CryoLetters **42 (1)**, ii-iii (2021)

EDITORIAL: COMING INTO PERSPECTIVE

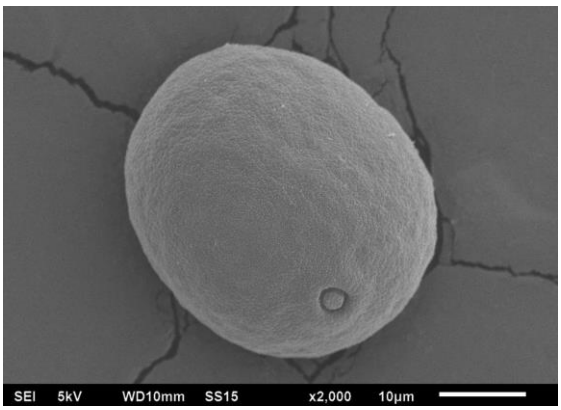
Hugh W. Pritchard

Executive Editor, CryoLetters

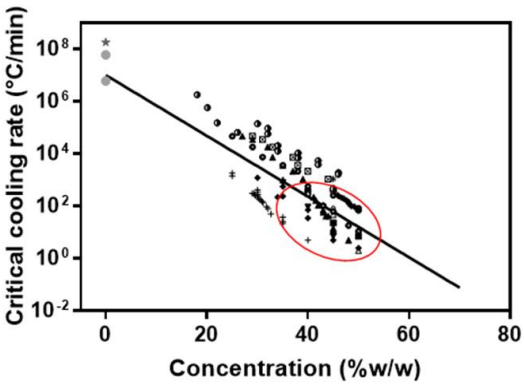
CryoLetters launched its series of PERSPECTIVES articles in the middle of 2020. They aim to provide an overview of recent developments in cryobiology and cryobiotechnology that will appeal to a wide readership. These mini-reviews are available as Open Access articles, at no cost to the authors. The first five PERSPECTIVES, that are briefly reviewed here, covered mechanisms of protection, nanotechnology for protection, cooling and warming dynamics and pollen banking.

Pollen preservation is an important tool for the maintenance of plant genetic resources and can promote improved efficiency in breeding programs and support germplasm conservation and exchange. For example, the technique has the potential to overcome challenges such as flowering asynchrony between different parent genotypes, or when insufficient pollen is available in nature. Dinato et al. (1) explain that pollen of many plant species have already been successfully cryopreserved in liquid nitrogen. Similar to seeds, maintaining pollen viability after exposure to ultra-low temperatures demands adjustment of the sample moisture so that freezable water is removed. Methods are summarised for the successful pollen cryopreservation for representatives of 30 plant genera. Examples cover forestry, horticultural and agricultural species, cryo-stored for up to four years.

For plant propagules that tolerate drying, such as the pollens of many species, the rates of cooling and rewarming are not particularly critical for cryo-success. However, this is not the case when manipulating usually hydrated cells to avoid ice crystallisation and successfully enter the vitrified state. As Han and Bischof (2) show, the critical rate for cooling is highly dependent on the cryoprotective agent (CPA) concentration. Amongst a broad range of CPAs, as concentration increases from 20 to 50%, the critical cooling rate (CCR) that should be exceeded falls by about three orders of magnitude. Similar dependencies are evident for critical warming rates (CWR). The authors suggest a convenient mathematical expression for CCR and CWR that can guide general use for developing cryoprotective



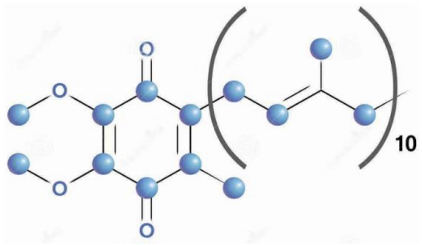
Pollen of Bahia grass, *Paspalum notatum* (1)



Relationships between CCRs and CPA concentrations (2)

protocols, and also highlight the critical need for further studies on CPA cocktails and tissue systems in which CPAs may behave differently and/or may not be fully equilibrated to the loaded CPA.

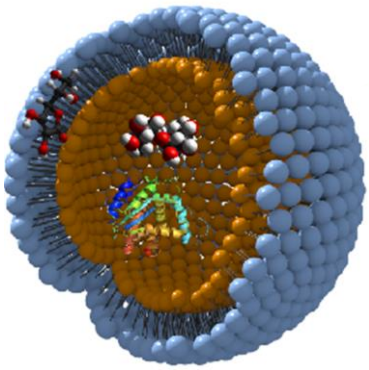
The choice of CPA is made in relation to the properties of cell types for preservation. Sperm cryopreservation underpins artificial insemination (AI) within elite livestock breeding programmes, and the protection of endangered



Co- coenzyme Q10 (3)

species. However, cooling often has some detrimental effects on sperm, including reduced motility. As Appiah et al. (3) note, micro-domain changes in the membrane on cooling can result in impaired function and this could be counteracted by the presence of coenzyme Q10. Consequently, treatment of sperm with this vital lipophilic molecule has been attempted and found to improve sperm quality of various species during cryopreservation and the cooled-stored condition. Although it remains unclear exactly how this antioxidant protects sperm, the authors highlight potential protective mechanisms of CoQ10.

Delivering CPAs to their target site of action is now a focus of nanotechnology development. Stewart et al. (4) review nanoparticle-mediated delivery of cryoprotectants for action during cooling down and warming back. An example is the use of synthesized poly(N-isopropylacrylamide-co-butyl acrylate)-based cold responsive nanoparticles for the intracellular delivery of trehalose and successful cryopreservation of human adipose-derived stem cells. Cells could take up the trehalose-laden nanoparticles at 37°C and trehalose could be released out of the nanoparticles inside cells during the cooling process of cryopreservation.



Polymeric nanoparticle encapsulation (4)

Examples of nanotechnology and antioxidant delivery to protect sperm during cryopreservation (5)

Active molecule	Nano-technology type	Function during cryopreservation
Ellagic acid	Liposomes	Improved motility; membrane function and viability.
Cholesterol	Cyclo-dextrin	Protection against oxidative damage.
Vitamin E	Poly-ethylene glycol	Protection during chilling at 4°C

Nanoparticle-mediated delivery of trehalose can be used in combination with other techniques, such as hydrogel encapsulation and magnetically heating (4). Taouzinet et al. (5) describe the wide range of methodologies now being developed and tested for the delivery of cryoprotectants and other protective compounds, particularly those impermeable to the cell membrane. Based on examples given for eight species, sperm cryopreservation has benefitted from the nanotechnology delivery of a range of active molecules, including ellagic acid, cholesterol and vitamin E.

REFERENCES

1. Dinato NB, Santos IRI, Vigna BBZ, de Paula AF and Fávero AP (2020) *CryoLetters* **41** (**3**), 115-127.
2. Han Z & Bischof JC (2020) *CryoLetters* **41** (**4**), 185 – 193.
3. Appiah MO, Asante-Badu B, Zhao J, Liu H, Wang J & Lu W (2020) *CryoLetters* **41** (**5**), 246- 256.
4. Stewart S, Arminan A & He X (2020) *CryoLetters* **41** (**6**), 308 – 316.
5. Taouzinet L, Fatmi S, Lahiani-Skiba M, Skiba M & Iguer-Ouada M (2021) *CryoLetters* **42 (1)**, 1-12 (2021).