

Stem Cells

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MICROBIOLOGY SEM 2 CC4, Unit-5

Stem cell, an undifferentiated cell that can divide to produce some offspring cells that continue as stem cells and some cells that are destined to differentiate (become specialized). Stem cells are an ongoing source of the differentiated cells that make up the tissues and organs of animals and plants.

Stem cells have the remarkable potential to develop into many different cell types in the body during early life and growth. In addition, in many tissues they serve as a sort of internal repair system, dividing essentially without limit to replenish other cells as long as the person or animal is still alive. When a stem cell divides, each new cell has the potential either to remain a stem cell or become another type of cell with a more specialized function, such as a muscle cell, a red blood cell, or a brain cell.

Properties

- **Self-renewal:** the ability to go through numerous cycles of cell division while maintaining the undifferentiated state.
- **Potency:** the capacity to differentiate into specialized cell types. In the strictest sense, this requires stem cells to be either totipotent or pluripotent—to be able to give rise to any mature cell type, although multipotent or unipotent progenitor cells are sometimes referred to as stem cells. Apart from this, it is said that stem cell function is regulated in a feedback mechanism.
- They are capable of dividing and **renewing** themselves for long periods;
- They are **unspecialized cells** capable of renewing themselves through cell division, sometimes after long periods of inactivity.
- They can give rise to **specialized cell types**.

Potency specifies the differentiation potential (the potential to differentiate into different cell types) of the stem cell

1. A **multipotent cell** can give rise to several types of mature cell.
2. A **pluripotent cell** can give rise to all types of adult tissue cells plus extraembryonic tissue: cells which support embryonic development.
3. A **totipotent cell** can give rise to a new individual given appropriate maternal support.
4. **Oligopotent stem cells** can differentiate into only a few cell types, such as lymphoid or myeloid stem cells.
5. **Unipotent cells** can produce only one cell type, their own, but have the property of self-renewal, which distinguishes them from non-stem cells (e.g. progenitor cells, which cannot self-renew).

Until recently, scientists primarily worked with two kinds of stem cells from animals and humans: embryonic stem cells and non-embryonic "**somatic**" or "**adult**" stem cells. The functions and characteristics of these cells will be explained in this document. Scientists discovered ways to derive embryonic stem cells from early mouse embryos more than 30 years ago, in 1981. The detailed study of the biology of mouse stem cells led to the discovery, in 1998, of a method to derive stem cells from human embryos and grow the cells in the laboratory. These cells are called human embryonic stem cells. The embryos used in these studies were created for reproductive purposes through *in vitro* fertilization procedures. When they were no longer needed for that purpose, they were donated for research with the informed consent of the donor. In 2006, researchers made another breakthrough by identifying conditions that would allow some specialized adult cells to be "reprogrammed" genetically to assume a stem cell-like state. This new type of stem cell, called induced pluripotent stem cells (iPSCs), will be discussed in a later section of this document. adult cells to be "reprogrammed" genetically to assume a stem cell-like state. This new type of stem cell, called induced pluripotent stem cells (iPSCs), will be discussed in a later section of this document.

Stem cells are important for living organisms for many reasons.

- In the 3- to 5-day-old embryo, called a blastocyst, the inner cells give rise to the entire body of the organism, including all of the many specialized cell types and organs such as the heart, lungs, skin, sperm, eggs and other tissues.

- In some adult tissues, such as bone marrow, muscle, and brain, discrete populations of adult stem cells generate replacements for cells that are lost through normal wear and tear, injury, or disease.
- Given their unique regenerative abilities, stem cells offer new potentials for treating diseases such as diabetes, and heart disease.
- Laboratory studies of stem cells enable scientists to learn about the cells' essential properties and what makes them different from specialized cell types. Scientists are already using stem cells in the laboratory to screen new drugs and to develop model systems to study normal growth and identify the causes of birth defects.
- Stem cell research is one of the most fascinating areas of contemporary biology, but, as with many expanding fields of scientific inquiry, research on stem cells raises scientific questions as rapidly as it generates new discoveries.

What are embryonic stem cells?

Embryonic stem cells (often referred to as ES cells) are stem cells that are derived from the inner cell mass of a mammalian embryo at a very early stage of development, when it is composed of a hollow sphere of dividing cells (a blastocyst). Embryonic stem cells from human embryos and from embryos of certain other mammalian species can be grown in tissue culture. The ES cells also are called **pluripotent cell**.

Properties:

- Derived from five-day old human embryos before specialised tissues of the body begin to form.
- May be grown indefinitely in culture in the primitive embryonic state.
- Retain the property of pluripotency during extended growth in vitro.
- Derived directly and at high frequency from pluripotent cell populations in vivo
- Grow indefinitely in vitro (express telomerase)
- Maintain normal karyotype
- Cloned lines capable of differentiation into a wide range of somatic and extraembryonic tissues in vivo and in vitro-at high frequency and under a range of conditions
- Capable of colonising all tissues including germ line after blastocyst injection to give chimeric offspring

Embryonic stem cells have important applications

- In biomedical research
- Basic studies of early human development and its disorders-birth defects, childhood cancers
- Functional genomics in human cell
- Discovery of novel factors controlling tissue regeneration and repair
- In vitro models for drug discovery and toxicology
- Source of tissue for transplantation medicine

What laboratory tests are used to identify embryonic stem cells?

At various points during the process of generating embryonic stem cell lines, scientists test the cells to see whether they exhibit the fundamental properties that make them embryonic stem cells. This process is called characterization.

- Growing and subculturing the stem cells for many months. This ensures that the cells are capable of long-term growth and self-renewal. Scientists inspect the cultures through a microscope to see that the cells look healthy and remain undifferentiated.
- Using specific techniques to determine the presence of transcription factors that are typically produced by undifferentiated cells. Two of the most important transcription factors are Nanog and Oct4. Transcription factors help turn genes on and off at the right time, which is an important part of the processes of

cell differentiation and embryonic development. In this case, both Oct 4 and Nanog are associated with maintaining the stem cells in an undifferentiated state, capable of self-renewal.

- Using specific techniques to determine the presence of particular cell surface markers that are typically produced by undifferentiated cells.
- Examining the chromosomes under a microscope. This is a method to assess whether the chromosomes are damaged or if the number of chromosomes has changed. It does not detect genetic mutations in the cells.
- Determining whether the cells can be re-grown, or subculture, after freezing, thawing, and re-plating.

How are embryonic stem cells stimulated to differentiate?

As long as the embryonic stem cells in culture are grown under appropriate conditions, they can remain undifferentiated (unspecialized). But if cells are allowed to clump together to form embryoid bodies, they begin to differentiate spontaneously. They can form muscle cells, nerve cells, and many other cell types. Although spontaneous differentiation is a good indication that a culture of embryonic stem cells is healthy, the process is uncontrolled and therefore an inefficient strategy to produce cultures of specific cell types.

So, to generate cultures of specific types of differentiated cells—heart muscle cells, blood cells, or nerve cells, for example—scientists try to control the differentiation of embryonic stem cells. They change the chemical composition of the culture medium, alter the surface of the culture dish, or modify the cells by inserting specific genes. Through years of experimentation, scientists have established some basic protocols or "recipes" for the directed differentiation of embryonic stem cells into some specific cell types (Figure 1). (For additional examples of directed differentiation of embryonic stem cells, refer to the 2006 NIH stem cell report.)

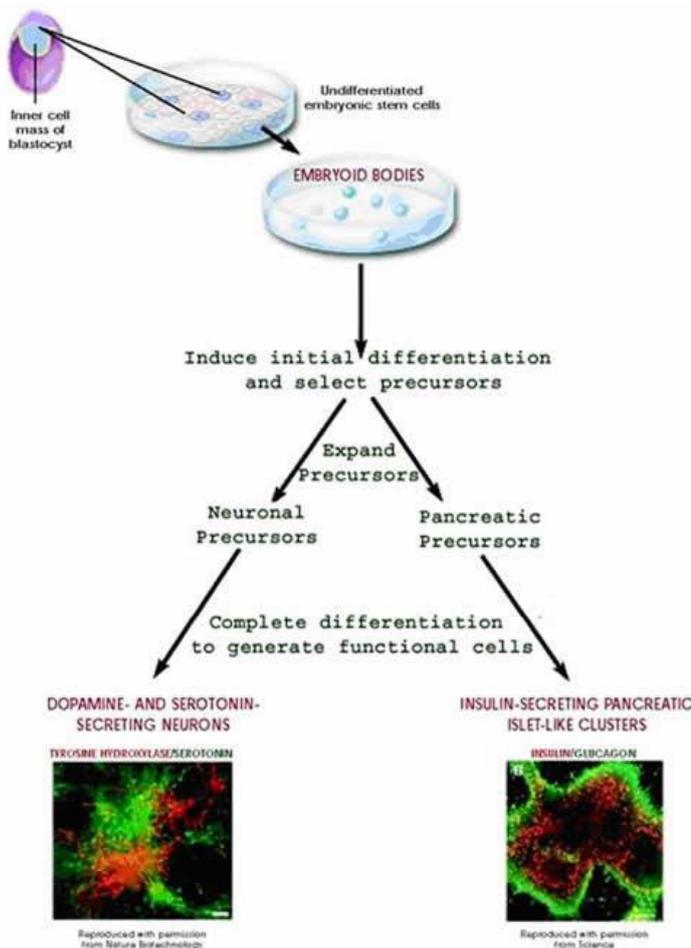


Figure 1. Directed differentiation of mouse embryonic stem cells. (© 2008 Terese Winslow)

If scientists can reliably direct the differentiation of embryonic stem cells into specific cell types, they may be able to use the resulting, differentiated cells to treat certain diseases in the future. Diseases that might be treated by transplanting cells generated from human embryonic stem cells include diabetes, traumatic spinal cord injury, Duchenne's muscular dystrophy, heart disease, and vision and hearing loss.

What are 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.

Where are adult stem cells found, and what do they normally do?

Adult stem cells have been identified in many organs and tissues, including brain, bone marrow, peripheral blood, blood vessels, skeletal muscle, skin, teeth, heart, gut, liver, ovarian epithelium, and testis. They are thought to reside in a specific area of each tissue (called a "stem cell niche"). In many tissues, current evidence suggests that some types of stem cells are pericytes, cells that compose the outermost layer of small blood vessels. Stem cells may remain quiescent (non-dividing) for long periods of time until they are activated by a normal need for more cells to maintain tissues, or by disease or tissue injury.

Typically, there is a very small number of stem cells in each tissue and, once removed from the body, their capacity to divide is limited, making generation of large quantities of stem cells difficult. Scientists in many laboratories are trying to find better ways to grow large quantities of adult stem cells in cell culture and to manipulate them to generate specific cell types so they can be used to treat injury or disease. Some examples of potential treatments include regenerating bone using cells derived from bone marrow stroma, developing insulin-producing cells for type 1 diabetes, and repairing damaged heart muscle following a heart attack with cardiac muscle cells.

What tests are used to identify adult stem cells?

Scientists often use one or more of the following methods to identify adult stem cells: (1) label the cells in a living tissue with molecular markers and then determine the specialized cell types they generate; (2) remove the cells from a living animal, label them in cell culture, and transplant them back into another animal to determine whether the cells replace (or "repopulate") their tissue of origin.

Importantly, scientists must demonstrate that a single adult stem cell can generate a line of genetically identical cells that then gives rise to all the appropriate differentiated cell types of the tissue. To confirm experimentally that a putative adult stem cell is indeed a stem cell, scientists tend to show either that the cell can give rise to these genetically identical cells in culture, and/or that a purified population of these candidate stem cells can repopulate or reform the tissue after transplant into an animal.

What is known about adult stem cell differentiation?

Normal differentiation pathways of adult stem cells. In a living animal, adult stem cells are available to divide for a long period, when needed, and can give rise to mature cell types that have characteristic shapes and specialized structures and functions of a particular tissue. The following are examples of differentiation pathways of adult stem cells (Figure 2) that have been demonstrated *in vitro* or *in vivo*.

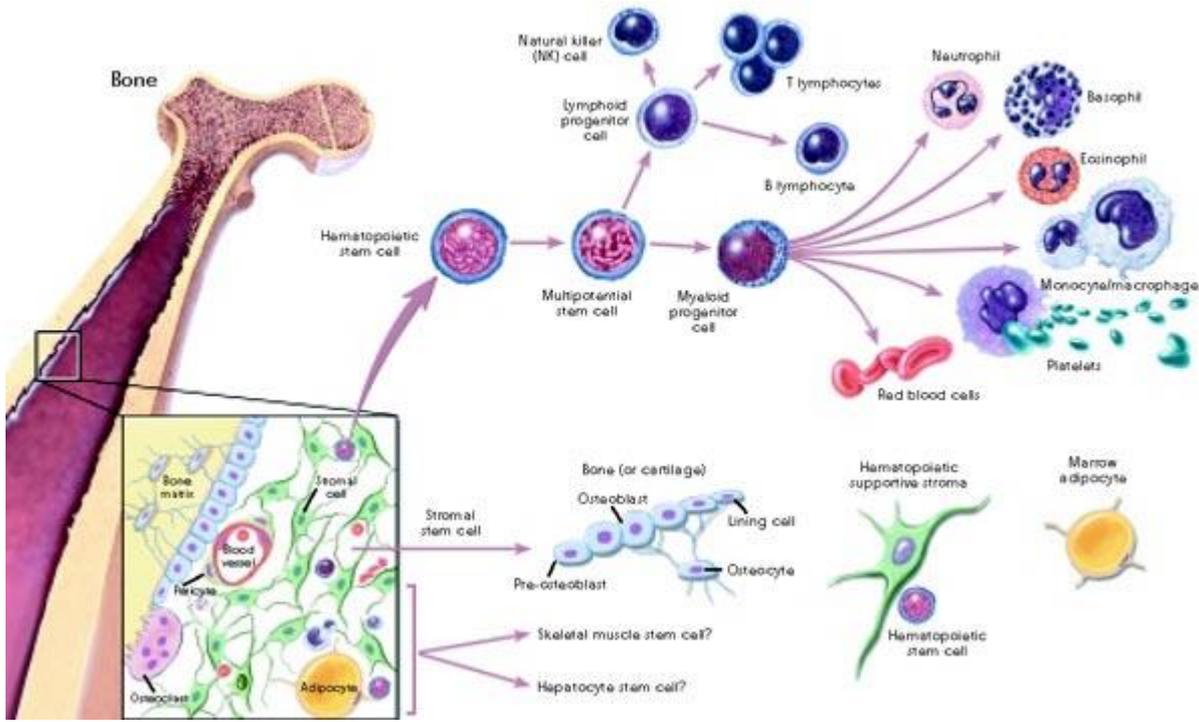


Figure 2. Hematopoietic and stromal stem cell differentiation. Click here for larger image. (© 2008 Terese Winslow)

- **Hematopoietic stem cells** give rise to all the types of blood cells: red blood cells, B lymphocytes, T lymphocytes, natural killer cells, neutrophils, basophils, eosinophils, monocytes, and macrophages.
- **Mesenchymal stem cells** have been reported to be present in many tissues. Those from bone marrow (bone marrow stromal stem cells, skeletal stem cells) give rise to a variety of cell types: bone cells (osteoblasts and osteocytes), cartilage cells (chondrocytes), fat cells (adipocytes), and stromal cells that support blood formation..
- **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.

Transdifferentiation. A number of experiments have reported that certain adult stem cell types can differentiate into cell types seen in organs or tissues other than those expected from the cells' predicted lineage (i.e., brain stem cells that differentiate into blood cells or blood-forming cells that differentiate into cardiac muscle cells, and so forth). This reported phenomenon is called transdifferentiation.

Although isolated instances of transdifferentiation have been observed in some vertebrate species, whether this phenomenon actually occurs in humans is under debate by the scientific community. Instead of transdifferentiation, the observed instances may involve fusion of a donor cell with a recipient cell. Another possibility is that transplanted stem cells are secreting factors that encourage the recipient's own stem cells to begin the repair process. Even when transdifferentiation has been detected, only a very small percentage of cells undergo the process.

What are 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.

What are the differences between embryonic and adult stem cells?

One major difference between adult and embryonic stem cells is their different abilities in the number and type of differentiated cell types they can become. Embryonic stem cells can become all cell types of the body because they are pluripotent. Adult stem cells are thought to be limited to differentiating into different cell types of their tissue of origin.

Embryonic stem cells can be grown relatively easily in culture. Adult stem cells are rare in mature tissues, so isolating these cells from an adult tissue is challenging, and methods to expand their numbers in cell culture have not yet been worked out. This is an important distinction, as large numbers of cells are needed for stem cell replacement therapies.

Scientists believe that tissues derived from embryonic and adult stem cells may differ in the likelihood of being rejected after transplantation. We don't yet know for certain whether tissues derived from embryonic stem cells would cause transplant rejection.

Embryonic	Adult
Most potential	Organ specific
Can form all cell types	Can form few cell types
Immortal in culture	Limited life span
Plentiful	Hard to isolate
Pluripotent cells	Multipotent / oligopotent cells