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## **Cryopreservation for the cell selection using water channels AQPs combination with ultra-quick freezing**

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Cryopreservation has been only empirically related to cell type and freezing conditions such as cooling rates and freezing solutions. In this report, we found that membrane damage caused by ultra-quick freezing is rescued by expression of aquaporin (AQP), leading to freezing tolerance. It is suspected that AQP can contribute to the improvement in cryopreservation by increasing membrane water permeability even under very low temperatures since AQP-mediated facilitated diffusion of water molecules is temperature independent. It was then examined if the freezing tolerance associated with AQP expression could be applied as a cell selection method. The cells expressing AQP either exogenously or endogenously were exclusively selected after ultra-quick freezing and thawing. Finally it is showed that CHO cells transiently transfected with Endothelin receptor A and AQP4 were also selected and concentrated by multiple cycles of freezing/thawing, which was confirmed with calcium imaging in response to endothelin. Taken together, it is proposed that this simple but efficient and safe method may be applicable to the selection of mammalian cells for applications in regenerative medicine as well as cell-based functional assays or drug screening protocols. Interestingly, also was discovered that multiple steps of ultra-quick freezing/thawing exclusively selects only stably transfected cells, which imply that stably transfected cell lines can be mimicked by this strategy. Taken together, this simple but efficient and safe method may be applicable to the selection of mammalian cells for applications in drug screening protocols as well as cell-based functional assays.

### **Biography**

Yasuhiro Kato has completed his PhD at the age of 27 years from Nagoya University and postdoctoral studies from JSPS (Japan Society for the Promotion of Science) and Tokyo Dental College. He is the assistant professor at Keio University School of medicine. He has published some papers for function of membrane protein and development of cell freezing methods using Aquaporin water channels.

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