Factors regulating stem cell behaviour – P3

Effect of different culture condition on development of stem cell like cells from IVF derived early developing embryos in buffalo
S. Bag¹, B.C. Das¹, R. Chhetri¹, G. Puri¹, A.C. Majumdar¹

¹IVRI, Physiology & Climatology, Bareilly, India

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So far it has been difficult to generate a proven stem cell line in animal which has been attributed to the lack of proper culture condition. The present experiment was carried out to study the role of different culture conditions on multiplication of early stage IVF derived embryonic cells of buffalo. The IVF embryos of 32-cell stage were made zona free. The clumped blastomere were cultured on inactivated murine embryonic fibroblast (MEF) with culture condition viz. (C-I) DMEM+ITS +FBS +LIF + SCF, (C-II) DMEM+ITS +FBS +LIF + SCF + IGF1, (C-III) DMEM+ITS +FBS +LIF + SCF +IGF1+bFGF4. The blastomere were cultured at 37°C, 5% CO2 and 90% relative humidity in CO2 incubator. Once the isolated blastomere clumped made stem cell clone, they were passaged mechanically. In the first culture condition, blastomere did not maintain for long time in culture. In culture condition-II, the multiplication was better as compared to C-I but no cell line could be derived in this culture condition also. In culture condition C-III, the cell clones could be propagated upto 3rd passage. The stem cell like clone were positive for Oct, AP and Nanog expression. The results indicated that multiplication of buffalo IVF derived embryonic cells from early stage embryos were better when they were cultured in presence of LIF, SCF, IGF1 and bFGF4 than culture condition excluding these cytokines.