BIOCHEMICAL PROCESSES
(CM4710)

STEM CELLS:
Science and Applications

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Presentation Outline

• Definition
• Sources
• Maintenance
• Differentiation
• Tissue engineering
• Applications
  – Cures of health problems
  – Understanding of health problems
DEFINITION

• Stem cells are multipotent cells with the ability to self-renew and differentiate into specialized cells in response to appropriate signals [1].

Those cells can be cultured to divide indefinitely

Those cells can turn into many – and perhaps all - cells types

Stem cells properties

SOURCES

• Embryos

Human: Blastocysts (5 to 7 days old embryo) from fertility clinics

• Adult form of the species

• Human: Most tissues have endogenous stem/progenitor cells (known as adult stem cells)
FUNCTION OF STEM CELLS IN THE ADULT FORM OF THE SPECIES

- Upon injury to the organ, stem cells can proliferate and differentiate at the damaged site [1]
Stem cell distribution in the adult human body

- Brain
- Blood
- Cornea
- Retina
- Heart
- Skin
- Dental pulp
- Bone marrow
- Blood vessels
- Skeletal muscle
- Intestines
- Umbilical cord

MAINTENANCE

- Ability to divide indefinitely
- It will be maintained by adding fresh culture media

Embryonic stem cells are cultured on gelatin-coated T25 flasks prepared using 5 mL of 0.1% v/v gelatin (gelatin 2%) diluted in 0.01 M phosphate-buffered 0.15 M saline (pH 7.4 PBS) without calcium and magnesium. The flasks were incubated for 15-30 min at 37 °C. Embryonic stem cells are cultured in DMEM (Invitrogen) cell culture medium containing 10% FBS (batch tested for embryonic stem cells, Invitrogen), L-Glutamine 2 mM, mercaptoethanol 1 microliter/mL, LIF 1 or 10^3 microliter/mL, and penicillin/streptomycin 1% v/v [7]
MAINTENANCE

- Ability of being able to differentiate in all types of cells
- It will be maintained by addition of substances that prevent differentiation
- Leukemia inhibitory factor (LIF) maintains embryonic stem cells pluripotent [3]

DIFFERENTIATION

- Neuronal stem cell
  - Glial cells
  - Neurons
- Cord blood stem cells
  - Platelets
  - Red blood cells
  - White blood cells
  - Mesenchymal cells
- Bone marrow
  - Miocytes
  - Hematocytes
**DIFFERENTIATION**

Effects of Indole Fatty Acids on the Differentiation of Neural Stem Cell Derived Neurospheres [4]

<table>
<thead>
<tr>
<th>compd</th>
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<th>% neurons relative to control</th>
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<td>+</td>
<td>100</td>
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<td>+</td>
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<td>+</td>
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<td>H</td>
<td>12</td>
<td>124</td>
<td>+</td>
</tr>
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<td></td>
<td>H</td>
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<tr>
<td></td>
<td>H</td>
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<tr>
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<td>13c</td>
<td>5-MeO</td>
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<td>108</td>
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<td></td>
<td>5,6-MeO</td>
<td>15 (without -OH)</td>
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<td>+</td>
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Figure 1. Optical micrographs showing P19 EC cell culture inside an empty zebrafish chorion. (a) From the left, intrusion of a micropipet into a chorion egg to create an opening for the release of the yolk and remaining inner mass. During the process, the chorion expands more than twice its original volume without any surface splitting. (b) By day 4, an embryonic body (EB) was successfully formed inside the chorion by the adhesions among P19 EC cells. A carefully controlled micropipet can deliver the required number of cells into the chorion for one EB formation. (c) By day 7, membrane shrinkage is likely visible because of a deficiency of medium supply inside the chorion. A crescent-shaped cavity is clearly observable inside the cell mass. (d) By day 10, the shape is recovered by continued cell proliferation. (e) A complete occupation of the whole inner space can be observed by day 15, and proliferating cells can be seen extruding through the injection opening. The fluid-filled cavity is observed by the dense cell mass. (f) Optical micrograph of neural cells with elongated filopodia recovered on day 15. (g) Rotating cardiac cell masses were found on day 15. The scale bar in frame g is also applicable to frame f.

**DIFFERENTIATION**

Using the Chorions of Fertilized Zebrafish Eggs as a Biomaterial for the Attachment and Differentiation of Mouse Stem Cells [5]
DIFFERENTIATION

E-Cadherin induces Hepatopacific phenotype and maturation of embryonic stem cells in conjunction with hepatotrophic factors [6]

Small molecules that induce Cardiomyogenesis in embryonic Stem cells [1]

<table>
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<th>Chemical Structure</th>
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<th>Optical Activity</th>
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<tr>
<td>Cardiogenol B</td>
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<td>+++</td>
</tr>
<tr>
<td>Cardiogenol C</td>
<td>O</td>
<td>0.1 μM</td>
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</tr>
<tr>
<td>Cardiogenol D</td>
<td>O</td>
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+++ ~ 10-25% cells are positive for MHC after 7 days.
++++ ~ 25-40% cells are positive for MHC after 7 days.
+++++ ~ 40-55% cells are positive for MHC after 7 days.

TISSUE ENGINEERING

- Engineering bone-like tissue in vitro using human bone marrow stem cells and silk scaffolds [8]

- Three-dimensional collagen gel networks for neural stem cell-based neural tissue engineering [9]

- Human mesenchymal stem cells tissue development in 3D PET matrices [10]
APPLICATIONS

Cure of health problems

• Heart disease: Adult bone marrow stem cells injected into heart arteries are believed to improve cardiac functions

• Leukemia: Patients treated with stem cells from bone marrow and umbilical cord blood emerged free of disease

• Rheumatoid arthritis: Adult stem cells may be helpful in jump-starting repair of eroded cartilage.

APPLICATIONS

Understanding of health problems

• Large-scale expansion of mammary epithelial stem cells aggregates in suspension bioreactors [11]

• Motivation: Self renewal and differentiation are currently hypothesized to result in uncontrolled cell division and, in turn, breast tumor formation

• Results: Successful development of expansion protocols for Mammary epithelial cells aggregates in suspension bioreactors that would produce enough aggregates to study breast tumor formation
Bibliography