Influence of DMSO on crystallization of water at low temperatures and its impact on cell survival

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The processes of solidification and melting of water-DMSO mixtures (up to 10% vol. of DMSO), which represent a cryopreservation medium for freezing biological cells,[1] were investigated. The positron annihilation lifetime spectroscopy allows to monitor the temperature dependence of the lifetime of the orthopositronium probe, which is sensitive to phase changes or changes in structure. It has been shown that with increasing concentration of DMSO in water, there is an increased induction of the amorphous fraction in the freezing mixture.[2] However, this fraction recrystallizes during slow heating, which is manifested by hysteresis between the temperature-dependent lifetime of the ortho-positronium probe during heating and cooling. This recrystallization can be avoided by rapid heating. We did the same experiment for 10% DMSO in water in the presence of a lipid bilayer, which represents a biological membrane model whose integrity is compromised during water crystallization. The presence of recrystallization induced by slow heating in the temperature range -110 °C to -60 °C was confirmed (Fig.1). This temperature range is bounded by two thermal events, namely the glass transition of water/DMSO mixture and the melting of eutectic water-DMSO complexes. In the past, decreased cell viability was reported when heating through this temperature range was slow (5 K/min), which was also explained by recrystallization of water.[3] The ability of DMSO to amorphize water is an advantageous property as this form of frozen water does not pose a risk to membrane systems and can form a protective layer around the cells regarding ice crystals.

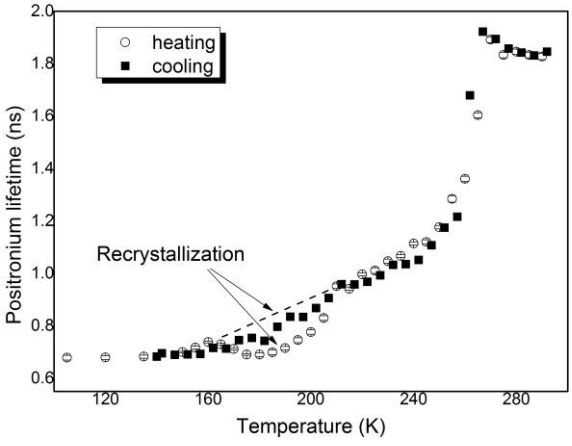

Fig.1. 10 % DMSO with lipid bilayer

[1] J. Lakota and P. Fuchsberger, *Bone Marrow Transplantation* **18(1)**, 262-3 (1996)
[2] K. Čechová, I. Maťko, J. Rusnák et al., *RSC Advances* **9**, 34299-34310 (2019)
[3] W. F. Rall, D. S. Reid and C. Polge, *Cryobiology* **21(1)**, 106–121 (1984)