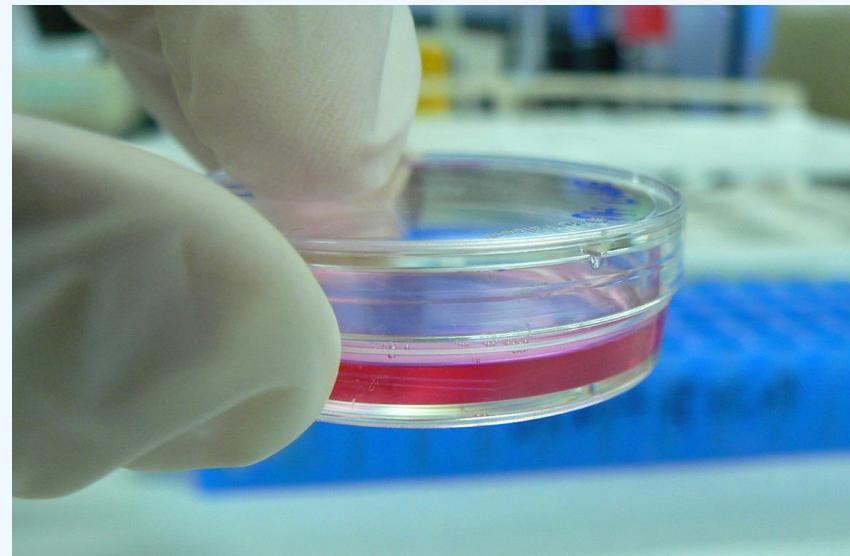


# Lec4 \ Cell Culture Techniques

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## cell line

Cell culture is the process by which cells are grown under controlled conditions, generally outside their natural environment



# Cell line

Most cells require a **surface or an artificial substrate** (adherent or **monolayer** culture), whereas others can be grown free **floating** in culture medium (**suspension** culture). The lifespan of most cells is genetically determined, but some cell culturing cells have been “transformed” into immortal cells which will reproduce indefinitely if the optimal conditions are provided.

## Cell strains:

## سلالات الخلايا

**Cell strains** are cells that have been adapted to culture but, **unlike** cell lines, have a finite division potential. Non-immortalized cells **stop dividing** after 40 to 60 population doublings and, after this, they lose their ability to proliferate, which is a genetically determined event known as **senescence**.

A cell strain is derived either from a **primary culture** or **subpopulation** of a cell line by the selection or cloning of cells having **specific properties** or characteristics which must be defined. A cell strain often acquires **additional genetic changes** subsequent to the initiation of the parent line.

## Finite and continuous cell line :

Normal cells usually divide only a limited number of times before losing their ability to proliferate and reach senescence (aging); these cell lines are **known as finite**. However, some cell lines become **immortal** through a process called **transformation**, which can occur spontaneously or can be **chemically or virally induced**. When a finite cell line undergoes transformation and acquires the ability to divide indefinitely, it becomes a **continuous cell line**.

## Finite and continuous cell line :

Attempts have been made to culture almost every tissue, including neuronal cells, bone, cartilage and hair cells. However, **human fibroblasts are easier** to culture than epithelial cells. Different epithelial cells show different responses to culture conditions.

### Reasons for failure or difficulty in epithelial tissue culture سبب فشل او صعوبة زراعة الانسجة الظهارية

Despite advances in culturing techniques, **human epithelial cells could not be maintained** in culture for long time periods. The problem is the tendency of human cells to undergo senescence after a certain cell division.

## There are two types of Cell Line or Cell Strain:

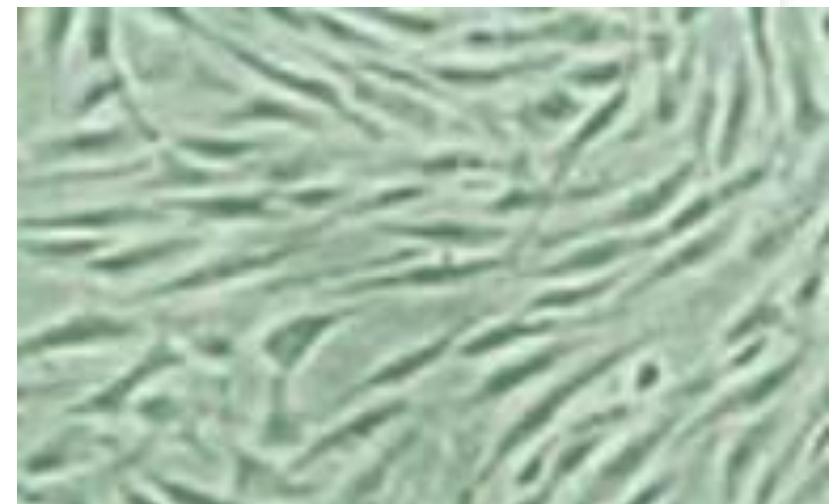
Finite cell Lines	Continuous Cell Lines
<ul style="list-style-type: none"><li>• Have a limited life span</li></ul>	<ul style="list-style-type: none"><li>• Have unlimited life span, Exhibit heterogeneity</li></ul>
<ul style="list-style-type: none"><li>• They grow in monolayer form</li></ul>	<ul style="list-style-type: none"><li>• They grow in monolayer or suspension form</li></ul>
<ul style="list-style-type: none"><li>• Exhibit the property of contact inhibition</li></ul>	<ul style="list-style-type: none"><li>• Absence of contact inhibition</li></ul>
<ul style="list-style-type: none"><li>• The growth rate is slow</li></ul>	<ul style="list-style-type: none"><li>• The growth rate is rapid</li></ul>
<ul style="list-style-type: none"><li>• The doubling time is around 24-96 hours</li></ul>	<ul style="list-style-type: none"><li>• The doubling time is 12-24 hours</li></ul>

## Morphology of Cells in culture:

Cells in culture can be divided into **three** basic categories based on their shape and appearance (morphology).

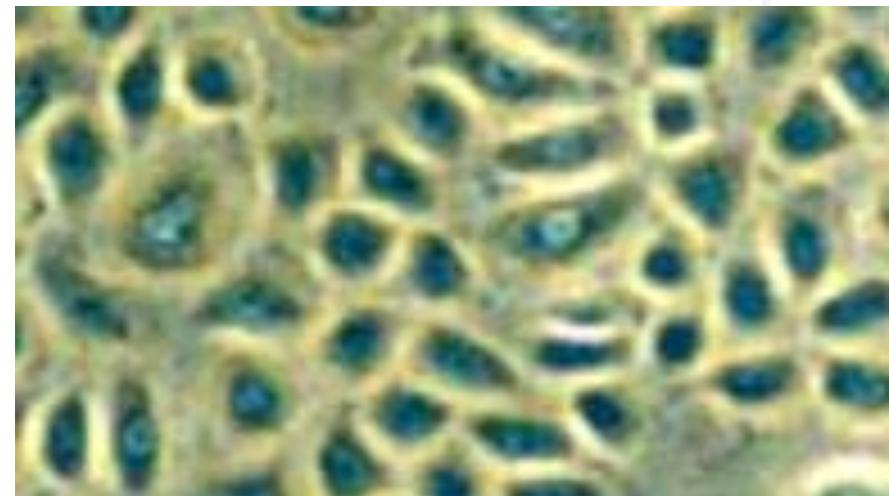
# Morphology of Cells in culture:

1. Fibroblastic (or fibroblast-like) cells: they are bipolar or **multipolar**, have **elongated shapes**, and grow **attached** to a substrate.



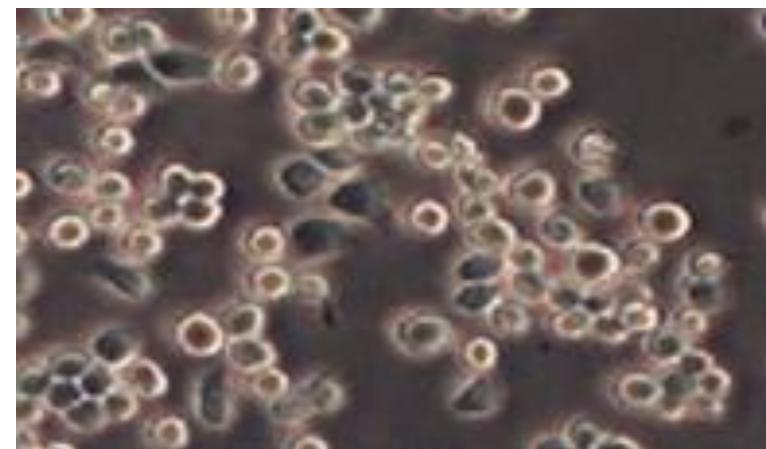
# Morphology of Cells in culture:

2. Epithelial-like cells: they are polygonal in shape with more **regular dimensions**, and grow **attached** to a substrate in discrete patches.



# Morphology of Cells in culture:

3. Lymphoblast-like cells: they are **spherical** in shape and usually grown in **suspension** without attaching to a surface.



## cell culture in two and three dimensional :

Research in tissue engineering, stem cells and molecular biology primarily involves cultures of cells on **flat plastic dishes**. **This technique is known as two-dimensional (2D) cell culture.**

From the advance of polymer technology, today's standard plastic dish (commonly known as the Petri dish) had arose for the 2D cell culture. However, various researchers today also utilize laboratory **culture flasks** and **conicals**.

## **cell culture in two and three dimensional :**

In addition, cell culture in three dimensions (3D) has been described as "Biology's New Dimension". Currently, there is an **increase in the use of 3D cell cultures in research areas, including drug discovery, cancer biology, regenerative medicine, nanomaterials assessment and basic life science research.**



Petri Dishes



Different sizes of cell culture flasks

## cell culture in two and three dimensional :

The 3D cell culture is **unlike** 2D cell culture environments; it is an artificially created environment in which cells are permitted to grow or interact with their surroundings *in vitro* in all three dimensions, similar to how they would *in vivo*.

The **3D cultures** are usually grown in **bioreactors**, small capsules in which the cells can grow into **spheroids**, or 3D cell colonies. Approximately 300 spheroids are usually cultured per bioreactor.



Samples of Bioreactors



Sample of Bioreactor