

Interactive comment on “Highly active and stable fungal ice nuclei are widespread among *Fusarium* species” by Anna T. Kunert et al.

Anonymous Referee #2

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General Comments:

The authors report on *Fusarium* species ice nucleation activity utilizing two standard, well established methods. 112 strains from 65 species were analyzed using LINDA and TINA and identified 18 strains with initial ice nucleation activity between -3.5 and -12°C . It was further demonstrated that freeze-thaw cycles did not impact ice nucleation activity and filtration of the samples suggests that the proteinaceous compound responsible for *Fusarium* ice nucleation forms cell-free aggregates whose nucleation efficacy is impacted by size. Additional ozone and NO_2 studies were done to further evaluate the stability of the compounds in the atmosphere and showed no variation from untreated samples.

This work provides valuable insight into the biodiversity of ice nucleation active *Fusar-*
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ium species and aids in the understanding of ice nucleating particles and bioaerosols as a whole. This work is aligned with the scope of the journal and can be accepted after the noted changes have been addressed.

Specific comments:

Abstract: Indicate the biological relevance of *Fusarium* and its ice nucleation activity. This is discussed well in the introduction but will help to bridge the first few sentences of the abstract.

Methods 2.1: How were the initial samples obtained? Could their original environment (crop vs. airborne, etc.) shed light on IN frequency?

Line 21: Additional, more recent, studies have contributed to this understanding of IN as well. (Failor et al. 2017, Hanlon et al. 2017, Stopelli et al. 2017, 2015, Joly et al. 2014).

Line 24-6: Failor et al. (2017) further expanded on known gammaproteobacteria IN.

Line 118: Was the range of incubation times necessary to reach a specified optical density? If so, that indication would be useful. If not, elaborate of reasoning for the times.

Line 119: Be specific for the 0.5°C freezing point depression. Is it 0.5°C or $0.5\pm x^{\circ}\text{C}$.

Results 3.1: This would be an interesting point to note the original sampling locations for the various strains and could further demonstrate the cosmopolitan nature of these IN-active species should any tends be identified.

Lines 154-5: This is a risky assumption to make. Prior to the Failor et al. study, all bacterial IN were thought to be proteinaceous. Exposing a selection of the species to high heat could support this claim.

Lines 184-6: With the drastic decrease in activity after the 300,000 MWCO filter and then again after 100,000, could the protein not be larger, but when damaged or broken

still retains some ice nucleation activity?

Lines 195-6: Why would single proteins in the atmosphere be unlikely? Please elaborate on this statement.

Line 216: Change "...and the fungus could safe energy." to "...and the fungus could save energy."

Figure 1. Inclusion of the positive control SnoMax curve would be beneficial here. Any incidence of spontaneous freezing of the negative control should also be noted (if any occurred with the methods you used).

Figure 3. You note in the text that SnoMax has been shown to decrease after exposure. Did you see this same result, or did you not use SnoMax because of this interaction?

References:

Failor, K.C., Schmale III, D.G., Vinatzer, B.A., and Monteil, C.L.: Ice nucleation active bacteria in precipitation are genetically diverse and nucleate ice by employing different mechanisms, *ISMEJ*, 11(12), 2740–2753, doi:10.1038/ismej.2017.124, 2017.

Hanlon, R., Powers, C., Failor, K.C., Monteil, C.L., Vinatzer, B.A., and Schmale III, D.G.: Microbial ice nucleators scavenged from the atmosphere during simulated rain events, *Atmospheric Environment*, 163, 182-189, doi:10.1016/j.atmosenv.2017.05.030, 2017.

Joly, M., Amato, P., Deguillaume, L., Monier, M., Hoose, C., and Delort, A.-M.: Quantification of ice nuclei active at near 0°C temperatures in low-altitude clouds at the Puy de Dôme atmospheric station, *Atmospheric Chemistry and Physics*, 14, 8185-8195, doi:10.5194/acp-14-8185-2014, 2014.

Stopelli, E., Conen, F., Morris, C. E., Herrmann, E., Bukowiecki, N. and Alewell, C.: Ice nucleation active particles are efficiently removed by precipitating clouds, *Scientific Reports*, 5, 16433, doi:10.1038/srep16433, 2015.

Stopelli, E., Conen, F., Guilbaud, C., Zopfi, J., Alewell, C. and Morris, C. E.: Ice nu-

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cleators, bacterial cells and *Pseudomonas syringae* in precipitation at Jungfrauojoch, *Biogeosciences*, 14(5), 1189–1196, doi:10.5194/bg-14-1189-2017, 2017.

Interactive comment on *Biogeosciences Discuss.*, <https://doi.org/10.5194/bg-2019-276>, 2019.

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