Independent Cryonics Educators Program

4.1: Minimizing ice crystals using vitrification

Vitrification is when a liquid becomes a glass-like solid without ice crystal formation.

A **cryoprotectant solution** reduces ice formation.

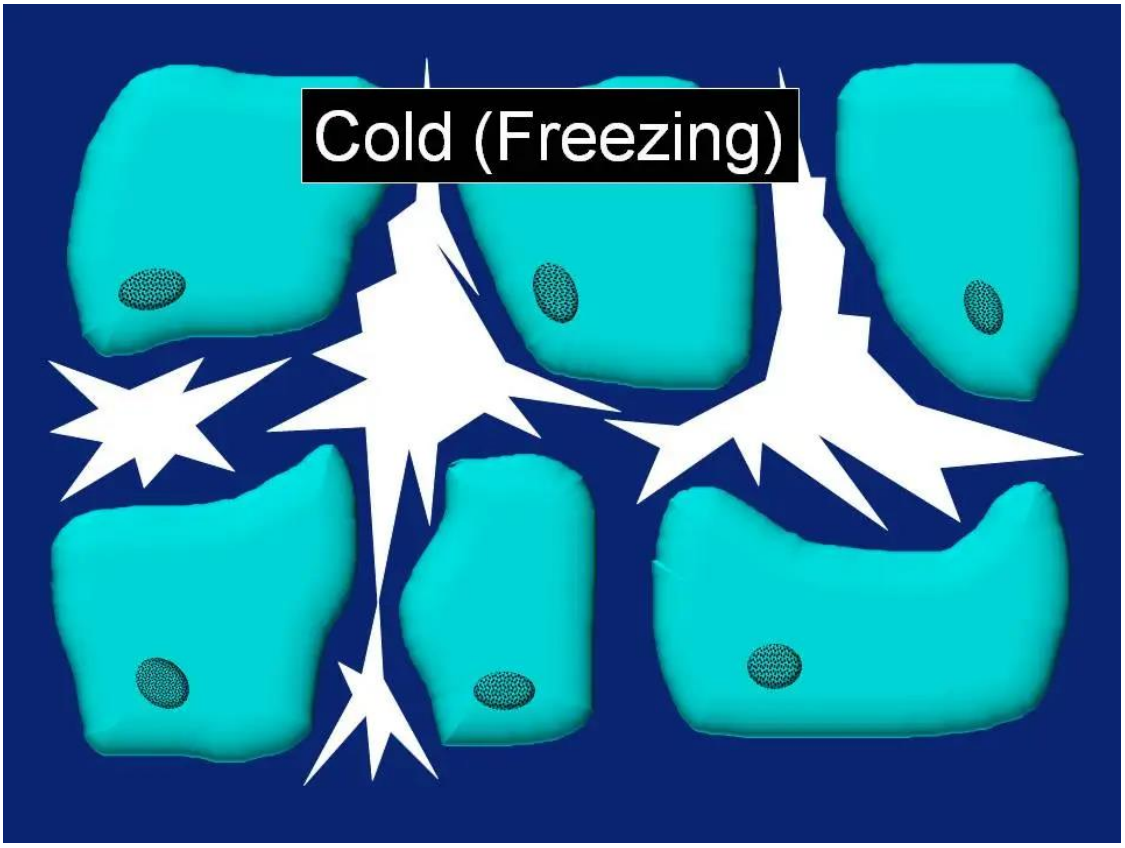
A **vitrification solution** consists of cryoprotective agents that is sufficiently concentrated to enable complete or almost-complete vitrification of tissue at the cooling rates employed for that purpose. [See Explainer 3.3]

Freezing implies the formation of ice crystals out of the water. That water is part of a solution with other molecules. When tissue is cooled below the freezing point, water molecules come together and form ice crystals. Ice crystals can cause some damage to cell membranes. However, most of the damage done by ice results from the way ice forms outside the cell, dehydrating and shrinking the cell. This concentrates other molecules into a harmful solution. Cells need a very specific environment to remain viable.

As ice forms, water expands by only a few percent. Examined on a cellular level, ice first forms outside cells during cooling. As water leaves the cells ("osmotic dehydration") and forms ice between cells, the cells dehydrate and shrink. As this process continues, ice crystals squeeze the cells, causing physical damage.

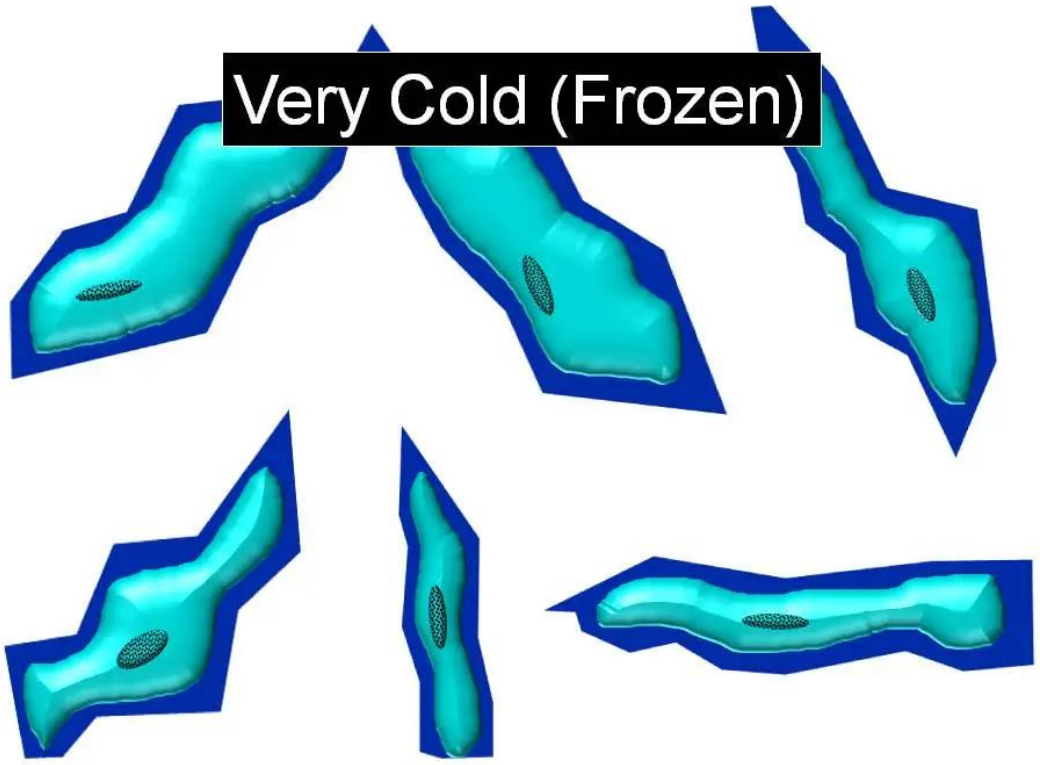
When talking about cryonics, people often say that we *freeze* people. If that is taken to imply the formation of ice crystals, it is not that simple. Although that sometimes happens to varying degrees, it is not the aim of good cryonics practice. To the extent that freezing occurs in a cryonics patient, it is due to factors beyond control. These include unavoidable delays in accessing the patient and beginning the procedure; poor circulation; and medical conditions such as aneurysm or stroke that impede the flow of cryoprotectants into the brain.

It is important in cryonics to prevent water from turning into ice. This is challenging because the human body consists of approximately 60% water. The percentage varies between different tissues in the body. Muscles contain more water than fatty tissue. Your brain, lungs, heart, liver, and kidneys are 65-85% water. Water makes up about half of your blood volume.



Now, consider what happens when we introduce cryoprotectants – small molecules that easily penetrate inside cells and that depress the freezing point of water. When cryoprotectants are used, the freezing point of the unfrozen solution drops sooner and faster, limiting the total amount of ice that forms and eventually stops forming. Cryoprotectants come in differing concentrations.

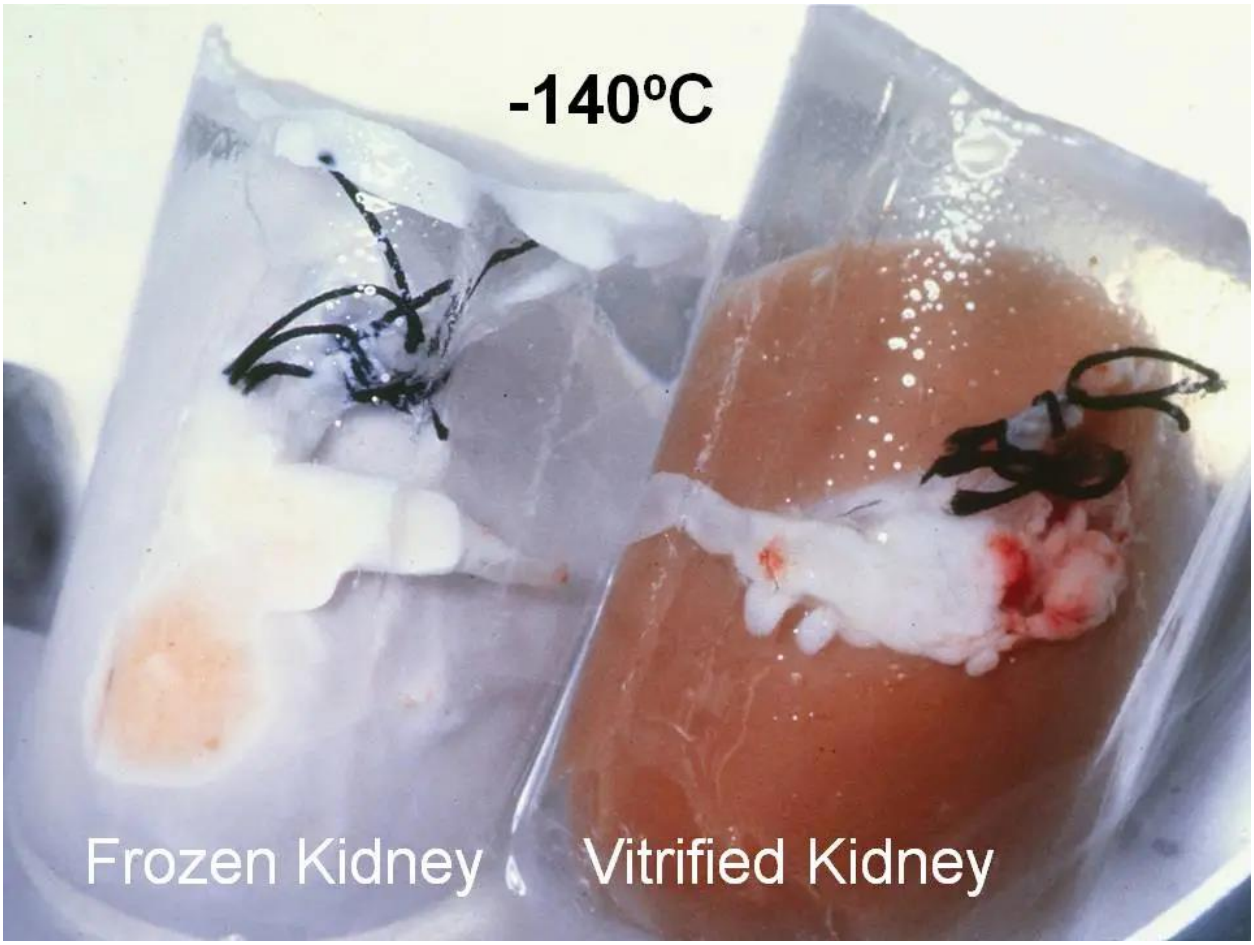
Up until around 2000-2002, Alcor used cryoprotectants with increasingly high concentrations. Every time a more concentrated solution was used, less ice formed. Alcor now uses a **vitrification solution**. When the high concentration of a vitrification solution is combined with rapid cooling, ice formation *can* be completely avoided. (This is not to say that full vitrification *will* be achieved in every case or even most cases.)



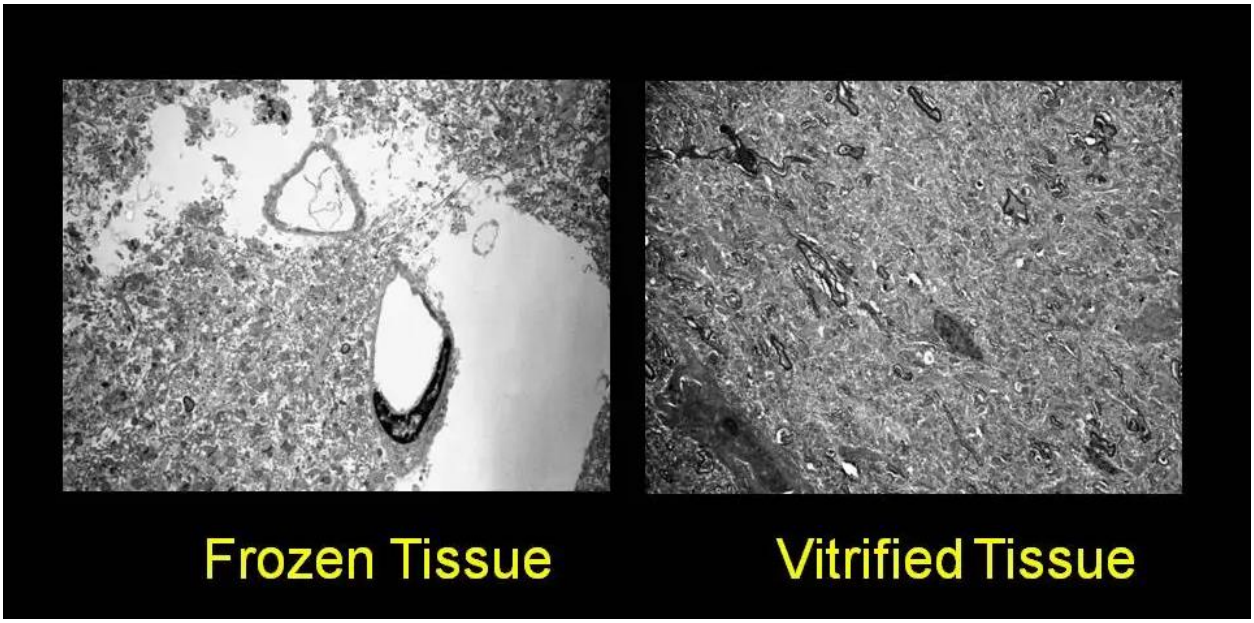
Vitrification means that water becomes solid without freezing. A vitrification solution is a solution of cryoprotective agents that is sufficiently concentrated to enable complete or almost-complete vitrification of a living system at the cooling rates employed for that purpose.

The combination of rapid cooling and high cryoprotectant concentration to completely avoid ice formation was first suggested in the 1984 paper, “Vitrification as an Approach to Cryopreservation”. When a vitrification solution is used, molecules just move slower and slower as they are cooled. As the temperature drops below -100° C (the “glass transition temperature”), molecules become locked in place and a solid is formed. Once cells are vitrified, cellular activity stops. This prevents any future damage to the tissue. Something that has been vitrified can theoretically be stored for almost any length of time.

Entire organs have been vitrified and stored at temperatures as low as -140° C. Among tissues that have been successfully vitrified are embryos, ova, skin, corneas, pancreatic islets, blood cells, and blood vessels. A rabbit kidney has been vitrified at -135°C and successfully transplanted with long term survival. Vitrification is now widely regarded as the most promising approach for long-term banking of organs. The image below shows the difference between a frozen kidney and a vitrified kidney.



Scientists are working to reduce the toxicity of the cryoprotectants used to make tissues vitrify. This would allow the banking of organs for transplantation. At Alcor, toxicity does still occur with vitrification of human organs, but we anticipate this to be reversible with future molecular repair technology.



[09/19/22]

References & Resources

https://www.alcor.org/library/what-is-vitrification/

“Vitrification as an approach to cryopreservation”, G. M. Fahy, D. R. MacFarlane, C. A. Angell, H T Meryman. *Cryobiology*, 1984 Aug;21(4):407-26.

“Physical and biological aspects of renal vitrification”, Gregory M. Fahy,* Brian Wowk, Roberto Pagotan, Alice Chang, John Phan, Bruce Thomson, and Laura Phan. *Organogenesis* 5:3, 167-175; July/August/September 2009.

“M22 Implementation”
https://www.alcor.org/2005/10/m22-implementation/

“Cryoprotectant Toxicity: Facts, Issues, and Questions,” Ben Best. *Rejuvenation Research*, 2015 Oct 1; 18(5): 422-436.
https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4620521/#:~:text=High%20levels%20of%20penetrating%20cryoprotectants,increasingly%20toxic%20as%20concentration%20increases

Next: 4.2: Cerebral ischemia: the 4–6-minute brain-death myth

---

ICE Program Index

Independent Cryonics Educators Program - Alcor
