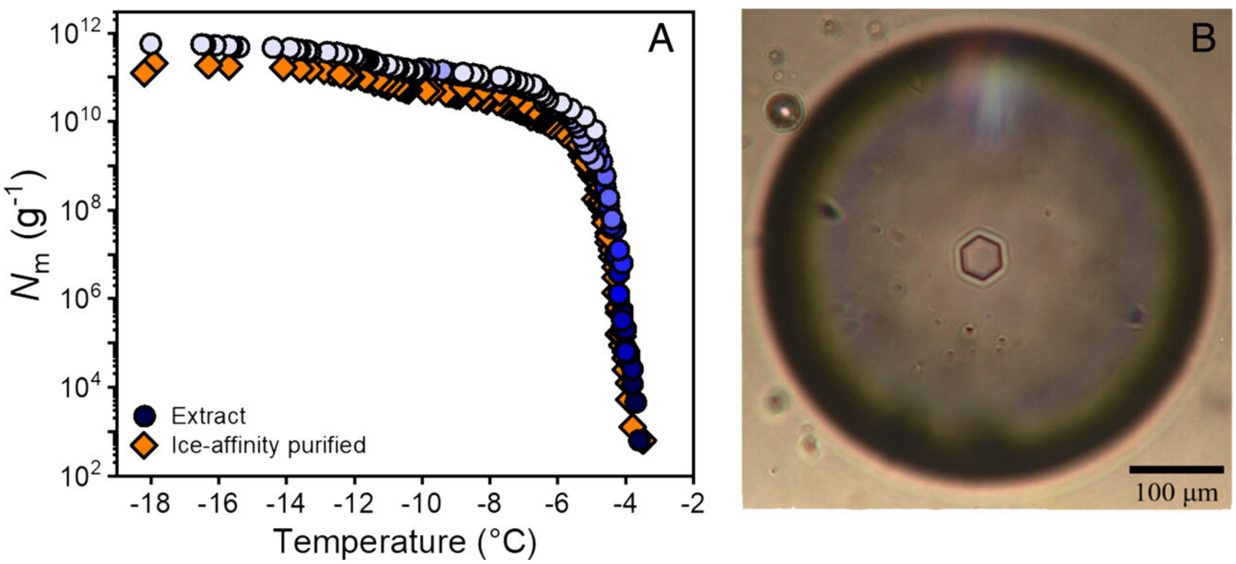
Forming ice: There's a fungal protein for that

November 13 2023



Freezing experiments of aqueous extracts containing fungal ice nucleators from *F. acuminatum*. (*A*) Shown is the cumulative number of ice nucleators per unit mass of *F. acuminatum* (N_m) for extracts containing ice nucleators from spores and mycelial surfaces (blue) and for ice-affinity purified ice nucleators (orange). (*B*) Cryomicroscopic image of a hexagonal ice crystal grown in a *F. acuminatum* IN extract. Credit: *Proceedings of the National Academy of Sciences* (2023). DOI: 10.1073/pnas.2303243120

The way ice forms is a lot more interesting than you think. This basic physical process, among the most common in nature, also remains somewhat mysterious despite decades of scientific scrutiny.

Now, new research from the University of Utah, with Germany's Max Plank Institute for Polymer Research and Idaho's Boise State University, is shedding fresh light on the role of biological agents—produced by fungi of all things—in ice formation.

Contrary to what we have been taught in school, water won't necessarily freeze at 0 degrees Celsius (32 degrees F) because of the energy barrier inherent in phase transitions.

Completely pure water won't freeze until it cools to as low as –46 C. Water molecules require particles on which to build the crystals that lead to ice, a process called nucleation. Organisms have evolved various ways to control ice formation as an adaptation to survive in cold environments.

So, the most efficient ice-nucleating particles are biological in origin, produced in bacteria and fungi, or even by insects, but the molecular basis and precise mechanisms of "biological ice nuclei" have not been well understood.

A fungus' ability to control ice

Valeria Molinero, a theoretical chemist with the University of Utah's College of Science, is at the forefront of sorting out this mystery, which holds potential implications for improving our understanding of how life affects precipitation and climate.

In a new study she co-led, an international team of researchers explores the characteristics and properties of fungal ice nucleators, revealing that they are made up of small protein subunits and play a role in both promoting and inhibiting ice growth.

"They are proteins that are excreted to the environment and these particles are extremely effective for ice nucleation," said Molinero, director of the university's Henry Eyring Center for Theoretical Chemistry. "But the way the organism benefits from these ice nucleation abilities is not known and it doesn't exist in all the possible variants of the organism. We don't know why they form ice or whether there's an advantage."

The multidisciplinary team homed in on a species of fungus called Fusarium acuminatum and discovered it produces ultra-minute proteins that aggregate into larger particles. Their findings are published this week in the *Proceedings of the National Academy of Sciences*.

According to co-lead author Konrad Meister, the mechanism of forming larger aggregates from smaller building blocks is found in other organisms besides fungi.

"Nevertheless, we were surprised by the small size of the fungal protein building blocks compared to their efficiency," said Meister, a professor of chemistry at Boise State. "Other known and similarly efficient ice-making proteins from other organisms, for example, are 25 times larger."

How organisms evolve in different ways to achieve the same outcome

Bacteria and fungal proteins can spur ice formation at temperatures as warm as -10 to -2 degrees. Some bacteria are so effective at promoting ice that they are put to work in products that ski areas use for snowmaking.

Molinero is intrigued that so many different kinds of organisms have evolved similar ice-nucleating capabilities that she originally titled the paper "E pluribus unum," meaning "out of many, one," but the journal insisted they drop the Latin.

"If you look across kingdoms that can nucleate ice, there are insects, lichen, bacteria, and fungi. All of these seem to have evolved independently, very potent ice nucleants" she said. "All of the ice nucleation in nature that is extremely effective seems to be done by proteins, although the size of the individual ice nucleating proteins vary a lot among organisms."

The ecological advantages of ice nucleation and its role in cloud formation and precipitation are not yet fully understood and pose a significant gap in our grasp of the interplay between climate and life, according to the study. The research can improve the efficiency of the food-freezing process, snowmaking, and cloud seeding.

However, with the team's discoveries come many more questions, such as why and how these proteins aggregate.

"The other question is whether they're doing this on purpose or is it just that there's a protein that they produce for something else, but it has this property," said Molinero.

Resolving these fundamental questions will require teamwork, bringing together investigators with expertise in various areas of chemistry, biophysics, and biology.

"This is the positive message. Solving the puzzle of biological control of ice formation drives scientists to collaborate," Molinero said. "Each of us has a piece of the knowledge, but all together we can do so much. It's been fun."

More information: Ralph Schwidetzky et al, Functional aggregation of cell-free proteins enables fungal ice nucleation, *Proceedings of the*

National Academy of Sciences (2023). DOI: 10.1073/pnas.2303243120

Provided by University of Utah