Response to the interactive comment by Referee #1:

We thank referee #1 for his or her thoughtful comments and feedback. Please find below our responses and suggestions for the manuscript revision, with the referee comments in black, our answers in red, and suggested changes and/or additions to the manuscript in blue.

This study shows the seasonal cycle of immersion-mode ice nucleating particles (INPs) in boreal forests in Finland. Since few studies have reported the seasonal cycle of atmospheric INPs in a forested site based on year-round measurements, the INP datasets presented here are unique and will be valuable for atmospheric science communities. This study also suggests that the seasonal cycle of INPs would be attributable to biogenic aerosol particles and provides two new INP parameterizations related to ambient temperature and ice nucleation active site (INAS) approach. However, since evidences to support these points are insufficient, the authors should conduct an indeep discussion based on other available datasets (e.g., meteorological data, WIBS and L-RoF-AMS data) to quantify the possible contributions of biogenic aerosol particles to INP populations. I would like to support the publication if the authors can answer the following requests/comments.

Major Comments:

1) Further detailed discussion is necessary to investigate a possible relationship between INPs and primary biological aerosol particles (PBAPs). For example, this study uses ambient temperature data as a proxy of changes of the season. As illustrated in Figure 4, it seems that the variation of INPs active at given temperatures are somewhat related to that of ambient temperatures. On the other hand, the comparison with other meteorological parameters is not performed, despite the fact that many earlier studies (e.g., Heald and Spracklen, GRL 2009; Hoose et al., ERL 2010; Huffman et al., ACP 2013; Wright et al., Aerosol Sci. Technol. 2014) have suggested that humidity would be a key factor influencing the release of PBAPs and/or INPs. In addition, as cited in the Introduction section of this paper, rain events might also be an important factor. The authors should conduct further comparison with other meteorological parameters.

We agree to the referee that correlations to other parameters may be worth considering. In fact we have already explored more than shown in the first manuscript version, but did not find strong correlations. Nevertheless, we have followed the referees suggestion and extended Figure 4 (in the new version, this is Figure 5) by including three additional panels showing the time series of relative humidity (Fig. 5c), wind speed (Fig. 5d) and precipitation (Fig. 5e) and ajusted the description accordingly. We included the following section in the manuscript:

Figure 5c shows the comparison of the INP time series with the time series of relative humidity (RH) measured 35 m above ground. Over the entire time period, no clear relationship between RH and INP concentrations is observed. For a shorter time period from June 2018 to September 2018, there seems to be some correlation of the INP concentration with the RH, during which the peaks in INP concentration in June corresponds to a RH peak. In Figure 5d, we compare the time series of INP concentrations with the time series of wind speed measured 34 m above the ground, which is also above the forest canopy. A relationship between measured INP concentrations and wind speed is not observed. In Figure 5e, the time series of INP concentration is compared to the occurrence of precipitation. Although other studies like Prenni et al. (2013), Huffman et al. (2013) and Iwata et al. (2019) report increasing INP concentrations during and after rain events in forested sites, we do not consistently observe this behavior. In this respect, increased INP concentrations are observed only during two of the strongest precipitation events in June and September 2018. However, it should be noted that in the cited studies INP concentrations were measured with higher time resolutions from minutes to hours. Huffman et al. (2013) reported increased INP and biological particle concentrations during rain events and up to one day after rain events. With our sampling (and therefore averaging)

time of 24 hours or more, rain-induced enhancements of INP concentrations may have been missed. Therefore, this type of sampling strategy may not be appropriate to deterministically link INP concentrations with rain events.

We do not exclude that the INP concentrations measured in the boreal forest are influenced by precipitation events for the whole year. However, our sampling set up is not appropriate to investigate the relationship more precisely.

2) In addition to the meteorological parameters and snow coverage/depth, the authors might need to check indicators showing the seasonal cycle of vegetation in/around the monitoring site (if available). For example, some earlier studies have used leaf are index (LAI) data as a measure of vegetation density (e.g., Heald and Spracklen, GRL 2009; Hoose et al., ERL 2010).

For our measurement site at SMEARII, the vegetation indexes NDVI (Normalized Difference Vegetation Index) and PRI (Photochemical Reflectance Index) are available, but only from 11 March 2018 to 11 September 2018. The NDVI and PRI data and the following interpretation is provided by Jon Atherton and Pasi Kolari from University of Helsinki (personal communication, October, 2020). For both vegetation indices, we calculated daily average values, which are averaged between 12:00 and 13:00 Finnish time, because the solar angle plays an important role for these indices. Unfortunately, LAI is not available. NDVI mainly tracks the development and loss of green leaf material including overstory trees and understory shrubs. We observe a trend in NDVI over the seasonal transition as the canopy slowly greens from winter to summer. During summer, when the color of the canopy does not change significantly, the NDVI remains relatively constant. The increase in NDVI in the transition period from winter to summer is observed some days later than the increase in INP concentrations. The increase in INP concentrations is nicely represented by increasing air temperature and melting snow in early April 2018 (see Figure 4), whereas a strong increase in NDVI is observed in the second half of April. It has to be considered, that the NDVI is affected by snow resulting in smaller NDVI values. The PRI measures the abundance of photoprotective pigments called carotenoids, which help the tree to deal with excess light, which is potentially harmful. We observed a PRI increase in May, which indicates a carotenoid change of the trees. A relationship to the increasing INP concentrations in April is not observed.

We do not think that these comparisons are of great benefit to the story outlined within the manuscript and have decided not to include the comparison in the paper. However, we are glad it can be made available in this publicly accessible discussion and gratefully acknowledge the support of Jon Atherton and Pasi Kolari by providing and interpreting the NDVI and PRI data.

3) After checking the points described in the above Comments 1 and 2, I would like to suggest reconsidering the INP parameterization related to ambient temperature (equation 1). I admit that this parameterization seems to successfully reproduce the INP number concentrations at this site as compared with other existing parameterizations. However, it is skeptical if this parameterization is scientifically meaningful, while it may be technically useful for reproducing only INP number concentrations near the ground level of this site. Given that this study emphasizes the possible contribution of PBAPs to INP populations, the development of parameterizations related to PBAPs and/or factors influencing the release of PBAPs would be more scientifically valuable. For example, given that previous model studies have used humidity and LAI to simulate the release of PBAPs, it might be interesting to evaluate if these parameters would also be useful for developing new INP parameterizations.

As the time series of vegetation indices available for SMEAR II do not show the same pattern as the time series of INP concentrations, and RH only correlates with INP concentrations in summer months, we do not get a better parameterization of INP concentrations using these parameters. Moreover, ground-level ambient air temperature represents the variability in measured INP concentrations better than other meteorological parameters or the vegetation indices. We are aware, that a physical mechanism that could explain this behavior remains unclear and that this parameterization is only empirical. We report this relationship simply as an observation and provide a description that technically describes INP concentrations quite well in this environment. We also see this result as a motivation for further studies and measurements regarding these findings.

4) As for the datasets during the HyICE-2018 campaign, the WIBS data would be potentially powerful, because these data might be useful as an indicator of biogenic aerosol particles. Unfortunately, however, the explanations and discussion on these data are insufficient. The authors should add detailed explanations and discussion regarding the WIBS data. For example, the WIBS has multiple channels as illustrated in Figure A2, but I could not find any detailed explanations and discussion about these data in the main text. I assume the categorization, such as FL1, FL2, and FL3, is based on Savage et al. (AMT, 2017). If so, the authors should clarify the source of this definition and then clearly explain the characteristics of each channel provided in Figure A2 (for example, what materials are expected to be detected using FL1, FL2, FL3, and their combinations?). In addition, the authors may need to discuss what channel was most sensitive to the variations of INPs.

Thanks for this comment. We agree that a more detailed description of the WIBS instrument and a more thorough discussion of the WIBS data is important to underpin the results of this study. Therefore, we have added a more detailed description of the WIBS instrument to Section 2.3. "Additional Instrumentation at SMEARII", which addresses the question and suggestion about the WIBS and also addresses the associated comments of the other referees.

The WIBS-NEO (Droplet Measurement Technologies, Longmont, CO, USA) is a bioaerosol sensor that provides information on the fluorescence properties, size and asphericity ratio of individual aerosol particles. It operates with an inlet flow of 0.3 l min^{-1} and detects particles with diameters between 500 nm and 30 µm. From 11 March 2018 to 2 April 2018, the WIBS was located about 50m from the aerosol filter sampling line used for the INP analysis. There, it was attached to a total aerosol inlet, which is characterized in Vogel (2018). On 3 April 2018, the WIBS was moved and installed directly next to the filter sampling line and attached to a PM10 inlet, which is described in Schmale et al. (2017). For the WIBS data analysis, particles from 0.5 µm to 10 µm were considered. To analyse the fluorescence of the particles, the WIBS sensor utilizes two xenon flashlamps as excitation light sources (optically filtered at wavelengths of 280 nm and 370 nm) and two emission detection channels (wavelength bands 310 – 400 nm and 420 – 650 nm). Optical size information is acquired utilizing elastic scattering from a continuous wave laser with a wavelength of 635 nm and a photomultiplier tube located orthogonally with respect to the laser. The excitation pulses are fired into the sample volume at different times and both detection channels record the emission(s) from both excitations, leading to three distinguishable excitation-emission combinations (the 370 nm light saturates the 310 – 400 nm detection channel and therefore does not provide any information). Thus, the fluorescence can be divided into 7 unique fluorescence groups based on the excitation-emission wavelength pairs and their combinations, after Perring et al. (2015) and Savage et al. (2017): A (only FL1: excitation 280 nm, emission 310 – 400 nm), B (only FL2: excitation 280 nm, emission 420 – 650 nm), C (only FL3: excitation 370 nm, emission 420 – 650 nm), AB (FL1 + FL2), BC (FL2 + FL3), AC (FL1 + FL3) and ABC (FL1 + FL2 + FL3).

The WIBS performs an empty-chamber background signal check every 8 hours, during which the excitation pulses are fired into the optical chamber without any present particles. The background check collects a multitude of emission intensities that form a baseline for particle fluorescence. In this study, a particle is considered fluorescent, if the associated emission peak intensity is larger than $FT + 9\sigma$. FT is the mean value of the forced trigger intensities and σ is their standard deviation.

A more commonly used method would be to compare the emission peak intensity to $FT + 3\sigma$. However, some non-biological particle types such as wood smoke, African dust and black carbon are weakly fluorescent and therefore might satisfy the lower threshold value, leading to an overestimation of biological particle concentration. Furthermore, the stricter threshold only marginally affects the detection efficiency of biological particles, because they tend to have stronger fluorescence (Savage et al., 2017). More detailed descriptions on the WIBS are also available in Savage et al. (2017) and Perring et al. (2015).

We also added the following text to the description on Figure 4 (old Figure 3b) to Section 3.2 Comparison to meteorology and aerosol properties to explain the characteristic of the different excitation-emission wavelengths pairs:

The time series of the number concentration of particles with a fluorescence signal in other fluorescence groups is shown in the Appendix in Fig. A2. In this Figure, the strongest seasonal increase in the transition period from winter to summer is observed in the group ABC. Consequently, this fluorescent group correlates best with the measured INP concentrations (see Figure 4). The characteristics of each fluorescence group are comprehensively investigated and reported in Savage et al. (2017), who examined the fluorescence emissions of different types of pollen, fungi, bacteria, biofluorophores, dust, HULIS (humic-like substances), PAH (polycyclic aromatic hydrocarbons), soot and brown carbon. Using the $FT + 9\sigma$ threshold for defining a particle as fluorescent, nearly all dust and HULIS types show no fluorescence signal at all. Some of the soot and brown carbon types only show weak signals in A, and B, BC and A, respectively. Nearly all of the bacteria types show fluorescence only in group A. The fluorescence of fungal spores are also mainly detected in group A, but also in AB and ABC. The investigated biofluorophores show mainly fluorescence in the groups BC (Riboflavin, NAD), A (Pyridoxamine), AB (Tryptophan) and ABC (Ergosterol). PAHs show fluorescence mostly in groups ABC and A. Finally, most pollen types show fluorescence in groups ABC and AB. Some pollen types also show a fluorescence signal in groups A and B.

We have also modified the discussion on the comparison of WIBS data to INP in the same Section as follows:

Such a correlation is also supported by the peaks of pollen and PBAP concentrations observed in snow-free periods in spring and in autumn, and by the increases of the organic aerosol mass concentration and fluorescent particle numbers of group ABC observed in spring. According to the study of Savage et al. (2017), we assume particles of fluorescence group ABC to be mainly pollen, particles containing PAH or Ergosterol, or fungal spores. ~~Fluorescent particles are expected to be primarily of biological origin except for a few percent, which could arise from non-biological materials (Pöhlker et al., 2012; Savage et al., 2017).~~

As was suggested by Savage et al. (2017), we have also introduced the categorization of fluorescence groups using A, B, C, AB, BC, AC, ABC instead of FL1, DL2, FL3, FL1+FL2, FL2+FL3, FL1+FL3, FL1+FL2+FL3, as this appeared to be less confusing. We changed the legend of Fig. A2 accordingly.

5) The explanations on the L-RoF-AMS data are also insufficient. The authors should briefly explain what kinds of organics were measured using the L-RoF-AMS and how they obtained organic mass concentrations presented in Figure 3.

Thanks for the comment. We agree that the description and discussion of the L-Tof-AMS needs to be more detailed and precise. We have added a more detailed description of the L-ToF-AMS instrument to Section 2.3. "Additional Instrumentation at SMEARII", which addresses this comment and also the associated comments regarding the L-Tof-AMS description from other referees:

The size-resolved chemical composition of ambient aerosol was measured with the L-ToF-AMS. Its application in the same campaign has been described in Paramonov et al. (2020). It builds on the functionality and characteristics of the high-resolution ToF-AMS (DeCarlo et al., 2006). However, due to the longer time-of-flight chamber, the L-ToF-AMS, has a better resolution (8000 M/ΔM) than the standard ToF-AMS (2000 M/ΔM in V-mode). Detailed descriptions of the instrument, measurements and data processing are available in other publications (Canagaratna et al., 2007; DeCarlo et al., 2006). In general, the L-ToF-AMS measures the size-resolved, non-refractory composition of submicron aerosols, including organic, sulfate, nitrate, ammonium and chloride. The aerodynamic lens has a 100% transmission range of 75-650 nm (in vacuum aerodynamic diameter; Liu et al. (2007)) and focuses particles into a narrow beam that impacts the surface of a porous tungsten vaporizer heated to 600°C, followed by ionization by a 70eV electron source. Ions are detected by a long time-of-flight mass analyzer (Tofwerk AG). The sample flow of 0.09 l min^{-1} is extracted from an extra suction flow (3 l min^{-1}) that is used to avoid aerosol losses in the inlet line. A PM2.5 cyclone mounted at the inlet removes large particles to avoid clogging the critical orifice (100µm), and before entering the L-ToF-AMS, the samples are dried by a Nafion dryer to keep the RH below 30%.

The L-ToF-AMS data were analyzed using standard ToF-AMS data analysis toolkits (Squirrel V1.61B and PIKA1.21B) using Igor Pro software (V6.37, WaveMetrics Inc.). To calculate mass concentrations an ionization efficiency (IE) was determined using 300 nm, size-selected, dry ammonium nitrate particles, and a relative ionization efficiency (RIE) for ammonium of 3.7 was determined. The default relative ionization efficiency (RIE) values of 1.1, 1.2, 1.3 and 1.4 for nitrate, sulfate, chloride and organics, respectively, were applied. A composition-dependent collection efficiency (CE) was applied based on the principles proposed by Middlebrook et al. (2012).

We have also added the following text to the discussion of Figure 4 (old Figure 3b) in Section 3.2 "Comparison to meteorology and aerosol properties."

The non-refractory organic components measured by the AMS include the commonly observed primary organic aerosol (POA) and oxygenated organic aerosol (OOA).

Specific Comments:

6) I could not find any descriptions of the objective of this study in the Introduction section. I thought that most sentences in the first paragraph of the Methods section (Lines 75-88) should be included in the Introduction section.

We agree that the manuscript would benefit from a stronger definition of the objectives of this study – thanks for this comment. We add the following text to the introduction:

The main objective of this study is to investigate and describe the variability and seasonal trends in INP concentrations and INP temperature spectra in a boreal forest environment. The absence of anthropogenic and/or dust aerosol sources in the boreal region motivates the additional

investigation of biogenic ice nucleation activity and reveals the relevance of boreal forest areas as an important INP source. The comprehensive instrumentation provided at the measurement site at the SMEARII station allows comparisons between INP measurements with simultaneous measurements of many meteorological variables. These measurements are complemented by measurements characterizing the sampled aerosol number concentrations, size distributions and chemical compositions in order to elucidate the potential origin and nature of the INPs. Heat treatments of the suspensions prior to INP analysis also help identifying the nature of INPs. We aim to improve the parameterizations describing atmospheric INP concentrations in the boreal forest by considering seasonal dependences in the formulations. Finally, this study provides motivation for further continuous long-term studies of INP in different environments across the globe.

In addition, we have shifted the description of the SMEARII station from the Methods to the Introduction, as the referee suggested.

7) Sections 2.1: What is the top height of the inlet? It is also important to provide the information on vegetation of the SMEARII site (e.g., tree canopy heights, dominant vegetation types) and the difference of the heights between the inlet and canopy.

For the top height of the inlet of the aerosol sampling line, we added the following sentence to Section 2.1 "Aerosol filter sampling":

The inlet height is approximately 4.6 m above ground and therefore approximately 17.2 m below the forest canopy.

Concerning the dominant vegetation species and canopy height, we have added the following information to the introduction into the description of the SMEARII station:

The boreal forest around the SMEARII station is dominated by Scots pine trees (Hari and Kulmala, 2005). In summer 2018, the canopy height of pines at SMEARII was determined to be 21.8 m.

8) Line 189 (and Figure 3): What is the definition of snow coverage? Where and how was the snow coverage measured?

We have added the following explanation to Section 3.2 "Comparison to meteorology and aerosol properties" to explain the definition of snow coverage applied here:

We have defined snow coverage as measured snow depth > 1 cm, where snow depth was measured by a Jenoptik SHM30 snow depth sensor, which is based on an opto-electronic laser distance sensor, in open field about 500 m southeast of the aerosol collection area of SMEARII.

9) Line 190: The authors would need to briefly explain about how pollen and other PBAPs reported by Manninen et al. (2014) have been measured at the SMEARII site.

We have added the following explanation to Section 3.2 "Comparison to meteorology and aerosol properties" to explain the measurements of PBAP in Manninen et al, 2014:

Manninen et al. (2014) collected aerosol samples in a Hirst-type volumetric spore trap (Burkard Manufacturing Co. Ltd.; Hirst, 1952), located at SMEARII 3 m above the forest canopy. The trap is driven by a clockwork and collects aerosol particles larger than approximately 3 µm on an adhesive, transparent, plastic tape with a sampling flow rate of approximately 10 l min^{-1}. The analysis of the collected particles was performed according to standard methodology adopted by the Finnish pollen

information network and following the principles of the European Aeroallergen Network (www.polleninfo.org/) and Rantio-Lehtimäki et al. (1994).

10) Line 208 (and Figure 4b): What is the definition of snow depth? Where and how was the snow depth measured?

See answer to Comment 8.

11) Figure 1: The size of numbers in x-axis and y-axis are too small.

We have increased the size of the axis description in Figure 1.

12) Figure 3a: What is the unit of y-axis? As for pollen and PBAP data, since pollen is a kind of PBAPs, I think these should be treated as "pollen" and "other PBAPs". Also, since the pollen and other PBAP data were obtained in 2003/2004, these data should not be included in Figure 3a. I would like to suggest making another figure if they want to show the pollen and other PBAP data in 2003/2004.

We thank the referee for this suggestion. The units for PBAP and INP concentrations has been added (m^{-2} and std l^{-1}, respectively). The fraction of NPF event days has no unit. The legend was changed from "PBAP" to "other PBAP". To improve the comparison of data measured in different years, the x-axis labels were adjusted by removing the year(s) and instead the corresponding years are shown in the legend. Now, the presentation of pollen and PBAP data from 2003/2004 together with INP, NPF and snow data from 2018-2019 should be improved. We think it is useful to have this data in one panel, as we focus on general seasonal trends and not on quantitative values measured in specific years. Other improvements to this Figure has been done considering other referee comments.

13) Figure 3b: What is the unit of y-axis? What is the source of fluorescent particles in this figure? I guessed that it would be the same as "all" in Figure A2 according to a description "all three lase-channel combinations of WIBS" in the main text (lines 205- 206). However, these curves in Figures 3b and Figure A2 seems to be quite different. Please clarify this point (see also Comment 4).

We did not use "all", but "FL1+FL2+FL3" (group ABC), which agrees with the curve in Figure A2. We have now completely modified the Figure. For details see answer to comment 14) below. A complete comparison, including aerosol number concentration and surface concentration is too much for a single panel, and thus we prepared a separate Figure including several panels showing INP concentrations compared to ABC fluorescent particle concentration, mass concentrations of organics, PM10 number concentration and PM10 surface concentration (see new Figure 4). We have added corresponding units to the axis labels, where they were missing.

14) Figure 3b: I would like to suggest including the data on the number and/or surface area concentrations in PM10 (the data in Figure A1) during the HyICE period in this figure. Then, the authors should discuss if the variations of INPs are indeed related to organics and fluorescent particles, rather than PM10 particles.

Thanks for this suggestion. Continuing from above in the new Figure 4 panel (a) shows a comparison of INP concentrations to ABC fluorescent particles, panel (b) to organic mass concentrations, panel (c) to PM10 number concentrations and panel (d) to PM10 surface concentrations.
We have added additional description to Section 3.2. "Comparison to meteorology and aerosol properties:"

Both, the organic aerosol mass concentration measured by the L-ToF-AMS and the number concentration of fluorescent particles measured by WIBS, tend to increase during the transition

period from winter to summer (Fig. 4a and b). The number concentration of atmospheric PM10 aerosol presented in Fig. 4c does not show this trend. However, a slight seasonal trend is visible in the PM10 surface concentration (Fig. 4d).

We have also added a short discussion of the previous observation later to the same Section:

As the surface concentration of PM10 particles is more sensitive to larger particles, the increase in PM10 surface concentration (see Fig. 4d) indicates that the observed seasonal increase may be due to larger particles, which are expected to be mainly of biogenic origin.

15) Figure 6d: How did the authors apply the parameterization by Ullrich et al. (2017)? Did they measure the surface area concentrations of mineral dust particles at this site? Or did they simply apply this parameterization to the measured total aerosol surface area concentrations in PM10? Please clarify this point.

The parameterization of Ullrich et al. (2017) for the INAS density of mineral dust is only temperature dependent. Therefore, for the application of this parameterization, we only used the activation temperatures in the Ullrich et al. (2017) formulation. When we established a new parameterization for the INAS densities of the measured boreal forest INPs (see Eq. (2)), we used the same exponential relation of INAS density and activation temperature as suggested in Ullrich et al. (2017). For calculating the INAS densities used to establish this new parameterization, the PM10 surface area concentrations measured by APS/DMPS including all atmospheric aerosol particles were used.

16) Figure A1a: I could not understand how to see this figure. What do the authors mean by "percentage deviation"? Please clearly explain the meaning of this word.

Thanks for this comment. The old Figure A1a was meant to explain and show the difference in percent between the number/surface concentration of total aerosol and PM10 aerosol. As the Figure seems to be more confusing, we decided to remove this Figure and just describe in the text that the difference between total aerosol and PM10 aerosol is lower than 1% (line 151, revised manuscript). The old Figure A1b was separated into two panels and compared to INP concentration at 257 K in the same way as meteorological parameters are compared to INPs and are now the new Figure A1.

We referred to the new Figure in Section 3.2 “Comparison to meteorology and aerosol properties,” as follows:

The number and surface concentration of PM10 atmospheric aerosol for the entire time period from March 2018 to May 2019 is shown in the Appendix in Fig. A1.

17) What are the valid temperature ranges of equations 1 and 2? Did you apply these equations to all available INP data or INP data in a specific temperature range when you made Figure 7?

Regarding equation 1, it is mentioned in line 494 that INP-T-spectra between 250-265 K are used. Therefore, the parameterization is valid for this temperature range. Also, for the INAS density parameterization in equation 2, the spectra in the full T range (250-265 K) was used and is therefore valid in the same temperature range.

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