Ministry of Higher Education and Scientific Research University of Baghdad College of Science Department of Biology



# Practical Embryology 2021-2022

المرحلة الرابعة - الدراستين الصباحية والمسائية الفصل الدراسي الاول تدريسيي المادة : أ.د.جبار حميد ينزيل أ.م.د.لينا عبد المطلب صالح م.د.ياسمين لطيف السعدي م. لمياء عبدالرضا فاضل م.م. بشرى رشيد ابراهيم م.م. هند قصي م.م. أسراء محمد مبارك م.م.أسراء حسن علي م.م. أمراء محمد مبارك م.م.

# Lab. 1 MICROSCOPES

# **MICROSCOPES**

Most important magnifies objects (makes objects look bigger) help scientists study objects and living things too small to see with the naked eye.



# Parts of microscope

# 1.Ocular (lens) eyepiece

The lens of the microscope that you look through.

# 2.Course adjustment

The large knob on the microscope that you turn to bring the object into focus.

# 3.Fine adjustment

The small knob on the microscope that brings the image into focus.

# 4.Arm

The part of the microscope supporting the body tube.

# 5.Head

The part that holds the eyepiece and the objective lenses.

# **6.Nosepiece**

The part at the bottom of the body tube that holds the objective lenses and allows them to be turned.

# 7. Objective lens

Magnifying lenses with different magnification power include:

# a- very high power

The lens that magnifies the object the greatest amount (usually 100x) called oil lens

# b-high power objective lens 40x

c-middle power lens that magnifies the object usually 10x to 20x.

# d- low power that magnifies the object as scanner lens, 4x

# 8.Stage

The flat part below the objectives lens where the slide is placed.

# 9.Clip

The part that holds the slide in place so it doesn't move.

# 10.Diaphragm

The part that controls the amount of light entering the field of view.

# **11.Light source**

The lamp (or mirror) under the stage that sends light through the object being viewed.

# 12.Base

The bottom part that supports the rest of the microscope.

# **Magnification**

In order to ascertain the total magnification when viewing an image with a compound light microscope, take the power of the objective lens which is at 4x, 10x or 40x and multiply it by the power of the eyepiece which is typically 10x.

Therefore, a 10x eyepiece used with a 40X objective lens, will produce a magnification of 400X. The naked eye can now view the specimen at a magnification 400 times greater.

# Lab. 2 Introduction to practical embryology

**Embryology** (from Greek embryon, "the unborn, embryo"; and logia) is the branch of biology that studies the prenatal development of gametes (sex cells), fertilization, and development of embryos and fetuses. Additionally, embryology includes the study of congenital disorders that occur before birth, known as teratology. Embryology has a long history. Aristotle proposed the currently accepted theory of epigenesis, that organisms develop from seed or egg in a sequence of steps. The alternative theory, preformation, that organisms develop from pre-existing miniature versions of themselves, however, held sway until the 18th century. Modern embryology developed from the work of von Baer, though accurate observations.

# Animal models in embryology

All human and animal embryos go through very similar stages of early development. The major difference appears to be how long it takes to reach each of these same stages many research animal species that have been used as models of development.

There many systems in study the timeline of embryonic development of different animal species. Carnegie stages are named after the famous USA institute which began collecting and classifying embryos in the early 1900's. Human stages are based on the external and/or internal features of the embryo, and are not directly dependent on either age or size.

The human embryonic period proper is divided into 23 Carnegie stages covering the first 8 weeks after fertilization. This period is most of the first trimester and the second and third trimester is called Fetal Development. Other features used in this classification include: ranges of age in days, number of segments (somites) present, and embryonic size (CRL, crown rump length).



#### Vertebrate Axis Formation

Animal bodies have externally visible symmetry. However, the internal organs are not symmetric. For example, the heart is on the left side and the liver on the right. The formation of the central left-right axis is an important process during development. This internal asymmetry is established very early during development and involves many genes. Even as the germ layers form, the ball of cells still retains its spherical shape. However, animal bodies have lateral-medial (left-right), dorsalventral (back-belly), and anterior-posterior (head-feet) axes.



# The Cell Reproduction

One of the most significant characteristics of the living organisms is the ability to reproduce. All living cells and organisms reproduce, producing offspring like themselves, and pass on the hereditary information contained in their DNA molecules. If the organisms were unable to reproduce they would decrease in number and become extinct. The reproduction process is varied and complex, and depends upon the ability of individual cells to replicate. All cells arise from preexisting cells by some mechanism of cell reproduction or division. In conclusion, we can define the cell reproduction as the division of a parent cell into two daughter cells. This process includes the number of events, that illustrated as in the following:

- ✤ The replication of DNA.
- ✤ The equal distribution of DNA to the two daughter cells.
- ✤ The division of the rest of cell material into two daughter cells.

#### **The Cell Reproduction Types**

The cell reproduction is divided into two main types: **asexual** and **sexual cell reproduction**. Typically, in asexual reproduction, a single cell gives rise to a genetic duplicate of the parental cell, without any genetic contribution from another individual, while sexual cell reproduction involves the genetic recombination of two cells.

#### - Asexual Reproduction

Asexual reproduction is the production of offspring from a single organism, that are genetically identical to the parent because the offspring are all clones (copies) of the original parent. Hence, no gametes are formed and no fertilization is involved in the formation of a new organism. Asexual reproduction can form generations rapidly compared to sexual reproduction. This type of reproduction occurs in prokaryotic microorganisms (bacteria) and in some eukaryotic single-celled and multi-celled organisms.

# **Types of Asexual Reproduction**

Animals may reproduce asexually through various mechanisms such as fission, budding, fragmentation, or parthenogenesis.

#### 1-Fission

Two types of fission can be identified: binary fission and multiple fission.

### **A- Binary Fission**

Where every organelle is copied and the organism divides in two daughter organisms.

#### There are three types of binary fission:

a-Irregular binary fission, which can observe in Amoeba

#### b- Longitudinal binary fission, which can occur in Euglena

c- Transverse binary fission, which can observe in *Paramecium*.



The types of binary fission in protozoans

# **B- Multiple Fission**:

The nucleus is divided several times to produce multiple daughter cells, which occurs in protozoans like *Plasmodium*.



#### Multiple fission in *Plasmodium*

# 2-Budding

A new individual cell (organism) develops from an outgrowth of a parent cell (organism), splits off, and lives independently. The budding occurs in *Hydra*. In *Hydra*, a bud forms that develops into an adult, which breaks away from the main body.



#### **3-Fragmentation**

Fragmentation is the formation of a new organism from a fragment of the parent organism. Each fragment is capable of developing into a new organism. The fragmentation observed in planarians and starfish.



#### The Fragmentation in planarian

#### **4-Parthenogenesis**

Parthenogenesis is a form of asexual reproduction where an unfertilized egg develops into a complete individual. The resulting offspring can be either haploid or diploid, depending on the process and the species. Aphids, bees, amphibians, and some reptiles produce by parthenogenesis. Bees use parthenogenesis to produce haploid males (drones) and diploid females (workers). If an egg is fertilized, a queen is produced. The queen bee controls the reproduction of the hive bees to regulate the type of bee produced.

#### - Sexual Reproduction

Sexual reproduction is the combination of (usually haploid, or having a single set of unpaired chromosomes) reproductive cells from two individuals to form a unique offspring (usually diploid, having a pair of each type of chromosome).

Cells that reproduce sexually are characterized by **meiosis**, the nuclear division process by which sex cells (gametes) are formed. Every chromosome of a somatic cell occurs in a pair (diploid). During meiosis these diploid pairs of chromosomes duplicate and are separated so that each meiotic sex cell has only one chromosome (haploid) of each pair.

Meiotic divisions result in the production of haploid sperm and egg cells, each with one-half of the amount of the parental genome.

During the life cycle of sexually reproducing organisms, fertilization results in the fusion of haploid gametes (sperm and egg) producing the zygote, the zygote undergoes cellular differentiation, whereby cells become structurally, functionally, and biochemically distinct from each other.

9



#### Sexual reproduction in human

#### - Hermaphroditism

Hermaphroditism also referred to as intersex, occurs in animals where one individual has both male and female reproductive parts. Invertebrates, such as earthworms, slugs, tapeworms and snails, are often hermaphroditic. Hermaphrodites may self-fertilize or may mate with another of their species, fertilizing each other and both producing offspring. Self-fertilization is common in animals that have limited mobility or are not motile, such as snails, slugs and clams.

NO.	Asexual Reproduction	Sexual Reproduction
1	It happens in lower invertebrates	It happens almost in all types of
	and lower chordates and plants	animals and mostly in higher
	with simple organizations.	plants.
2	It is always uni-parental.	It is usually bi-parental
3	Gametes are not formed.	Gametes are always formed
4	No fertilization.	Fertilization takes place
5	It involves only mitosis.	It involves both meiosis and

		mitosis.
6	Daughter organisms are	Daughter organisms genetically
	genetically identical to the parent	differ from the parents.
	and called clones	
7	Multiplication occurs rapidly.	Multiplication is not as rapid as
		in asexual reproduction.
8	Since there is no variation, so it	Since there are variations, so it
	does not contribute to evolution	contributes to evolution of the
	of the species	species.

#### **Cell Division**

Growth and development of every organism depend on multiplication, enlargement and differentiation of its cells, beginning from the zygote.

Cell division increases the number of cells, both the somatic cells and the sex cells depended directly on cell division, Cell division provides the bases for one form of growth for both sexual and asexual reproduction, and for transmission of hereditary qualities from one cell generation to another. the division of the cells include two types:

#### 1- nuclear division (karyokinesis)

#### 2- cytoplasmic division (cytokinesis)

the nuclear material of the living body cells both somatic and reproductive cells requires division before the division of the cytoplasm, therefore there are two types of nuclear division

- 1- mitosis
- 2- meiosis

In the multicellular organisms there are two types of the nuclear division: -

1- Mitosis >>> a nuclear division occur in the somatic cells.

2-Meiosis >>> occur in gametes (sex cells).

In human there are 46 chromosomes (23 pairs); 22 pairs consist of homologous chromosomes called the autosomes chromosomes or somatic chromosomes.

The last pair (2 chromosomes, XY) are called the sex chromosomes



#### **Structure of chromosome**

In the nucleus of each cell, the DNA molecule is packaged into threadlike structures called chromosomes. Chromosomes are not visible in the cell's nucleus not even under a microscope when the cell is not dividing. However, the DNA that makes up chromosomes becomes more tightly packed during cell division and is then more visible under microscope.



A-The structure of chromosome

B- karyotype of human genome

# The cell cycle

- The cell cycle is an orderly sequence of events that occurs from the time when a cell is first formed until it divides into two new cells.
- Most of the cell cycle is spent in interphase. Cell cycle consist of two stages (interphase stage and mitosis stage)
- Following interphase, the mitotic stage of cell division occurs.
- The interphase stages:
  - A. **G1 stage** cell growth (increase the size of the cell), cell doubles its organelles, accumulates materials for DNA synthesis.
  - B. **S stage** DNA synthesis occurs, and DNA replication results in duplicated chromosomes.
  - C. G2 stage cell synthesizes proteins and enzymes needed for cell division.



The cell cycle of most living organisms

- The diploid cell (2n) mean the cell contains two chromosomes of each type (somatic cells).
- The haploid cell (1n) mean the cell contains one set of chromosome for each type (sex cells).
- In the life cycle of many animals, only sperm and eggs have the haploid number of chromosomes.
- The nuclei of somatic cells undergo mitosis, a nuclear division in which the number of chromosomes stays constant.



The single and duplicated chromosome

#### - The Mitotic Stage

Mitosis: is the process in which one cell divides giving 2 daughter cells with The mitosis consists of 4 phases sometimes 5 phases

#### <u>Lab. 3</u>

#### **1- Prophase**

- Appearance of thin-thread like condensing chromosomes, and the chromosomes become more visible. each chromosome composed of two sister chromatids bind together by the centromere.
- The nucleolus disappears.
- The nuclear membrane degenerate.
- Formation of the spindle apparatus (centrioles + microtubules) and migrate to opposite pole of the cell.



**Diagram of the prophase stage** 

#### 2. Metaphase

- The chromosomes line up in the equatorial plane.
- The sister chromatids are still bind together at the spindle fibers.
- The centromeres are still undivided.
- The chromosomes are still duplicated.



Diagram of the metaphase stage

# 3. Anaphase

- The centromeres are separated, this lead to the separation of the two sister chromatids and move towards the poles.
- Each one chromosome has 2 chromatids joined together by a centromere, the separated chromosomes are called the daughter chromosomes.



Diagram of the anaphase stage

# 4. Telophase

- After ending the migration of the daughter chromosomes, two new nuclei will form at each pole of the cell.
- New nuclear membranes reform around each group of the daughter chromosomes.
- Disappear the spindle fiber.
- Reformed two new nucleoli.
- The chromosomes again become long, and recoiled.



Diagram of the telophase stage

#### <u>Lab. 3</u>

#### **5-Cytokinesis**

The cytokinesis means the division of the cytoplasm of the cells after complete the mitosis to form 2 daughter cells, with the same DNA contents of the parent cells (diploid).



Diagram of the cytokinesis stage

# **Meiosis**

Meiosis: is the division of the sexual cells to form 4 daughter cells with the half number of chromosomes (haploid cells 1N) therefore sometimes called the reduction division, as in the production of gametes and plant and fungi spores

- Meiosis includes two nuclear divisions and four haploid nuclei result.
- The meiosis consists of two stages: -
- Meiosis I & Meiosis II
- the cells before the meiosis undergo a replication process of DNA to become diploid cells with 4N instead of 2N, therefore their chromosomes called tetrad in certain time during meiosis.
- The meiosis occurs in the primary spermatocytes in the testis and primary oocytes in the ovaries.

# 1. Meiosis I Includes:

# 1- Prophase I

- It is the longest phase of meiosis; in the human female it persists in each primary oocytes from a fetal age of 12-16 weeks until ovulation during puberty in 12-15 years.
- The prophase includes the following 5 substages: -
- 1) Leptotene, 2) Zygotene, 3) Pachytene, 4) Diplotene and 5) Diakinesis.
- 1. Leptotene stage
  - The nucleus enlarges.
  - the chromatin material condenses, the chromosomes become threadlike.
  - Each chromosomes consists of 2 sister chromatids join by the centromere.



Leptotene stage

# 2-Zygotene

The homologous chromosomes (each one has 2 sister chromatids) pair or synapse, the joining chromosomes are called **bivalent**.



Zygotene stage

#### Lab. 3

#### **3-Pachytene stage**

- The 2 sister chromatids of each one of the homologous chromosomes become visible, the bivalent appears with 4 strands (chromatids) this called the **tetrad**.
- Two of the non-sister chromatids (one from mother and one from father) of the homologous chromosomes exchange some genes or portions of chromosomes, this called **crossing over** or **recombination**.



Pachytene stage

#### **4-Diplotene stage**

The paired homologous chromosomes begin to separate, but not completely, because remain united at their points of exchanging the genes, these points called the **Chiasmata** (single= chaisma).

**Chiasmata:** - the sites or points, at which exchange of genes or segments of chromosomes has occurred during crossing over.

#### 5- Diakinesis stage

- This is the final stage of the prophase I, the chiasma moves to the end of the chromosome.
- The nucleolus disappears
- The nuclear membrane degenerate.



Diplotene & Diakinesis stage

# 2. Metaphase I

The synapsed chromosomes line into the metaphase plate of the cell.



# 3. Anaphase I

- The homologous chromosomes are completely separate from each other and move towards opposite poles of the cell.
- The centromeres that hold the 2 chromatids of the one chromosome do not divide as they do in mitosis. Therefore, the 2 chromatids do not separate and move together to the poles of the cell.



Anaphase I

#### 4. Telophase I

- The nucleolus and nuclear membrane are formed.
- The cytokinesis occurs resulting 2 daughter cells, but each one contains one chromosome from each homologous pair (2n,2c) this mean the nuclei contain double amount of DNA, but one chromosome of the bivalent.



**Diakinesis I** 

#### 2. Meiosis II

The same four phases seen in meiosis II – prophase II, metaphase II, anaphase II, and telophase II. The period or time between meiosis I and meiosis II is called Interkinesis.

#### Lab. 3

No replication of DNA occurs during interkinesis because the DNA is already duplicated.

# 1. Prophase II

- The chromosomes condense again.
- The nuclear membrane degenerate.
- Form the spindle fibers.



prophase II

# 2. Metaphase II

The diploid number of chromosomes, each consisting of 2 chromatids joined at the centromeres are arranged in the metaphase plate.



metaphase II

# 3. Anaphase II

The centromere which bind the sister chromatids separate, the sister chromatids move to the opposite pole as chromosomes.



Anaphase II

# 4. Telophase II

- Haploid set of the chromosomes at two poles uncoil and form the chromatin.
- The nucleolus reappears.
- Nuclear envelope reforms.
  - Also after the Telophase II the cytokinesis occur, resulting 4 daughter cells each one has 1 chromatid, and haploid number of chromosomes.



**Telophase II** 

# Most important differences between mitosis and meiosis

Mitosis	Meiosis
1-mitosis occurs continuously in the body or somatic cells.	1-meiosis occurs in the germ cells during the process of gametogenesis
2-the whole process complete in one sequence after one round of DNA replication.	2-the whole process complete in two successive division which occur one after the other.
3-the prophase is short duration and does not include any sub stages	3-the prophase is longer duration and it complete in six successive stages.
4-the homologous chromosomes duplicate into two chromatids, the chromatids separate and form new chromosomes.	4- only one type of chromosome either maternal or paternal moves to the daughter cells.
5-no synapse take place between the homologous chromosomes.	5-paining or synapse occurs between homologous chromosomes.
6-duplication of chromosomes take place in the early prophase.	6-duplication of splitting of chromosomes take place in late prophase.
7-no Chaismata formation or crossing over take place.	7-chiasmata formation in crossing over process take place during meiosis I
8-no exchange of the genetic material between the homologous chromosomes.	8-the exchange of the genetic material take place between the chromatids of homologous chromosomes
9-the chromosome number remain the same in the newly formed daughter cells.	9-chromosome number is reduced into half in the newly formed daughter cells.
10-two daughter cells are formed for each one parent cell	10-four daughter cells are formed for each one parent cell

# Gametogenesis

Gametogenesis is the process by which germ cells are converted into highly specialized sex cells (gametes) that are capable of uniting at fertilization and producing a new individual.

Gametogenesis consists of two types:

#### 1) Spermatogenesis in a male.

2) **Oogenesis** in a female.

There are two types of gametes:

1- male gametes (sperm).

2- female gametes (ovum).

### Primordial Germ Cells(PGCs)

Both male and female gametes are derived from the primordial germ

cells(PGCs) during the embryogenesis.

Primordial germ cells (PGCs) are the embryonic precursors of the gametes that are formed in the epiblast and that move to the wall of the yolk sac and then migrate from the yolk sac toward the developing gonads (ovaries or testes). Mitotic divisions increase their number during their migration.

# **Spermatogenesis**

Spermatogenesis is the process by which stem cells (PGCs) develop into mature spermatozoa (sperm) in the male genital system during the embryonic development. There are two phases:

- (1) Spermatocytogenesis includes Mitosis and Meiosis
- (2) Spermeiogenesis
- We study the spermatogenesis in cross sections of human testis
- The human testis consists of many lobes opened in one end into the vas deferens or sperm ducts.

• Examine the slide of C.S. in the human testis and notice the following cells during Spermatocytogenesis process:

1) Spermatogonium (plural=spermatogonia)

- have spherical or oval nuclei and rest on the basement membrane of the seminiferous tubules
- Undifferentiated germ cells.
- Derived from the mitotic division of the PGCs.
- Large-sized and rich in chromatin.
- Contain duplicating chromosomes Diploid cells (2n).
- Each one enters many divisions of mitosis to form two daughter cells the primary spermatocytes.

#### 2) Primary Spermatocytes

- Bigger than the spermatogonia
- Locate beyond the spermatogonia
- The nucleus in the center of the cytoplasm.
- Contain duplicating chromosomes Diploid cells (2n).
- Do not enter another mitosis but enter the **meiosis I.**
- These cells develop into 2 haploid secondary spermatocytes (1n)

#### 3) Secondary Spermatocytes

- Smaller than the primary spermatocytes.
- Locate next to the primary spermatocytes
- The nucleus in the center of the cytoplasm.
- Enter the **meiosis II**.
- Give 2 haploid **spermatids** (1n).

# 4) Spermatids

- Spherical small (round) cells.
- The nucleus not in the center of the cytoplasm.
- Some of them have a **tail bud.**
- Not capable of functioning as male sperms.
- They are Haploid cells (1n).
- Don't inter another divisions
- Inter another process called Spermeiogenesis.

# Spermeiogenesis

Spermeiogenesis is the metamorphosis of spherical **spermatids** into elongated **spermatozoa** (**sperm**). No further mitosis or meiosis takes place. During this process the following events occur:

- Formation of the acrosome.
- Formation of the flagellar apparatus.
- Most excess cytoplasm (the residual body) is separated and left in the Sertoli cell.
- Spermatozoa are released into the lumen of the seminiferous tubule.
- A small amount of excess cytoplasm (the cytoplasmic droplet) is shed later in the epididymis.

# 5) Spermatozoa (Sperm)

- Very active and mobile cell.
- Found near the cavity of the seminiferous tubules.
- Long head, thin and dark body, long tail.





The typical spermatozoa (sperm) in human.

We will study two models; human & Ascaris male
 Examine and draw the C.S. in seminiferous tubules in human mature male
 Notice the characteristics of the all spermatocytes in the lumen of the
 seminiferous tubule



Examine and draw the L.S. in *Ascaris* mature male
 Study all the structures with labels as in the chart in your Lab.

> Examine human sperm smear under the light microscope.

### Exercise inside Lab.

Complete the diagram of spermatogenesis below in mature human male with writing the following items inside the diagram:

- 1- Name of each cell?
- 2- Diploid or haploid cell with no. of N?
- 3- Type of division (mitosis or meiosis)?
- 4- How many sperms are formed from one cell in spermatogenesis?



#### **Oogenesis**

Oogenesis is a process of growth and development of the germ cells in the female genital system to produce mature cells during the embryonic development and after puberty. We study the Oogenesis in the C.S. of the reproductive system of *Ascaris* female because of the 2 large chromosomes as model for oogenesis in animals, female *Ascaris* has two ovaries. It is long, thin and coiled tube-like structures, the oocytes do not begin meiosis until the sperm touches and penetrate it in contrast to mammals, where meiosis is completed partially before fertilization.

• Examine the unfertilized egg of Ascaris



The process of oogenesis includes the following stages:

- Examine the diagnostic characteristics and draw each cell
  1) Oogonium (2n, 2c)
- a- Developed from the germinal zone of the ovaries
- b- Small cells arrange around ovary as wheel structure (rachis)
- c- Each cell has a cone shape, the up toward the rachis.
- d- Small nucleus near the bottom of the cell.
- e- Has duplicated chromosomes with 2n,2c
- f- Enter the mitosis to produce the primary oocytes

#### 2) Primary Oocyte (2n,2c)

We can study this cell in the cavity of the ovary

- a- Larger than the oogonium.
- b-After penetration of sperm it enters the meiosis I
- c- After the sperm enters the oocyte, each homologous chromosome binds together and each one consists of 2 chromatids, so this called the tetrad\_near the surface of the oocyte.
- d-after complete the meiosis I, produce 2 daughter cells, one is the secondary oocyte (1n) and the other is the first polar body (1n) attaches to the inner surface of the cell inside perivitelline space
- Examine C.S. in the oviduct of *Ascaris* under the light microscope to notice the primary oocyte.



#### Lab. 4

# 3) Secondary Oocyte (1n, 2c), examine in the uterus of the Ascaris female

- Enter the meiosis II to produce 2 daughter cells, one is the mature ovum (1n) and the second is the second polar body(1n)
- The sperm is still in the cytoplasm of this cell.
- The first polar body attaches to the inner surface of the secondary oocyte.
- The sperm is in the center of the cell.
- Examine C.S. in the uterus of *Ascaris* under the light microscope to notice the secondary oocyte.

# 4) mature ovum (1n,1c)

- A cell contains 2 chromosomes only (1n).
- The first and second polar bodies appear at the inner surface
- The sperm is in the center of the cell.



# 5-Male and Female Pronuclei (zygote)

- This cell contains both the male and female nuclei at the center of the ovum.
- At the final stage of this process the 2 nuclei will fuse to form the zygote and the number of chromosomes retains 2N.
- The male pronucleus is larger than the female pronucleus.
- The zygote enters the cleavage (mitosis)
- Examine C.S. in the uterus of *Ascaris* under the light microscope to notice late cleavage of zygote



# Exercise inside Lab.

Complete the below diagram of oogenesis in mature human female with writing the following items:

1-Name of each cell?

2-Diploid or haploid cell with no. of N?

3-Type of division (mitosis or meiosis)?

4-How many polar bodies are formed from one cell in oogenesis?


## **Early Embryogenesis of Amphioxus**

#### **Classification of Amphioxus**

Kingdom: Animalia Phylum: Chordata Subphylum: Cephalochordata Species*: Branchiostoma lanceolatum* 



Development of *Branchiostoma* (Amphioxus) plural amphioxi, or amphioxuses also called lancelet, is indirect because it has a larval stage during late embryogenesis, the transformation of egg, which having less yolk, into a complex and differentiated animal is easier to follow than in any other vertebrate. The early development of *Branchiostoma* is of great phylogenetic significance because it resembles with those of invertebrates like Echinodermates on one hand and vertebrates on the other. Amphioxus have separate sexes and the fertilization is external.

Amphioxus lives buried in sand seafloor of the shallow and coastal waters. This animal shows 26 -28 pairs of gonads along the body at the both side without ducts. When mature the overlapping tissue of the gonad will rupture and the gametes are liberated into atrium of amphioxus. They will come out into the water through the atriopore during their spawning season.

## **Organization of the Gametes:**

The primordial germ cells PGCs of testes or ovaries undergo spermatogenesis or oogenesis to produce sperms or eggs.

#### Spermatozoa in Amphioxus:

- ✤ The mature male sex cell called Spermatozoa.
- It is 15 to 20 μm in length and shows typical flagellate spermatozoa with three regions: spherical head, very short middle piece and long tail.



Amature spermatozoa of amphioxus

#### **Ovum in Amphioxus**

The unfertilized ovum or secondary oocyte of Amphioxus is 0.10 mm - 0.12 mm in diameter. The cytoplasm has little amount of yolk granules except in the cortex of the egg and around the nucleus, therefore the egg type of the amphioxus called **microlecithal egg**. Around plasma membrane surrounds the cytoplasm there is a

mucopolysaccharide membrane it is called vitelline membrane. In between these two layers (plasma membrane & vitelline membrane) the perivitelline space is present. However, the amount of yolk in the ooplasm is few so sometimes its ovum called **isolecithal eggs** but the spread of the yolk granules in the vegetal pole are more than in the animal pole, therefore the egg of the amphioxus called **telolecithal egg**. The nucleus is present towards the **animal pole**, whereas the opposite pole is called **vegetal pole**. The vegetal pole becomes posterior-dorsal side of the embryo. The Animal pole becomes anterior-ventral side of the embryo. Hence a gradient polarity is established in the egg.



An unfertilized ovum of amphioxus

## **Fertilization in Amphioxus**

Fertilization in amphioxus is external, taking place in the surrounding sea water where eggs and spermatozoa are shed. Before fertilization, the ovum has outer thin vitelline membrane enclosing a peripheral cytoplasmic layer, central yolky cytoplasm mainly towards the vegetal pole and a fluid-filled germinal sac or nucleus towards the animal pole.

As soon as the egg comes in contact with water the vitelline membrane will separate from the plasma membrane. The egg is surrounded by a number of sperms. One sperm will make its entry through the contents of the egg from the vegetal pole. At this time a number of changes take place in the corticoplasm. The egg nucleus undergoes second maturation division. Second polar body is pushed into the pervitilline space. Both the male and female pronuclei will unite a zygote nucleus is formed.

1- Examine a preserved specimen (w.m) of the adult amphioxus, and notice the gonads through the body wall. The gonads have a cuboidal shape, 26-28 pairs along the body at the both sides, the male and female are similar morphologically.

## 2- Examine C.S. in female gonad (ovary) and notice:

The ovary locates between the atrium and the body wall

• Notice the sections in the ovary and notice oocytes in different growth stages



## **3- Examine C.S. in male gonad (testis)**

The testes locate between the atrium and the body wall

• The testis contains high number of small cells that called the spermatocytes, which produce the sperms.

### 4- Examine the zygote that represent one-cell stage

\*Draw with label microscopic slide and plaster model in your lab.



# **Cleavage of the Amphioxus**

The egg of amphioxus is **isolecithal** and the cleavage is of **holoblastic** type means cytokinesis completely separates. About one hour after fertilization, the egg is divided into 2 blastomeres.

## Now let us study and examine the stages and planes of the cleavage of the

## **Amphioxus zygote**

## • The stages and plane of the cleavage: -

**1.** The first cleavage **the 2-cell stage:** is meridional plane cuts the egg produce 2 equal sized blastomeres (equal holoblastic cleavage).



**2.** The second cleavage **the 4-cell stage:** is meridional plane and produces 4 equal sized blastomeres (equal holoblastic).

**3.** The third cleavage, **the 8-cell stage:** is transverse (horizontal) cleavage, produce 8 unequal sized blastomeres; 4 small blastomeres are called **micromeres** at the animal pole, and the other 4 larger are **macromeres** at the vegetal pole.

**4**. The fourth cleavage, the **16-cell** stage: is meridional cleavage, produce 16 unequal sized blastomeres **,8** are **micromeres**, and the other **8** are **macromeres**.



**5.** The fifth cleavage, **the 32-cell stage:** is produced by the transverse (horizontal) cleavage produce 32 blastomeres arranged in four tires with graduation in size. At the beginning of the seventh cleavages the arranges of division are decline become arithmetically rather than geometrically.

#### 6. Morula stage

The blastomeres constitute the embryonic stage called, Morula is an early stage embryo consisting of blastomeres arranged in a solid ball.





Morula more than 32-cell in amphioxus.

#### 7. Blastula stage

The blastula is a hollow sphere of blastomeres has special characteristics:

- inner fluid-filled central cavity called the blastocoel formed during an early stage

of embryonic development in animals

-the blastomeres are arranged in a single layer, called the blastoderm.

## \* Draw the microscopic slide with label the blastomeres and blastocoel.



Blastula stage in amphioxus

# **Embryogenesis of frog**



## Introduction

The frog has been historically being used as an amphibian animal model of development due to the ease of observation from the fertilized egg through to tadpole stage. The later metamorphosis of the tadpole to frog has also been studied for hormonal controls and limb development.

#### The classification of frog

Kingdom: Animalia Phylum: chordate Subphylum: vertebrate Class: Amphibia

# Reproduction

- In frog the sexes are separate.
- The fertilization is external (oviparous female) and occur in the water outside the body of female.
- Frogs lay eggs in water the eggs can hatch in a few days to a few weeks convert to tadpole during a process called metamorphosis



Frog Life Cycle

- In male, the testes are paired and located in the coelomic cavity near the kidney.
- Histologically, the testes are composed of numerous of fine convoluted tubules, called seminiferous tubules.
- These seminiferous tubules which are lined by germinal epithelium which includes spermatogonial cells and sertoli cell.
- This spermatogonial cells undergo the spermatogenesis and produce spermatozoa.
- In female, gonads are two ovaries, attached to the dorsal side of the body cavity, they are multi-lobed, each lobe of the ovary is hollow.
- The ovaries different in size depending on the time of the year, during the breeding season (summer) the ovaries become large and after ovulation in spring they become small.
- Histologically, the ovaries consist of oocytes, each oocyte surrounded by a single layer of flattened cells called the follicle cells.

- The follicle cell produces a non-cellular vitelline membrane around the oocyte.
- The oogenesis in amphibians like those of lower vertebrate, produce of large number of eggs at one time during the year.

# The eggs

- In the frogs, the oocyte undergoes oogenesis and the secondary oocyte will be ovulation in to the oviduct.
- The mature eggs are highly pigment on the surface of the animal pole, and white yolk in the vegetal pole. So the ovary has appearance as a black.
- During the development of eggs in the oviduct the eggs surround by noncellular and transparent vitelline membrane, probably derived from both ova the follicle cell.
- After the egg is fertilized this membrane become separated from the egg and the space become between is then known as periviteline space, filled with a fluid.
- During movement of eggs in the oviduct, many layers of jelly called tertiary egg membrane will be adding around the eggs, the jelly membranes swollen in the water.

# The functions of the jelly membranes are:

**1-** They serve to bind the eggs to each other to protect them from the flow of water.

**2-** The jelly layers protect the eggs from the effect of the heat, radiation of the sun rays.

**3-** They protect the eggs from mechanical injures and make the eggs bad taste to the aquatic animals.

**4-** The jelly layers help the eggs to float on the surface of water, jelly contains air bubbles which help in respiration of the ova.

- The amount of the yolk in frog eggs increased gradually and accumulates at on pole (vegetal pole) lead to pushing the nucleus at one pole (animal pole), therefore; the eggs of the amphibians are **mesolecithal** and **telolecithal eggs**.
- The pigment granules are found in the animal pole more than in the vegetal pole.
- The nucleus of the egg called **germinal vesicle**.
  - \* Draw the unfertilized egg of frog under light microscope



## Fertilized egg (zygote)

- After the spawn (a cluster of frog eggs stick together at a form called spawn) transport outside in the water, the male excretes a suspension of sperms over the eggs to sexual copulation occur.
- The frog spermatozoon always enters to the egg in animal hemisphere.
- Formation (zygote) one-cell stage.



## Cleavage

The process of cleavage or cellulation happens through repeated mitotic divisions. These divisions result in cells called blastomeres. In later stages of development, the blastomeres occupy different regions and differentiate into several types of body cells.

## The planes of the cleavage:

Depending on the position of the cleavage furrow the planes of cleavage are named.

## 1-The first cleavage ...... <mark>2-cell stage</mark>

- The plane of cleavage is named meridional cleavage. It cuts the egg through its median animal-vegetal polar axis and results in two equal-sized blastomeres. therefore, its type called equal holoblastic cleavage
- The results are 2 blastomeres
- Draw this stage:

## 2-The second cleavage ...... 4-cell stage

- Like the first cleavage, equal holoblastic cleavage
- Meridional plane of cleavage.
- The results are 4 blastomeres
- Draw this stage:



## 

- unequal holoblastic cleavage.
- latitudinal plane of cleavage but not in the equatorial of the egg near to the animal pole more than the vegetal pole
- The results are 8 blastomeres, therefore the cells in the animal pole called micromeres, and the cells in the vegetal pole called macromeres.
- Draw this stage:



## 

- unequal holoblastic cleavage, divide yolkless micromeres more rapidly than yolk-rich macromeres
- Meridional plane of cleavage.
- The results are 16 blastomeres
- Draw this stage:



# Blastula stage

- After **4-5<sup>th</sup> cleavage stages** a small space called the blastocoel appears between the blastomeres of the morula near the animal pole.
- Non-center blastocoel but it near to the animal pole.
- The blastomeres arrange themselves into an epithelium called blastoderm.
- The blastoderm over the blastocoel are two-cell thick and small with outer layer contain a black pigment, these cells are the micromeres (animal pole).
- the blastoderm under the blastocoel are big and rich in yolk called the macromeres (vegetal pole).
- Draw this stage:



## Gastrula stage

The process of gastrulation is a continuous activity succeeding, cleavage. During this process the blastodermal cells begin to move. They wander and occupy their prospective organ forming zones. During this movement at one region on the blastula, the cells wander inside and occupy the blastocoelic cavity

- The process of gastrulation in frog is more complicated than Amphioxus.
- Examine the L.S section in early gastrula and notice the following
- a small cleft under the equatorial region of the embryo near the vegetal pole, this cleft called the blastopore.

## Draw this stage (early gastrula)



# Late gastrula

- At a specific region below the equator the blastoderm cells move toward the interior of the blastula. As the cells move further inside, an invagination happens. The blastopore become deeper to form the gastrocoel or archenteron inside The opening of the archenteron on the surface of the blastula is called the blastopore.
- The endodermal and mesodermal cells inter the gastrula through the blastopore into the gastrocoel until the blastopore closed by an endodermal plug this called the yolk-plug.
- The roof of the yolk-plug represents the dorsal lip of the blastopore.
- The floor of the yolk-plug represents the ventral lip of the blastopore.

- The blastocoel contracts until the end of the gastrulation, it recognized only as a narrow slit.
- Draw this stage (late gastrula)

# Organogenesis:

- Neurulation process (neural tube formation)

Neurulation is the process of neural tube formation, the dorsal side of the gastrula is lined by ectodermal cells. The presumptive area of the nervous system gets differentiated from the rest of ectoderm

Stages of neurulation:

#### A- Neural plate stage

The mid-dorsal part of the ectoderm become flattened and differentiate from the rest of the ectoderm to form a neural plate.

• Draw this stage:



## **B-** Neural folds

The edges of the neural plate become thickened and grow up called neural folds and between them there is a groove called a neural groove.

• Draw this stage:



# **B-** Neural tube

- The neural folds meet each other in the midline and fuse together and form a tube called the neural tube.
- The epidermal ectoderm of each side also meets and fuses at the midline over of neural tube.
- The embryo in this stage called the neurula.
- The neurula is the stage of the neurulation in the embryo.
- Draw this stage:



# Tail bud stage

- With the progress of development of the embryo of the frog, the embryo begins stretch in length in the posterior part of the body and this elongation become the rudiment of the tail, called **tail bud stage**.
- In this stage the Length of the Frog larva is 3-4 mm.
- The embryo after hatching from the egg another stage of embryogenesis called the larval stage, this stage called the **tadpole**.
- Tadpole lack to mouth unable to feed, and take the nutrients from yolk still present in the endodermal cells of the gut.
- The body of the embryo is elongated beyond the blastopore this region represents the tail bud.
- The blastopore later gives rise into the anal opening therefore this animal called deuterostomia or development to the mouth open this animal called protostomia.
- In this stage we study the internal parts of the embryo.
- Study the serial sagittal and cross sections through the body of the tail bud stage of the embryo.

# Draw a model of the elongated frog embryo

# I- Sagittal sections through tail bud frog embryo

We will study the organogenesis of the frog embryo from the sagittal sections in tail bud frog

## 1- Nervous system

The neural tube differentiates to a wide vesicle called the brain vesicle .

The brain vesicle later differentiates into the 3 parts:

A- Prosencephalon (forebrain)

B- Mesencephalon (midbrain)

C-Rhombencephalon (hindbrain)

Draw with label Sagittal sections in tail bud stage of frog embryo showed nerve system.

#### 2- Digestive system

- Examine the foregut, midgut and hindgut
- The hindgut end with the anus
- Examine the invagination of the stomodeum (ectoderm)
- Examine the evagination of the foregut (endoderm) called the oral evagination
- Examine the invagination of the ectoderm above called the stomodeum
  Stomodeum + oral evagination = oral plate → mouth opening
- Notice the liver diverticulum at the dorsal side of the foregut
- Draw with label Sagittal section in tail bud stage of frog embryo showed digestive system.

Draw with label Sagittal sections in tail bud stage of frog embryo showed liver diverticulum.

# **II-Cross section through tail bud frog embryo**

## 1- C.S Through olfactory placodes of tail bud frog embryo

Examine the:

- Prosencephalon, thick wall of neural ectoderm.
- Ectoderm in the outside.
- The ectoderm becomes thicker at the lateral side to form the olfactory pit.
- Draw with label cross section in tail bud stage of frog embryo showed olfactory placodes.



## 2- C.S through optic vesicles of tail bud frog embryo

Examine the: -

- Mesencephalon.
- The two optic vesicles are arising from the lateral sides of the prosencephalon forming a cup-shape structure called the optic cup.
- Later the optic cup forms the optic vesicles.
- Draw with label cross section in tail bud stage of frog embryo showed optic vesicles.



## 3-C.S. through auditory vesicles of tail bud frog embryo

Examine the: -

• Examine the rhombencephalon, thick lateral sides but thin in the roof.

- The notochord under the rhombencephalon.
- The auditory vesicles locate at the lateral sides of the rhombencephalon.
- Draw with label cross section in tail bud stage of frog embryo showed auditory vesicles.

# 4- Cross section through heart of tail bud frog embryo

Examine the: -

- The heart is under the pharynx.
- The heart consists of 2 layers, the internal is thin called the endocardium and the outer is thick called the myocardium and intermediate pericardium.
- Draw with label cross section in tail bud stage of frog embryo showed heart.



## 5- C.S through midgut and liver of tail bud frog embryo

- Notice the spinal cord
- Notochord is under the spinal cord
- Intermediate mesoderm which form later the pronephric tubules
- Midgut is under the notochord
- Liver diverticulum is under the gut
- Draw with label cross section in tail bud stage of frog embryo showed midgut and liver.

## 6- Cross section through hindgut of tail bud frog embryo

Examine the: -

- Notice the spinal cord.
- Notochord under the spinal cord.
- Hindgut under the yolk.
- Draw with label cross section in tail bud stage of frog embryo showed hindgut.



## 7- Cross section through tail bud of tail bud frog embryo

Examine the: -

- Notice the small spindle shape.
- The final part of the spinal cord.
- The final part of the notochord.
- Mesoderm along the sides of the spinal cord and the notochord.
- Draw with label cross section in tail bud stage of frog embryo showed tail bud.



## **Gastrulation in amphioxus**

During gastrulation, cell movements result in a massive reorganization of the embryo from a simple spherical ball of cells, the blastula, into a multi-layered organism. During gastrulation, many of the cells at or near the surface of the embryo move to a new, more interior location. The primary germ layers (endoderm, mesoderm, and ectoderm) are formed and organized in their proper locations during gastrulation. Endoderm, the most internal germ layer, forms the lining of the gut and other internal organs. Ectoderm, the most exterior germ layer, forms skin, brain, the nervous system, and other external tissues. Mesoderm, the middle germ layer, forms muscle, the skeletal system, and the circulatory system.

#### • Early Gastrula

The blastomeres at the vegetal pole are big, therefore they begin to invaginate inside the blastocoel toward the animal pole. A new cavity appears called the gastrocoel (archenteron)

#### • Late Gastrula

The invagination of the cells at the vegetal pole has complete, the blastocoel disappears and the formation of the gastrocoel has complete. The external opening of the gastrocoel is called the blastopore. The embryo in this stage consists of two layers, the external layer is called the ectoderm, and the internal layer called the endoderm.

\*Draw with label microscopic slide and plaster model of early & late gastrula mark the three germinal layers and the gastrocoel



## Early Embryo

The embryo becomes long and flat at the dorsal side and concave at the ventral side.

The anterior side of the embryo is recognized from the anterior neuropore.

The anterior neuropore leads to a narrow canal called the neurocoel (neural canal) along the dorsal side of the body.

Notice the cavity of the archenteron that gives rise to the intestine.

Notice the notochord over the intestine and down the neural canal.

## \*Draw plaster model of the early embryo of the amphioxus

## Organogenesis

After the completion of gastrulation, the embryo enters into organogenesis stage this is the process by which the ectoderm, mesoderm, and endoderm are converted into the internal organs of the body.

## 1- Formation of the Neural Tube (Neurulation)

The process of Neurulation consists of 3 stages:

a- **Neural plate stage:** The ectoderm is flattened and thickening along the dorsal region of gastrula and form a plate called the neural plate.

b-**Neural folds stage:** The neural plate sinks below the level of the remainder of the ectoderm, the lateral edges of ectoderm grow above the neural plate as folds the cavity between them is called **the neural groove.** 

c- **Neural tube stage:** The neural folds are fusing over the neural groove and the neural plate rolls itself into a hollow tube called **the neural tube** and the cavity of the tube is called **the neurocoel (neural canal)** which forms in the future the central canal of the neurous system in the adult amphioxus.





# 2- Development of mesoderm and coelom

mesodermal bands separated from the endoderm by dorsal evagination into the residual blastocoel. The lateral part of mesoderm form segments with cuboidal masses along the length of the animal of cells called **the mesodermal pouches** on both dorsal sides of the endoderm tube then developed to **mesodermal** 

## \* Examine the Plaster Models to notice the mesodermal pouches and sacs



The mesodermal sacs grow down between the ectoderm & the endoderm and join under gut, the mesoderm become two layers and the space between is called the coelom

## \* Examine the Plaster Models and mark the two layers of mesoderm and coelom

In the amphioxus the mesoderm develops into 2 regions, each region will give certain derivatives as below:

### A. Dorsal Mesoderm B. Lateral Mesoderm

## **A- Dorsal Mesoderm**

The dorsal part of the mesoderm gives the somites. These somites will differentiate into **3** regions

1-Myotome; grow around notochord and give the muscles of the body

2- Dermatome ; grow under the epidermis posterior to myotome give the dermis

of skin

**3- Sclerotome** ; grow around neural tube differentiate into skeletal sheath of connective tissue

## \* Examine the Plaster Models and mark all the layers of dorsal mesoderm

#### **B-** Lateral mesoderm

After the mesodermal somites grow ventrally they differentiate into two parts The part which remains near the epidermal ectoderm is called the **somatic (parietal) mesoderm**. While the part which remains closely associated with endoderm is called **visceral (splanchnic) mesoderm**, the coelom is between them.

\* Examine the Plaster Models and mark all the layers of lateral mesoderm



## **3-Development of Notochord**

Side by side of neurulation the notochord begins formed is an embryonic structure common in all members of the phylum Chordata. The mesoderm and gut separated from each other, the notochord cells grow mid-dorsally in the roof of the archenteron these cells evaginate dorsally at the anterior end of the embryo and separated from endoderm this evagination continues slowly towards the caudal end and converts into solid, round cord below the neural tube and between the mesodermal somites.

\*Examine the plaster models to study the notochord development in amphioxus embryo

# 4- Development of endoderm and alimentary canal

After the notochord and mesodermal segments dissociate themselves from the endoderm, the free edges of the endoderm fuse together along dorsal mid-line, the endoderm becomes a closed sac; the cavity of this sac converted into the cavity of the alimentary canal which opened with the exterior by mouth and anus at later stage of development.

#### \*Draw the endoderm and alimentary canal



## **Gastrulation in amphioxus**

During gastrulation, cell movements result in a massive reorganization of the embryo from a simple spherical ball of cells, the blastula, into a multi-layered organism. During gastrulation, many of the cells at or near the surface of the embryo move to a new, more interior location. The primary germ layers (endoderm, mesoderm, and ectoderm) are formed and organized in their proper locations during gastrulation. Endoderm, the most internal germ layer, forms the lining of the gut and other internal organs. Ectoderm, the most exterior germ layer, forms skin, brain, the nervous system, and other external tissues. Mesoderm, the middle germ layer, forms muscle, the skeletal system, and the circulatory system.

#### • Early Gastrula

The blastomeres at the vegetal pole are big, therefore they begin to invaginate inside the blastocoel toward the animal pole. A new cavity appears called the gastrocoel (archenteron)

#### • Late Gastrula

The invagination of the cells at the vegetal pole has complete, the blastocoel disappears and the formation of the gastrocoel has complete. The external opening of the gastrocoel is called the blastopore. The embryo in this stage consists of two layers, the external layer is called the ectoderm, and the internal layer called the endoderm.

\*Draw with label microscopic slide and plaster model of early & late gastrula mark the three germinal layers and the gastrocoel



## Early Embryo

The embryo becomes long and flat at the dorsal side and concave at the ventral side.

The anterior side of the embryo is recognized from the anterior neuropore.

The anterior neuropore leads to a narrow canal called the neurocoel (neural canal) along the dorsal side of the body.

Notice the cavity of the archenteron that gives rise to the intestine.

Notice the notochord over the intestine and down the neural canal.

## \*Draw plaster model of the early embryo of the amphioxus
# Organogenesis

After the completion of gastrulation, the embryo enters into organogenesis stage this is the process by which the ectoderm, mesoderm, and endoderm are converted into the internal organs of the body.

# 1- Formation of the Neural Tube (Neurulation)

The process of Neurulation consists of 3 stages:

a- **Neural plate stage:** The ectoderm is flattened and thickening along the dorsal region of gastrula and form a plate called the neural plate.

b-**Neural folds stage:** The neural plate sinks below the level of the remainder of the ectoderm, the lateral edges of ectoderm grow above the neural plate as folds the cavity between them is called **the neural groove.** 

c- **Neural tube stage:** The neural folds are fusing over the neural groove and the neural plate rolls itself into a hollow tube called **the neural tube** and the cavity of the tube is called **the neurocoel (neural canal)** which forms in the future the central canal of the neurous system in the adult amphioxus.





# 2- Development of mesoderm and coelom

mesodermal bands separated from the endoderm by dorsal evagination into the residual blastocoel. The lateral part of mesoderm form segments with cuboidal masses along the length of the animal of cells called **the mesodermal pouches** on both dorsal sides of the endoderm tube then developed to **mesodermal** 

# \* Examine the Plaster Models to notice the mesodermal pouches and sacs



The mesodermal sacs grow down between the ectoderm & the endoderm and join under gut, the mesoderm become two layers and the space between is called the coelom

# \* Examine the Plaster Models and mark the two layers of mesoderm and coelom

In the amphioxus the mesoderm develops into 2 regions, each region will give certain derivatives as below:

## A. Dorsal Mesoderm B. Lateral Mesoderm

# **A- Dorsal Mesoderm**

The dorsal part of the mesoderm gives the somites. These somites will differentiate into **3** regions

1-Myotome; grow around notochord and give the muscles of the body

2- Dermatome ; grow under the epidermis posterior to myotome give the dermis

of skin

**3- Sclerotome** ; grow around neural tube differentiate into skeletal sheath of connective tissue

# \* Examine the Plaster Models and mark all the layers of dorsal mesoderm

#### **B-** Lateral mesoderm

After the mesodermal somites grow ventrally they differentiate into two parts The part which remains near the epidermal ectoderm is called the **somatic (parietal) mesoderm**. While the part which remains closely associated with endoderm is called **visceral (splanchnic) mesoderm**, the coelom is between them.

\* Examine the Plaster Models and mark all the layers of lateral mesoderm



## **3-Development of Notochord**

Side by side of neurulation the notochord begins formed is an embryonic structure common in all members of the phylum Chordata. The mesoderm and gut separated from each other, the notochord cells grow mid-dorsally in the roof of the archenteron these cells evaginate dorsally at the anterior end of the embryo and separated from endoderm this evagination continues slowly towards the caudal end and converts into solid, round cord below the neural tube and between the mesodermal somites.

\*Examine the plaster models to study the notochord development in amphioxus embryo

# 4- Development of endoderm and alimentary canal

After the notochord and mesodermal segments dissociate themselves from the endoderm, the free edges of the endoderm fuse together along dorsal mid-line, the endoderm becomes a closed sac; the cavity of this sac converted into the cavity of the alimentary canal which opened with the exterior by mouth and anus at later stage of development.

#### \*Draw the endoderm and alimentary canal



# Introduction

#### -Classification of chicken:

Kingdom: Animalia Phylum: Chordata Class: Aves Genus: Gallus Species: G. gallus Subspecies: G. g. domesticus

In a mature hen it is approximately 25 to 27 inches long. The yolk is completely formed in the ovary. When a yolk is fully developed, its follicle ruptures, releasing it from the ovary. It then enters the infundibulum, the entrance of the oviduct All of the other parts of the egg are added to the yolk as it passes through the oviduct. The chalazae, albumen, shell membranes, and shell are formed around the yolk to make the complete egg, which is then laid. This complete cycle usually requires a little more than 24 hours. About 30 minutes after the egg is laid, another yolk is released and the process repeats itself. After the fertilization the chicken embryo develops and hatches in 20 to 21 days.

# The early embryogenesis of chick and frog is different in: -

1- the egg of the chick is rich in yolk which used in all the incubation time (21 days) until hatching.

2- the embryological stages in chick are more like the mammals.

3- the development of the chick is considered direct development, this unlike the embryo of the frog.

4- Cleavage planes cannot penetrate the dense yolk so only the animal pole undergo the cleavage

5- Cleavage produces a cap of cells sitting on top of yolk- called blastoderm So the blastoderm in the chick is the equivalent of the blastula in the frog

## Reproduction

The sexes in chick are separated. The sexual dimorphism is well marked between male and female.

## Testes

- The adult rooster has testes that are paired and ovoid.
- The testes surrounded by connective tissue called tunica albuginea
- Each testis contains large amount of seminiferous tubules, that the spermatogenesis occurs in it.
- The spermatozoa of chick have typical morphology, large head, mid piece and short tail compare of human sperm.

# \* Draw a sample rooster sperm



# **Ovaries**

The reproductive system of the female chicken is in two parts: the ovary and oviduct. Unlike most female animals, which have two functioning ovaries, the chicken usually has only one. The right ovary stops developing when the female chick hatches, but the left one continues to mature The oviduct is a tube like organ lying along the backbone between the ovary and the tail. The adult hen has paired ovaries and oviducts, only one ovary and one oviduct on the left side of the body is active, the other on the right side remains degenerated, this is one of the factors of the flying adaptation.

**Oocyte** contains large nucleus and surrounded by cuboidal cell shape called **follicle cell**.

#### Eggs:

When the egg leaves the ovary at the ovulation, it is enveloped by the vitelline membrane, which is secreted by the cytoplasm of the egg.

- It is oval in shape.
- 3cm wide and 5cm long.
- Have 95% of calcium carbonate and porous to exchange of gases occur, soft and flexible in laid and soon become hard.
- The egg of the hen is macrolecithal and telolecithal. The polarity of the egg is well-marked, the animal pole having very small of active cytoplasm and nucleus in the form of disc called the **blastodisc** or **germinal disc** while the vegetal pole contains rich amount of yolk

## **Fertilization:**

The fertilization is internal, upper part of oviduct. Inside the oviduct, the spermatozoa lie in wait to fertilize the egg to active the egg to complete the meiosis II.

- The egg continues down the oviduct the shells (Albumen and calcareous) are added by the lining of the oviduct.
- After fertilization the cleavage occur inside the oviduct until the morula stage. After the fertilization the egg leave the oviduct toward the uterus the morula completes the cleavage to blastula. The egg is lay at the next day (about 18 h. after leaving the ovary).
- The development of the embryo is stop after laying until the incubation occur.



The egg of chick showed germinal disc nucleus

\*Examine un-incubated egg and note the blastoderm

- The blastoderm after fertilization is divided into 2 areas: -
- the central area is light called the area pellucida represent the site of the embryo.
- around the area pellucida there is a dark area adhere on the yolk called the area opaca.
- Draw with label w.m of blastoderm of un-incubated chick egg.



## - w.m chick embryo at 13h incubation

Examine w.m of chick embryo at 13h of incubation and notice:

- Long linear structure extended from the area opaca towards the area pellucida, this structure is called the primitive streak.
- The primitive streak is not well observed at this stage.
- The formation of the primitive streak represents the beginning of gastrulation.
- Draw with label Chick embryo at 13h incubation.

# - w.m of chick embryo at 16h incubation (primitive streak stage) Examine w.m of chick embryo at 16h incubation and notice:

- The area opaca and the area pellucida are more oval shape.
- The formation of the primitive streak is complete, therefore this stage called the primitive streak stage
- Examine the primitive streak and notice:
- Inside the middle of the primitive streak there is a groove called the primitive groove.
- The primitive groove ends with a pit called the primitive pit, at each side there are primitive ridges.

- The two primitive ridges meet to form the primitive knot (Hensen's node).
- Draw with label Chick embryo at 16h incubation (primitive streak stage).

w.m of chick embryo at 18h incubation (head process stage)
Examine w.m of chick embryo at 18h incubation and notice:

- The primitive streak become shorter and thicker
- Notice the area anterior to the Hensen's node:
- Thick plate at each side this is The neural plate.
- The head process and the notochord are under the neural plate.
- Anterior to the head process is a light area that represent the proamnion.
- Draw with label Chick embryo at 18h incubation (head process stage).

# Serial cross sections of the chick embryo at 18h incubation.

## 1- Examine cross section through neural plate and notochord notice:

- Thick ectoderm.
- Thick ectoderm represents the neural plate.
- The notochord under the neural plate and above the endoderm.
- The three germ layers are more obvious far away the neural plate.
- Very thin endoderm.
- Each side of the section is supported by the yolk.
- Draw with label C.S. of the neural plate in chick embryo at 18h incubation.

# 2-Examine C.S. through primitive groove of 18h incubation of chick embryo notice:

- Primitive pit.
- Primitive knot as a thick high layer.
- In front of the primitive knot there is the neural plate.
- Under the neural plate there is the notochord as a rode of cells between the neural plate and the endoderm.
- The three germ layers not clear at the primitive pit.
- Draw with label cross section through primitive groove of 18h incubation of chick embryo.

# **Embryogenesis of chick**

# - w.m. of Chick embryo at 24 h. incubation (4-5 pairs of somites)

- Over growth in head process and foregut
- The neural folds are more visible.
- The notochord along the primitive streak and under the neural groove.
- Notice 4-5 pairs of somites.
- Notice the area opaca: the side near the area pellucida is more dark and spotted because appearance the blood island from the mesoderm this region called area opaca vasculosa
- the outer side of the area opaca is light without blood island.
- The proamnion is free of mesoderm.
- The head process originates from the ectoderm and endoderm, grow over the proamnion.
- Examine the w.m. slide of Chick embryo of 24h. incubation

# 1-Sagittal Sections in Chick Embryo of 24h incubation:

# Examine a sagittal section in chick embryo of 24h. incubation

1- Serial Cross Sections (C.S.) through chick embryo of 24h incubation:

**a-** Examine the C.S. through the head process in Chick embryo of 24h. incubation under light microscope and notice the followings:

- The head process locates over the ectoderm of the blastoderm, separated by subcephalic pocket.
- The head process is surrounded by the ectoderm.
- At the dorsal to the head process the ectoderm forms the neural folds, between them the neural groove.
- Under the neural groove there is the notochord.
- Under the notochord there is the foregut like crescent shape.
- Notice the thickness of the splanchnic mesoderm near the endoderm of the anterior internal portal of the foregut this is represent the primary indicator of the formation of the heart in the embryo.
- Draw with label C.S. through the head process in chick embryo of 24h. incubation.

b- Examine the C.S. through the anterior intestinal portal in chick embryo of 24h. incubation.

# c- Examine the C.S. through the somites in chick embryo of 24h. incubation.

- Thick mesoderm at the lateral sides of the neural folds this represent the somites
- The somites bind with the intermediate mesoderm
- Notice the binding of the intermediate mesoderm with the lateral mesoderm which splits into somatic & splanchnic mesoderm
- The neural groove wider than at the previous section
- Notice the notochord under the neural groove
- The endoderm at this section is thin and adhere to the yolk
- Ectoderm + somatic mesoderm = **Somatopleure.**
- Endoderm + splanchnic mesoderm = **Splanchnopleur**.

**•** Draw the C.S. through the somites in chick embryo of 24h. incubation

## d-Examine the C.S. through primitive pit.

- The neural folds & neural groove disappears
- •The primitive pit at the center
- No notochord is obvious
- Difficult to notice the three germinal layers at the primitive pit region, but we can at the margins of the section
- •Notice the blood island at the area opaca at the margins of the section.

#### Draw the C.S. through the primitive pit in chick embryo of 24h. incubation