Author's response to Referee #2

We thank reviewer #2 for the valuable comments on our manuscript. Below we provide detailed answers to the individual point giving in black the original comments, in red our answer and in blue the changes in the manuscript.

Specific comments

1) The time series of INP data measured using the PINE chamber are unique and potentially valuable. However, this work appears to repeat essentially the same work as reported by Schneider et al. (2021) and Brasseur et al. (2022). I strongly encourage that the authors reconsider if they can add new topics and findings using their data sets. Then, the authors should significantly update the contents of the manuscript in order to clarify the difference between this study and other earlier studies.

We disagree that we report that same work as the two previous papers. Schneider et al., 2021 measured at temperatures higher than -25°C, where it is known from literature that the INP population is often dominated by biogenic aerosol in terrestrial environments. Furthermore, they present daily filter measurements to evaluate a day-by-day and also seasonal variability. In our manuscript we present measurement at temperatures lower than -24°C, the temperature range that was not covered by Schneider et al., 2021 and our data are given in a time resolution of 6 min, allowing us to average over 1h and by that investigate an hour-by-hour variability, which is driven by different factors than the long-term variability. The different driving factors are highlighted by (1) the fact that the Schneider et al., 2021 parameterization does not fully represent our data, and overestimates the INP concentration especially for lower temperatures that were not covered by their study and (2) very low correlation coefficient with the ambient air temperature, which Schneider et al., 2021 showed to be high for their measurements. Brasseur et al., 2022 compare four selected days, for a time of maximum 12h, the INP concentrations measured by the various instruments with various parameterizations. Their Measurement Report was meant to give an overview of the campaign rather than a full analysis. We apply the parameterization on two months of continuous measurements and by that present a wider variability. For this reason, we think that it is essential to provide the evaluation of the parameterizations on the full dataset. In order to emphasize the importance of our manuscript, we added a paragraph at the end of the introduction in L. 60:

The study on the same campaign presented by Schneider et al. (2021) focused on the temperature range higher than -25 °C and seasonality, while here we present measurements at temperatures below -24 °C and investigate the hourly variability. Paramonov et al. (2020) performed measurements in the same temperature range, but over a different time period. Their measurements were from end of February 2018 to beginning of April 2018, whereas ours were from mid-March until mid May, including the time of snow melt and early spring. Only

four days of our measurements are included in the measurement report from Brasseur et al. (2022), challenging several literature parameterizations. In our present manuscript, we extend this analysis for the full 2 months to better understand the variability of the dataset.

2) In my understanding, cloud-chamber-type instruments other than PINE (e.g., SPIN, PINC) had also been used to measure the time series of INPs active below -24 degree C at this site during the HyICE-2018 campaign. The authors should compare the difference between the PINE and the other instruments in the lower temperature regime and evaluate the strengths (and limitations) of the PINE. For example, to my knowledge, Paramonov et al. (2020) already reported the full INP data sets active around -30 degree C measured using a PINC during the overlapping period.

During the HyICE-2018, PINE, PINC and SPIN measured the INP concentration at temperatures lower than -24°C. However, there are substantial differences in the data coverage and availability so there we have not done a full comparison on all data. There were a few occasions within HyICE-2018 where we purposefully performed intercomparison tests where we all aimed at reporting in overlapping temperatures and times. These are reported in Brasseur et al. (2022) and it would not be appropriate to repeat those intercomparisons here. That said, we have added the data from PINC in figure 2a based on comment 13. SPIN covered the same time period as PINE, but data are only available for the 2 days that were part of the intercomparison of Brasseur et al., 2022. Other data are not publicly available. PINC measured from end of February until April 2 and by that covered the time period where everything was covered in snow. In contrast to that, PINE provided data during the snow covered period, the transition period and the snow free period. By that PINE covers more variability in potential aerosol sources, especially from local emissions. As mentioned in our answer to comment 1), we provide a detailed discussion in the revised manuscript. We also added the data of PINC in figure 2 and changed the according discussion. We don't think that it is valuable to add the SPIN data in this figure, because the two available days are not representative.

L. 168 … and CFDC measurements of PINC from the same campaign (Figure 2a)...

L. 172: PINE and PINC data overlap at the lower end of the covered temperature range and fully agree in the span of measured INP concentration.

3) Please explain more details for the WIBS data. It is unclear how the WIBS fluorescent particle data were applied to the Tobo et al. (2013) (2) parameterization. As a result, it is difficult to judge whether the authors' explanations that "this means that the measured INP concentration ~ from different sources (Lines 250-252)" and "a second parameterization proposed by Tobo et al. (2013), linking ~ but they are not identical (Lines 373-376)" are indeed reasonable. For example, Tobo et al. (2013) used the data on fluorescent particles larger than 0.5 um measured using UV-APS that employs an excitation wavelength of 355 nm and detects laser-induced

fluorescence spectra in the range of 420–575 nm). Did they use the same setup as the UV-APS?

The WIBS-neo used for the measurements of fluorescent particles during HyICE-2018, has two excitation laser and two detection bands. The wavelength of laser 2 (370nm) is close to the wavelength of the UV-APS laser. The detection band of the UV-APS falls within the second detection band of the WIBS of 420nm – 650nm, but is missing the higher wavelengths. The size cut of both instruments is 0.5 um. We are aware, that the two instruments have their differences, but we still think that it is worth to challenge the Tobo 2013 parameterization based on the total concentration of fluorescent particles, since the measurement environments have similarities. We added in the methods section more details of the WIBS, and discuss this specific parameterization keeping the differences between the two instruments in mind.

L. 150 ff: WIBS (Wideband integrated bioaerosol sensor) measures particle fluorescence with two excitation lasers (wavelength of 280 nm and 370 nm), and emission is monitored in two detection bands (310 nm to 400 nm and 420 nm to 650 nm). ...

L. 158: WIBS detected particles with an optical diameter larger than 0.5 µm.

L. 226: ... Tobo 2013 (2) uses the concentration of fluorescent particles with a diameter larger than 0.5 µm measured with a UV-APS with an excitation wavelength of 355 nm and an emission wavelength range between 420 nm and 575 nm ...

L. 228: Even though fluorescent particle measurements to develop the Tobo 2013 (2) parameterization were done with a different instrument, it is interesting to apply this parameterization on our data due to the similarity of the measurement environments. Due to the setup of WIBS it provides measurement of seven different combinations of excitation wavelength and detection band wavelength, whereas the UV-APS provides only one channel. By that the WIBS can capture more various bioaerosols and the INP concentrations predicted by Tobo 2013 (2) should be seen as an upper estimation.

L. 249: As discussed before, this is the upper estimated of the INP concentration due to the differences in UV-APS and WIBS, and the predicted INP concentrations can be even lower.

4) Brasseur et al. (2022) announced that "forthcoming studies would explore atmospheric vertical profiles of INPs, INP sources and transport modeling, plausible links between INP abundance and NPF, and the ice nucleation activity of boreal biology such as flora and fungi". If this work is a part of the forthcoming studies, the authors should present some more details for the comparison of INPs with NPF. Since the information on the correlation coefficients only (Figure 6) is obviously insufficient, the authors should show the time series of the INP and NPF data.

We originally took the approach of keeping the manuscript succinct, but see the value of this additional analysis. We have expanded on this topic with an additional section and an additional figure:

3.4.1 INP concentrations during new particle formation events
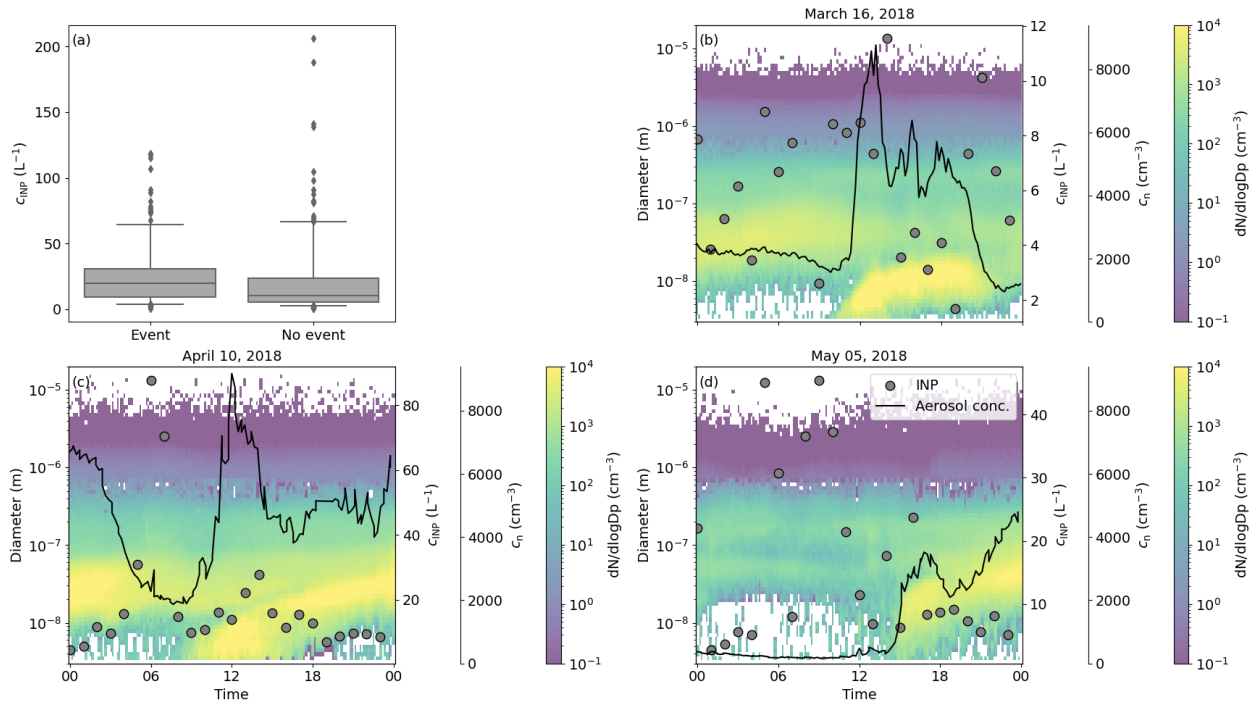
Figure 7. Overall comparison of the INP concentration during NPF events and outside NPF events together with three case studies. (a) Boxplot of the INP concentration on days flagged NPF event days and days without an NPF event. The borders of the box represent the 25th and 75th percentile, the horizontal line inside the box the median, the whiskers the 5th and 95th percentile and the single points all the data outside of that. (b), (c) and (d) are three cases (March 16, April 10 and May 5) of NPF events of category Ib, where the background of each panel shows the particle number size distribution, with the diameter in the left y-axis and dN/dlogDp as the colorbar. The first right y-axis is the INP concentration and the second right y-axis the aerosol number concentration measured by the DMPS and APS. Please note that the y-axis of the INP concentration varies, due to the high variability of INP.

The low correlation between INPs and the concentration of small particles (Fig.6) suggests that particles formed during new particle formation (NPF) events do not contribute to the INP population. During NPF events, gaseous species nucleate to form a critical cluster which then grows via gas to particle conversion to particles of tens of nanometers in diameter over the course of hours Kulmala et al., 2001. These newly formed particles are very high in number concentration and can be detected based and the aerosol size distribution and the increase in aerosol number concentration. SMEAR II is known for its NPF events and the analysis and categorization of these events is based on Dal Maso et al., 2005, who introduced category Ia, Ib

and II events, which designates the intensity of an event. The occurrence of NPF days during HyICE-2018 was already presented in Brasseur et al., 2022, where individual days were flagged as NPF days, independent of their category. To investigate the connection between NPF events and INPs, we flagged the hourly INP data measured with PINE at a freezing temperature between -27 °C and -30 °C with the different NPF event categories. In case none of the three NPF categories was detected, we set the flag to no NPF event. By comparing the INP concentration on NPF event days (independent of the category) and non-NPF event days (Fig.7a), a similar median and variability of the INP concentration is seen in the two boxes. This indicates that NPF events have no effect on the INP population.

In order to explore the NPF events in more detail, we plot time series of PINE INP measurements alongside aerosol concentrations and the particle number size distribution on three days, one from each month of the campaign. The case on March 16 (Fig.7b) shows a steep increase in the aerosol number concentration at 12:00 local time from about 2000 cm-3 to approximately 8500 cm-3, which characterizes the start of a NPF event, which is also visible in the high concentration of small particles at the same time. The aerosol concentration decreases to the initial value at 23:00 local time. During the whole day the INP concentration is scattered between 2 L-1 and 12 L-1 without showing a distinct pattern. In the other two cases on April 10 and May 5 (Fig.7c and d) the aerosol concentration also shows a steep increase when NPF sets in and lower aerosol concentration outside the NPF event. In these two cases the INP concentration reaches higher values during phases of lower aerosol concentrations and drops when NPF is initiated. This may be because NPF events are favored when the aerosol surface area concentration is low, which is also when the INP concentrations would generally be expected to be lower. This analysis strengthen the conclusion that particles formed during NPF events in the size range up to 50 nm in diameter do not contribute to the INP population.

5) The possible links between INPs and low RH are potentially interesting. On the other hand, many earlier studies have reported elevated INP concentrations under high RH conditions at a forested site in Colorado (Huffmann et al., 2013; Prenni et al., 2013; Tobo et al., 2013) and other locations (Wright et al., 2014). I would suggest showing figure(s) that could explain the relationship between INP and RH data. Then, please discuss the discrepancy between this study and the above studies if there was indeed a clear negative correlation between the INP and RH data.

We have added an addition section to cover this in more detail and place the result in the context of the literature:

3.4.2 The link between high INP concentrations and low relative humidity
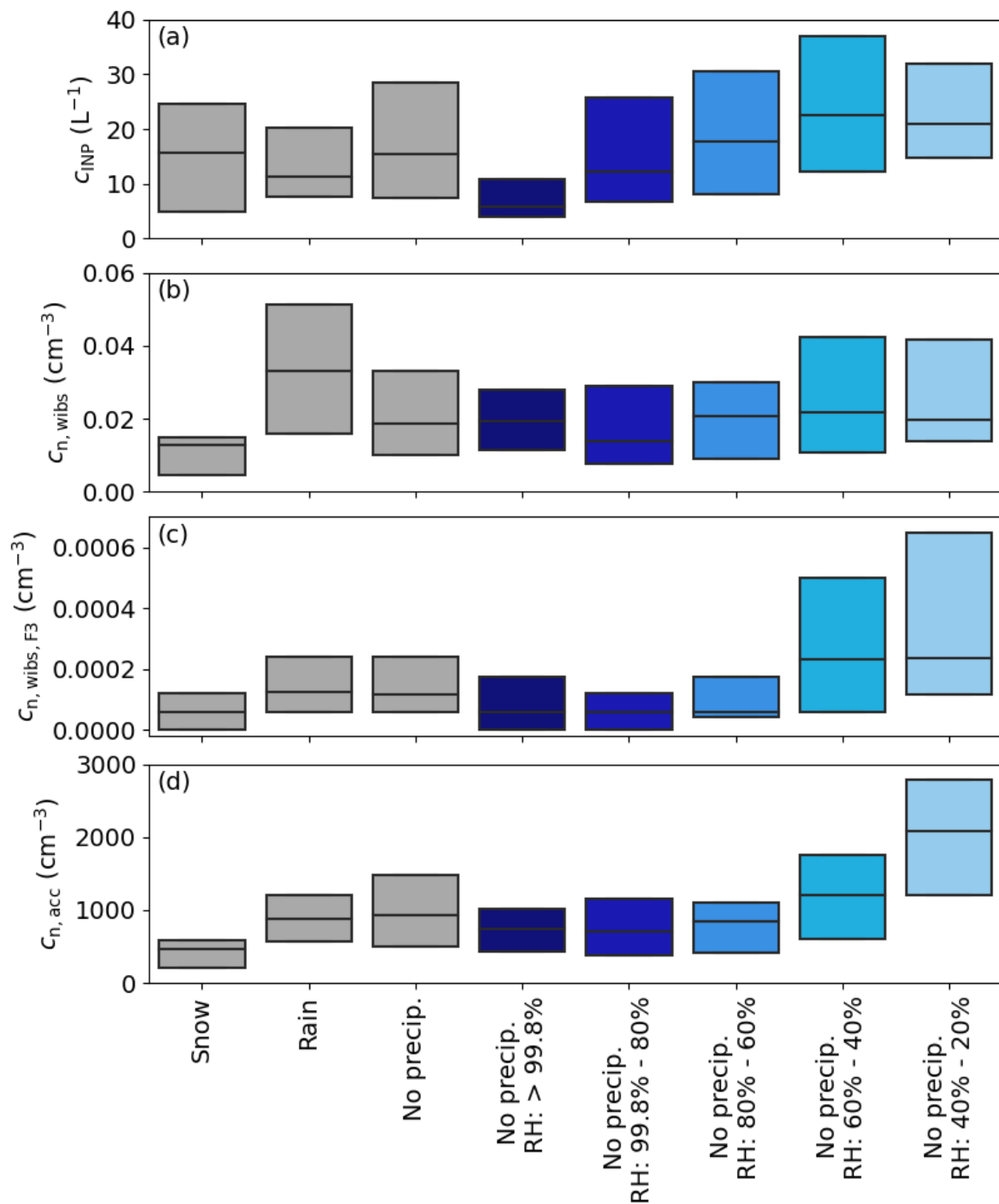
Figure 8. The relationship between aerosol, precipitation and humidity. (a) The INP concentration, (b) fluorescent particle concentration measured by WIBS, (c) fluorescent particle concentration measured in the F3 channel of WIBS and (d) the particle concentration in the accumulation mode are divided into several precipitation and relative humidity categories. The precipitation is split

into cases of snow fall, rain fall and no precipitation (including also no snowfall). The data of the 'no precip.' category were again divided into different relative humidity (RH) classes, which are RH > 99.8 % (saturated conditions, considering some uncertainty of the measurements), 99.8 % > RH > 80 %, 80 % > RH > 60 %, 60 % > RH > 40 %, 40 % > RH > 20 %. The boundaries of the boxes are the 25th and 75th percentile. For clarity, whiskers and outliers are not shown.

In Section 3.4 we discuss the observation of a negative correlation between the INP concentration and the relative humidity (RH), which is potentially linked to an increased release of bioaerosol from lichen (Armstrong, 1991; Tormo et al., 2001). To better understand potential connections between the INP concentration, relative humidity and aerosol sources, the data are grouped based on precipitation into the three categories snow fall, rain fall, and no precipitation (snow fall and rain fall). The data of the last category are further split into five different RH ranges: > 99.8 % (which considers saturated conditions including the uncertainty of measurement), 99.8 % > RH > 80 %, 80 % > RH > 60 %, 60 % > RH > 40 %, 40 % > RH > 20 % (Fig.8a). (Fig.8a) reveals no substantial change in the INP concentration between snow fall, rain fall, and no precipitation. However, when dividing the data points without precipitation into the RH categories, the lowest INP concentrations with a median of 5 L−1 are measured for the highest RH. For the following groups the median INP concentration increases until it reaches 20 L−1 at a RH of 60 % and lower. A similar trend with RH is observed for the aerosol concentration in the accumulation mode and the WIBS F3 channel. The WIBS F3 channel can point to increased concentration in NAD(P)H (Savage et al., 2017), a co-enzyme linked to energy metabolism in cells and is an indication for living biological organisms. In contrast, the total fluorescent particles concentration of WIBS showed no clear increase in the mean concentration with decreasing RH (Fig.8b). In other locations fluorescent bioaerosol concentrations were found to increase markedly with increasing RH, which may be related to RH-dependent fungal spore release mechanisms (Toprak and Schnaiter, 2013; Gabey et al., 2010; Timothy P. Wright and Petters, 2014). Hence, the bioaerosol released in the Boreal forest of Southern Finland appears to be different to other locations. The WIBS results and the correlation with RH are consistent with release of bioaerosol particles from lichens. Lichens can reproduce asexually through the production of diaspores. These diaspores contain living components of lichen, the mycobiont and photobiont, that can then colonise new locations (Hale, 1974). These diaspores would therefore contain NAD(P)H since they are metabollically active. In addition, as mentioned above, release of diaspores has been shown to be negatively correlated with RH (Armstrong, 1991; Tormo et al., 2001). Hence, the hypothesis that lichen derived bioaerosol contribute to the INP population is consistent with both the F3 channel (NAD(P)H of WIBS and the negative RH dependence.

A remarkable feature of Fig.8 is that for the fluorescent particle concentration, the WIBS F3 concentration and the aerosol concentration in the accumulation mode, the lowest values are

detected during snowfall, while for the INP concentration this is not the case. This implies that many aerosol types are preferentially removed during snow fall events, but the INP concentration is not affected to the same extent. When snowing, the ambient RH would be expected to be well below 100% (with respect to water), for example, if the air were at ice saturation the RH would be about 90% at -10°C, conditions under which we might expect to start to see lichens producing aerosol. However, the low bioaerosol concentration from the WIBS is inconsistent with this idea.

During rain fall, an enhanced concentration of fluorescent particles is measured by WIBS, however it is not reflected in the INP concentration. Previous studies reported an increase of INPs during and after rain events during measurements in Colorado in summer. The highest increases in fluorescent particle and INP concentrations were observed during intense rain events with rain rates more than 3 mm/5min (Tobo et al., 2013). Less pronounced increases were observed during less intense events and at lower measurement temperatures, which are equal to our covered temperature range (Huffman et al., 2013; Prenni et al., 2013). The overall rain rate during our measurement period was about 0.03 mm/5min and by that two orders of magnitude lower than during the measurements in Huffman et al. (2013); Prenni et al. (2013); Tobo et al. (2013). Moreover, they report measurements in summer, when rain fell on dry soil and by that released the biological particles. During the HyICE-2018 campaign, the soil was always covered by snow or wet, hence release of biological particles might have been hindered.

Technical comments

6) I would suggest changing the title appropriately, because other related literatures (Paramonov et al. 2020; Brasseur et al., 2023) have already reported INP data below -24 degree C during the HyICE-2014.
Our strong preference is to keep the title. It contrasts strongly with the title used by Paramonov (Condensation/immersion mode ice-nucleating particles in a boreal environment) and emphasizes the different conclusion (i.e. bioaerosols). It is also very different to the title used by Brasseur: 'Measurement report: Introduction to the HyICE-2018 campaign for measurements of ice-nucleating particles and instrument inter-comparison in the Hyytiälä boreal forest'

7) Please indicate the full name and the abbreviated name in the same manner (e.g., see "Portable Ice Nucealtion Experiment (PINE)" in Line 59 and "PINC (Portable Ice Nucleation Chamber)" in Line 104.
We changed the abbreviations of PINC and PINCii in L.103-104 as suggested
... Portable Ice Nucleation Chamber (PINC, operated by ETH Zurich; Kanji et al. (2013)) and Portable Ice Nucleation Chamber ii (PINCii, operated by University of Helsinki and University of Gothenburg ...

8) Please explain some more details for the experimental setup for the PINE. For example, what is the cut-off diameter of the inlet?

Section 2.2 contains the results of the inlet characterization of PINE in L. 107 ff, that were performed during the campaign. Also, the overview paper (Brassuer et al., 2022) contains many of the details around the experimental setup. We rephrased the pertinent sentence to that points out the cut-off diameter test was done specifically for the PINE inlet and not for the inlet in general.

L. 108: An additional impactor was not installed. The PINE inlet was characterized onsite using an OPC (MetOne, GT 526S) directly at the aerosol inlet and at the entrance of PINE to compare the number concentration in five different size bins of particles behind the inlet with those entering PINE. With that the transmission efficiency was calculated, which showed ...

9) I assume that the PINE uses an OPC that can detect larger aerosol particles, liquid cloud droplets and ice crystals as single particles (Line 84), but the size range covered by this OPC is not clear. It is also unclear how the PINE used here discriminated ice crystal signals from ambient aerosol particles and liquid droplets?

We agree with the reviewer, that additional information is valuable for the reader and added them in L. 85 ff.

The OPC detects larger aerosol particles, liquid cloud droplets and ice crystals as single particles, based on the size and shape dependent scattering signal. Due to their spherical shape, liquid cloud droplets are detected with their actual size, while the aspheric ice crystals show a higher scattering signal and are thus detected a larger diameter than cloud droplets. Aerosol particles have been proved to be detected at smaller diameters than ice crystals and can thus be differentiated from ice crystals. A detailed discussion can be found in Möhler et al. (2021). During the HyICE-2018, the OPC was set to measure particles with a diameter between 0.6 μm and 40 μm.

10) Lines 96-97: Please explain if the INP number concentrations presented here were corrected to the values at standard temperature and humidity conditions (0 degree C and 1 atom) or not.

The INP concentrations presented in the manuscript are given in stdL^-1. We added a sentence in L. 97.

... and corrected for standard conditions (i.e. all ice-particle concentrations are reported at standard temperature and pressure).

11) Line 99: What do you mean by "the uncertainty for PINE is given as 20 %"?

We intended to say that the uncertainty of the INP concentration is given as 20%, with further explanation given in Möhler et al., 2021. The sentence in L. 99 was adjusted to the following:

The uncertainty for the INP concentration is given as 20 % (Möhler et al., 2021) and is not displayed in the following figures to keep them clear.

12) Lines 111-115: Please specify the RH setup for mixed-phase cloud conditions in the PINE chamber.

We have added the following to explain that the RH in the chamber is defined by the presence of cloud droplets.

L. 115 ff: In this campaign we operated PINE under mixed-phase conditions, that is we always generated a liquid cloud and counted the number of ice crystals that grew out of the liquid cloud. During an expansion, the temperature inside the cloud chamber is lowered around 6 °C within 40 s. It is evident that water saturated conditions are reached with the formation of a liquid cloud. Any ice crystals that form during the expansion grow rapidly in the strongly ice-supersaturated environment. In contrast to other instruments, the relative humidity during the expansion was not directly measured since the appearance of droplets defines the RH in the chamber.

13) Figure 2a: I would suggest including INP data derived from SPIN and PINC during the HyICE-2014 campaign.

The PINC data are available in Paramonov et al., 2020, so we included them as a grey bar in figure 2a. SPIN data are only available for a few hours for two days from Brasseur et al., 2022, so they do not represent the full range of INP concentrations measured with SPIN.

14) Lines 262-264: Please explain how the PINE chamber captures deposition and condensation nucleation in the Methods section.

Deposition nucleation occurs while water saturation is not exceeded. In the case of the measurements presented here, this only includes a few seconds before cloud formation sets in and would be seen as an ice signal before the formation of the liquid cloud. While in principle PINE can be used below water saturation, we do not do it here hence have not included an explanation in the methods. In the statement in L. 262-264, we wanted to imply that with PINE we are not limited to one freezing mechanism in contrast to INSEKT.

15) Lines 271-272: Please revise this description if the temperatures were between -27 and -30 degree C as explained in the Figure 6 caption.

We changed the sentence in L. 271-272 to the following:

To further investigate potential connections between ice nucleation activity and meteorological variables and aerosol properties at temperatures between -27 °C and -30 °C, correlation coefficients are calculated.

16) Line 284: What do you mean by "FP3 channel"? Please also check my comment 3.

Also reviewer #1 commented on that and we inserted an explanation in L. 287 ff.

An aerosol particle entering WIBS gets excited by two lasers with a different wavelength and can be detected in two different detection bands. Depending on its composition, fluorescence is detected in only one or more laser detection band channels. The F3 channel is fluorescence

triggered by the 370 nm laser and detected in the 420 nm to 650 nm band. Particles detected only in this channel can be assigned to NAD(P)H, which is a tracer for the viable biological fraction (Savage et al., 2017).