

STEM CELLS AND APPLICATIONS





Introduction

- Stem cells are those cells that have the capability of self-renewal and differentiation.
- Self-renewal is the ability of cells to proliferate without the loss of differentiation potential and without undergoing senescence (biologic aging).
- A stem cell is a blank cell/ precursor cell that can give rise to multiple tissue types such as a skin, muscle, or nerve cell.
- A stem cell is essentially the building block of the human body

Features of Stem Cells

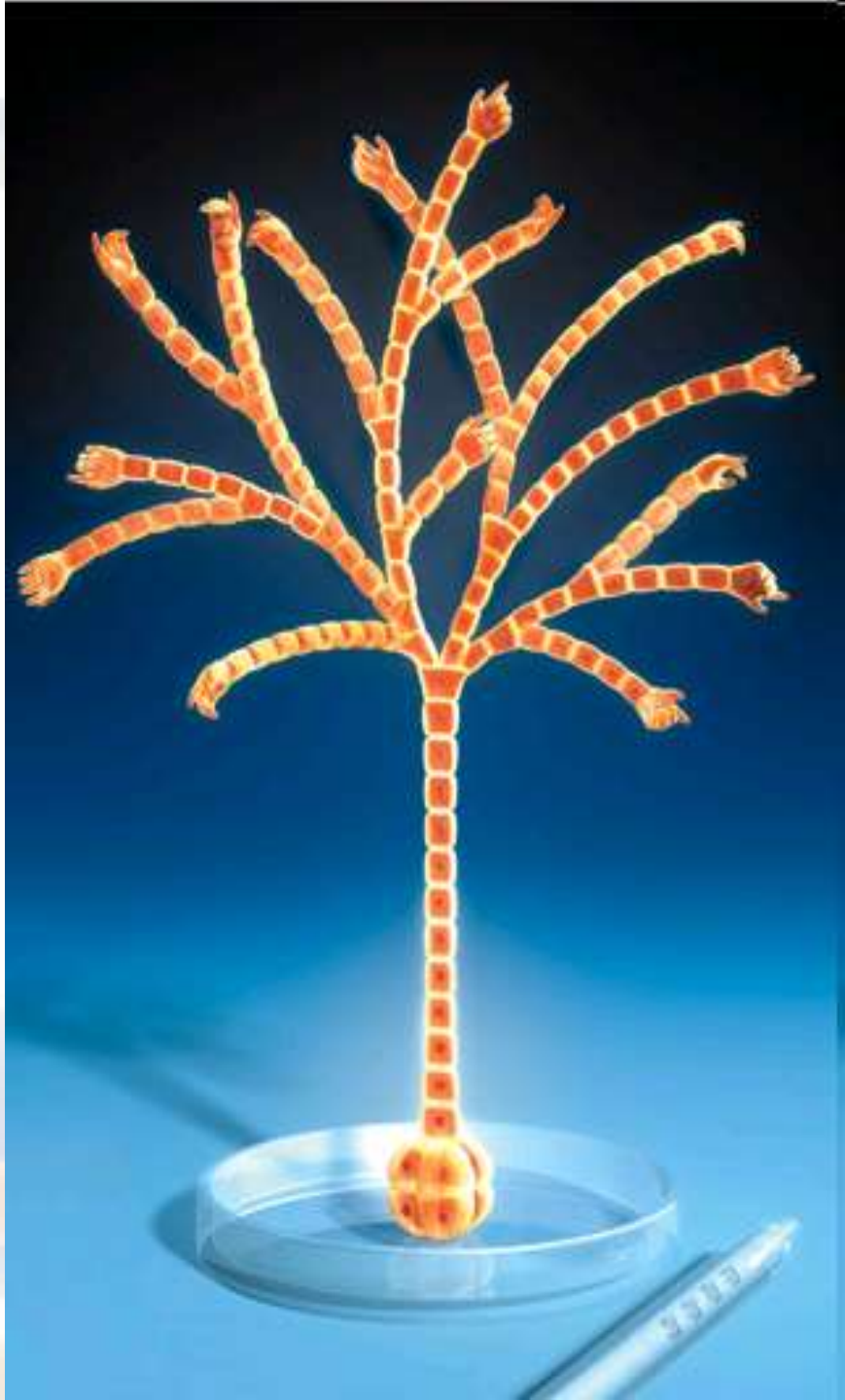
1. Stem Cells are very unique cells.
2. Stem Cells have the amazing ability to develop into several distinct cell types in the body.
3. Stem Cells can be used as a repair system for the body.
4. Stem Cells can theoretically divide without limit in a living organism in order to replenish various types of cells.
5. When a stem cell divides, each new cell has the potential to either remain a stem cell or become another type of cell with a more specialized function (i.e. a muscle cell, a red blood cell, a brain cell, etc.).

Three unique properties of stem cells

■ Stem cells are capable of dividing and renewing themselves for long periods;

■ They are “unspecialized” and they can give rise to specialized cell types.

■ A stem cell is "uncommitted," until it receives a signal to develop into a specialized cell.



Stem Cells

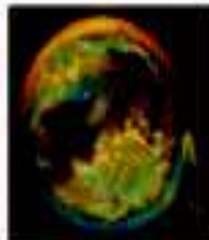
Human Developmental Continuum →



Single-cell
Embryo



3-day
Embryo



5-7 day
Embryo



4-week
Embryo



6-week
Embryo



Infant



Adult

David A. Prentice

Embryonic Stem
(ES) cells
Totipotent

Embryonic Germ
(EG) cells
(primordial germ cells)
Pluripotent

Fetal Tissue
Stem cells
*Pluripotent
or
Multipotent*

Cord Blood Stem cells
Placental Stem cells
Pluripotent or Multipotent

"Adult" Stem cells
*Pluripotent
or
Multipotent*

Teratocarcinoma
(germ cell tumor)

Embryonal
Carcinoma
(EC) cells
Pluripotent

Distinguishing Features of Progenitor/Precursor Cells and Stem Cells.

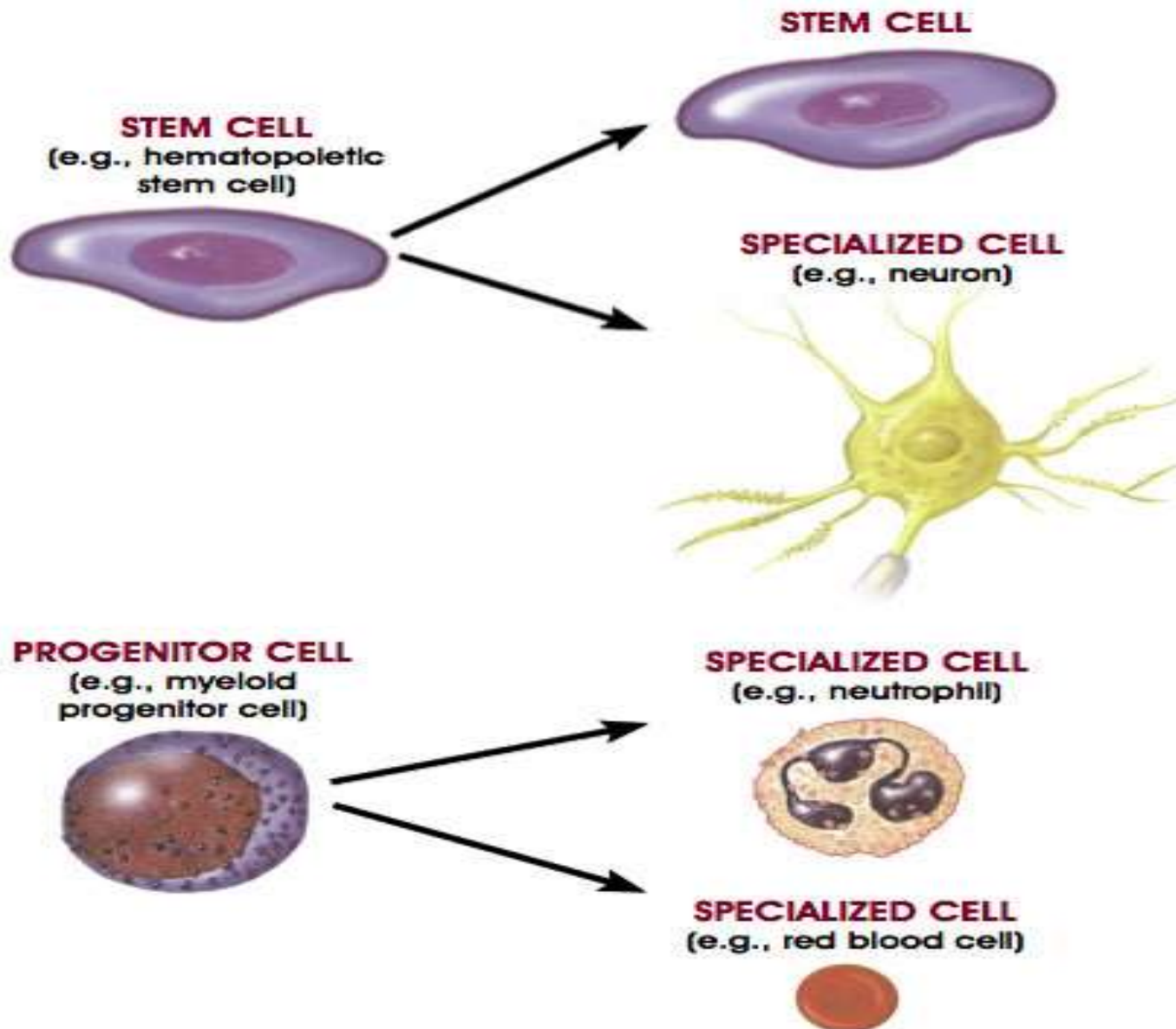
A stem cell is an unspecialized cell that develops into a variety of specialized cell types.

a stem cell divides and gives rise to one additional stem cell and a specialized cell.

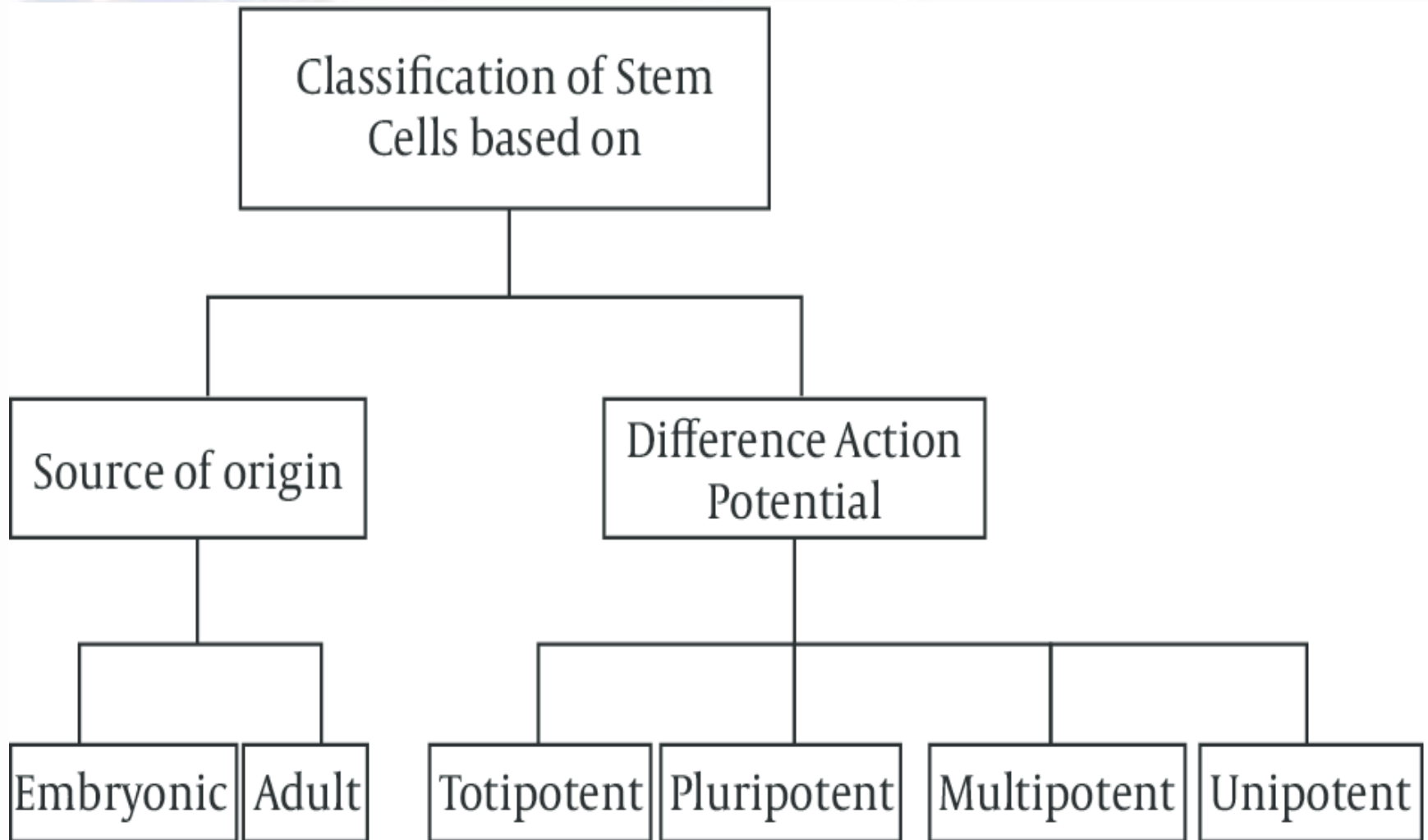
Example: a hematopoietic stem cell produce a second generation stem cell and a neuron.

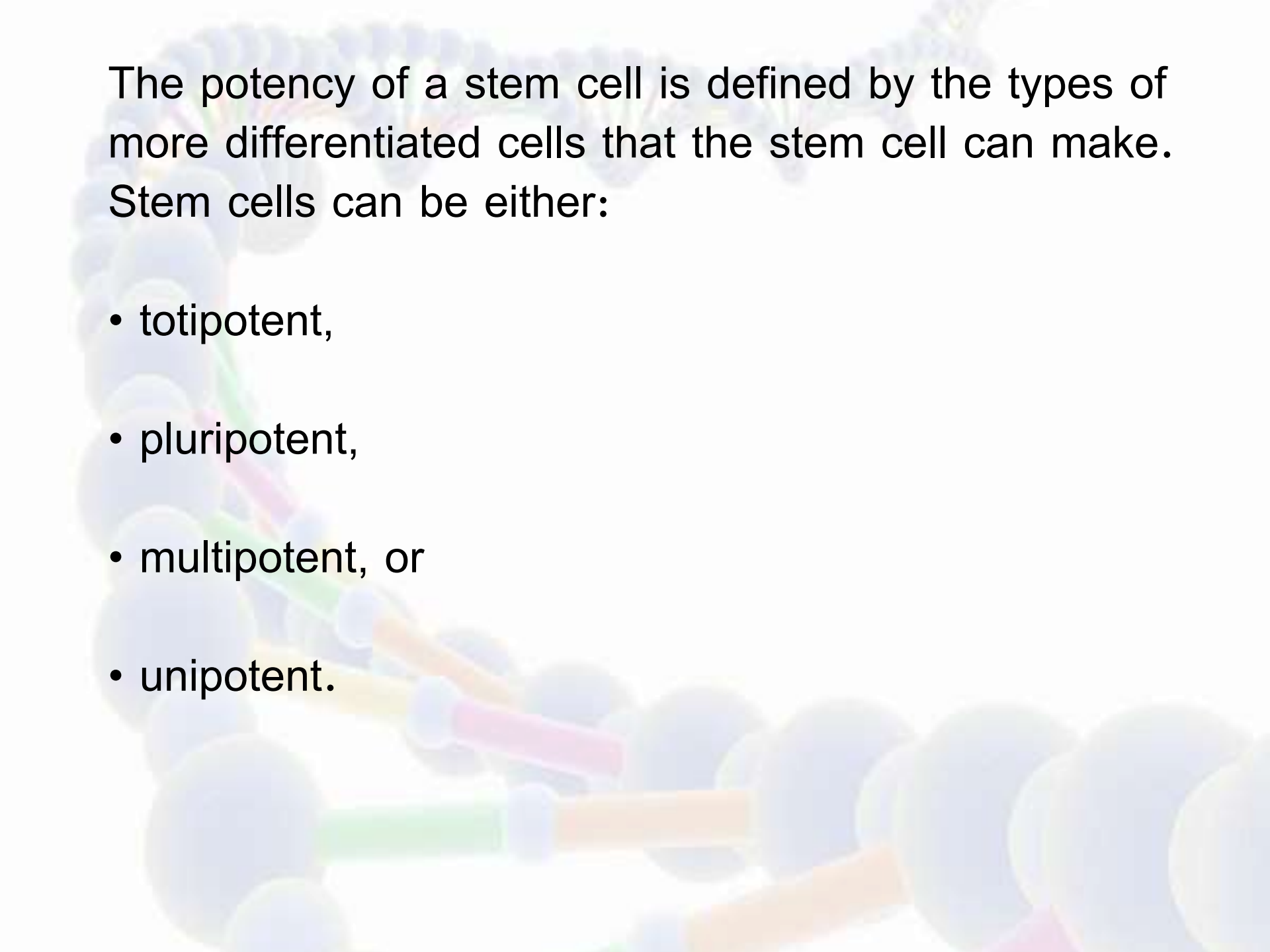
A progenitor cell (a precursor cell) is unspecialized that is capable of undergoing cell division and yielding two specialized cells.

Example: a myeloid progenitor/precursor cell undergoing cell division to yield two specialized cells (a neutrophil and a red blood cell).



classification



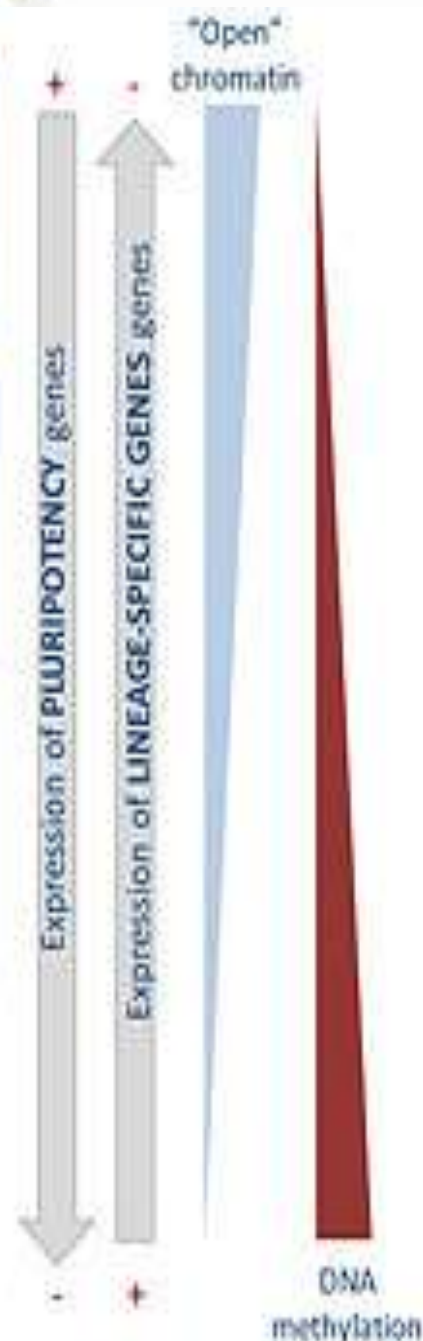
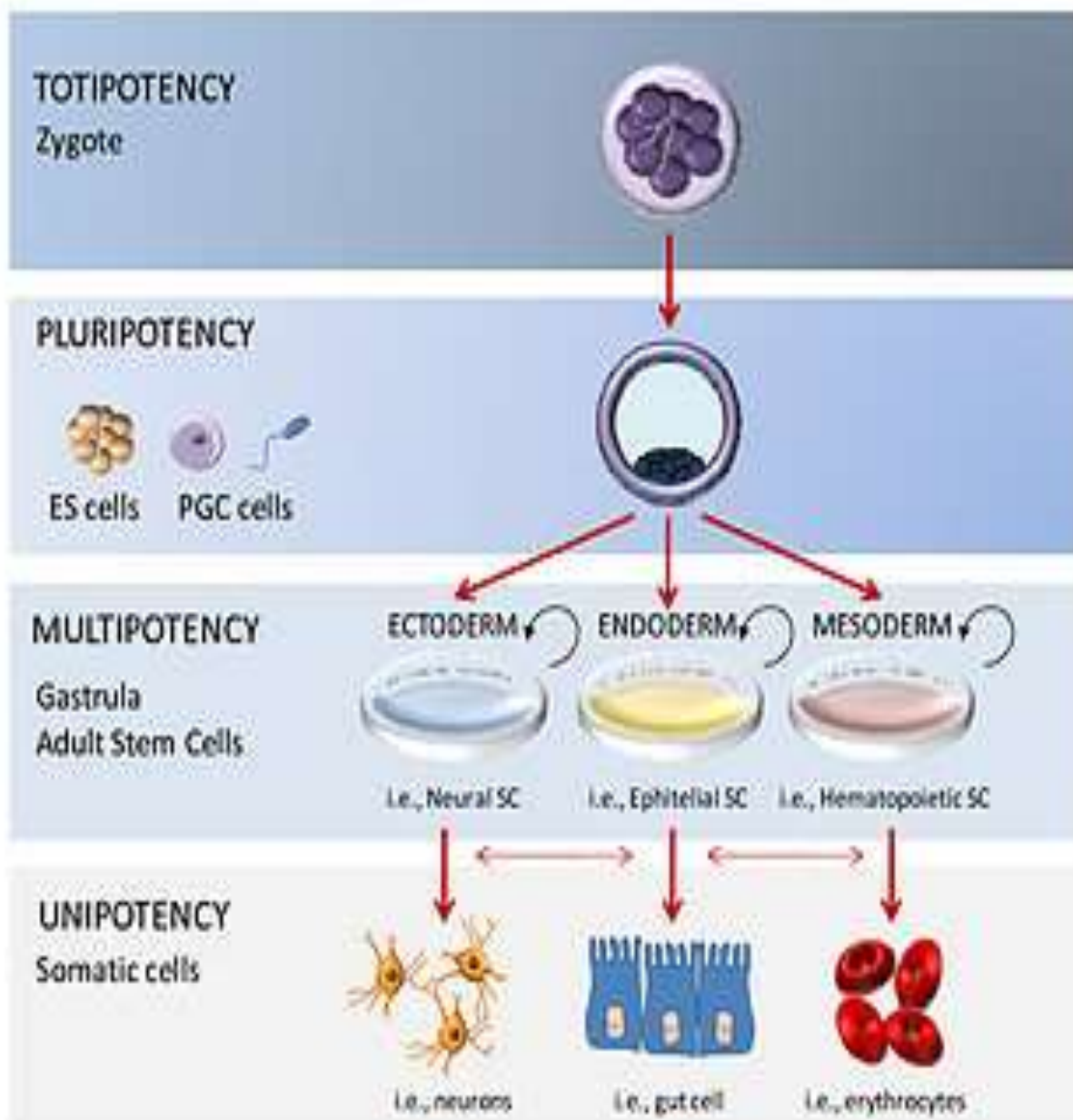


The potency of a stem cell is defined by the types of more differentiated cells that the stem cell can make. Stem cells can be either:

- totipotent,
- pluripotent,
- multipotent, or
- unipotent.

Totipotent cells have the capability to produce all cell types of the developing organism, including both embryonic and extraembryonic (eg, placenta) tissues.

- **Pluripotent** cells can only make cells of the embryo proper, but make all cells of the embryo including germ cells and cells from any of the germ layers. Therefore, they can make any cell of the body.
- **Multipotent** cells can only make cells within a given germ layer. For example, multipotent stem cells from a mesodermal tissue like the blood can make all the cells of the blood, but cannot make cells of a different germ layer such as neural cells (ectoderm) or liver cells (endoderm).
- **Unipotent** cells make cells of a single cell type. An example is a germ cell stem cell that makes the cells that mature to become egg or sperm, but not other cell types.



Embryonic Type stem cells

Embryonic stem cells (ES cells) are pluripotent stem cells derived from the inner cell mass of a blastocyst, an early-stage preimplantation embryo.

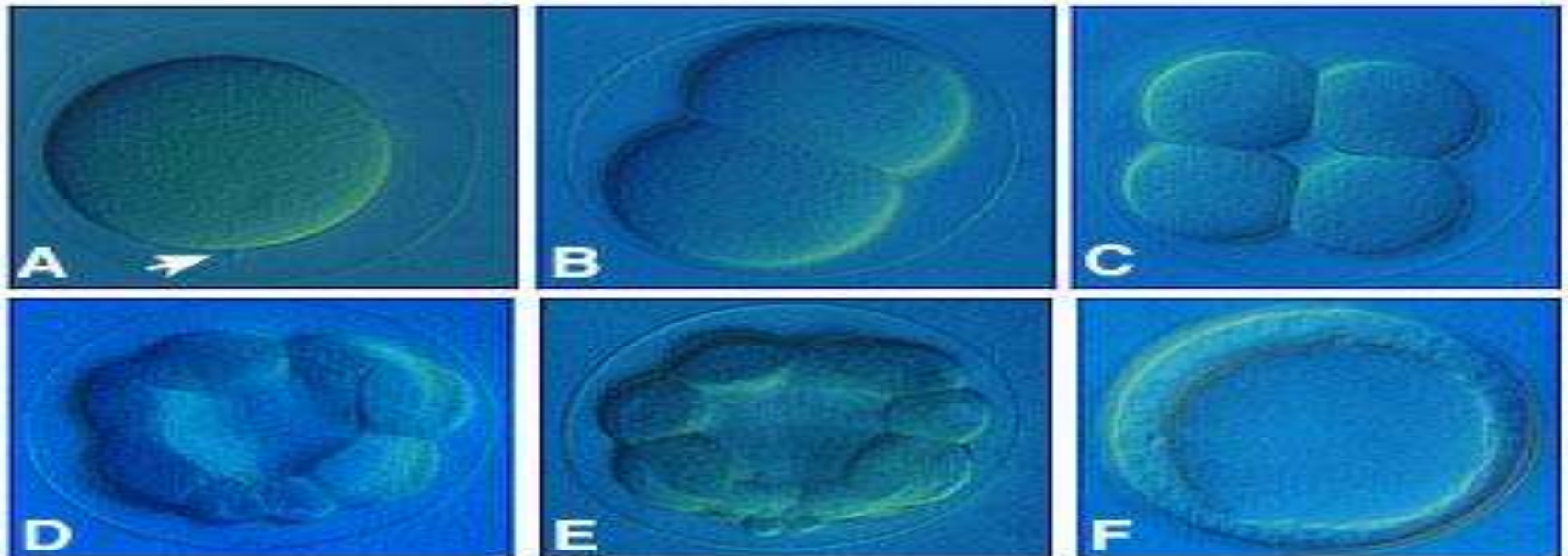
Human embryos reach the blastocyst stage 4–5 days post fertilization, at which time they consist of 50–150 cells.

Isolating the embryoblast or inner cell mass (ICM) results in destruction of the blastocyst, which raises ethical issues, including whether or not embryos at the pre-implantation stage should be considered to have the same moral or legal status as more developed human beings.

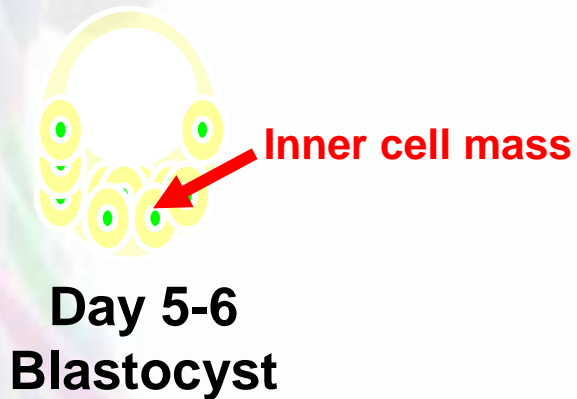
Human ES cells measure approximately 14 μm while mouse ES cells are closer to 8 μm

- Embryonic stem cells of the inner cell mass are pluripotent, that is, they are able to differentiate to generate primitive ectoderm, which ultimately differentiates during gastrulation into all derivatives of the three primary germ layers: ectoderm, endoderm, and mesoderm.
- These include each of the more than 220 cell types in the adult body.
- Pluripotency distinguishes embryonic stem cells from adult stem cells found in adults; while embryonic stem cells can generate all cell types in the body, adult stem cells are multipotent and can produce only a limited number of cell types.
- If the pluripotent differentiation potential of embryonic stem cells could be harnessed in vitro, it might be a means of deriving cell or tissue types virtually to order.
- This would provide a radical new treatment approach to a wide variety of conditions where age, disease, or trauma has led to tissue damage or dysfunction.

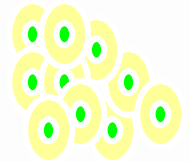
Additionally, under defined conditions, embryonic stem cells are capable of propagating themselves indefinitely in an undifferentiated state and have the capacity when provided with the appropriate signals to differentiate, presumably via the formation of precursor cells, to almost all mature cell phenotypes. This allows embryonic stem cells to be employed as useful tools for both research and regenerative medicine, because they can produce limitless numbers of themselves for continued research or clinical use



How to Derive an Embryonic Stem Cell Line?



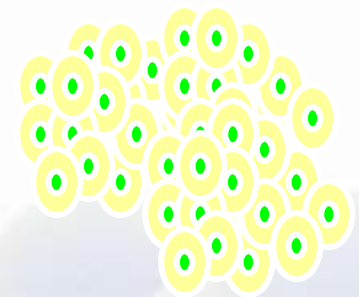
(destroys embryo)
ETHICS?



Culture cells

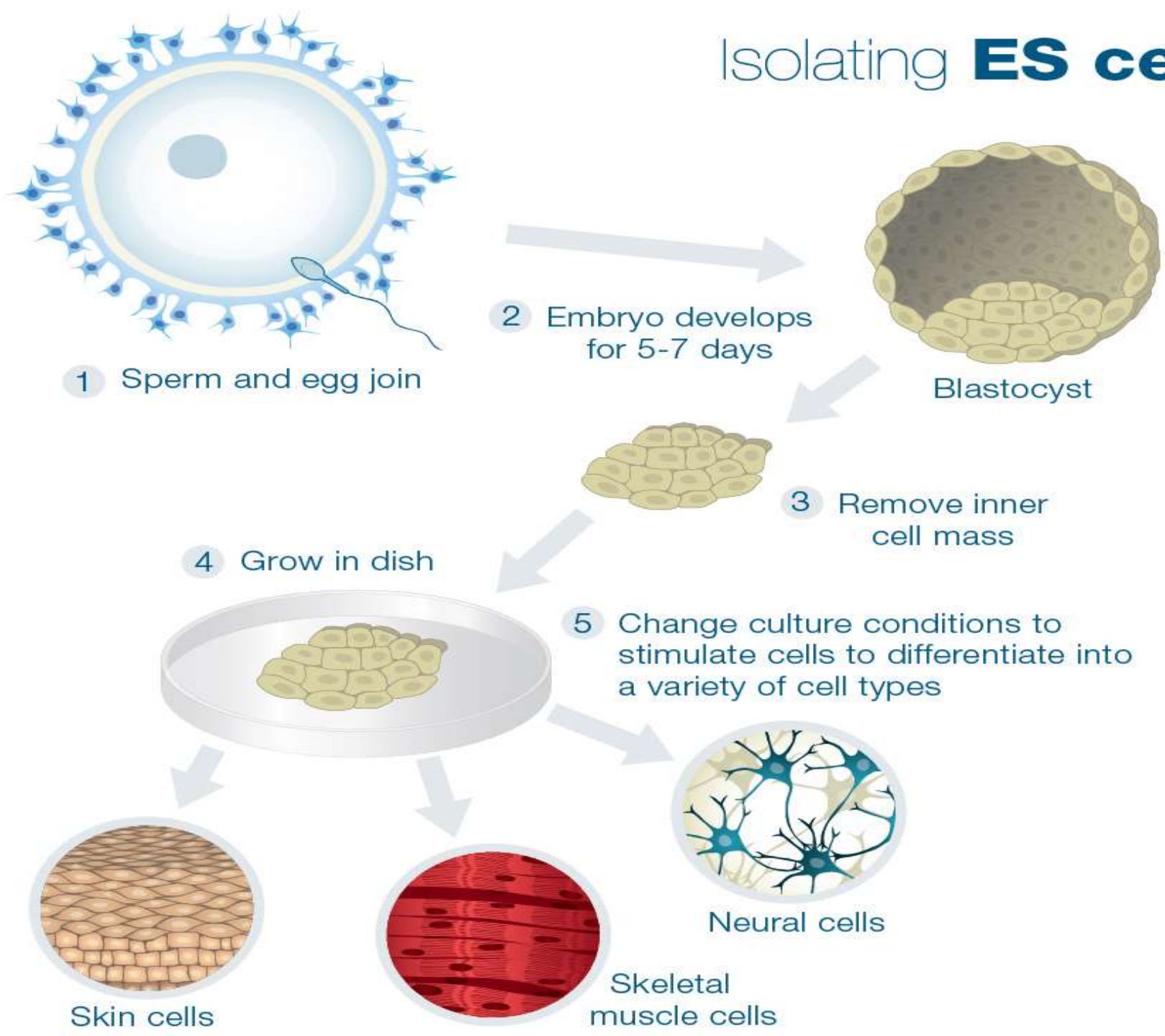


A stem cell line is composed of a population of cells that can replicate themselves for long periods of time *in vitro* (out of the body)



**An embryonic
stem cell
clone**

Isolating **ES** cells



Advantages of Embryonic Stem Cell

1. Flexible – appear to have the potential to make any cell.

2. Immortal – one embryonic stem cell line can potentially provide an endless supply of cells with defined characteristics.

3. Availability – embryos from *in vitro* fertilization clinics.



Disadvantages of Embryonic Stem Cell

1. **Difficult to differentiate uniformly and homogeneously** into a target tissue.
2. **Immunogenic** – embryonic stem cells from a random embryo donor are likely to be rejected after transplantation
3. **Tumorigenic** – capable of forming tumors or promoting tumor formation.
4. **Destruction of developing human life.**

adult stem cells

- An **adult stem cell** is thought to be an **undifferentiated** cell, found among differentiated cells in a tissue or organ.
- The adult stem cell can renew itself and can differentiate to yield some or all of the major specialized cell types of the tissue or organ.
- The primary roles of adult stem cells in a living organism are to maintain and repair the tissue in which they are found.
- Scientists also use the term **somatic stem cell** instead of adult stem cell, where somatic refers to cells of the body (not the germ cells, sperm or eggs).
- Unlike **embryonic stem cells**, which are defined by their origin (cells from the **preimplantation**-stage embryo), the origin of adult stem cells in some mature tissues is still under investigation.

Differentiation pathways of adult stem cells

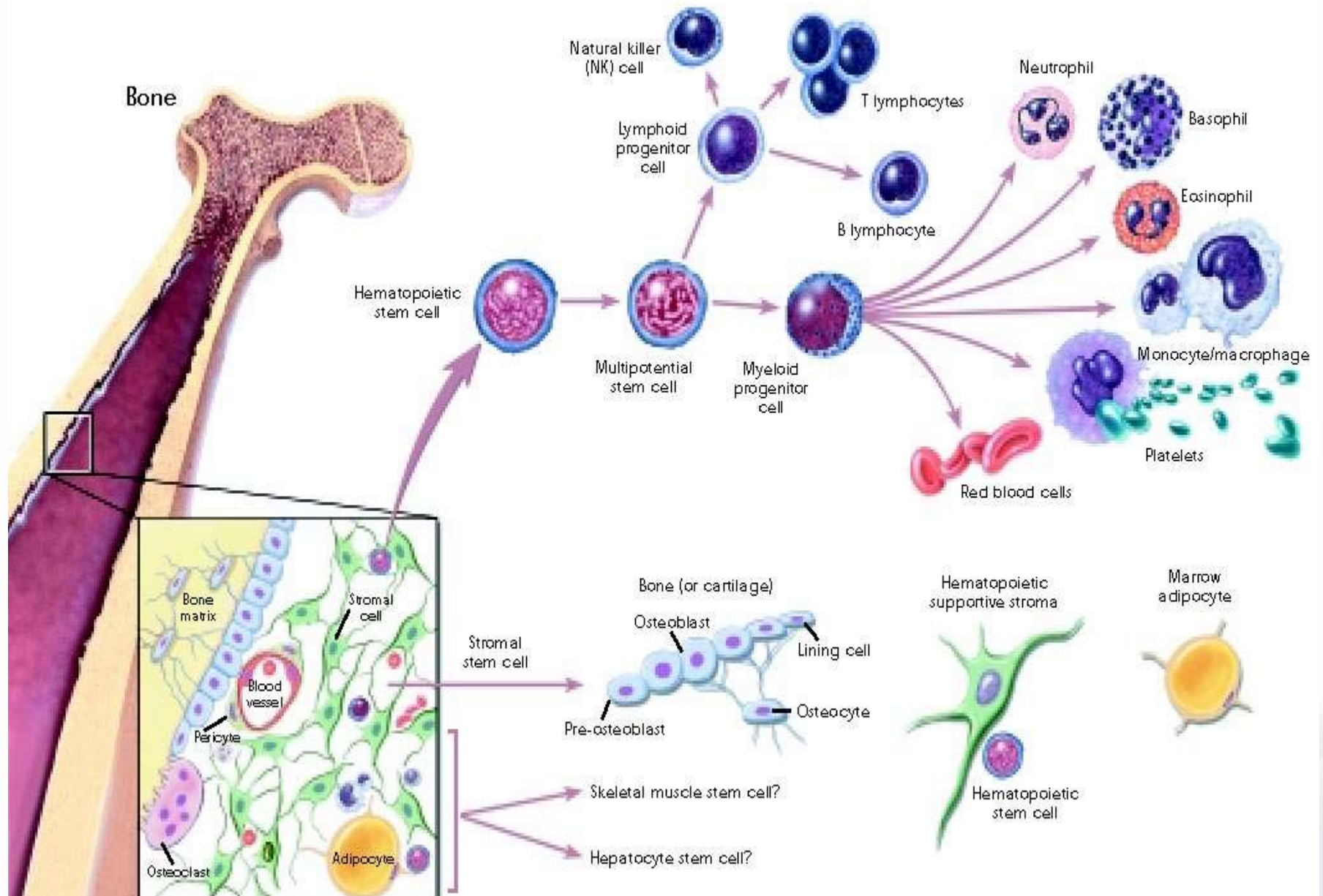
Neural stem cells in the brain give rise to its three major cell types: nerve cells (neurons) and two categories of non-neuronal cells — astrocytes and oligodendrocytes.

Epithelial stem cells in the lining of the digestive tract occur in deep crypts and give rise to several cell types: absorptive cells, goblet cells, Paneth cells, and enteroendocrine cells.

Skin stem cells occur in the basal layer of the epidermis and at the base of hair follicles.

The epidermal stem cells give rise to keratinocytes, which migrate to the surface of the skin and form a protective layer.

The follicular stem cells can give rise to both the hair follicle and to the epidermis



Advantages of Adult Stem Cell

1. **Adult stem cells from bone marrow and umbilical cords appear to be as flexible as the embryonic type**
2. **Somewhat specialized** – inducement may be simpler.
3. **Not immunogenic** – recipients who receive the products of their own stem cells will not experience immune rejection.
4. **Relative ease of procurement** – some adult stem cells are easy to harvest (skin, muscle, marrow, fat)
5. **Non-tumorigenic**—tend not to form tumors.
6. **No harm done** to the donor.



Disadvantages of Adult stem cells




- 1. Limited quantity** - can sometimes be difficult to obtain in large numbers.
- 2. Finite** - may not live as long as embryonic stem cells in culture.
- 3. Less flexible** - may be more difficult to reprogram to form other tissue types



Why are adult stem cells preferable to embryonic stem cells?

- Adult stem cells naturally exist in our bodies, and they provide a natural repair mechanism for many tissues.
- They belong in the microenvironment of an adult body, while embryonic stem cells belong in the microenvironment of the early embryo, where they tend to cause tumors and immune system reactions.

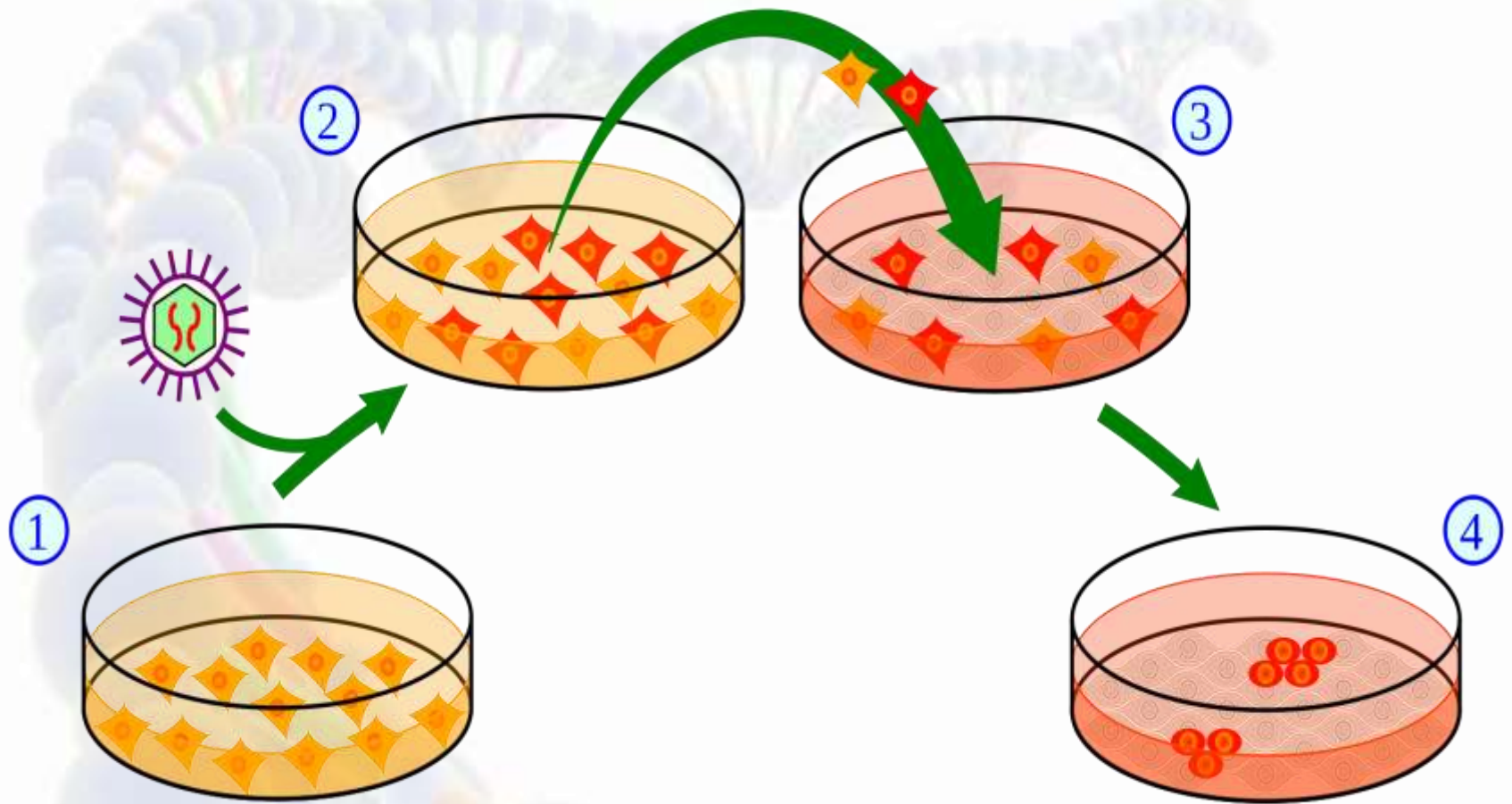
COMPARISON OF THE DIFFERENT SOURCES OF STEM CELLS

	Embryonic Stem Cells		Adult Stem Cells
Attributes	 <p>In Vitro Fertilization</p> <ul style="list-style-type: none"> • can produce all cell types • relatively easy to identify, isolate, maintain, and grow in the laboratory • large source of "excess" blastocysts from IVF clinics 	 <p>Nuclear Transfer</p> <ul style="list-style-type: none"> • can produce all cell types • relatively easy to identify, isolate, maintain, and grow in the laboratory • stem cells may be genetically matched to patient 	 <p>Adult Tissues</p> <ul style="list-style-type: none"> • demonstrated success in some treatments • stem cells may be genetically matched to patient
	<p>Limitations</p> <ul style="list-style-type: none"> • limited number of cell lines available for federally funded research • risk of creating teratomas (tumors) from implanting undifferentiated stem cells 	<ul style="list-style-type: none"> • not yet achieved with human cells • risk of creating teratomas (tumors) from implanting undifferentiated stem cells 	<ul style="list-style-type: none"> • produce limited number of cell types • not found in all tissues • difficult to identify, isolate, maintain, and grow in the laboratory
Ethical Concerns	<ul style="list-style-type: none"> • destruction of human blastocysts • donation of blastocysts requires informed consent 	<ul style="list-style-type: none"> • destruction of human blastocysts • donation of eggs requires informed consent • concern about misapplication for reproductive cloning 	<ul style="list-style-type: none"> • no major ethical concerns have been raised

Induced pluripotent stem cells

- **Induced pluripotent stem cells (iPSCs)** are adult cells that have been genetically reprogrammed to an embryonic stem cell–like state by being forced to express genes and factors important for maintaining the defining properties of embryonic stem cells.
- Although these cells meet the defining criteria for pluripotent stem cells, it is not known if iPSCs and embryonic stem cells differ in clinically significant ways.
- Mouse iPSCs were first reported in 2006, and human iPSCs were first reported in late 2007.
- Mouse iPSCs demonstrate important characteristics of pluripotent stem cells, including expressing stem cell markers, forming tumors containing cells from all three germ layers, and being able to contribute to many different tissues when injected into mouse embryos at a very early stage in development.
- Human iPSCs also express stem cell markers and are capable of generating cells characteristic of all three **germ layers**.

- Although additional research is needed, iPSCs are already useful tools for drug development and modeling of diseases, and scientists hope to use them in transplantation medicine.
- Viruses are currently used to introduce the reprogramming factors into adult cells, and this process must be carefully controlled and tested before the technique can lead to useful treatment for humans.
- In animal studies, the virus used to introduce the stem cell factors sometimes causes cancers.
- Researchers are currently investigating non-viral delivery strategies.
- In any case, this breakthrough discovery has created a powerful new way to "de-differentiate" cells whose developmental fates had been previously assumed to be determined.
- In addition, tissues derived from iPSCs will be a nearly identical match to the cell donor and thus probably avoid rejection by the immune system.
- The iPSC strategy creates pluripotent stem cells that, together with studies of other types of pluripotent stem cells, will help researchers learn how to reprogram cells to repair damaged tissues in the human body



(1) Isolate and culture donor cells. (2) Transduce stem cell-associated genes into the cells by viral vectors. Red cells indicate the cells expressing the exogenous genes. (3) Harvest and culture the cells according to ES cell culture, using mitotically inactivated feeder cells (lightgray). (4) A small subset of the transfected cells become iPS cells and generate ES-like colonies.



Source of Stem cells

- Stem cells may be derived from autologus, allogeneic or xenogenic sources.
- Histocompatibility is prerequisite for transplantation of allogeneic stem cells.
- Fetal tissue is the best current tissue source for human neural stem cells, however ethical issues are a major concern.

Potential sources of stem cells are:

- fetal tissue that becomes available after an abortion
- excess embryos from assisted reproductive technologies such as commonly used in fertility clinics
- embryos created through in vitro fertilization specifically for research purpose, and
- embryos created asexually as a result of the transfer of a human somatic cell nucleus to an egg with its own nucleus removed.
- Other sources of stem cells are those from umbilical cord blood, and bone marrow.
- In addition, neural stem cells, haematopoietic stem cells and mesenchymal stem cells can be harvested from fetal blood and fetal tissue.

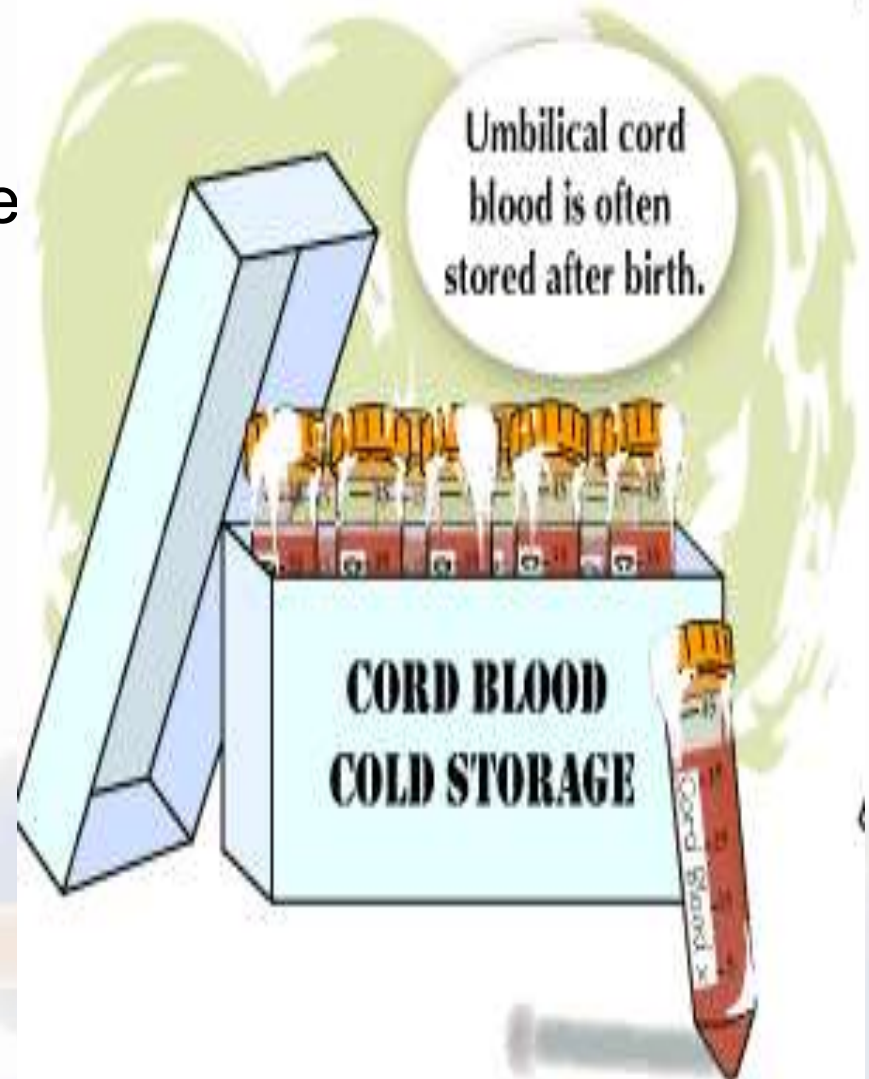
Placenta a Source of Stem Cells

Placental stem cells, like umbilical cord blood and bone marrow stem cells, can be used to cure chronic blood-related disorders such as sickle cell disease, Thalassaemia, and leukaemia.



Umbilical Cord Blood Stem Cell Transplant

Umbilical cord blood stem cell transplants are less prone to rejection than either bone marrow or peripheral blood stem cells. This is probably because the cells have not yet developed the features that can be recognized and attacked by the recipient's immune system.





Applications

- **Stem cells can be used to study development**
- Stem cells may help us understand how a complex organism develops from a fertilised egg.
- In the laboratory, scientists can follow stem cells as they divide and become increasingly specialized, making skin, bone, brain, and other cell types. Identifying the signals and mechanisms that determine whether a stem cell chooses to carry on replicating itself or differentiate into a specialized cell type, and into which cell type, will help us understand what controls normal development.
- Some of the most serious medical conditions, such as cancer and birth defects, are due to abnormal cell division and differentiation.
- A better understanding of the genetic and molecular controls of these processes may yield information about how such diseases arise and suggest new strategies for therapy. This is an important goal of stem cell research.

Stem cells have the ability to replace damaged cells and treat disease

- This property is already used in the treatment of extensive burns, and to restore the blood system in patients with leukaemia and other blood disorders.
- Stem cells may also hold the key to replacing cells lost in many other devastating diseases for which there are currently no sustainable cures.
- Today, donated tissues and organs are often used to replace damaged tissue, but the need for transplantable tissues and organs far outweighs the available supply.
- Stem cells, if they can be directed to differentiate into specific cell types, offer the possibility of a renewable source of replacement cells and tissues to treat diseases including Parkinson's, stroke, heart disease and diabetes.
- This prospect is an exciting one, but significant technical hurdles remain that will only be overcome through years of intensive research.

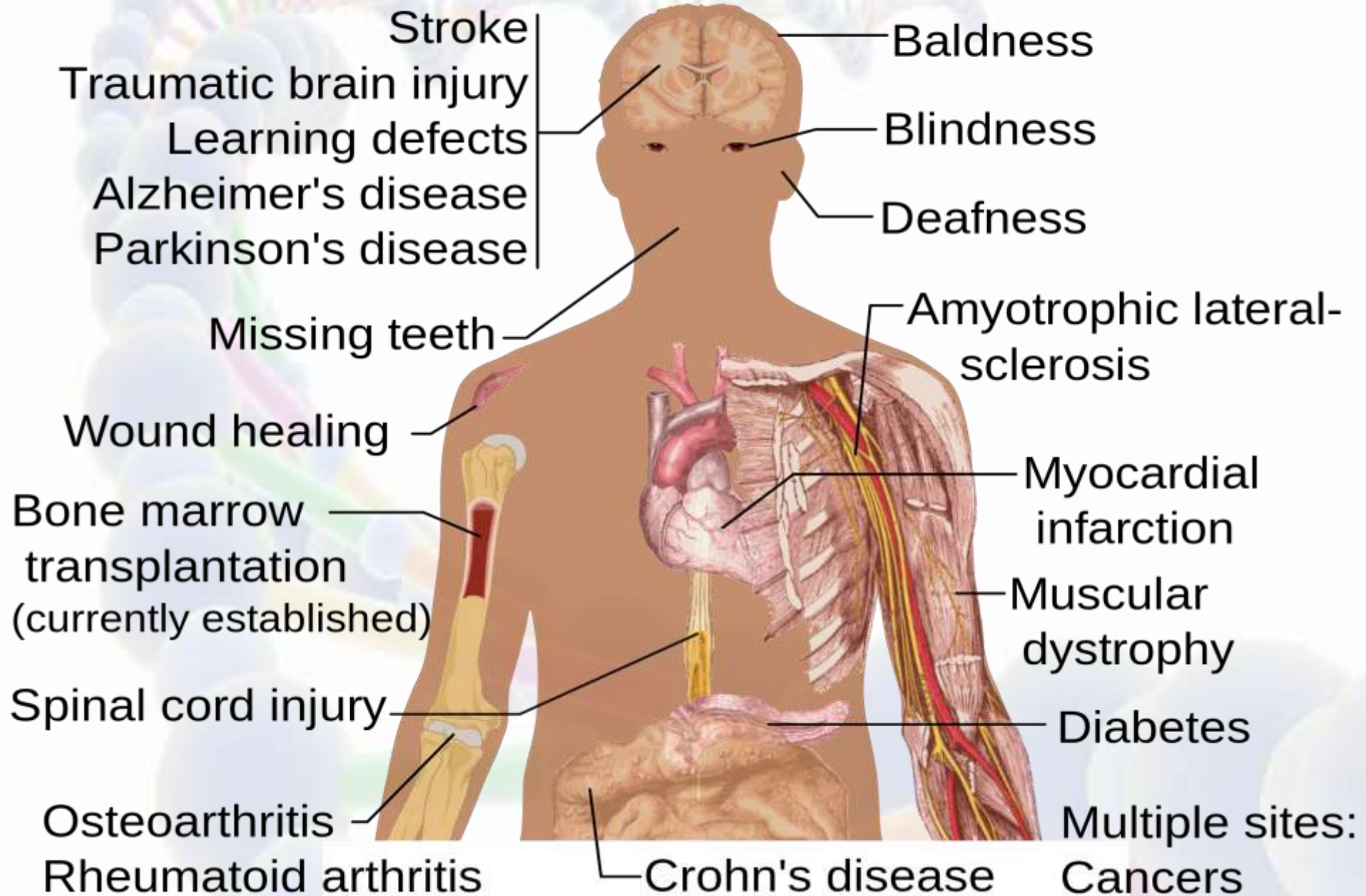
Stem cells could be used to study disease

- In many cases it is difficult to obtain the cells that are damaged in a disease, and to study them in detail.
- Stem cells, either carrying the disease gene or engineered to contain disease genes, offer a viable alternative.
- Scientists could use stem cells to model disease processes in the laboratory, and better understand what goes wrong

Stem cells could provide a resource for testing new medical treatments

- New medications could be tested for safety on specialized cells generated in large numbers from stem cell lines – reducing the need for animal testing.
- Other kinds of cell lines are already used in this way.
- Cancer cell lines, for example, are used to screen potential anti-tumour drugs.

Potential uses of **Stem cells**



POSSIBLE USES OF TISSUE DERIVED FROM STEM CELLS TO TREAT DISEASE

Cell type	Target disease
Neural (nerve) cells	Stroke, Parkinson's disease, Alzheimer's disease, spinal cord injury, multiple sclerosis
Heart muscle cells	Heart attacks, congestive heart failure
Insulin-producing cells	Diabetes
Cartilage cells	Osteoarthritis
Blood cells	Cancer, immunodeficiencies, inherited blood diseases, leukemia
Liver cells	Hepatitis, cirrhosis
Skin cells	Burns, wound healing
Bone cells	Osteoporosis
Retinal (eye) cells	Macular degeneration
Skeletal muscle cells	Muscular dystrophy

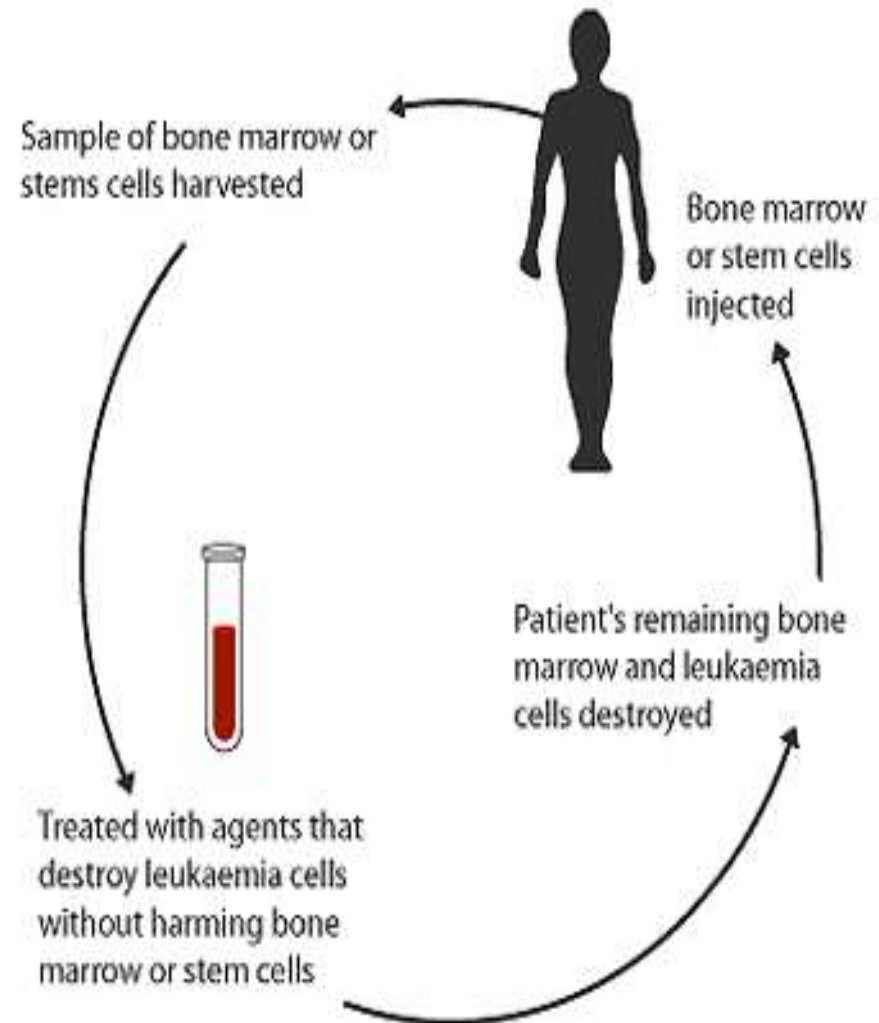


Cell therapy.

- Treatment of neural diseases such as Parkinson's disease, Huntington's disease and Alzheimer's disease.
- ◆ Stem cells could be used to repair or replace damaged neurons.
- ◆ Repair of damaged organs such as the liver and pancreas.
- ◆ Treatments for AIDS.

How Does Cell Therapy Work?

- Stem cells can be used to generate healthy and functioning specialized cells, which can then replace diseased or dysfunctional cells.
- It is similar to the process of organ transplantation only the treatment consists of **transplanting cells instead of organs**.



How Does Cell Therapy Work?

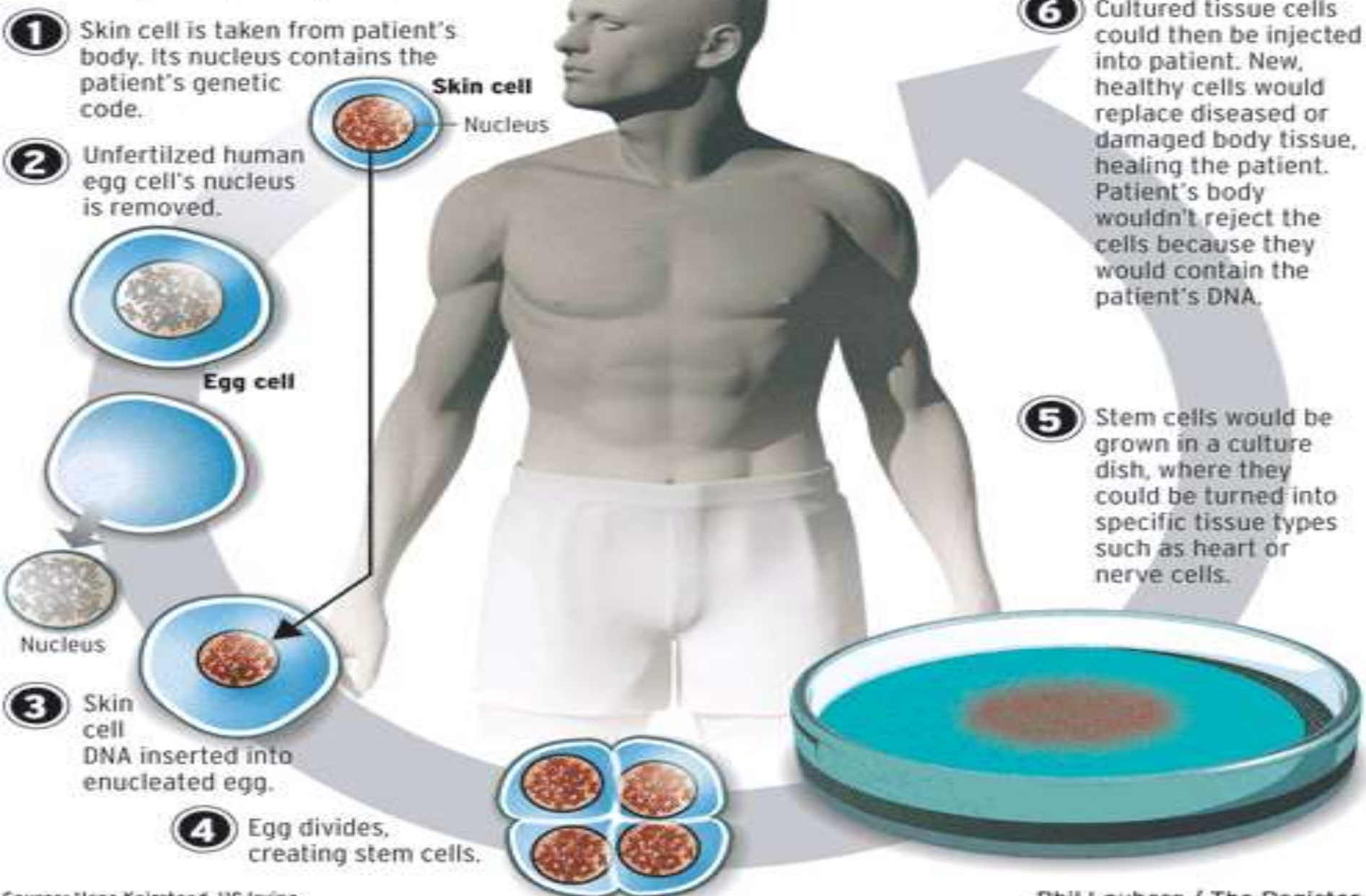
Bone marrow transplants are an example of cell therapy in which the stem cells in a donor's marrow are used to replace the blood cells of the victims of leukemia.

Cell therapy is also being used in experiments to **graft new skin cells** to treat serious burn victims, and to **grow new corneas** for the sight-impaired.

In all of these uses, the goal is for the healthy cells to become integrated into the body and begin to function like the patient's own cells.

How therapeutic cloning could work

Cloning human tissue has never been done, but one way it might be performed:

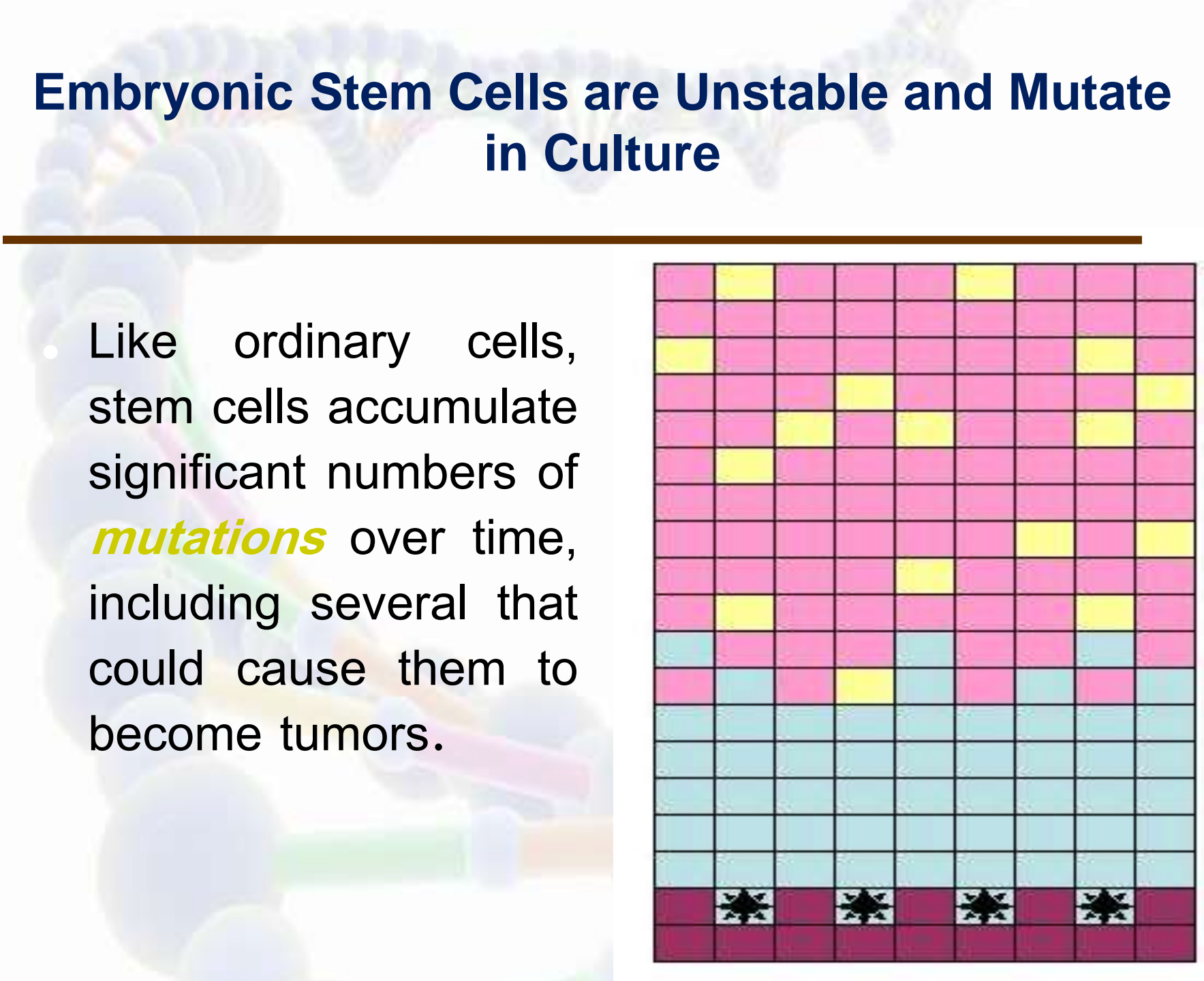


Obstacles of Stem Cell Research

- **How to find the right type of stem cells?**
- **How to put the stem cells into the right place?**
- **Will the stem cells perform the desired function in the body?**
- **Differentiation protocols for many cell types have not been developed.**

[illegible]

- [illegible]





Ethical debate

- Harvesting ES cells destroys the blast cyst
- “This is murder”
- ES cell research requires human cells
- Could create a commercial market for human cells
- “This devalues life”



Destroying life to cure some one – Ethical ?

- If stem cells have such potential to relieve suffering, why are so many people so upset about their use? The reason is that the most powerful type of stem cell  embryonic stem (ES) cells  can only be obtained from human embryos. Many people think that it's wrong to create and destroy human embryos to treat disease



Religious Debate over Harvesting Embryonic Stem Cells

- The pro-life group generally believes that:
 - Personhood happens at, or shortly after, conception.
 - Thus, they consider the removal of stem cells from an embryo -- a procedure which kills the stem cells -- to be a form of murder of a human being.
 - They argue that no potential health benefits to even hundreds of millions of people can justify the murder of other humans.



**Day 5-6
Blastocyst**



Why we should support *Can help several disabled*

- Human embryonic stem cell (HESC) research offers great promise of cures for otherwise incurable conditions: spinal cord injuries, ALS, Alzheimer's, Parkinson's, etc.



Shall be Clone Humans ?

- Arguments for and against human cloning research. Should we ban human cloning? Why investors are moving away from human cloning and why human cloning now looks a last-century way to fight disease. Why some people want to clone themselves or even to clone the dead.



