

# Anti-ice nucleation peptide applied for cell stock solution

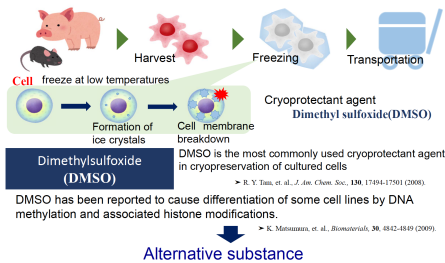
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## Introduction

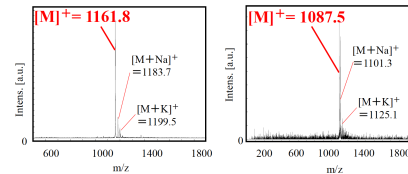
Generally, cells or tissue are transported frozen



## Characterization of synthetic peptide

GL and GPL peptides were synthesized by solid phase peptide synthesis procedure. These peptides were synthesized on Alko-PEG resin using a handmade standard manual Fmoc-protocol with a DMT-MM activation procedure.

GLLGPLGPRGLL (C<sub>54</sub>H<sub>93</sub>N<sub>15</sub>O<sub>12</sub>) Fw=1162.4 **GP peptide**  
 GPLGPLGPL (C<sub>52</sub>H<sub>84</sub>N<sub>12</sub>O<sub>12</sub>) Fw=1087.3 **Control**



## Cell cytotoxicity test

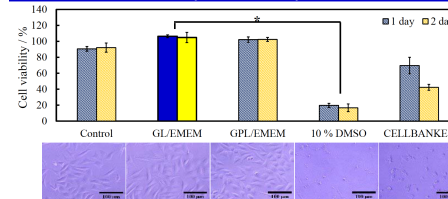
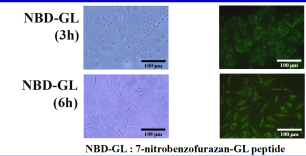
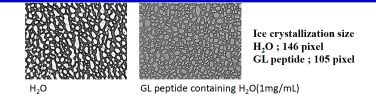


Fig. 9 Results of cell viability and cell morphology (Incubation time 2days)  
 Cell density: 1.0 × 10<sup>5</sup> cells/mL  
 Control: EMEM (-)  
 GL/EMEM and GPL/EMEM showed **no cytotoxicity** compared to DMSO containing media.  
 Control: EMEM (-)  
 GL/EMEM: 4.0 mg/mL GLEMEM (-)  
 GPL/EMEM: 4.0 mg/mL GPL/EMEM (-)  
 10% DMSO: 20% DMSO/EMEM (-)  
 CELLBANKER: CELLBANKER

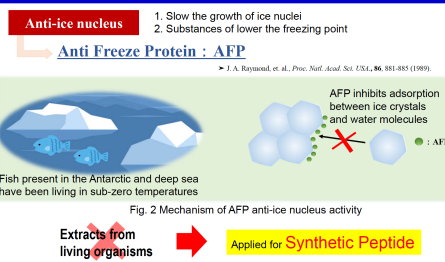
## Effect of anti-ice nucleus activity peptide



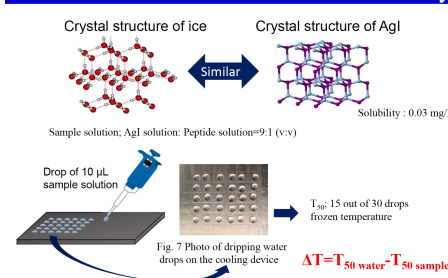
## Ice recrystallization inhibitory activity



## Introduction



## Measurement of anti-ice nucleus activity



## Refrigerated storage test (L929)(cell suspension)

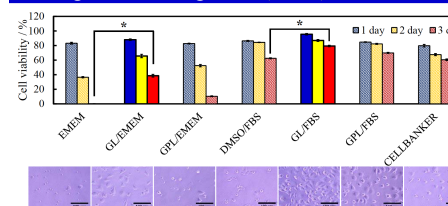
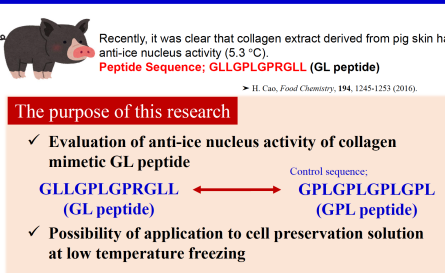


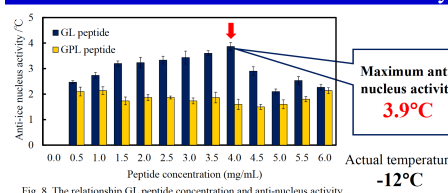
Fig. 10 Results of cell viability and cell morphology refrigerated in cell suspension system (Incubation time: 2 days)

- The addition of GL peptide significantly improved cell viability.
- L929 refrigerated with the addition of GL peptide maintained its proliferative capacity.

## Introduction



## Measurement of anti-ice nucleus activity



The GL peptide and the extract from collagen had same anti-ice nucleus activity.

## Refrigerated storage test (hMSC)(cell suspension)

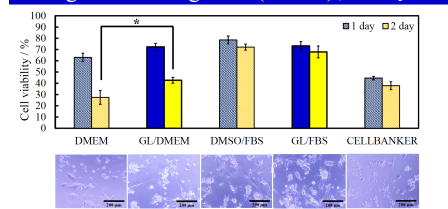


Fig. 11 Results of cell viability and cell morphology refrigerated in cell suspension system (Incubation time: 2 days)

- The addition of GL peptide significantly improved cell viability.
- hMSC refrigerated with the addition of GL peptide maintained its proliferative capacity.

