Answer to reviewer's comments on the manuscript by Thomas Häusler, Lorenz Witek, Laura Felgitsch, Regina Hitzenberger and Hinrich Grothe "Heterogeneous freezing of super cooled water droplets in micrometre range- freezing on a chip"

All three referees have suggested that the manuscript would be more suitable for AMT. After correspondence with the handling editor and Chief editors of both AMT and ACP, the editors recommended to proceed with the regular review process in ACPD as a manuscript under consideration for publication as a Technical Note in ACP. Therefore the title of the manuscript has been changed to: *"Technical note: Heterogeneous freezing of super cooled water droplets in micrometre range- freezing on a chip"*

The revised manuscript was uploaded separately and is available as *author comment*.

Reviewer 3

The authors would like to thank the reviewer for the constructive comments!

Comment

1. As the use of a gold surface is the most novel aspect of this approach, it really warrants more discussion. Why was gold chosen? Was its surface already known to induce poor/zero ice nucleation properties? No discussion or references regarding this are provided. Silicon was found to strongly induce ice nucleation, so what is special about gold's surface? Might other (precious?) metals have similar desirable properties as gold? Gold is soft and easily scratched, which might present an important issue for the long term use of gold substrates for droplet freezing.

Answer:

Since silicon became INA after the FIB treatment, another metal had to be chosen to apply the cavity-pattern. Gold (beside quicksilver and palladium) is the metal with the lowest standard free energy to form oxides, it is considered inert. That is why we chose gold to coat the silicon surface in first place and later on to produce a freezing chip completely made out of gold. The fact that gold has poor/zero INA is now stressed in the manuscript. However we did not investigate the INA of other metals. Small scratches are quickly introduced on the surface but they do not seem to influence the freezing behaviour. Damaging the cavity pattern by scratching the surface is still given. Therefore a freezing chip made out of a harder metal with the same ice nucleation inactivity will be part of further investigations.

Changed text on page 5 line 29 ff:

"Peckhaus et al. (2016) found no effect of a silicon substrate on ice nucleation. After the RIE-treatment, however, a shift of the freezing temperature of ultrapure water from -37,5°C to approx. -20°C was found. This shift might have been caused by a reaction of the etching agents with the silicon surface leading to an ice nucleation active compound. After the etching process, a gold layer (thickness 500 nm) was sputtered on top of the pattern, leading to an ice nucleation neutral surface. As an alternative to a gold sputtered silicon plate, a pure gold chip of similar dimensions was ion milled with a Focused Ion Beam (FIB) to introduce the same kind of pattern. Due to the thermodynamically stability of pure gold, no ice nucleation active compounds are formed on the surface during the introduction of the cavity pattern. Therefore no further treatments of its surface are necessary. If the surface of the gold sputtered silicon plate is scratched accidentally and the silicon is exposed, the chip becomes ice nucleation active again. Small scratches on the surface of the pure gold chip as well as the slight surface irregularities in the cavities were not found to have any influence on the INA. Anyway they have to be avoided to not damage the cavity pattern.”

Comment

2. Some more details on how the pure water was prepared would also be useful. Was the water just taken directly from the commercial Milli-Q water generator, with no additional filtration etc.? I think these are important details that are often not reported by the ice nucleation community, but the quality of the water can severely limit how low in temperature the system can go before “pure” water freezing starts to interfere. If the water was taken directly from the system, this is more evidence that it is the gold substrate that allows this system to achieve freezing temperatures of pure water very close to the homogeneous limit. This would provide further important evidence to the community that it is the substrate (such as commonly used hydrophobic cover slips) that is the main cause of the higher freezing temperature of pure water commonly observed in cold stage systems, as opposed to impurities in the water itself.

Answer:

For each set of experiment, the water was directly taken from the type 1 water generator (MilliQ Merck Simplicity 2012) and stored in a laboratory clear glass bottle for a maximum time of about 6 hours.

Changed text on page 6 line 23 ff:

“For each set of experiment, the water was directly taken from the generator and stored in a laboratory clear glass bottle for a maximum time of about 6 hours.”

Comment

3. I was surprised that none of Markus Petters' groups significant measurements from their cold stage were mentioned here. They have done a lot of work on validating their system, and developing protocols for analyzing the data, and have used both droplets immersed in oil and also droplets in air. The design and evaluation of other similar cold stage systems should also be discussed, as this will better convey the unique aspects of the system reported here (Budke and Koop, 2015; Mason et al., 2015; Petters and Wright, 2015; Tobo, 2016; Whale et al., 2015; Wright et al., 2013; Wright and Petters, 2013). Some important lessons regarding how to properly evaluate a cold stage system can also be learned from these other papers, and should be incorporated here to properly evaluate their system.

Answer:

The correct approach to evaluate a cold stage and to compare the data is now given. The ice nucleation active surface/mass site densities are now provided in Figure 6 and 8, compared, described and discussed. A more detailed evaluation of other cold stage systems is not provided, since manuscript is going to be submitted as a *technical note*.

Changed text on page 3 line 24 ff:

"The ice nucleation activity can be also well expressed by referring to the mass of INP per droplet (n_m) instead of the surface per droplet. This is often used when the surface of the investigated INP is not accurately quantifiable."

Changed text of chapters *Results* and *Conclusion*:

See the revised manuscript as uploaded separately as *author comment*.

Changed text on page 13 line 4 ff:

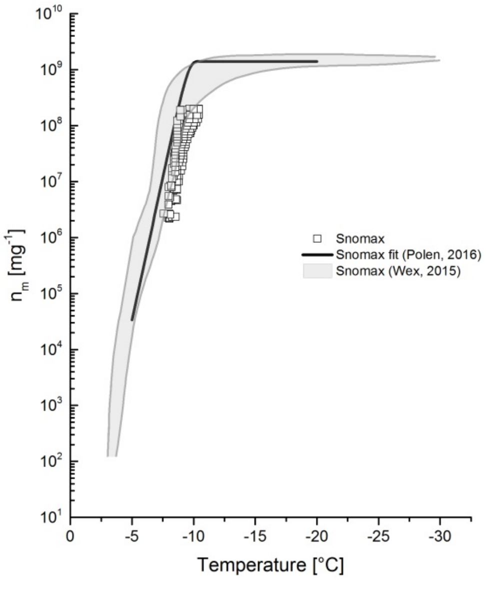

Figure 6 The ice nucleation active mass site density n_m of Snomax$^{®}$ determined with the freezing ship is in consistence with the results published by Wex et al. (2015) and Polen et al. (2016). A shift of the n_m values to lower temperatures due to degradation processes can be observed and is in agreement with Polen et al. (2016).

Changed text on page 14 line 6 ff:

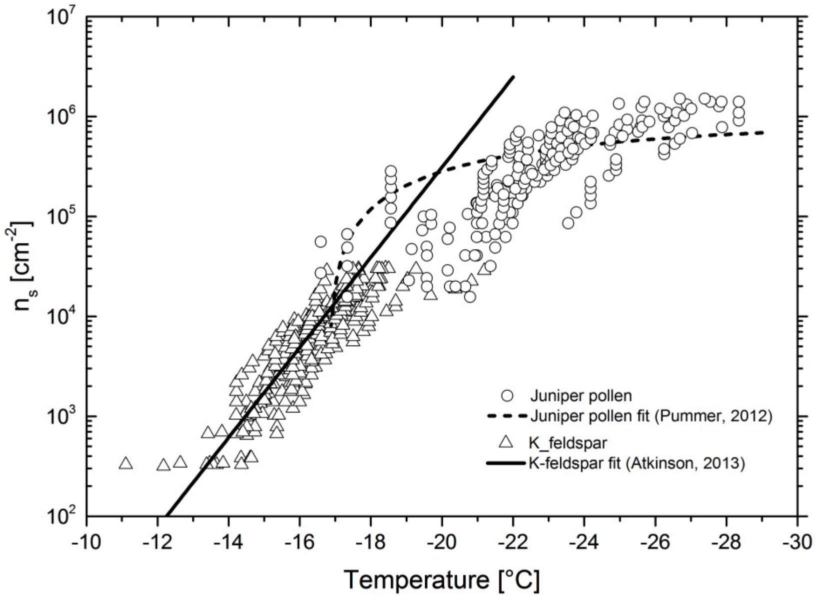

Figure 8 Comparison of ice nucleation surface site densities n_s of measurements done using the freezing chip with already published data. The n_s values of K-feldspar fit well to the published data of Atkinson et al. (2013). Minor deviations of the obtained juniper and birch pollen values compared to Pummer et al. (2012) can be seen.

Comment

4. The temperature control system design seems to be a simple and effective one, but very little is said regarding the accuracy and precision of the temperature control. There will be some lag and offset between the setpoint temperature and what is actually produced in the substrate where the droplets reside. How significant are these, and do they vary in time during the temperature cooling cycle? There might also be a significant temperature gradient across the gold substrate droplet array. These all need to be measured and discussed. Other groups usually validate their systems by also measuring the melting points of a series of compounds to test the accuracy of the temperature control of the droplets.

Answer:

The systems cooling rate is set to 2K/min between 0°C and -40°C. The targeted and actual temperatures are monitored constantly (online tracking). Within this cooling rate and temperature range, no delays, lags or offsets were observed. This was achieved by adjusting the PID controller and using a K-type thermocouple with an uncertainty of ±0,1°C.

Using the measured T_{50} values of ultrapure water and the homogeneous freezing temperature of 40µm water droplets at -37°C according to Pruppacher and Klett, (1997) as a reference, a standard deviation of the temperature of ±0,5°C was calculated (confidence level of 90%, ten degrees of freedom, t-distribution= 1,812). The evaluation is now mentioned in the manuscript.

No temperature gradients within the cold stage were observed when using multiple temperature measuring points simultaneously. Temperature differences within the observed area (~0,2mm^2) were not recorded.

The temperature range (about -5°C to -38°C) of heterogeneous ice nucleation was covered by investigated the chosen INPs with the freezing chip. The additional investigation of the melting point was not rated to show necessary additional validation of the setup/technique.

Changed text on page 5 line 19 ff:

“The uncertainty of the temperature was calculated by using the homogeneous freezing temperature of -37°C of water droplets with a diameter of 40µm calculated by Pruppacher and Klett (1997) using the classic nucleation theory, as a reference and comparing it to the obtained $T_{99,9}$ values (temperature where 99,9% of the droplets are frozen) of ultrapure water using the freezing chip. Applying a confidence level of 90%, a standard deviation of the temperature of ±0,5°C was calculated (t- distribution: 1,812). “

Comment

5. Similarly, what cooling rates can this system achieve, and what cooling rate is typically used? The cooling rate can have significant effects on the observed freezing temperature, and must be accounted for (Broadley et al., 2012; Mason et al., 2015; Wright et al., 2013).

Answer:

The used cooling rate is now given in the manuscript. Cooling rates between 0,1 K/min and 10K/min can be achieved.

Changed text on page 6 line 24 ff:

“The temperature control was set to a cooling rate of 2K/min for all measurements.”

Comment

6. An advantage of using isolated droplets is the ability to perform droplet refreeze experiments, which can provide important insights into the nature of the ice nucleants (Polen et al., 2016; Vali, 2008, 2014; Wright et al., 2013). It would strengthen this manuscript if its refreeze capabilities were also tested and assessed.

Answer:

The freezing chip offers a great technique for refreezing experiments, which will be the focus of a future study but are out of scope of the current manuscript.

Comment

7. The analysis and discussion of the actual droplet freezing temperature curves measured is frankly very thin, and does not provide a meaningful evaluation of the system's immersion freezing measurement capabilities. The authors are aware of the important effects of INP concentration on the measured freezing temperatures, but only discuss these effects qualitatively when comparing their results to published data. A proper quantitative analysis is require here so that results from different cold stage systems that use different droplet sizes and INP concentrations can be properly compared to this new system. The n_s framework could be used, as is commonly done in the IN community now (Hiranuma et al., 2015; Wex et al., 2015). n_s is attractive in its simplicity for accounting for how changes in total particle surface area change the observed droplet freezing temperature, but it does contain some significant issues when applied to droplet freezing measurements from a cold stage. It was recently reported that varying the INP in water concentration can cause the n_s values retrieved from cold stage measurements to change significantly (Beydoun et al., 2016). This means that n_s will not always properly normalize for changes in INP surface area, as it is intended to do. As different IN groups use different droplet volumes and INP concentrations, this may explain the persistent disagreement in n_s values obtained by different groups for the same INP system (Emersic et al., 2015; Hiranuma et al., 2015). Caution in using n_s is thus warranted, but the authors must quantitatively account for how different particle concentrations affect the measured freezing temperatures when comparing their results to literature data.

Answer:

The correct approach to evaluate the data is now given. The ice nucleation active surface/mass site densities are now provided in Figure 6 and 8 (see Comment 3), compared, described and discussed. Furthermore the limits of interpretation of freezing spectra via n_s described by Beydoun et al. (2016), is now mentioned in the manuscript.

Changed text on page 3 line 25 ff:

“Beydoun et al. (2016) showed that shifts to colder freezing temperatures caused by reducing the particle concentration or surface area present in the droplet, cannot be fully accounted for by normalizing to the available surface are or mass ($n_{s/m}$). However this needs to be accounted when the measurements are made in conjunction with just single-particle/atmospheric concentration analysis techniques.”

“A comparison of T_{50}, n_s and n_m values of the INPs investigated by using the freezing chip and previously published data is plotted in Figure 6, 8 and 9.

Changed text of chapters *Results* and *Conclusion*:

See the revised manuscript as uploaded separately as *author comment*.

Comment

8. Issues of particle coagulation and sedimentation when working at high particle concentrations are another concern in cold stage systems, and high concentrations are here (Beydoun et al., 2016; Broadley et al., 2012; Emersic et al., 2015). As this method only has an oil film at the top of the droplet perhaps the ice nucleants will always be available to induce freezing even if they settle to the bottom of the droplet. That would be a unique advantage of this approach that is worth evaluating and discussing.

Answer:

We could only speculate on coagulation and/or sedimentation of INP in the suspension, so we do not feel confident to discuss this interesting issue in the manuscript.

Comment

9. The analysis and discussion of the median freezing temperatures observed for the various systems tested here needs some major attention if these results are to be used to credibly evaluate the system's performance. First, the expected homogeneous freezing temperature expected for the droplet volumes used here should actually be calculated. I suspect the authors will find that their measured temperature is very close to that expected for the droplet sizes used here, but the droplet's volume must be accounted for.

Answer:

The comparison of homogeneous freezing temperatures of ultrapure water measured via the freezing chip, is now correctly related to the freezing temperature to 40µm droplets calculated by Pruppacher and Klett, 1997 (CNT) and is provided in Figure 9- Ultrapure water. For further evaluation, ice nucleation surface and mass site densities are now provided in Figure 6 and 8 (see above).

Changed text on page 14 line 1 ff:

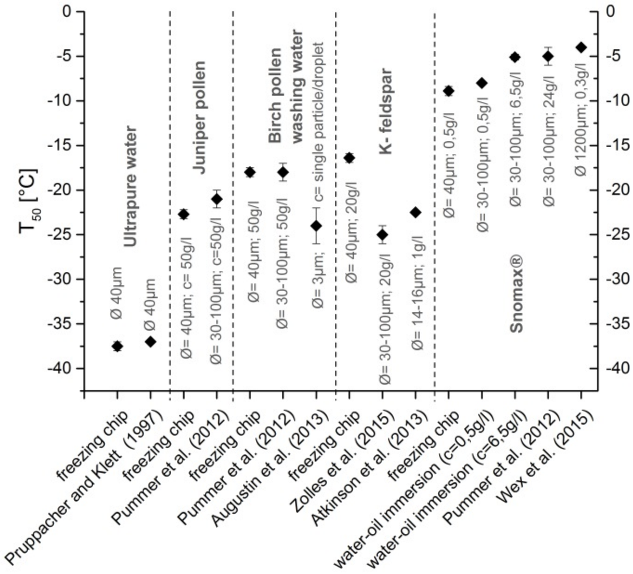

Figure 9 T_{50} values of several INPs compared with already published values. Diameters of the droplets and concentrations at each experiment are given in the figure.

Comment

10. The discussion of Snomax does not reflect our current state of understanding of this widely used ice nucleant. It is known that Snomax contains at least 3 different types of ice nucleants, and that this causes at least two different freezing temperature regimes to be observed, as the concentration of Snomax is varied (Pandey et al., 2016; Turner et al., 1990, 1991). More significantly, it was recently found that the most efficient ice nucleants in Snomax that induce freezing ∼-3 C degrade in time, while the ice nucleants that freeze ∼-8 C are stable (Polen et al., 2016). So the fact that Snomax was observed to freeze around -9 C here can be explained by the degradation of the more active ice nucleants, and/or the use of low concentrations that mean most droplets do not contain the rare more active ice nucleants. Given the high concentrations used here, I suspect it is the former, but the authors can confirm this by comparing the total amount of Snomax present in their droplet volumes with the range reported by (Polen et al., 2016).

Providing information about the age of their Snomax sample and how it was stored will also help to clarify this. Note that the instability of Snomax's ice nucleation properties makes it a risky choice as an ice nucleation standard for comparison to other immersion freezing systems. The IN community really lacks good INP standards. The much higher freezing temperatures observed for microcline here might be explained by the milling of the material, but this makes the use of this system to compare to other results rather meaningless, as they are not comparing the same INP type. The authors should test their system using a better behaved type of INP that has been studied by other groups, to properly evaluate their system. Illite NX (Hiranuma et al., 2015) or Arizona test dust (A1 Ultrafine fraction) are some good possibilities. Measuring melting points of different pure compounds should also be performed.

Answer:

Thank you for these critical and important information! Measurements done with Snomax are now more detailed compared/discussed with results published from Polen et al. (2016) and Wex et al. (2015). The age of Snomax is now provided. Furthermore surface/mass site densities of Snomax and K-feldspar are now provided to correctly compare the results of our study to those obtained by other authors.

Changed text on page 7 line 9 ff:

"Polen et al. (2016) reported that the most efficient ice nucleus in Snomax (induces freezing at about -3°C) degrades in time. Therefore the freezing temperature was expected to be at ~-8°C where the less active but more stable ice nucleus triggers ice formation."

Changed text of chapters *Results* and *Conclusion*:

See the revised manuscript as uploaded separately as *author comment*.

11. Figure 1 doesn’t really add to the paper and could be omitted.

Answer: Figure 1 was removed.

12. The roughness of the gold wells and surface shown in Fig. 4 is quite notable. Based on the very low freezing temperature observed for pure water it seems that this does not create ice active surface sites on the gold, but this should be discussed as it provides further evidence of the desirable (and unique?) properties of gold as a substrate for ice nucleation measurements.

Answer:

The surface roughness seems not to influence the freezing process. But no further investigations were done concerning the dependency of different roughnesses to the ice nucleation. The fact that gold has poor/zero INA is now stressed in the manuscript. Small scratches are quickly introduced on the surface but they do not seem to influence the freezing behaviour.

Changed text on page 6 line 33 ff:

“If the surface of the gold sputtered silicon plate is scratched accidentally and the silicon is exposed, the chip becomes ice nucleation active again. Small scratches on the surface of the pure gold chip as well as the slight surface irregularities in the cavities were not found to have any influence on the INA. Anyway they have to be avoided to not damage the cavity pattern.”

References

Atkinson, J. D., Murray, B. J., Woodhouse, M. T., Whale, T. F., Baustian, K. J., Carslaw, K. S., Dobbie, S., O'Sullivan, D., and Malkin, T. L.: The importance of feldspar for ice nucleation by mineral dust in mixed-phase clouds (vol 498, pg 355, 2013), Nature, 500, 491-491, 2013.
Beydoun, H., Polen, M., and Sullivan, R. C.: Effect of particle surface area on ice active site densities retrieved from droplet freezing spectra, Atmos Chem Phys, 16, 13359-13378, 2016.
Peckhaus, A., Kiselev, A., Hiron, T., Ebert, M., and Leisner, T.: A comparative study of K-rich and Na/Ca-rich feldspar ice-nucleating particles in a nanoliter droplet freezing assay, Atmos Chem Phys, 16, 11477-11496, 2016.
Polen, M., Lawlis, E., and Sullivan, R. C.: The unstable ice nucleation properties of Snomax (R) bacterial particles, J Geophys Res-Atmos, 121, 11666-11678, 2016.
Pruppacher, H. R., and Klett, J. D.: Microphysics of Clouds and Precipitation, Kluwer Academic Publishers, Dordrecht, 1997.
Pummer, B. G., Bauer, H., Bernardi, J., Bleicher, S., and Grothe, H.: Suspendable macromolecules are responsible for ice nucleation activity of birch and conifer pollen, Atmos Chem Phys, 12, 2541-2550, 2012.
Wex, H., Augustin-Bauditz, S., Boose, Y., Budke, C., Curtius, J., Diehl, K., Dreyer, A., Frank, F., Hartmann, S., Hiranuma, N., Jantsch, E., Kanji, Z. A., Kiselev, A., Koop, T., Mohler, O., Niedermeier, D., Nillius, B., Rosch, M., Rose, D., Schmidt, C., Steinke, I., and Stratmann, F.: Intercomparing different devices for the investigation of ice nucleating particles using Snomax (R) as test substance, Atmos Chem Phys, 15, 1463-1485, 2015.