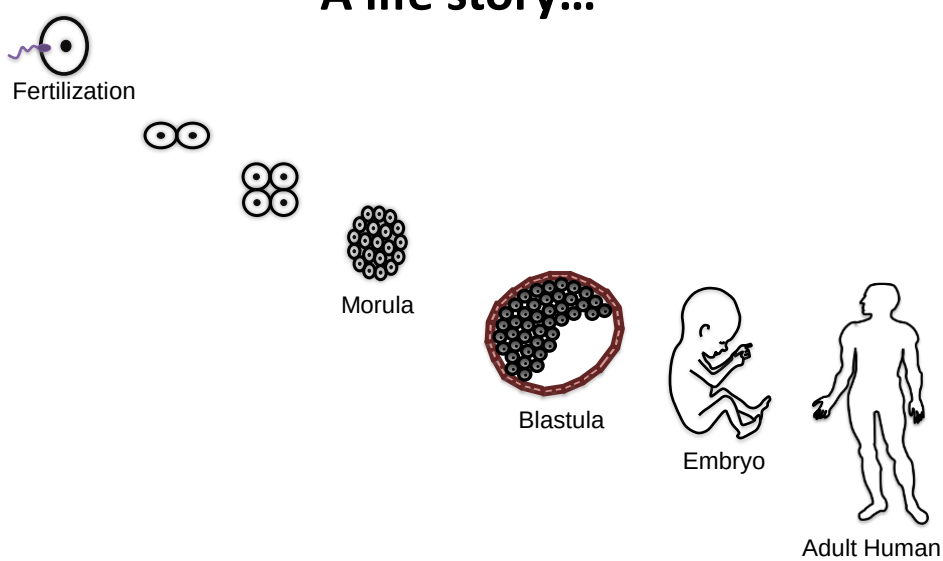
DEVELOPMENTAL PRINCIPLES IN ONTOGENESIS

- PROLIFERATION
- DIFFERENTIATION
- INDUCTION
- MIGRATION
- APOPTOSIS

All these principles (except induction) take part in the development of adult organisms.

Dr. Wasay Mohiuddin

A life story…

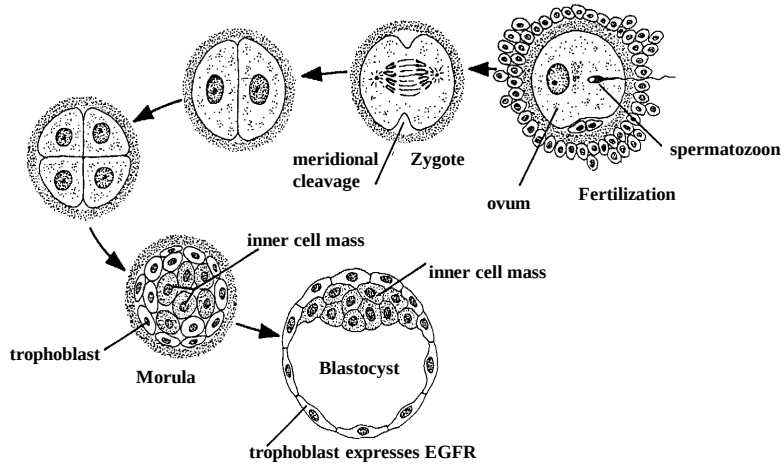


IMPORTANT DEFINITIONS

- **ONTOGENESIS:** The origin and development of an individual organism from embryo to adult.
- **PROLIFERATION:** Multiplication of cells through repetitional cellular divisions.
- **DIFFERENTIATION:** The normal process by which a less specialized cell develops or matures to possess a more distinct form and function.
- **INDUCTION**: In development, an interaction between two cell lineages to alter the developmental fate of one or both of them.
- **MIGRATION:** Movement of a population of cells from one place to another as in the movement of neural crest cells during morphogenesis.
- **APOPTOSIS:** Apoptosis is a programmed cell death.

PROLIFERATION = multiplication of cells through repetitional cellular divisions (mitosis).

Fertilized egg must divide many times to form multicellular organism, i.e. $35x10^{12}$ cells. Rapid proliferation is characteristic of early developmental stages.

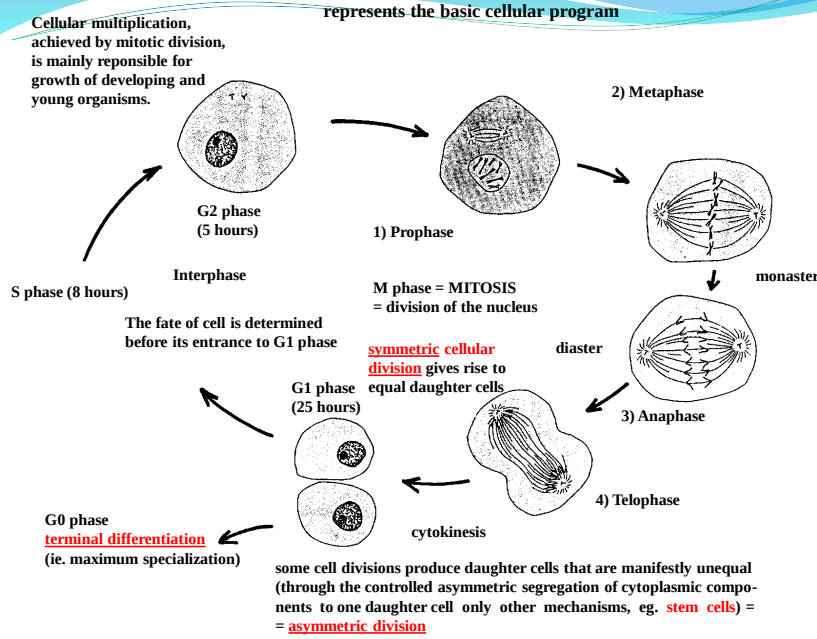


Cleavage: Fertilization triggers an unusually rapid sequence of cell divisions of a single giant cell (zygote) that cleaves, without growing, to generate an embryo consisting of a large amount of smaller cells.

In this process, DNA is the principal macromolecule to be synthesized. The development of an embryo can begin soon after fertilization; as the cytoplasm of the oocyte contains a large amount of mRNAs. This enables the embryos´ development not to be retarded by waiting for transcription of new genes (maternal mRNAs are used to produce the necessary proteins during early phases of ontogenesis). The cleaving embryo is almost entirely dependent on the RNAs, proteins, membranes, and other materials that accumulated in the egg.

THE CELL CYCLE

represents the basic cellular program



This cellular division is strictly by growth factors (GFs), growth inhibitors and other controlling molecules.
Growth factors can act on cells in differ ent phases of the cell cycle. Each cell responds to specific combinations of signaling molecules incl. Different cells can respond differently to the same signal (e.g. growth factor).

Growth factor (GF)		Cells proliferating in response to GF
EGF	epidermal GF	keratinocytes, trophoblast, neuroepithelial c. etc.
NGF	neural GF	neuroblasts, neurite outgrowth
PDGF	platelet-derived GF	CT cells, neuroglia
FGF	fibroblast GF	many cell types
IGF-1	insulin-like GF 1	colaborate with other GFs
IL-2	interleukin-2	activated T lymphocytes
	erythropoietin	precursors of red blood cells
VEGF	vascular endothelial GF	endothelial cells
HGF	hepatocyte GF	hepatocyte

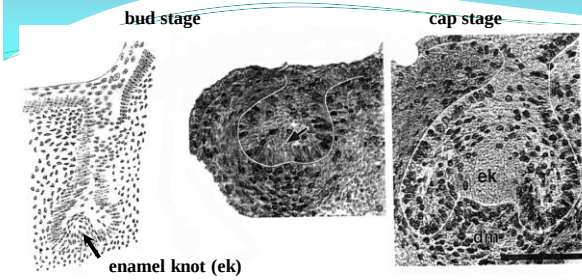
Growth factors can influence only those cells that possess specific receptors for the particular growth factor.

The receptors for most growth factors are transmembrane Tyrosine-specific protein kinases. Through their activation, growth factors trigger cascades of intracellular signals and activate new genes.

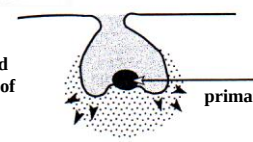
Some growth factors may also act as differentiation factors.

The growth of developing and young organisms consists of:
- **proliferation**
- **enlargement of individual cells during their differentiation**
- **production of extracellular matrix**

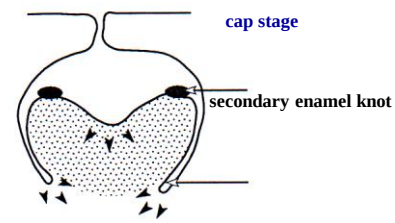
TOOTH MORPHOGENESIS



Primary enamel knot = embryonic structure formed by cluster of non-dividing cells situated in the centre of the developing tooth. Cells of the enamel knot do not proliferate but they produce FGF-4, which stimulates proliferation of surrounding inner enamel cells.



Proliferation of inner enamel epithelium results in folding and downgrowth around the enamel knot. Bilateral proliferation of inner enamel epithelium (in combination with non-dividing enamel knot) is responsible for formation of a characteristic bell-shape of the enamel organ - tooth shape development (incl. its crown).



The differential rates of growth of different parts of developing structures and uneven distribution of proliferating and resting areas (e.g. in the epithelium) are necessary for formation of a resulting shape of these structures (e.g. brain vesicles etc.).

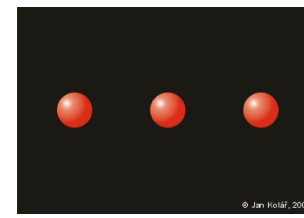
CHARACTERISTICS OF STEM CELLS

- Biological properties:
- Unlimited self-renewal
- Production of differentiated elements

} asymmetric division

- High proliferative capacity (telomerase)
- Broad differentiation potential (toti-, pluri-, multipotency)
- Plasticity

The oldest (earliest) cellular elements produced during ontogenesis



- Primitive morphology
  (lack of specific markers)
- Limited pool (of a defined size)
- Localisation
- Hierarchical arrangement of individual SC generations

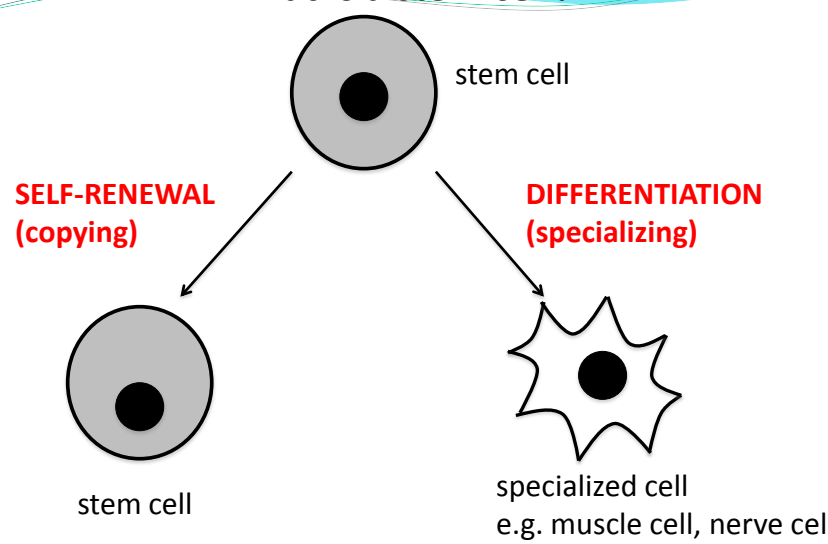
STEM CELL

= undifferentiated cell capable of production of differentiated cells and at the same time of unlimited self-renewal. Stem cells give rise to all tissues. In adulthood, they are responsible for maintenance of tissue homeostasis and for tissue regeneration. These functions are performed by the stem cells throughout the entire lifespan, i.e. in all developmental stages. In the course of development, different stem cells are generated. The generations of stem cells are hierachically arranged – older (i.e. more primitive) stem cells have broader developmental potential.

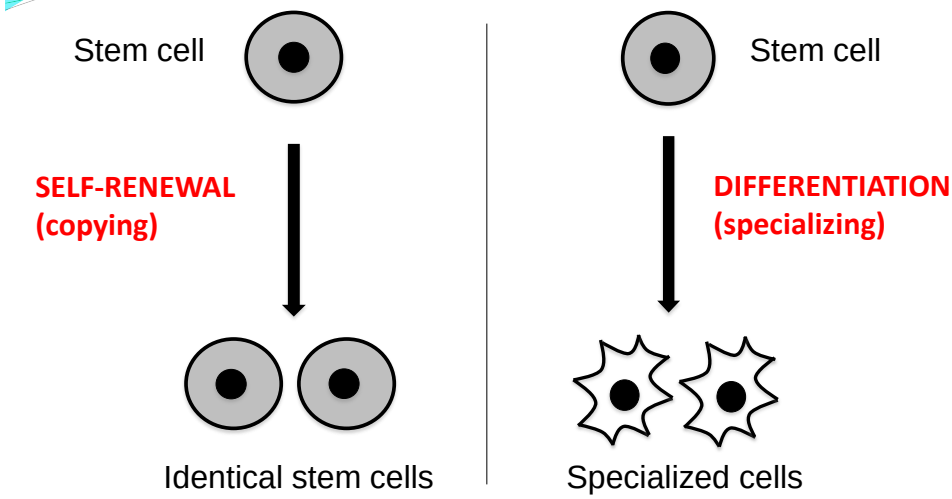
FUNCTION of SCs:

- units of development - biological organization of developing tissues
- units of tissue maintenance
- units of regeneration
- units of transplantation
- units of selection (in evolution)

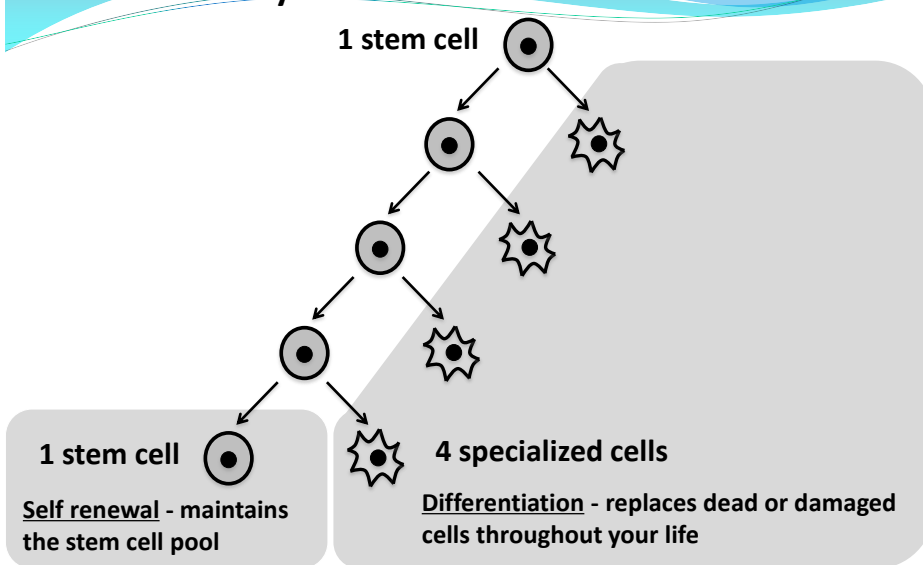
What is a stem cell?



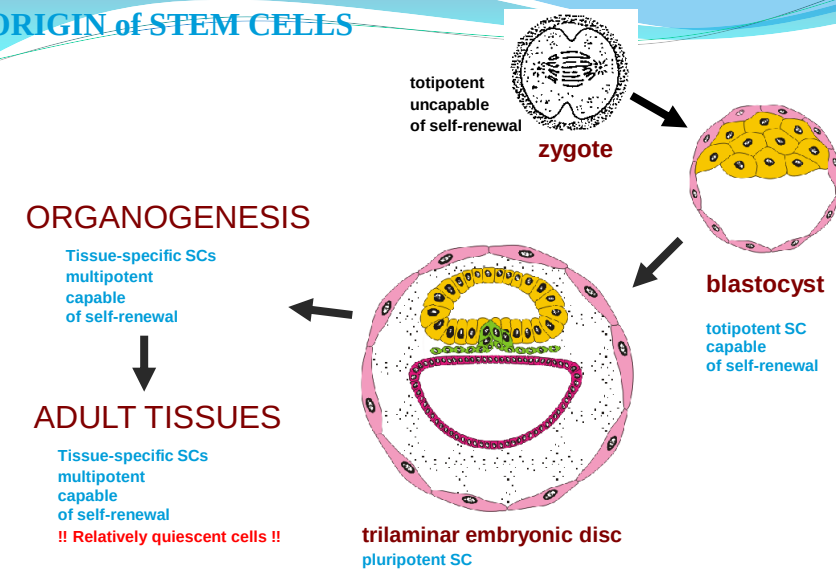
What is a stem cell?



Why self-renew AND differentiate?



ORIGIN of STEM CELLS



DIFFERENTIATION Lat. *differo* (to differ)

= The process of gradual specialization of cells to functions definitively different from those of original cell type.

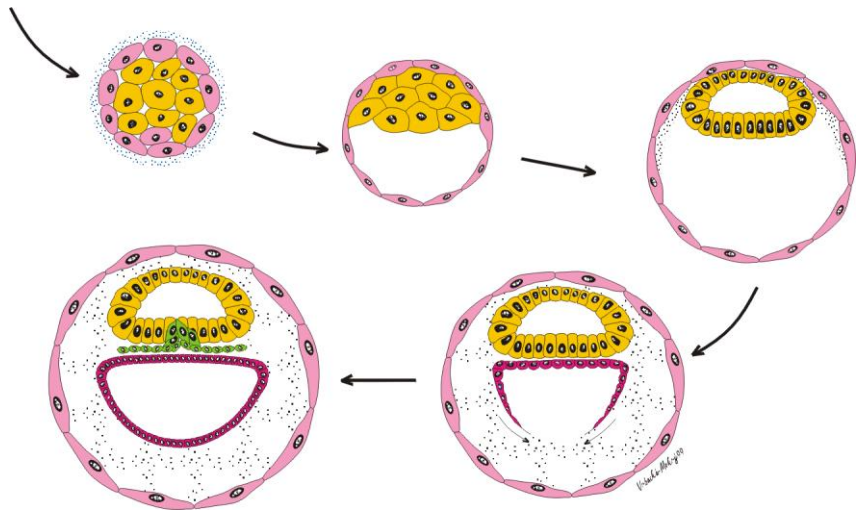
Ectoderm → neuroectoderm (neural crest) → odontoblasts

The cells of the vertebrate body exhibit more than 200 different modes of specialization.
The cells are specialized for specific functions.

Example:
In the neuromuscular junction there are three types of cells involved:

- **The muscle cell/fibre performs contraction**
- **The nerve cell stimulates the muscle to contract**
- **Schwann cells form myelin sheath that isolates a nerve fibre**

The development of tissue begins at the moment of fertilization, ie. upon the union of a spermatozoon with an ovum. Male and female pronucleus fuse and form the first nucleus of the first cell (zygote) of the new organism. All tissues develop from one zygote.



Morula → Blastocyst (the trophoblast) → epiblast beneath it hypoblast → the bilaminar embryonic disc intraembryonic mesoderm → trilaminar embryonic disc
From three germ layers develop all groups of tissues, ie. their cells are multipotent.

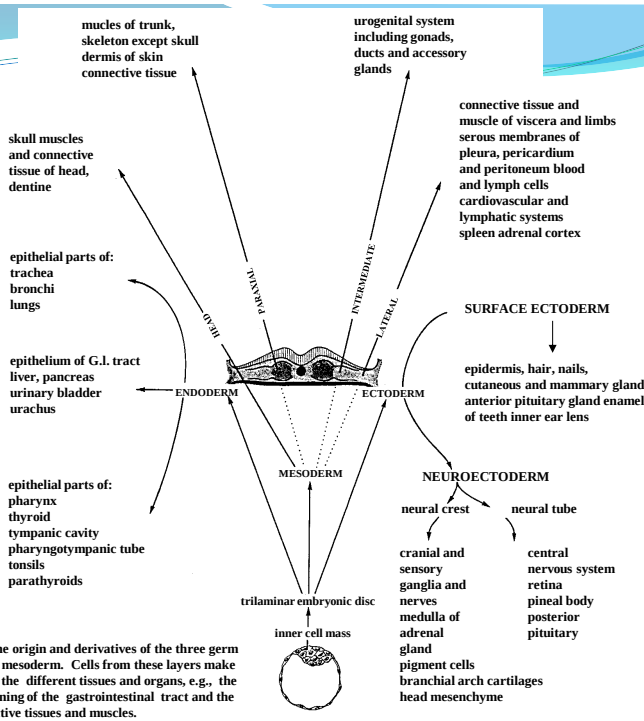
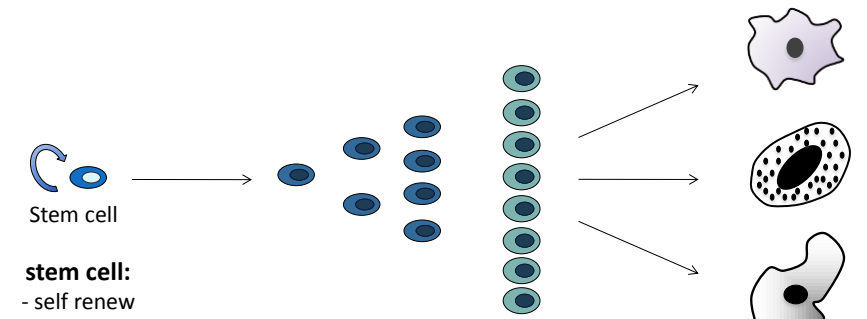


Figure 6-2. Schema illustrating the origin and derivatives of the three germ layers ectoderm, endoderm,and mesoderm. Cells from these layers make contributions to the formation of the different tissues and organs, e.g., the endoderm form the epithelial lining of the gastrointestinal tract and the mesoderm gives rise to its connective tissues and muscles.

Principles of renewing tissues



Stem cell

stem cell:
- self renew
- divide rarely
- high potency
- rare

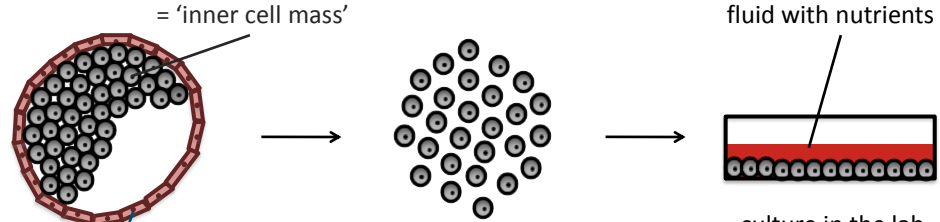
committed progenitors:
- "transient amplifying cells"
- multipotent
- divide rapidly
- no self-renewal

specialized cells:
- work
- no division

Embryonic stem (ES) cells

blastocyst

cells inside
= 'inner cell mass'



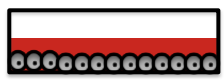
fluid with nutrients

embryonic stem cells taken from the inner cell mass

culture in the lab to grow more cells

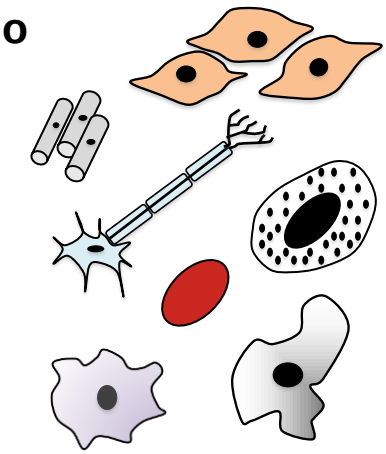
outer layer of cells
= 'trophectoderm'

Embryonic stem (ES) cells: What they can do



embryonic stem cells

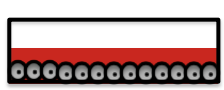
differentiation



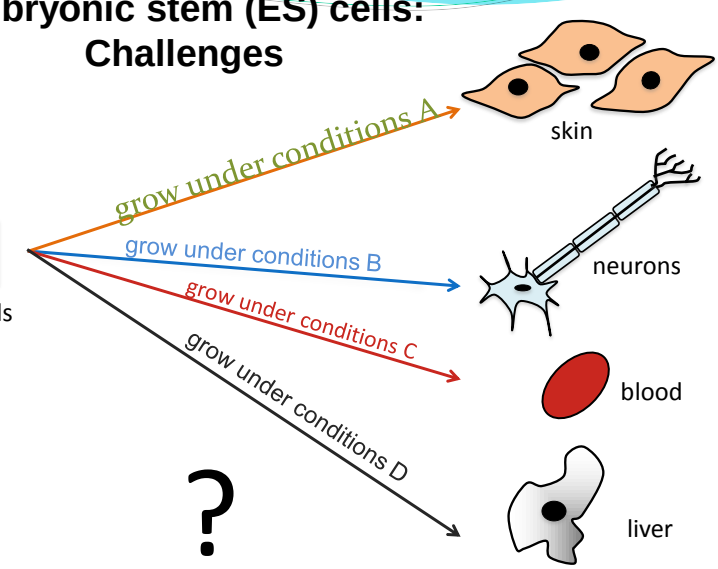
all possible types of specialized cells

PLURIPOTENT

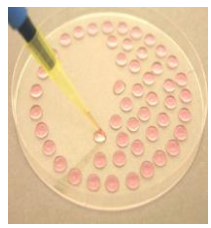
Embryonic stem (ES) cells: Challenges



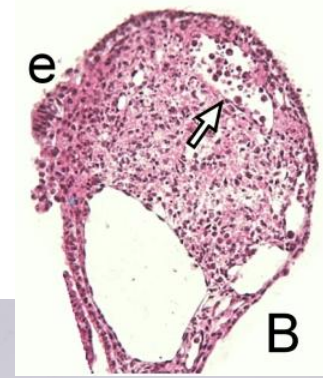
embryonic stem cells

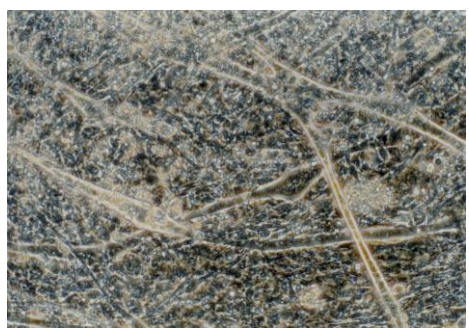


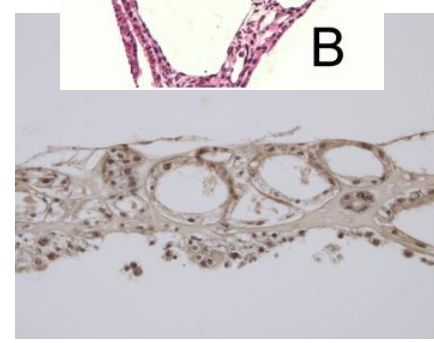
EMBRYONIC STEM (ES) CELLS (1981, 1998)



ES-D3

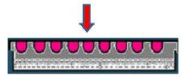




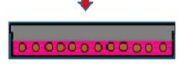


Embryonic stem cells differentiation

D3 ES cultured in T-25 Flask with LIF

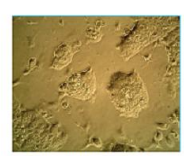


ESD3 in hanging drop without LIF

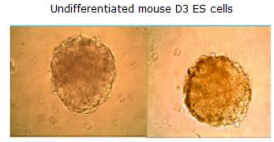


EBs cultured in Petri dish

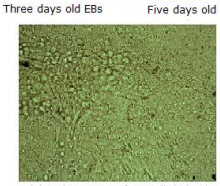
EBs plated in 24 well plates for differentiation



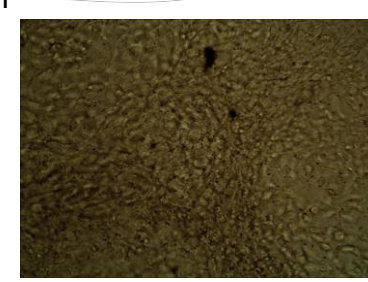
Undifferentiated mouse D3 ES cells



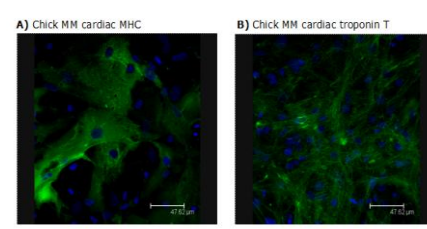
Three days old EBs Five days old EBs



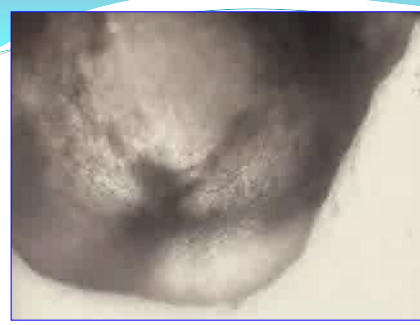
Light microscopy of ES cell derived cardiomyocyte (magnification x25)



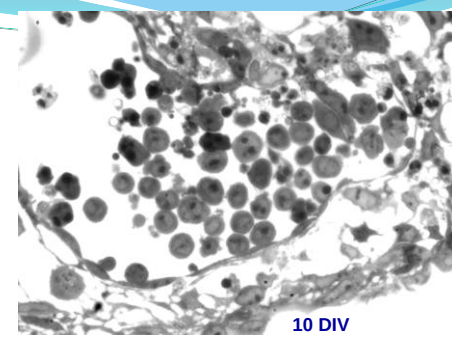
Contracting cardiomyocytes



Cardiac cell staining

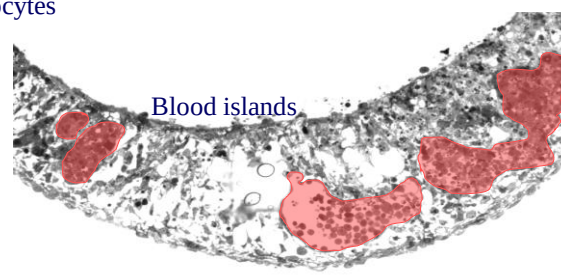


Cardiomyocytes

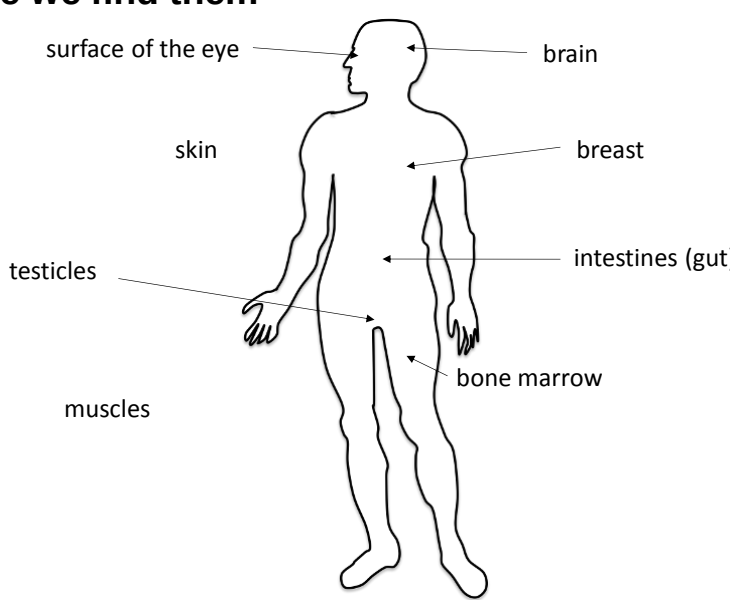


EMBRYOID BODIES

- Myogenesis
- Vasculogenesis
- Hematopoiesis in vitro



Tissue stem cells: Where we find them

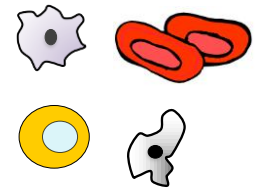


Tissue stem cells: What they can do

blood stem cell

found in bone marrow

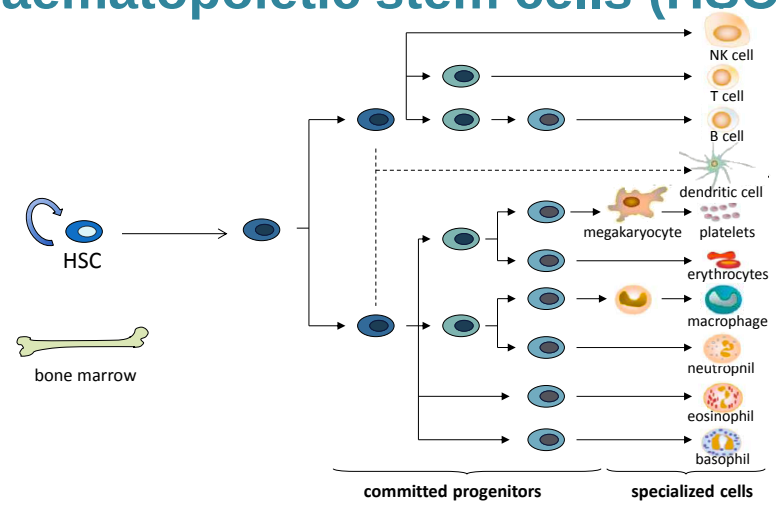
differentiation



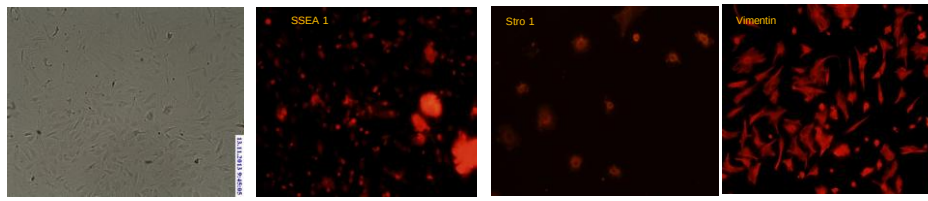
only specialized types of blood cell: red blood cells, white blood cells, platelets

MULTIPOTENT

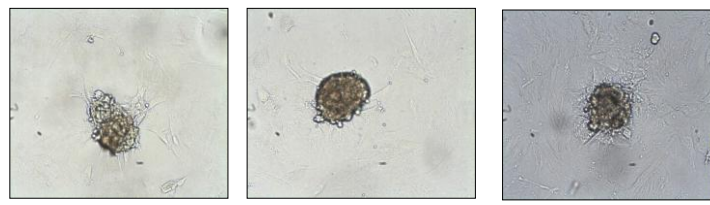
Tissue stem cells:
Haematopoietic stem cells (HSCs)



Bone marrow Stromal stem cells

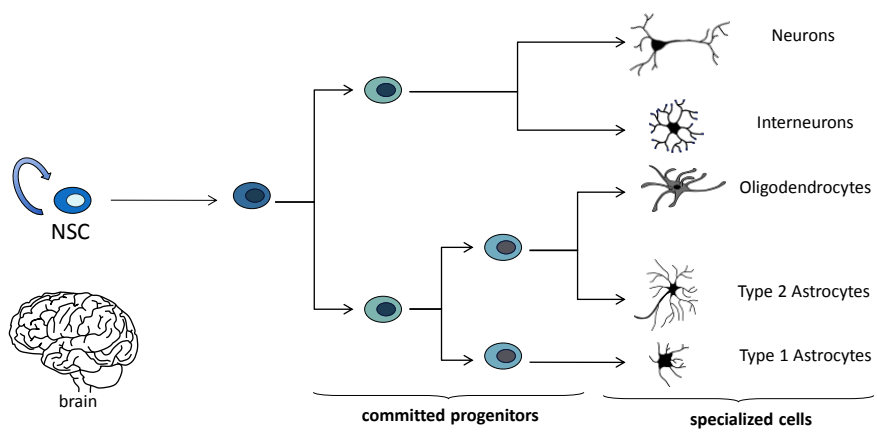


Stromal stem cells

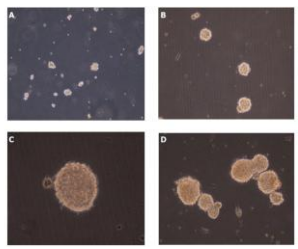


Stromal stem cells cardiac differentiation

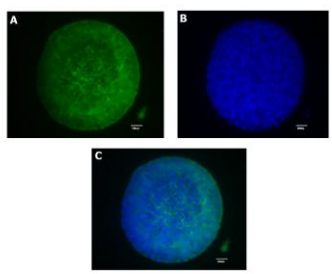
Tissue stem cells:
Neural stem cells (NSCs)



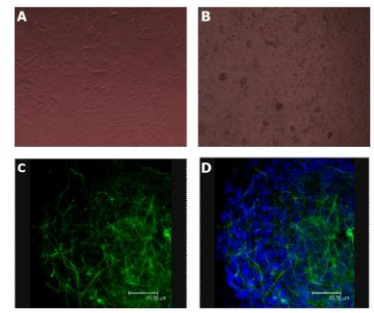
Neural Stem Cells culture



NSC neurospheres formation



Neurons



Glial cells

Tissue stem cells:

Gut stem cells (GSCs)