

## **Embryology**

**Embryo:** is a stage in animal development between the fertilized egg (zygote) and hatching or birth.

**Embryology:** is the studying of embryos and different process and the development of animals.

**Or:** The study of different processes of animal development, before, during, and after embryonic period.

The life of any organism starts by fertilization and ends by death.

Fertilization needs sperms and ova both carrying  $\frac{1}{2}$  number of chromosomes.

The preparation of these gametes called as **Gametogenesis**.

The zygote changes into complete individual who has organs, by division process and tissue differentiation and cellular differentiation, all these processes are called as **Development**.

We have two types of embryology:

- 1- Phylogeny: development of phylum.
- 2- Ontogeny: development of individual or life history of animal.

To differentiate between 1&2 embryology is defined as:

The study of ontogeny (development of individual) (no.2), can be used as an evidence for phylogeny (development of phylum) (no.1).

### **Types of Embryology:**

- 1- **Descriptive Embryology:** is the science which describe the formation of the embryo and what happen insides it and the time of each stage.

This was studied by eyes then lenses then microscope.

- 2- **Comparative Embryology:** is the science which compares different developing stages of embryos in different animals.
- 3- **Experimental Embryology:** they try to analysis the embryo and its different organs and the reason of their development.

- 4- **Developmental physiology:** studying all the physiological activities of all parts of the embryo.
- 5- **Analytical Embryology:** by using genes and nucleic acid and their functions as a base to analyze different activities.
- 6- **Teratology:** including abnormal development of organs or the loss of organs and studying causes.

## **History of Embryology:**

**Aristotle** 340 b.c. described the formation of embryo inside the egg as follows:

At first the heart is formed, then blood, then blood vessels, then organs.

These formations are done by condensation and coagulation. This theory is called as **Epigenesis theory**.

He also said that: there is a sort of soul which is responsible for condensation & coagulation and this soul come from the father, but the mother is responsible for growing the embryo.

During the 17<sup>th</sup>. & 18<sup>th</sup>. Century a **Preformation theory** was found and there is no condensation and coagulation, but the embryo is performed which is a minute form in the ovum, then this embryo grows into adult.

This theory was explained in many ways:

- 1\_ The Egg contains 200 million micro-eggs, one shall developed, others will transfer to the next generation.
- 2\_ Encasement theory: means that members of different generations are kept inside the egg one inside other like boxes.
- 3\_ By discovering sperms, scientists thought that the sperm carries the embryo, While the ovum provides it with place and nutrition.
- 4\_ Spalanazani found that both ovum and sperm are necessary for initiating of embryo's development.

Preformation theory was dominant to the 19<sup>th</sup>. Century.

**Bears' Law:** The more general features that are common to all the members of a group of animals, which appear in the embryo, developed earlier than the more special features which distinguish the various members of the group.

**Biogenetic Law:** an animal in its individual development try to repeat all characters & stages passed by its ancestors.

It can be put in short words. Ontogeny (development of embryo) repeats phylogeny (development of phylum).

**The two main theories are correct and used today, because:**

- a- The DNA is transferred through generations, so this is an evidence for Preformation theory.
- b- The embryo organs are not appear at the same time; they appear one after the other. This is an evidence for Epigenesis theory.

**Chordate Embryology or Developmental Biology includes following stages:**

- 1- Gametogenesis: **a-** Spermatogenesis. **b-** Oogenesis.
- 2- Fertilization: Union of ovum + sperm = zygote.
- 3- Cleavage: multiple divisions without growth.
- 4- Blastulation: hollow spherical embryo.
- 5- Gastrulation: embryo has 3 germ layers: ectoderm, mesoderm and endoderm.
- 6- Organogenesis: formation of organs.
- 7- Growth: increase in size.
- 8- Differentiation: **a-** Morphological differentiation. **b-** Histological differentiation.  
**c-** Chemical differentiation.

**The Germ Cells**

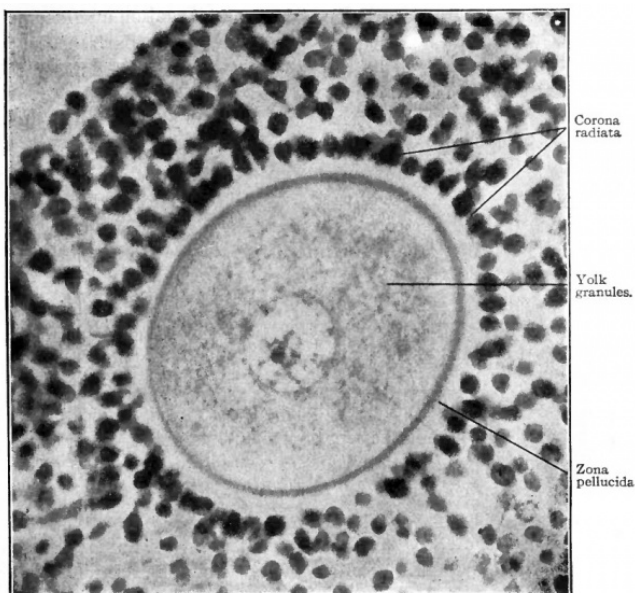
During the life of an individual **germ cells** are formed at certain times .These cells contain the same elements as the somatic cells, that is, nuclear and cytoplasmic components, but there are differences in internal organization which make these cells alone capable of producing a new member of the species. The female carries the ovum (**ovium, female sex cell or germ cell**), the male carries the spermatozoon (**spermium, sperm, male sex cell or germ cell**).

**The Ovum**

The ovum is among the **largest** cells in the animal body, but varies in size from a fraction of a millimeter in some of the **invertebrates** and in mammals to several inches in the **largest birds**. The differences in size are due to **differences in the amounts of food or yolk stored within the egg**.

**The human ovum**

The human ovum as an example of ova containing **a small amount** of yolk , it is not spherical in shape but **ovoid**, with an average diameter of slightly less than 0.2 mm. Surrounding the ovum is the **zona pellucida**, a thick, highly refractive membrane. The outside of this membrane one or two layers of the epithelial cells of the **Graafian follicle** are arranged radially as the **corona radiata**.



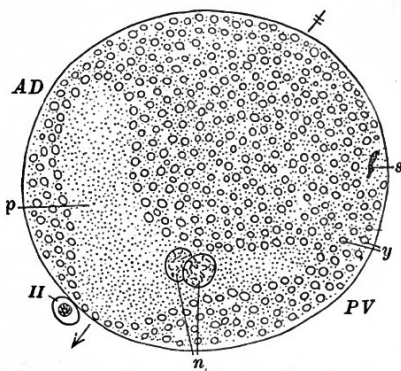
**a section of the ovary of a 12-year old girl.**

The egg cytoplasm called the **vitellus**, the **vitelline** is more **opaque** and more **coarsely granular** than the cytoplasm of most cells. It contain **yolk globules**, these globules are suspended in the cytoplasm and composed of **fatty** and **albuminous substances** that are later utilized in the growth of the embryonic cells. The **yolk globules** are congregated near the **center of the cell**, while a zone of cytoplasm destitute of yolk forms the **peripheral portion** of the ovum. The nucleus is situated near **center** of the ovum amidst the yolk granules.

### Amphioxus egg

The ovum of *Amphioxus* contains only a **small** quantity of yolk, being regarded as a **meiolecithal ovum**, this material is situated off center and the nucleus lies **outside** of the yolk.

Which consider the animal pole and also the side which will be the anterior part of the embryo. The sperm enters the egg at the **vegetative** pole. The sperm nucleus and centrosome then traverse the yolk area to meet the mature egg nucleus .



**Fig. A section through an *Amphioxus* ovum.**

The arrow indicates the direction of the polar axis. AD, antero-dorsal region; PV, postero-ventral region; N, male and female pronuclei; p, yolk-free area; S, tail of sperm; y, yolk area; II, second polar body.

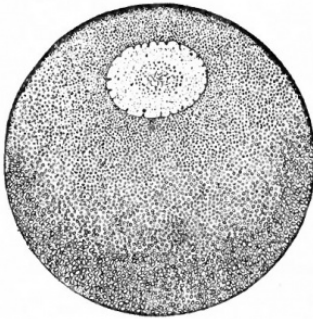
### Frog egg

The frog's egg will serve as an example of an ovum with a **moderate amount** of yolk suspended in the cytoplasm.

Each egg is spherical and measures from **1.5 to 3 mm.** in diameter. The half of the external cell is black because of the presence of pigment granules, and the remainder is nearly white. A **delicate vitelline membrane**, not easily seen, surrounds each ovum. This is a **true cell membrane**.

The animal pole is toward the top of the egg, the nucleus is situated nearer the animal pole, in the center of the cytoplasmic mass. The yolk globules can be seen in the lower part of the figure.

The nucleus of the frog's ovum is **eccentric**, situated nearer the animal than the vegetal pole. Being thus situated it obviously tends to occupy the **center of the cytoplasmic mass**.



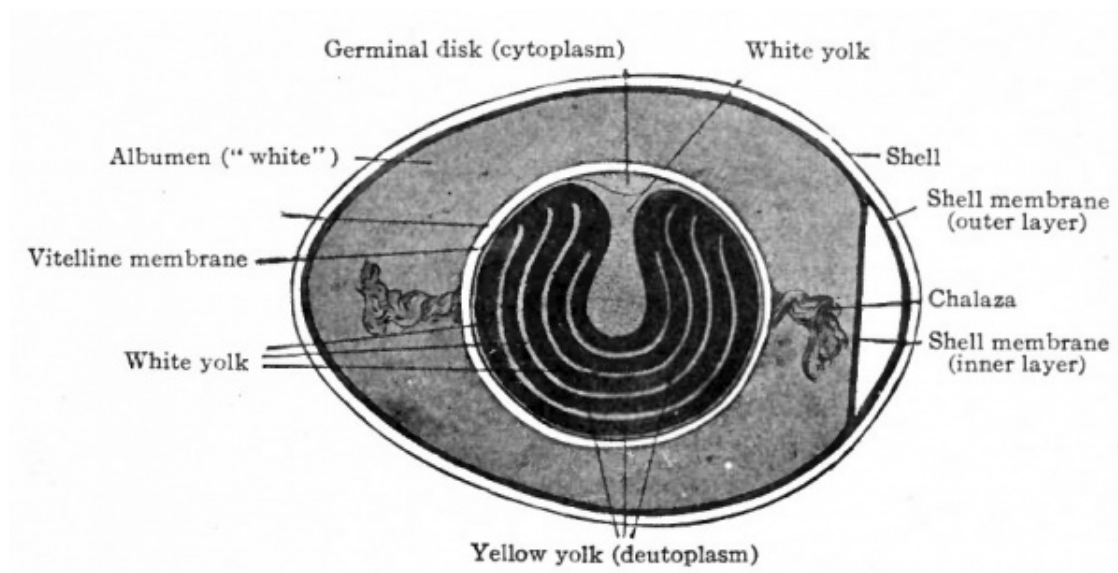
**The section of frog egg**

### **Bird egg**

The freshly laid **hen's egg** may be chosen as an example of a large ovum with a **relatively great quantity of yolk**. The outer covering is the **shell**, a **calcareous substance**. If the shell is broken the tough shell-membrane appears; this is a double layer with a considerable air space between the layers at the larger end of the egg. Under this membrane is the thick layer of **albuminous** substance with a denser twisted portion, **the chalaza**, at each end of the egg. All these structures are secondary egg membranes secreted around the ovum proper by the epithelium of the oviduct during its passage through that organ.

The ovum proper consists of the large spherical mass of yolk, **25 mm.** or more in diameter, and a small disk of cytoplasm, which rests upon the yolk.

At this time the yolk mass is quite similar to that of the egg after laying, and the small disk of cytoplasm containing a single flat nucleus is **attached** to one side of the yolk. While a few small yolk granules are suspended in the cytoplasm, there is transition from the cytoplasmic disk to pure yolk. The greater part of the yolk don't contains cytoplasm but consists of nutritive substances which are important for growing embryo.



**Fig. 3. Diagram of a vertical section through an unfertilized hen's egg. Bonnet.**

The presence of the large quantity of yolk in the ova of birds and reptiles is correlated with the **long period** during which embryos of these animals undergo development within their shells before hatching and attaining ability to get their own food.

In the case of the frog the moderate amount of yolk in the egg serves as food for the growing embryo until it becomes a free-swimming larva or tadpole.

An embryo of a mammal develops for a long period in the uterus of its mother from an ovum with scanty yolk, **but provision is made for drawing nourishment directly from the maternal blood during this time.**

A simple classification of ova is made on the **basis of the amount and distribution of the yolk content.**

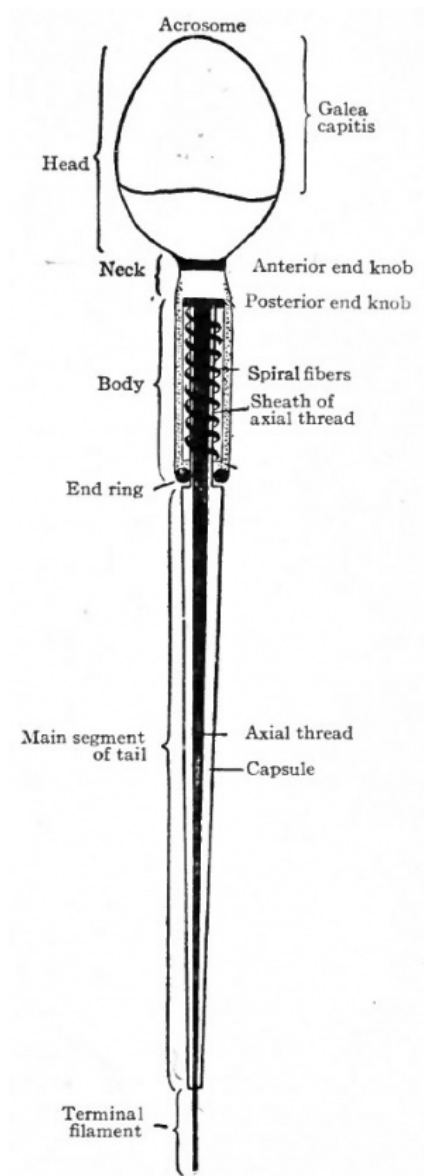
1-The term **meiolecithal** is used, to designate ova in which **the yolk granules are few** (many invertebrates, Amphioxus, mammals).

2- **Mesolecithal** ova are those which contain moderate quantities of yolk (amphibians).

3-Ova that possess large yolk content are classed as **polylecithal** (certain fishes, reptiles, birds)

In case the yolk is accumulated in greater quantity toward one pole the ovum is **telolecithal**, while in case of nearly **uniform** distribution it is **homolecithal** as mammal.

## Spermatozoon



**Fig. Diagram of a human spermatozoon.**

Compared with the ovum the spermatozoon is a small cell bearing **little resemblance** to the typical cell.

It is so small in most animals that the volume ovum of the same species larger the volume of sperm several **hundred thousand times**. Its shape and structure are associated with its high degree of motility. All spermatozoa of vertebrates are of the **flagellate type**.

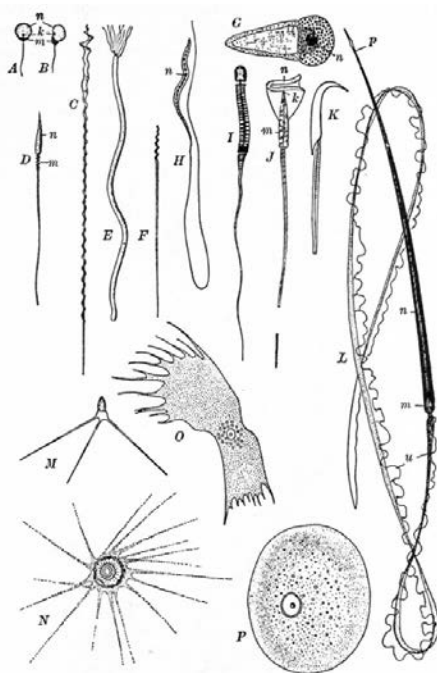
The human spermatozoon composed of **head**, a **middle piece or body**, and a **tail**. On side view the head is oval, a little narrower at the front end.



The nucleus is situated in the head, **nearer the attachment of the body**, and a thin layer of cytoplasm, the **galea capitis**, surrounds the nucleus and is continued forward as the **acrosome**. The head being much smaller than a red blood corpuscle. The body is attached to the broader end of the head and is cylindrical.

The body contains a **delicate fibrile cord**, the **axial thread**, which is continued throughout the tail. Surrounding the axial thread is a capsule of cytoplasm which, does not extend to the tip of the tail, thus leaving the axial thread naked for a short distance. In the body the cytoplasm contains a **spiral fiber**, perhaps of a mitochondrial nature. The body contains the **centrosome** which takes the form of a **double structure**; one part, the **anterior end knob**, is attached to the posterior surface of the head close to the nucleus, the other part, the **posterior end knob**, is situated farther back. A derivative of the centrosome, is the **end ring** which marks the boundary between body and tail.

In the tubules of the mammalian testis, where the spermatogenic cells develop into the mature spermatozoa, the sperms are not motile. They acquire some degree of motility in the tubules of the epididymis and the highest degree only after they are mixed with the secretions of the prostate gland and other accessory sex glands. The number of spermatozoa produced by an individual is greater than the number of ova.



Various types of spermatozoa. ( للأطلاع )

A, B, A teleost; C, D, bird; E, F, snail; G, Ascaris; H, an annulate; I, bat; J, opossum; K, rat; L, salamander; M, N, O, P, crustaceans, k, End knob; w, middle piece; u, undulatory membrane.



# Sexual Cycles in Mammals

In vertebrates, the breeding activities of males & females are seasonal, and their gametes will be mature at the same time.

The related hormones which are secreted by pituitary gland (LH & Progesterone) and estrogen, play an important role in stimulating and maturing and ovulation.

So the young are born at a time of the year when conditions for their growth are most favorable.

## **Estrous cycles:**

Females of mammals which has seasonal reproduction, undergo many short sexual cycles during the year, these are called as estrous cycles, where ovulations take place during them.

The period of estrous cycles is different in animals. It takes few days in mouse and rat, but in dogs it is 3-4 months .....etc.

Some mammal's females undergo more than one estrous cycles in the year, so they are called as **multi estrous cycles animals**.

Or they undergo one estrous cycle only during the year, so they called as **mono estrous cycle animals**.

## **Phases of estrous cycles in mammals:**

According to the ovarian changes, in mammals, estrous cycle is divided into two phases:

### **1- Follicular phase:**

The pituitary gland secretes FSH (follicular stimulating hormone) for developing Graafian follicles.

Also follicular cells secrete Estrogen hormone, which acts for maintenance of endometrial lining of the uterus by increasing its thickness.

### **2- Luteal phase:**

After ovulation and releasing of eggs, a Graafian follicle changes into corpus luteum. While the pituitary gland secretes LH (Luteinizing hormone) which stimulates the corpus luteum to secrete progesterone hormone which is responsible for producing milk; it is also maintain the endometrial lining of the uterus better than estrogen.

Now both estrogen and progesterone will stop the FSH (which is released from pituitary gland) in order to stop the formation and growing of follicles and eggs.

## **If the female is pregnant:**

Generally, in mammals, the egg is fertilized in the upper part of the oviduct.

After few days, it reaches the uterus and prepare itself for implantation. The embryo at those days continues cleavage while it moves inside the oviduct. So the embryo becomes blastula stage when it reaches the uterus.

The pituitary gland continues secreting LH, the corpus luteum remains functional, secreting progesterone, so the ovulation will stop during pregnancy.

After 3 months, the placenta will secrete the progesterone hormone.

## **If the female is not pregnant:**

If mammalian female will not be pregnant, the pituitary gland will stop secreting LH, so the corpus luteum will be degenerated in 10 days, and pituitary gland will start releasing FSH which stimulate the follicles to grow and mature, and a new sexual cycle will start.

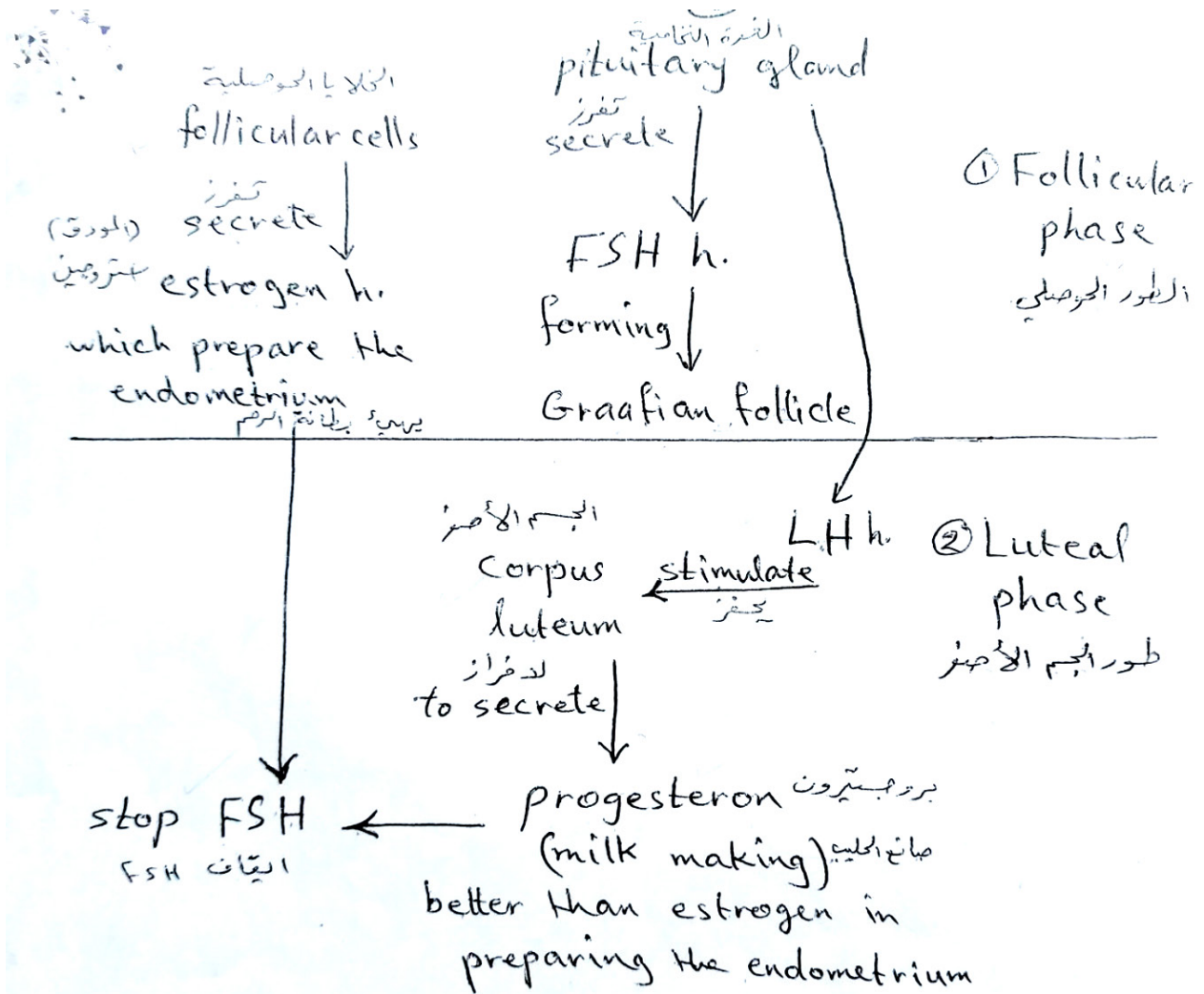


Diagram of estrous cycle phases according to ovarian changes (in mammals).

# **Fertilization**

**Fertilization** means that the contents of an ovum and a sperm unite to form a single structure (zygote), which has a complete number of chromosomes. This is considered as a complex and critical step.

## **Types of fertilization according to the number of sperms entering the ovum:**

**1- Monospermic fertilization:** if one sperm enter and fertilize the egg.

It is common in vertebrates.

**2- Polyspermic fertilization:** if more than one sperm enter the egg but one will fertilize the egg, others will degenerate.

It is rare in vertebrates. e.g. reptiles & birds.

The fertilization always happens in aquatic medium to help sperms for swimming (in random way).

## **Types of fertilization according to the position of fertilization:**

**1- External fertilization:** occurs in aquatic medium outside the bodies of male and female parents, in aquatic animals.

**2- Internal fertilization:** occurs inside female reproductive tract which has fluid media inside it. e.g. birds & mammals.

## **Chemical of fertilization:**

It is known for many years that eggs and sperms of most animals are chemically attracted to one another. These eggs release substances into the surrounding medium that bring about agglutination of the sperms and the adherence of the sperm to the egg. These substances are considered to be more of a trapping agent for the sperm than a chemotaxic factor.

The factors that mediate sperm-egg interaction even before they make contact, were identified in 1914 by Lillie, who proposed the **Fertilization theory**. He observed that sperms of sea urchin agglutinated around unfertilized eggs and activated their motility.

This reaction was specific since some sperms from related species were unaffected. This factor is called as **fertilizin**, which continuously secreted by the egg before fertilization, but it is now clear that **fertilizin** is the constituent of both jelly coats and such as vitelline membrane and plasma membrane.

For sperms to be agglutinated by **fertilizin**, they must have receptor sites on their plasma membrane.

These receptor sites or **antifertilizin** interact with **fertilizin** molecules, like key and lock type interaction.

This interaction is currently called **adhesive recognition**.

## **Importance of fertilization:**

- 1- It activates the zygote to initiate the process of embryonic development.
- 2- It restores the diploid number of chromosomes to the embryo.
- 3- It recombines the heredity characters from parents.
- 4- It detects the sex of embryo, male or female.

## **The basic requirements of fertilization:**



- 1- The fertilization requires fluid medium in marine animals or some body-fluid in viviparous animals.
- 2- The life-span of gametes are limited. Eggs which are shed into water, must be fertilized immediately within few minutes.  
  
Eggs that are fertilized within the body of the female, generally have a longer life-span. E.g. the human egg can be fertilized for at least 24 hours after ovulation.
- 3- To increase the probability of fertilization, the number of sperms must exceed the number of ova.

### **Steps of fertilization in animals:**

#### **1- Formation of fertilization cone:**

The sperms swim randomly to reach the ovum by means of chemo taxis. As the acrosome touch the ovum, the fertilization cone will be formed at this area by the ovum, near the vegetal pole.

#### **2- Swallowing of the sperm:**

The fertilization cone try to swallow the sperm by phagocytosis (Like amoeba when eats by pseudopodia).

#### **3- The penetration of ovum by sperm:**

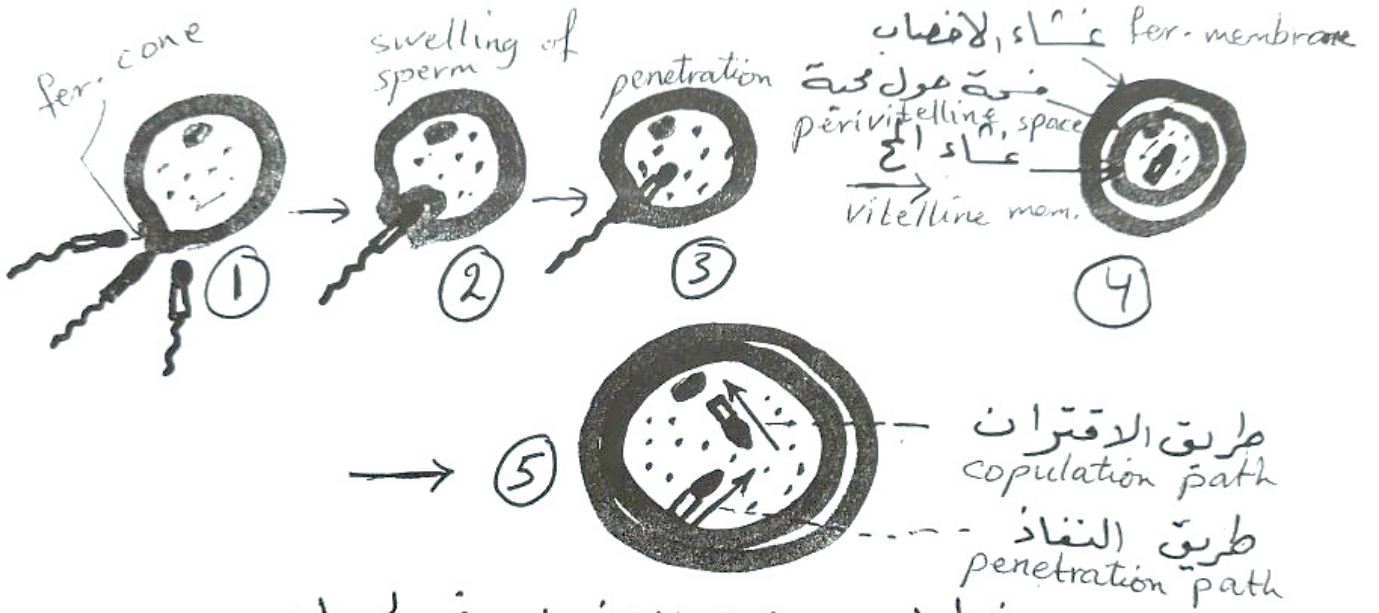
The head and the middle piece of the sperm will be inside the ovum, but the tail will be outside the vitelline membrane.

#### **4- Formation of fertilization membrane:**

The plasma membrane will be separate from the vitelline membrane which is surrounded by perivitelline space, which is filled with water and causes swelling of plasma membrane. This will prevent any excess sperms to penetrate the plasma membrane of the ovum.

#### **5- Union of the sperm and ovum:**

The pathway of sperm after penetration the ovum is called as penetration path. After that the sperm rotate  $180^\circ$ , so the nucleus will be behind the middle piece. Then the sperm will go toward the ovum nucleus by a new direction called as copulation path by chemo taxis. Now the nuclear membranes of both sperm and ovum will degenerate, and both nuclei will fused together forming a new nucleus, which has  $2N$  chromosome, and new nuclear membrane, and the ovum now is called as zygote.



خطوات عملية الاخصاب في الحيوان  
Steps of fertilization in Animal

# Cleavage

**Cleavage:** is the mitotic division of the zygote. It is also called as segmentation. Cleavage follows fertilization or any way which stimulate the ovum to divide. As a result the embryo will consist of many blastomeres. Cleavage starts with division of nucleus first, followed by division of cytoplasm.

During the mitotic division of somatic cells, the resulting cells increase in size to reach the size of the mother cell, then they will divide mitotically. But in the case of cleavage, there is no growth in the resulting blastomeres. And the total size of the embryo is the same as the size of the zygote. This indicate that there is no quantitative change in the cytoplasm of blastomeres.

## **Characteristics of Cleavage:**

- 1- Cleavage is characterized by rapid, successive divisions without intervening period of growth. So it takes a short time.
- 2- Cleavage begins with zygote and ends with blastula, which has the same size of the zygote.
- 3- The ratio of the volume of: nucleus / cytoplasm is raised in comparison with somatic cells.
- 4- Cleavage is a biological process having energy which is coming from its stored materials (yolk).
- 5- The period of one cleavage is varied in different animals, it is 20 minutes in Goldfish, 1 hour in Frog, 12-20 hours in Mammals.

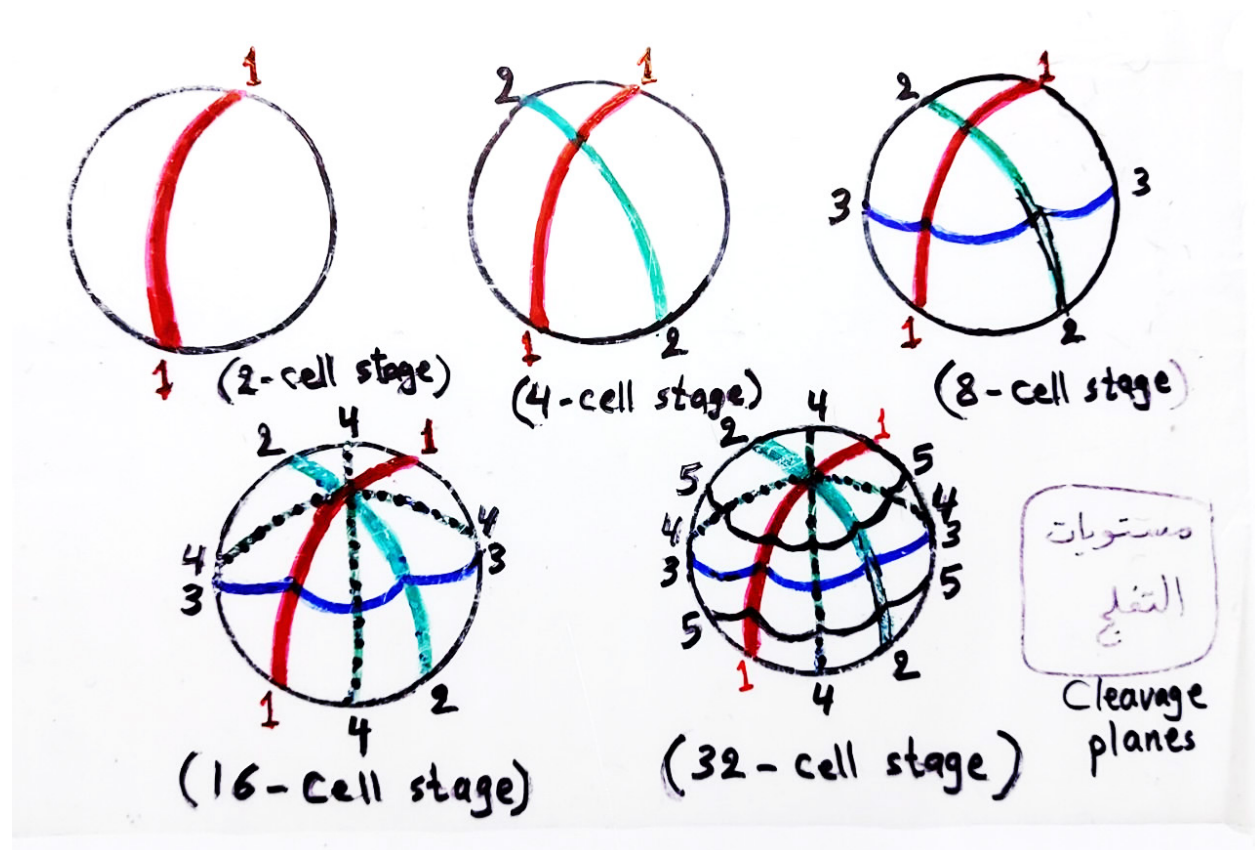
## **Determinate Cleavage & Indeterminate Cleavage:**

Some blastomeres have certain characters. In four cell stage, if we separate one cell of the four, and we let this cell to develop, we see that this cell in some animal species will form a part of the animal but not the whole animal. This mean that the material which forms the embryo is not distributed equally in the blastomeres. So

this cleavage is called as **Determinate cleavage**. E.g. annelida & Mollusca. It is also called as mosaic development.

In other animals, scientists found that the separation of one cell in early cleavage, will lead to the formation of whole (complete) animal, but has smaller size. This means that the material forming the embryo distributed equally in all the blastomeres. This type of cleavage is called as **Indeterminate cleavage**. E.g. echinoderms, amphibian, mammals.

### The characters of the first five Cleavage planes:



1- **The First Cleavage** is vertical (longitudinal) plane, and median along the middle of the egg. It starts from the animal pole then goes down to the vegetal pole. This cleavage is holoblastic or total. The result is two equal blastomeres which remain sticking together.

2- **The Second Cleavage** is also vertical plane, at right angle to the first cleavage. The result is four blastomeres equal in size and sticking together.

3- **The Third cleavage** is horizontal plane locates between the two poles. It produces eight blastomeres.

4- **The Forth cleavage** is a pair of vertical planes, at right angel to each other, resulting in 16 blastomeres.

5- **The Fifth cleavage:** is a pair of horizontal and parallel to each other planes, one is in the animal hemisphere and the other is in the vegetal hemisphere. The result is 32 blastomeres, called as Morula.

Further cleavages are irregular, so, the first five cleavages are enough to study.

### **Factors affecting on cleavage:**

#### **1- Quantity of yolk:**

When the amount of yolk increases, the amount of active cytoplasm gradually decreases, and the position of nucleus is affected. The mitotic divisions of such a displaced nucleus result in unequal-sized blastomeres.

2- Cleavage planes try to divide the blastomeres equally.

This factor is the reverse of the first one, which try to retard and inhibit the process of cleavage, so division will be slower resulting in larger yolky cells. While the second factor try to divide every cell equally.

### **Kinds of cleavage according to the amount and distribution of yolk, they are:**

**A- Total or holoblstic cleavage:** the entire egg divides by each cleavage furrow. It has the following two types:

a- **Equal holoblastic cleavage:** occurs in the microlecithal and isolecithal eggs, it produces blastomeres of equal size. E.g. Amphioxus, Mammals.

b- **Unequal holoblastic cleavage:** occurs in mesolecithal and telolecithal eggs, produces unequal sized blastomeres which include many small-sized blastomeres called as micromeres, and few large-sized yolk macromeres.

E.g. Fishes and Amphibian.

## **B- Meroblastic cleavage:**

This is partial, incomplete, occurs in polylecithal and centrolecithal eggs. The furrows divide the small amount of active cytoplasm of animal pole and most of the egg remains undivided.

It has two types:

a- **Discoidal:** in polylecithal and heavily telolecithal egg, the cleavage is restricted to the germinal disc only. E.g. Reptiles and Birds.

b- **Superficial:** occur in centrolecithal eggs, the cleavage is restricted to the peripheral cytoplasm of egg. E.g.: Insects.

## **Patterns of cleavage:**

### **1- Regular cleavage:**

The dividing blastomeres arrange in regular manner, it has three types:

a- **Radial cleavage:** the blastomeres arrange in a radially symmetrical form.

E.g. Echinoderms.

b- **Spiral cleavage:** the blastomeres arranged in spiral form.

E. g.: Annelida, Mollusca.

c- **Bilateral cleavage:** the blastomeres arranged bilaterally to the plan of symmetry. E.g. Chordates.

### **2- Irregular cleavage:**

The blastomeres don't arrange as anyone of the three patterns above.

E.g. Coelenterate.

# **Blastulation**

**Blastula:** is a stage of embryonic development, appears as a spherical ball, made of layer or layers of cells, with a cavity inside called as blastocoel.

## **Development of blastocoel:**

After the formation of morula, further cleavages are irregular. Now the micromeres divide slightly faster than the macromeres, because they have fewer yolk granules. A jelly - filled cavity now appears at 16-blastomeres stage, and become distinct at 64-blastomere stage. At morula stage, the jelly material starts to absorb water and becomes quite fluid later. This will push the cells to the peripheral sides. This causes little increasing of blastula size. Now the blastomeres arrange around the blastocoel, which is completely separated from outside.

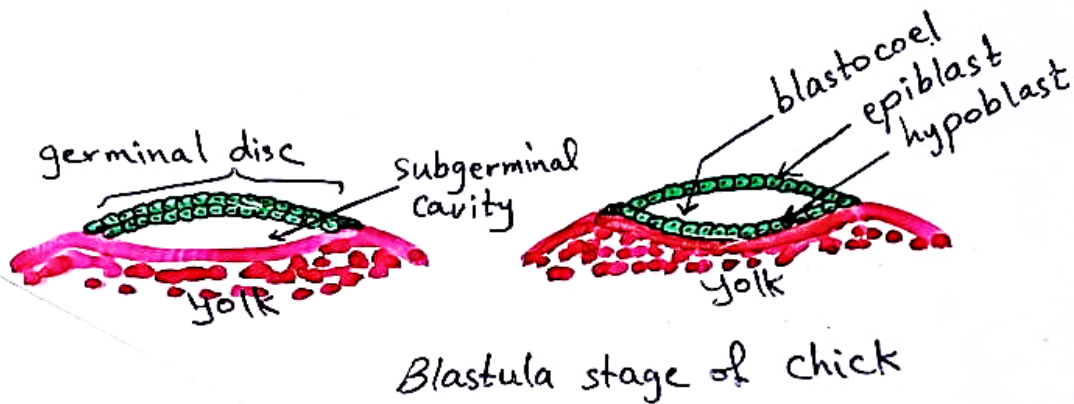
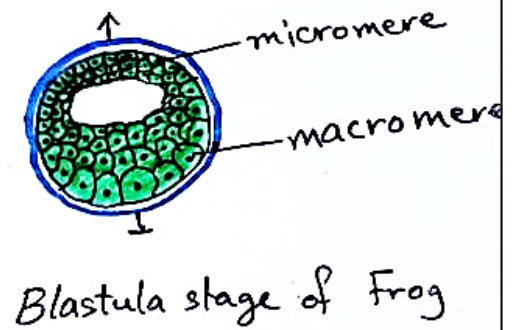
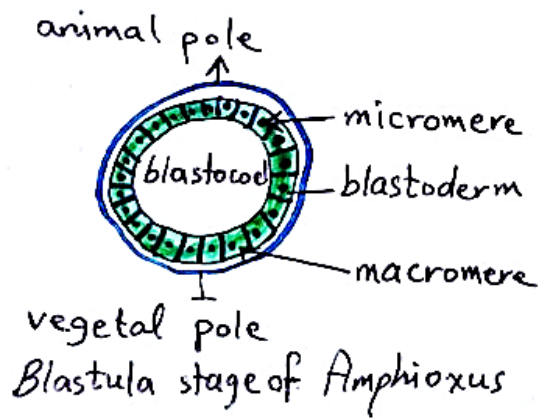
## **Blastulation in isolecithal egg (e.g. Amphioxus):**

There is little amount of yolk so the cleavage is approximately equal. A group of cells is forming a complete blastula with a single layer on the periphery of the embryo and a blastocoel. The cells are not exactly the same, the cells in the animal hemisphere are smaller than the cells in the vegetal hemisphere.

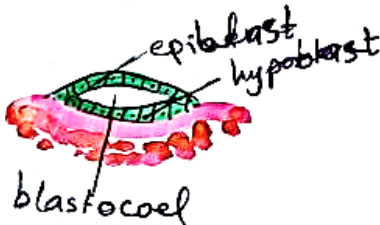
## **Blastulation in moderately telolecithal egg (e.g. Frog):**

Due to the presence of moderate amount of yolk, division will be faster at animal pole, so cells are smaller, while division in vegetal pole is slower, so they are larger. The roof of blastula is made of 2 or more layers of blastomeres.





subgerminal cavity



Delamination  
Theory

Involution Theory

Infiltration Theory

Theories of hypoblast formation

## **Blastulation in heavily telolecithal egg (e.g. Bird):**

Blastulation takes place in germinal disc only. In central part of germinal disc, after cleavage, multilayered cells will separate from yolk, while marginal cells still attach to yolk. Due to separation, sub-germinal cavity develops between blastomeres and yolk. Outer blastomeres called epiblast.

Few blastomeres migrate down and arranged into a thin layer called hypoblast over the yolk. The cavity between epiblast and hypoblast is called blastocoel, and these 3 structures form the blastula.

## **There are 3 theories about the formation of hypoblast:**

### **1. Delamination theory:**

Splitting of cell layers into 2 layers with blastocoel between them.

### **2. Involution theory:**

Some cells involute peripherally creep on the yolk and forming a new layer (hypoblast) and a new cavity is blastocoel.

### **3. Infiltration theory:**

Some cells of epiblast separate as single or group of cells downward and settle on the yolk.

## **Fate Maps**

Scientists did experiments on blastula to determine growth and direction of presumptive cells in future. Long time ago, scientists used simple methods then complex methods. They could draw fate maps of many animals.

## **Methods of preparation of fate maps:**

### **1- Using natural marking:**

Amphibian egg has two different natural pigments. The black one covers the animal hemisphere and it has the ability to absorb sun light which release heat that used in

embryonic development. This pigment with its cells will form ectoderm. The vegetal hemisphere is yellow in color (has yellow pigment) will form endoderm.

## **2- Using artificial markings:**

The first person who used vital stains was Vogt, 1925, e.g. Nile blue & Natural red.

**Vital stain:** is a harmless dye which colours the cells (blastomeres) without interfere with cell process or mitosis. These stains are extracted from natural plants.

For example: to construct a fate map of amphibian, a small piece of agar or cellophane (both are called as stain carriers) is stained with a vital stain and is pressed against a chosen area of the blastula for a short period. The stain diffuses from the piece of agar or cellophane into the blastomeres, which will retain their coloration for several days. so it is possible to follow the movements of gastrulation by continuous observations.

## **3- Carbone particles marking:**

This method was used by Spratt, 1946, by applying tiny particles of carbon to the surface of the embryo. They stick to the surface of the cells and can be used as markers.

## **4- Radio-active labelling method:**

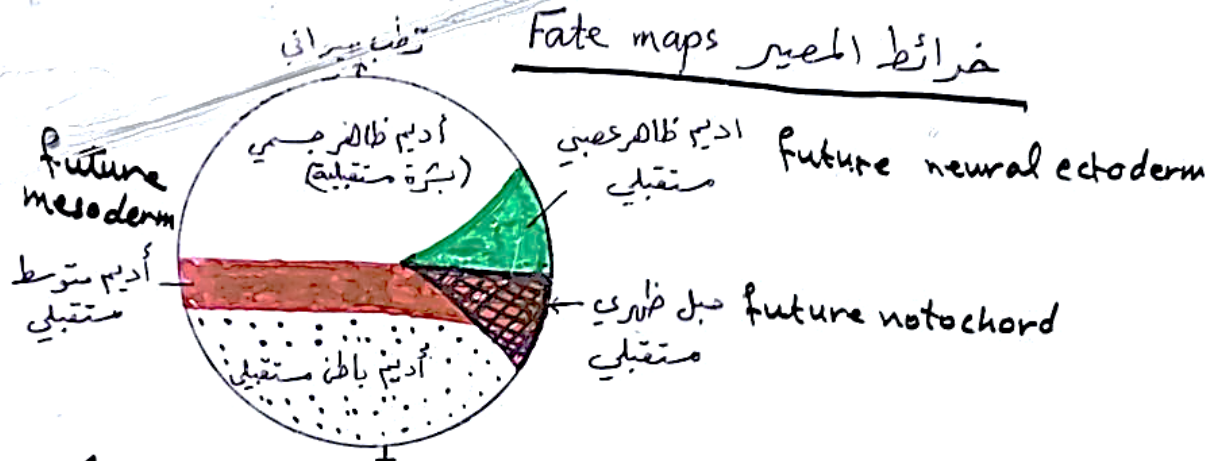
When the substance tritiate thymidine is used in very little amount, it will not be harmful for cells. This will incorporate with the DNA in the nucleus prior to each cell division, then radioactivity will be examined by Radioautographs.

## **Conclusions from fate maps:**

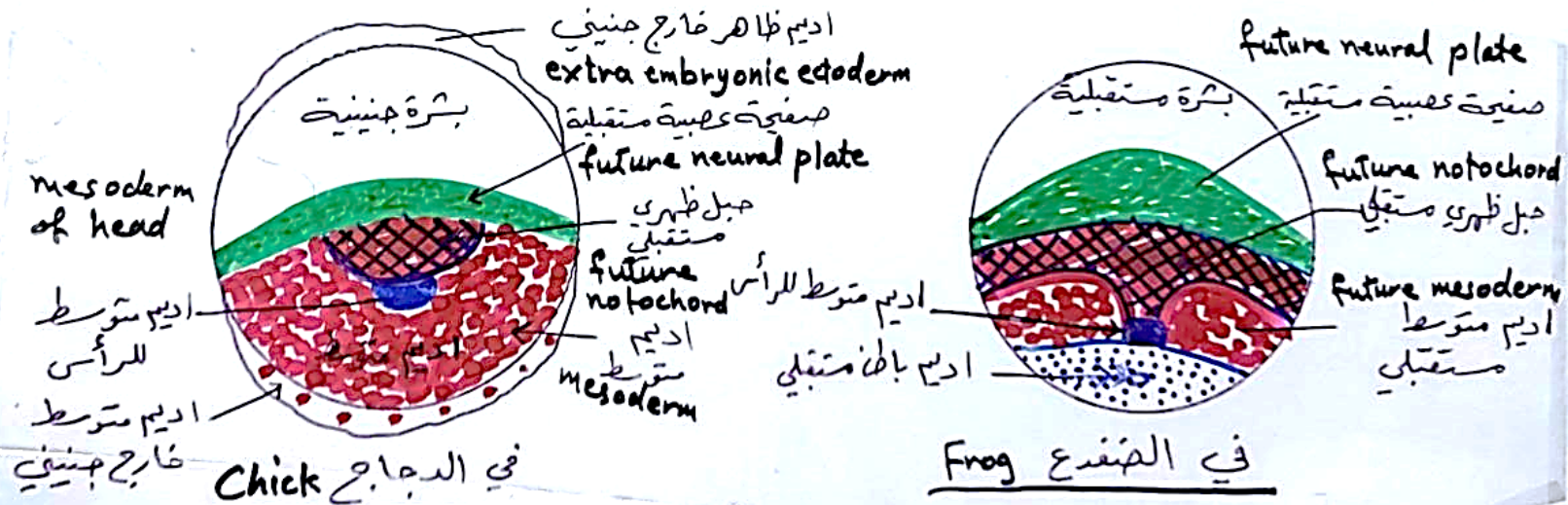
After studying of fate maps, we conclude:

- 1- In Amphioxus & Amphibian, the whole blastula will form the embryo. While in Reptiles, Aves and Mammals, some parts of blastula will form the embryo, and other parts of it will be assistant as extra embryonic structures.
- 2- The blastoderms of animal hemisphere will form ectoderm. The blastoderms of vegetal hemisphere will form endoderm. The blastoderms which will form mesoderm, will be in lateral position.

## خرائط المصير Fate maps



### في الرميح Amphioxus



### في الدجاج Chick

### في الضفدع Frog

Gastrulation is a process of changing the embryo from blastula (hallow ball) into embryo with 3 germinal layers (ectoderm, mesoderm & endoderm), having a new cavity called as archenteron.

## Types of gastrula:

1- Some invertebrates have gastrula with double layer, so they are called as

**Diploblastic animals.**

2- In vertebrates, they have gastrula with three layers, so they are called as

**Triploblastic animals.**

## Morphogenic movements:

The movement of cells and changing of their size, shape and position, to form the three germinal layers, is called as: **Morphogenic movements.**

Morphogenic movements are affected by 3 factors depending on the type of the eggs:

1- Size of blastocoel.

2- Thickness of blastula wall.

3- Size of cells.

**Morphogenic Movements** have two types:

A- Epiboly

B- Emboly

**Epiboly:** is the ability of ectodermal sheets to expand into the free areas over a substratum, they are usually outside, and tend to envelope and surround inner cells. These ectodermal sheets expand toward a single point on the surface.

**Emboly:** is just opposite to epiboly. A sheet migrates inside a cell group or beneath another layer because of local changes in cell shape or motility. Emboly includes:

- 1- **Invagination**, is punching of an expanding layer at a specific point.
- 2- **Involution**, is turning of an expanding layer at a specific point and enters inside to expand and forms new layers.
- 3- **Ingression**, when small groups of cells separate as single or many cells from a primary ectodermal layer and migrate into blastocoel or other embryonic spaces as a new layer.

## **Gastrulation in Amphioxus**

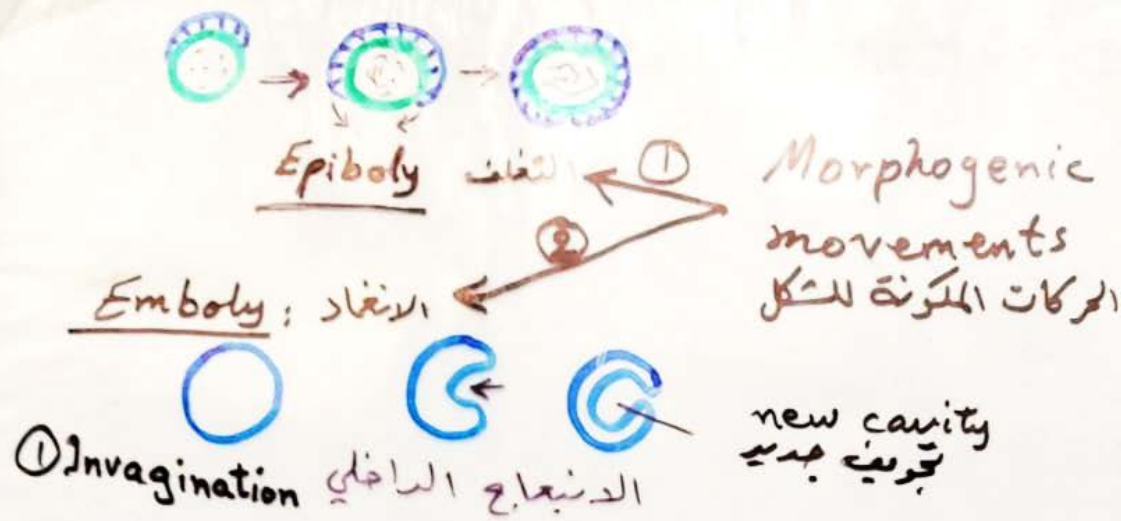
- 1- The blastula of amphioxus.
- 2- The vegetal pole cells become flattened.
- 3- Invagination of the flattened cells inside the blastocoel, because the animal Pole cells divide faster than those of the vegetal pole.
- 4- A double layer cup shape is formed with a new cavity called as **archenteron**.
- 5- As the archenteron enlarges, the blastocoel disappears.

The blastopore becomes narrow. The macromeres become inside the gastrula to form its inner wall (hypoblast). Some micromeres also go inside the gastrula by turning along the dorsal side of the blastopore lip to form the roof of archenteron, which will form later on the **notochord**.

In the median dorsal side of the gastrula, an elongated plate of cells will formed, called as **neural plate**, which will form neural tube.

The blastopore is triangle in shape, has one dorsal lip, and 2 lateral lips. Over the dorsal lip, a group of notochord cells will appear, and over them there is a group of neural cells which will form neural tube.





### Gastrulation of Amphioxus



## **The future of blastopore:**

It represents the future mouth in coelenterates.

It represents the future mouth and anus in annelida, crustacean, & Mollusca.

It represents the future anus in echinoderms & chordates.

## **Gastrulation in Amphibia**

### **A- Early Gastrula:**

By the time epiboly starts, there appears a small depression at the junction between micromeres & macromeres. This is the beginning of dorsal lip of blastopore. The depression become deeper and a small cavity will appear, it is the archenteron.

### **B- Mid Gastrula:**

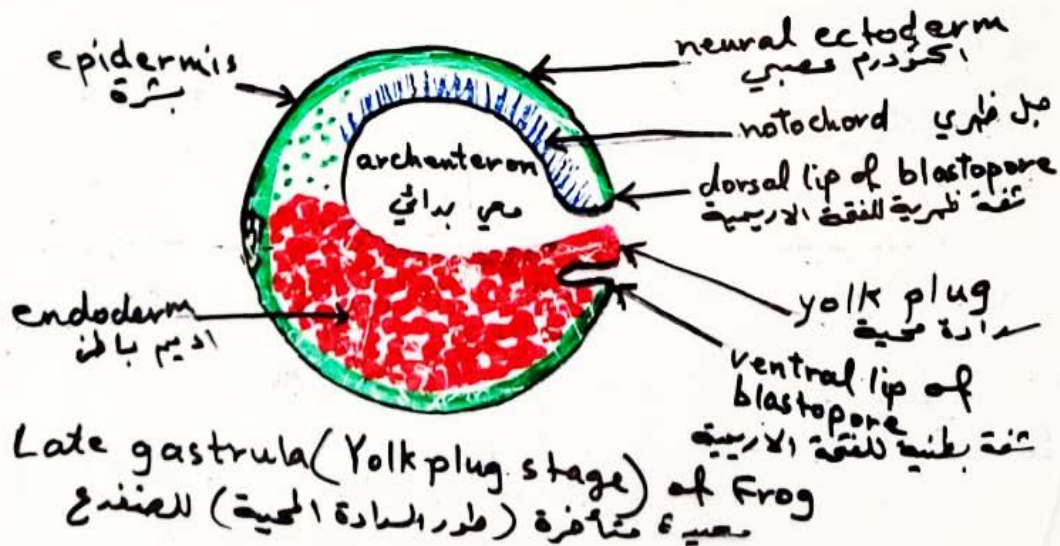
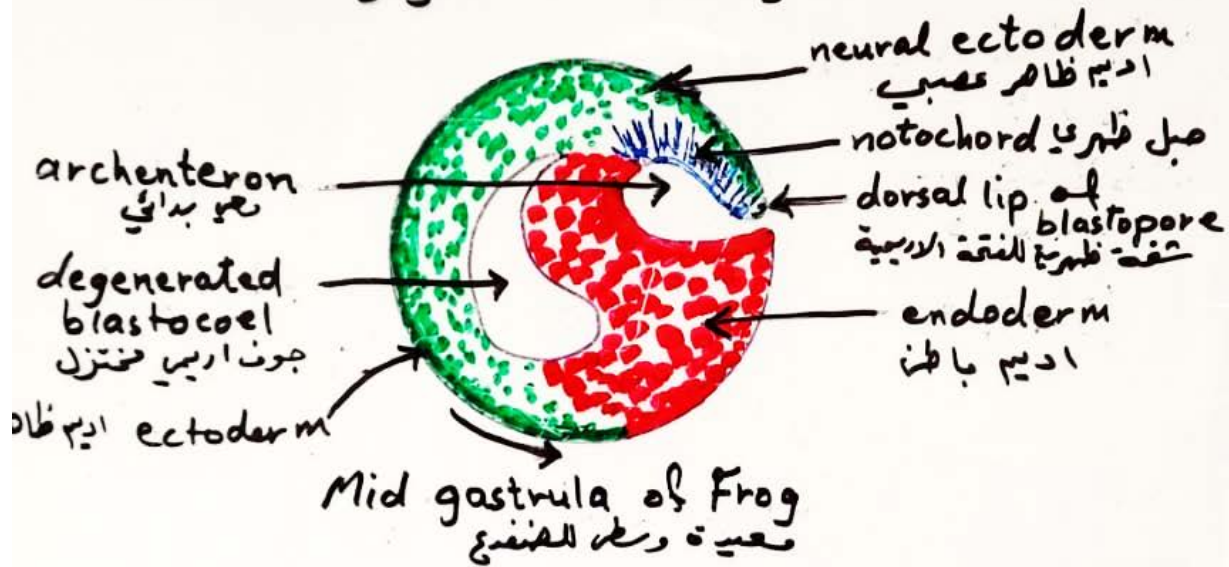
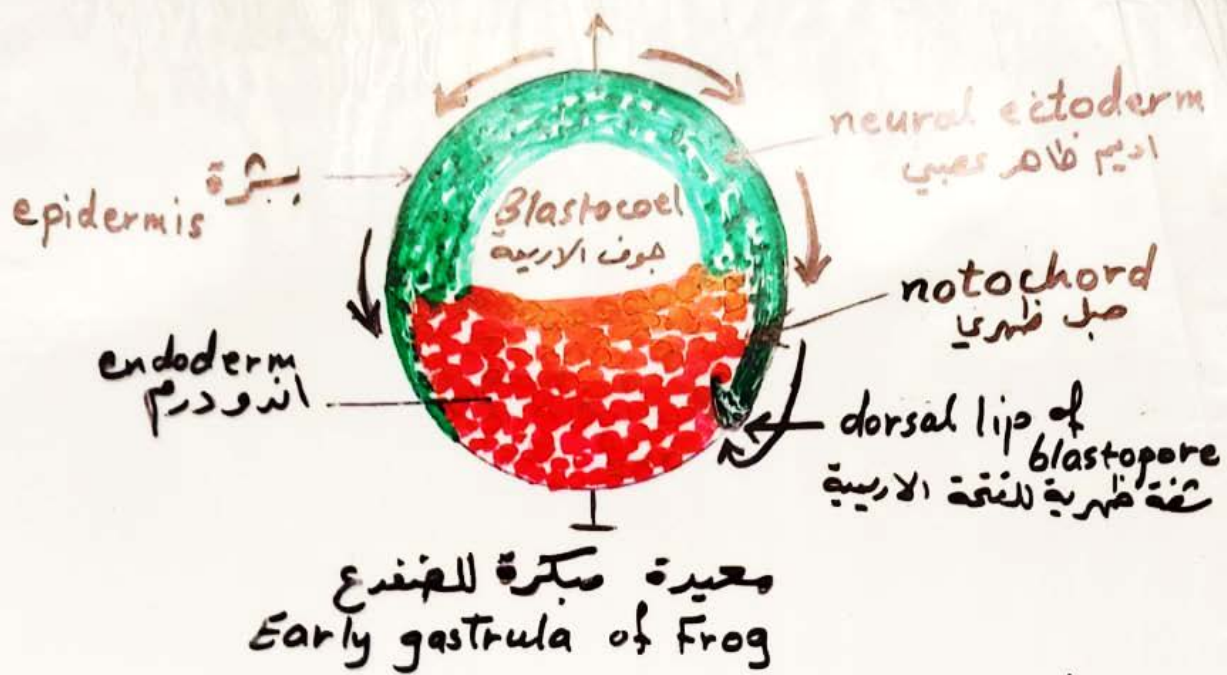
As the development progress, the archenteron enlarges in size, while blastocoel decrease in size. Epiboly continues to cover more areas of the embryo surface, neural ectoderm will be in the superficial dorsal median position of the embryo. Notochord cells (mesoderm) continue their involution through the dorsal lip of blastopore to be in a dorso-median internal position over the archenteron.

### **C- Late Gastrula (Yolk Plug stage):**

- 1- Epiboly continues and the ventral lip of blastopore is formed.
- 2- Ectodermal cells will cover all the embryo except a small area (blastopore), which shows endodermal cells from inside.
- 3- The blastocoel degenerates.
- 4- Mesodermal structures involute through lateral lips of blastopore to take a position between ectoderm & endoderm.

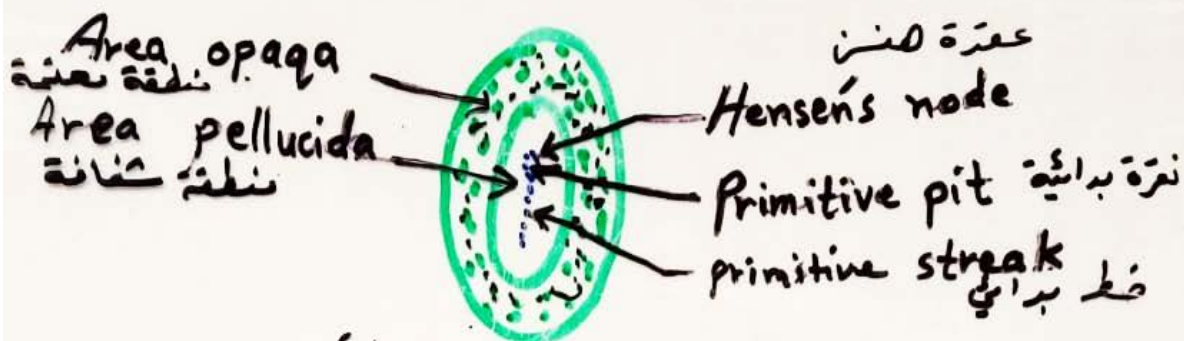
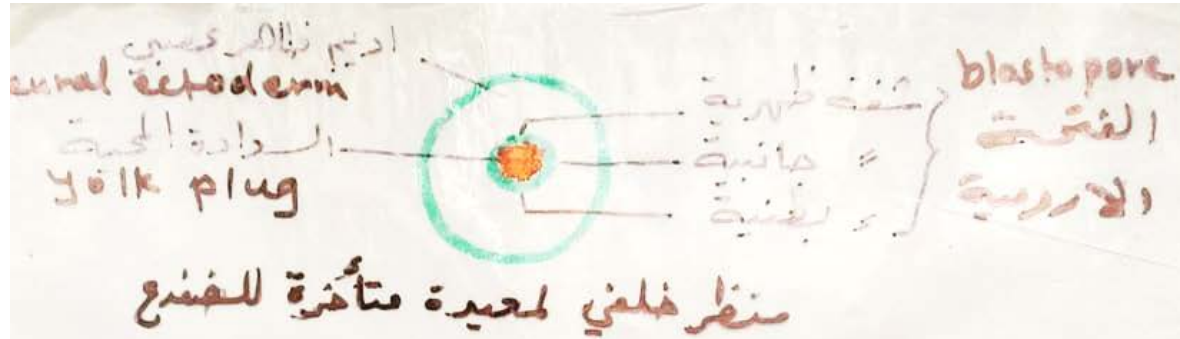
**Blastopore has 4 lips:** one dorsal, one ventral, & 2 lateral.



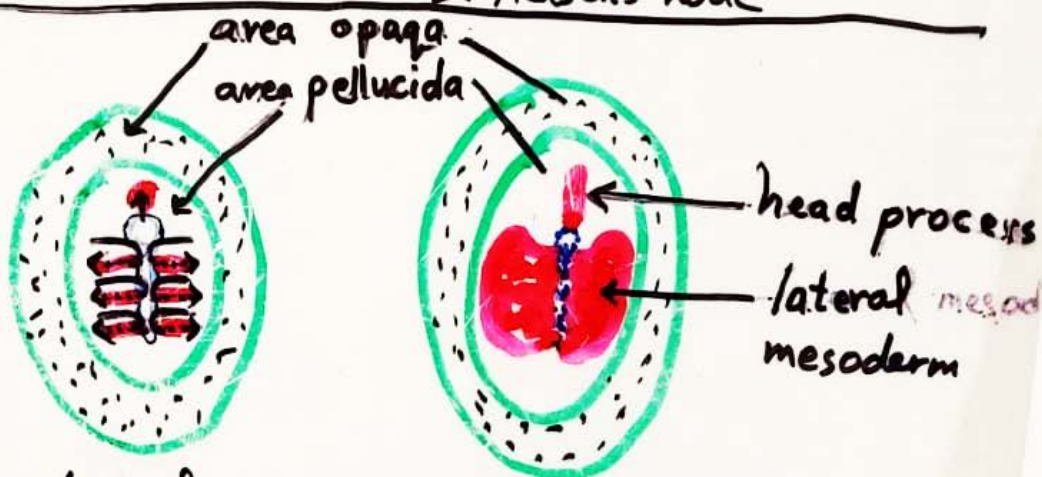


## Gastrulation in Chick

- 1- In blastula, epiblast is the future ectoderm & future mesoderm. Hypoblast is the future endoderm.
- 2- The epiblast cells increase in number, so they move and go inside the blastocoel through a groove called as **primitive groove**.
- 3- The large number & crowding of cells make two folds in either side of the primitive groove, each one called as **primitive fold**. The three structures (2 folds & groove) called as **primitive streak**.
- 3- The anterior part of the primitive streak becomes thick and has a pit in the middle called as **Henson`s node**.
- 4- The dividing cells of epiblast, coming from the two lateral sides, will enter through primitive streak then direct laterally in both sides forming a shape of butterfly wings, which is made of mesodermal layer and called as **lateral mesoderm**.
- 5- The dividing cells coming from anterior side will enter Henson`s node, then extend anteriorly, will form the **head process** which will form the **notochord**.



منظر سطحي لجنين الطيور ايوغح عقدة هسن والخط البدائي  
Surface view of chick embryo showing primitive streak & Hensen's node



Mesoderm formation of chick gastrula  
تكوين طبقة الميزودرم في معينة الطيور (الدجاج)