

Supplementary Material for Long-term xeno-free culture of human pluripotent stem cells
on hydrogels with optimal elasticity

Akon Higuchi^{1,2,3,4}, Shih-Hsuan Kao¹, Qing-Dong Ling^{5,6}, Yen-Ming Chen¹, Hsing-Fen Li¹, Abdullah A. Alarfaj⁴,
Murugan A. Munusamy⁴, Kadarkarai Murugan⁷, Shih-Chang Chang⁸, Hsin-Chung Lee^{8,9}, Shih-Tien Hsu¹⁰, S.
Suresh Kumar¹¹, Akihiro Umezawa²

¹Department of Chemical and Materials Engineering, National Central University, No. 300 Jhongli, Taoyuan, 32001
Taiwan

²Department of Reproduction, National Research Institute for Child Health and Development, 2-10-1 Okura,
Setagaya-ku, Tokyo 157-8535, Japan

³Nano Medical Engineering Laboratory, RIKEN, 2-1, Hirosawa, Wako, Saitama 351-0198, Japan

⁴Department of Botany and Microbiology, College of Science, King Saud University, P.O. Box 2455,
Riyadh 11451, Saudi Arabia

⁵Cathay Medical Research Institute, Cathay General Hospital, No. 32, Ln 160, Jian-Cheng Road, Hsi-Chi City,
Taipei, 221, Taiwan

⁶Graduate Institute of Systems Biology and Bioinformatics, National Central University, No. 300, Jhongda RD.,
Jhongli, Taoyuan, 32001 Taiwan

⁷Division of Entomology, Department of Zoology, School of Life Sciences, Bharathiar University, Coimbatore,
Tamil Nadu, India

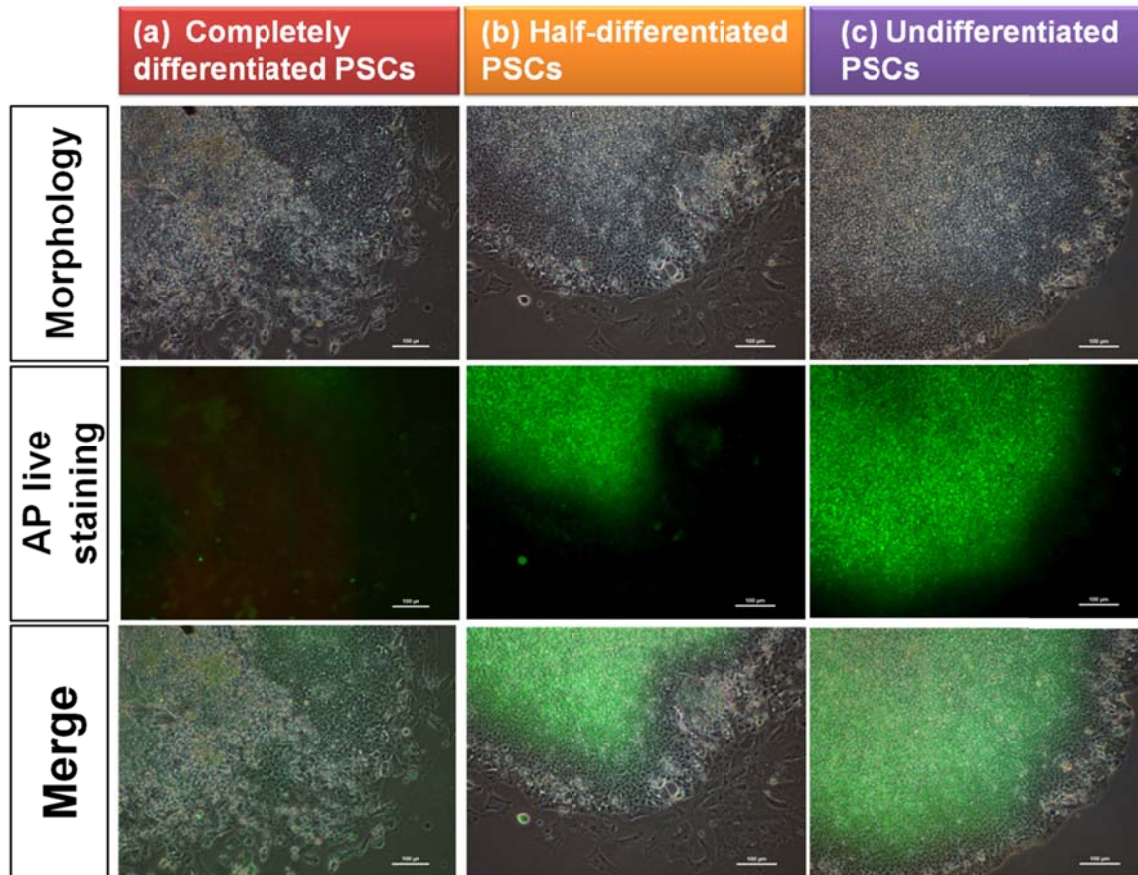
⁸Department of Surgery, Cathay General Hospital, No.280, Sec. 4, Ren'ai Rd., Da'an Dist., Taipei, 10693, Taiwan

⁹Graduate Institute of Translational and Interdisciplinary Medicine, College of Health Science and Technology,
National Central University, No. 300, Jhongda RD., Jhongli, Taoyuan, 32001 Taiwan

¹⁰Department of Internal Medicine, Taiwan Landseed Hospital, 77, Kuangtai Road, Pingjen City, Taoyuan 32405,
Taiwan

¹¹Department of Medical Microbiology and Parasitology, Universiti Putra Malaysia, Serdang 43400, Slangor,
Malaysia

SUPPLEMENTARY FIGURES



Supplementary Figure 1. Definition of completely differentiated PSCs (a), partially differentiated PSCs (b), and pluripotent (undifferentiated) PSCs (c). No alkali phosphatase (AP) activity was detected in the completely differentiated cells (a). In contrast, while the edge of the colony of the partially differentiated cells did not exhibit alkali phosphatase activity, alkali phosphatase activity was detected in the center of the colony (b). The pluripotent cells exhibited good colony morphology and alkali phosphatase activity in most cells (c).