

Establishment of a feeder-free culture system for porcine induced pluripotent stem cells ⁽¹⁾

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Abstract

The present study established the feeder-free culture system for porcine induced pluripotent stem cells (piPSC) by using Matrigel[®], an extracellular matrix used to replace the feeder cells in the traditional stem cell culture, and stem cell culture medium named StemFit Basic02+2i (S+2i) and mTeSR1+2i (M+2i), StemFit Basic02 and mTeSR1 supplemented with CHIR99021 and SB431542. The results showed that S+2i and M+2i maintained the cell morphology and showed high nuclear cytoplasmic ratio, indicating that piPSC showed an undifferentiated state. The pluripotency markers expression of Oct4, Sox2, and Nanog was very high, the expression of SSEA-1, TRA-1-60, and TRA-1-81 was low, but the expression of SSEA-3 and SSEA-4 was very weak. The expression of pluripotency genes was significantly improved, and S+2i exhibited the best results. After embryoid body formation, piPSC *in vitro* differentiated into three germ layer cells, expressing GFAP, SMA, and AFP antigen. Teratomas were successfully formed in both S+2i and M+2i, but their ability to differentiate into three germ layers was still quite limited. Taken together, this study provides a new method for long-term culture of piPSC. This culture system not only solves the problem of preparing feeder cells in a traditional culture system, but also improves the cell pluripotency and utility which is better than traditional feeder-dependent culture systems.

Key words: Feeder-free culture system, Induced pluripotent stem cells, Pigs.

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