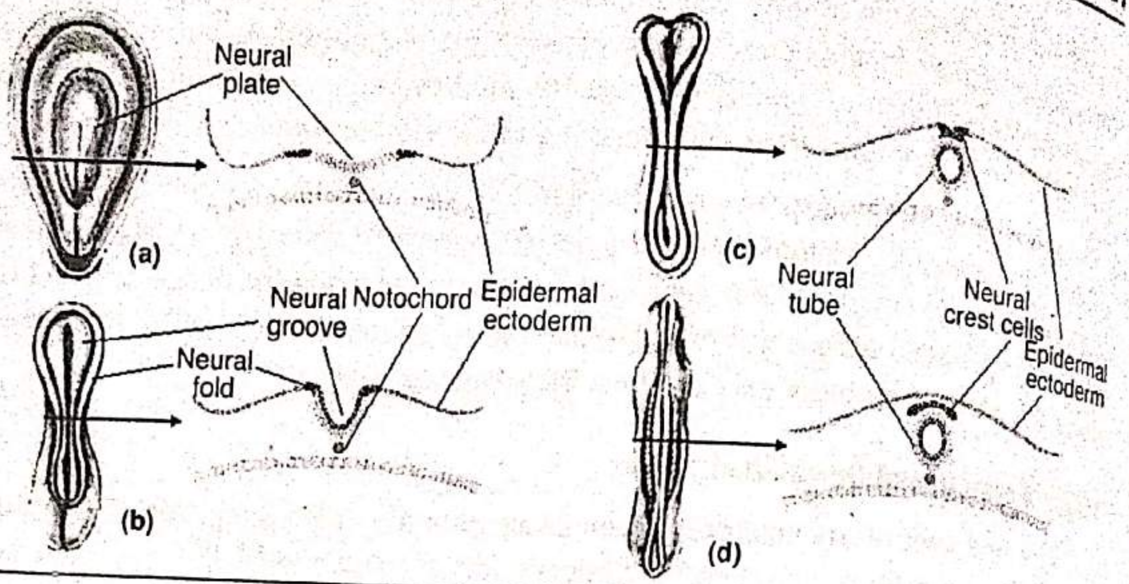

Fig. 15.9 : *Formation of neural tube in mammals involves closure of neural tube at several points.*

accumulation of a determinant on left side of the node. The nature of this determinant is obscure.

15.8 NEURULATION IN MAMMALS

Like other vertebrates, formation of neural tube in mammals from neural plate occurs through primary and secondary neurulation. Primary neurulation occurs up to sacral vertebrae level after which secondary neurulation occurs.

Formation of neural plate and bending of the plate in mammals is like birds. But closure of neural tube in mammals takes place at several points along the anterior-posterior axis. In human embryo of 22-days, closure of neural tube occurs at three points (Fig. 15.9). Closure of human neural tube requires an interaction between genetic and environmental factors. Genes like *Sonic hedgehog, Openbrain* etc. are involved in the closure of neural tube. At the same time, cholesterol and vitamin 12 (folic acid) are also essential.

15.9 PLACENTA IN MAMMALS

Placenta is associated with viviparity that is, giving birth to living young ones. When the embryo develops within the body of mother, it has reduced supply of yolk stored in the egg. Then the embryo has to maintain a functional relationship with maternal tissue for deriving nutrients from the body of mother. This functional relationship is established as an organ jointly formed by maternal and foetal tissue, called **placenta.**

From embryonic side, placenta is formed by extraembryonic membranes. Chorion completely surrounds the complex embryo and the other extraembryonic membranes. So chorion serves as interface between embryonic tissue and maternal tissue. The chorion is vascularized by the mesoderm layer of either yolk sac or allantois. In marsupials, the yolk sac vascularizes the chorion and the

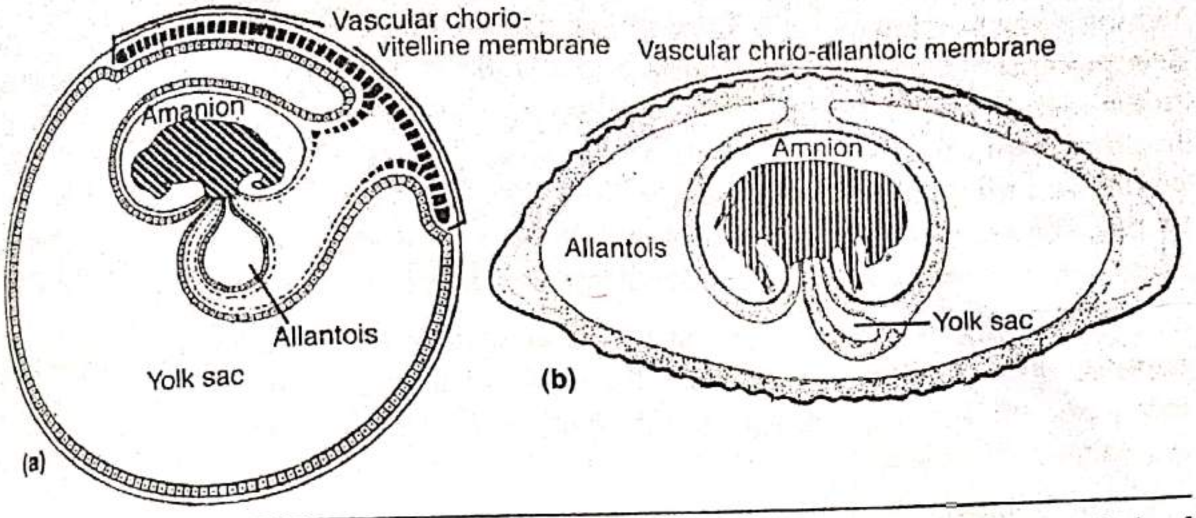

Fig. 15.10 : *According to involvement of extraembryonic membranes, mammalian placenta is of two types – (a) chorio-vitelline (opossum) and (b) chorio-allantoic (pig).*

chorio-vitelline [15.10(a)]. In higher eutherian mammals, allantois supply the vascular support to the chorion and the placenta is called **chorio-allantoic** [15.10(b)].

Chorion sends out finger-like projections called villi, which invade the maternal tissue. The distribution pattern of the villi on the chorionic sac determines the shape of placenta. On this basis, placenta are classified into following types (Fig.15.11):

1. **Diffuse:** villi are retained over the entire chorion; found in lemur, pig etc.
2. **Cotyledinary:** villi are grouped in prominent rosettes, that are well spaced and separated by smooth chorion. eg. deer, cattle, sheep.
3. **Zonary or Annular:** villi occupy the middle of the chorionic sac like a girdle band. Found in carnivores.
4. **Discoidal:** villi are restricted to one or two (**bidiscoidal**) disc-shaped area(s) on the chorion. Found in rodents, bats, primates.

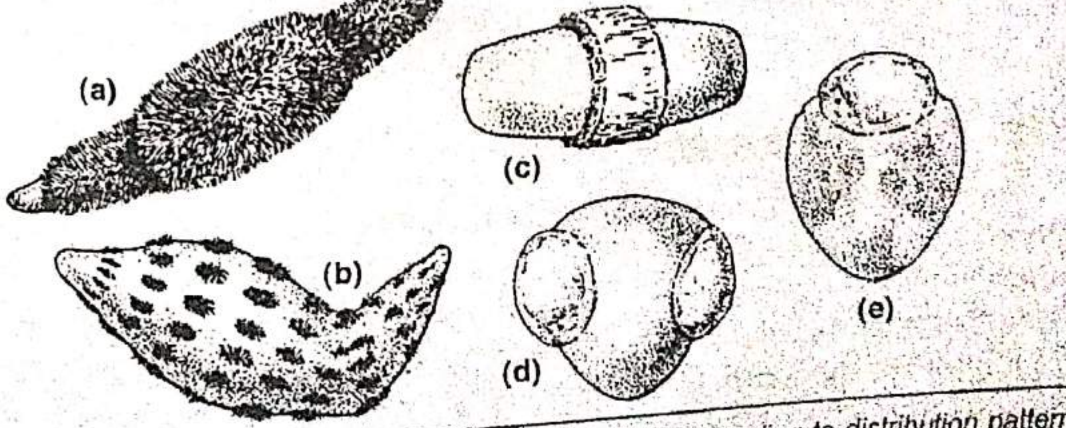

Fig. 15.11 : *Types of placenta observed in eutherian mammals according to distribution pattern of villi on the chorionic sac – (a) diffused (b) cotyledinary (c) zonary (d) bidiscoidal and (e) discoidal.*

Chorionic villi lie in apposition with the maternal uterine lining. When the villi and uterine lining just make a contact and do not fuse with each other, the association is called **semiplacenta**. At the time of parturition, the villi are withdrawn without any damage to the uterine wall. This type of placentation is called **non-deciduate** and found in pig. In other mammals, the chorionic villi make close contact with uterine lining, which is eroded according to intimacy of villi and uterine wall. Such type is called **true placenta** or placenta vera, and during parturition, uterine wall is damaged causing hemorrhage. This type of placentation is called **deciduate** and found in rabbits, man etc.

The intimacy between chorionic villi and uterine lining varies with species and depends upon the depth up to which the villi invade uterine mucosa. In fact, intimacy means reduction in barriers between the foetal and maternal circulation. On this basis, a structural classification of placenta includes five classes (Fig. 15.12):

1. **Epithelio-chorial placenta:** The chorio-allantoic membrane is in contact with intact uterine wall. All of the three tissue layers of the foetus and mother (epithelium, connective tissue and endothelium) are seen in this association and there is no chance of hemorrhage during parturition. Observed in pig, horse, lemurs etc.
2. **Syndesmo-chorial:** Chorionic villi invade through the uterine epithelium and come in direct contact with the vascular uterine mucosa. Observed in ruminants. During child birth, the villi are withdrawn without damaging the maternal blood vessels, hence no loss of blood.
3. **Endothelio-chorial:** This type of placenta is advanced than the previous two considering intimacy. Loss of uterine mucosa allows the chorionic villi to reach the endothelium of the maternal blood vessels. Observed in carnivores.
4. **Hemo-chorial:** A more intimate placenta is found in insectivores, bats and primates. Chorionic villi penetrate through the endothelium of the uterine blood vessels. So, maternal blood comes in direct contact of the chorionic villi and bleeding occurs during child birth.
5. **Hemo-endothelial:** Most intimate type of placenta found in higher rodents, like guinea pig, rabbit etc. Chorionic villi are bathed in maternal blood sinuses and loss their epithelium and connective tissue layers retaining only the endothelium lining of their blood vessels. In fact, there is only one barrier between foetal and maternal circulation – the endothelial lining of the foetal blood vessels. Severe bleeding occurs during child birth.

15.10 PLACENTA IN RABBIT

Placenta of rabbit is chorio-allantoic, discoidal, hemo-endothelial and deciduate. After fertilization, formation of three germ layers in rabbit takes about 8 days. During this period, blastocyst surrounded by zona pellucida travels through the fallopian tube and enters into uterus. Within uterus, the zona pellucida is eroded and the blastocyst becomes free, also called the **hatching of blastocyst** (Fig. 15.13). The trophoblast layer of the embryo undergoes rapid change once it comes in contact with the maternal tissue. Within the uterus, the blastocyst comes in contact of uterine wall and the place is called **maternal**

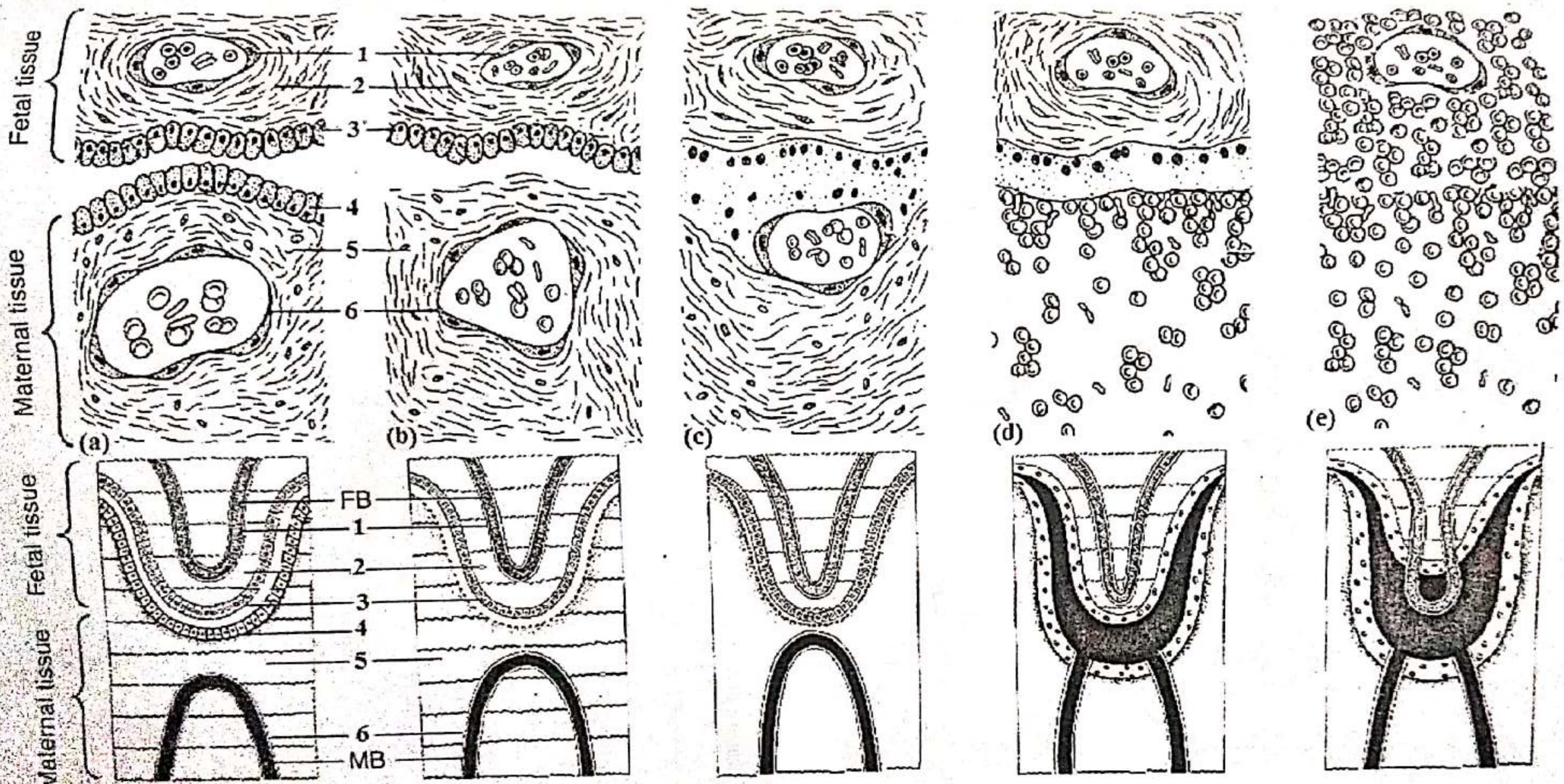

Fig. 15.12 : Classes of placenta found in mammals on the basis of intimacy between chorionic villi and maternal uterine lining. (a) Epithelio-chorial, (b) Syndesmo-chorial (c) Endothelio-chorial (d) Hemo-chorial and (e) Hemo-endothelial. FB = Foetal blood; 1 = foetal endothelium; 2 = foetal connective tissue; 3 = chorionic epithelium; 4 = uterine epithelium; 5 uterine endometrium; 6 = uterine endothelium; MB = Maternal blood.

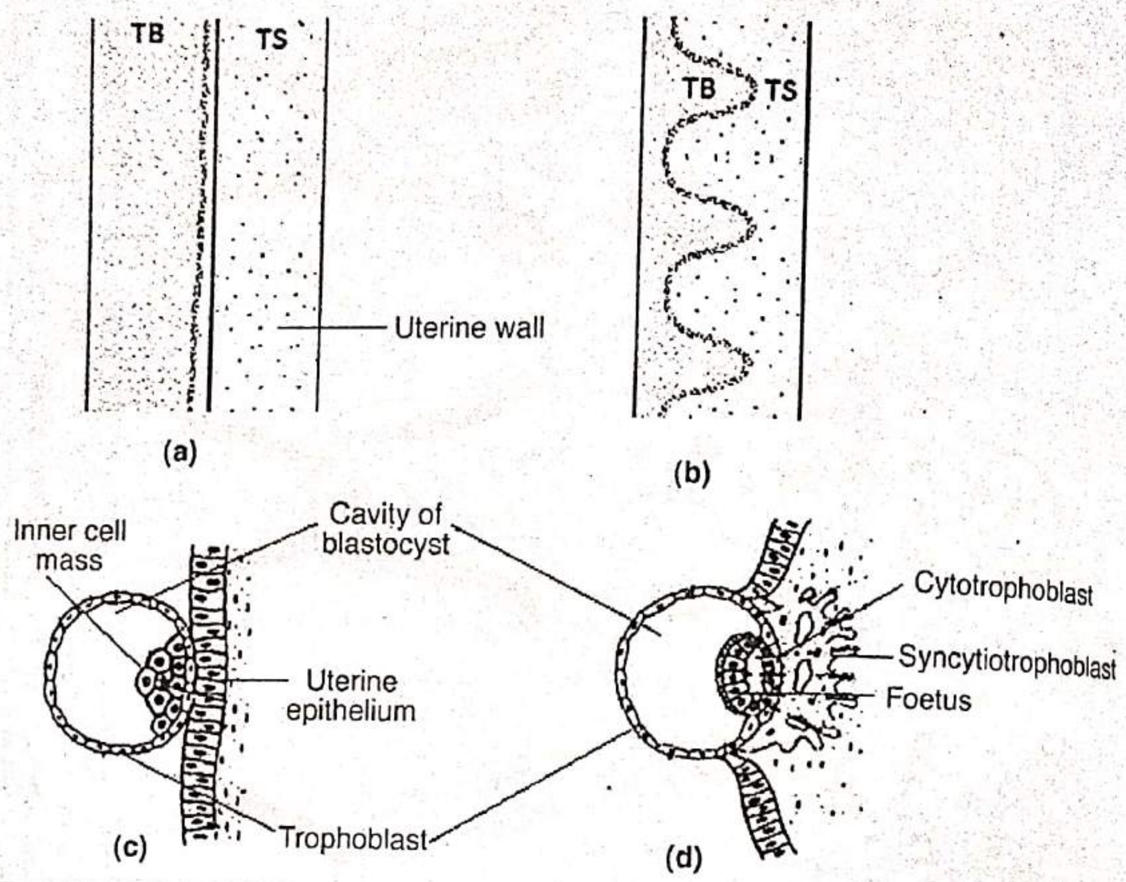

Fig. 15.13 : *Formation of placenta in rabbit (a) & (b) schematic view of relation between chorion of foetus and uterine wall of mother. TB = Trophoblast, TS = Trophospongia. (c) & (d) corresponding stages of implantation.*

site of implantation (Fig. 15.13). Due to rapid cell division, trophoblast becomes thick and two-layered. The inner layer is made up of well-defined cells called **cytotrophoblast**, but the outer layer is syncytial in nature called **syntrophoblast** (Fig. 15.14).

As blastocyst grows, much area of uterine wall comes in contact with blastocyst. The uterine wall at the site of implantation becomes thick, vascularized and spongy. Such uterine wall is called **trophospongia**. Proteolytic and lysosomal enzymes from trophoblast act on the uterine wall to form a frothy surface called **histotroph** that surround the embryo. Initially, the developing foetus of rabbit derives nutrition from this histotroph.

Development of minute villi from chorion makes the contact between blastocyst and uterine wall more intimate, because these villi penetrate the trophospongia. These villi are called **primary villi** because they have only epithelium layer, no connective tissue. In the mean time, many irregular lacunae develop in the syntrophoblast, which are filled by the blood of damaged uterine wall. Primary villi are extended in these blood-filled lacunae and derive nutrition.

Now allantois joins chorion and vascular chorio-allantoic membrane is formed. Allantoic blood vessels and mesoderm enter into the primary villi and they become

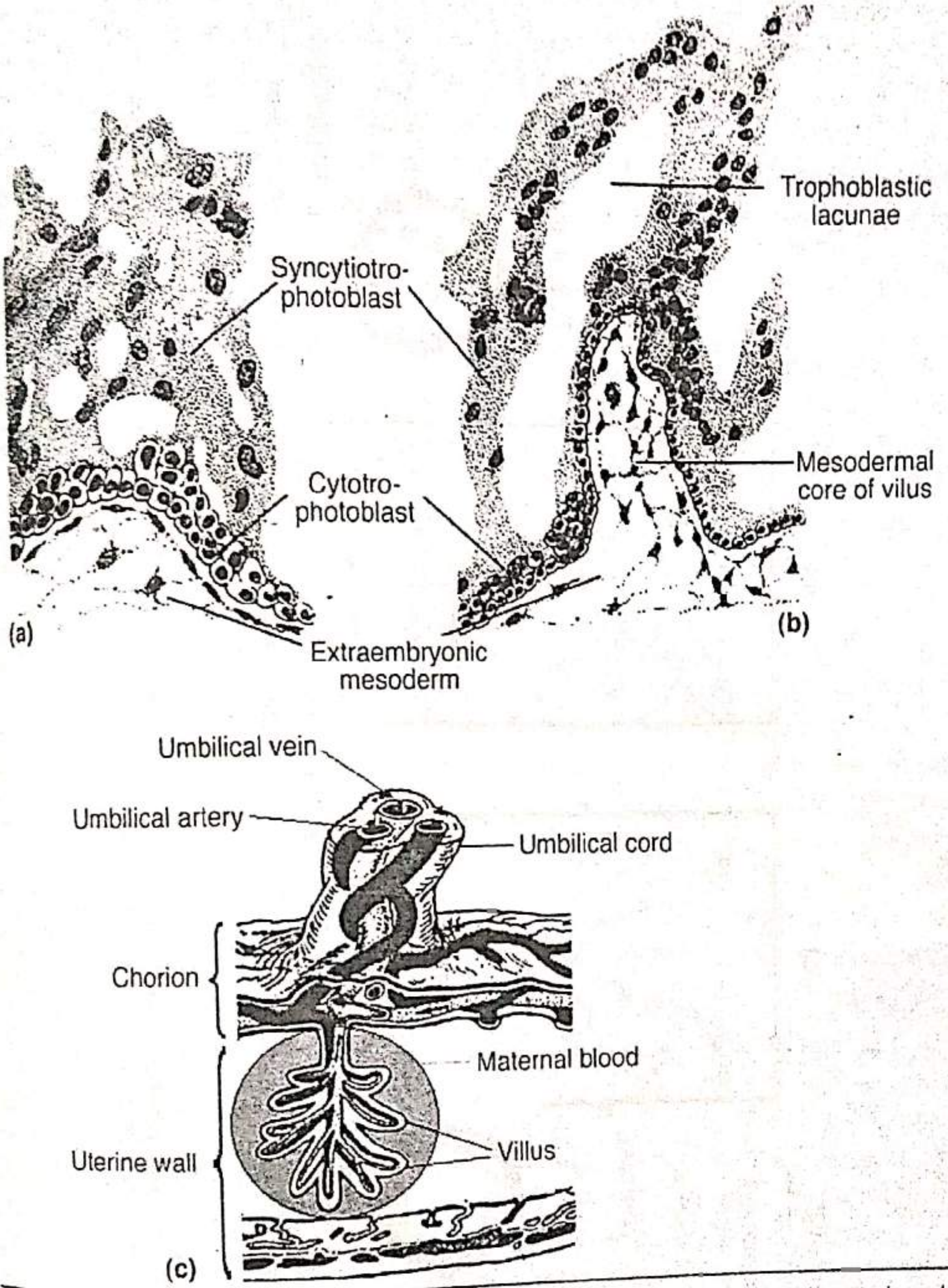

Fig. 15.14 : *Progressive changes in villus in rabbit. (a) Primary villus without mesenchymal core, (b) Secondary villus with mesenchymal core, (c) a portion of Hemochorial placenta*

secondary villi. Soon the secondary villi undergo branching followed by branching of blood vessels. Such villi are called **tertiary villi** (Fig. 15.15). In the final phase of placenta development in rabbit, the epithelium and connective tissue layers of chorion disappear, and the only endothelium lining of foetal blood vessel separates the foetal and maternal circulation.

Initially villi develop from whole surface of chorion but later, the villi facing site of implantation persist, all others disappear. So villi are restricted to a disc-shaped area

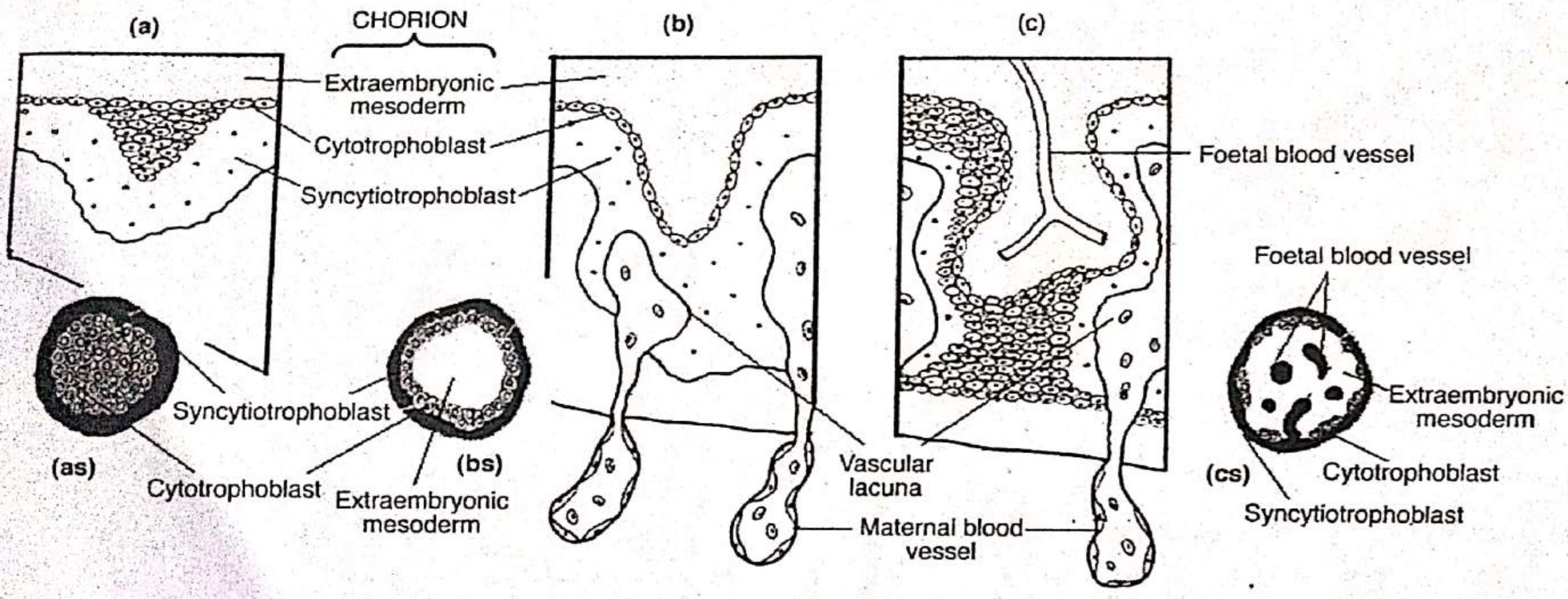

Fig. 15.15 : *Transformation of villi – (a) primary, (b) secondary and (c) tertiary (as), (bs) and (cs) are sections of corresponding villi.*

on the chorion of blastocyst. Again, progressive invasion of the villi through epithelium, connective tissue and endothelium of maternal blood vessels make the contact more intimate and strong; and initial epithelio-chorial placenta of rabbit becomes hemo-chorial and then hemo-endothelial. In the mean time, mucosa layer of the uterus expand over the blastocyst. The region of endometrium strongly anchor the villi from blastocyst is called **decidua basalis**. The portion of endometrium expand over the blastocyst is called **decidua capsularis**. Endometrium of remaining portion of uterus is called **decidua paraietalis.**

15.11 PLACENTA IN MAN

Fertilization in man occurs in the ampulla of oviduct. The zygote begins a trip down the oviduct toward the uterine cavity. During this trip, rapid mitotic cell division of zygote produces a solid sphere of blastomeres called morula. While surrounded by the zona pellucida, the morula continues its movement through the oviduct. At about 5th day after fertilization, the solid morula transform into a hollow ball of cells – the blastocyst. The blastocyst enters the uterine cavity and it takes about 7 days after fertilization.

15.11.1 Implantation

Within the uterine cavity, the blastocyst is free-living for 2 days. During this time, the blastocyst is bathed in the secretion of uterus. At this phase, the endometrium is in secretary phase and the endometrial glands secrete glycogen-rich fluid called **uterine milk**. The blastocyst derives metabolic nutrients, ions and oxygen from this uterine milk for survival and continued growth. There is a limitation of size of the free-living blastocyst after which such supply becomes inadequate. Before reaching that critical stage, the blastocyst interacts locally with the uterine endometrium and get attached to the wall. This step is called implantation (Fig 15.16) and occurs about 6-7 days after fertilization.

After entering the uterus, the blastocyst is positioned for implantation. The location of implantation site within the uterus tends to be characteristics of each species. Generally, the blastocyst implants in either the posterior portion of the fundus or the body of the uterus. The blastocyst is oriented in such a manner that the inner cell mass faces the endometrium. But the mechanism by which the implantation site is determined is little known. Two major factors seem to be involved in the orientation of blastocyst to the endometrium. First is the shape of the blastocyst and its response to the contraction of the uterus. The second is specific adhesiveness of the trophoblast; only certain regions of trophoblast are adhesive. In polytocous mammals, having several embryos at a time, a spacing mechanism operates to ensure the distribution of blastocysts along the axis of the whole uterus, rather than clustering at one site.

Implantation is prerequisite to placentation. During implantation, two phases of interactions occur between the embryo and uterus – adhesion and invasion. During adhesion, the zona pellucida disintegrates and freed embryo attaches to the luminal surface of the uterine wall. This is achieved by the formation of junctional complexes between endometrium of uterus and the trophoblast of the embryo. During invasion, a trophoblast-originated cytolytic activity leads to disruption of uterine epithelia. This

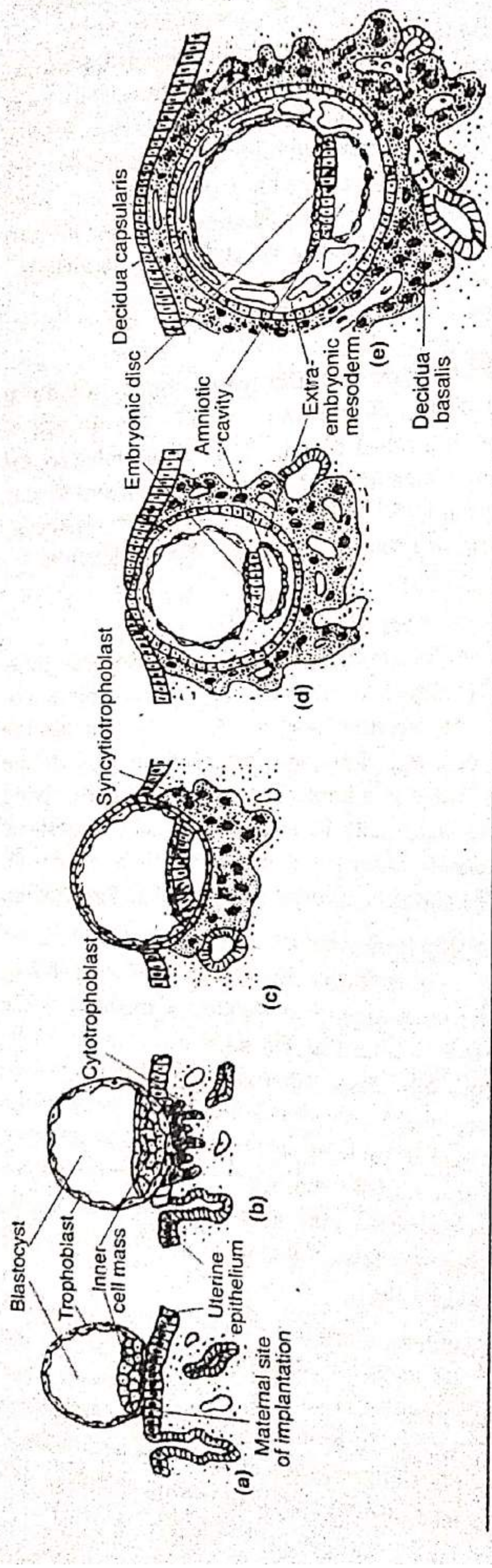

Fig. 15.16 : Stages of implantation of human embryo in uterine wall.

is followed by the intrusion of trophoblastic processes into the basal lamina and underlying uterine stroma.

Events subsequent to attachment vary with species and accordingly implantation may be classified into two categories (Box. 15.1).

In many species, blastocyst reaching uterus but implantation does not occur for a prolonged time. This extension of pre-implantation period is called delayed implantation, during which development of blastocyst becomes relatively quiescent. The phenomenon of delayed implantation is natural and widespread in mammals, and found in diverse groups. When it occurs due to natural suppression of endogenous secretion in females engaged in suckling young of previous litter, it is referred to a **facultative delayed implantation**. Such phenomenon is found in armadillo, rat, mice gerbil and help to prevent excessive metabolic drainage of lactating mother. In these animals, if litters are removed or exogenous estrogen is injected to the mother, implantation occurs within 24 hours. In many other species, such as elephant, fruit bat, brown bear, delayed implantation is an essential and normal part of pregnancy called **obligate delayed implantation** or **diapause**. In these species, blastocyst remains un-implanted

Box. 15.1 : Types of implantation

According to intimacy of attachment, implantation may be of two types (Fig. 15.17):

Invasive Implantation : When the blastocyst breaks through the uterine epithelium and invades the underlying stroma extensively. Seen in primates, rodents, carnivores. Free-living phase of blastocyst of these mammals is relatively short and invasive embryos tend to be smaller during attachment.

During invasive contact, relatively few tropho-ectodermal cells are involved in contact. The foetal tissue not only erodes the epithelium, but also stromal cells, connective tissues and even the wall of blood vessels (in man, rabbit). It seems that a primary signal from the foetus is effectively transferred to the maternal stroma and then amplified. The stromal response to invasive contact seems to anticipate the event by developing protective device, controlling the limit and depth of invasion. An earliest visible response to this event is an increased vascular permeability in the stromal tissue, followed by edema and compositional changes in the intercellular matrix. There are changes in the morphology of stromal cells, and progressive branching and ingrowth of capillaries. These changes are easily visible in man and rodents, and called **decidualization**. The endometrial component of placenta in decidualization is **decidua**.

The rapid progression of changes in the endometrium underlying implantation site is probably stimulated and organized by signal from the blastocyst. The signal could be low molecular weight secretion from the blastocyst. More likely, the blastocyst triggers the adjacent uterine tissue to secrete a decidualizing molecule, which then spread through the stroma. Histamine and prostaglandin are suggested as probable decidualizing molecules. Because both are present in the endometrium, and can induce decidualization, if injected locally. However, if endometrial epithelium and stroma are primed with progesterone and then exposed to high level of estrogen, maximum decidual response is shown by the conceptus of rat and human.

Non-invasive implantation : In this type of implantation, epithelia more or less retained integrity; therefore, uterine epithelium not the underlying stroma, limits and controls the invasion of conceptus. This type of implantation is found in ungulates. Attachment of the blastocyst to the uterine wall is

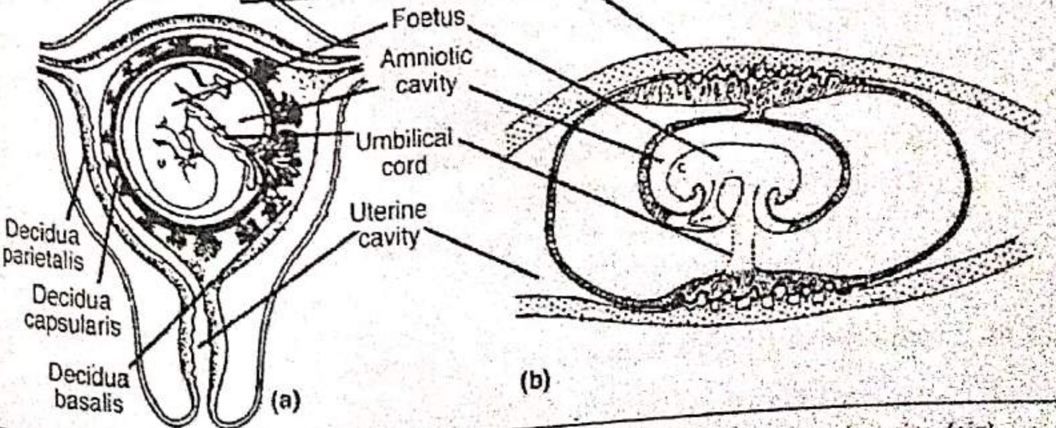

Fig. 15.17 : *Types of implantation – (a) invasive (human) and (b) non-invasive (pig)*

relatively late. So, free-living blastocyst continues to draw the so-called uterine milk and grows greater in pre-implantation size than that of invasive embryos. Apparently, no decidual response is induced in uterine stroma, but certain changes prove the recognition of embryonic presence. These changes include distinct cellular morphology and increase in vascularity.

According to the relation of chorionic sac to the uterine wall, three types of implantation are found in mammals –

(a) Superficial or Central: growth of chorionic sac results into contact with lining of main uterine cavity, such as hoofed mammals and monkey.

(b) Eccentric: the chorionic sac is a fold or crypt of endometrium that closes off from the main cavity such as rat, squirrel, cat etc.

(c) Interstitial: chorionic sac penetrates through the uterine lining, such as guinea pig, apes, man.

for a prolonged period, several weeks or months. Such delay has definite adaptive advantages. For example, among hibernating fruit bats of Southern Hemisphere, mating and fertilization occur in March, when food is available plenty. The embryo remains as blastocyst until next September. And then, blastocyst is implanted and reactivates for young birth in December, when food and seasonal conditions are optimal for new young as well as the lactating mother.

15.11.2 Hormonal control of implantation

Uterus must be prepared for receiving the blastocyst and to support implantation. Estrogen dominated uterus is hostile for blastocyst and may cause its premature death. Normally, progesterone increases rapidly from the pre-ovulatory period. Under progesterone domination, uterus is ready for friendly interaction with blastocyst. For optimal uterine sensitization, high level of estrogen is necessary when progesterone is dominating hormone. Estrogen stimulates the uterine epithelium in two ways; firstly it stimulates a glandular secretion – like proteolytic enzymes, glucose, amino acid etc., which cause embryonic activation. Secondly, estrogen sensitizes the uterine epithelial cells to receive the blastocyst signal to commence decidualization. Thus it appears that embryo first stimulates the progesterone-primed uterus, which in turn secret embryotrophic factors for successful attachment and implantation of the embryo itself. hCG (human chorionic gonadotropin) is another hormone produced by trophoblast cells. hCG acts like LH, prevents corpus luteum from disintegration. Sustained corpus luteum continues secretion of progesterone and estrogen, which maintain the uterine lining in a secretary state and thus prevent menstruation.

15.11.3 Formation of placenta in man

Within a few hours of implantation and establishment of contact with the uterine wall, human blastocyst destroys the adjacent epithelial layer of uterus and sink into the underlying connective tissue. The trophoblast of blastocyst plays a vital role in the formation of placenta. With immediate contact with maternal tissue, blastocyst starts proliferation rapidly and gives rise to two distinct layers of cells. The outer layer is syncytiotrophoblast, closer to uterine wall and comprised of multinucleate syncytial types of cells. The deeper layer, closer to the embryo, is cytotrophoblast with distinct cell boundaries. Cytotrophoblast serves as a proliferative source for generating more trophoblastic cells. Syncytiotrophoblast secretes enzymes that digest and liquefying the endometrial cells and enable the blastocyst to penetrate deeper into uterine wall. During this phase, endometrial secretion nourishes the burrowing blastocyst.

Lack of compact cellular nature allows syncytiotrophoblast to develop system of cavities called trophoblastic lacunae. As a result of this change, the syncytiotrophoblast becomes a meshwork of irregular strands with interstices in between. The strands now penetrate deeper into the uterine wall and reach the maternal blood vessels. Proteolytic action of trophoblastic cells breaks down the endothelium of maternal capillaries; thereby maternal blood flows into trophoblastic lacunae. Maternal blood circulation in the lacunae provides nutrients to the growing foetus.

In the next step, the underlying cytotrophoblast penetrates into strands of syncytiotrophoblast forming a cellular core. The cytotrophoblast is followed by underlying extraembryonic mesoderm along with allantoic blood vessels. Finally, trophoblast layer develops several finger-like ramifying branches called **chorionic villi,** which make intimate connection with maternal circulation. Diffusible nutrients and oxygen from maternal blood collected in the lacunae pass to the foetus through chorionic villi. In human, chorionic villi develop uniformly around the chorionic sac, but only decidua basalis is penetrated by them. Thus human placenta includes decidua basalis (maternal part) and chorion frondosum (foetal side) [Fig. 15.18].

15.12 PHYSIOLOGY AND FUNCTIONS OF PLACENTA

In viviparous animals, placenta helps to transfer the nutrients directly from maternal source. In the first week, the human embryo depends upon the diffusion of materials from the endometrium. In second week, the embryo receives metabolites through the syncytiotrophoblast from maternal blood circulating through intervillous space. After fourth week, embryonic heart starts to pump blood unidirectionally and embryo becomes dependent upon placenta. The transport of substances between the placenta and the maternal blood is facilitated by the great surface area of the placenta. There are scattered areas where the barrier between fetal and maternal blood is extremely thin, measuring only a few micrometers. These areas, sometimes called **epithelial plates,** are apparently morphological adaptations designed to facilitate the diffusion of substances between the fetal and maternal circulations (Fig. 15.19). The transfer of substances occurs both ways across the placenta. The bulk of the substances transferred from mother to foetus consist of oxygen and nutrients. The placenta represents the means for the final elimination of carbon dioxide and other foetal waste materials into the maternal circulation. Main functions of placenta are:

- The main function of placenta is to mediate the metabolic exchange between mother and foetus. It provides the foetus with nutrition and respiration.

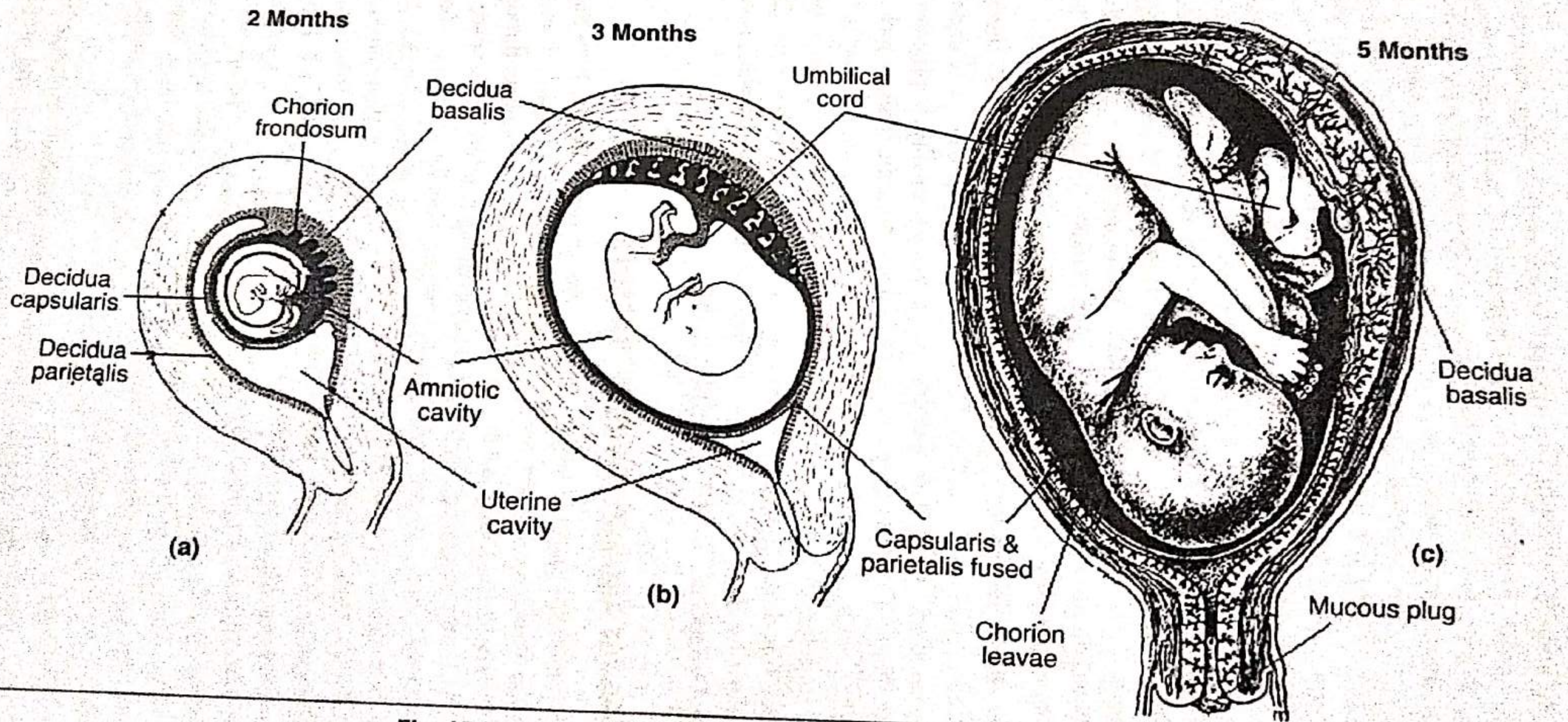

Fig. 15.18 : *Progressively full formed placenta in man within uterus.*

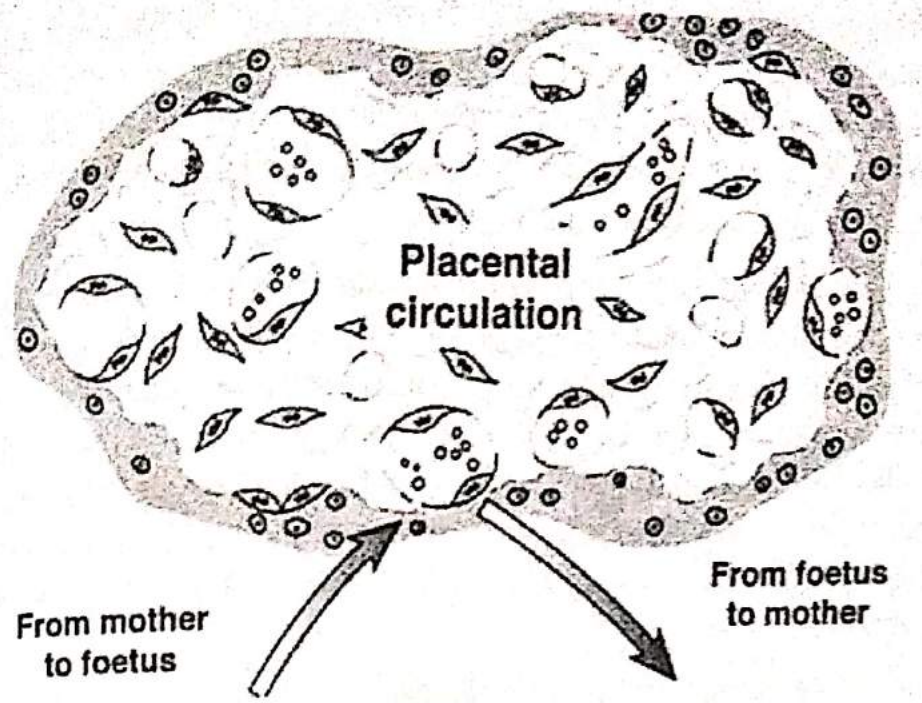

Fig. 15.19 : *Placenta facilitates the exchange of various substances between the fetal and maternal circulations.*

Placenta is an avenue for excretion of foetal waste products, but at the same time, serves as barrier between foetal and maternal blood stream.

- Placenta is an important endocrine organ and secretes a number of hormones essential for embryonic development. The first hormone is human chorionic gonadotropin (hCG), secreted by syncytiotrophoblast tissue. hCG is a peptide hormone and causes other cells of placenta as well as ovary of mother to produce progesterone – essential for embryonic implantation and maintenance of embryo. Among other hormones, placenta secretes estrogen, human placental lactogen (hPL), human placental growth hormone (hPGH), corticotrophin-releasing hormone (CRH).
- A very important function of human placenta is to prevent immunological rejection of embryo by the mother. the placenta enjoys privileged protection from maternal immune system. Mother and foetus act co-operatively to down-regulate the functions of immune cells while allowing innate host immune

defenses to stay intact. Discovery of Tumor Necrosis Factor (TNF–superfamily) in the trophoblast cells provides much insight into the immunology during mother-foetus interface. TNF α is found important for placental homeostasis, cytotrophoblast cell differentiation and defense from pathogens. These cytokines help placental growth and development.

- Many substances traverse placenta at different ways and different rates. Water, gases and simple solutes pass across the semipermeable placental barrier through diffusion at varying rates. Substances other than water with rapid diffusion rate include urea, uric acid, simple amines, oxygen, and carbon dioxide. Sugars, essential amino acids, water soluble vitamins pass through placenta by active or facilitated transport by enzymatic carriers. Foetal hemoglobin has the capacity to bind more oxygen than maternal hemoglobin at same oxygen tension.
- Some enzymes produced by placenta prevent trans-placental passage of some material potentially dangerous to foetus. Similarly, large molecules like plasma proteins, antigen-antibodies and many drugs are denied to pass through placental membrane. Thus placenta not only maintains the foetus, but also the chief responsible for the intrauterine welfare of the foetus.

QUESTIONS

1. Describe the process of cleavage in mammals with suitable diagram.
2. With necessary diagram, discuss the process of gastrulation in mammals.
3. Discuss the process of formation of dorsal-ventral, anterior-posterior and left-right axes in mammals.
4. Define placenta. With example and diagram, classify placenta in mammals according to distribution of villi and intimacy between foetal and maternal tissue.
5. Describe the process of formation of placenta in rabbit. Mention major functions of placenta.
6. *Write short note on:* Blastocyst, Amniotic cavity, Neurulation in mammals, Deciduate placenta, Types of placentation, Implantation
7. MCQ

   i) Compaction is a phenomenon found in cleavage of

   (a) chick. (b) zebra fish.
   (c) *Xenopus*. (d) mouse.

   ii) Purpose of zona pellucida envelope of mammalian blastocyst is to

   (a) supply nutrient to embryo. (b) prevent premature implantation.
   (c) prevent desiccation of embryo. (d) protect inner cell mass.

iii) Embryo proper in mammals develop from

(a) blastocyst. (b) inner cell mass.

(c) trophoblast. (d) hypoblast.

iv) In formation of placenta in eutherian mammals, vascular support to chorion comes from

(a) yolk sac. (b) allantois.

(c) amnion. (d) none of the above.

v) Most intimate type of placenta is

(a) hemo-chorial. (b) hemo-endothelial.

(c) syndesmo-chorial. (d) endothelio-chorial.

vi) The region of uterine endometrium strongly anchor the villi from blastocyst is called

(a) decidua basalis. (b) decidua capsularis.

(c) decidua parietalis. (d) decidua vera.

vii) Chorionic villi supported by mesoderm and allantoic blood vessels is called

(a) primary. (b) tertiary.

(c) secondary. (d) none of above.

viii) One important hormone secreted by placenta embryonic development is

(a) hCG. (b) ACTH.

(c) GHRH. (d) oxytocin.