Temporal changes in snow algal abundance on surface snow in Tohkamachi, Japan

Yukihiko ONUMA1, Nozomu TAKEUCHI1 and Yukari TAKEUCHI2

1 Department of Earth Sciences, Graduate School of Science, Chiba University, Chiba 263-8522, Japan
2 Tohkamachi Experimental Station, Forestry and Forest Products Research Institute, Tohkamachi 948-0013, Japan

(Received May 5, 2016; Revised manuscript accepted July 13, 2016)

Abstract

Snow algae are cold-tolerant photosynthetic microbes growing on snow and ice. In order to investigate the factors affecting snow algal growth, the temporal changes in algal abundance on surface snow were studied over four winters in an experimental station in Niigata Prefecture, Japan, where seasonal snow is usually present from late December to early April. Snow algae appeared on the snow surface in February, and the initial algae were likely to be deposited on the snow by winds. The timing of the algal appearance varied among years, from early-February in 2011 to late-February in 2015, and is likely to be determined by a period of no snowfall and air temperatures above the melting point. Algal abundance generally increased until the disappearance of snow. The maximum algal concentration was found in 2011, which corresponds to the year when the period from algal appearance to the disappearance of snow was the longest (80 days) among the four winters. The results suggest that snow algae keep growing unless snowfall occurs and air temperature drops to freezing point, and that the algal abundance is likely to be correlated with the duration of algal growth. The algal growth curve in 2011 could be reproduced by a Malthusian model with a growth rate of 0.22 d⁻¹.

Key words: snow algae, algal growth, snowpack, Tohkamachi, snow melting

1. Introduction

Snow algae are autotrophic microbes observed commonly on melting glaciers and snowfields worldwide. They are adapted to cold environments, and therefore, are able to grow on snow and ice. The most common snow algae are flagellates belonging to the green algae of the order Volvocales (Kol, 1968). They usually play a role as primary producers in snowfield and glacier ecosystem, and thus sustain heterotrophic organisms living on snow and ice (e.g. Kohshima, 1987; Hoham and Duval, 2001).

Algal blooms can change the color of the snow surface to red or green, and reduce snow albedo, thus accelerating snow melting. On seasonal snowfields, red-pigmented snow algae, such as *Chlamydomonas (Cd.)* nivalis and *Chloromonas (Cr.)* nivalis, can often bloom on the snow surface, and acceleration of snow melting by the algal blooms has been reported worldwide (Hoham and Duval, 2001; Takeuchi et al., 2006; Nedbalová et al., 2008). Lutz et al. (2014) reported surface albedos for red snow to be 49% and 44% for green snow, which were lower than the albedo of clean snow (75%), on a Greenland glacier. If the algal distribution on a snowfield is patchy, then algae do not generally affect mean albedos of the snowfield overall, as suggested by Thomas and Duval (1995). However, in some cases, snow algae can be widely distributed on the snow surface and they can reduce substantially the surface albedo (Takeuchi et al., 2006).

Reduction of the surface albedo and enhancement of melting caused by algae has also been reported on the ice surfaces of the Greenland Ice Sheet (Yallop et al., 2012; Lutz et al., 2014). Thus, understanding snow algal ecology is important for the evaluation of surface albedo and melting rates of snow and ice.

Essential conditions for the growth of snow algae are the occurrence of liquid water, solar radiation, and nutrients. When snow melts, snow algal cells grow in the liquid water film surrounding the snow grains (Fukushima, 1963). Pollock (1970) reported that water in liquid phase must continuously be present in the snow for several days before the substantial growth of algae, because air temperature below the freezing point ceases their metabolism. One percent of incident photosynthetically active radiation (PAR) can penetrate at a depth of 1 m in wet snow, allowing algal photosynthesis and germination (Curl et al., 1972). Hoham (1980) revealed that the spatial variation of nutrients might correlate with the spatial distribution of snow algae. Field observations showed that nutrient depletion (particularly nitrate) caused shifts in the life cycle phases of snow algae (Hoham et al., 1989). These environmental
Factors change seasonally and are likely to affect the growth of snow algae. However, there is little information on the temporal changes in snow algae on a snowpack and the factors affecting the algal growth.

Only a few studies have focused on the temporal changes in snow algal communities. Takeuchi (2013) reported the temporal changes in snow algae on the Gulkana Glacier in Alaska during the melting season. According to this study, the snow algal community changed seasonally with changing environmental conditions such as air temperature and snow depth. Lutz et al. (2014) reported that the dominant algal community on surface snow rapidly changed with the rising of the snow line, on a glacier in southeast Greenland.

Snow algal blooms commonly occur on seasonal snowpacks in Japan. Because of strong westerlies in winter, the western coastal regions of Japan are generally covered with abundant snow from December to March. In the mountain regions, the snow depth often exceeds 2-3 m and snow cover remains until July or August. Fukushima (1963) reported on the cell morphology and abundance of snow algae in various mountain regions in Japan and recorded over 10 snow algae species. Some of the species, including Chloromonas fukushima sp. nov. and Chloromonas tenuis sp. nov. have been identified by DNA and microscopic analysis (Matsuzaki et al., 2014). Segawa et al. (2005) reported that the snow algae abundance increased during season on mountain snow in Japan. However, it is still not clear how the meteorological or snow conditions control the temporal changes in snow algae. Because their blooms can affect surface albedo and snow melting, it would be worth to reproduce the seasonal change of algal growth with a simple numerical model. However, numerical growth modeling has never been applied to snow algae.

In the present study, we aimed to quantify the temporal changes in snow algal abundance in an experimental snowfield in Niigata-prefecture, Japan. The algal abundance on the snow surface was quantified biweekly, until the disappearance of the snow cover over four winters. The temporal changes and inter-annual variation of the growth curve were discussed in relation to the meteorological data and the physical and chemical conditions of the surface snow. Furthermore, we attempted to reproduce the observed seasonal change of snow algae with a simple growth model.

### 2. Study site and Methods

The study was conducted at Tohkamachi Experimental Station, Forestry and Forest products Research Institute (37° 08’ N, 138° 46’ E, 200 m above sea level) in Niigata Prefecture, Japan (Fig. 1). The site is located on a river terrace along the Shinano River. This region is well known as one of the snowiest areas in Japan owing to strong winter westerlies from Siberia. This area is usually covered with snow from December to April, and the annual maximum snow depth often exceeds 2 m. During winter, the meteorological conditions and physical properties of snowpack are monitored for the purposes of disaster prevention as well as for glaciological and meteorological studies in the observation field of Tohkamachi Experimental Station covering an area of 1.35 ha (Fig. 1) (Takeuchi et al., 2014). These meteorological and snow pit observations have continuously been conducted since 1918.

The hourly air temperature, solar radiation, wind speed, relative humidity, air pressure, snow depth, and precipitation measured automatically at the field, were used in this study. The temperature sensor was installed at a height of 4 m above ground level. The detailed settings of the meteorological sensors are described in Takeuchi et al. (2014). The diurnal amount of snowfall was calculated from the precipitation and the ratio of snow versus rain according to the protocol of Yamazaki (1998, 2001). The ratio was calculated from air temperature, relative humidity, and air pressure.

The snow pit observations were conducted in the field at about 10 days intervals during the winter season. The snow pit was made every time at the same spot.
The snow type, density, temperature and liquid-water content of snow from the surface to the bottom of a snow pit were observed. The detailed methods for the observations are also described in Takeuchi et al. (2014).

Surface snow collection was conducted every 10 days, simultaneously with the snow pit observations, in 2011, 2013, 2014, and 2015. The absence of samples in 2012 is because of lack of funding for this research. The snow samples were collected from 3-5 randomly selected surfaces surrounding the snow pit. The sampling area and depth were 80-400 cm² and 2-4 cm, respectively, and the data were recorded for each collection. All snow samples were preserved in Whirl-Pak® bags (Nasco, Fort Atkinson, WI, USA). Snow samples from a snow pit (depth=121 cm) in the field were collected from the surface (depth=0-2 cm), the subsurface (depth=2-10 cm), and other layers (depth=10-121 cm) on April 6, 2015. The samples from the other layers were collected at 10 cm intervals, except for the layer closest to the bottom of the pit (depth=110-121 cm). Twenty-nine snow samples were collected from the snow pit.

The samples were kept frozen and transported to a laboratory at Chiba University, Japan, and stored in a freezer (−20°C) until further analysis. The snow samples were melted in room temperature for analyses of algal cell count. Electrical conductivity (EC) and pH of each sample were measured with a portable pH-conductivity meter (F-54, Horiba, Japan).

Algal abundance was estimated as cell concentration based on the cell counts and melted water volume of the samples used for the counting. The cell counts were conducted with an optical microscope (BX51, Olympus, Japan). A volume of the water sample (20-1000 µL) was filtered through a hydrophilic membrane filter (0.45 µm JHWP01300, Millipore, Billerica, MA, USA), and the number of algae cells in the entire filter area was counted. Counting was conducted 1-3 times for each sample. From the average number of the counted cells and the volume of filtered water, the algal cell concentration (cells mL⁻¹) was obtained.

3. Results

3.1 Meteorological conditions

Meteorological observation showed that air temperature generally decreased from December to January, and then increased gradually in February, March, and April. For example, in the winter of 2011, the monthly mean temperature for December, January, February, March, and April was 3.8, −1.4, 0.8, 1.4, and 6.7°C, respectively (Fig. 2a). Daily mean temperatures in the winter of 2011 ranged from −1.1 to 10.4°C in December, −3.7 to 1.4°C in January, −1.2 to 7.6°C in February, −2.8 to 6.4°C in March, and 2.7 to 12.1°C in April, suggesting that snow melting occurred irregularly in January and February, and more continuously in March and April.

Solar radiation also decreased from December to January, and then increased gradually. For example, the monthly mean solar radiation in December, January, February, March, and April in the winter of 2011 was 611.5, 523.1, 1177.5, 1500.0, and 1946.6 W m⁻², respectively (Fig. 2a). The daily mean solar radiation varied largely depending on weather conditions. For example, in 2011 it ranged from 9.6 to 120.9 W m⁻² in December, 17.0 to 117.6 W m⁻² in January, 159.0 to 2035.7 W m⁻² in February, 328.0 to 2833.0 W m⁻² in March and 334.0 to 3089.7 W m⁻² in April, indicating that both sunny and cloudy conditions...
were frequent during that winter.

Precipitation occurred as both snow and rain, throughout the seasons. Total amounts of snow and rain in the winter of 2011 were 192.6 and 201.4 mm in December, 674.3 and 21.2 mm in January, 112.0 and 38.5 mm in February, 237.2 and 38.8 mm in March, and 29 and 123.1 mm in April, respectively (Fig. 2b).

Wind speed ranged from 0.4 to 2.1 m s$^{-1}$ in daily mean and did not show a clear seasonal trend. For example, the monthly mean wind speed in December, January, February, March, and April in the winter of 2011 was 1.0, 1.1, 0.9, 1.1, and 1.0 m s$^{-1}$, respectively (Fig. 2c). The daily mean wind speed in 2011 ranged from 0.5 to 19 m s$^{-1}$ in December, 0.6 to 15 m s$^{-1}$ in January, 0.4 to 13 m s$^{-1}$ in February, 0.6 to 21 m s$^{-1}$ in March, and 0.6 to 16 m s$^{-1}$ in April, suggesting that wind speed almost didn’t change in winter.

These meteorological conditions varied among the four years (Figs. 2, 3, 4, and 5). The mean air temperature from January to April in the four winters was highest in 2015 (3.4°C) and lowest in 2011 (1.9°C). The amount of snowfall also differed among four winters. The total

---

**Fig. 3.** Meteorological conditions and snow algal abundance at the Tohkamachi experimental station in the winter of 2013. (a) Daily mean air temperature and solar radiation, (b) snowfall and snow depth, (c) daily mean wind speed, (d) algal cell concentration. Error bars=standard deviation. Open and solid marks for algal cell concentration indicate fresh and granular snow, respectively (d).

**Fig. 4.** Meteorological conditions and snow algal abundance at the Tohkamachi experimental station in the winter of 2014. (a) Daily mean air temperature and solar radiation, (b) snowfall and snow depth, (c) daily mean wind speed, (d) algal cell concentration. Error bars=standard deviation. Open and solid marks for algal cell concentration indicate fresh and granular snow, respectively (d).
snowfall in the four months from January to April was highest in 2011 (1026 mm) and lowest in 2014 (627 mm). The mean solar radiation and wind speed during the same period did not vary significantly among the years, ranging from 118 to 130 W m\(^{-2}\) (mean 124 W m\(^{-2}\)) and 1.0 to 1.1 m s\(^{-1}\) (mean 1.0 m s\(^{-1}\)), respectively.

3.2 Physical and chemical conditions of the snowpack

The snow depth in the field indicated that snow started to accumulate in December, reached maximal depth in February, melted in March and April, and then finally disappeared in late April (Tables 1, 2, 3, and 4; Figs. 2b, 3b, 4b, and 5b). In 2011, the period of snow coverage in the field was from December 7 to April 29, and the maximum snow depth was 302 cm on January 31. The surface snow type was mostly fresh snow in January and granular snow from February to April in all four winters. The volumetric liquid-water content on the snow surface in 2011 was almost 0% until February, increased to 1.0% in February 4, then gradually kept increasing up to 7.9% (April 25) until the snow disappeared. The water content for the four winters ranged from 0.0 to 5.2% (mean: 2.4%) in the period from December to March, and from 3.5 to 8.4% (mean: 5.6%) in April. The temporal pattern of the variations in snow depth and water content for the other three winters followed a similar pattern to the winter of 2011. However, the timing of the snow disappearance varied among the years, ranging from April 11 in 2014 to April 29 in 2011. The maximum depth of snow was greatest in 2011 (302 cm), and lowest in 2014 (183 cm), corresponding to the years of later and earlier timing of the snow disappearance, respectively.

EC for the surface snow ranged from 0.6 to 103.6 µS cm\(^{-1}\) and did not show a clear seasonal trend over the four winters (Tables 1, 2, 3, and 4). The pH for the surface snow ranged from 4.51 to 6.95 over the four winters and did not show a clear seasonal trend during the study period. There was no significant difference in the mean value of EC or pH among the four winters.

3.3 Snow algae in snowpack

Microscopic observation revealed that two morphological types (A and B) of snow algal cells appeared on the snow surface in all four winters (Fig. 6). The type A cell (Fig. 6A) was an ovoid shape with a ribbed cell wall. The cell length was 36.5 ± 6.4 µm (mean ± SD) and the width was 21.2 ± 2.8 µm. The chloroplast was located in the middle of the cells and the orange cytoplasm was located in the outer side of the cells. The type B cell (Fig. 6B) was also an ovoid shape, but no ribbed cell wall was observed. The cell size was smaller than the type A cell; the cell length and width was 20.4 ± 2.2 µm and 14.6 ± 1.8 µm, respectively. The chloroplast was located in the middle of the cells.

The vertical profiles of snow algal abundance in a snow pit (depth=121 cm) on April 6, 2015 showed that the algal cell concentration peaked at the surface, decreased with depth, and was zero at a depth below 50 cm (Fig. 7a). The total algal cell concentration ranged from 1.3 × 10\(^6\) to 2.8 × 10\(^2\) cells mL\(^{-1}\) at the surface snow whereas it was 0 cells mL\(^{-1}\) at a depth below 50 cm from the snow surface. Type A cells were observed at a depth above 50 cm and type B cells were at a depth above 40 cm. The proportion of type A cells to the total algal cell concentration in the snow above 40 cm ranged from 25 to 60%. The snow type was granular without any ice layer and snow temperature was 0°C at all depths in the snowpack. The snow density and volumetric liquid-water
Table 1. List of algal abundance and chemical and physical properties of surface snow in the winter of 2011. The asterisk at the sampling date indicates the first day when the snow algae were observed in the winter.

| Sampling date | Algal cell concentration (cells mL\(^{-1}\)) | Electrical conductivity (\(\mu S\) cm\(^{-1}\)) | pH | Water content of snow (%) | Type of snow |
|---------------|-----------------------------------------------|-----------------------------------------------|----|--------------------------|-------------|
| 5-Jan         | 0.0 ± 0.0                                     | 17.5 ± 4.4                                    | 6.0 ± 0.7 | 1.4                       | Fresh       |
| 14-Jan        | 0.0 ± 0.0                                     | 7.6 ± 0.1                                     | 5.8 ± 0.3 | 0.0                       | Fresh       |
| 25-Jan        | 0.0 ± 0.0                                     | 29.0 ± 0.9                                    | 5.0 ± 0.3 | 0.0                       | Fresh       |
| *4-Feb        | 0.7 ± 0.6                                     | 13.2 ± 1.9                                    | 5.5 ± 0.2 | 1.0                       | Granular    |
| 14-Feb        | 0.0 ± 0.0                                     | 14.8 ± 0.5                                    | 5.4 ± 0.1 | 0.0                       | Fresh       |
| 25-Feb        | 1.0 ± 0.0                                     | 4.6 ± 0.2                                     | 6.7 ± 0.2 | 3.8                       | Granular    |
| 7-Mar         | 0.4 ± 0.6                                     | 10.6 ± 0.9                                    | 5.9 ± 0.2 | 4.2                       | Granular    |
| 16-Mar        | 2.9 ± 2.7                                     | 23.7 ± 4.3                                    | 5.6 ± 0.1 | 0.4                       | Fresh       |
| 25-Mar        | 6.6 ± 6.3                                     | 4.4 ± 0.2                                     | 6.1 ± 0.2 | 3.2                       | Granular    |
| 5-Apr         | 3.4 ± 1.4                                     | 3.9 ± 0.1                                     | 5.8 ± 0.4 | 3.9                       | Granular    |
| 15-Apr        | 263 ± 270                                     | 4.7 ± 0.8                                     | 6.3 ± 0.3 | 3.3                       | Granular    |
| 26-Apr        | 2059 ± 1739                                   | 5.1 ± 1.5                                     | 6.1 ± 0.2 | 7.9                       | Granular    |

Table 2. List of algal abundance and chemical and physical properties of surface snow in the winter of 2013. The asterisk at the sampling date indicates the first day when the snow algae were observed in the winter.

| Sampling date | Algal cell concentration (cells mL\(^{-1}\)) | Electrical conductivity (\(\mu S\) cm\(^{-1}\)) | pH | Water content of snow (%) | Type of snow |
|---------------|-----------------------------------------------|-----------------------------------------------|----|--------------------------|-------------|
| 25-Jan        | 0.0 ± 0.0                                     | 17.9 ± 6.9                                    | 5.7 ± 0.6 | 1.5                       | Granular    |
| 4-Feb         | 0.0 ± 0.0                                     | 72.1 ± 29.5                                   | 4.7 ± 0.1 | 1.5                       | Fresh       |
| *15-Feb       | 0.3 ± 0.6                                     | 6.5 ± 0.3                                     | 6.6 ± 0.3 | 1.2                       | Granular    |
| 25-Feb        | 0.0 ± 0.0                                     | 64.1 ± 12.6                                   | 5.2 ± 0.2 | 0.0                       | Fresh       |
| 5-Mar         | 0.0 ± 0.0                                     | 60.5 ± 1.5                                    | 4.5 ± 0.1 | 0.2                       | Fresh       |
| 15-Mar        | 0.0 ± 0.0                                     | 7.5 ± 0.5                                     | 6.5 ± 0.2 | 0.0                       | Granular    |
| 26-Mar        | 0.0 ± 0.0                                     | 56.0 ± 5.9                                    | 4.8 ± 0.3 | 0.8                       | Fresh       |
| 5-Apr         | 2.8 ± 1.0                                     | 11.8 ± 14.0                                   | 6.2 ± 0.1 | 3.5                       | Granular    |
| 16-Apr        | 8.1 ± 6.4                                     | 2.5 ± 0.5                                     | 6.7 ± 0.2 | 8.4                       | Granular    |
Table 3. List of algal abundance and chemical and physical properties of surface snow in the winter of 2014. The asterisk at the sampling date indicates the first day when the snow algae were observed in the winter.

| Sampling date | Algal cell concentration (cells mL⁻¹) | Electrical conductivity (µS cm⁻¹) | pH | Water content of snow (%) | Type of snow |
|---------------|--------------------------------------|-----------------------------------|----|---------------------------|--------------|
| 6-Jan         | 0.0 ± 0.0                            | no data                           | no data | 0.0           | Fresh       |
| 15-Jan        | 0.0 ± 0.0                            | no data                           | no data | 0.0           | Fresh       |
| 24-Jan        | 0.0 ± 0.0                            | 15.8 ± 4.0                        | 5.2 ± 0.1 | 0.0           | Granular    |
| 5-Feb         | 0.0 ± 0.0                            | no data                           | no data | 0.0           | Fresh       |
| 14-Feb        | 0.0 ± 0.0                            | 2.5 ± 0.7                         | 6.0 ± 0.0 | 0.0           | Fresh       |
| *25-Feb       | 4.5 ± 4.4                            | 1.9 ± 0.5                         | 6.1 ± 0.1 | 0.8           | Granular    |
| 5-Mar         | 4.8 ± 3.3                            | 0.6 ± 0.3                         | 6.7 ± 0.2 | 4.9           | Granular    |
| 17-Mar        | 8.6 ± 7.9                            | 1.1 ± 0.4                         | 6.6 ± 0.8 | 1.4           | Granular    |
| 25-Mar        | 4.8 ± 3.4                            | 1.3 ± 0.8                         | 6.9 ± 0.1 | 3.9           | Granular    |
| 4-Apr         | 459 ± 960                            | 0.6 ± 0.1                         | 6.5 ± 0.1 | 5.4           | Granular    |

Table 4. List of algal abundance and chemical and physical properties of surface snow in the winter of 2015. The asterisk at the sampling date indicates the first day when the snow algae were observed in the winter.

| Sampling date | Algal cell concentration (cells mL⁻¹) | Electrical conductivity (µS cm⁻¹) | pH    | Water content of snow (%) | Type of snow |
|---------------|--------------------------------------|-----------------------------------|-------|---------------------------|--------------|
| 5-Jan         | 0.0 ± 0.0                            | 16.9 ± 6.8                        | 5.3 ± 0.4 | 4.8           | Granular    |
| 15-Jan        | 0.0 ± 0.0                            | 19.3 ± 6.6                        | 5.2 ± 0.2 | 0.0           | Granular    |
| 26-Jan        | 0.0 ± 0.0                            | 14.3 ± 8.1                        | 5.8 ± 0.3 | 1.1           | Granular    |
| 5-Feb         | 0.0 ± 0.0                            | 38.5 ± 2.0                        | 5.0 ± 0.1 | 0.0           | Granular    |
| 16-Feb        | 0.0 ± 0.0                            | 103.6 ± 60.6                      | 5.3 ± 0.2 | 0.5           | Fresh       |
| *25-Feb       | 1.3 ± 1.5                            | 3.6 ± 0.5                         | 6.3 ± 0.3 | 4.0           | Granular    |
| 5-Mar         | 0.3 ± 0.6                            | no data                           | no data | 0.5           | Fresh       |
| 16-Mar        | 0.7 ± 0.6                            | 5.3 ± 0.4                         | 6.2 ± 0.4 | 5.2           | Granular    |
| 25-Mar        | 8.3 ± 7.6                            | 8.8 ± 1.4                         | 6.0 ± 0.1 | 4.2           | Granular    |
| 6-Apr         | 85 ± 123                             | 4.6 ± 0.7                         | 5.6 ± 0.1 | 5.6           | Granular    |
| 15-Apr        | 31 ± 11                              | 3.9 ± 1.3                         | 6.0 ± 0.0 | 6.9           | Granular    |
| 24-Apr        | 50 ± 44                              | 2.7 ± 0.4                         | 6.0 ± 0.1 | 5.7           | Granular    |
content ranged from 465 to 557 kg m$^{-3}$ and 4.0 to 5.8% in the snow pit, respectively, and did not show any clear trend with depth (Figs. 7b and c).

### 3.4 Temporal changes in algal cell concentration of surface snow

Total cell concentration of snow algae on the surface indicated that the algae appeared in February, but the timing of appearance varied among the four winters (Tables 1, 2, 3, and 4). The algae appeared on February 4 in 2011, which was the earliest appearance among the four winters. Algae appeared on February 15 in 2013, and on February 25 in 2014 and 2015.

The algal concentration generally increased in March and reached a maximum in April. In 2011, the algal abundance was $6.8 \times 10^4$ cells mL$^{-1}$ when the algae first appeared on February 4, and then increased to $6.6 \times 10^0$ cells mL$^{-1}$ on March 25, although it decreased occasionally, on March 3 and April 5 (Fig. 2d). The algae finally reached the maximum value of $2.1 \times 10^3$ cells mL$^{-1}$ on April 26, which was just before the disappearance of the snow cover on April 29. No change of snow color to red or green was observed during the observation period in 2011. In the other three winters, the algal abundance gradually increased after their appearance, however, the maximum abundances were much smaller than those in 2011 (Figs. 3d, 4d, and 5d). A statistical test showed that the temporal changes in the algal cell concentration were significant in 2011 (one-way ANOVA, $F=2.22$, $P<0.01$). Statistical tests showed that temporal changes in the algal abundance also were significant in the winter of 2013 (one-way ANOVA, $F=2.71$, $P<0.01$), but were not significant in the winters of 2014 and 2015 (2014: $F=2.32$, $P>0.05$; 2015: $F=5.99$, $P>0.05$).

The duration from the appearance of algae to the maximum algae concentration on the surface also varied among winters. It was longest in 2011 (80 days), was 60 days in 2013, 38 days in 2014, and 40 days in 2015.
4. Discussion

4.1 Life cycle of snow algae in Tohkamachi

Snow algal cells observed in this snow field were likely Chloromonas (Cr.) nivalis, which are commonly reported from snowfields in Japan, as well as worldwide (Muramoto et al., 2008). The shape, size, and pigmentation of the type A algal cells corresponded with those of the cysts of Cr. nivalis as reported by Remias et al. (2010). Cr. nivalis is a species characterized by a wide tolerance to environmental factors, growing both at shade (spruce or broadleaf trees) and open exposure sites (Komárek and Nedbalová, 2007; Nedbalová et al., 2008), and can be seen on snowfields in Japanese mountains during the melting season (Muramoto et al., 2008). Most commonly, the immotile mature cysts populate the snow surface, and they have in abundance the secondary carotenoid astaxanthin that causes the orange color of the cytoplasmic lipid bodies (Remias et al., 2010), which could be seen in type A cells. The shape and pigmentation of type B cells was similar to those of the vegetative cells of Cr. nivalis, reported by Nedbalová (2008). These motile cells have been recorded in the field only for short period of time because they quickly cease cellular divisions and start a rapid process of cyst formation into robust spores (Remias et al., 2010). Both type A and B cells were observed during the entire melting period at the study site, suggesting that cell division and cyst formation of the algae occurred throughout the melting season.

The snow conditions over the four winters suggest that the snow algae did not originate from the ground surface under the snowpack, but from the atmosphere. Resting cells of snow algae can be transported by wind or animals from distant places (up to hundreds of kilometers), or they develop into motile cells and swim up to the surface from the ground soil under the snow (Müller et al., 2001; Remias, 2012). In order to transform motile cell from resting cell, solar radiation as well as water is required (Hoham, 1980). According to Curl et al. (1972), one percent of incident PAR can reach at a depth of 1 m in wet snowpack and may promote photosynthesis and algal germination. When snow algae appeared on the snow surface of the study site, the snow depth was deeper than 1 m in all of the seasons (244 m, 203 m, 143 m, and 270 m in 2011, 2013, 2014, and 2015, respectively) and appeared to be too deep for snow algal spores germination at the ground surface under the snowpack. Furthermore, the snow pit observation on April 6, 2015 did not record any algal cells in the snow layers below the depth of 50 cm (Fig. 7a, 14 samples, snow depth=121 cm). Marshall and Chalmers (1997) reported that cells of Cd. nivalis could be found in air samples at 1 m above ground level in Antarctica. These results suggest that the snow algae at the study site are likely from the atmosphere by wind transport although further study is necessary.

The meteorological conditions and algal cell concentrations suggest that snow algae start to grow when the period of no snowfall and air temperature above 0°C exceeded approximately 24 hours. Since fresh snow coverage inhibits photosynthesis of the snow algae below the snow, no new snowfall for a certain period appears to be important to initiate snow algal growth. The records showed that no snowfall over 0.1 mm occurred for at least 29 hours before the algal appeared in all four winters. Furthermore, snow melting is required for the algal growth as suggested by previous studies (Fukushima, 1963). The surface was melting granular snow in all four winters, when the algae appeared on the surface. The hourly air temperature exceeded 0°C for 24 hours, or longer, just before the algal appearance in four winters. The periods when air temperature exceeded 0°C were 25 hours in 2011, 28 hours in 2013, 24 hours in 2014, and 104 hours in 2015. Wind speed did not show a clear seasonal trend over the four winters, and there appeared to be no relationship between the wind speed and the timing of algal appearance. Therefore, the prevalence of these conditions for longer than 24 hours is likely to be the minimum requirement to initiate the snow algal growth.

4.2 Annual variations in algal growth curves and maximum abundance

The differences in algal growth curves among the four winters can be explained by the duration of algal growth in each winter. The algal growth curve in 2011 showed that the algae increased exponentially until the disappearance of snow and the maximum algal abundance was the greatest among the four winters. In contrast, the algal growth curves in the other three years showed that the algae slightly increased in March and April, and the maximum algal abundances were much smaller than those in 2011. In 2011, the timing of algal appearance was earlier and the accumulated snowfall in January was greatest (674.3 mm), which could result in a longer growth period for the snow algae. In fact, the period from algal appearance to snow disappearance was 84 days in 2011, but was 62, 45, and 60 days in 2013, 2014, and 2015, respectively. The total hours of air temperature above 0°C after the algal appearance were also longest in 2011 (1415 hours), but were 1065, 888, and 1291 hours for 2013, 2014, and 2015, respectively. This also suggests that snow algae grew continuously for a longer duration in 2011 than in the other three winters. In the other three winters, the algal growth appeared to be interrupted frequently by occasional snowfall and by a decrease of air temperature below the freezing point. Other conditions, such as snow temperature, solar radiation, EC, pH, and water content, did not show any seasonal trend and any difference among the four years, suggesting that none of these conditions directly affect the algal growth curve. Therefore, snow algal abundance is likely to keep increasing as far as the snow melts when there is no
fresh snow coverage, and the exponential increase and greater algal maximum abundance in 2011 are probably because of the longer duration of algal growth.

4.3 Approximation of the algal growth curve

In general, an increase of microbial cells can simply be expressed by a differential equation called Malthusian model, which is defined with an initial cell concentration and a growth rate of the microbe and has been applied to observational microbial abundance (Lavoie et al., 2005). Based on the assumptions that there is no inflow or outflow of algal cells on the snow surface, and that light, nutrients, and habitable space for the algae are not limited, a growth curve of algal cell abundance can be expressed by the following equation (Cui and Lawson, 1982):

\[ X = X_0 e^{\mu(t-t_0)}, \]

where, \( X \) and \( X_0 \) are population densities of algal cells at \( t \) and \( t_0 \) respectively; \( t \) is the number of days, and \( \mu \) is the growth rate of algal cells in \( \text{d}^{-1} \). \( t_0 \) is defined as the day of the first appearance of algae on the snow surface. Because snow algae can grow only when surface snow melts, \( t \) includes only the days when daily mean air temperature was higher than 2°C. The temperature of melting point (2°C) was determined based on a previous study (Kojima et al., 1983). This equation was fitted by a least-squares regression to the observational algal cell concentrations in 2011. Although the type A and B algal cells observed in this study is likely to be different life stage of Cr. nivalis, the cell concentrations used for the regression were the total of types A and B cells since the cell divisions occur in the stage of vegetative cells. The maximum cell abundance in 2011 was 2059 cells mL\(^{-1}\), which was lower than those of Chloromonas sp. reported previously in mountain snow (e.g. 3.0×10\(^3\) cells mL\(^{-1}\), Hoham et al., 1993), probably due to earlier timing of snow disappearance in Tohkamachi. However, the model suggests that the cell abundance would reach the level of 10\(^5\) cells mL\(^{-1}\) if snow remained in another 21 days. Result suggest that this simple numerical model can reproduce the seasonal change of algal growth and also possibly predict their effect on surface albedo and melting of snow although further research is required to evaluate the parameters of the equation.

5. Conclusions

Investigation of snow algae on the snowpack in Tohkamachi, Japan over four winters revealed the temporal and inter-annual changes in their abundance. Two morphological types of snow algae were observed in the site, and they are likely to be Cr. nivalis, which is a common species on seasonal snow surfaces in Japan, as well as worldwide (Muramoto et al., 2008). The timing of the algal appearance on the snow surface varied from February 4 to February 25 in the four winters. The meteorological conditions suggest that the timing of algal appearance is determined by a continuous period of no fresh snow coverage and air temperatures above 0°C: the snow algae appeared on the surface when that period exceeded approximately 24 hours. The algal growth curves varied among the four years, and the maximum algal concentration was greatest in 2011. The difference in algal growth curves among the four winters can be explained by the duration of algal growth in each winter. The duration, which corresponds to the period from algal appearance to snow disappearance, was the longest (84 days) in 2011, probably because of a larger amount of snowfall during this year. Thus, snow algal abundance is likely to keep increasing as far as the snow melts and there is no fresh snow coverage, and the exponential increase and greater maximum abundance in 2011 are probably because of the longer duration of algal growth. The growth curve of algae in 2011 could be reproduced in another 21 days. Result suggest that this simple numerical model can reproduce the seasonal change of algal growth and also possibly predict their effect on surface albedo and melting of snow although further research is required to evaluate the parameters of the equation.
acknowledgments

we would like to thank yasoichi endo, shoji niwano and shigei murakami for the snow sampling and snow pit observations during four winters in the tohkamachi experimental station, forestry and forest products research institute. we also thank two anonymous reviewers and an editor (masahiro hori) for helpful suggestions that greatly improved this manuscript. this study was conducted by a collaborative project between chiba university and forestry and forest products research institute supported by japan society for the promotion of science (jspqs) grant-in-aid for scientific research (no. 23221004, 02624707, and 26241020).

references

cui, q. and lawson, g. j. (1982) study on models of single populations: an expansion of the logistic and exponential equations. j. theor. Biol., 89, 645–659. doi: 10.1016/0022-5193(82)90043-6.
curl, h. jr., hardy, j.t. and ellermeier, r. (1972) spectral absorption of solar radiation in alpine snow fields. ecology, 53, 1189–1194. doi: 10.2307/1936433.
fukushima, h. (1963) studies on cryophytes in japan. j. yokohama mun. univ. ser. c, nat. sci., 43, 1–146.
hoham, r. w. (1980) unicellular chlorophytes-snow algae. in phytogelagete (cox er, ed), 61–84.
hoham, r. w. and duval, b. (2001) microbical ecology of snow and freshwater ice. snow ecology, cambridge university press, 168–228.
hoham, r. w., yatsko, c. p., germain, l. and duval, b. (1993) snow algae and other microbes in several alpine areas in new england. in proc. 50th annual eastern snow conf., 196–200.
hoham, r. w., laursen, a. e., clive, s. o. and duval, b. (1993) snow algae and other microbes in several alpine areas in new england. in proc. 50th annual eastern snow conf., 165–173.
kojima, k., moyotomo, h. and yamada, y. (1983) estimation of melting rate of snow by simple formulae using only air temperature in (japanese). low temp. sci., 42, 101–110.

Kohshima, S. (1987): Glacial biology and biotic communities. In Kojima, K., Motoyama, H. and Yamada, Y. (1983): Estimation of meteorology and snow pit observations at Tohkamachi in Niigata Prefecture, Japan (8) (2009-10 to 2013-14, five winter periods) (in Japanese). Bull. Forestry Forest Prod. Res. Inst., 13, 271–334.

Thomas, W. H. and Duval, B. (1995): Sierra Nevada, California, USA, snow algae: snow albedo changes, algal-bacterial interrelationships, and ultraviolet radiation effects. Acta. alp. usa, 8, 033002, doi: 10.1088/1748-9326/8/3/033002.

Takeuchi, N. (2013): Seasonal and altitudinal variations in snow algal communities on an Alaskan glacier (Gulkana glacier in the Alaska Range) Environ. res. lett., 8, 035002, doi: 10.1088/1748-9326/8/3/035002.

Takeuchi, N., dial, r., kohshima, s., segawa, t. and uetake, j. (2006) spatial distribution and abundance of red snow algae on the harding icefield, Alaska derived from a satellite image. Geophys. Res. Lett., 33, L21502, doi: 10.1029/2006GL027819.

Takeuchi, Y., Endo, Y., niwano, S. and murakami, S. (2014): Data of meteorology and snow pit observations at tohkamachi in Niigata Prefecture, Japan (6) (2009-10 to 2013-14, five winter periods) (in Japanese). Bull. Forestry Forest Prod. Res. Inst., 13, 271–334.

Thomas, W. H. and Duval, B. (1995): Sierra Nevada, California, USA, snow algae: snow albedo changes, algal-bacterial interrelationships, and ultraviolet radiation effects. Acta. alp. usa, 8, 033002, doi: 10.1088/1748-9326/8/3/033002.

Yallop, M.L., Anesio, A.M., Perkins, R.G., Cook, J., telling, J., pagan, D., macFarlane, J., stibbl, M., Barker, G. and Bellas, C. (2012): Photophysiology and albedo-changing potential of the ice algal community on the surface of the Greenland ice sheet. Isme J., 6, 2302–2313, doi: 10.1038/ismej.2012.107.

Yamazaki, T. (1998): A multi-layer heat balance model of snow cover adaptable to intensely cold regions (in Japanese). Seppyo, 60, 131–141, doi: 10.3331/seppyo.60.131.

Yamazaki, T. (2001): A one-dimensional land surface model adaptable to intensely cold regions and its applications in eastern Siberia. J. Meteorol. Soc. jap., 79, 1107–1118, doi: 10.2151/jmsj.79.1107.