**Editor (Roberto Ambrosini):**

Thank you much for the review of our manuscript. We have addressed all the comments. Please see below for our point-by-point responses to the Editor's comments (in blue and preceded by "REPLY:").

The manuscript would benefit from a clearer separation between the results and the discussion. Currently, the RDA and the GLMs are mentioned in the discussion only, while they should be moved to the Results. Dunn's test is a post-hoc test useful for multiple comparisons after a Kruskal-Wallis test. It seems to me that your analyses compare only two groups, so the Mann-Whitney U-test is more straightforward. The results should not markedly differ, however. In contrast, if you are comparing more than two groups, you should report the outcome of the Kruskal-Wallis test first, and then those of the Dunn's test

REPLY: We sincerely thank the Editor for these helpful comments. We have reorganized the manuscript to more clearly separate the Results from the Discussion. Specifically, we added a new subsection, "3.6 Environmental factors associated with algal morphology and community structure," and moved the results of the linear models and RDA, together with Table 1 and Figure 8, from the Discussion to this subsection. The corresponding Discussion sections now focus on interpretation of these results. We also agree that Dunn's test was not the most appropriate method for the planned two-group comparisons. Comparisons between soil-based and ice-based surface snow were therefore reanalyzed using Mann–Whitney U-tests. Because surface and subsurface samples from the ice-based snowpack were collected at the same sampling sites, these observations were paired and were reanalyzed using Wilcoxon signed-rank tests. These changes were applied to algal abundance, spherical cell size, and nutrient concentrations. The Methods, Results, and Figure 4 caption have been revised accordingly, and the relevant U or V statistics are now reported together with P values. The paired analyses resulted in some changes from the original Dunn's test results, including a significant difference in spherical cell abundance between the surface and subsurface layers, and we revised the corresponding Results and Discussion accordingly. You will read "Comparisons between surface samples from soil-based and ice-based snowpacks were conducted using Mann–Whitney U-tests, whereas paired comparisons between surface and subsurface samples collected at the same ice-based sites were conducted using Wilcoxon signed-rank tests." in Lines 185–187 of the revised manuscript.

Line 174: please specify which distribution you are assuming for the data in the GLMs. If you are assuming a Gaussian distribution (the default in the glm procedure in R), state it clearly or simply call these models "linear models". Please also check model assumptions and briefly mention the outcome of these checks in the results.

REPLY: The models assumed a Gaussian error distribution, and we have therefore followed the Editor's suggestion and refer to them simply as "linear models" throughout the revised manuscript. We also added a description of the model-assumption checks to the Methods and briefly report the outcome of these checks in the Results. You will read "Spherical and non-spherical cell concentrations were log(x + 1)-transformed prior to analysis, whereas spherical cell size was analyzed without transformation. Model assumptions were assessed using standard residual diagnostic plots." in Lines 189–191 of the revised manuscript. You will also read "Inspection of residual diagnostic plots did not indicate substantial violations for the main surface-snow models, although several potentially influential observations were identified. Sensitivity analyses excluding these observations did not alter the principal results for the surface models, including the negative association between non-spherical cell abundance and snow depth. Some associations in the subsurface models were more sensitive to influential observations and were therefore interpreted cautiously." in Lines 287–291 of the revised manuscript.

Line 179: please specify how many permutations.

REPLY: We have now specified that the significance of the overall RDA model, constrained axes, and explanatory variables was assessed using 999 permutations. You will read "The significance of the overall model, constrained axes, and explanatory variables was assessed using permutation tests with 999 permutations." in Lines 195–196 of the revised manuscript.

Line 195 and elsewhere: please report the values of the pivot quantities for statistical tests (z-values for the Dunn's tests, U-value for the Mann-Whitney test) and the relative degrees of freedom (when necessary). Please avoid reporting only P-values.

REPLY: We reported the relevant test statistics together with P values. Specifically, U statistics were reported for Mann–Whitney U-tests, V statistics for paired Wilcoxon signed-rank tests, t statistics with the corresponding degrees of freedom for linear models, and F statistics with degrees of freedom for the RDA permutation tests. We also revised the manuscript throughout to avoid reporting p values alone.

Lines 291-297 should go to a dedicated session of the Results. In addition, please report

the degrees of freedom for the F-statistics throughout the text.

REPLY: We moved the RDA results from the Discussion to the newly created Results subsection "3.6 Environmental factors associated with algal morphology and community structure." We also added the degrees of freedom to the reported F statistics. You will read "RDA explained 72.2% of the total variation in algal community composition (adjusted $R^2 = 0.555$, Fig. 8), and the overall constrained model was significant ($F_{3,5} = 4.32$, $p = 0.009$). Among the constrained axes, RDA1 was significant ($F_{1,5} = 9.41$, $p = 0.009$), whereas RDA2 ($F_{1,5} = 3.53$, $p = 0.187$) was not significant. Sequential permutation tests showed significant effects of snow depth ($F_{1,5} = 6.24$, $p = 0.009$) and elevation ($F_{1,5} = 6.61$, $p = 0.006$), whereas nutrients were not significant ($F_{1,5} = 0.12$, $p = 0.907$)." in Lines 291–296 of the revised manuscript.

Legend to Figure 8: Please specify whether this is a correlation or a distance biplot.

REPLY: Corrected as suggested. You will read "Figure 8: Redundancy analysis (RDA) correlation biplot of algal community composition based on Hellinger-transformed relative abundance data." in the revised manuscript.

Line 360: Table 1 should go to the results

REPLY: We moved Table 1 from the Discussion to the newly created Results subsection "3.6 Environmental factors associated with algal morphology and community structure". You will read "Linear models showed that spherical cell size at the snow surface decreased significantly with elevation ($t_{26} = -2.62$, $p = 0.014$), whereas non-spherical cell abundance decreased significantly with snow depth ($t_{26} = -2.56$, $p = 0.017$; Table 1)." in Lines 282–283 of the revised manuscript.

**Reviewer 1:**

Thank you much for the review of our manuscript. We have addressed all the comments. Please see below for our point-by-point responses to the reviewers (in blue and preceded by "REPLY:").

This study uses microscopy and DNA sequencing to examine the spatial heterogeneity of snow algae across an elevation gradient in Alaska. The study presents an important comparison of supraglacial snow compared to snow over soils and presents evidence that factors related to snowmelt including snow depth and elevation are important factors in snow algae phenotype as well as potential underlying substrate (e.g., ice vs. soil). The manuscript and data are exceptionally well written and presented and the data and findings will be of interest to a broad audience.

REPLY: We sincerely thank the reviewer for the careful evaluation of our manuscript and for the positive and encouraging comments. We are pleased that the reviewer found the comparison between supraglacial snow and snow overlying soils, as well as the analyses of snow-algal spatial heterogeneity, to be important and of broad interest.

I have a handful of comments for improvement:

Line 67: Deposition could still be local. Perhaps a more specific description is needed here.

REPLY: We agree that "locally sourced cells" was ambiguous because it could include cells deposited onto the snow surface from nearby sources. We corrected and changed the sentence as suggested. Now you can read "Within this framework, snowmelt duration can be viewed as an approximation of the time available for in situ algal growth, while snow depth may influence the relative contribution of cells associated with the underlying substrate to surface snow communities." in Lines 68–70.

Lines 105-106: Why the different depths? Based on Figure 2, snow algae are in the snowpack below 2 cm.

REPLY: We collected the upper 2 cm of red snow because this depth has commonly been used in previous snow algae studies and therefore allowed comparison with published data. However, previous observations indicated that snow algae can also occur below the upper 2 cm of the snowpack (Schuler and Mikucki, 2023, *Arctic, Antarctic, and Alpine Research*, doi.org/10.1080/15230430.2023.2233785). At ice-based sites, where the

snowpack was sufficiently deep, we therefore additionally collected samples extending to a depth of 3 cm to better capture the contribution of subsurface algal cells. In contrast, snowpacks overlying soil were often shallow, and deeper sampling would have increased the risk of contamination by soil particles and soil-associated organisms. We therefore restricted sampling at soil-based sites to the upper 2 cm. We have added this explanation to the Methods and clarified that the sampling depths differed according to snowpack conditions and the underlying substrate. Now you can read "Red snow was collected from a 10 × 10 cm area using a stainless-steel scoop. The upper 2 cm was defined as the surface layer because visible red-snow blooms are generally concentrated in the uppermost few centimetres of the snowpack, and a 0–2 cm layer has commonly been used to quantify surface snow algal communities (Onuma et al., 2022). However, recent studies have shown that snow algae can also occur below the immediate surface, including within deeper snow layers (Schuler and Mikucki, 2023). We therefore additionally collected the underlying 2–5 cm layer as a subsurface sample at the ice-based sites. Subsurface sampling was not conducted at soil-based sites because the snowpack was generally shallow and deeper sampling could have introduced soil particles and soil-associated organisms (Fig. 2)" in Lines 108–114.

Line 130: Is the 100 cells true for each sample, for both geometric size and diameter?

REPLY: The minimum of 100 cells applies only to measurements of spherical-cell diameter, which were conducted separately for each sample to characterize cell-size distributions. For the other algal morphotypes, size distributions were not analyzed; instead, the dimensions of at least five cells were measured using samples in which each morphotype was abundant for geometric cell-size calculation. We added information about the measurement of geometric size. Now you can read "For geometric cell-size calculation, the dimensions of at least five cells were measured for each algal morphotype using samples in which the respective morphotype was abundant." in Lines 139–141.

Line 149: Remove reaction (redundant with PCR)

REPLY: Corrected as suggested.

Line 210: I like this comparison but I'm confused about the surface vs. subsurface samples. I think the methods need to be updated to better describe the sample collection.

REPLY: As described in our response to the previous comment, we have revised the sample-collection section to define the surface and subsurface sampling intervals explicitly and to explain why subsurface samples were collected only at ice-based sites.

For figure 3, nitrate and ammonium cannot be 0. They would be below the limit of detection. These data points should not be shown on the graph because these numbers do not exist to plot.

REPLY: We have removed these data points from Figure 4. We have also revised the figure caption to clarify that nitrate and ammonium were measured in all samples, but values below the respective detection limits are not shown. Corrected Figure 4 and its caption was shown below.

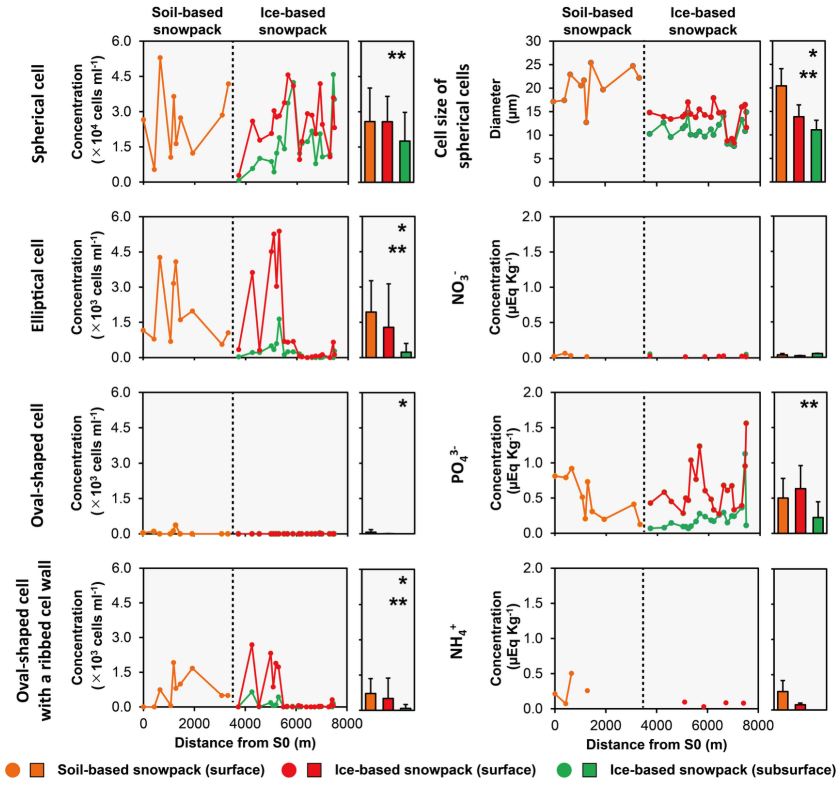



Figure 4: Spatial variability of algal abundance, size of spherical cell, and nutrient concentrations. Spatial distributions are shown in the left panels, and comparisons among the on-glacier snowpack (surface and subsurface) and the glacier forefield (snow surface) are shown in the right panels. * indicates a significant difference between the on-glacier snowpack and the glacier forefield (Mann-Whitney U-test, $p < 0.05$), and ** indicates a significant difference between the surface and subsurface within the on-glacier snowpack (Wilcoxon signed-rank test, $p < 0.05$). $NO_3^-$ and $NH_4^+$ concentrations below the respective limits of detection were not plotted.

Line 232: Abundant is probably more accurate than dominated

REPLY: Corrected as suggested.

**Reviewer 2:**

Thank you much for the review of our manuscript. We have addressed all the comments. Please see below for our point-by-point responses to the reviewers (in blue and preceded by "REPLY:").

The present manuscript explores the morphological and taxonomic variability of red snow algae in different elevational gradients and substrate types (soil-based or ice-based) across Alaskan glacier and its forefield, using molecular as well as microscopic approaches. The study provides valuable insights into the composition of snow algae in various habitats, as well as information on variations in algal communities responsible for red snow across elevation and substrate gradients. I also value the comparison and discussion on different approaches and their outputs. The manuscript is well written, with only minor language inconsistencies that are easy to correct.

REPLY: We sincerely thank the reviewer for the positive and encouraging assessment of our manuscript. We appreciate the reviewer's recognition of the value of combining microscopic and molecular approaches to examine the morphological and taxonomic variability of red snow algae across elevational and substrate gradients. We are also pleased that the comparison of these approaches and their respective outputs was considered informative.

My main concern lies in the analysis of soluble nutrients, which are evaluated throughout the study. The approach used to collect soluble nutrients, especially phosphorus, is not commonly used, so validation of this method for these types of samples is needed or needs to be referred in the study. This concern stopped me from further review of the data and results connected to soluble nutrients. Also, when evaluating surface and subsurface data, I miss further information on radiation conditions from the sampling campaigns, as cells of snow algae can migrate within the snow layer. I believe that such data could contribute substantially to a better evaluation of the results. I describe all my comments in point-by-point below.

I must also stress that I am not an algologist by specialization. I recommend this paper to be seen by a specialist in snow and glacier algae, as I am not fully available to review the algal information and data in detail.

REPLY: We thank the reviewer for identifying these important concerns. The comments regarding the collection and preservation of samples for soluble nutrient analysis,

particularly phosphorus, and the potential influence of radiation conditions on the vertical distribution of snow algal cells were highly valuable. In response, we have clarified and revised the relevant methodological descriptions and added further information and discussion where appropriate.

Abstract

L16: the sentence, as currently written, is not fully consistent with results which state that (e.g., L195) mean concentration of spherical cells between environments were similar. Please revise it so that its meaning is clear and consistent with the results.

REPLY: The original sentence referred to differences in morphotype composition rather than differences in the absolute concentration of spherical cells. We have revised the sentence to clarify that it describes the relative proportions of spherical and non-spherical cells. Now you can read "Microscopic observations revealed that the ice-based snowpack was characterized by a greater proportion of spherical cells, whereas the soil-based snowpack was characterized by a greater proportion of non-spherical cells and larger spherical cells." in Lines 16–18.

Methods

L110 + Sect. 2.3: For the chemical analysis of $PO_4^{3-}$, acid-prewashed bottles are commonly used to prevent adsorption of P to the vessel surface, for example, and further bias in analyses. Were your vessels acid-washed before sampling?

REPLY: The sample tubes were not acid-washed before use. However, before collection of the final aliquot, each tube was rinsed twice with the corresponding filtered sample. In addition, Milli-Q water blanks analyzed together with the samples showed no detectable $PO_4^{3-}$ peak. We have added these sample-handling details to the revised Methods. Now you can read "For chemical analysis, the meltwater was filtered immediately after melting through a 0.45 μm ion-free disposable filter designed for ion-chromatographic sample preparation (25AI ChromatoDisk, GL Sciences, Japan) to minimize the potential influence of particulate material on the dissolved solute measurements, following procedures used in previous studies of snow and glacier samples (Terashima et al., 2017; Takeuchi et al., 2019; Román et al., 2026). In addition, samples were also transferred into 6 ml tubes for molecular analysis at selected sites located (S1, S4, S7, S9, S10, S14, S22, S25, and S30). Samples for chemical and molecular analyses were transported frozen and stored in a freezer (−20 °C) at Chiba University and thawed immediately before analysis whereas samples for microscopic observations were stored at room temperature." in Lines 116–123.

My main concern is freezing samples for chemical analysis. Even though the samples were filtered, samples for P analysis are commonly stored in the refrigerator, not frozen, as freezing might break down fine particles, for example, and affect the final P concentration in the analysed water. Has your approach been validated for this type of sample? If so, providing a reference to the particular validation or detailed justification for the particular approach would help to increase the reliability of your results.

REPLY: Although we did not conduct a direct comparison between refrigerated and frozen aliquots of the samples analyzed in this study, comparable filtration and storage procedures have been used in previous chemical analyses of snow and glacier samples. Takeuchi et al. (2019; "Variations in Phototroph Communities on the Ablating Bare-Ice Surface of Glaciers on Brøggerhalvøya, Svalbard", Frontiers in Earth Science, 7:4, doi:10.3389/feart.2019.00004) used a 0.45 µm ion-free disposable ChromatoDisk to filter glacier meltwater before measuring soluble ions, including $PO_4^{3-}$, by ion chromatography. Terashima et al. (2017; "Microbial Community Analysis of Colored Snow from an Alpine Snowfield in Northern Japan Reveals the Prevalence of Betaproteobacteria with Snow Algae", Frontiers in Microbiology, 8:1481, doi:10.3389/fmicb.2017.01481) filtered colored-snow samples and stored them at −20 °C before chemical analyses that included total phosphate. More directly comparable to the present study, Román et al. (2026; "Unveiling the Large Coverage of Red Snow Algae Blooms in Antarctic Coastal Snowfields", Communications Earth & Environment, 7:53, doi:10.1038/s43247-025-03156-6) filtered red-snow samples, stored the filtrates frozen in PE tubes, and subsequently measured $PO_4^{3-}$. No artificial increase in $PO_4^{3-}$ attributable to frozen storage was reported in these studies. We have therefore added the filter specifications, tube-rinsing procedure, storage and thawing conditions information, and the relevant references to the revised Methods. Nevertheless, we acknowledge that these published applications provide methodological precedents rather than a direct refrigerated-versus-frozen validation of the specific samples analyzed in the present study. As this concern overlaps with the preceding comment, the corresponding revisions to the Methods are the same as those described in our response above.

L136: I might have overlooked it, but it is not clear to me at which section the quantitative comparison of morphotype composition with molecular taxonomic composition by calculating volumetric biomass for each morphotype is present.

REPLY: The morphotype composition shown in Fig. 6d was calculated on the basis of relative volumetric biomass and was compared with the molecular taxonomic composition shown in Fig. 6c. We have revised the Methods to provide the calculation explicitly.

Now you can read “The average cell counts and the volume of sample examined were used to calculate the concentration of each morphotype per unit volume of snow meltwater (cells $mL^{-1}$). For each morphotype, volumetric biomass (µL $L^{-1}$) was calculated by multiplying its cell concentration by its mean cell volume. For samples subjected to molecular analysis, morphotype composition based on relative volumetric biomass was compared with molecular taxonomic composition based on relative sequence abundance.” in Lines 146–148.

L177: As discussed further, all morphotypes can vary in their species composition, also some snow algae species revealed cryptic diversity (i.e. Remias et al. 2023). How was the pooling of all non-spherical cells for further analyses and evaluation justified? They might differ in their metabolic/environmental requirements and pooling such data into a single category may limit the explanatory power of further analyses and evaluation of results.

REPLY: We agree that pooling the non-spherical morphotypes was not intended to imply that they were taxonomically or ecologically equivalent. In our samples, each individual non-spherical morphotype occurred only sporadically and at low abundance. Their separate site-level proportions could therefore not be estimated reliably, and retaining each morphotype as an independent category would have produced several highly sparse categories. We pooled them only to provide a broad compositional comparison between the dominant spherical cells and the less abundant non-spherical cells in Fig. 6d. The pooled category was not used to infer that its constituent morphotypes shared the same metabolic or environmental requirements. We have revised the Methods to clarify the limited purpose of this pooling and have added a statement to the Discussion acknowledging that it may obscure taxon-specific ecological patterns, particularly given the potential cryptic diversity of snow algae (Remias et al., 2023). Now you can read “Because each non-spherical morphotype occurred only sporadically and at low abundance, their separate site-level biomass proportions could not be estimated reliably. They were therefore pooled into a descriptive category (“non-spherical cells”) only for the broad comparison with spherical cells. This pooling does not imply taxonomic or ecological equivalence among the constituent morphotypes.” in Lines 191–194 and “The individual non-spherical morphotypes occurred at low abundance and were therefore pooled for the broad compositional comparison. However, this pooled category should not be interpreted as a single taxonomic or functional group. Morphologically similar snow algae may comprise genetically distinct lineages with different ecological characteristics (Remias et al., 2023), and pooling the non-spherical morphotypes may therefore obscure taxon-specific distribution patterns.” in Lines 325–329.

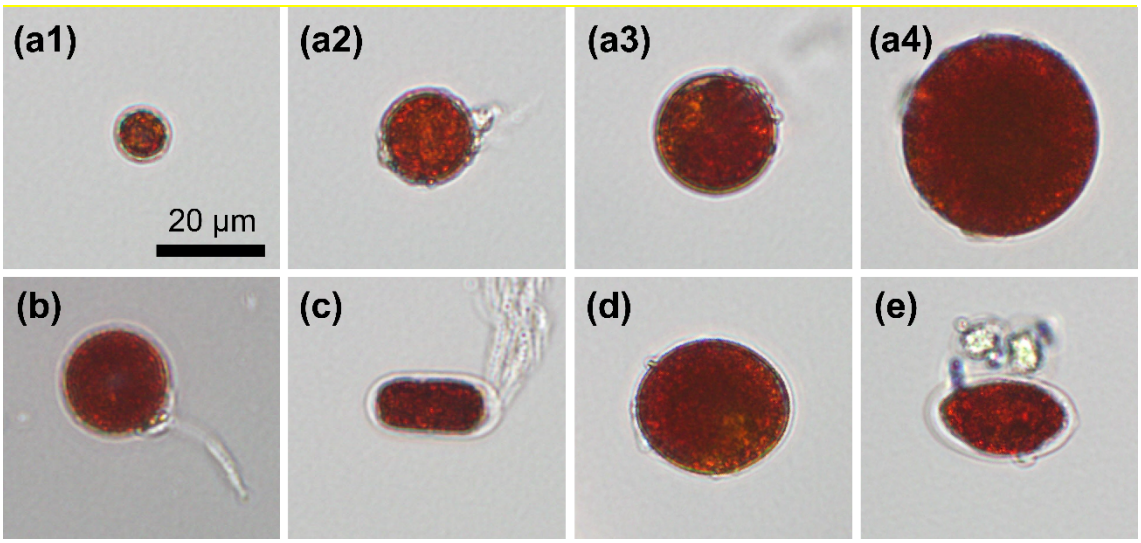
Results

3.1. I suggest calling the section “Red snow algae morphotypes in snow samples” as the section refers solely to morphotypes.

REPLY: Corrected as suggested.

L190: I would recommend including the scale bar size directly in the picture to help with the orienting in the figure.

REPLY: We added a scale bar size in the figure as suggested, and deleted the sentence “A scale bar is 20 µm.” from the caption. Now you can see the modified Figure 3 below.




Sect. 3.3. While reading this section, I am wondering why subsurface samples weren’t taken and analysed also for soil-based samples if the reported lowest depth was 4 cm in all sites and subsurface samples were taken approx. 3–5 cm deep? I cannot find any justification for this decision as such results might be very valuable for the study.

REPLY: As also explained in our response to Reviewer 1, we revised the Methods to clarify the rationale for collecting subsurface samples only from the ice-based snowpack. Although snow depth at the soil-based sites was at least 4 cm, these snowpacks were generally shallow and close to the underlying soil. Collecting deeper samples therefore posed a greater risk of contamination by soil particles and soil-associated organisms. For this reason, sampling at the soil-based sites was restricted to the surface layer. We have added this explanation to the revised Methods and clarified the sampling depths and definitions of surface and subsurface samples. Now you can read “Red snow was collected from a 10 × 10 cm area using a stainless-steel scoop. The upper 2 cm was defined as the

surface layer because visible red-snow blooms are generally concentrated in the uppermost few centimetres of the snowpack, and a 0–2 cm layer has commonly been used to quantify surface snow algal communities (Onuma et al., 2022). However, recent studies have shown that snow algae can also occur below the immediate surface, including within deeper snow layers (Schuler and Mikucki, 2023). We therefore additionally collected the underlying 2–5 cm layer as a subsurface sample at the ice-based sites. Subsurface sampling was not conducted at soil-based sites because the snowpack was generally shallow and deeper sampling could have introduced soil particles and soil-associated organisms (Fig. 2)" in Lines 108–114.

Fig. 6 + 7. The orientation in the figures while reading the results is rather difficult. For the better orientation, I recommend adding a heading along the y-axis describing the data origin (e.g., 18S rRNA, potentially red pigmented, etc.).

REPLY: As recommended, we changed Figures 6 and 7 to indicate the origin and category of the data, including 18S rRNA data and potentially red-pigmented algal taxa. Because this information is now shown directly in the figures, we removed the corresponding explanatory text from the figure captions to avoid redundancy. Now you can see modified figures 6 and 7 below.

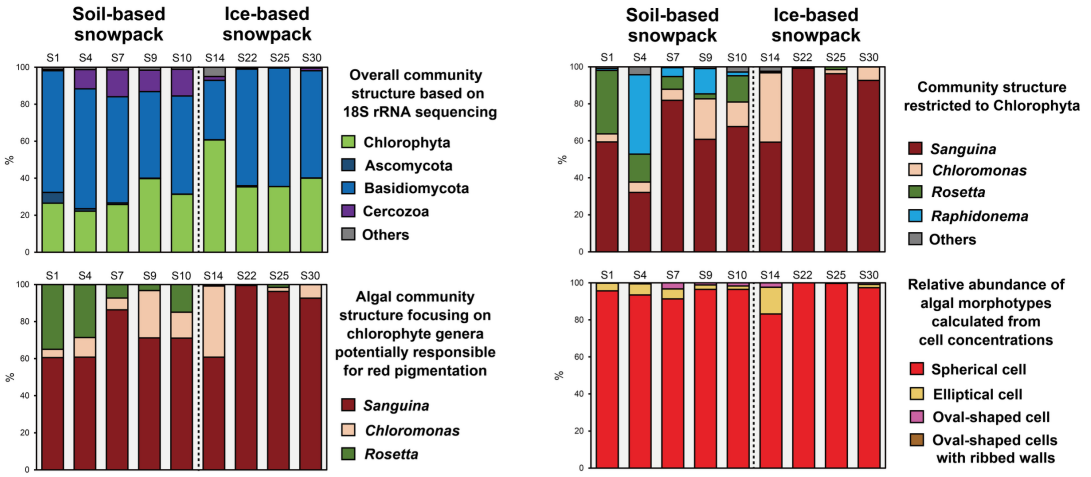



**Figure 6: Microbial community structure on the snow surface based on 18S rRNA gene sequencing and morphological observations. Community structure at each elevation is presented as stacked bar charts normalized to 100%. For the molecular dataset, only phyla or genera accounting for >5% of the total reads were included in the analysis.**

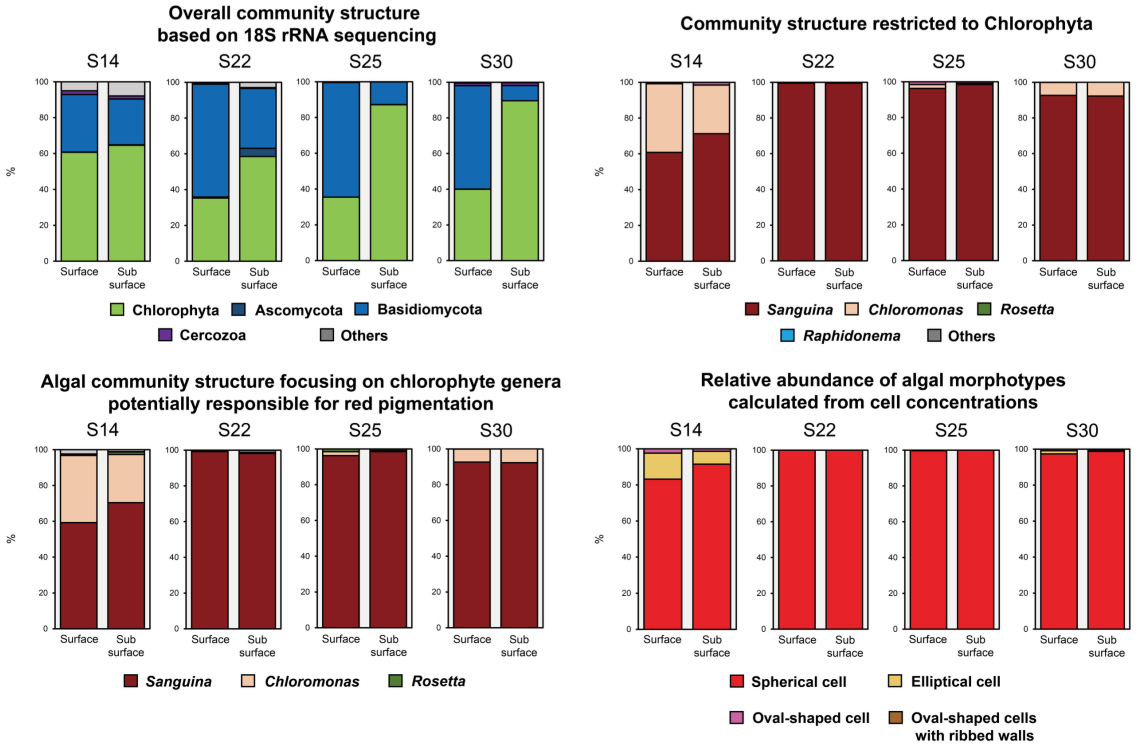



**Figure 7: Microbial community structure on the surface and subsurface in ice-based snowpack. Community structure is presented as stacked bar charts normalized to 100% for each site, comparing the snow surface (left) and snow subsurface (right) across four sites. For the molecular dataset, only phyla or genera accounting for >5% of the total reads were included.**

Section: 3.5, as highlighted above. I would like to see evidence that the methodological approach used in this study has been validated for this type of samples as higher P than N in snow samples is at least unexpected.

REPLY: As detailed in our responses to the two comments above, the sample tubes were rinsed twice with the corresponding filtered sample, and no detectable $PO_4^{3-}$ peak was observed in the Milli-Q water blanks. Moreover, comparable filtration and frozen-storage procedures have been used for phosphate measurements in previous studies of snow and red-snow samples, without a reported increase in $PO_4^{3-}$ attributable to freezing. We have added these methodological details and supporting references to the revised Methods.

Figures in overall: The study uses similar colours for different variables in various figures. For example, Fig. 5 and Fig. 6 use the same red colour which make the orientation harder for reader. I would suggest choosing colour-schemes to be unified for the same variables to make the orientation in figures easier.

REPLY: As shown in the revised figures described above, we changed the color schemes in Figures 5 and 6 to avoid using similar colors for different variables and to improve

readability. In contrast, the colors used in Figures 3 and 4 represent the same sampling-site categories and are already applied consistently across both figures; therefore, these color assignments were retained.

Discussion

L274–275: The sentence needs to be reworded slightly to make the meaning clearer. I would also recommend dividing it into shorter sentences to make it easier to read. I am not sure what information the authors wanted to convey to the reader, as both the soil-based site and S14 had a higher proportion of non-spherical cells and Chloromonas.

REPLY: We have divided the sentence into shorter statements and clarified that these sites showed both higher relative abundances of Chloromonas and Rosetta and a higher proportion of non-spherical cells, whereas the upper ice-based sites were characterized by Sanguina dominance and a greater proportion of spherical cells. Now you can read "Across the glacier valley, *Chloromonas* and *Rosetta* were relatively abundant at the soil-based site and the lower ice-based site, including S14. These sites also showed a higher proportion of non-spherical cells in the morphotype data. In contrast, *Sanguina* dominated at the upper ice-based sites, where spherical cells accounted for a greater proportion of the algal community (Fig. 6c, d)." in Lines 310–313.

L279: "Previous studies support this explanation." I think this sentence is unnecessary, particularly given the paragraph's logic. The authors discuss the fact that the spherical morphotype is taxonomically heterogeneous, yet they continue to refer to studies supporting the spherical cell type for Sanguina.

REPLY: We deleted it from the revised manuscript as suggested.

L284: "Therefore, morphology alone can merge taxonomically distinct algae into the same category, especially for spherical cells." In my opinion this sentence is redundant with L278.

REPLY: We agree that this sentence was redundant with the preceding discussion. We have therefore deleted it from the revised manuscript to avoid repetition.

L322: I miss the reference for the sentence on modulation of local growth.

REPLY: We have added a reference (Almela et al., 2024, "Laboratory Experiments Suggest a Limited Impact of Increased Nitrogen Deposition on Snow Algae Blooms," Environmental Microbiology Reports, 16, doi:10.1111/1758-2229.70052) supporting the potential influence of nutrient availability on snow algal growth. Because direct evidence

that nutrients regulate life-stage transitions in red snow algae remains limited, we also revised the sentence to avoid making this broader claim. Now you can read “However, nutrient availability may still influence local algal growth once cells reach the snow surface (Almela et al., 2024).” in Lines 355-356.

Sect. 4.4. As radiation can affect the migration of algal cells, I would like to see more information and data on light conditions during sampling, as this information could substantially contribute to evaluating the results. This is particularly important when samples are taken within four days at which the light conditions could potentially differ.

REPLY: Radiation conditions were not measured during the sampling , and we therefore cannot directly evaluate their effects on the vertical distribution of algal cells. However, the red-pigmented stages that typically dominate red-snow blooms are generally non-motile cysts or resting spores rather than actively swimming cells. This applies particularly to *Sanguina* and to the mature red cyst stages of many snow-inhabiting *Chloromonas* species (Procházková et al., 2019; Hoham and Remias, 2020). Therefore, short-term active migration of the abundant red cells observed in this study in response to radiation conditions during sampling was likely limited. Nevertheless, some snow algae possess motile stages during their life cycles. Moreover, molecular data do not allow us to determine the life stage of each detected taxon. We therefore acknowledge that radiation conditions before and during sampling could have influenced the distribution of some algal stages. We have added this limitation and revised the Discussion to avoid attributing the observed vertical patterns directly to active migration. Now you can read “The taxonomic composition also differed between the surface and subsurface layers. Although Sanguina was detected in both layers, the relative abundance of Chloromonas was higher at the surface. This pattern indicates taxonomic differentiation between layers but does not identify the mechanism responsible for the observed distribution. The red spherical cells associated with Sanguina represent non-motile cyst stages (Procházková et al., 2019; Hoham and Remias, 2020), making short-term active migration during sampling unlikely. However, motile stages of other taxa, passive redistribution with meltwater, and environmental conditions preceding sampling may have contributed to the vertical distributions. Although solar radiation, a critical factor shaping vertical microhabitats, was not directly measured during the sampling period and should therefore be considered when interpreting the vertical patterns, our findings suggest that vertical positioning is influenced by both physical transport and taxon-specific traits.

Overall, these results underscore that the subsurface snowpack serves as an essential, persistent seed bank that feeds surface blooms as snowmelt progresses. Because surface-

only observations risk underestimating total algal biomass and missing early-stage dynamics, incorporating subsurface biomass reservoirs into spatial models will be crucial for accurately predicting future red snow blooms and their feedback on glacier albedo." in Lines 397–409.