



شرکت سلول بافت زیست (سبز)

Differentiation Protocols

Differentiation to Neural Cells:

DMEM
FBS 10%
IBMX 0.5mM
dbcAMP 1mM
Retinoic Acid 10^{-6} M

Differentiation to Osteocytes:

DMEM
FBS 10%
Dexamethasone 10^{-7} M
Beta-Glycerol-Phosphate 10mM
Ascorbic Acid bi-Phosphate 50 ug/ml

Differentiation to Chondrocytes:

DMEM
FBS 10%
TGF-beta 10 ng/ml
Ascorbic Acid bi-Phosphate 50 ug/ml
Dexamethasone 10^{-7} M
bFGF 10ng/ml

Differentiation to Endothelial Cells:

M199
FBS 10%
VEGF 20 ng/ml
IGF-1 20 ng/ml

Differentiation Protocols

Differentiation to Adipocytes:

DMEM
FBS 10%
IBMX 0.5 mM
Dexamethasone 250 nM (0.25 uM)

Insulin 66 nM
Indomethacine 0.2 mM

Differentiation to Motor Neuron:

For 4 days:

DMEM
FBS 10%
Retinoic Acid 10^{-6} M
Shh 300 nM

For another consequent 4 days:

Neurobasal medium
FBS 10%
B₂₇: 2.5%

Differentiation to Myofibroblast:

DMEM
FBS 10%
TGF beta 1: 5 ng/ml

Differentiation Protocols

For adipogenic differentiation, Insulin and Indomethacine could be omitted or be used interchangeably. Dexamethasone is also functional at the range of 100 nM to 1uM.

Prepare a range of 1000-10000 cells per well of a 24-well plate for each differentiation assay.

For Chondrocyte differentiation, coat the cells with Poly-L-Lys for 45 minutes in 37°C and then remove it before seeding the cells. An alternative method is to seed 2×10^5 cells in a 15 ml conical polypropylene tube and then add about 1 ml of the differentiation media. In both methods the cells are expected to form a cell clump, which should be formalin fixed and paraffin embedded for IHC.

In the case of endothelial differentiation, before seeding the cells, the wells should be coated with fibronectin (5 ug/ml) for 45 minutes in 37°C. Do not remove it.

Duration of differentiation for endothelial, Myofibroblast, adipogenic and neural cells is 2 weeks and for osteocytes 3 weeks. For chondrocyte differentiation this time is 21-30 days.

Retinoic Acid is extremely light sensitive.

The second 4 days period in motor neuron differentiation, is for the maturation of the cells and could be omitted.

For Neural Differentiation, coat the wells with gelatin before seeding the cells.