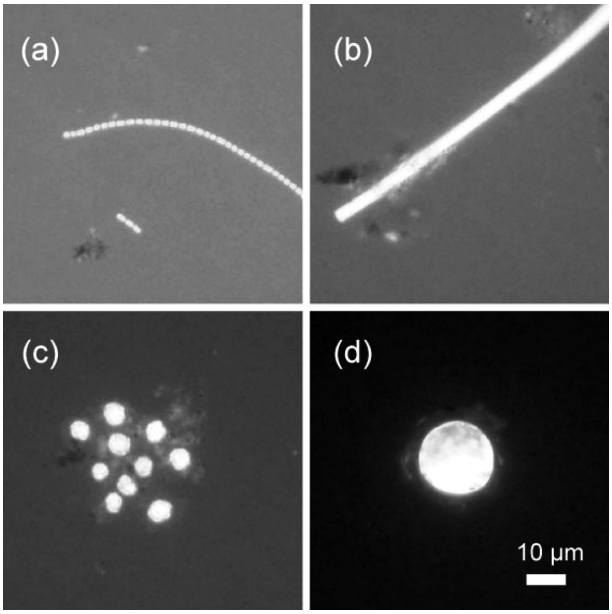

Fig. 7. Photographs of snow algae observed in the ice core of Rikha-Samba Glacier. a. Oscillatoriaceae cyanobacterium 1 (depth: 1.33 m); b. Oscillatoriaceae cyanobacterium 2 (depth: 1.33 m); c. Chroococcaceae cyanobacterium (depth: 1.33 m); d. Unknown Alga (depth: 0.83 m). All photographs were taken with a fluorescence microscope.

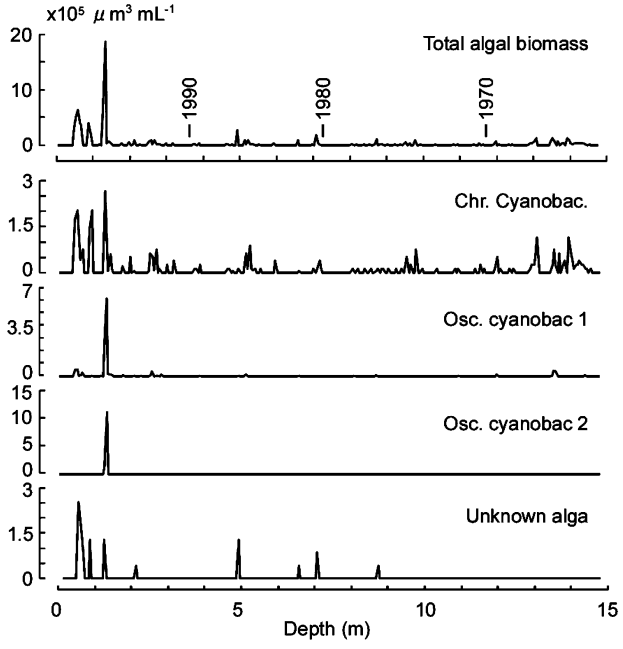

Fig. 8. Profiles of algal cell volume biomass for total and each taxon in the ice core of Rikha-Samba Glacier.

The total cell volume biomass of the algae in the ice core varied from 0.0 to $19\times10^5\mu m^3$ mL^{-1} (Fig. 8). The depth profile of the biomass showed spikes. Three remarkable spikes were observed between 0.4 and 1.4 m in depth, and the other spikes were relatively smaller. The biomass profile also differed among the taxa. The Chroococcaceae cyanobacterium frequently appeared from top to bottom of the ice core, whereas the other three taxa rarely did so.

4. Discussion

4.1 Spatial variations in a snow algal community on the glacial surface

Most of the algae observed on this glacier corresponded to those previously reported on other Himalayan glaciers. According to the previous reports on the other Himalayan glaciers: Yala Glacier (5100–5400 m a.s.l.) by Yoshimura *et al.* (1997) and AX010 Glacier (4800–5300 m a.s.l.) by Takeuchi *et al.* (1998), *M. berggrenii* and *C. brébissonii* are dominant species on the glaciers. *M. berggrenii* is observed not only in the Himalayas, but in many other cold regions in the world. Remias *et al.* (2009) have noted two variations in the cell size of *M. berggrenii*: the smaller one is *M. berggrenii* var. *alaskana* (length: 8–16 μm, width: 8–9 μm) and the larger one is a standard species (length: 20–30 μm, width: 12–13 μm). The alga found on this glacier is likely the smaller one. Oscillatriaceae cyanobacteria are also common taxa on the ice surface of the glaciers, although it is difficult to say that these cyanobacteria are the same taxa based on morphological identification. The Unknown Alga found in this study may be *Trochiscia* sp. or a zygospore of a *Chloromonas* sp., which are also common in the snow surface of the Himalayan glaciers (Yoshimura *et al.,* 1997; Takeuchi *et al.,* 1998). Only the Nostocaceae cyanobacterium in this study was not reported on the two glaciers in previous reports.

The altitudinal change of algal biomass on the glacial surface also showed the same trend as those reported on other Himalayan glaciers. The total cell volume biomass decreased as elevation increased on Yala and AX010 Glaciers as well as on this glacier (Yoshimura *et al.,* 1997; Takeuchi *et al.,* 1998). As suggested in the previous reports, this relationship arises from an altitudinal gradient of the surface conditions affecting algal growth. In the case of the Himalayan glaciers, availability of water and/or length of the snow-free period are the most likely conditions affecting the seasonal biomass. Thus, algal biomass may be correlated positively with the length of the surface melting period in summer.

The community structure of snow algae also results from the change of environments on the glacial surface. Snow algae on glaciers are usually classified into four specialized types (e.g., Yoshimura *et al.,* 1997): snow-environment specialists (observed on the snow surface), ice-environment specialists (observed on bare ice), generalists (observed on both snow and ice), and opportunists (observed on a specific area of the ice or snow surface). Based on the distribution of each species on this glacier and on the previous reports showing the classification of snow algal species on Himalayan glaciers, *M. berggrenii,* Chroococcaceae cyanobacteria, and Oscillatriaceae cyanobacterium 1 can

be categorized as generalists, Unknown Alga as a snow-environment specialist, and *C. brebissonii,* Nostocaceae cyanobacterium, and Oscillatriaceae cyanobacterium 2 as an ice-environment specialist. The variable conditions of the glacial surface favoring growth of certain species are likely to result in the differences in the algal community among sites.

4.2 Dating of the ice core

The core was dated using records of tritium, dust particle concentrations, and stratigraphy. Density and stratigraphy showed significant melt features in the ice core. The lack of seasonal signals on hydrogen stable isotopes and chemical species is likely due to disturbance of melt water percolation. In contrast, there were many clear spikes in dust concentration, which may correspond to annual layers because insoluble dust particles are less affected by melt water percolation. Many studies on ice cores have used dust signals to date annual layers, in particular on Asian ice cores (*e.g.,* Thompson *et al.,* 1989; Han *et al.,* 2006). The dry fallout of dust particles is a common phenomenon in this region and usually occurs in the winter dry season (*e.g.,* Kang *et al.,* 2000). The effect of percolating water on insoluble particles in snow has been examined in previous studies, and particles larger than 7.5μm in diameter were less affected and could stay at the original layer, whereas particles smaller than 5μm could be moved down by melt water (Uetake *et al.,* 2006). Dust analyzed in this study was larger than 5μm and contained a significant amount of large particles ranging up to 100 μm. Thus, dust spikes in the profiles are likely to show the boundaries of annual layers. Tritium records showed a maximum at a depth of 14.5 m, which most likely corresponded to the time of the atmospheric nuclear test in 1963 (Fig. 6). At this depth, the year determined by stratigraphy and dust profiles was consistent with the year 1963.

The dating showed that the entire length of the ice core spanned 37 years. However, since such a large portion of the core was refrozen ice, hiatuses in the annual layers might have occurred. Furthermore, there are some unclear small dust peaks, which might cause misreading of the annual boundary. Thus, the dating possibly includes an error of 1 or 2 years.

4.3 Temporal variations in snow algae derived from the ice core

The annual variation in the total algal biomass of the snow algae showed that the biomass was high in 1965–67, 1977, 1981, 1986, and 1992–98, and it reached particularly high levels in the late 1990s (Fig. 9). The biomass in 1990, 1988, 1980, 1973, and 1962 was under the detection level. The total annual biomass varied from 0.0 to 96$\times10^9\mu m^3\ m^{-2}\ a^{-1}$ (mean, 4.4$\times10^9\mu m^3\ m^{-2}\ a^{-1}$).

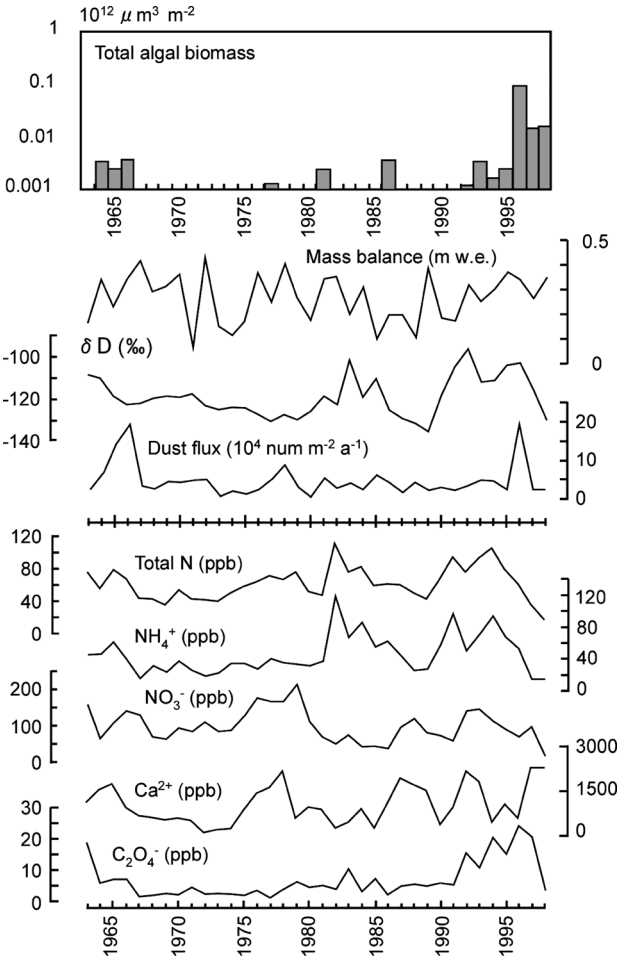

Fig. 9. Annual variations in algal volume biomass, mass balance, dust flux, soluble ions (total nitrogen, NH_4^+, NO_3^-, Ca^{2+}, $C_2O_4^-$), and hydrogen stable isotope (δD), for 37 years derived from the ice core of Rikha-Samba Glacier.

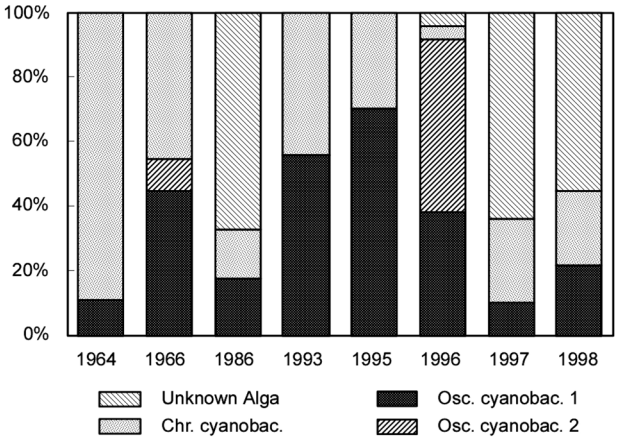

Fig. 10. Difference in community structure of snow algae in specific years derived from ice core of Rikha-Samba Glacier. Community structures shown here are from years in which the algal biomass was higher than $2\times10^9\mu m^3\ m^{-2}\ a^{-1}$.

The increase in the 1990s was particularly significant: the mean biomass in the 1990s was almost 16-fold greater than that before 1990 (17×10^9 versus 1.1×10^9 $\mu m^3\ m^{-2}\ a^{-1}$).

The variation in snow algal biomass in the ice core may indicate differences of algal growth in each year; however, there are many other possible factors that might affect the algal biomass in the ice core. One of them is spatial variation in algal biomass on the glacial surface. According to the surface algal biomass examined in this study, the standard deviation of the spatial biomass variation averaged 5.5×10^9 $\mu m^3\ m^{-2}$ in the upper snow area (sites S6 and S7). The range of the annual biomass before the 1990s (0.0μ 3.9 $\times10^9\mu m^3\ m^{-2}\ a^{-1}$) was smaller than the standard deviation, so spatial variation cannot be excluded from the possible factors affecting the biomass before the 1990s. However, the mean biomass in the 1990s ($17\times10^9\mu m^3\ m^{-2}\ a^{-1}$) significantly exceeded the standard deviation. Therefore, the biomass increase in the 1990s is unlikely to be due to spatial variation, but probably reflects the algal growth on the glacial surface during that period of time.

The biomass variation could also be affected by decomposition after the algae were buried in the glacial snow and/or ice. Yoshimura *et al.* (2006) have shown the variation of snow algal biomass in a 7 m deep ice core on the Yala Glacier (5350 m a.s.l.) in the Himalayas, and have mentioned that algal cells are possibly decomposed by autolysis or bacterial decomposition in layers deeper than 5 m. The Yala Glacier is a temperate glacier: the temperature of all glacial ice is near the melting point (between -1 and 0℃ from the surface to 60 m depth; Watanabe *et al.,* 1984). This higher ice temperature may allow the diagenesis of algal cells in the deeper part of the glacier. In contrast, the Rikha-Samba Glacier in this study is a cold continental-type glacier; the ice temperature in the accumulation zone ranged from -7 to -4℃ between the surface and 23.25 m deep (Fujita *et al.,* 1997; Fujii *et al.,* 1996). Therefore, algae in the ice core of this glacier could be preserved better than that of the Yala Glacier.

The ice core record showed that the species composition of snow algae also varied annually. Figure 10 shows the species composition of snow algae in the years in which the algal biomass was higher than $2\times10^9\mu m^3\ m^{-2}\ a^{-1}$. The two algae, Oscillatriaceae cyanobacterium 1 and Chroococcaceae cyanobacterium, appeared in all of the years, whereas the other two taxa in only certain years. The common two taxa were the dominant species in the upper part of the glacier in the altitudinal distribution (Fig. 5). This difference in algal community structure is likely to result from variation in the algal community grown each year. Although spatial variation in algal community may also result in the difference among the annual layer, the results of examination of the surface algal community showed that species composition was spatially uniform over the surface. On the basis of the species composition, the communities can be classified into three types: Type 1 is a community consisting of two species: Oscillatriaceae cyanobacterium 1 and Chroococcaceae cyanobacterium (1964, 1993, 1995). Type 2 is a community consisting of three species, two of Type 1 and Unknown Alga (1986, 1997, 1998), while Type 3 is a community including Oscillatriaceae cyanobacterium 2 (1966, 1996).

These significant annual variations in algal communities could be further explained by the shift in the altitudinal distribution of snow algae on the glacial surface. For example, the large biomass in 1996 ($96\times10^9\mu m^3\ m^{-2}\ a^{-1}$) is equivalent to that on the glacial surface between 5600 and 5700 m a.s.l. (Fig. 4). This suggests that the surface conditions at the drilling site in 1996 were similar to those on the glacial surface 200–300 m lower than the drilling site. Thus, the algal distribution on the glacial surface in 1996 may have shifted 200-300 m upward. The community structure observed in each annual layer in the core also corresponded to that observed on the glacial surface. The algal community in 1996 and 1966 was Type 3, which included an alga of Oscillatriaceae cyanobacterium 2. This alga was an ice-environment specialist, observed in the middle to lower part of the glacial surface (Fig. 5). Thus, the surface conditions of the drilling site in 1996 and 1986 might be similar to the ice surface in the middle to lower part of the glacier in 1998. The algal community in 1986 and 1997 was a Type 2 community, which is of the same species composition as at the surface of the drilling site in 1998 (Fig. 10). Thus, the surface conditions in 1986 and 1997 appeared to be similar to those in 1998. The surface conditions in the years of smaller or no algal biomass are likely not suitable for algal growth. Such conditions might correspond to a surface at an elevation higher than the drilling site. Thus, the variation in algal community in the ice core suggests that altitudinal distribution of snow algal communities could shift up or down on the glacial surface from year to year. The shift may depend on the physical or chemical conditions on the glacial surface in each season.

The algae found in the ice core were only four out of the seven taxa observed on the glacial surface. The other three algae may not grow at the top of the glacier during that period of time or readily decomposed in the ice. *M. berggrenii,* which was the dominant species in the middle part of the glacier (Fig. 5), was not found in the ice core, although the other algae in the middle to lower part of the glacier were observed. *M. berggrenii* may be decomposed or destroyed easier than the other species

4.4 Comparison of algal variations to mass balance, dust, and soluble ions in the ice core

The physical and chemical conditions that affect annual growth of snow algae on the glacier may be recorded in the ice core. One of the possible factors

affecting the algal community on the surface is net mass balance (net accumulation) of the glacier. Annual net mass balance of the glacial surface generally increases as altitude increases, and also depends on the amounts of accumulation and ablation. Intensive ablation in summer possibly causes more algal growth because algal growth requires liquid melt water. Our previous study showed that altitudinal variation in an algal community on the glacial surface corresponded well to the surface mass balance (Takeuchi *et al.,* 1998). As compared with mass balances obtained from the annual layers of the ice core, however, the algal biomass was not correlated with the mass balance (Fig. 9).

The melting history of Himalayan glaciers (1965–1996) has been extracted by Fujita *et al.* (2006) based on two net balance records of glaciers located approximately 200 km east of the Rikha-Samba Glacier. Their results showed that glacial melting increased during 1969–71, 1980–81, and 1990–92. Snow algae can be expected to grow intensely in these periods. However, these periods do not coincide with the years of high algal biomass in the ice core. Furthermore, there is no similarity between variations in the algal community and air temperatures of Kathmandu (the capital of Nepal, 500 km east of the glacier) and the 500 hPa NCEP/NCAR reanalysis data of Fujita *et al.* (2006). Stable isotopes in the ice cores could also be indicative of climate conditions each year since they usually reflect regional air temperatures and also monsoon activity (*e.g.,* Thompson *et al.,* 2000). However, hydrogen stable isotopes in most of this ice core seem to be modified by melt water percolation and runoff, so they can not be used as a climate signal.

The dust concentration is also a possible factor in the temporal variations in the snow algal community, since dust contains algal spores and also provides the main nutrient source for algal growth on the glacial surface (Yoshimura *et al.,* 1997; Marshall and Chalmers, 1997). The variations in dust flux peaked in the mid-1960s and mid-1990s (Fig. 9). These dust peaks coincide with the years in which algal biomass was high and the Type 3 algal community appeared. Such similarities suggest that the annual variations in snow algae were affected by the flux of the wind-blown dust. However, since the peak of dust flux in the 1990s was comparable to the peak in the 1960s, the significant increase in biomass during the 1990s can not be explained by dust flux variations alone.

Compared with concentrations of soluble ions in the ice core, the emergence of the algal community did not appear to coincide with most ions. Total soluble nitrogen (NH_4^+, NO_3^-, and NO_2^-), which is derived from biogenic or agricultural sources in lower parts of the valleys (Shresta *et al.,* 1997), is expected to affect algae since it can serve as a nutrient for them. The variations in NH_4^+, NO_3^-, and NO_2^- showed different trends, and total nitrogen increased in the early 1980s and 1990s (Fig. 9). However, the variation did not agree with that of algal biomass. The temporal variations in carboxylic acids (formate, acetate, and oxalate; only oxalate is shown in Fig. 9) showed an increasing trend in the 1960s and 1990s, which coincided with an increase of algal biomass. However, the variation in carboxylic acids is unlikely to cause the algal variation because the carboxylic acids do not appear to be derived from sources other than the glacier, but rather from biological activity on the glacial surface.

Although our results could not definitively demonstrate the factors affecting algae, variation in the algal community may be due to the combination of some physical and chemical factors. In particular, the significant increase in algal biomass during the 1990s is likely due to the recent changes in glacial environments so often reported in this region. Glacial shrinkage and climate warming have recently been reported in the Himalayan region. Mass-balance studies in the Himalayas have shown that glacial shrinkage was particularly significant in the 1990s (Fujita *et al.,* 2001a, 2001b). Recent increases in oxygen- or hydrogen-stable isotopes are commonly observed in many ice cores from Asian glaciers, suggesting recent climate warming (*e.g.,* Thompson *et al.,* 1989, 1993, 2000). Both the shrinkage of glaciers and climate warming indicate that the melting intensity of glacial surfaces has recently increased. Such changes can increase liquid water availability on the glacial surface and extend the growth period of snow algae. Furthermore, dramatic increases of soluble ions such as ammonia since the 1950s have been reported on glacial snow in the Himalayan region due to anthropogenic emissions from enhanced agricultural activities and energy consumption over Asia (*e.g.,* Kang *et al.,* 2002). These changes could make the surface conditions more favorable to the growth of algae.

It should be noted that the discussion on algal variation in the ice core is based on the assumption that diagenesis of algal cells did not occur in the glacial ice. Although the physical conditions at the drilling site could support this assumption as mentioned previously, there is no direct evidence. More studies on the diagenetic process of snow algae in glacial ice are warranted. Otherwise, genetic and/or carbon and nitrogen stable isotope analyses on the biogenic material in glacial ice would be more useful to study paleo-ecology on glaciers, since these results are less influenced by diagenesis. However, there is little information about genetic and carbon/nitrogen stable isotopes of glacial organisms on Himalayan glaciers, so further basic studies are required.

Acknowledgments

We wish to thank the staff of the Department of

Hydrology and Meteorology, Government of Nepal and the members of the CREH project for supporting our fieldwork. We are also indebted to Dr. Karl Kreutz for his stable isotope analysis, Drs. Yoshiyuki Fujii and Makoto Igarashi for their tritium analysis, Mr. Toshiaki Miyajima for his assistance with the microscopy analysis, and anonymous reviewers for valuable suggestions, which significantly improved this manuscript. The fieldwork was supported by a Grant-in-Aid for scientific research (No. 21681003, 19310020, 09490018), and the laboratory work by the research project "Ice core analysis with cryo-microbes on glaciers in an arid Chinese region," supported by the Special Coordination Funds for Promoting Science and Technology from the Japanese Ministry of Education, Culture, Sports, Science and Technology, and by the research project.

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