Review:

"Ice-nucleating particles active below -24 °C in a Finnish boreal forest and their relationship to bioaerosols"

Summary:

In their manuscript, Vogel and colleagues measured the ice nucleation activity of aerosols collected during a two-months field campaign in Hyytiälä, Finland, in spring 2018. They hypothesize that the measured ice-nucleating particles (INPs) active below -24°C originate from local biological sources. They measured INPs online, using a portable expansion chamber and compared it to offline droplet freezing measurements. The authors argue that the INPs below -24°C are derived from biological sources due to the alignment of their INP data with a parameterization of INPs in a pine forest in Colorado, a correlation with autofluorescing particles at sizes larger than 0.5 um and speculate that the negative correlation between relative humidity (RH) and INPs is connected to INPs originating from lichens. In addition, they observed no correlation between small particles and INP concentrations concluding that new particle formation is not the source of INPs.

Field studies like this are critical for the ice nucleation community. This study holds particular significance as biological INPs in large biosystems like the boreal forest are understudied, leaving much to be discovered. Thus, the work presented merits publication in Atmospheric Chemistry and Physics after addressing the following comments.

GENERAL COMMENTS:

- The manuscript lacks discussion on contrasting results from previous research, such as the study by Prenni et al., 2013, which indicates a correlation between increased biological INP concentration and high RH. The field study by Prenni et al., 2013 was conducted in a forest in Colorado, USA with the presence of pine trees as well. Since the authors argue in their manuscript that pine forests in Colorado and Finland can be compared (parameterization comparison), I miss the discussion about these contrasting results.

- Paramonov et al., 2020 measured INPs at the same research station in the same year and proposed that INPs were influenced by long-range transport, a result that is different from the outcome of this study. A more thorough discussion on the differences in outcomes between studies would enhance the manuscript.

- The authors speculate that the INPs originate from lichens. However, their argument, primarily based on the negative RH correlation and lichens not being covered by snow, warrants further elaboration. In general, biosystems are rather complex and the emission of bioparticles from the biosphere to the atmosphere depends on multiple factors (Pasanen et al., 1991). It is worthwhile noting that also certain fungal spores, like those from the genus *Cladosporium*, are released into the atmosphere at low RH (Sabariego et al., 2000). Further, *Cladosporium* have been documented to nucleate ice at temperatures consistent with those presented in this study (Iannone et al., 2011). Additionally, besides lichens being not covered by snow, INPs could also originate directly from trees. Seifried and Reyzek et al., 2023 discovered biological INPs on the surface of Scots pines (bark and branches) capable of nucleating ice even below -25°C. Kokkila et al., 2002 mentions that Scots pine are the dominant tree species at the Hyytiälä research station. These points should be included in the discussion.

SPECIFIC COMMENTS:

Abstract:

- Line 6: To provide a clearer overview, I recommend inserting numbering (i), (ii), (iii) before each argument supporting the hypothesis that INPs below -24°C originate from biological sources.

Introduction:

- The Introduction is well written and the state of the art is well described.
- Line 37: Typo "rel~~a~~y"
- Line 50: Typo "bioaerosols" instead of bio aerosols

Methods:

- I suggest including a brief description or equation explaining how c_INP (L^{-1}) is calculated to ensure that readers understand it is measured per litre of air rather than per litre of liquid volume, as reported for some immersion freezing assays.
- Line 103: Typo "ETH" instead of EHT
- Line 122-124: missing space between units "L min^{-1}"
- Line 125: missing space between number and unit "6 min"
- Line 140: What is nanopure water? Please provide more details about the water used in the offline ice nucleation assays.
- Line 140: The freezing data is presented in °C in all graphs, so I recommend maintaining consistency by using the same temperature unit throughout the manuscript, including for the cooling rate. In addition, there is a space missing between the units "°C min^{-1}"
- Line 141: Consider rephrasing the sentence as not the well itself freezes but the content. Maybe: "[…] to a temperature at which all droplets freeze."
- Line 143: change unit for cooling rate
- Table 1: Last column, last cell: What do you mean by "end"? May 11, 2018?
- Line 156: add excitation and emission wavelengths

Results & Discussion:

- The months April and May are renowned for their elevated pollen concentrations, with species such as *Betula pendula* exhibiting notably high levels, especially towards the end of May in the southern regions of Finland (see e.g. Manninen et al., 2014). Could these pollen have contributed to the INP population?

- Figure 2, caption: Put listing items (a), (b), etc., before the sentence to match the style used in Figure 3.
- Figure 6 and line 284: It is not explained in the manuscript what FP3 for WIBS measurements is referred to. Please add the details.
- Line 285: missing space between the comma and “which”
- Line 295: consider rephrasing from “large concentrations” to “high concentrations”
- Line 316: Do you mean at around 14:00 (2 pm)? If so, use style hh:mm (same for time in line 335)
- Line 352: Typo “Figure 9” instead of “figure 9”

Conclusion:

- Are the aerosols after PINE collected and could be measured in the future with offline spectrum to obtain the entire freezing spectrum from the same aerosol population?

References:

Prenni, A. J., Tobo, Y., Garcia, E., DeMott, P. J., Huffman, J. A., McCluskey, C. S., ... & Pöschl, U. (2013). The impact of rain on ice nuclei populations at a forested site in Colorado. Geophysical Research Letters, 40(1), 227-231.

Paramonov, M., Drossaart van Dusseldorp, S., Gute, E., Abbatt, J. P., Heikkilä, P., Keskinen, J., ... & Kanji, Z. A. (2020). Condensation/immersion mode ice-nucleating particles in a boreal environment. Atmospheric Chemistry and Physics, 20(11), 6687-6706.

Pasanen, A. L., Pasanen, P., Jantunen, M. J., & Kalliokoski, P. (1991). Significance of air humidity and air velocity for fungal spore release into the air. Atmospheric Environment. Part A. General Topics, 25(2), 459-462.

Sabariego, S., Díaz de la Guardia, C. & Alba, F. (2000). The effect of meteorological factors on the daily variation of airborne fungal spores in Granada (southern Spain). Int J Biometeorol 44, 1–5. https://doi.org/10.1007/s004840050131

Iannone, R., Chernoff, D. I., Pringle, A., Martin, S. T., & Bertram, A. K. (2011). The ice nucleation ability of one of the most abundant types of fungal spores found in the atmosphere. Atmospheric Chemistry and Physics, 11(3), 1191-1201.

Seifried, T. M., Reyzek, F., Bieber, P., & Grothe, H. (2023). Scots Pines (Pinus sylvestris) as Sources of Biological Ice-Nucleating Macromolecules (INMs). Atmosphere, 14(2), 266.

Kokkila, T., Makela, A., & Nikinmaa, E. (2002). A method for generating stand structures using Gibbs marked point process. Silva Fennica, 36(1), 265-277.

Manninen, H. E., Bäck, J., Sihto-Nissilä, S. L., Huffman, J. A., Pessi, A. M., Hiltunen, V., ... & Petäjä, T. (2014). Patterns in airborne pollen and other primary biological aerosol particles (PBAP), and their contribution to aerosol mass and number in a boreal forest. Boreal Environment Research, 19, 383.