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Subpollen particles (SPP) of birch as carriers of ice nucleating macromolecules

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Wind pollinated trees such as birch trees release large amounts of pollen to the atmosphere during their blooming season in early spring. Due to the large size of pollen (birch pollen diameter: 20-25 μm) and short residence time in the atmosphere, their impact on cloud formation was believed to be negligible. However, in recent years studies have shown that ice nucleating materials, so called ice nucleating macromolecules (INM), much smaller in size can be extracted from pollen. At the same time there is evidence from medical studies that pollen can rupture under conditions of high humidity in the atmosphere and expel cytoplasmic material including starch granules, commonly referred to as subpollen particles (SPP). INM or SPP are much smaller and potentially more numerous than pollen and could significantly affect cloud formation in the atmosphere.

In this study, we focus on birch pollen and investigate the relationship between pollen grains, INM and SPP. According to the usage of the term SPP in the medical field we define SPP as the starch granules contained in pollen grains. We develop an extraction method to generate large quantities of SPP and investigate their ice nucleation activity. To our knowledge, this is the first study to investigate the ice nucleation activity of isolated SPP. We show that INM are only loosely attached to SPP and that purified SPP are not ice nucleation active: after several times of washing SPP with ultrapure water the ice nucleation activity completely disappears. In addition, we study the chemical nature of the INM with fluorescence spectroscopy and quantify the protein concentration with the Bradford assay. Fluorescence excitation-emission maps indicate a strong signal in the protein range (maximum around $\lambda_{\text{ex}} = 280 \text{ nm}$ and $\lambda_{\text{em}} = 330 \text{ nm}$) that correlates with the ice nucleation activity. In contrast, with purified SPP this signal is lost. The protein concentration ranges from $77.4 \mu\text{g mL}^{-1}$ for highly concentrated INM to below $2.5 \mu\text{g mL}^{-1}$ for purified SPP. The results thereby indicate a linkage between ice nucleation activity and protein concentration. Purified SPP are not ice nucleation active but could, however, act as carriers of INM and distribute those in the atmosphere.