

Chapter 5

Gastrulation

5.5. Basic Concepts

During gastrulation a single wall of cells, the **blastoderm**, gives rise to the three germinal layers: **ectoderm**, **mesoderm** & **endoderm**. The presumptive mesoderm and endoderm tissues move from the exterior into the interior of the embryo. (*Presumptive tissues are those destined to become certain tissues or organs in the embryo during the course of ordinary development*).

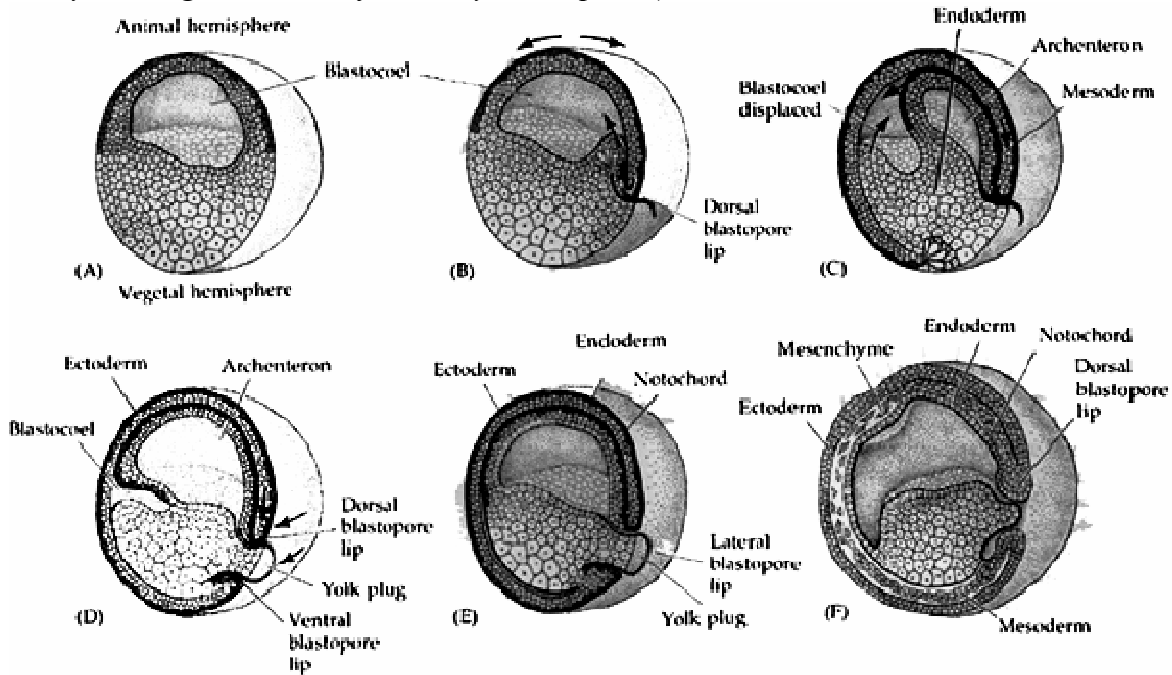


Fig.5.1. Overview of Gastrulation

(a) Fate Maps

Fate Maps are maps of presumptive tissues that can be mapped on the blastula. The presumptive tissues can be mapped by using vital dyes*, carbon particles, fluorescent proteins or by ³H-Thymidine labelling of explants, which are then implanted into an unlabelled embryo. The fates of the marked tissues are then traced during development. (*Vital dyes do not harm or change development*). By these methods it is possible to construct a diagram of the blastula with the fate of each part of the embryo marked. These are called **fate maps**.

(b) Clonal Analysis

Clonal analysis is a technique where the fate of single **founding cell** is followed as it forms a **cell clone**, which is its surviving descendants. **X-ray induced somatic crossing over** is used in *Drosophila* larvae to create a genetic marker in single embryonic cell. The descendants of that cell are then mapped on the *Drosophila* adult. Using vital dyes, Vogt (1925) constructed a fate map for an amphibian, which has three main areas, area

around the animal pole, marginal area around the equator and an area around the vegetal pole. The **animal pole** corresponds to the **future anterior** end of the embryo and the **vegetal pole**, the **posterior** end. The side where the **marginal zone is widest** is the **dorsal side** and the **opposite side** is **ventral side**.

The animal pole region is divided into the prospective epidermis and the prospective neural plate (*also called the neural ectoderm or medullary plate*) which will become the nervous system. The marginal zone is subdivided into the:

1. prospective notochord (chordomesoderm)
2. somites- which will form the segmental muscles of the body
3. lateral mesoderm which will be the mesodermal lining of the coelom and parts of the visceral organs
4. heart mesoderm
5. head endoderm will be the lining of the mouth, pharynx and gill region.

The vegetal region contains the prospective endoderm which will form the hindgut and midgut. Therefore, prior to **gastrulation** the **blastula** is arranged with the future germinal layers (**ecto-, meso- & endoderm**) on the exterior. A rearrangement of these regions is necessary to place the parts of the embryo into their proper locations. Gastrulation is the process which leads to the formation of the three germinal layers and the placement of these three germinal layers into proper relationship with each other in the embryo.

5.2. The Process of Gastrulation

There are nine basic cell movements in this process. Three cause the epithelium to expand and spread out on the surface or within an embryo: epiboly, intercalation (*radial & lateral*) and convergent extension. Three other types displace cells into the interior: invagination, ingression and involution. The organisms detailed in the book use these movements in a variety of ways.

We will be looking at gastrulation in **amphibians, chick** and **humans**.

5.3. Gastrulation in Amphibians

A small depression forms just below the gray crescent on the dorsal side of the embryo. This is the beginning of the blastopore. Involution begins at the dorsal blastopore lip and then also spreads laterally and ventrally. This results in the formation of a circular blastopore, which surrounds a mass of yolk-filled endodermal cells called the yolk plug. The lip of the blastopore above the yolk plug is the dorsal lip; the lip below is the ventral lip and those to the sides are the lateral lips. While the blastopore is encircling the vegetal endoderm, the heart mesoderm, the head mesoderm, and the chordomesoderm (notochord), in that order, are involuting over the dorsal lip of the blastopore to the inside; they then pass anterior along the interior dorsal surface. A new cavity, the **archenteron**, is forming below the involuting tissues. It expands at the expense of the blastocoele. When gastrulation is complete the archenteron will have obliterated the blastocoele. The lateral mesoderm involutes over the lateral lips of the blastopore to the interior; it will move anteriorly between the endoderm & ectoderm. The entire blastopore rotates down toward the vegetal pole during gastrulation. As the presumptive mesoderm and endoderm are leaving the exterior and passing to the interior, the ectoderm (*epidermis & neural plate*) spreads over the surface of the embryo by epiboly. These tissues will eventually cover the entire exterior of the embryo, but will not pass to the interior through the blastopore.

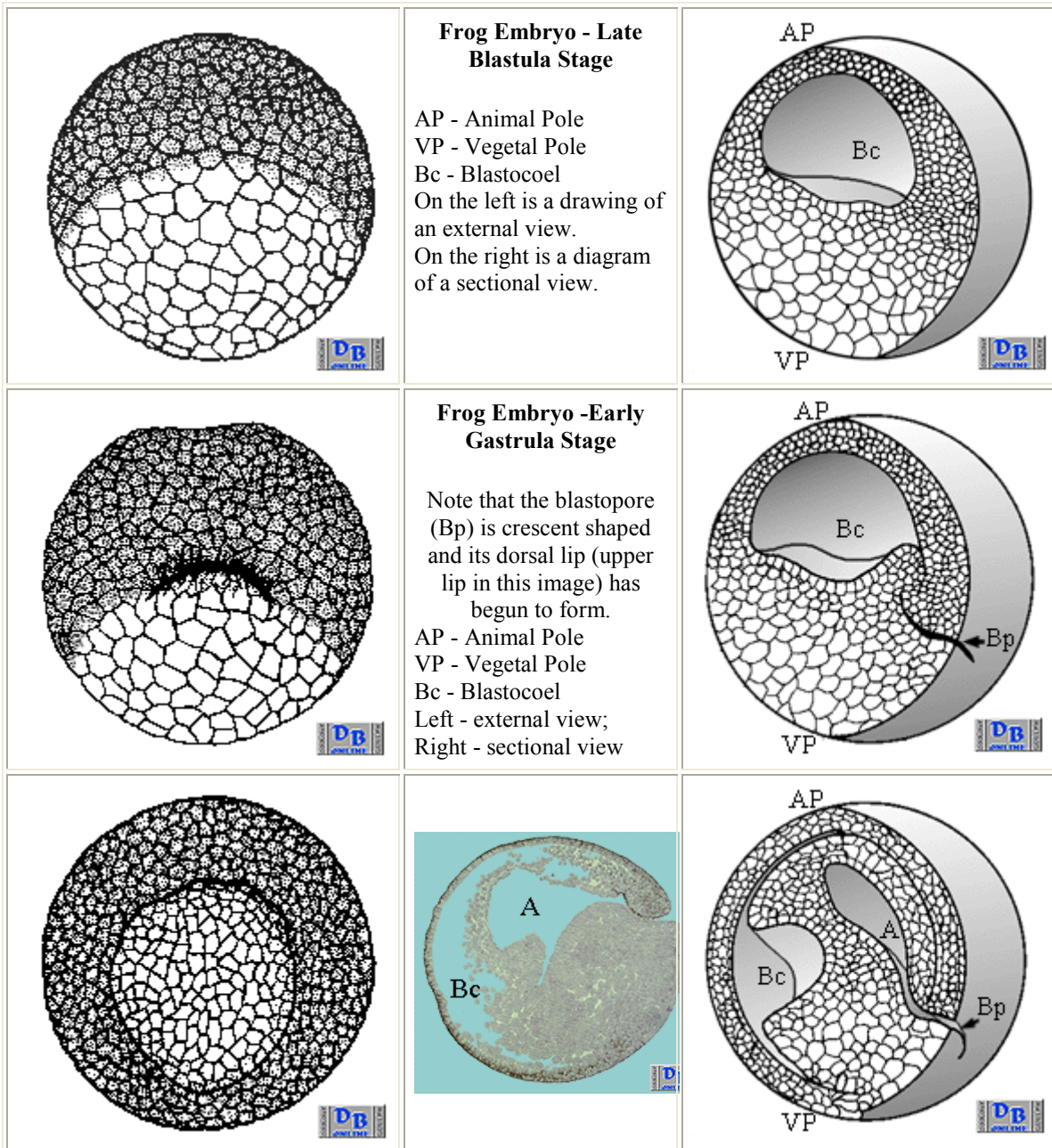


Fig.5.2a. Frog Embryo - Mid Gastrula Stage

Above Left - drawing of external view. Micromeres of the animal pole continue to overgrow the macromeres of the vegetal pole. The blastopore which first appeared crescent-shaped, becomes horseshoe-shaped, and eventually circular.

Above Center - tissue section of mid gastrula showing the formation of a new cavity, the archenteron (A) and the displacement of the blastocoel (Bc). Note that the appearance of the blastocoel is exaggerated due to shrinkage of cells during fixation and the subsequent processing of the specimen for sectioning.

Above Right - sectional diagram showing the blastopore (Bp), archenteron (A), and blastocoel (Bc). AP = Animal Pole; VP = Vegetal Pole

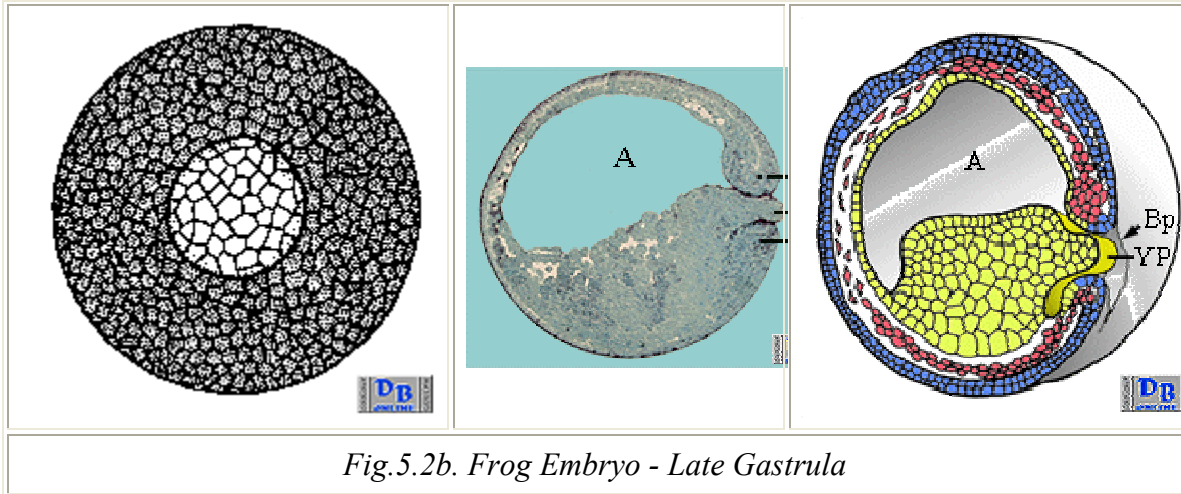


Fig.5.2b. Frog Embryo - Late Gastrula

When gastrulation is complete,

1. The prospective notochord lies on the mid-dorsal roof of the archenteron under the neural plate (which will become the nervous system).
2. The blastopore is nearly closed.
3. The endoderm lies on the floor of the archenteron.
 - a. The endoderm from the cephalic region will become the fore gut.
 - b. The endoderm from the vegetal region will become the mid and hind gut.
4. The dorsal exterior is covered with the neural plate (future nervous system).
5. The remainder of the surface is covered with ectoderm (future epidermis).

5.4. Gastrulation and Formation of 1° Organ

Rudiments in Birds (Aves)

The cells of the epiblast start converging on the posterior part of the area pellucida. As they do, they form a thickening along the midline of the epiblast, called the **primitive streak**. The primitive streak grows anteriorly along the posterior 3/5th of midline of the area pellucida. The primitive streak is complete at the 18 hour-stage.

A **primitive groove**, forms along the midline of the primitive streak and a thickening, the **primitive knot (Hensen's (node) knot)**, develops at the anterior end of the primitive streak. As the converging epiblast cells reach the primitive groove they migrate through the groove into the interior of the blastocoel and come to lie between the epiblast and hypoblast.

The cells migrate to the inside singly and not as a sheet; this type of gastrulation movement is referred to as **ingression**. The presumptive endoderm, from the epiblast, starts ingression even before the primitive streak is fully formed. These cells penetrate the hypoblast. As a result, a large part of the hypoblast around and anterior to Hensen's node is replaced by cells from the primitive streak. The presumptive notochord tissue ingresses over Hensen's node and moves straight anterior from Hensen's node. These notochordal cells can be distinguished from the other cells and are called the **head process (notochordal process)**.

The presumptive somite cells ingress through the anterior region of the primitive streak. From here, they move outward and forward to form a strip of somite mesoderm on each side of the notochordal mesoderm. The presumptive lateral plate mesoderm moves through the primitive groove in the posterior part of the primitive streak. From there these

cells ingress to the interior and then move outward and forward to each side of the somites.

Birds have *no cavity homologous* with the *archenteron* seen in amphibians. The influx of cells through the primitive groove slows and the primitive streak begins to shrink. Hensen's node is actually carried back as the primitive streak regresses. The regression is from anterior to posterior. The primitive streak is analogous with the blastopore in amphibians, in that cells on the surface, which are presumptive internal organs, migrate through the blastopore and the primitive streak to the inside of the embryo.

The regressing of the primitive streak is analogous to the closing of the blastopore. As the Hensen's node is carried backward the presumptive notochordal cells continue to ingress through Hensen's node, thus elongating the notochordal (head) process in a posterior direction. In the same manner the strips of somite mesoderm & sheets of lateral plate mesoderm are elongated, moving anterior to posterior.

As Hensen's node moves posteriorly a strip of neuroectoderm is laid down on the surface in the wake of the receding primitive streak. The strip of neuroectoderm also extends anterior from where Hensen's node was located originally. In the region of the embryo from which the primitive streak has receded, the embryo is now in three distinct principle germ layers.

1. The overlying layer is now pure **ectoderm** (a center strip of **neuroectoderm** surrounded by **epidermis** (*general body ectoderm*)).
2. The center layer is a **mesodermal** sheet, with the notochord as a central rod and lateral sheets of mesoderm.
3. The hypoblast has been replaced by the **endoderm**, which will be the lining of the gut and yolk sac.

5.5. Early Development of Mammals

The development of the mammalian embryo is unique. Within the class there is considerable variation in the details of embryology. We will not attempt a complete survey. In mammals the extraembryonic membranes are analogous in origin and function to the chick, but their development is different. The extraembryonic membranes form prior to the embryonic development, in contrast to the chick in which they develop simultaneously.

The eggs of mammals are fertilized in the upper part of the oviduct. As the zygote descends the oviduct it is beginning cleavage. As it reaches the uterus, the embryo either implants on the uterus or actually sinks into the uterine wall (as is the case in humans). This is called **implantation**. Cleavage in mammals is complete but the synchrony is lost very early.

In most mammals, cleavage leads to the formation of a **morula** (*Gk. little mulberry*). The cells of the morula become differentiated into the **inner cell mass** and the outer, **trophoblast** (*Browder: trophoctoderm*). As development continues a space appears between the inner cell mass and trophoblast, called the **blastocoele**. The embryo at this stage is the **blastocyst**. It is at this stage that the embryo implants itself into the uterine wall lining, the **endometrium**. The embryo proper will develop from the inner cell mass. The trophoblast will contribute to the chorion.

The cells of the inner cell mass further differentiate into an inner **hypoblast** (endoderm) and outer **epiblast** (ectoderm). The **yolk** (vitelline) **sac** forms by the spreading of the hypoblast cells below the blastodisc. There is no yolk in the yolk sac, only fluid. The

edges of the epiblast spread on the trophoblast forming a roof over the embryo which is the future ectoderm of the **amnion**. Below the amniotic cavity is the blastodisc which is the **embryo** proper. It has a lower **hypoblast** layer and an upper **epiblast** layer. Gastrulation and the formation of the germinal layers occurs in a similar manner as with birds. A primitive streak forms, which has a Hensen's node. Cells migrate through the primitive streak between the epiblast and hypoblast. These cells form the mesoderm. The primitive streak regresses at the end of gastrulation, as in birds.

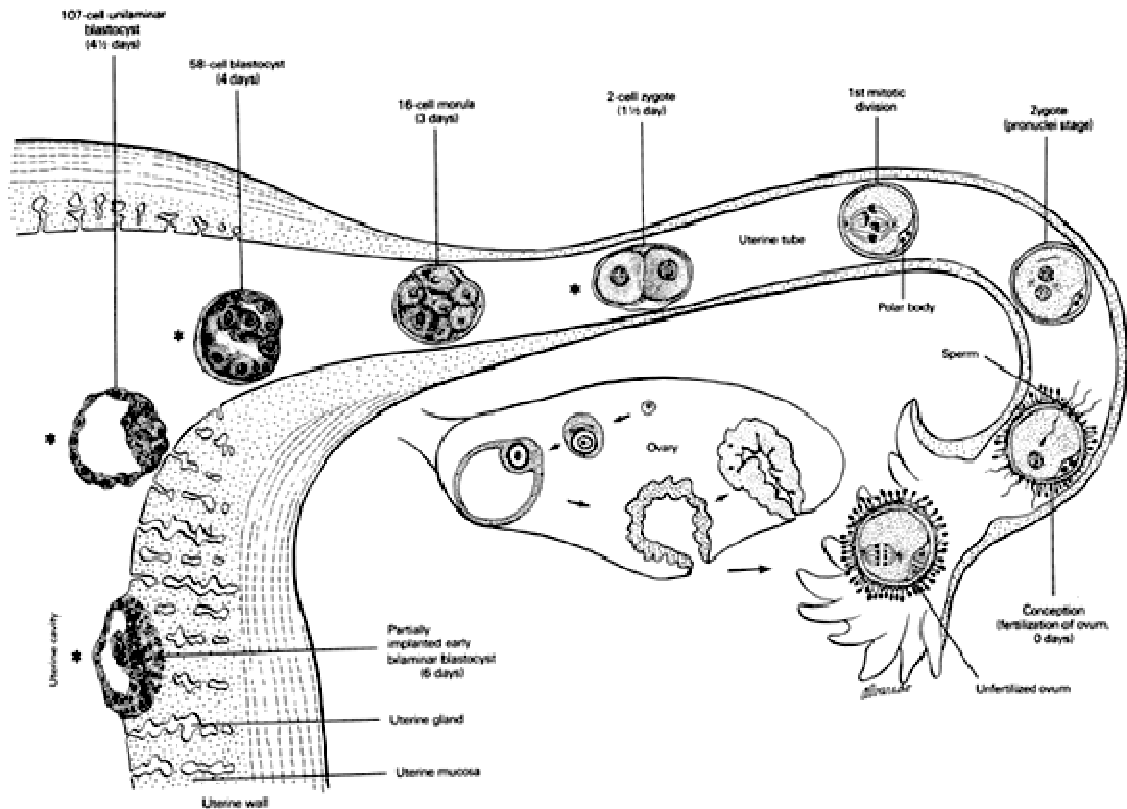


Fig.5.3. The Many Forms and Locations of the Human Embryo During the First Week

5.6. Metabolism During Gastrulation

There is a very slight overall volume increase during cleavage and gastrulation. Rapid mitosis continues during gastrulation, though at a slower rate than during cleavage. This necessitates continued synthesis and use of DNA and other nuclear components at the expense of cytoplasmic components. A sharp increase in O_2 consumption is noted during gastrulation. This increase in respiration is expected due to the increased expenditure of energy required for the morphogenetic movements. During gastrulation there is a sharp increase in protein synthesis. The newly synthesized proteins are qualitatively different from the yolk proteins and cleavage proteins. With an increase in protein synthesis, one would expect to find an increase in mRNA synthesis. This is the case, mRNA synthesis increases sharply at the approach of gastrulation. Some of the gastrula mRNA varies from some of the blastula mRNA, also qualitative difference in some of the mRNA being synthesized. Synthesis of tRNA and rRNA also increases during gastrulation.

5.7. Activity of Genes During Gastrulation

During cleavage, the cytoplasm exerted the most influence over the development of the

embryo. Enucleated fragments of eggs (which have been stimulated parthenogenetically) have been observed to develop up through the morula stage, but they cannot form a normal blastula nor undergo gastrulation without a nucleus. The nucleus plays an increasingly important role during gastrulation. Hybridization between different species demonstrates the effects that genes have during development. If the egg of one species is fertilized by sperm of another species and the pronuclei fuse, these are called true hybrids. Each blastomere has a maternal cytoplasm and a haploid set of chromosomes from each parent (maternal & paternal sets). If the two species in a hybrid cross have different rates of cleavage, it is always the cleavage rate of the maternal species which is expressed. However, the paternal genes are expressed later in development. The hybrid larvae do have intermediate characteristics between the two species. In true hybrids, any incompatibility of the sperm and egg nuclei does not become apparent until late cleavage or early gastrulation. If the hybrid is not viable this is the stage it will die.