

## ***Interactive comment on “Best practices for precipitation sample storage for offline studies of ice nucleation” by Charlotte M. Beall et al.***

**Charlotte M. Beall et al.**

cbeall@ucsd.edu

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The Authors would like to thank Gabor Vali for his helpful suggestions and discussion. A summary of our responses is below. Line numbers refer to the latest marked-up version of the manuscript.

Referee: “There is no indication in the paper of the kind of precipitation that was sampled. Presumably - vaguely deduced from the variations in the lengths of the sampling periods - a variety of precipitation types are included. Probably, some light rain to more showery situations were involved. Cases with all warm-rain processes and cases with ice origin may have been involved. This may justify the choice of ‘precipitation’ in the title rather than ‘rain’. If all events were from clouds with no ice-phase, a change in

C1

the title would be warranted to indicate so. This point isn’t very important to the main theme of the paper, but it could possibly make a difference for considerations of how the present results might apply to other situations.”

Thank you for this suggestion. A summary of the types of precipitation events that were sampled has been added to Table 1. Meteorological conditions associated with precipitation ranged from AR events to warm, low altitude rain clouds. It is true that the choice of “precipitation” in the title and throughout the text was motivated by ambiguity regarding the ice or liquid origin of the samples. Although all samples were collected as liquid at ground-level, it is possible that ice-processes were involved in the precipitating clouds (judging by the low cloud-top temperatures in the NWS satellite composite analysis).

The following updates have been made to the text: Sec. 2.1, Line 117: Satellite composites from the National Weather Service Weather Prediction Center’s North American Surface Analysis Products were used for synoptic weather analysis to generally characterize each rain event (see Table 1). Atmospheric river (AR) events were identified using the AR Reanalysis Database described in (Guan and Waliser, 2015) and (Guan et al., 2018).

Sec. 3.1, Line 165: Figure 1 shows INP concentrations of 15 coastal rain samples, collected in a variety of meteorological conditions including scattered, low coastal rain-clouds, frontal rain, and atmospheric river events (see Table 1). Observations generally fall within bounds of previously reported INP concentrations from precipitation and cloud water samples (grey shaded region, adapted from Petters and Wright, 2015). Observed freezing temperatures ranged from -4.0 to -18.4 °C, with concentrations up to the limit of testing at 105 INP L-1 precipitation. AIS measurement uncertainties are represented with 95% binomial sampling intervals (Agresti and Coull, 1998).

Referee: “The main constraint mentioned in the paper and explicitly stated in the conclusions is that the results refer to coastal environments. This is not as helpful as could

C2

be, since precipitation and aerosol sources on the coasts may still include a very broad variety.”

To provide broader context for our dataset, we have updated Figure 1 following the assumptions of Petters and Wright, 2015 to estimate in-cloud INP concentrations from precipitation samples (i.e. 0.4 g condensed water content m<sup>-3</sup>). The updated Figure 1 shows that the INPs observed here are comparable to spectra reported previously for a wide range of marine and coastal environments, including the Caribbean, Bering Sea, East Pacific and nascent sea spray aerosol (DeMott et al., 2016). As INP spectra in this temperature regime cluster distinctly by air mass type (e.g. Figure 1-10 in Kanji et al., 2017), Fig. 1 indicates that the air masses sampled in this study were likely primarily marine.

The following updates have been made to the text:

Title: “Best practices for precipitation sample storage for offline studies of ice nucleation in marine and coastal environments”

Sec. 3.1, Line 171: “ Following the assumptions in (Wright and Petters, 2015) to estimate in-cloud INP concentrations from precipitation samples (i.e. condensed water content of 0.4 g m<sup>-3</sup> air), observations of INP concentrations in fresh precipitation samples are additionally compared to studies of field measurements conducted in marine and coastal environments. Figure 1 shows that atmospheric INP concentration estimates compare with INP concentrations observed in a range of marine and coastal environments, including the Caribbean, East Pacific, and Bering Sea, as well as laboratory-generated nascent sea spray aerosol (DeMott et al., 2016).”

Sec. 3.1, Line 282: “Additionally, the INPs in this study compare with INPs observed in studies of marine and coastal environments (Fig. 1). As spectra in this regime (-5 to -20 °C and 10<sup>-5</sup> to ~10<sup>-1</sup> per L air, respectively) cluster distinctly by source type (see Fig. 1-10 in Kanji et al., 2017), Fig. 1 indicates that the dominant sources to air masses sampled in this study were marine. Considering that data in this study

C3

are characteristic of marine and coastal INPs previously reported over a wide range of marine environments (DeMott et al., 2016), we assume that the findings herein are relevant to samples from other marine and coastal environments.

Sec. 3.1: Line 294: If correspondence within 1 order of magnitude (or 2-3 °C) is desired, uncertainties associated with storage should also be considered in studies using samples from coastal or marine environments. Thus, uncertainty distributions provided in Tables 5-7 can be used to evaluate observed INP concentrations and responses to treatments in the context of potential changes due to storage. However, the degree to which INP sensitivity to storage varies by INP source (e.g. with soil-derived INP populations) remains to be tested.

Referee: “Separating measurement variability from actual changes is important. Figures 2-4 include indications of measurement reproducibility with the gray bars adjacent to the data point clusters. All of these bars are indicating values greater than unity. The caption to Fig. 2 says that the bars represent the ‘average difference between replicates’. How is this to be interpreted? What conclusion can drawn from these data?”

The following changes have been made in Sec 3.2 to describe how grey bars are to be interpreted and conclusions that may be drawn from the data. This section has also been updated to address the referee’s 1st comment on sample handling in “Minor points”:

Sec. 3.2, Line 218: Replicate samples were processed for each storage protocol so that impacts of sample handling can be distinguished from storage impacts. For example, if settling occurs in bulk rain samples that are then divided into smaller volumes prior to storage, INP concentrations may differ between replicates of the bulk sample. Thus, it is assumed that INP concentration changes that are greater than differences between replicates (grey bars in Figs 2-4) can be attributed to storage impacts.

Referee: “A lingering uncertainty in the paper about whether these treatments were applied to the fresh sample before division and storage, or just prior to INP measure-

C4

ment, is disconcerting. The discussion in lines following 218 seem to indicate that filtering was done before freezing for storage. It would be good to have the sequence better described.”

Thank you for bringing this to our attention. The following change has been made to Sec. 2.2 Storage Protocols:

Line 136: Heat treatments and filters were applied to samples just prior to processing (i.e. treatments were not applied to samples prior to storage).

Referee: “The overall effects of the treatments are given as, on the average, 59% of INPs were found resistant to heat and 69% passed the filters. These numbers are overly vague, as dependence of temperature can be expected as well as variations from sample to sample. While such detail will not alter the data, it is relevant to possible explanations of the results.”

This is a good point. The figures quoted above were calculated at the temperature of the next to last freezing event of the corresponding fresh sample (beyond which the data is not meaningful), again using the cumulative distribution. I recalculated the ratios of heat-treated and filtered to untreated INPs in fresh samples in the temperature intervals consistent with the rest of the manuscript (-9, -11, -13, -15 etc). This new way of calculating the filtered to untreated fractions yields a different answer regarding the general sizes of the INPs, interestingly, probably because smaller particles represent larger fraction of INPs only at the colder temperatures. It is also now necessary to discuss that we observed some enhancements in INPs after heating fresh samples (5 of the 15 samples). We have added the following detail and discussion:

Sec 3.1, Line 178: “In 5 of the 15 heat-treated samples, INP concentrations were increased by 1.9 – 13X between -9 and -11 °C (see Discussion). Excluding these 5 samples, the fraction of heat-resilient INPs varied between samples and generally increased with decreasing temperature. Geometric means and standard deviations of heat-treated:untreated INP ratios were  $0.40 \times \pm 1.9$ ,  $0.51 \times \pm 2.0$ , and  $0.62 \times \pm$

C5

2.1 at -11 , -13, and -15 °C respectively. Fractions of INPs < 0.45  $\mu\text{m}$  also varied between samples, with geometric means and standard deviations of  $0.48 \times \pm 1.73$ ,  $0.30 \times \pm 3.4$  and  $0.37 \times \pm 1.9$  at -11 , -13, and -15 °C respectively. Mean values of heat-resilient INP fractions and INPs < 0.45  $\mu\text{m}$  were calculated using the geometric mean, which is more appropriate than the arithmetic mean for describing a distribution of ratios (Fleming and Wallace, 1986).”

Discussion, Line 272: “The fractions of INPs < 0.45  $\mu\text{m}$  observed in this study varied between 52 and 63% at -11 and -15 °C, respectively. Excluding the five heat-treated samples in which INP concentrations were enhanced (e.g. 1.9 - 13X between -9 and -11 °C), the average fraction of non-heat-labile INPs varied between 40 and 62% at -11 and -15 °C, respectively. INP enhancements in heat-treated samples are unexpected, as heat-treatments are typically applied assuming that heat destroys proteinaceous (e.g. biological) INPs. The causes of INP enhancements in heat-treated samples are unknown and have only been reported in coastal precipitation samples (Martin et al., 2017) and nascent sea spray aerosol (McCluskey et al., 2018).

Discussion, Line 305: “Despite the range of enhancements and losses of heat-sensitive INPs observed in fresh samples, non-heat-labile INPs were generally less sensitive to storage than the total INP population., and with the exception of samples stored at room temperature, all techniques yielded similar results with fewer enhancements or losses. “

Discussion, Line 330: In this study, a large fraction (30% to 48%, on average) of INPs observed in fresh precipitation samples were < 0.45  $\mu\text{m}$ . Considering this and that INPs < 0.45  $\mu\text{m}$  exhibit significant losses across all storage types, there is a risk that filter-treatments on stored samples in this study would lead to the underestimation of INPs < 0.45  $\mu\text{m}$ .

Referee: On the level of internal consistency in the paper, it is worth asking how justified is the statement underlying conclusion 6 (line 280). Significantly greater losses

C6

are said to occur in storage for filtered samples. This is not really evident from a comparison of Fig. 2 with Fig. 4, or from the figures in Table 5 versus Table 7. Greater variability (larger 95% range) is found only for 'refrigeration' and 'freezing', while 'room temperature' and 'flash freezing' have narrower ranges and smaller standard deviations in Table 7 than in Table 5. Perhaps the claimed effect was clear for selected samples but not for the combined sample set.

This conclusion is based on the increased frequency of significant (Fisher's,  $p < 0.01$ ) data points on Figs 2 and 4. After making the suggested changes (see response to comment below), the 95% confidence intervals span losses  $> 1$  order of magnitude across all protocols and most temperature intervals. We have added the following text to explain how Tables 5-7 may be interpreted:

Discussion, Line 291: While mean INP changes are within a factor of  $\sim 2$  or less of fresh sample INP concentrations for all protocols except "Room temperature" (Table 5), none of the 4 storage protocols prevented significant losses or enhancements of INP concentrations in all samples (Fig. 2), indicating that INP concentration measurements on fresh precipitation are superior to measurements on stored samples. 95% confidence intervals in Table 5 span losses  $> 1$  order of magnitude in all protocols across multiple temperature intervals. As uncertainties  $< 1$  order of magnitude are necessary for the quantitative comparison between studies (DeMott et al., 2017), our results demonstrate that uncertainties associated with storage must be considered in studies of stored samples from coastal or marine environments. Thus, the uncertainty distributions provided in Tables 5-7 can be used to evaluate observed INP concentrations and responses to treatments in the context of potential changes due to storage. However, the degree to which INP sensitivity to storage varies by INP source (e.g. with soil-derived INP populations) remains to be tested.

Referee: "Tables 5-7 have some technical problems (see comment below on lines 185-189), but taking the data as is, most notable is the large range of variations for the corrections factors. Not just the 95% range, but even 50% spread: for the last line

C7

in Table 5, the 50% range is roughly 0.78 to 2.8. Experiments seldom lead to more accurate INP concentrations due to limitations in sample sizes (number of drops or vials). This reinforces the point that the results should be viewed as indications of the uncertainties associated with aging of samples during storage and not as correction factors that can usefully improve measured INP data in other studies. This argument is further supported by the potential for differences in the aging effects for precipitation at different times and locations. The current data provide help in weighing the importance of aging versus other sources of uncertainties in a given experimental design."

We agree that these results are better indications of the uncertainties associated with storage. Updates have been made to the tables, table legends and text to reflect this change:

Abstract: Finally, the estimated uncertainties associated with the 4 storage protocols are provided for untreated, heat-treated and filtered samples for INPs between -9 and -17 °C. Conclusion, Line 375: "2. Estimates of uncertainty attributed to storage impacts and 95% confidence intervals for INP measurements obtained from stored samples are provided (see Tables 5-7)."

Legend Table 5: Table 5. Estimate of uncertainty associated with storage impacts for INPs with activation temperatures between -9 and -17 °C measured in stored, untreated precipitation samples. Confidence intervals were derived from the log-normal distribution of changes observed in INP concentrations due to storage (see Fig. 2 and details in Sect. 3.2). Temperature intervals where datapoints were too few to derive confidence intervals are indicated with "NA". Changes in INP concentration corresponding to enhancements or losses greater than 1 order of magnitude (losses  $\leq -90\%$  or enhancements  $\geq +900\%$ ) in bold.

Legend Table 6: Table 6. Estimate of uncertainty associated with storage impacts for INPs with activation temperatures between -9 and -17 °C measured in stored, heat-treated precipitation samples. Confidence intervals were derived from the log-normal

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distribution of changes observed in INP concentrations due to storage (see Fig. 3 and details in Sect. 3.2). Changes in INP concentration corresponding to enhancements or losses greater than 1 order of magnitude (losses  $\leq -90\%$  or enhancements  $\geq +900\%$ ) in bold.

Legend Table 7: Table 7. Estimate of uncertainty associated with storage impacts for INPs with activation temperatures between  $-11$  and  $-19$  °C measured in stored, untreated precipitation samples. Confidence intervals were derived from the log-normal distribution of changes observed in INP concentrations due to storage (see Fig. 2 and details in Sect. 3.2). Temperature intervals where datapoints were too few to derive confidence intervals are indicated with "NA". Changes in INP concentration corresponding to enhancements or losses greater than 1 order of magnitude (losses  $\leq -90\%$  or enhancements  $\geq +900\%$ ) in bold.

Minor points: Referee: "How was the sample division done for different treatments? While this can be expected to be a step without risk of introducing discrepancies among the samples, it is not without such a possibility. Thus, the manner it was done should be described, as well as any tests done to assure that this step doesn't lead to artifacts."

The following updates have been made to the text: Sec. 2.2, Line 128: Prior to storage, 25 - 50 mL bulk sample aliquots were distributed directly from collection bottles into Falcon<sup>®</sup> tubes, shaking bottles  $\sim 10$  s between each distribution.

Sec. 3.2, Line 218: Replicate samples were processed for each storage protocol so that impacts of sample handling can be distinguished from storage impacts. For example, if settling occurs in bulk rain samples that are then divided into smaller volumes prior to storage, INP concentrations may differ between replicates of the bulk sample. Thus, it is assumed that INP concentration changes that are greater than differences between replicates (grey bars in Figs 2-4) can be attributed to storage impacts.

Referee: "Line 85 mentions samples getting divided into 24 bottles during collection. What is the relationship between this and the division of the samples for different treat-

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ments?"

This is a description of the precipitation collection device, which changes bottles on a rotating carousel at the specified time interval. There is no consistent relationship between the bottle numbers and sample division because sometimes we combined bottles corresponding to consecutive 1-hour sampling intervals in order to have enough volume for each of the sampling protocols, replicates, treatments, etc. This is described in Sec. 2.1: "The samples were distributed via the distributor arm into one of twenty-four 1-liter polypropylene bottles on an hourly time interval. Bottles were combined when the hourly precipitation volume was insufficient for sample separation and analysis ( $< 50$  mL)."

I have updated the last line to help clarify: Sec. 2.1, Line 112: "Bottles corresponding to consecutive 1-hour time intervals were combined when the hourly precipitation volume was insufficient for sample separation and analysis ( $< 50$  mL per bottle)."

Referee: "Figure 1 shows points near  $-5$ °C for one sample. This should be of special interest but the paper doesn't mention it. Was the sample unusable?"

We have added the following text to acknowledge the two warm-freezing observations (this sample was used in the storage experiments). Sec. 3.1, Line 176: "However, two of the warmest-freezing INP observations in Fig. 1 (at  $-4.0$  and  $-4.75$  °C) exceed temperatures commonly observed in marine-influenced atmospheres, precipitation and cloudwater samples

Referee: "Perhaps Fig.1 could be made less congested by showing only the interval  $0$ °C to  $-20$ °C." This is true, however, I think that seeing the whole Wright and Petters, 2015 and now DeMott et al., 2016 composite spectrum because it provides context for the regime we are observing.

Referee: "Is there more than one point included in Figs. 2-4 for a sample from the same rain event and time period? Unfortunately, one can't determine from the figures

C10

how many data points are shown for each temperature. More than the number of rain events? The number of points differs for different temperatures. Is this because of limits in the temperature range of freezing events?"

Yes, more than one point from the same sample is included in Figs. 2-4. This was motivated by the fact that a subset of the replicate samples exhibited differing sensitivity to storage (Fig. S5). However, replicates were not included in uncertainty factor calculations to avoid underweighting the small subset of samples that did not have replicates due to sample volume limitations. This is currently stated in the figure legends and table legends, and tables 2-4 show how many unique samples are represented in each figure vs how many replicates. The number of points differ due to limits of detection. There are typically fewer datapoints in the warmest and coldest temperature bin. At the warmest temperature bin, one of the samples (fresh or stored) is more likely to have 0 wells frozen, which would result in either a ratio of 0 or Inf. Ratios of zero were excluded because they are reflective of the limit of detection due to the number of droplets processed rather than a true lack of ice nucleating particles at this temperature. Similarly, datapoints tend to be fewer in the coldest temperature bin because in one or both of the samples (fresh and stored), all the wells had frozen.

The following has been added to the text: Sec. 3.2 Line 191: "Numbers of datapoints in Figs 2-4 differ across the temperature intervals due to limits of detection (i.e. ratios were not calculated at temperatures where zero or all wells were frozen in the fresh and/or stored sample)."

Figure 2 legend: All samples were processed at one or two time intervals between 1 and 166 days post-collection (see Figs S1-S4). For samples processed at two intervals, both replicate samples are represented in the figure for a total of 14, 16, 18 and 12 samples in (a), (b), (c) and (d), respectively (see Table 2 for summary of sample and replicate numbers). Significant data are also labelled to indicate the sample number (01-15, see Table 1), and replicate ("A" or "B", and "U" indicates there were no replicates for the sample). Figure 3 legend: All samples were processed at one or two

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time intervals between 1 and 166 days post-collection. For samples processed at two intervals, both replicate samples are represented in the figure for a total of 13, 16, 15 and 12 samples in (a), (b), (c) and (d), respectively (see Table 3 for summary of sample and replicate numbers).

Figure 4 legend: All samples were processed at one or two time intervals between 1 and 166 days post-collection. For samples processed at two intervals, both replicate samples are represented in the figure for a total of 13, 15, 16 and 12 samples in (a), (b), (c) and (d), respectively (see Table 4 for summary of sample and replicate numbers).

Figures 2-4 have been also been updated to show the numbers of samples represented for each protocol. Each filled data marker is now also labeled to show which sample and replicate it corresponds to.

Referee: "It would have been useful to identify by precipitation sample each data point in Figs 2-4, at least for the outliers. This would clarify, for example, if all the points with lowest values in Fig 2(a) are for the same sample or not, and if the same sample has the lowest points in Fig 2(b) and 2(c)"

This is a good idea, thank you. Figs 2-4 have been updated as suggested, with some text on each filled marker with the precipitation sample number and either the letter "A" or "B" to indicate which replicate it is. Samples without replicates are marked "U".

Referee: "Line 73: Were heat treatment and filtering applied before division for different storage temperatures, or just before INP analysis? One can assume it is the former, but the paper leaves it unspecified. Lines 101-102 still don't make clear what was done. Line 179 seems to indicate that filtration was done before storage."

Heat treatments and filtering was applied just before INP analysis, since this order is probably more common in e.g. field campaigns where samples must be frozen onsite before transport.

The following was added to Sec. 2.2, Line 136: "Heat treatments and filters were

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applied to samples just prior to processing (i.e. treatments were not applied to samples prior to storage).”

Referee: “Line 100: reference to ‘section above’ seems incorrect.”

This line has been corrected in Sec 2.2, Line 133: “INP measurements were made in two or three time steps: within two hours of collection, and once or twice after storing using one of four storage protocols described above, depending on volume.”

Referee: “Line 171: Is the ratio cited independent of the INP activity temperature?”

This ratio has been updated to reflect temperature dependence (see responses to referee comment beginning “The overall effects of the treatments are given as, on the average [..]”)

Referee: “Line 138: ‘... binned by 2°C increments ..’ seems odd for cumulative data. More likely, values are ‘determined (or calculated) at successive 2°C intervals’. If that is not the case, please explain. The word ‘binned’ appears in numerous places in the text.”

Thank you for bringing this to our attention. The following corrections have been made: Sec. 3.2, Line 189: INP concentrations of stored replicate samples are compared with original fresh precipitation samples in Figures 2-4, calculated in successive 2 °C increments between -7 and -19 °C.

Sec 3.2, Line 194: All stored: fresh ratios were calculated from cumulative INP distributions in 2 °C intervals, meaning that the INP concentration in each interval is inclusive of the concentration in the preceding (warmer) temperature interval. Thus, in this study, deviations observed in a stored sample are not necessarily independent, i.e. the sensitivity of INPs to storage in one temperature interval could impact the observed changes in each of the following (colder) temperature interval. For example, in the fresh untreated precipitation samples (see Fig. 1), the contribution of INPs from the preceding 2 °C interval ranges from 32 to 46% between -9 and -17 °C.

C13

Sec 3.2., Line 216: For each temperature interval containing data from at least two sets of replicate samples, the average difference in stored: fresh concentration ratios between replicates are represented with grey bars to indicate measurement variability.

Sec 3.2, Line 226: Finally, Fisher’s Exact Test was applied to frozen and unfrozen well fractions between each stored sample and its corresponding fresh sample at each of the 2 °C temperature intervals.

Sec 3.2, Line 232: Results in Fig. 2 show that significant enhancements or losses of INPs occurred for all stored samples between -9 and -17 °C, and that on average, stored samples exhibit INP losses (as indicated by the mean change in each temperature interval).” Figure 2 legend: Figure 2: Ratio of INP concentrations measured in untreated precipitation samples (stored: fresh), calculated in successive 2 °C increments between -19 and -7 °C. In temperature intervals containing stored: fresh ratios from at least two sets of replicate samples, grey bars represent the average difference between replicates. Stored sample frozen well fractions that passed Fishers Exact Test ( $p < 0.01$ ) for significant differences from original fresh sample frozen well fractions at each of the 5 temperatures are indicated with filled markers, and the mean change in each temperature interval is marked with a star.

Figure 3 legend: Figure 3: Ratio of INP concentrations measured in heated precipitation samples (stored: fresh), calculated in successive 2 °C increments between -19 and -7 °C. In temperature intervals containing stored: fresh ratios from at least two sets of replicate samples, grey bars represent the average difference between replicates  
Figure 4 legend: Ratio of INP concentrations measured in filtered (0.45 μm) precipitation samples (stored: fresh), calculated in successive 2 °C increments between -19 and -7 °C. In temperature intervals containing stored: fresh ratios from at least two sets of replicate samples, grey bars represent the average difference between replicates.

Referee: “Line 140: What does ‘significant’ refer to here? Maybe the authors meant ‘measured’.”

C14

This line has been corrected in Sec 3.2, Line 190: This temperature range was chosen for the analysis because most fresh precipitation samples exhibited freezing activity between -7 and -19 °C.

Referee: "Line 141: The cumulative values at any point are calculated by accounting for all freezing events (all frozen sample wells) at temperatures higher than the value at which the concentrations is evaluated, not just those of the preceding value at 2 °C higher temperature. Also, in line 146, 'each' should be replaced by 'all', and line 147 should be rephrased and clarified."

This line has been corrected in Sec 3.2, Line 194: All stored: fresh ratios were calculated from cumulative INP distributions in 2 °C intervals, meaning that the INP concentration in each interval is inclusive of the concentration in all of the preceding (warmer) temperature intervals.

Line 201: For example, in fresh untreated precipitation samples (see Fig. 1), 32% of the INP concentration calculated at -11 °C activated in one of the preceding (warmer) 2 °C temperature intervals. At -17 °C, this fraction is increased to 46%.

Referee: "Line 157: ' .. containing data from at least two sets of replicate samples ...' seems to say that data points shown include replicates from the same rain event. This is brought up again in lines 187-188 and in the caption to Fig. 2."

Yes, this line is intended to make clear that replicates are represented in Fig. 2-4. See response to comment beginning "Is there more than one point included in Figs. 2-4 for a sample from the same rain event and time period?"

Referee: "Lines 160-161: 'well counts' and 'well fractions' are not the same - please clarify."

This line has been corrected in Sec 3.2, Line 229: Finally, Fisher's Exact Test was applied to frozen and unfrozen well fractions between each stored sample and its corresponding fresh sample at each of the 2 °C temperature intervals. Stored sample frozen

C15

well fractions that were significantly different ( $p < 0.01$ ) from fresh sample frozen well fractions at each of the 5 temperatures are indicated with filled markers.

Referee: "Line 161: ' ... at each of the 5 temperatures ..' should probably be left out"

Line 228 has been corrected: "Stored sample frozen well fractions that were significantly different ( $p < 0.01$ ) from fresh sample frozen well fractions are indicated with filled markers."

Referee: "Line 163: Here it says that all stored samples showed significant changes whereas only a few points are shaded in Fig. 2"

Line 232 in Sec 3.2. has been corrected: "Results in Fig. 2 show that significant enhancements or losses of INPs occurred in all storage protocols between -9 and -17 °C, and that on average, stored samples exhibit INP losses (as indicated by the mean change in each temperature interval)."

Referee: "Line 179: The reference to Sect. 2.3 for detail is incorrect"

Line 248 in Sec 3.2 has been corrected: Effects of storage protocol on INP concentrations of filtered precipitation samples are shown in Figure 4 (0.45  $\mu\text{m}$  syringe filter, see Sect. 2.2 for details).

Referee: Lines 185-189: Tables 5-7 indicate the range of impacts that may be expected on the basis of the data presented in this paper. The correction factors here given appear to have been derived combining data from all temperatures for given storage and treatment type. This has an inherent multiplicity problem as data at successively lower temperatures include all data from higher temperatures. Thus, a value for, say, -11 °C is also incorporated into the values at -13 °C, -15 °C etc. so that the ratio at -11 °C is given multiple, though uneven, weights when combining all the values for -11 °C and lower into calculating a mean and standard deviation for the given treatment. Also, all data were included in calculating the values in Tables 5-7, not just the cases for which the differences observed were shown to be statistically significant.

C16



One may wonder what the results would be if only those cases were included.”

Thank you for bringing this to our attention. We agree that there is a multiplicity problem in combining the data from all intervals and have updated the tables to show average changes and confidence intervals for each 2 °C temperature increment. See also responses to p.4 comment beginning “Tables 5-7 have some technical problems [...]” about how the tables and text have been changed to reflect that these data represent uncertainty associated with storage rather than correction factors.

I also agree that it would be interesting to recalculate these figures using the significant (Fisher’s) datapoints only, but I couldn’t think of a way to justify the exclusion of the insignificant datapoints in a calculation. It is also likely that such a distribution would not be log-normal, and then it wouldn’t be meaningful to calculate the geometric mean and 95% confidence intervals.

Referee: “Line 192: What is meant by ‘in situ’ collection? Similarly, in line 241 ‘in situ dust’ is vague.”

Line 263 and Line 338 have been updated in the Discussion section:

The challenge in selecting a storage protocol for atmospheric samples (e.g. precipitation, cloud water, ambient atmosphere) is that the INP population composition is unknown, diverse, and the impact of any given technique on the different species may vary.

Considering that well-characterized IN-active dust and biological standards (Arizona Test Dust and Snomax<sup>®</sup>, respectively) are sensitive to storage conditions, it is possible that dust or biological INPs contributed to the observed INP losses.

Interactive comment on Atmos. Meas. Tech. Discuss., doi:10.5194/amt-2020-183, 2020.

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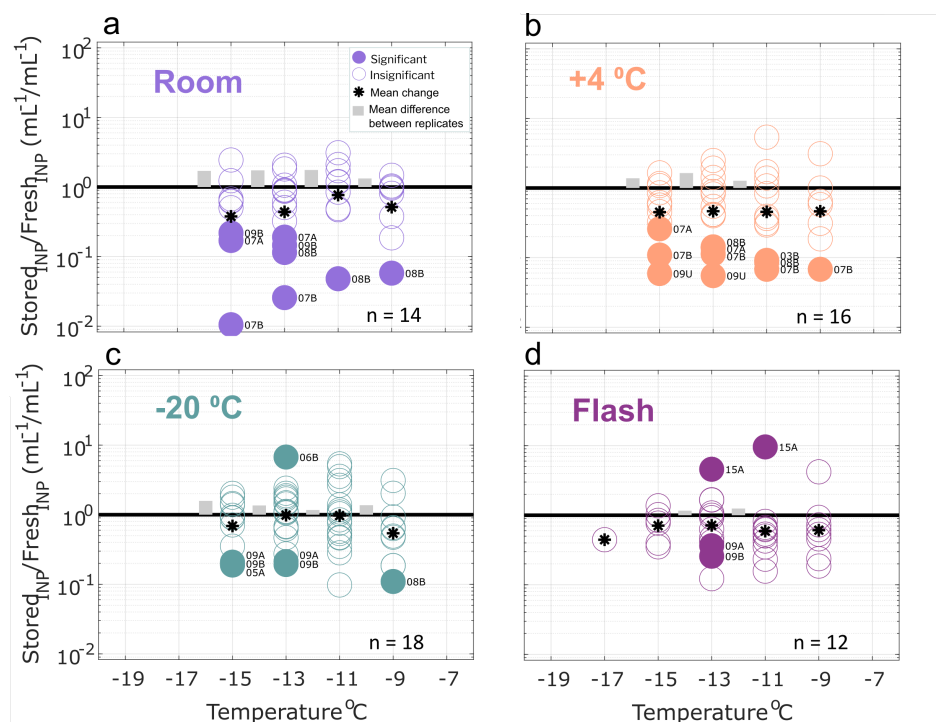


Fig. 1. Figure 2

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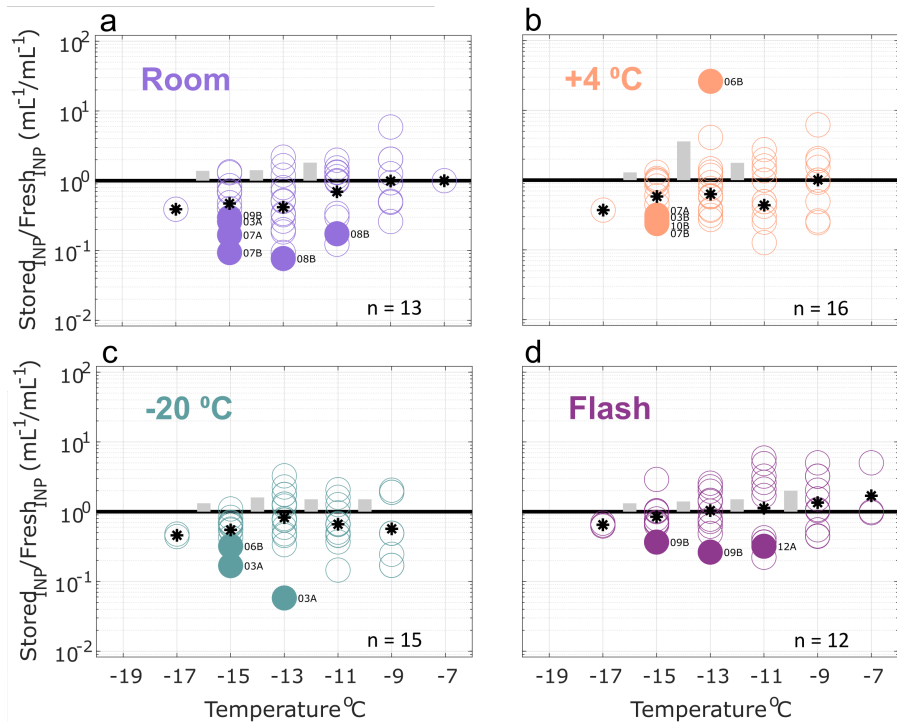


Fig. 2. Figure 3

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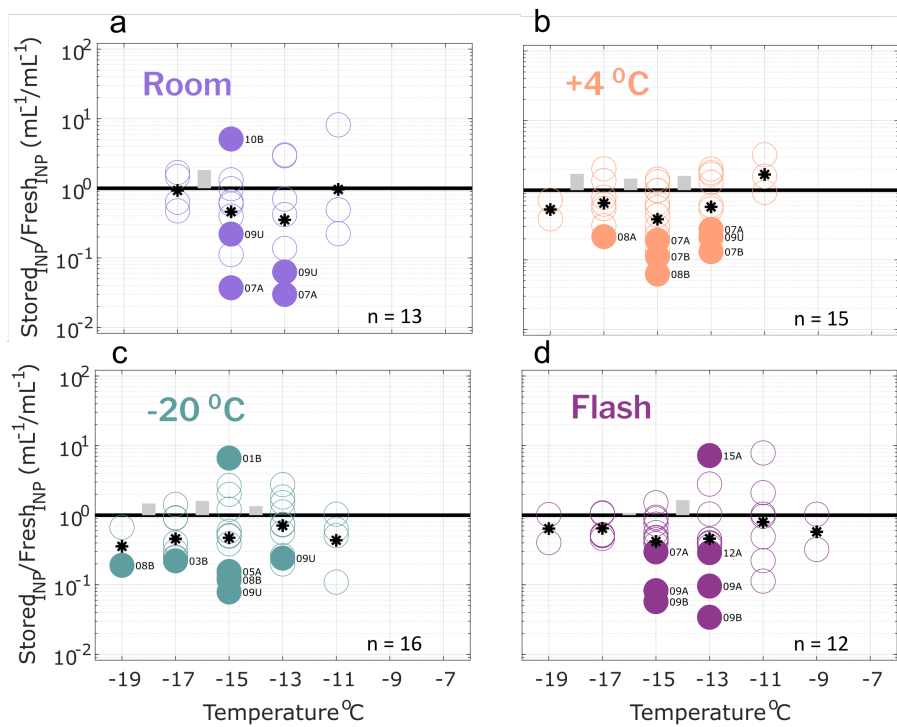


Fig. 3. Figure 4

C20