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Snow Algae of Mt. Watson

Fiona McBride
Western Washington University

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By Fiona McBride

Advised by Dr. Robin Kodner

Abstract

The taxonomy and life history of all the organisms that belong to the category of "snow algae" is complicated and contentious. Though scientists publish studies every year investigating these questions, many gaps remain in our collective knowledge. A study over two years investigated a newly described genus, *Sanguina*, in a subglacial habitat system on Mt. Watson. Using size and morphologic data we found yearly occurrences of cysts of both species and can conjecture about their life cycle changes according to microhabitat. Additional work will be done in sequencing and reviewing data before submission for publication.

Introduction

Algae is well known today for its criticality in ecosystems as a source of food and oxygen for almost all living things. But snow algae, an alpine and circumpolar group of organisms whose blooms turn snow green to orange to red, is rather less known. As such, the phycological community is still in uncharted territory when it comes to some of the details of the taxonomy and life history of these organisms.

A 2019 paper described a new genus, *Sanguina*, with two species: *S. aurantia and S. nivaloides* (Procházková et al.) Prior to this publication, *Sanguina* had been lumped in with the genus *Chlamydomonas*, despite differences in cyst morphology and genomics. The paper characterized external and internal cyst morphology, stating a lack of other life cycle stages that had been scientifically observed, as both *S. nivaloides* and *S. aurantia* seem to exist as cysts as soon as their blooms are visible.

Both species of *Sanguina*, as well as the titular *Chlamydomonas* cyst, were observed for two summers in a row in the late season on Mt. Watson in the North Cascades, Washington. In addition to dense blooms, what made this site interesting was the bright red meltwater pools (tarns) full of snow algae. Due to the variations in site conditions, from glacier to snow field to pools, Mt. Watson was a unique location to explore differences in algal communities by immediate environmental conditions over a small area.

Methods

In September of 2020 and August of 2021, day expeditions were taken up Mt. Watson in the Washington State North Cascades. The target site, visited each year, was directly downhill of Watson Glacier and consisted of several snow fields, once contiguous but separated into troughs after almost a full season of snowmelt. Melting had progressed further when samples were collected in 2021 than in 2020 despite the expedition happening almost a month earlier. The site also contains numerous tarns (temporary pools of melted snow) which were red in color due to the presence of snow algae. Some of these tarns had dried up but left a red residue behind. Snow was melting rapidly, and dark red areas signaled high algal content.

In 2020, the lowest snow field was sampled three times towards the middle and three times at the lower, melting edge. Additional samples were collected from the tarns. In 2021, samples were collected more broadly: from the middle of the snowfield directly underneath the glacier, the middle of a snow field in equivalent elevation to the subglacial field but not beneath the glacier, the middle and melting edge of the same, lower snow field from 2020, and several meltwater pools, wet and dry. Samples were collected fresh or preserved in Lugols and transported back to the lab freezer in insulated thermoses.

The primary method of data analysis for this study was done via Scanning Electron Microscopy. Samples were subsampled and dried on glass microscope slides, then promptly coated in gold palladium. Images were taken with a consistent image size and view field with a magnification of 3.46kx. Images were taken haphazardly (a fully systematic approach would have been inefficient due to cell clumping) but with an objective of taking a representative collection of what was in the sample. Images were analyzed in ImageJ. A trapezoid tool was used to outline each cell and measured for area; this was also done with the ellipse tool. As many of the cells had partially collapsed in the SEM vacuum, this tool was critical in estimating area visible by tracing the cell's outline to the greatest extent possible. Cells were assumed to be spherical, and an estimate of area calculated by ImageJ was used to estimate the radius and then the volume of each cell. Cells were also classified according to morphotypic features, with categorizations of *Sanguina* based on the work of Procházková et al. (2019). Over 800 cells were measured overall in this way.

Additional methods of sample processing involved light microscope imaging and surveys as well as an 18S sequence from each location (middle and melting edge of the lower snow field) from 2020.

Results

According to sequencing data and morphotypic classification, the site was dominated by *Chlamydomonas sp., Sanguina aurantia*, and *Sanguina nivaloides* cysts, with additional high counts of smooth or roughly smooth cells which could be earlier-stage cysts or vegetative forms of any of these cells (or something else entirely). *Chlamydomonas* was predominantly observed in the melting edge of 2020 samples, where it constituted a vast majority of the present cells and was only very sparsely present in the middle 2020 sample and all 2021 samples. Smooth cells dominated in the middle samples from 2020.

In 2021, smooth cells were dominant in the lower melting edge and tarn samples. Samples from snow field middles were primarily *Sanguina aurantia* cysts. *S. nivaloides* was present at roughly 15 percent in most of these samples.

In terms of cell size, smooth-walled cells and both species of *Sanguina* had roughly the same size distribution (averaging approx. 30 cubic microns), with *S. aurantia* having a greater proportion of large cells. Cell size seemed to increase on average with decreasing snow field elevation. In light microscopy images only, significant numbers of flagellated cells were discovered in tarn samples. These we were unable to classify taxonomically but are suspected to be *Sanguina*.

Full data analysis, images, and graphs to be published.

Discussion

The intent of the study had been to investigate life cycle changes in snow algae according to differences in microhabitat. In this regard it is inconclusive, as we do not have the data to characterize what species and life stage the smooth cells belong to – indeed, classifying any morphology with certainty is beyond our current means.

Instead, this study shed more light on the recently described *Sanguina* genus. Both known species are shown to cohabit the same location multiple years in a row. The size data expands on the knowledge of both species as well – in particular, *S. aurantia* is described as being small, about 5-10 microns in diameter (Procházková et al. 2019), but from our data *S. aurantia* typically had a diameter of at least 10-20 microns. The uneven distribution of size by site, the relative lack of *Chlamydomonas* in the second year, the appearance of swimmer cells in light images only, and the ambiguity of the smooth-walled cells are tantalizing discoveries with no real answer.

Further steps

In order to progress this study to a published work, several steps must be taken. Firstly, more in-depth sequencing must be done to more conclusively link morphotype with species. This may involve single-cell PCR and sequencing from a broader range of samples. The swimming cells should be more fully investigated, both with sequencing and imaging. Critical point drying may be necessary to ensure the least amount of damage in SEM. Additional relevant lab data and images from the Mt. Watson expeditions should be catalogued and incorporated into the analysis – tarn and rock samples are the most sparse and additional data would be useful.

As the distinctions in the literature between *Sanguina* and *Chlamydomonas* remain fuzzy, we consider furthering this understanding to be a vital part of the study.

Works Cited

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