Bulletin UASVM, Veterinary Medicine 65(1)/2008
pISSN 1843-5270; eISSN 1843-5378

LIF: ITS IMPLICATION IN MOUSE EMBRYONIC STEM CELLS PLURIPOTENCE

Pall Emoke[2]-, Lichner Zsuzsanna[1], I. Groza[2], Gocza Elen[2]

[1]Agricultural Biotechnology Center, Gödöllő, Hungary
[2]University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca, Romania
email: pallemoke@yahoo.com

Keywords: LIF; embryonic stem cells; differentiation; pluripotence

SUMMARY

Leukemia-inhibitory factor (LIF) is a multifunctional cytokine; which has pleiotropic biological effects on a diverse array of cell types. While spontaneous and random differentiation of ES cells can be easily triggered by the withdrawal of LIF from the medium. We used at our experiment the mouse ES cell line R1/E/NA with a normal karyotype; at 14th passages. To induce differentiation; the ES cells were washed with 1· PBS twice 12 h after the culture and then cultured without LIF in Iscove's modified Dulbecco's medium (Invitrogen) supplemented; on a 0;1% gelation –coated plate. The colony positive cells were fixed 14 days after the culture and stained for alkaline phosphatase (liver isozyme alkaline phosphatase).

Colony forming assay: was used for evaluate the incrase or decrase the number of morphologically distinct AP positive colonies. The spontaneous differentiation was induced by the withdrawal of LIF 12 h after plating ES cells. At 1 day after induction of differentiation; ES cells reached approximately 70% of confluence and still retained typical undifferentiated morphology. Most cells at 7 days displayed unspecified irregular shapes and a typical undifferentiated morphology was no longer observed. Several morphologically distinct populations of cells had appeared at 14 days. The colony forming assay test was made at 7; 14 and 21 days; observing a significant increase of positive alcalin-phosphatase colonies; which demonstrates the embryonic stem cells differentiation in comparison with the LIF treated culture where we noted compact colonies of 100% positive alcalin-phosphatase.

Spontaneous and random differentiation of embryonic stem cells can be easily triggered in the absence of LIF from the medium and without feeder layer (MEF- mouse embryonic fibroblast).

BIBLIOGRAPHY

1. Jeonghoon H.L. Ju-Seog; In-Sun C.;Yasushi; Snorri S.T.; 2005; Spontaneous differentiation of mouse embryonic stem cells in vitro: Characterization by global gene expression profiles; Biochemical and Biophysical Research Communications 332: 1061–1069;
2. Yusuke Y.; A. Tategaki; Mari Ogawa; Hiroyuki H.; Kensho N.; Sachiko A.; Haruo M.; Shuichi F.; 2006; Effect of novel monoclonal antibodies on LIF-induced signaling in chicken blastodermal cells; Developmental and Comparative Immunology 30: 513–522;
3. Keller G.; 2005; Embryonic stem cell differentiation: emergence of a new era in biology and medicine. Review; Genes&Dev. 19:1129-1155;
4. Palmqvist L.; Clive H. Glover; 2005;Correlation of murine embryonic stem cell gene expression profiles with functional measures of plurypotency; Stem Cells 23 : 663-680;
5. Lanza R.; 2004; Handbook of Stem Cells; vol.1.