Summary of the doctoral thesis mgr inż. Przemysław Smakulski

A use of the cryogenic particulate spray cooling method for biological material long-term preservation

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A biological material long-term preservation have to deal with many potential injuries caused mechanically and thermally during freezing process. The most dangerous from the perspective of cryopreservation are mechanical injuries caused by intra- or extracellular ice formation. Nowadays research on cryopreservation effectiveness goes into direction of new freezing techniques development, minimizing damage caused by excessive ice formation.

One of the proposed method is to use the micro-solid particulate spray for ultra-fast freezing of the biological material with extracellular ice formation (so called slow freezing method), or without creation of ice (glass freezing – vitrification method). Micro-solid particulate spray, among other methods of plunging a probe with specimen directly into a liquid nitrogen, omits a problem with poor heat transfer by film boiling in neuralgic for cells temperature level. In that way is possible to achieve higher heat transfer coefficients, what's directly influence on freezing process quality.

Following work includes a research on usage possibility of micro-solid nitrogen particulate spray for long-term preservation of the biological material.

In order to determine the ability of freezing by "slow freezing" as well as vitrification method, the tests series inside the Institute of Fluid Science Tohoku University (Japan) have been performed. Experimental tests allow to determine the quench temperature characteristics and to compare the resultant heat fluxes with boiling curve for pool boiling of liquid nitrogen. This results show approx. three times higher values of heat flux in comparison to liquid nitrogen in film boiling regime.

In the slow freezing method mass diffusion of a salt in saline solution plays a significant role. Salt concentration grows into direction of ice front propagation. In consequence, it causes the salt mass concentration difference between space intra- and extracellularly. That difference in chemical potential, allows the inner cell water to outflow via semipermeable cell's membrane, which is impermeable for salt. Thanks to that feature is it possible to achieve an equilibrium state for which internal space of the cell remains unfrozen in cryogenic temperatures.

Taking into consideration average size of the single cell (10⁻⁵ m) and difficulties to detect salt concentration changes inside the saline solution, the mathematical modelling of this process has been proposed. The mathematical model includes ice front propagation together with diffusive mass transport of the salt and cell membrane resultant movement. The model based on OpenFOAM numerical library with implementation of Phase Field Method was built.

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