

# **TOPIC : APPLIED ASPECTS OF IVF, STEM CELLS AND CLONING**

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SUBJECT : ZOOLOGY  
COURSE : BSC. (P) LIFE SCIENCE 1<sup>st</sup> Year  
TOPIC : APPLIED ASPECTS OF IVF, STEM CELLS AND CLONING

Room No. : A-210, A-214, A-114

# IN VITRO FERTILIZATION

- ▶ **IVF broadly deals with the removal of eggs from a women, fertilizing them in the laboratory and then transferring the fertilized eggs into uterus a few days later.**
- ▶ **Infertility due to the following causes may be considered for IVF**
  - ▶ **Failed ovulation induction**
  - ▶ **Tubal diseases**
  - ▶ **Endometriosis**
  - ▶ **Idiopathic infertility**

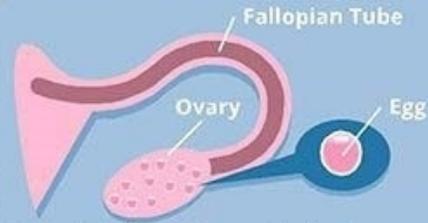
# How does IVF Work?

## 1 Consultation with a CRGH doctor



Before your treatment starts, you will discuss everything with your doctor

## 2 Blood tests and scans



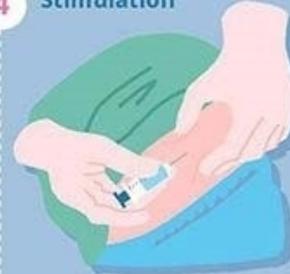
We need to look at your hormone levels and your ovarian follicles to decide a treatment plan that is right for you. Other investigations such as 3D SIS, 3D hycosy and endometrial biopsy may also be done

## 3 Semen analysis



IVF requires good quality sperm. The analysis looks at sperm morphology (shape) and motility (movement) as well as quantity

## 4 Stimulation



Injections will stimulate your body to develop multiple eggs

## 5 Monitoring



Scans and blood tests allow us to keep a close eye on your body's reaction to the drugs

## 6 Trigger injection



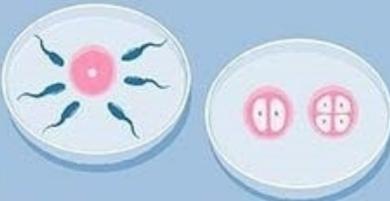
When hormones are at the right levels, another injection will trigger the follicles to release eggs

## 7 Egg collection and sperm collection



Eggs are collected via a needle passed through the vagina whilst you are under sedation. Sperm is collected manually or surgically

## 8 Fertilisation and embryo development



Eggs and sperm are placed in a petri dish in an incubator to fertilise. Embryologists keep a close eye on the embryos as they continue to develop

## 9 Embryo transfer



The best embryo(s) is/are transferred back into the uterus. Any viable embryos not transferred will be frozen for later use

## 10 Pregnancy test



Patients take a pregnancy test 16 days after embryo transfer. Whatever the outcome, CRGH will arrange further appointments and support for patients at this stage

# IVF

## ADVANTAGES

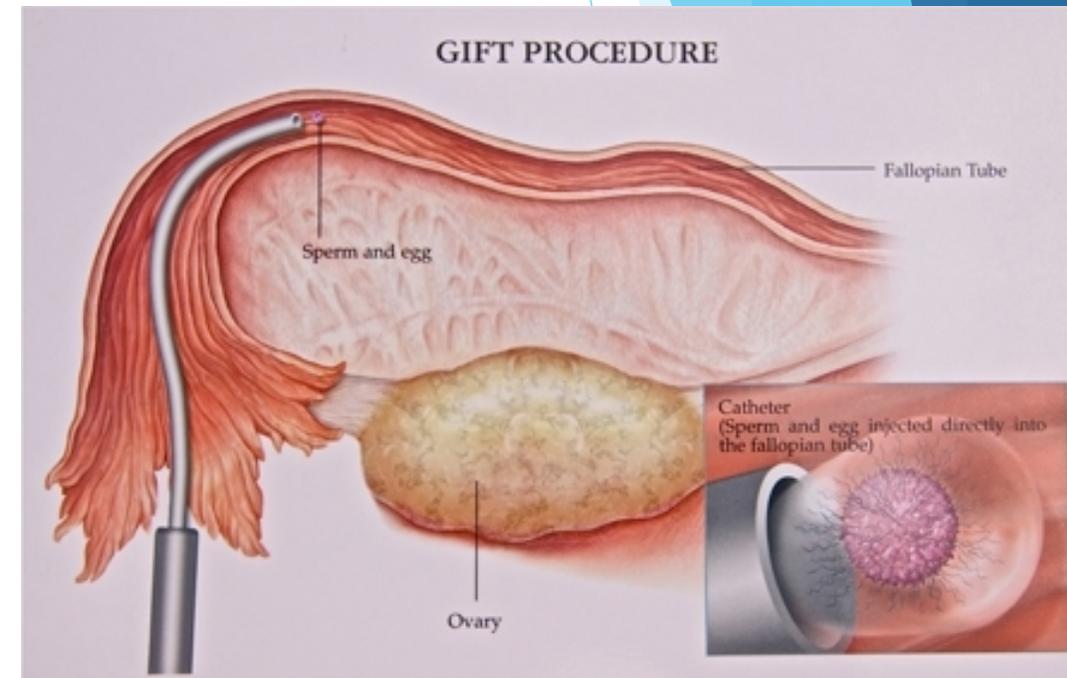
- ▶ **Fertilization is confirmed before implantation can occur.**
- ▶ **Gives women with damaged oviducts, the opportunity to carry their own foetus.**

## DISADVANTAGES

- ▶ **Implantation in the uterus does not always occur**
- ▶ **Higher risk of twins or triplets, which also increases the risk of complications and miscarriages.**
- ▶ **Side effects associated with the fertility medication**
- ▶ **Risk of ectopic pregnancy**

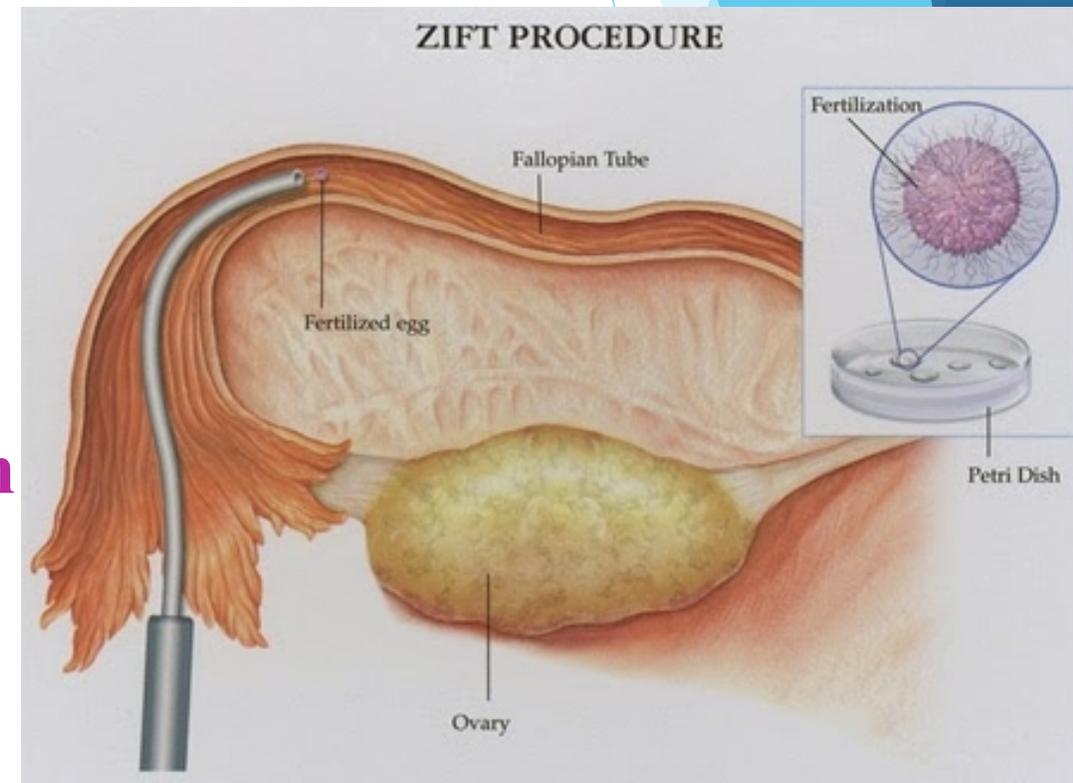
# GAMETE INTRA FALLOPIAN TRANSFER (GIFT)

- ▶ It involves the transfer of both sperm and unfertilized oocyte into the fallopian tube.
- ▶ This allows the fertilisation to naturally occur in vivo.
- ▶ Two oocyte along with 2-5 lakhs motile sperms are placed in plastic tube container.
- ▶ Then oocyte sperm combination is injected 4cm into distal end of fallopian tube.



# ZYGOTE INTRA FALLOPIAN TRANSFER (ZIFT)

- ▶ ZIFT combines aspects of both IVF and GIFT.
- ▶ Fertilization takes place outside the uterus and placed into the fallopian tubes.
- ▶ Protocols for ovarian stimulation are similar to those used for IVF and GIFT.
- ▶ Eggs are collected and fertilized by the partner's sperm in the laboratory.
- ▶ The zygote is transferred to the fallopian tube within 24 hours, when it is at 1 cell stage.

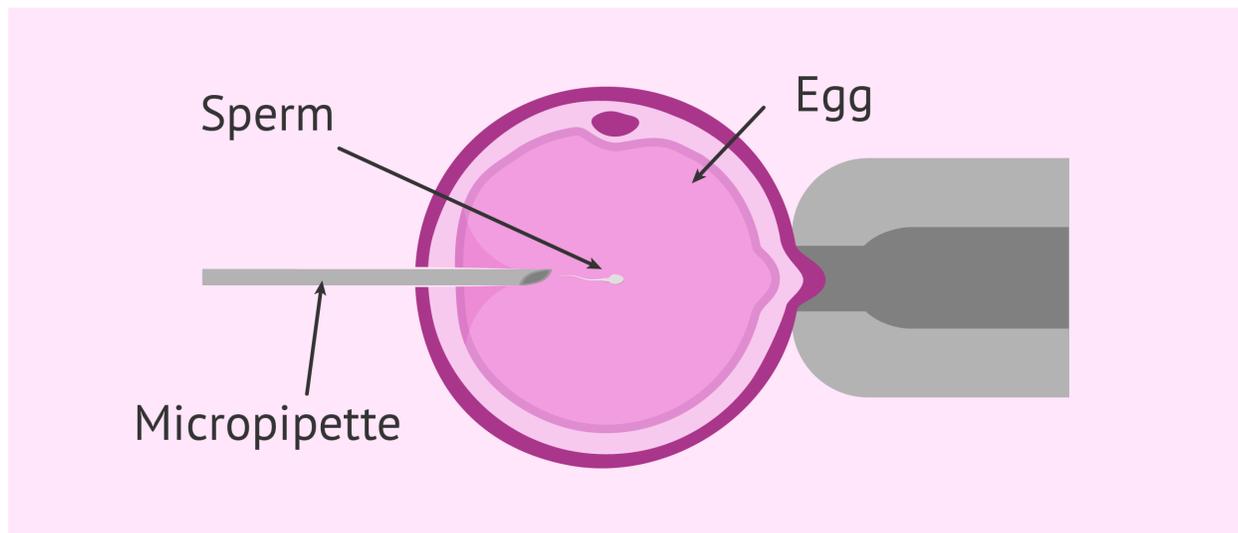


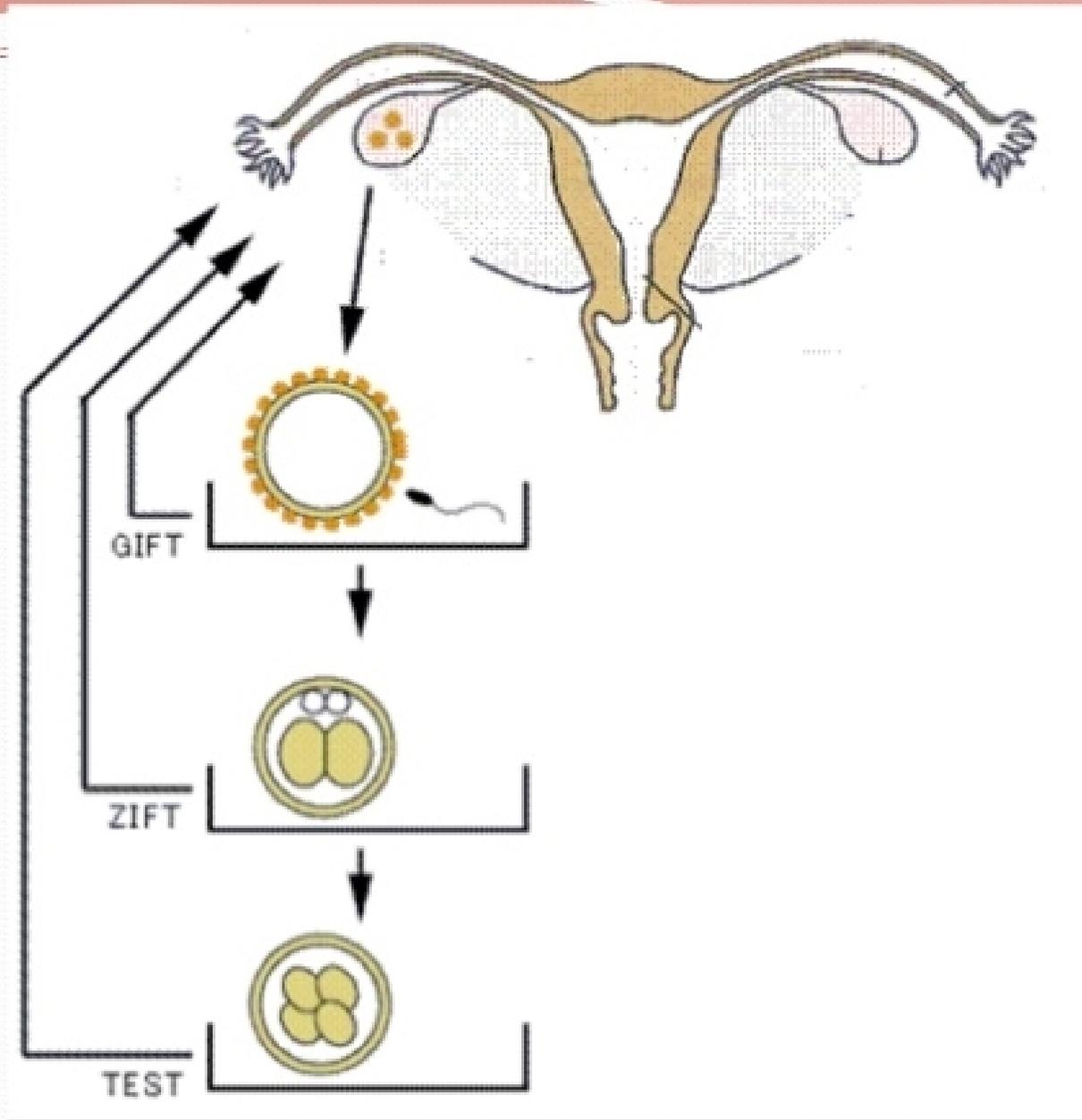
# TUBAL EMBRYO STAGE TRANSFER (TET)

- ▶ **It combines IVF with tubal transfer**
- ▶ **Embryos are placed into the women's fallopian tube.**
- ▶ **The embryos are transferred back into the woman 2 days after fertilisation. This is at the '2 cell or 4 cell' stage.**
- ▶ **TET allows embryos to make their way to the uterus for implantation.**
- ▶ **Its advantage over ZIFT is that it allows for the assessment of fertilisation and embryo quality.**
- ▶ **Success rate higher than ZIFT.**

# INTRA CYTOPLASMIC SPERM INJECTION (ICSI)

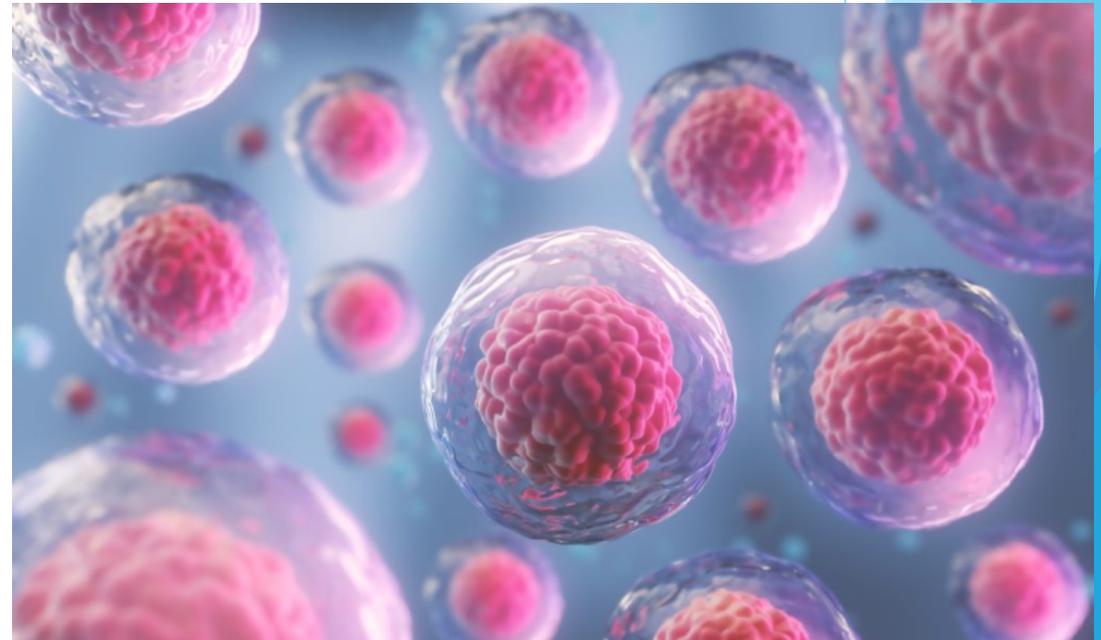
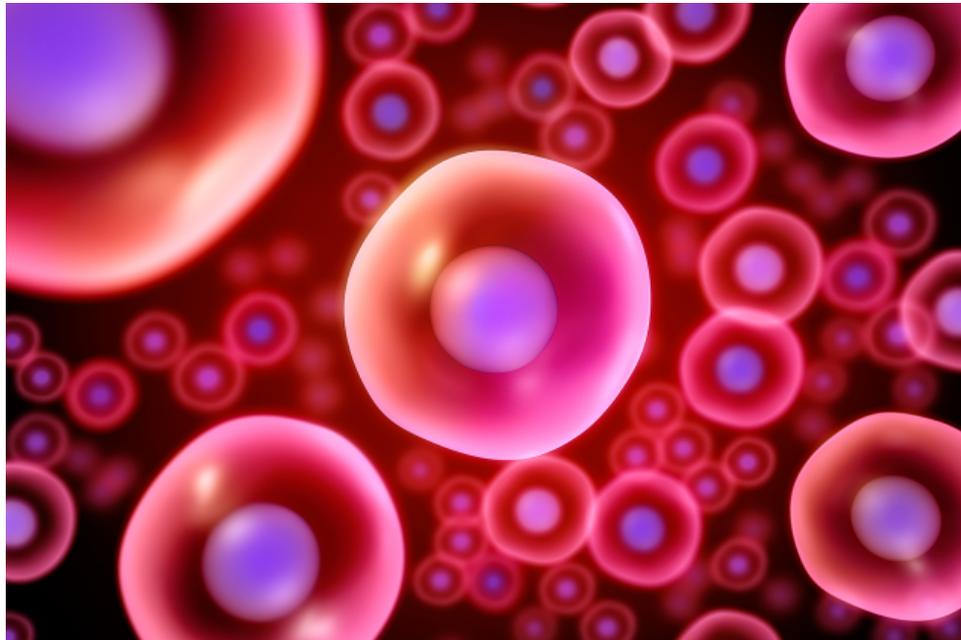
- ▶ Sperm is injected directly into the eggs in a laboratory.
- ▶ Used if infertility originates from the male such as :
  - ▶ Low numbers of sperm
  - ▶ Low sperm motility
- ▶ Single spermatozoan is directly injected into the cytoplasm of oocyte through the micropuncture of zona pellucida

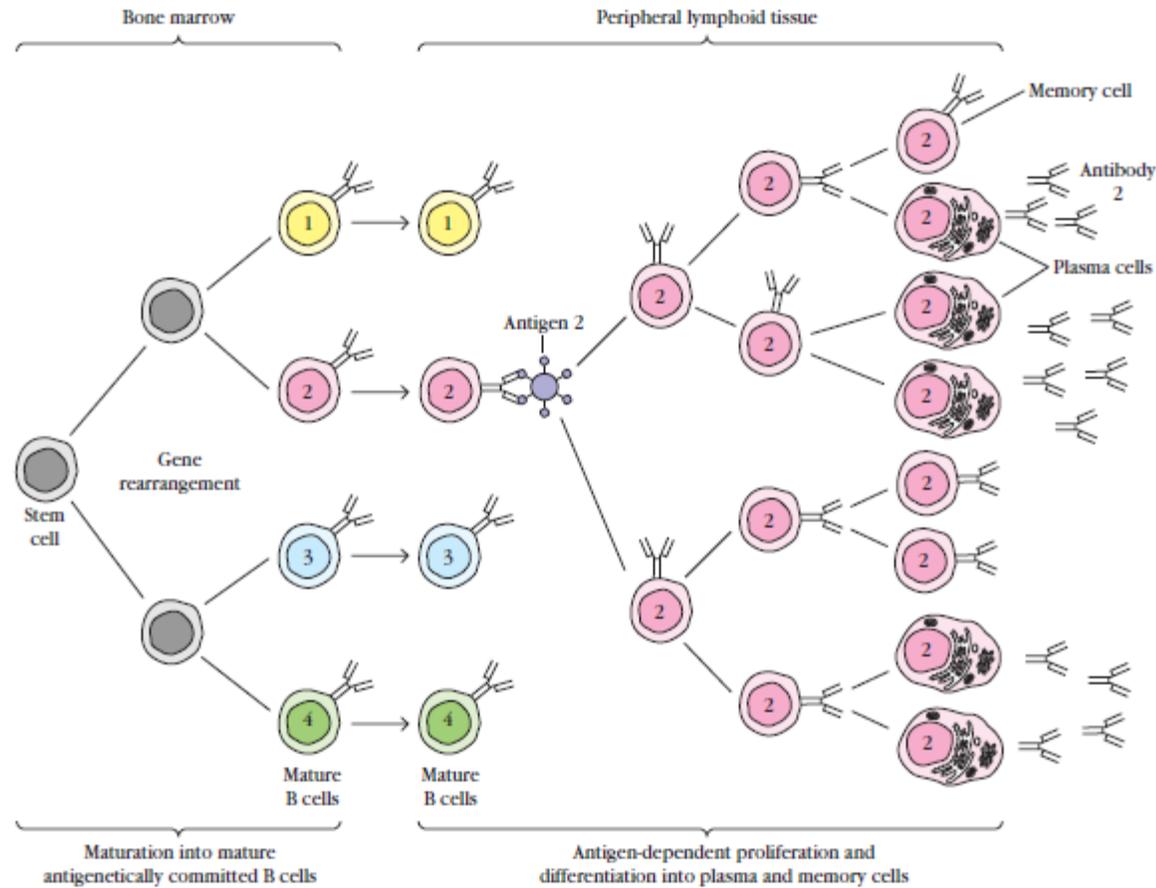




# STEM CELLS

- ▶ **Stem cells are one of the most fascinating areas of biology today.**
- ▶ **Stem cells are a special kind of cell that have the ability to divide indefinitely and have the potential to give rise to specialised cells that is any cell of the body.**



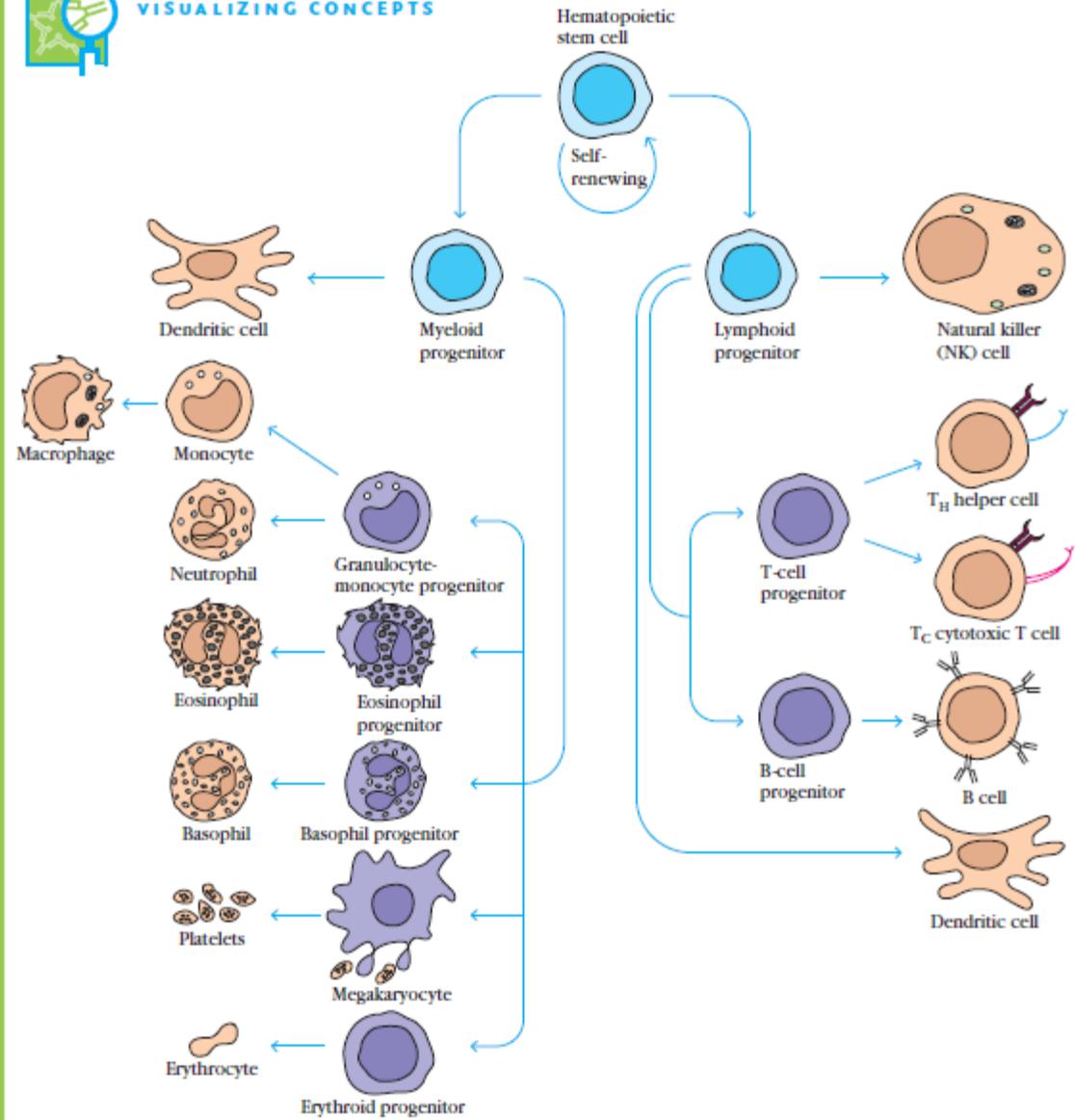


**FIGURE 1-10** Maturation and clonal selection of B lymphocytes. Maturation, which occurs in the absence of antigen, produces antigenically committed B cells, each of which expresses antibody with a single antigenic specificity (indicated by 1, 2, 3, and 4). Clonal selection occurs when an antigen binds to a B cell whose membrane-bound antibody molecules are specific for epitopes on that antigen. Clonal expansion of an antigen-activated B cell (number 2 in this ex-

ample) leads to a clone of memory B cells and effector B cells, called plasma cells; all cells in the expanded clone are specific for the original antigen. The plasma cells secrete antibody reactive with the activating antigen. Similar processes take place in the T-lymphocyte population, resulting in clones of memory T cells and effector T cells; the latter include activated  $T_H$  cells, which secrete cytokines, and cytotoxic T lymphocytes (CTLs).



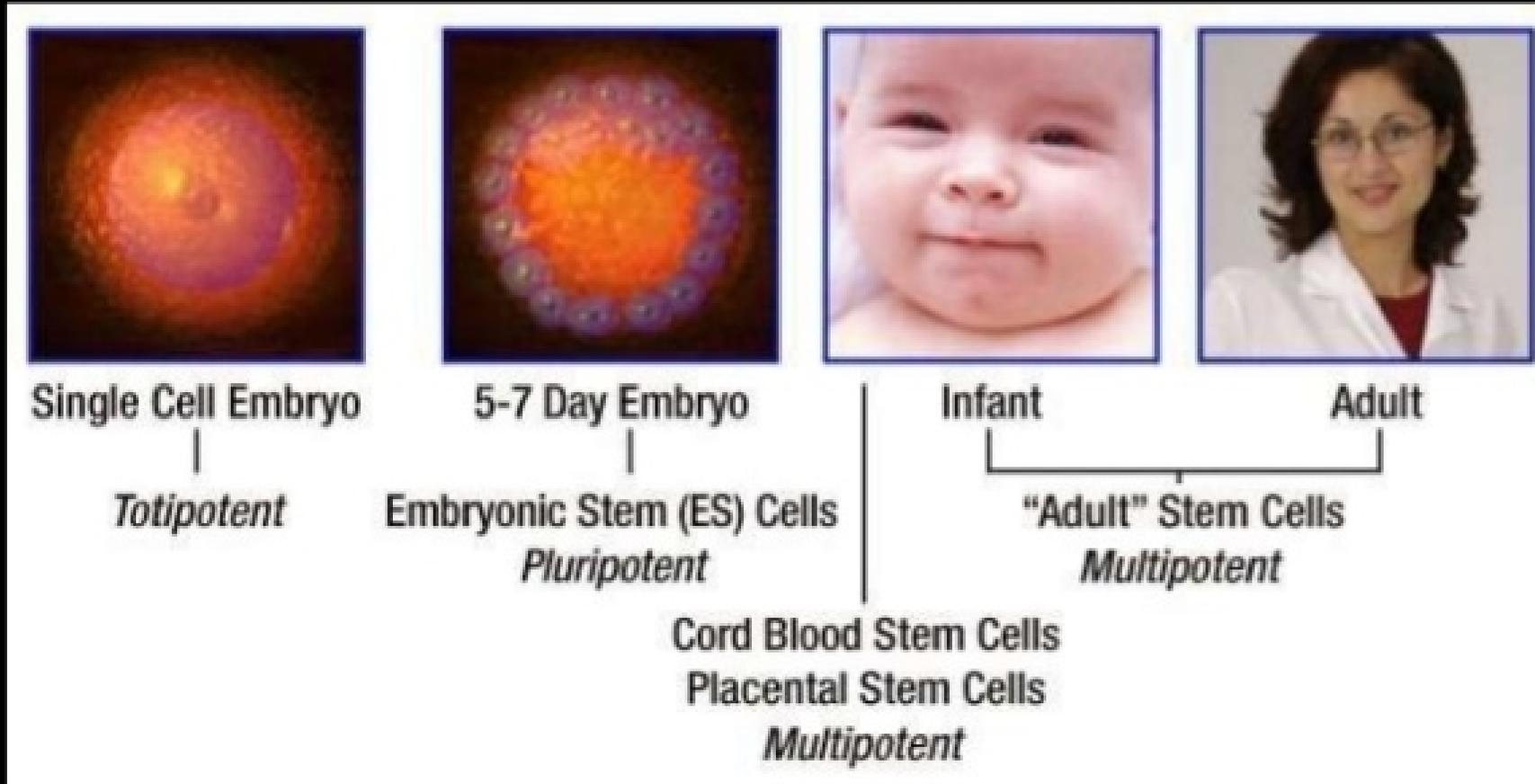
VISUALIZING CONCEPTS



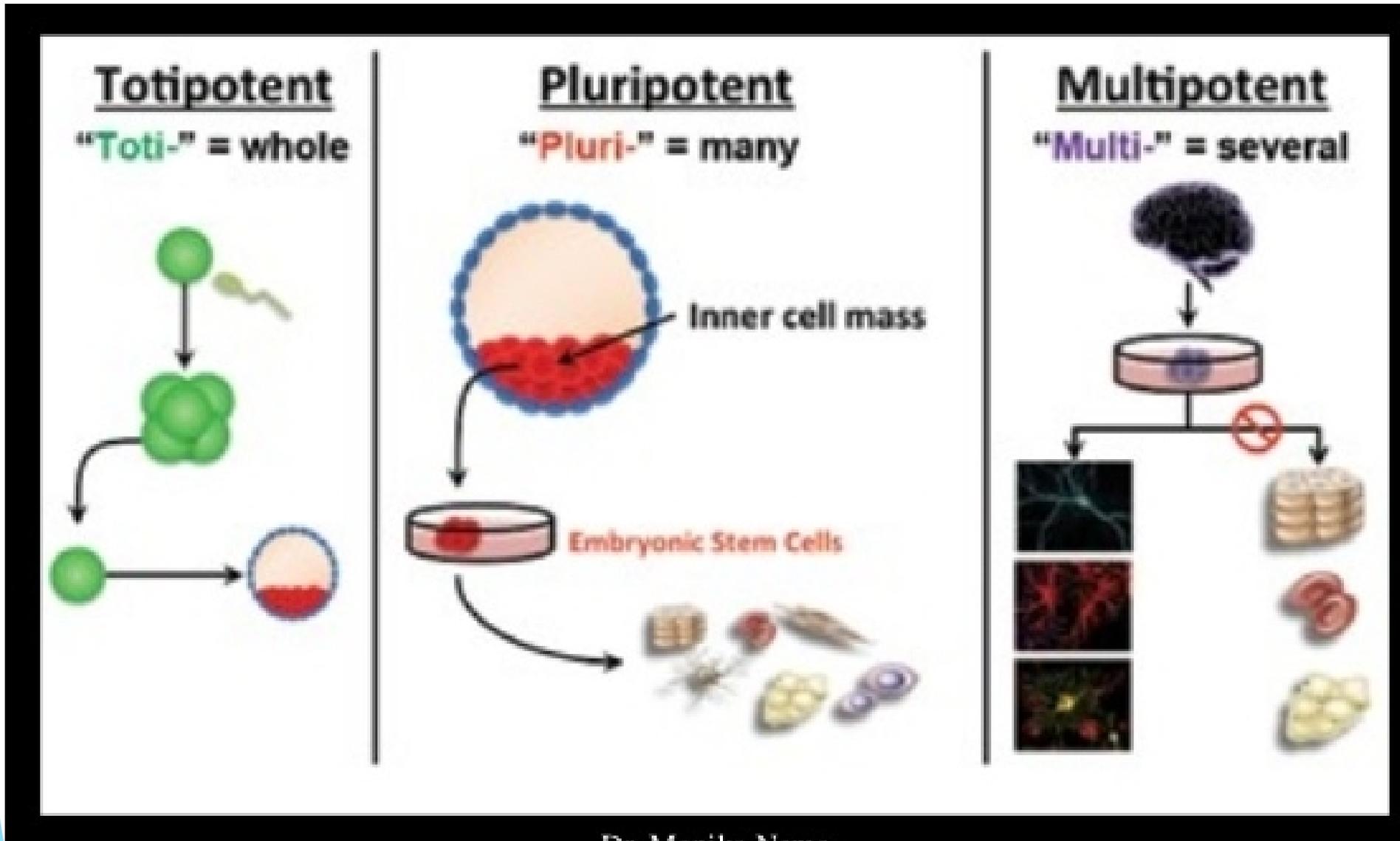
# STEM CELL CHARACTERISTICS

- ▶ ***BLANK CELLS*** : These are unspecialized cells that is they don't have any tissue specific structures that allow it to perform specialized function.
- ▶ ***PROLIFERATION*** : They are capable of dividing and renewing themselves for indefinite periods.
- ▶ ***DIFFERENTIATION*** : They have the potential to give rise to specialised cell types.
- ▶ ***PLASTICITY*** : Stem cell from one tissue may be able to give rise to cell types of completely different tissue , a phenomenon known as plasticity. Ex. Blood cells becoming neuron

# Stem cell timeline



# TYPES OF STEM CELLS





fertilised egg

This cell  
Can form the  
Embryo and placenta



totipotent stem cells

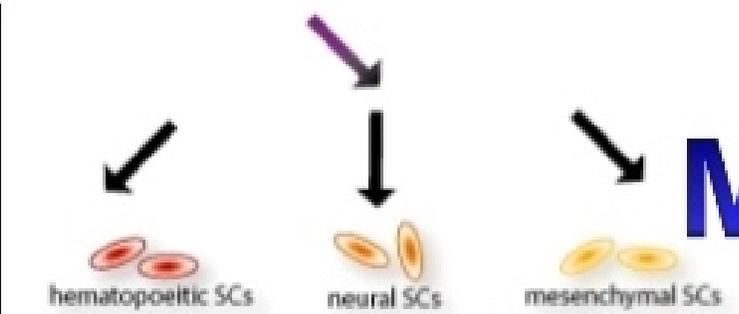
> **Totipotent**

This cell  
Can just form the  
embryo



blastocyst containing  
pluripotent stem cells

Pluripotent



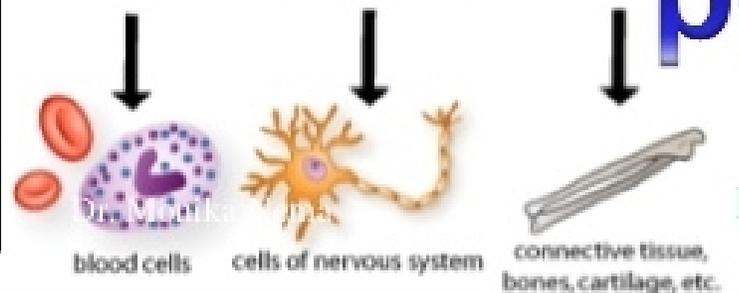
hematopoietic SCs

neural SCs

mesenchymal SCs

tissue-specific SCs

**Multi-  
potent**



blood cells

cells of nervous system

connective tissue,  
bones, cartilage, etc.

Fully mature

# TOTIPOTENT CELLS

- ▶ **These cells have potential to become any type of cell in the body or any cell of the extraembryonic membranes ex. Placenta**
- ▶ **The only totipotent cells are the fertilized egg and the first 4 or so cells produced by its cleavage (as shown by the ability of mammals to produce identical twins, triplets, etc.)**

# PLURIPOTENT CELLS

- ▶ **These are true stem cells with the potential to make any differentiated cell in the body (but probably not those of the placenta which is derived from the trophoblast)**
- ▶ **There are two types of them :**
  - ▶ **Embryo Stem cells : These are isolated from inner cell mass of the blastocyst.**
  - ▶ **Embryonic Germ cells : These are derived from the part of human embryo or foetus that will ultimately produce eggs or sperm (gametes)**

# MULTIPOTENT CELLS

- ▶ **These are the true stem cells but can only differentiate into a limited number of types.**
- ▶ **For ex. The bone marrow contains multipotent stem cells that give rise to all the cells of the blood but no other types of cells.**
- ▶ **These are found in adult animals : perhaps the most organs in the body (eg. Brain , liver) contain them where they can replace dead or damaged cells.**

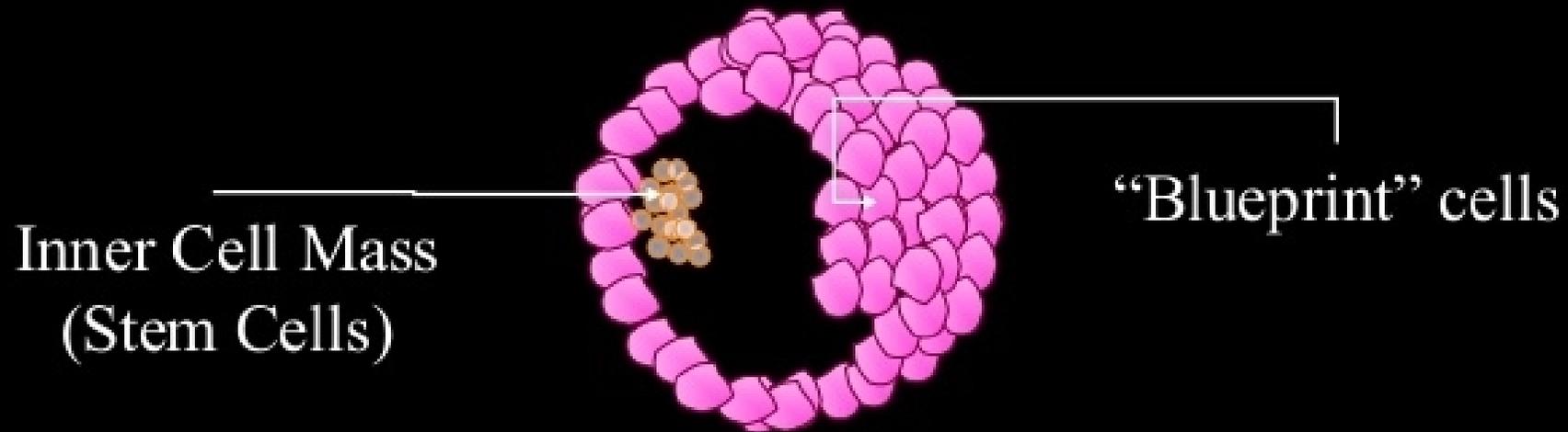
# USES OF STEM CELLS

- ▶ ***BASIC RESEARCH*** : It helps clarification of complex events that occur during human development and understanding molecular basis of cancer.
- ▶ ***BIOTECHNOLOGY*** : Stem cells can provide specific cell types to safety test of new drugs and screening of potential drugs.
- ▶ ***CELL BASED THERAPIES*** : *Regenerative therapy to Parkinson's , Alzheimer's , ALS , spinal cord injury , stroke, severe burns , heart disease , diabetes , osteoarthritis and Rheumatoid arthritis.*

**How are Embryonic  
Stem cells harvested ?**

# A primer on Human Embryonic Stem Cells

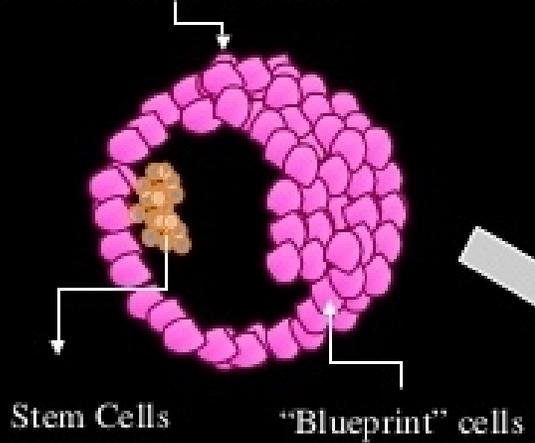
## Blastocyst - from In Vitro Fertilization Clinic



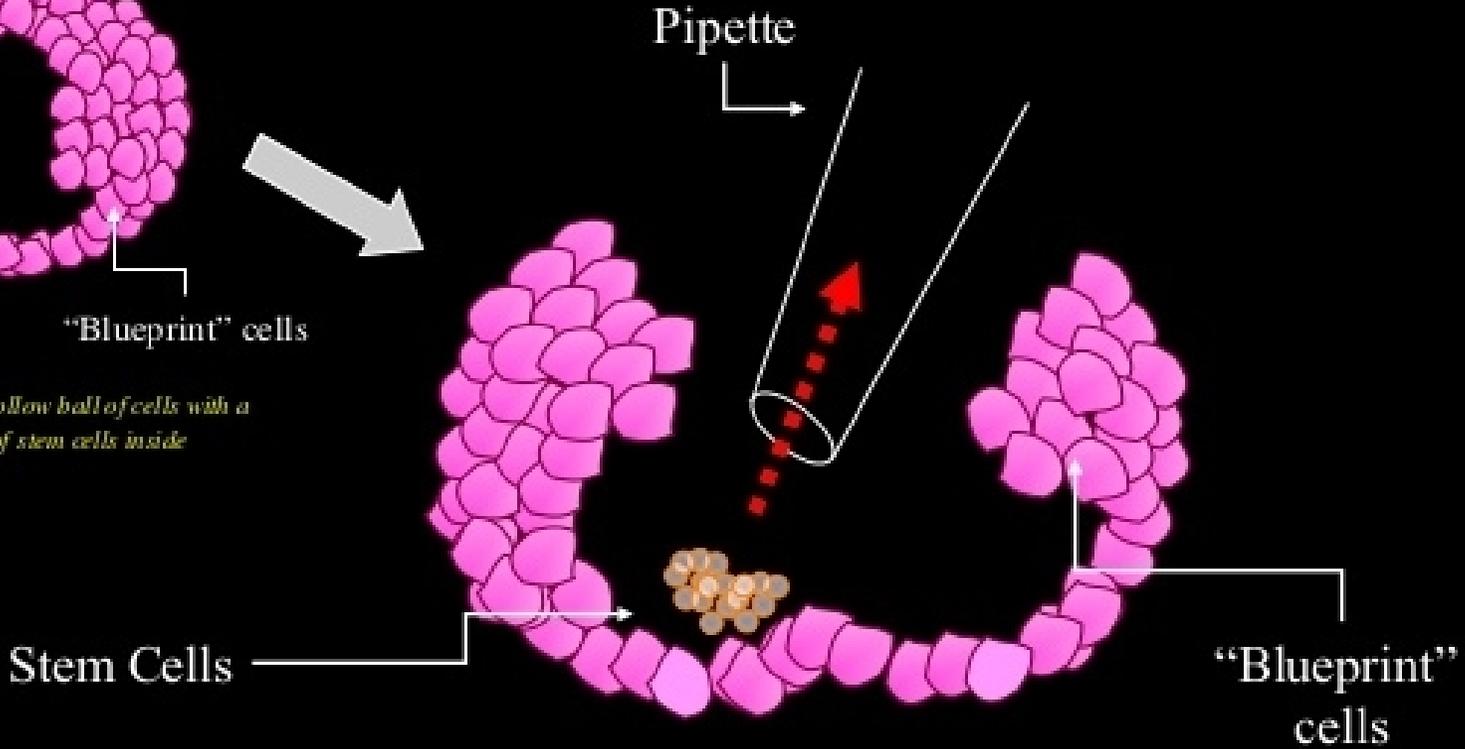
A Blastocyst is a hollow ball of cells with a small clump of stem cells inside

# Human Embryonic Stem Cells

Blastocyst -  
from In Vitro Fertilization Clinic



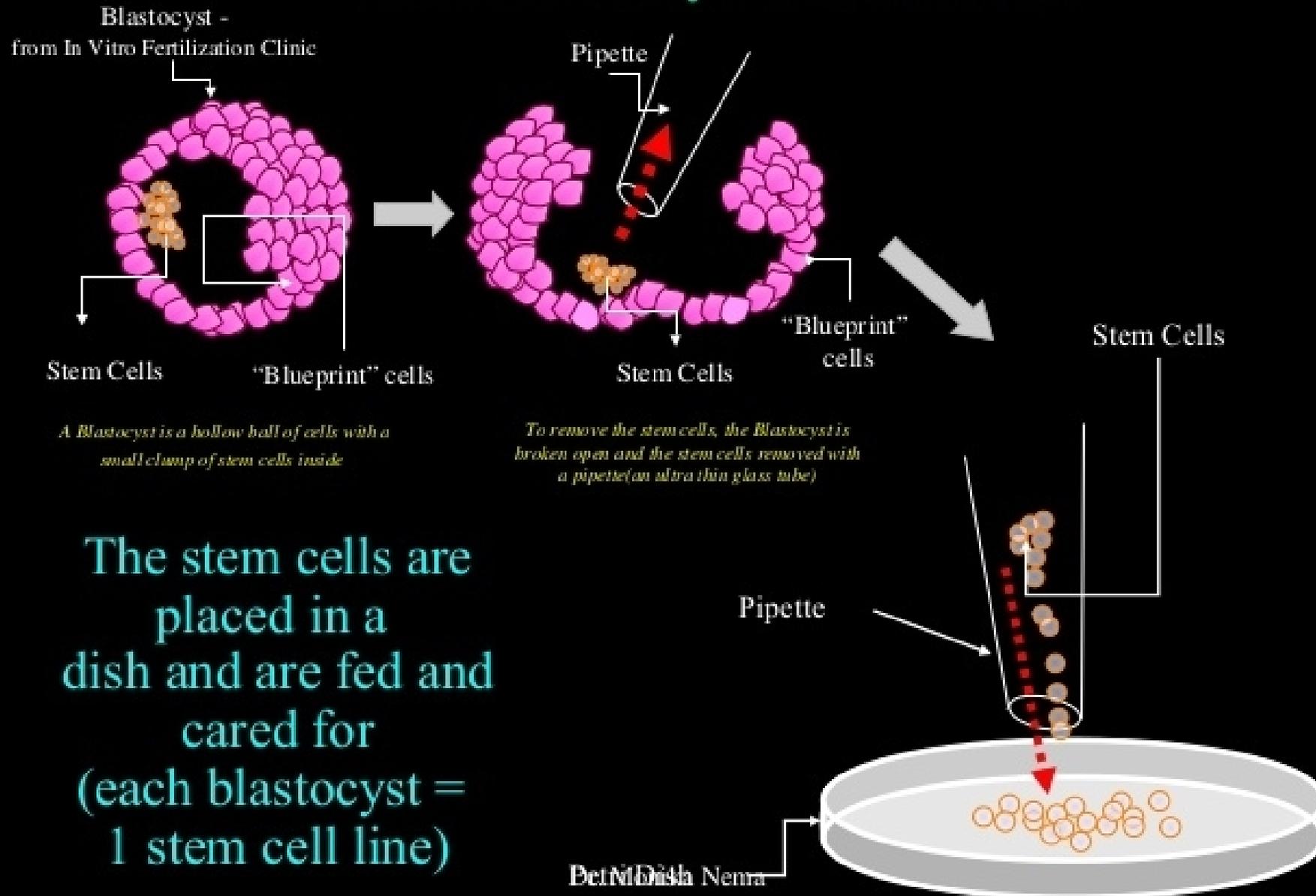
*A Blastocyst is a hollow ball of cells with a small clump of stem cells inside*



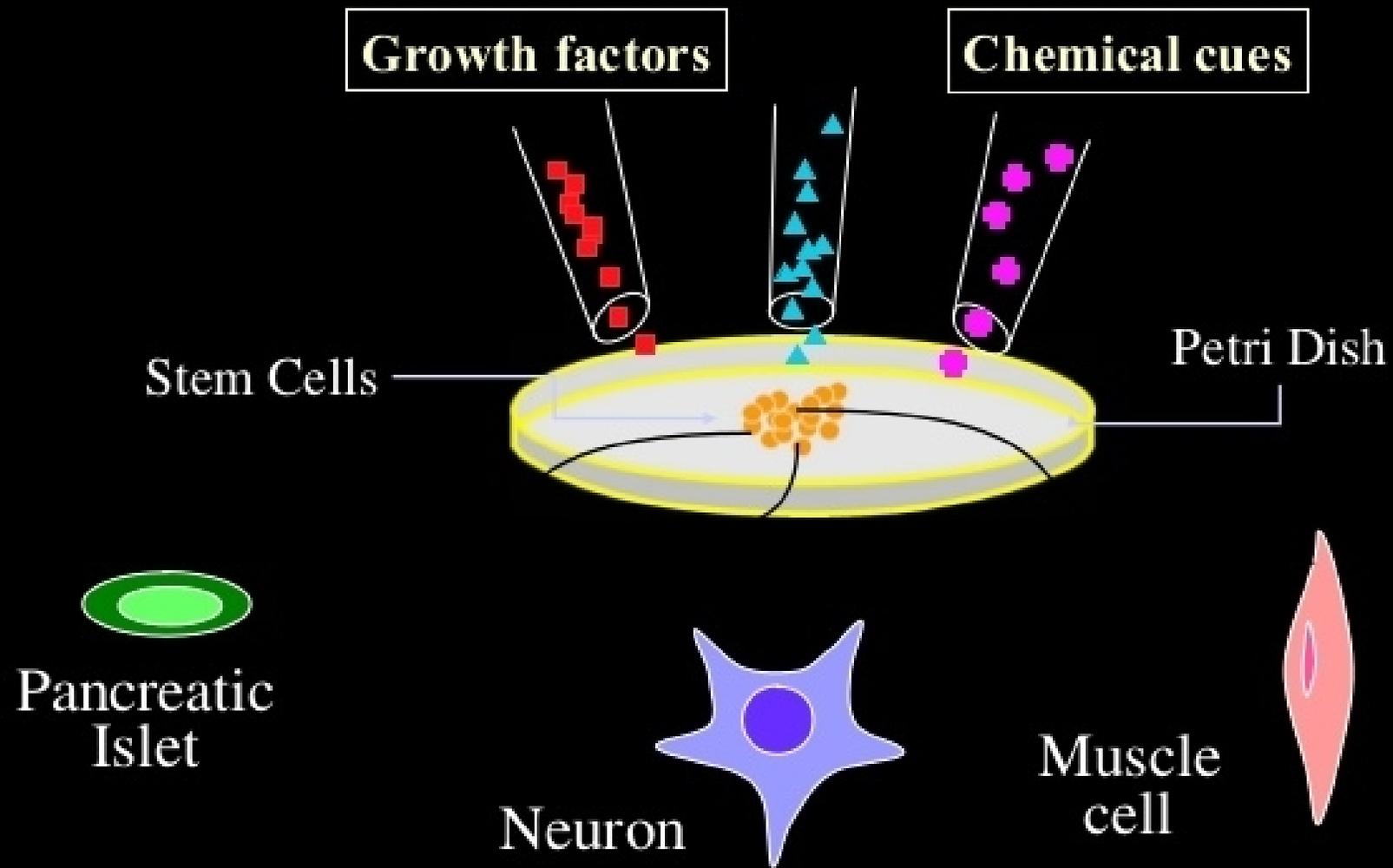
To remove the stem cells, the Blastocyst is opened and the stem cells removed with a pipette

Dr. Monika Nema

# Human Embryonic Stem Cells

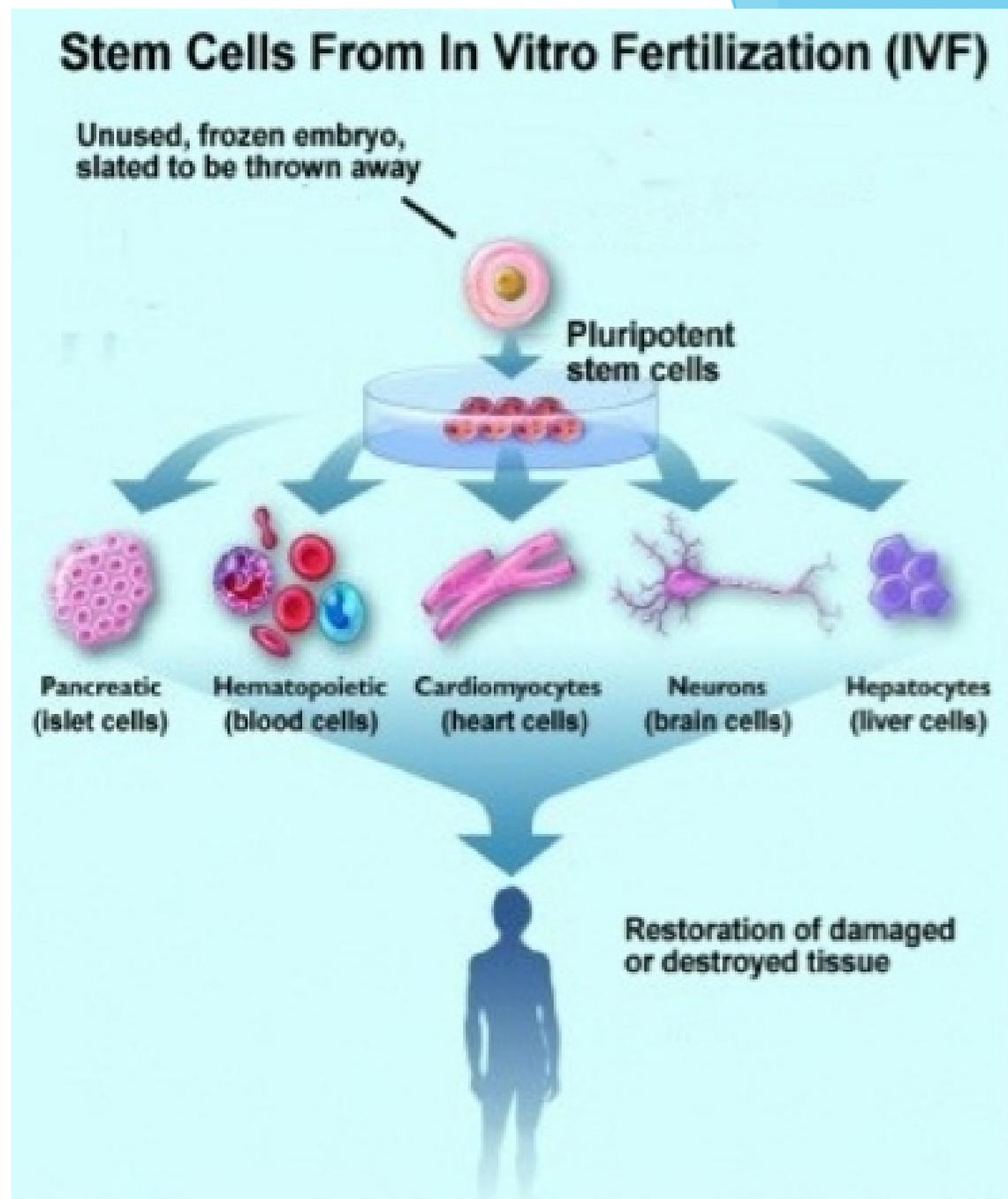


The stem cells are placed in a dish and are fed and cared for (each blastocyst = 1 stem cell line)



*Different chemicals / molecules are added to the stem cells to make them become specific types of cells.*

- ▶ Tens of thousand of frozen embryos are routinely destroyed when couples finish their treatment.
- ▶ Theses surplus embryos can be used to produce stem cells.
- ▶ Regenerative medical research aims to develop these cells into new , healthy tissue to heal severe illness.



# CLONING

- ▶ Cloning describes a number of different processes that can be used to produce genetically identical copies of a biological entity.
- ▶ Clones are organisms that are exact genetic copies. Every single bit of their DNA is identical.
- ▶ Human identical twins are clones of each other.
- ▶ Collectively refers to process used to create copies of DNA fragments.
- ▶ Animal cloning has been the subject of scientific experiments for years, but garnered little attention until the birth of the first cloned mammal in 1996, a sheep named Dolly.

# □ TYPES OF CLONING



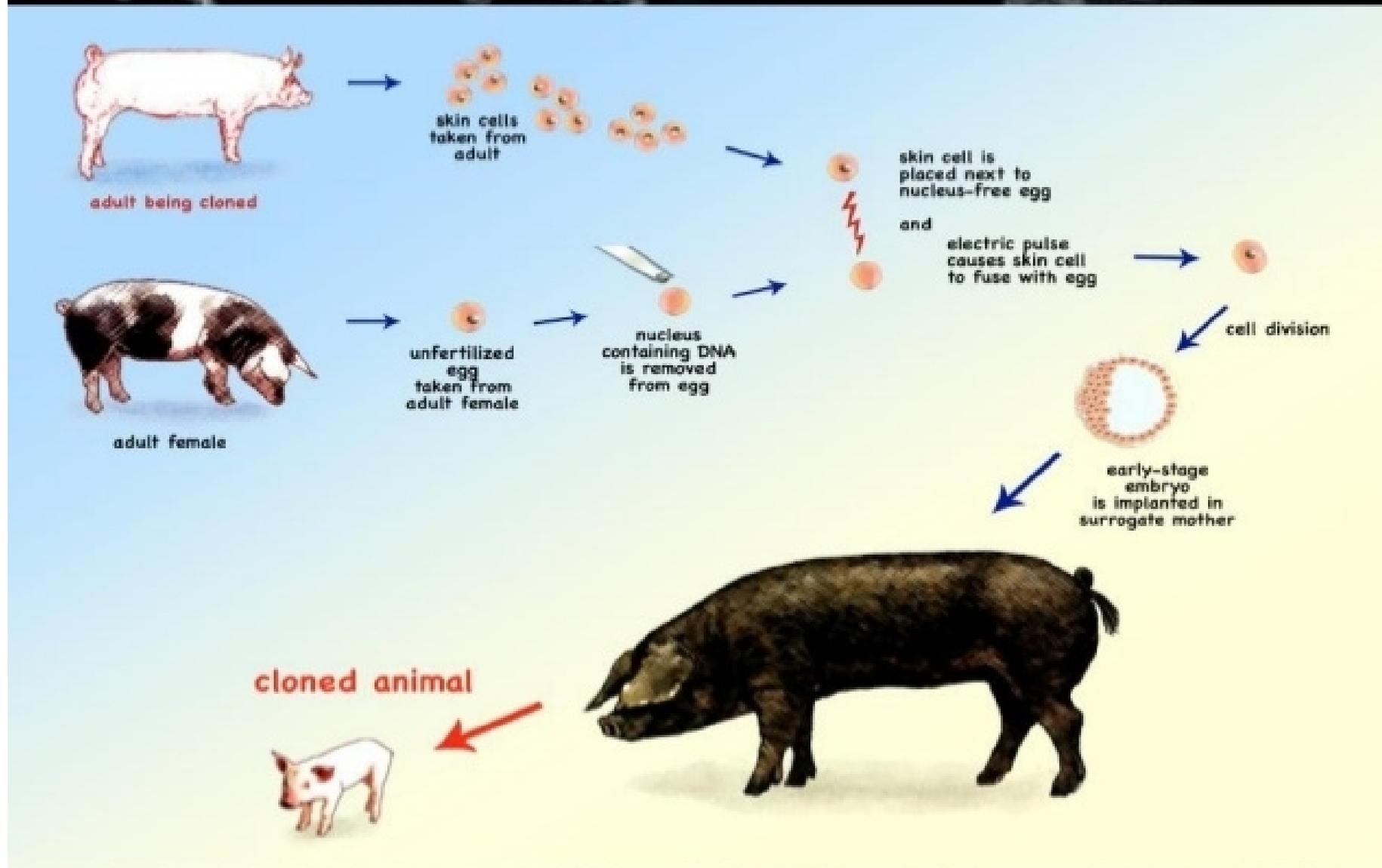
# Natural cloning :

- ▶ In nature , twins form very early in development when the embryo splits in two. Twinning happens in the first days after egg and sperm join, while the embryo is made of just a small number of unspecialized cells.
- ▶ Each half of the embryo continues dividing on its own , ultimately developing into separate , complete individuals. Since they developed from the same fertilized egg, the resulting individuals are genetically identical.

# Artificial Embryo cloning :

- ▶ Artificial embryo twinning is a relatively low-tech way to make clones. As the name suggests, the technique mimics the natural process that creates identical twins.
- ▶ Artificial embryo twinning uses the same approach, but it is carried out in a Petri dish instead of inside the mother. A very early embryo is separated into individual cells , which are allowed to divide and develop for a short time in the Petri dish. The embryos are then placed into surrogate mother, where they finish developing. Again, since all the embryos came from the same fertilised egg, they are genetically identical.

# REPRODUCTIVE CLONING



# ADVANTAGES OF CLONING :

- ▶ **It helps infertile couples to bear children.**
- ▶ **It has great contribution to organ replacement.**
- ▶ **It allows for growing eminent individuals.**
- ▶ **Cure for Diseases.**

# DISADVANTAGES OF CLONING :

- ▶ **It comes with a degree of uncertainty as of yet.**
- ▶ **It is expected to bring about new diseases.**
- ▶ **It might lead to problems in organ rejection.**
- ▶ **It decreases gene diversity.**
- ▶ **In-Breeding.**