ESTABLISHMENT OF CULTURE CONDITIONS FOR PRIMORDIAL GERM CELLS FROM TAIWAN COUNTRY CHICKEN

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Introduction

Primordial germ cells (PGCs), the progenitor cells of gametes, carry the genetic information throughout generations. Hence, PGCs hold the potential to produce transgenic animals and serve a tool for the conservation of endangered animals due to their germ line-transmissible ability. The aim of this study was to isolate, characterize and establish an optimal culture condition for Taiwan country chicken primordial germ cells (cPGCs).

Materials and Methods

Chicken embryos from the country chicken of National Chung Hsing University strain L2 were collected at stage 18-20 (3-3.5 days after incubation). The blood was drawn from the dorsal aorta of embryos using a fine glass micropipette, and then washed with DMEM containing 10% fetal bovine serum (FBS). The cPGCs were isolated by gradient centrifugation with 6.3% and 16% of Ficoll, and allocated into three culture media, i.e. DMEM-10%FBS-5% chicken serum (CS) supplemented with bFGF-LIF, bFGF-LIF-SCF, and bFGF-LIF-SCF-BMP4. Cell proliferation and expression of pluripotency markers were analyzed.

Results and Discussion

The cPGCs were cultured for 7 days and then subcultured every 5 days. Results showed that the total cell number decreased in three groups after culture, but the numbers in bFGF-LIF-SCF group are higher than those in the other groups after 2 passages. The immunocytochemical staining results demonstrated that some cultured cells expressed SSEA-1, indicating that the current three culture systems are able to maintain the cell pluripotency in short term.