

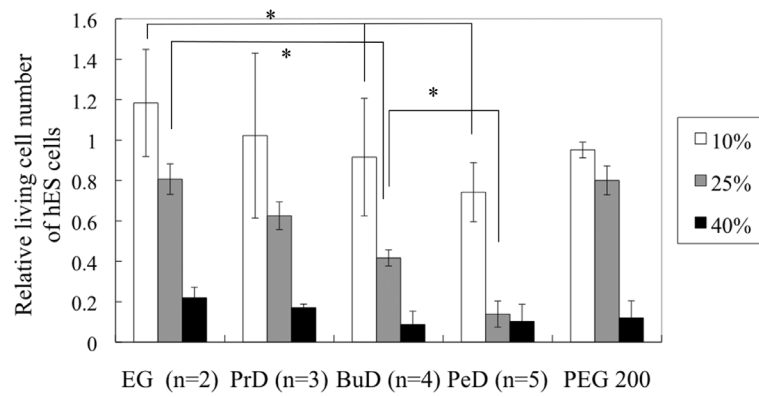
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**SUPPLEMENTARY MATERIAL**

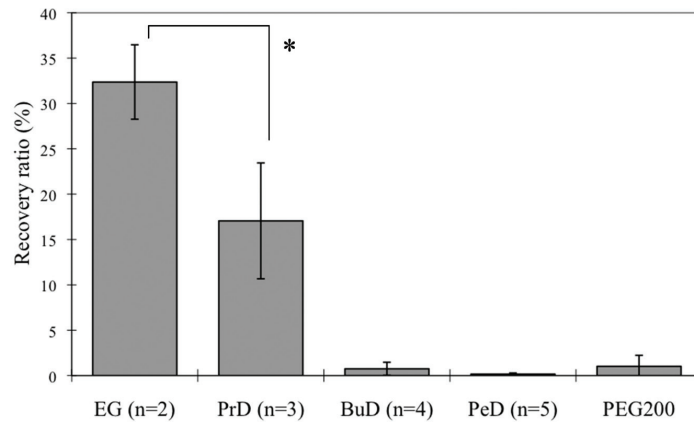
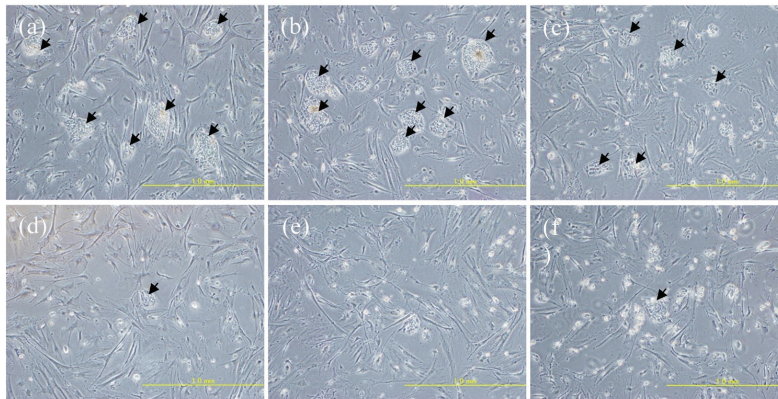
**corresponding to:**

**Highly efficient cryopreservation of human induced pluripotent stem cells using a dimethyl sulfoxide-free solution**

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**Supplemental Fig. 1.** Relative living cell numbers of hES cells (KhES-1) after exposure to 10, 25, and 40% (v/v) diol solutions. Cell numbers were normalized by the cell number of KhES-1 cells without exposure to the diol solutions. All data were expressed as mean  $\pm$  SD for N = 3. \* indicates the statistically significant differences ( $p < 0.05$ ).



**Supplemental Fig. 2.** Recovery of KhES-1 cells after after freezing and thawing. (A) Morphologies and recovery ratios of cryopreserved KhES-1 cells 1 day culture after a cooling-warming circle. (A) ; Colonies of KhES-1 cells, (a); KhES-1 cells without cryopreservation, KhES cells cryopreserved in (b); EG-, (c); PrD-, (d); BuD-, (e); PeD-, and (f); PEG200-based solutions. Arrows indicate KhES-1 cell colonies attached onto the SNL layer. Morphology was observed by a phase-contrast microscope. Scale bars: 1 mm. (B) Recovery ratios of KhES-1 colonies after cryopreservation in different diol solutions. Recovery ratios mean relative numbers of colonies after cryopreservation in diols solutions and those without cryopreservation. Results are expressed as mean + SD for N = 3. \* indicates a statistically significant difference ( $p < 0.05$ ).