

# Lecture 4

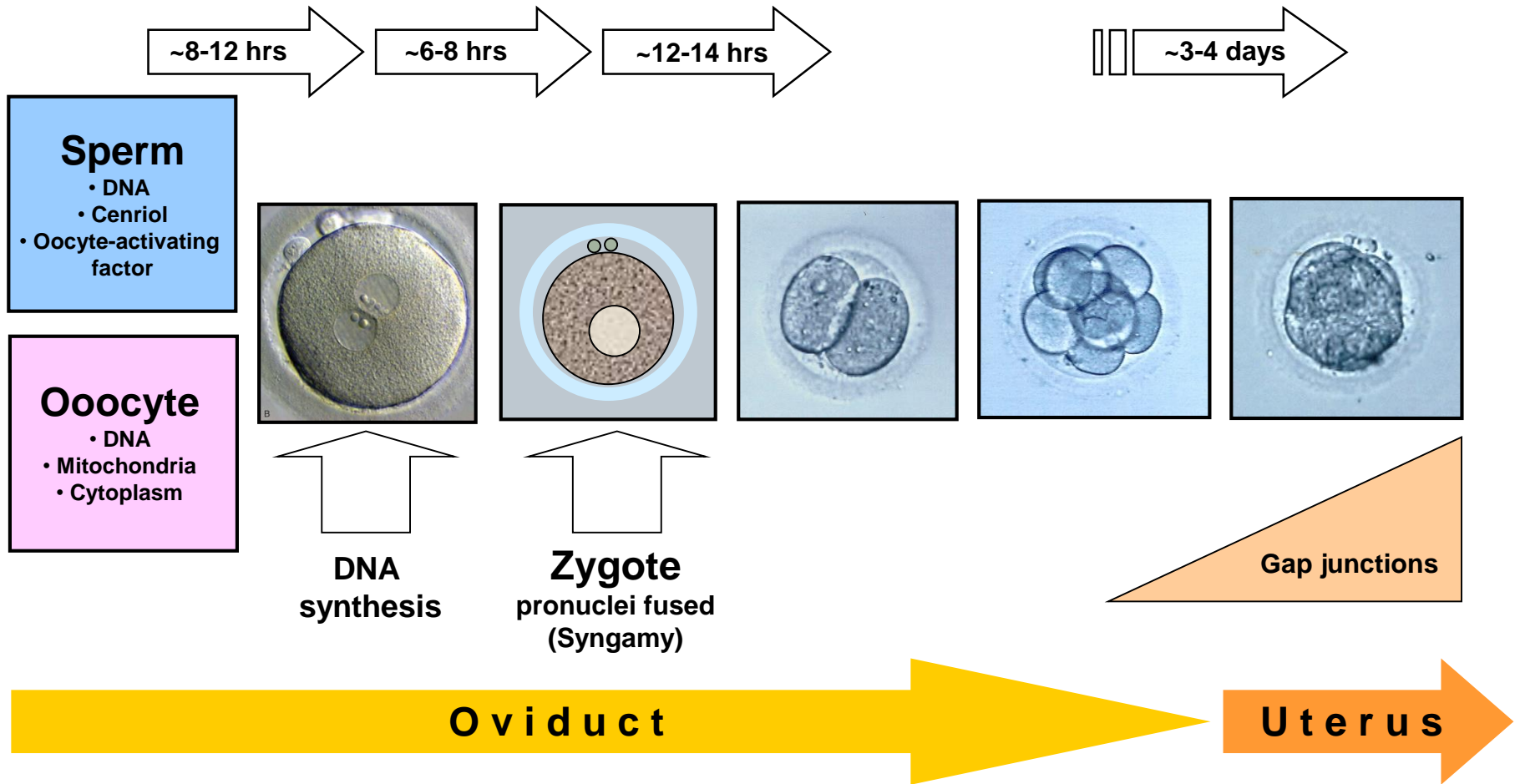
## Reproductive biology and Embryology

- Early embryo cleavages
- Implantation
- Somatic nuclear transfer– cloning
- Gastrulation
- Extraembryonal structures
- Fetal membranes
- Placenta

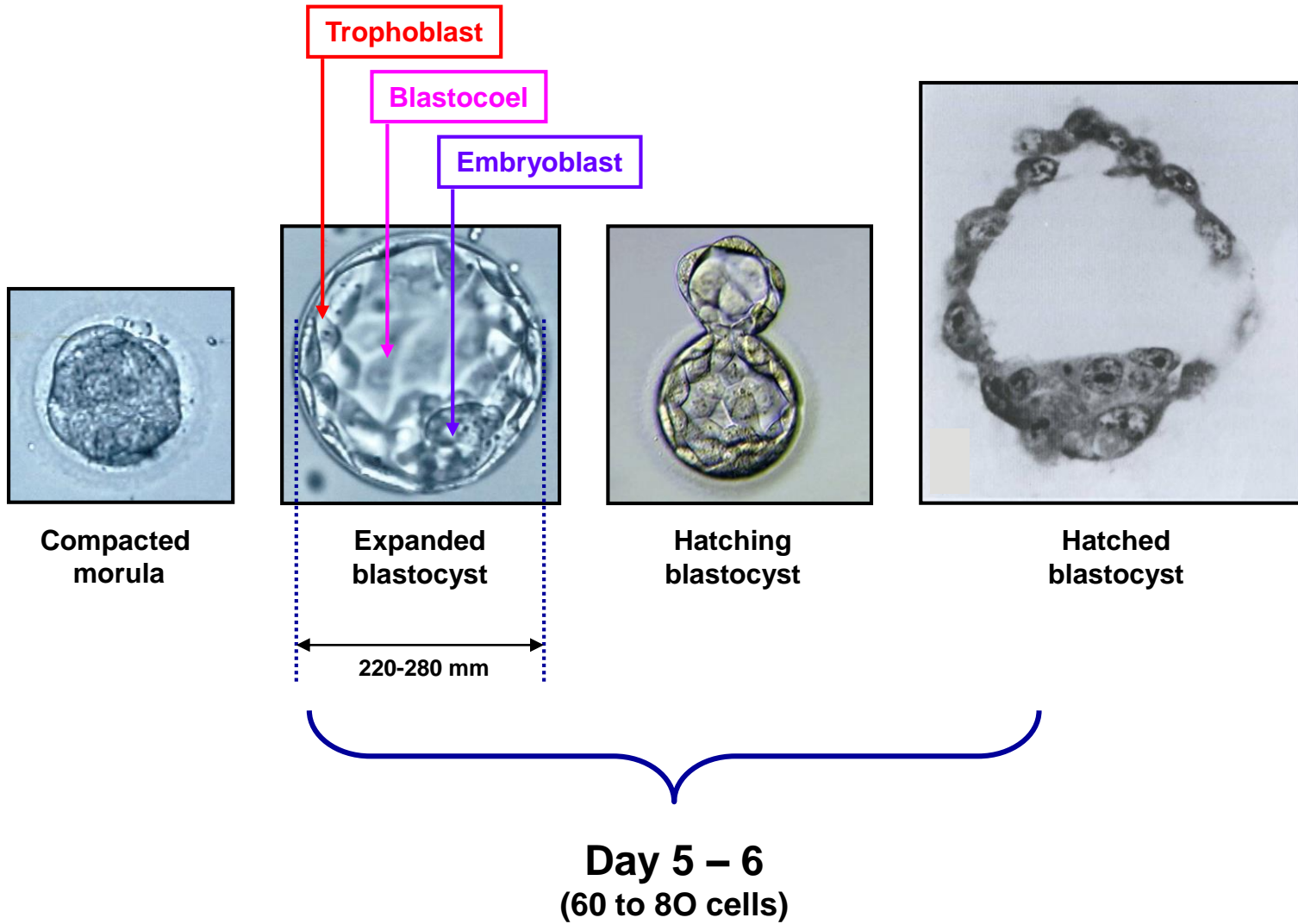
Brno, March 2020

# Fertilization

## Zygote formation and the first cleavages



# Blastocyst formation

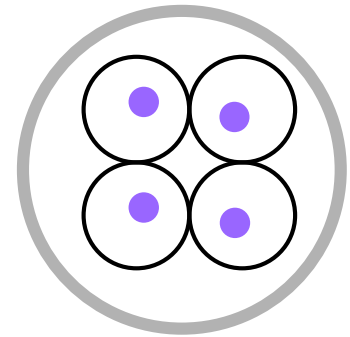
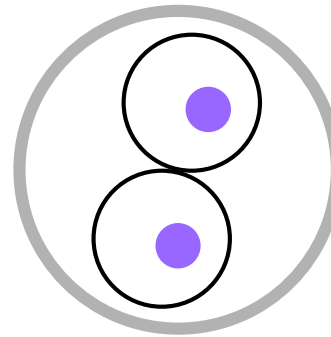
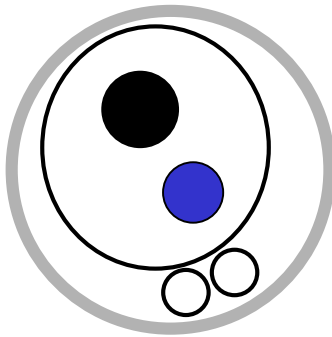
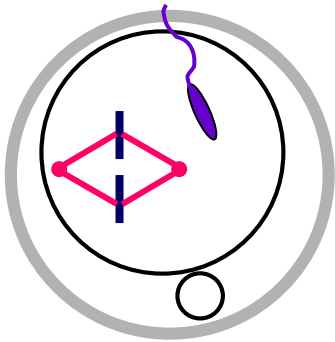
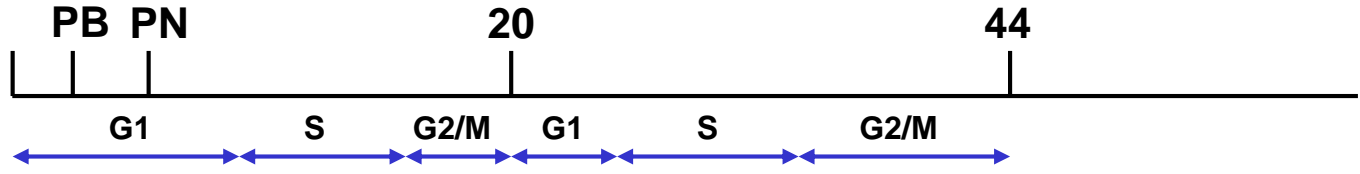


# Early embryogenesis of human embryo



Dr. Zuzana Holubcová  
Dept. Histology and Embryology + REPROFIT, Brno

# A potency of oocyte cytoplasm



Translation of maternal mRNA

Translation of zygotic mRNA

Zygotic transcription

Activation of embryonal genome

Repression of transcription

Significance of „enhancers“

# Activation of embryonal genome

**It is not a single discrete event**  
(first signs occur in zygote, in man it reaches its maximum in 4- to 8-cell embryo)

## Two types of transcripts

Transcripts that replace degraded maternal mRNAs

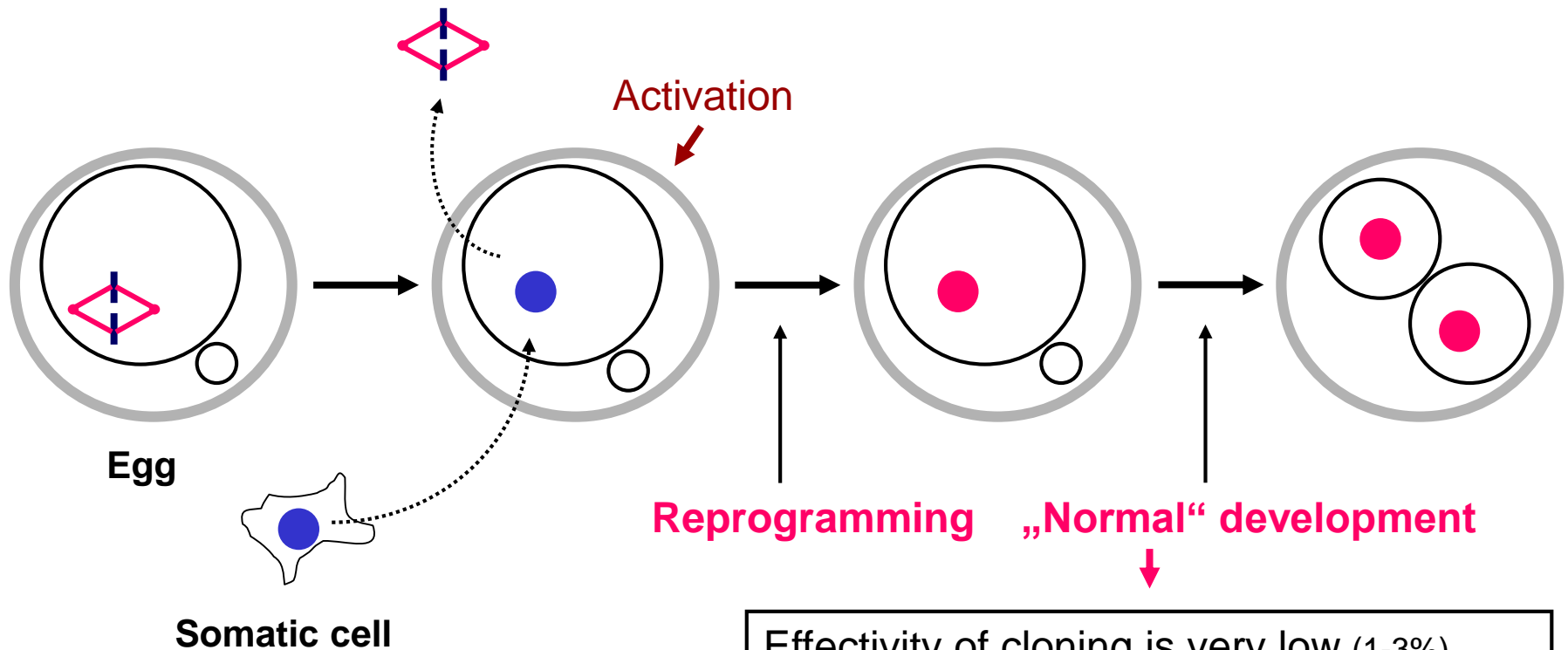
Novel transcripts that underlie **new pattern of gene expression**

**It is „responsible“ for establishment of totipotency of blastomeres**

**&**

**It represents phenomenon known as genome REPROGRAMMING**

# Nuclear transfer (cloning) - principle



Effectivity of cloning is very low (1-3%)

Reprogramming is slow and most likely incomplete (as the result, gene expression is often abnormal)

Effectivity of reprogamming depends on many factors (type of somatic cells, position in cell cycle phase, ...)

# Blastocyst implantation

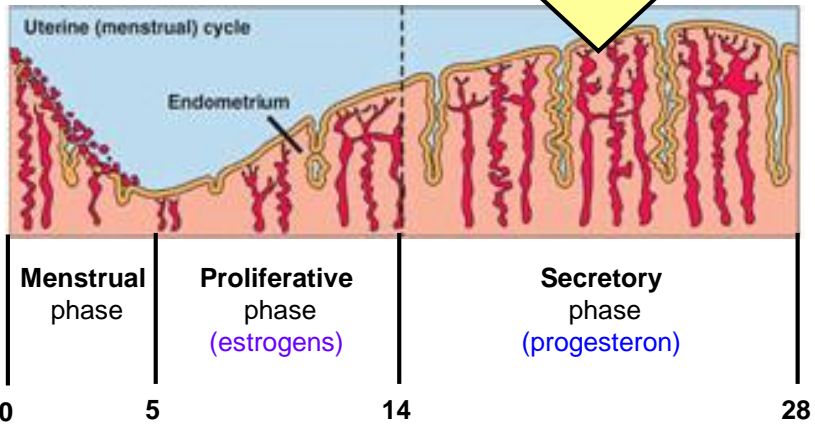


**Molecular players**

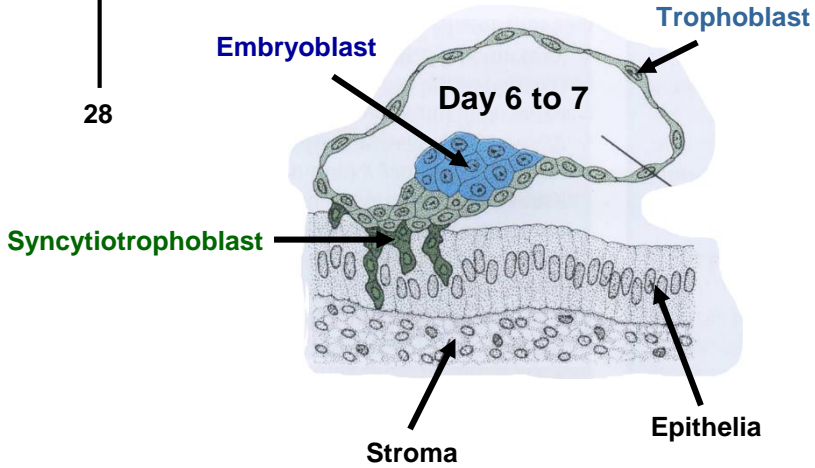
- Selectin on trophoblast + glycoproteins on epithelia
- Integrins + Laminin, Fibronectin
- IGF1, IGF2, VEGF
- Metalloproteinases + their tissue inhibitors
- Progesteron
- hCH
- Prostaglandins

Functional zone of endometrium

compact  
+  
spongius  
+  
basal

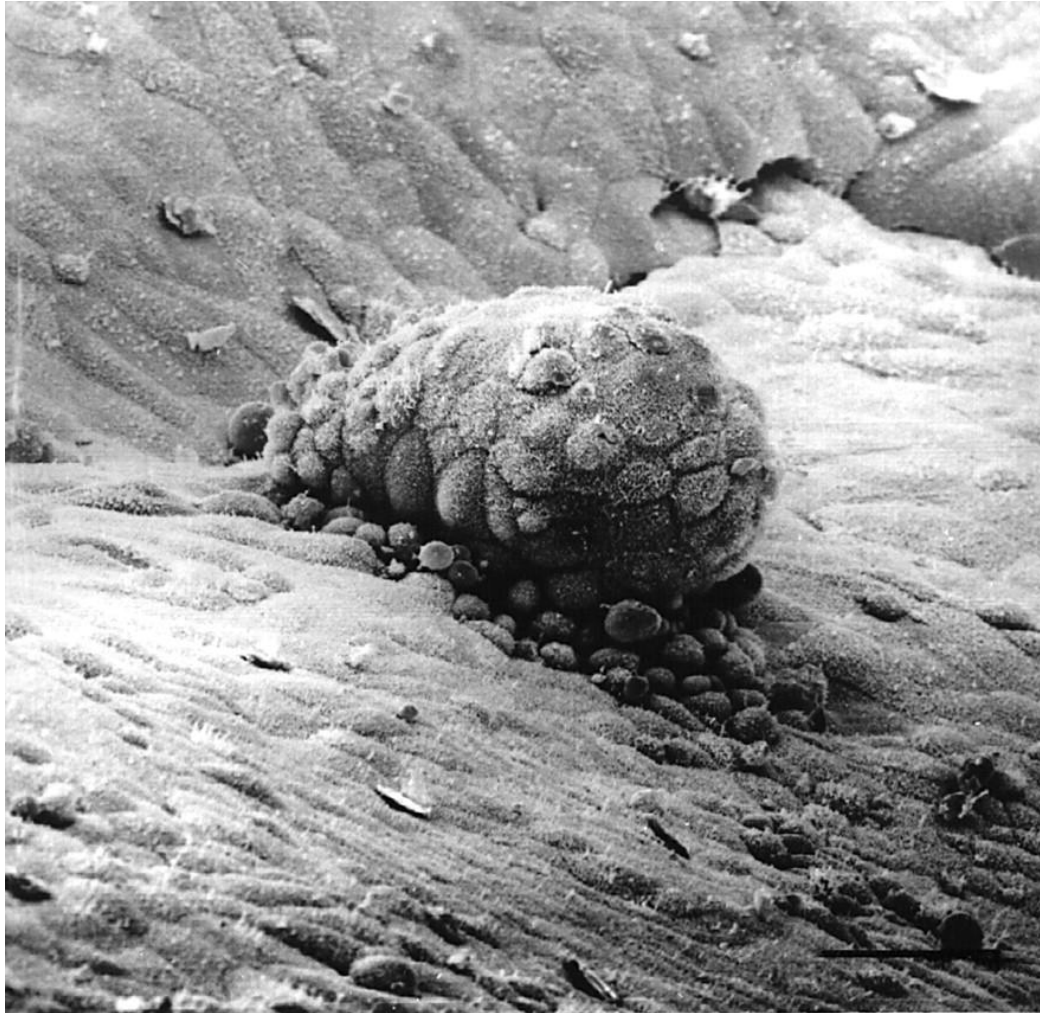


Ovulation  
followed by  
Fertlizaion





# Blastocyst implantation

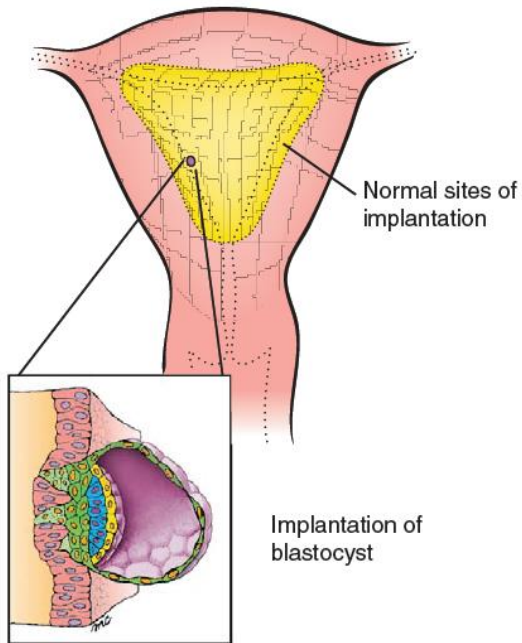


<http://myselfishgenes.blogspot.hu/2013/05/what-happens-to-my-embryos-if-they-do.html>

# Blastocyst implantation – place of implantation

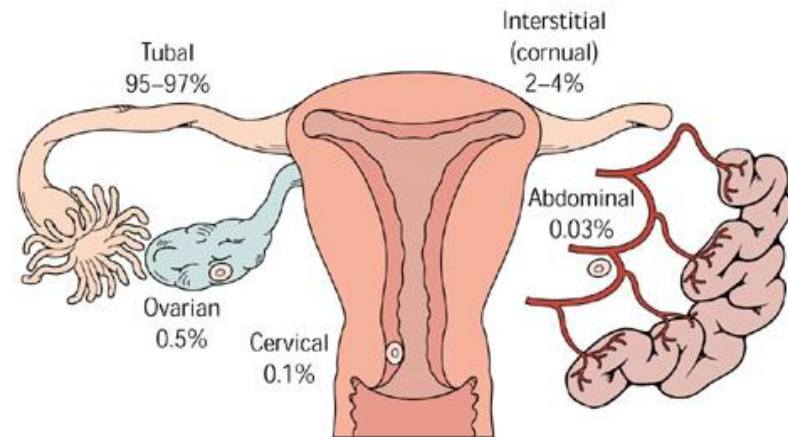
## Normal

(posterior / anterior wall of uterus)



## Abnormal

(0,25 až 1% of all implantations)



# Early development – The second week (1)

Completion of implantation + Further embryo development

**Trophoblast**

- Furtner invasion into endometrium
- Damage to capillaries and glands
- Engulfment of apoptosing cells (Fas/Fas ligand)

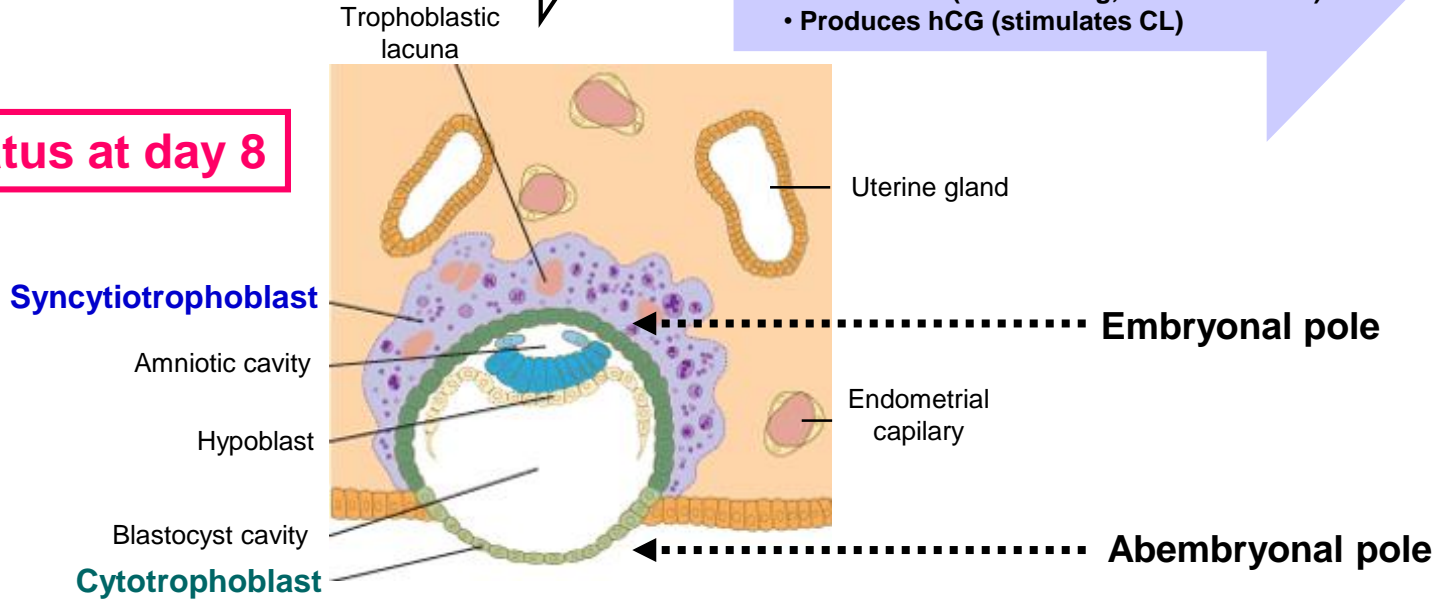
**Cytotrophoblast**

- Mitoticaly dividing cell (single layer)
- Supply for syncytiotrophoblast

**Syncytiotrophoblast**

- Fused cells (nondividing, multinucleated)
- Produces hCG (stimulates CL)

**Status at day 8**



# Early development – The second week (2)

## Decidual reaction of endometrium

Vessel number/density increases

Fibroblasts differentiate into glycogen- and lipid-containing cells

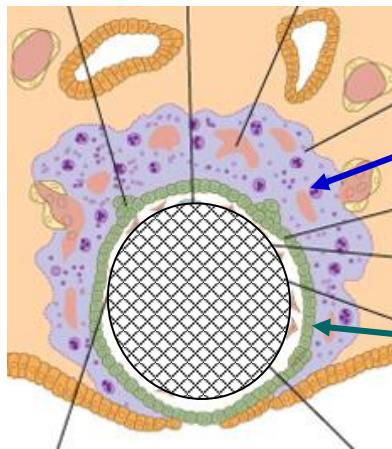
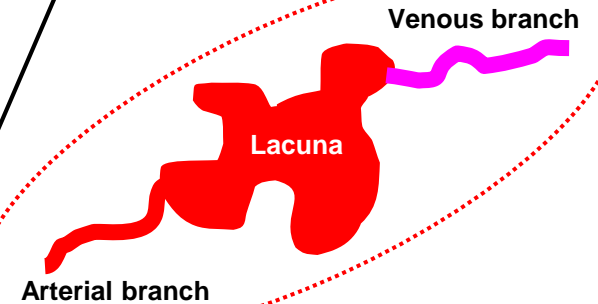
Fused lacunae create a network

+

Uterine vessels erode



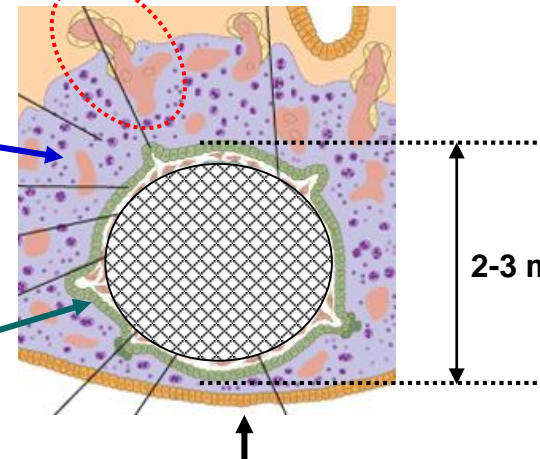
**Uteroplacental circulation**



**Syncytiotrophoblast**

Sponge-like appearance

**Cytotrophoblast**

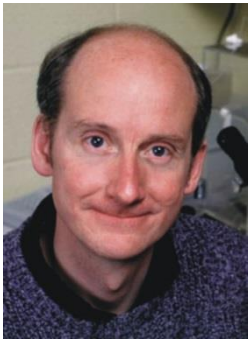


2-3 mm

Epithelization of the implantation site

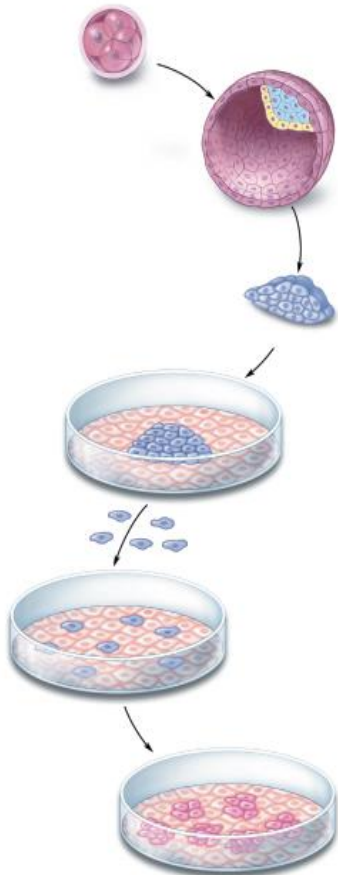
Status at day 10

Status at day 12



# Human Embryonic Stem (hES) Cells

(Thompson et al, 1998)

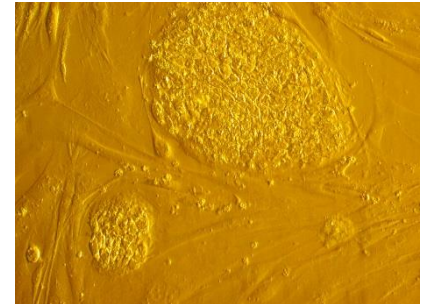
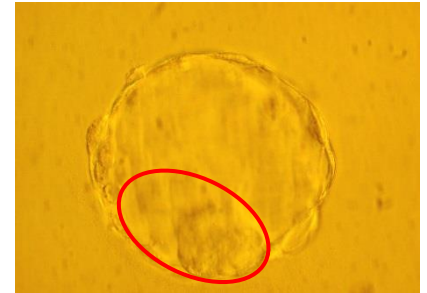


Early embryo at blastocyst stage

Isolated embryoblast (ICM - Inner Cell Mass)

Isolated embryoblast after placing to *in vitro* conditions (+ feeder cells + FGF2)

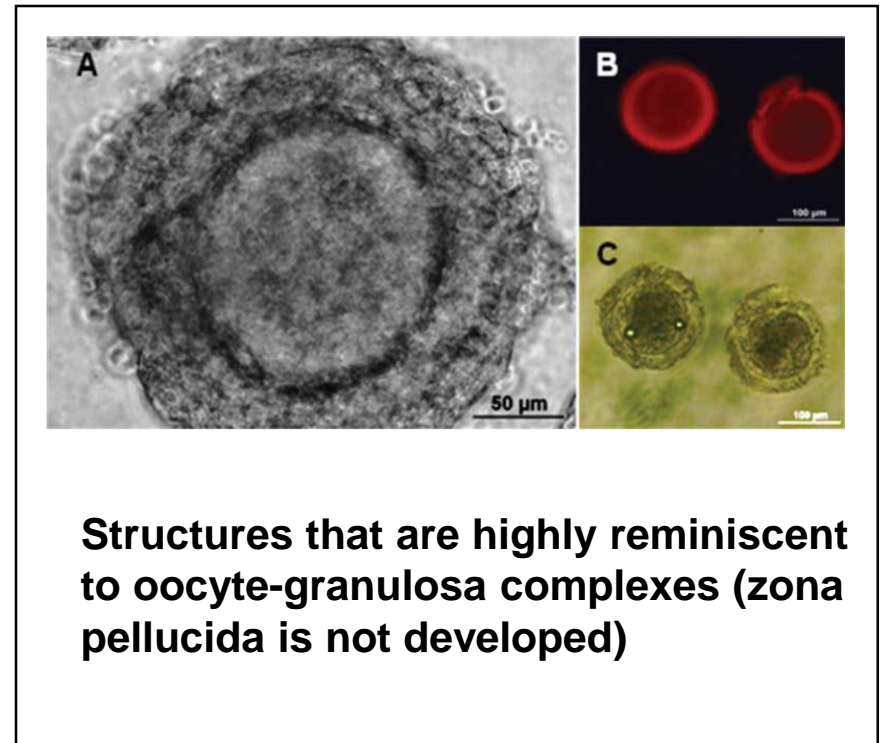
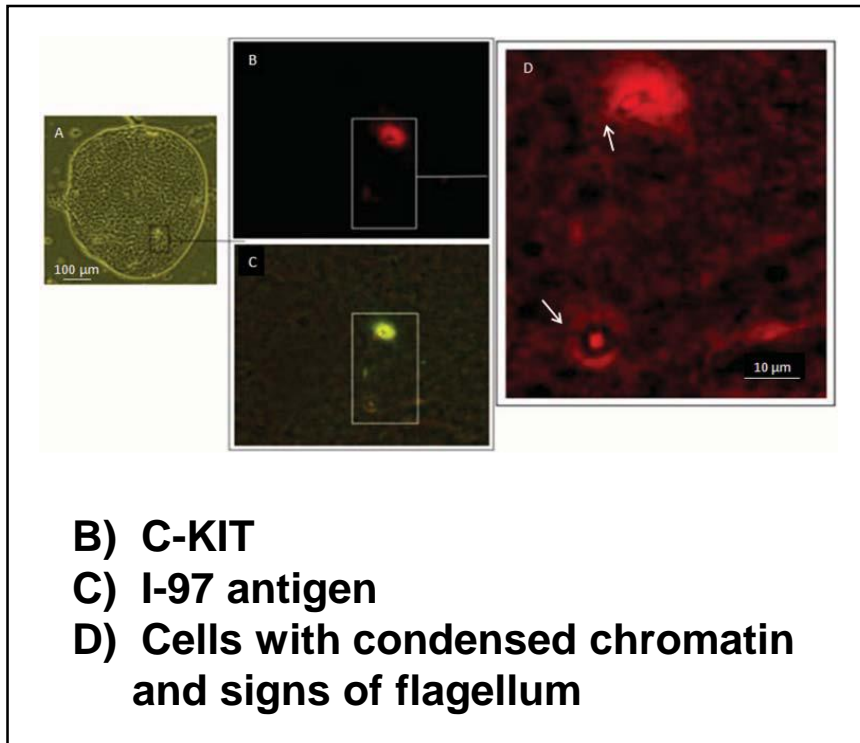
Propagation in culture by enzymatic disaggregation (repeated passaging)





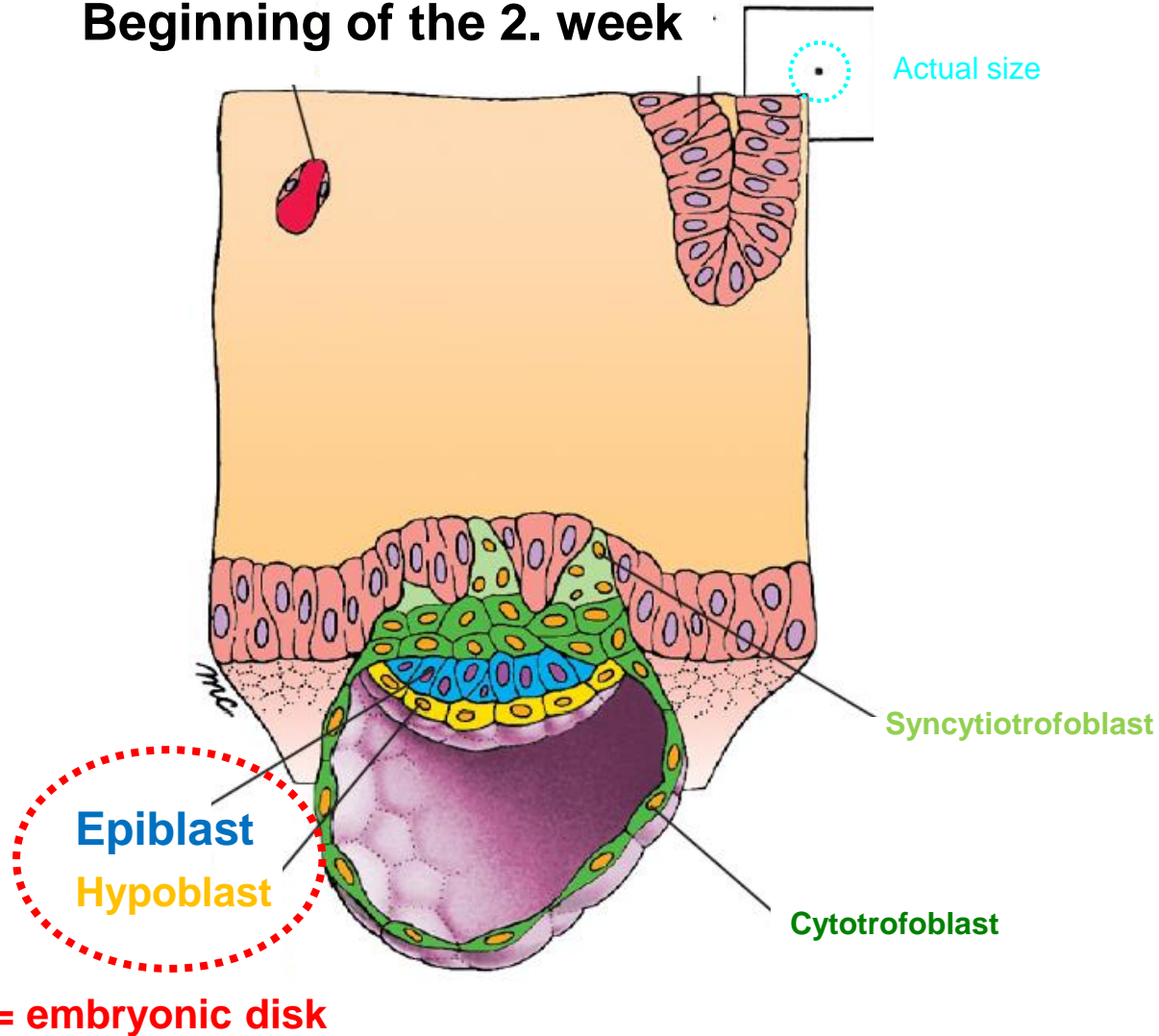
# Derivation of postmeiotic germ cells from hESC

Prof. Harry Moore, University of Sheffield, 2009



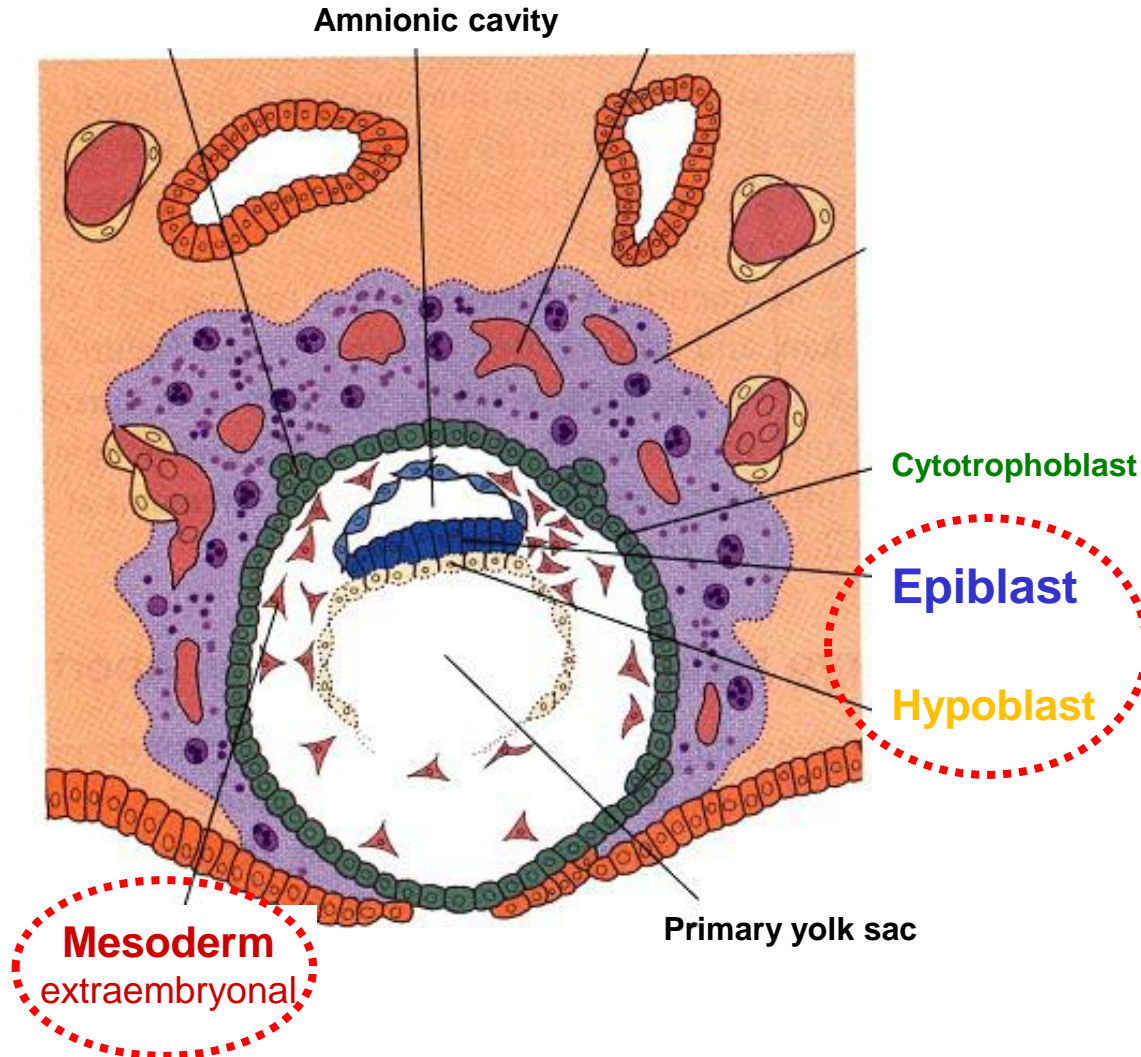
# Gastrulation – establishment of three germ layers

Beginning of the 2. week



# Gastrulation – establishment of three germ layers

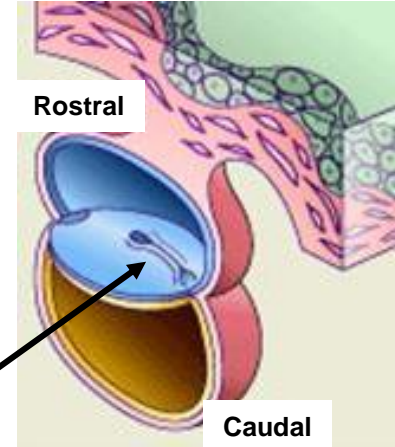
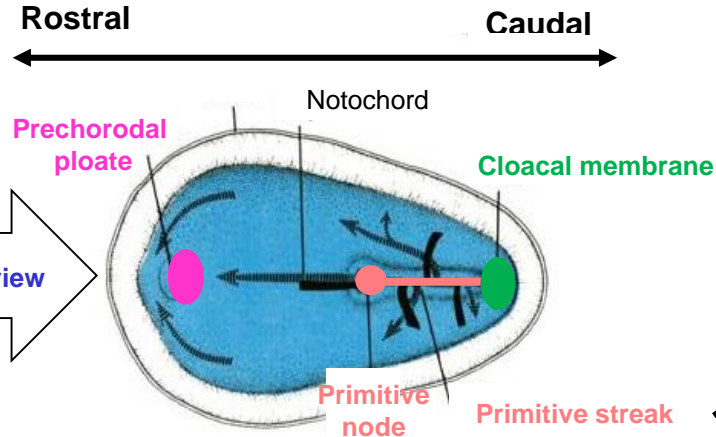
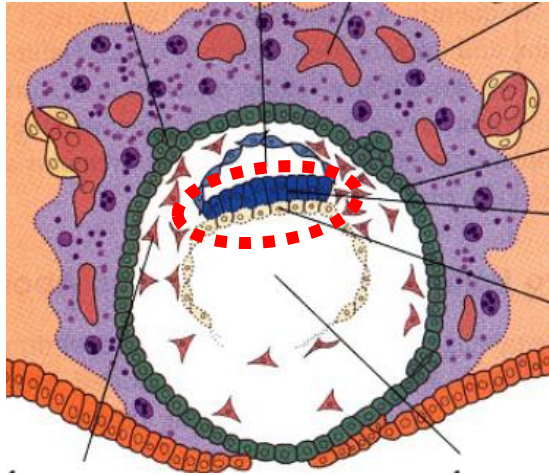
Day 8 to 9



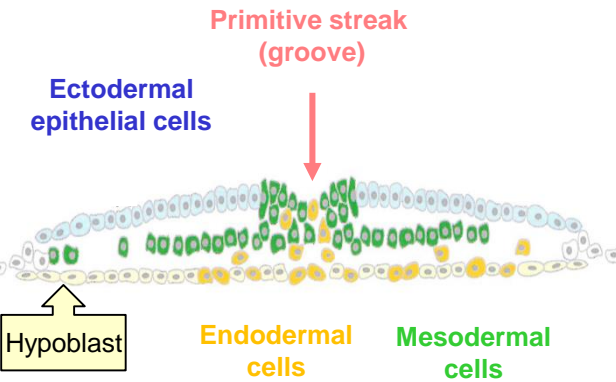


# Gastrulation – establishment of three germ layers

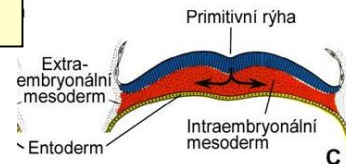
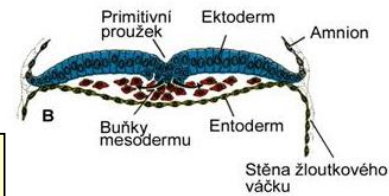
**Embryonic disk** – first at day 6 to 7



Transversal section



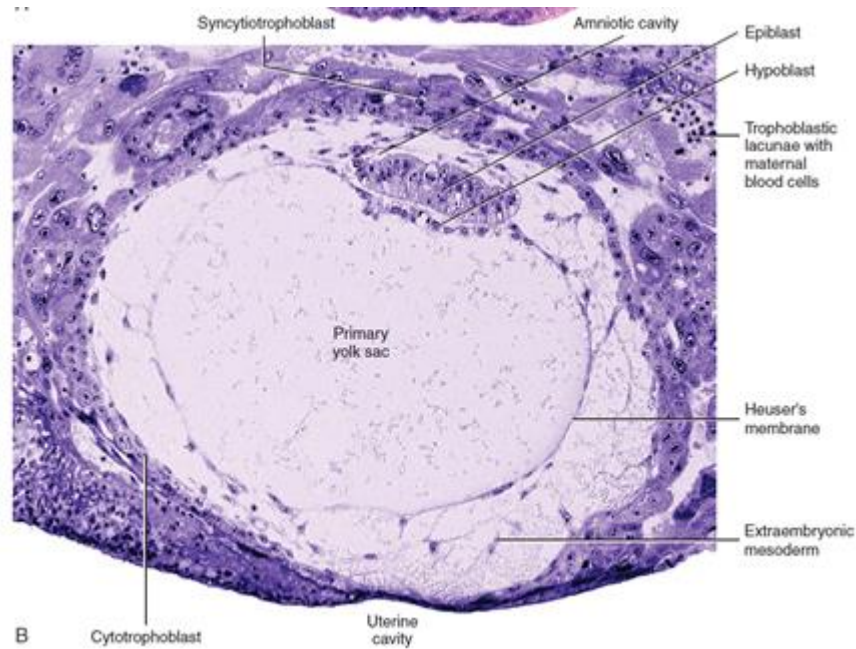
Specification of  
**Mesoderm**  
+ **Endoderm**  
(expanded view)



= epithelio-mesenchymal transition

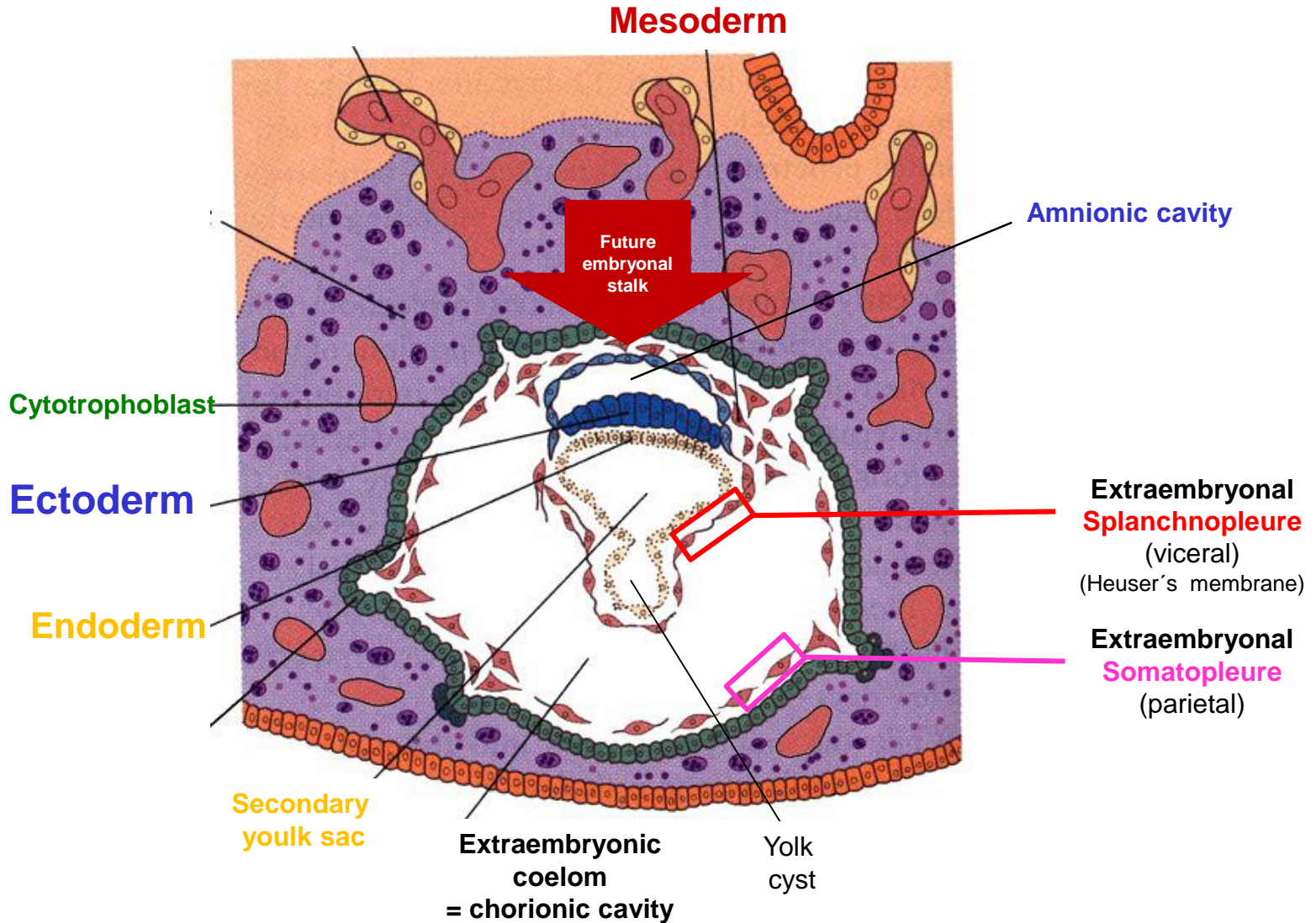
# Gastrulation – establishment of three germ layers

## Day 9 – primary yolk sac



# Gastrulation – establishment of three germ layers

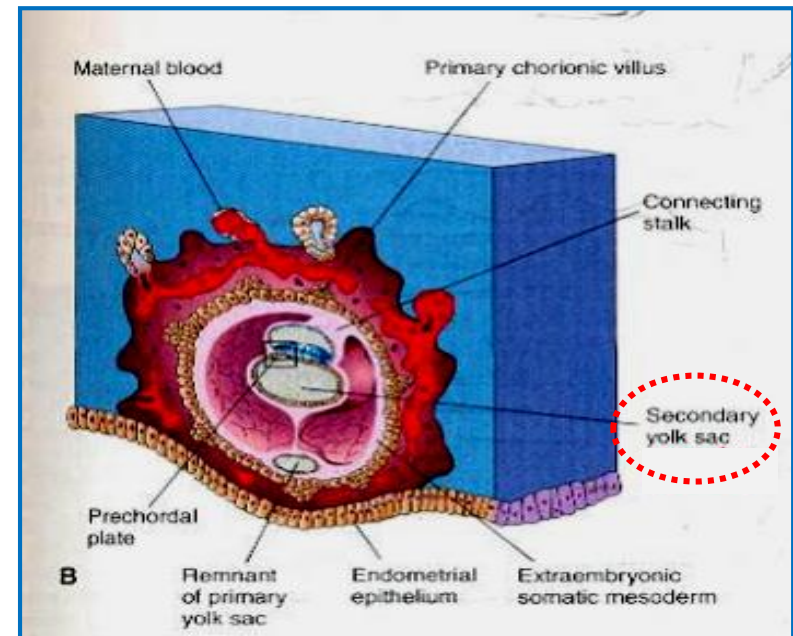
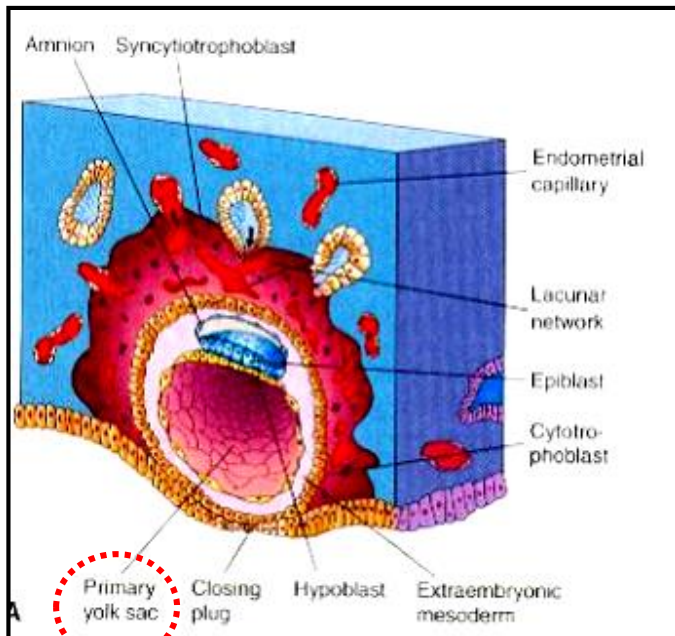
Day 12 - 13





# Extraembryonal structures – yolk sac 1

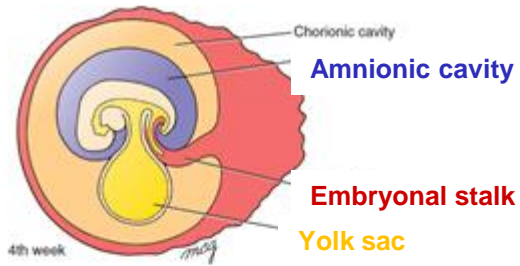
End of the 2. week



Wall of secondary yolk sac = **endoderm** + **mesoderm**

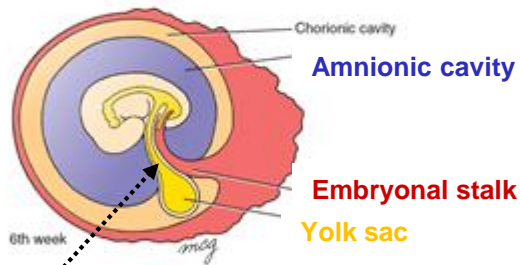
Heuser's membrane

# Extraembryonal structures – yolk sac 2

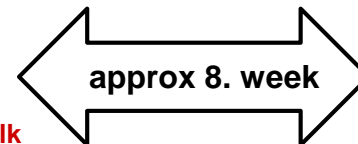
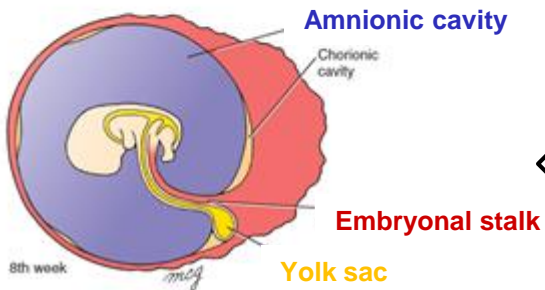
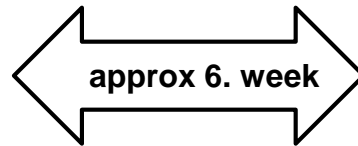


## Functions of yolk sac:

- does not contain yolk (oligolecithal egg)
- 3. week – hematopoiesis (since 6. week in liver)
- 3.- 4. week– PGC
- 4. week – incorporation into primitive gut
- since 6. week – loss of link to gut – obliteration
- abnormal persistence - Meckel diverticle

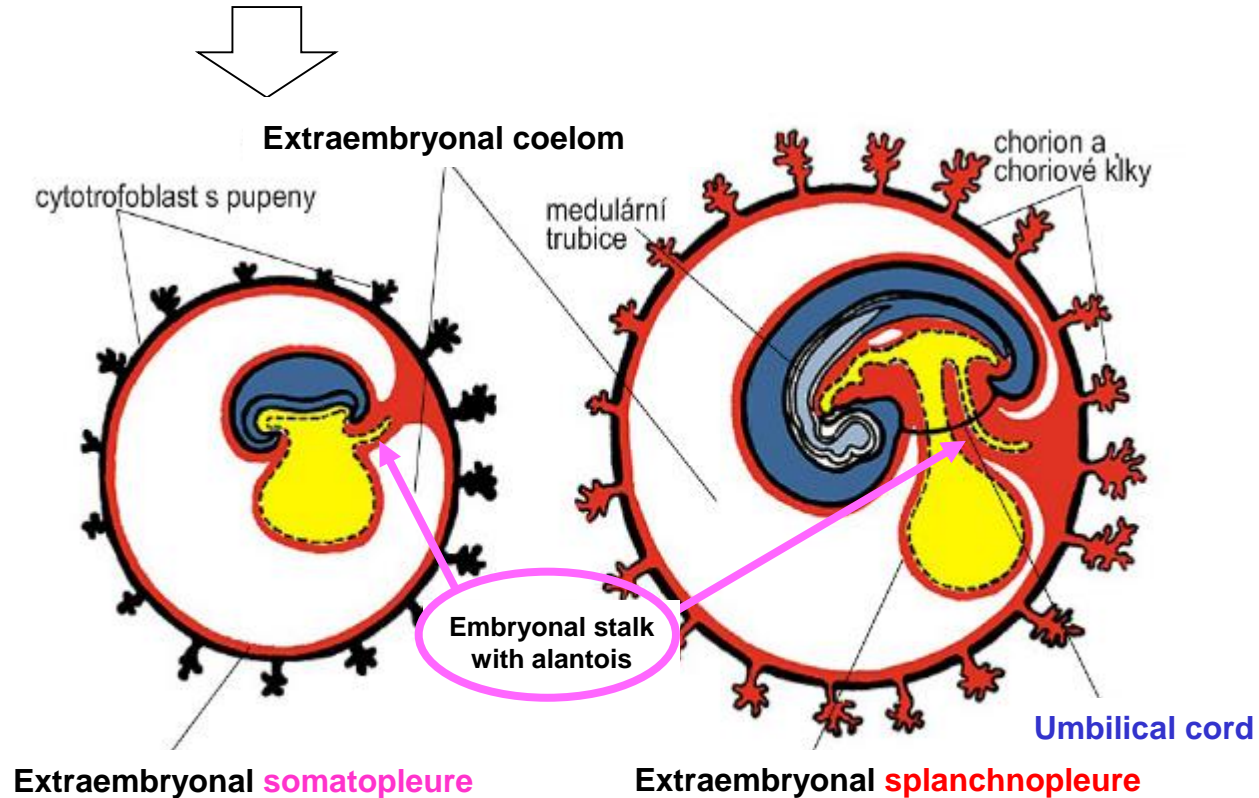


*Ductus omphaloentericus*



# Extraembryonal structures – alantois

Beginning of the 3. week – diverticle of caudal wall of yolk sac

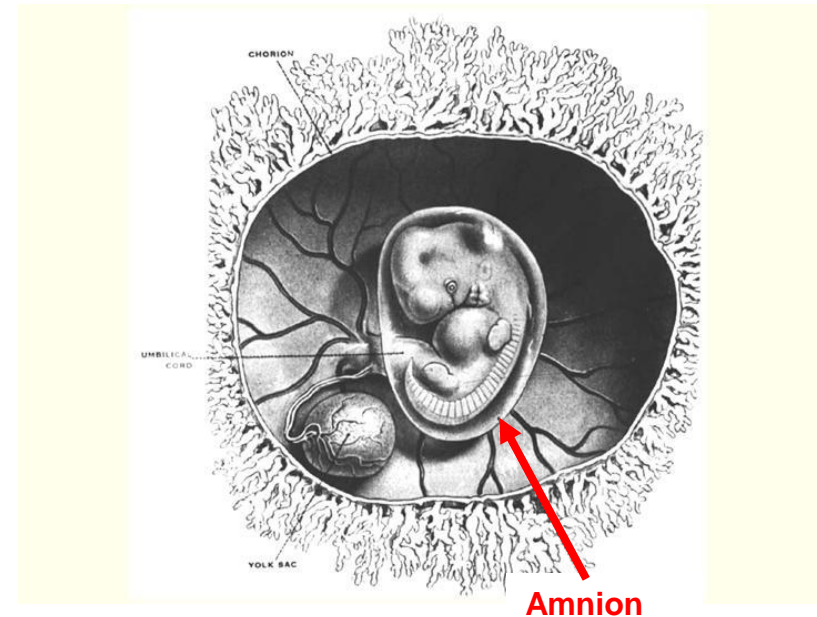
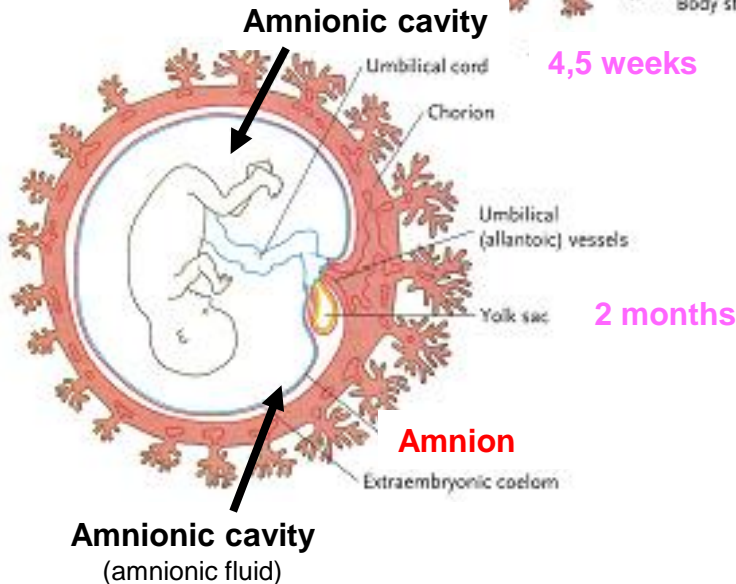
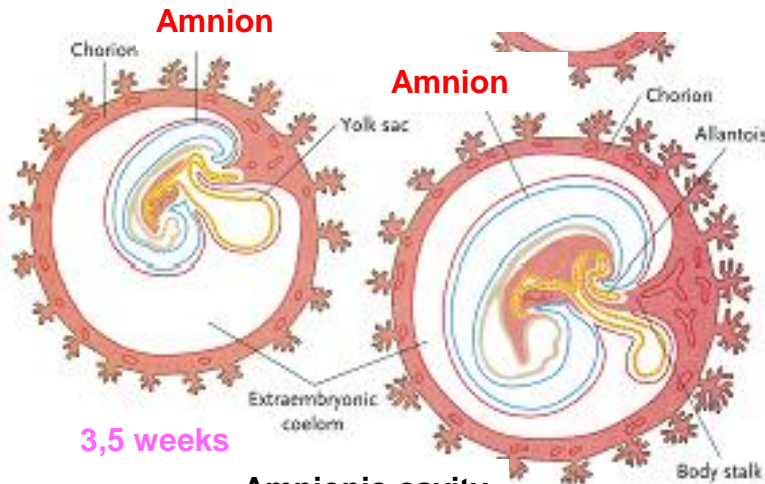


## Functions:

- In humans rudimentary structure
- **vessels of alantois – umbilical vein and arteries**
- 2. month – extraembryonal part degenerates
- 3. month – intraembryonal part – urachus (link to urinary bladder)

# Extraembryonal structures – amnion (internal fetal membrane)

thin, transparent = flat simple ectodermal epithelium + extraembryonal mesoderm (somatopleure)

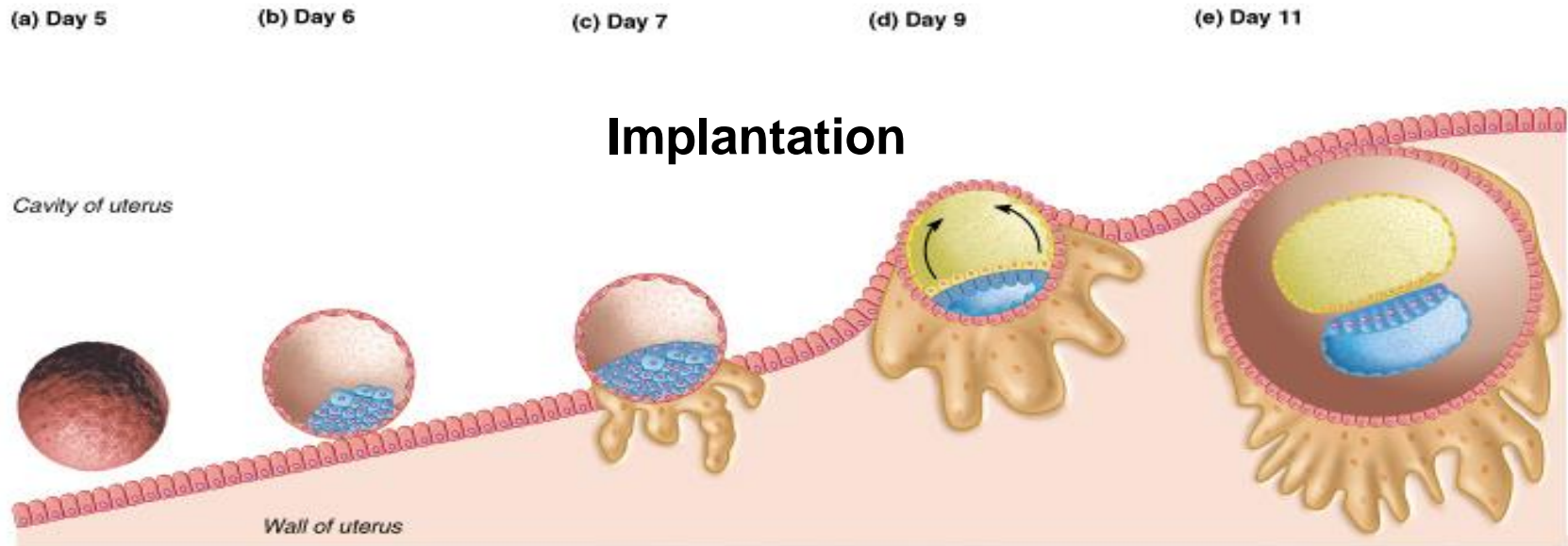


## Amniotic fluid:

- 99% water; 800 až 1000 ml in the last month of pregnancy
- source = diffusion from endometrium through amniochorion
- source = transfer through placenta
- source = transfer through skin of fetus
- source = excretion by fetal kidneys (since 11. week)
- very fast turnover – passage via fetus (digestive + respiratory tracts)
- function – free movement without adhesion (space for symmetrical growth)
- function – barrier (temperature, mechanical damage, infection)
- function – chemical homeostasis



# Extraembryonal structures – chorion (external fetal membrane)



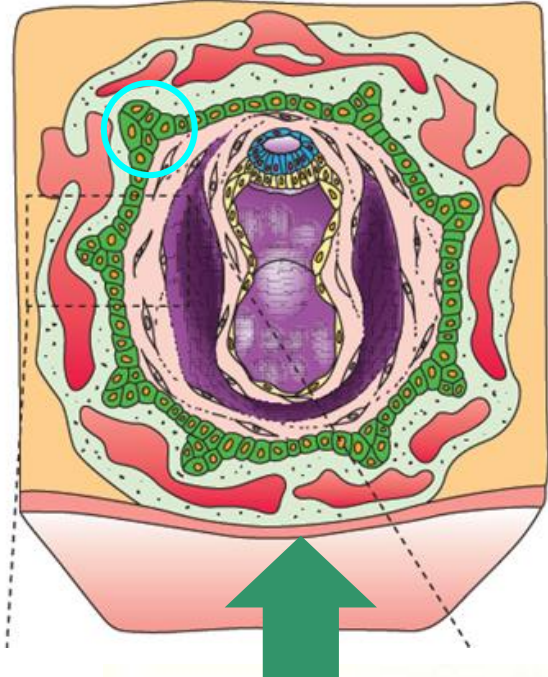
Copyright © Pearson Education, Inc., publishing as Benjamin Cummings.

Syncytiotrophoblast invades into surrounding stroma

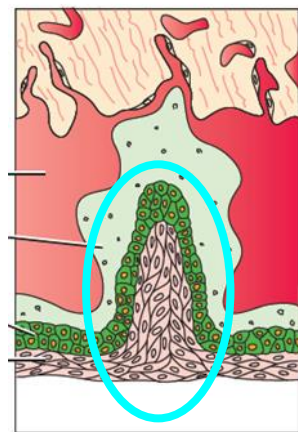


# Extraembryonal structures – chorion – chorionic villi

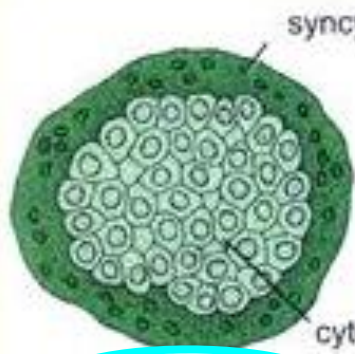
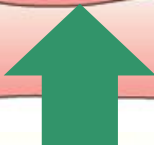
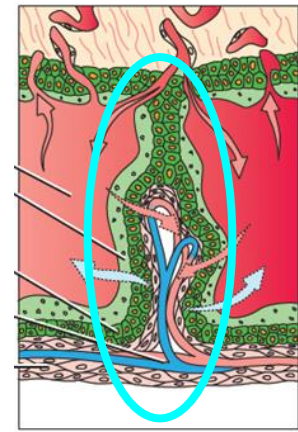
Day 11 až 13



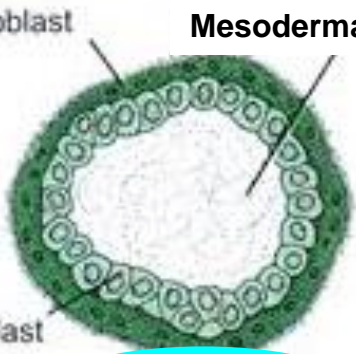
Day 16



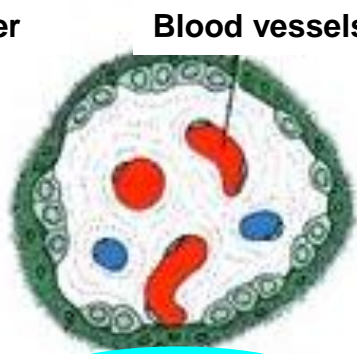
End of the 3. week



primary villus



secondary villus



tertiary villus

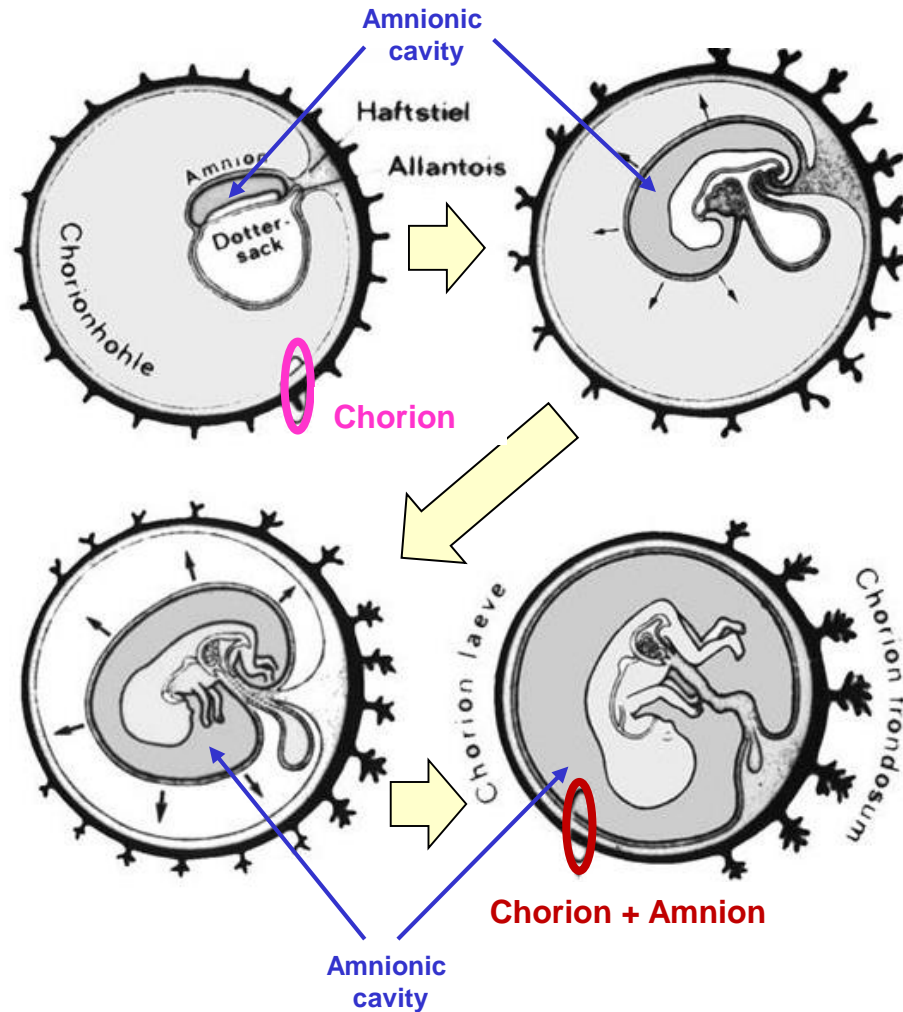
syncytiotrophoblast

Mesodermal center

Blood vessels

cytotrophoblast

# Extraembryonal structures – chorion – expansion of amnion

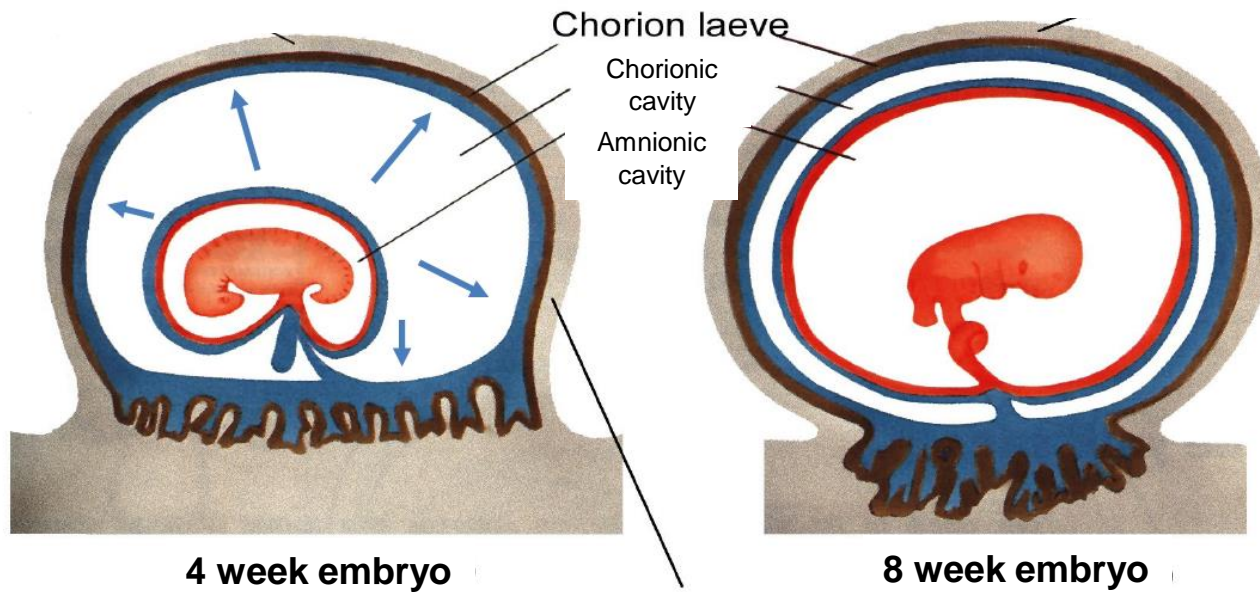


# Extraembryonal structures – growth of amnion and chorion

**CHORION** = cytotrofoblast + mezoderm (ex.)

**AMNION** = mezoderm (ex.) + ektoderm

## Growth of amnionic and chorionic cavities

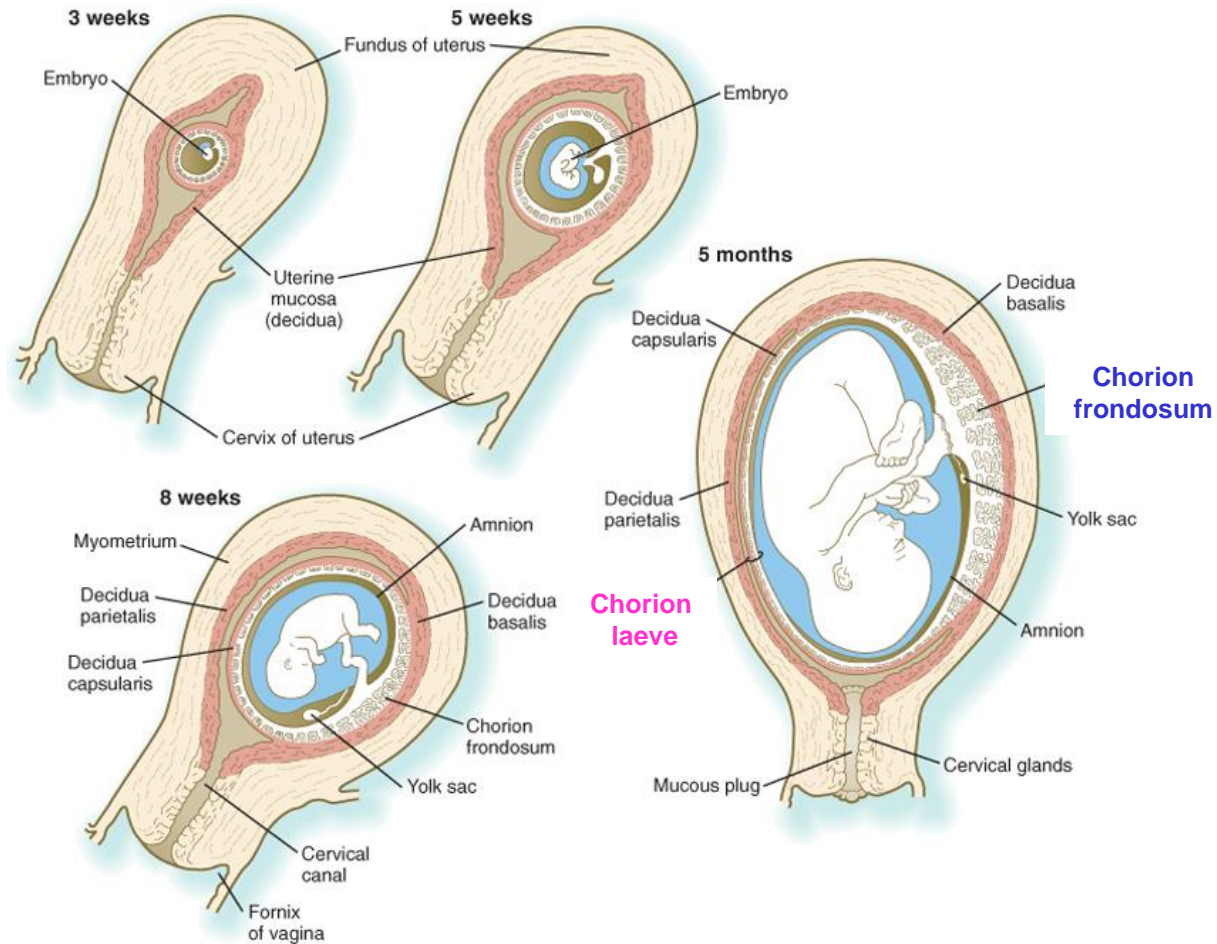




# Extraembryonal structures – chorion – *frondosum x laeve*

*Ch. frondosum – vilous*

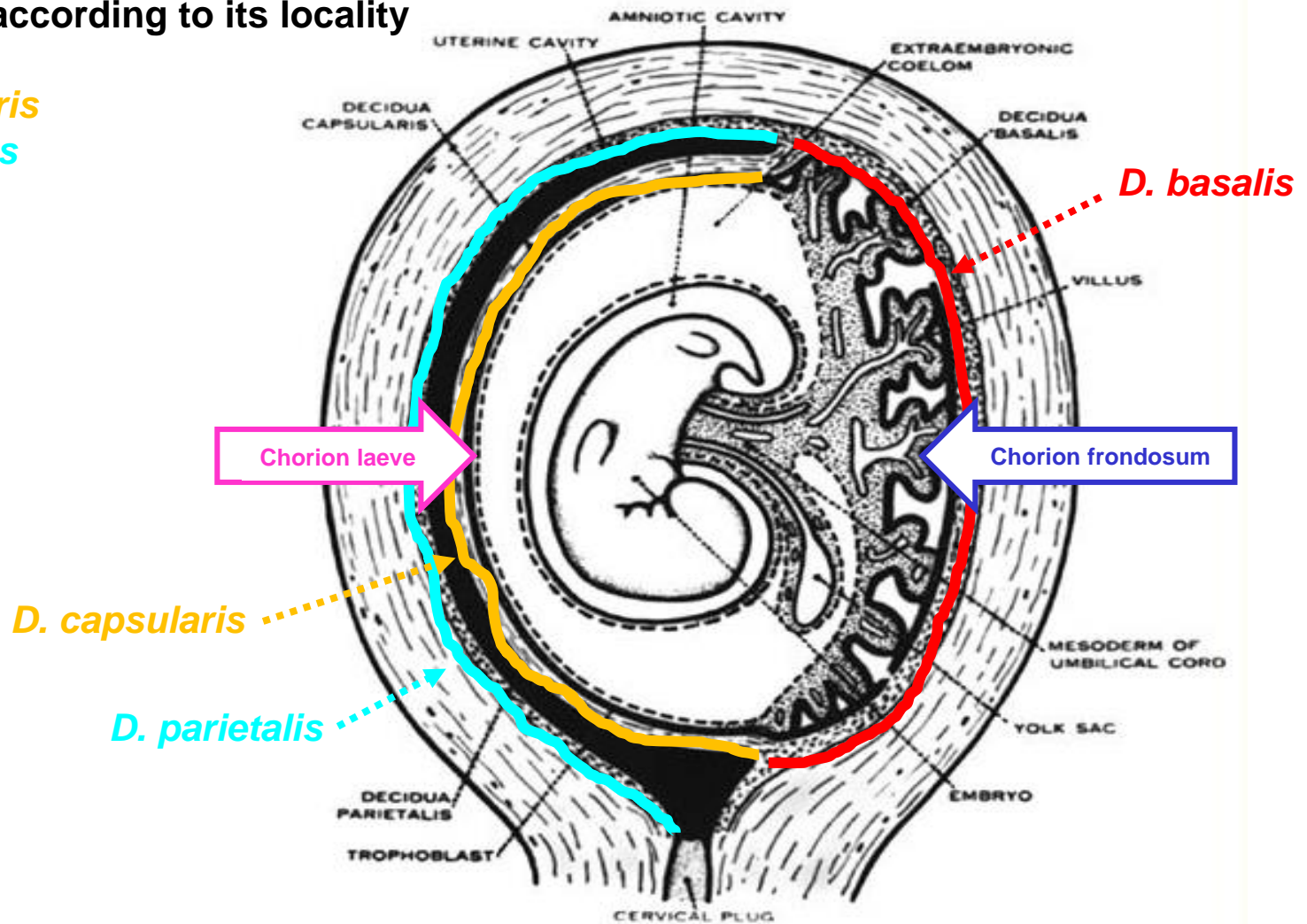
*Ch. laeve - smooth*



# Extraembryonal structures – chorion – decidua

Decidua – according to its locality

- *basalis*
- *capsularis*
- *parietalis*



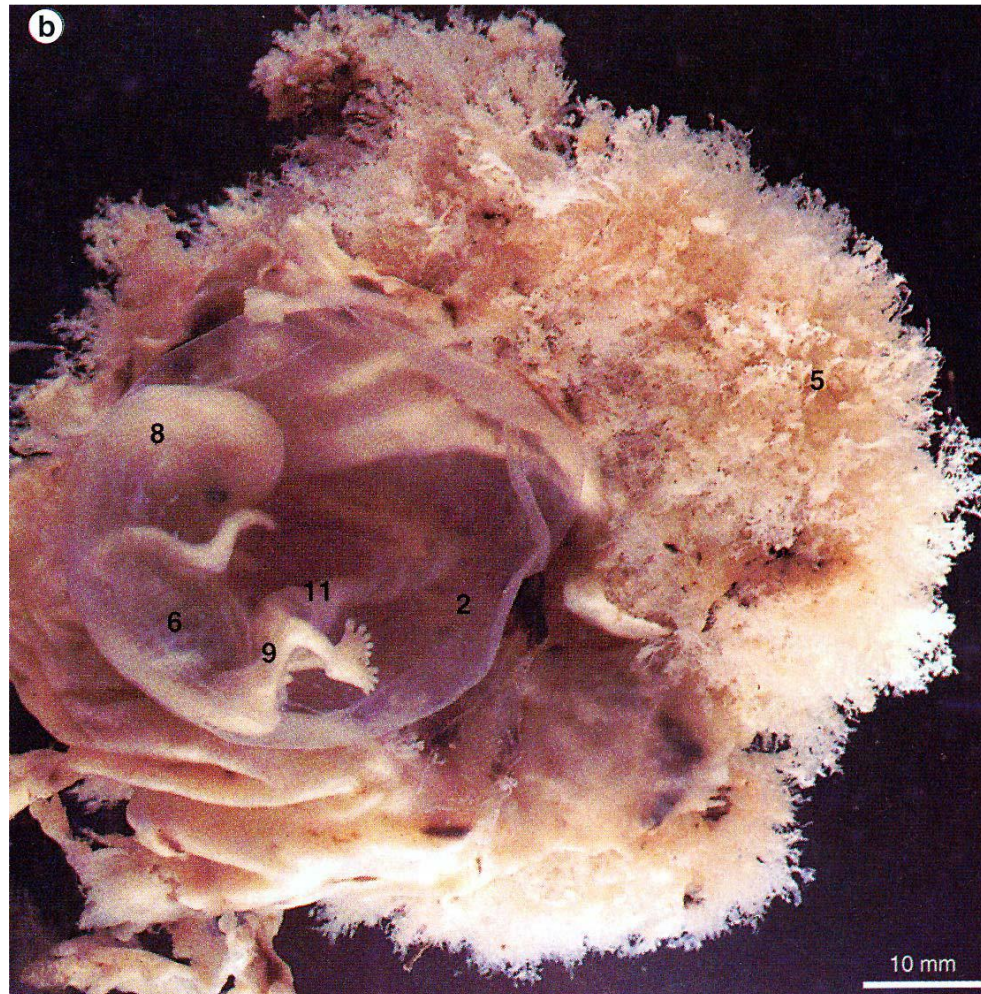
*Decidua basalis* – between embryo and myometrium

*Decidua capsularis* – between embryo and uterine cavity (becomes thinner)

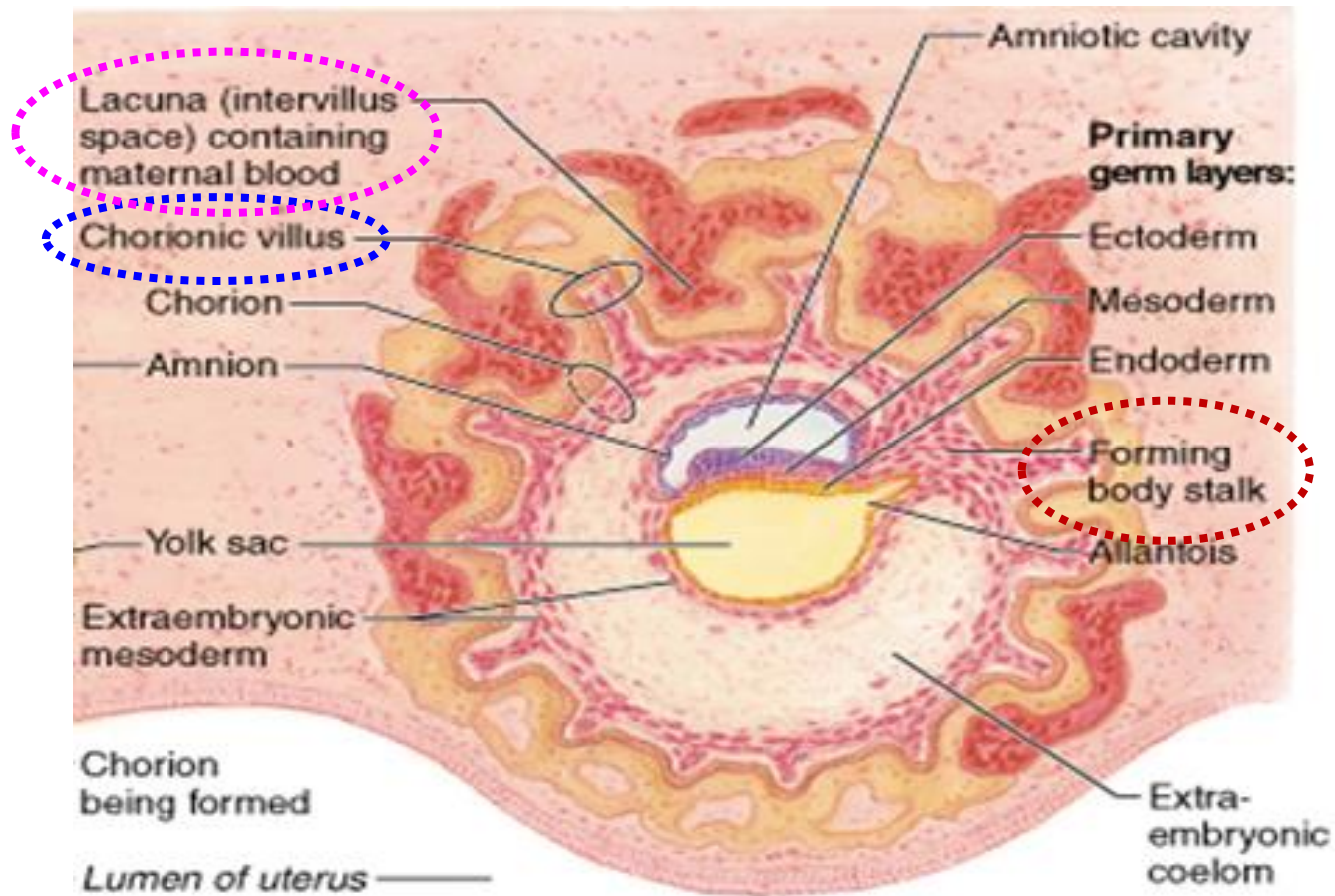
*Decidua parietalis* – opposite wall of uterus



# Extraembryonal structures – chorion – decidua



# Extraembryonal structures – chorion – placenta



(c) 16-day embryo

**Chorionic villi** - finger like projection of embryonic tissue that come in contact with bleeding endometrium

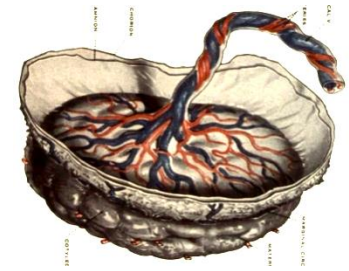
**Decidual cells** – fibroblast of endometrium (large, cuboidal, very active proteosynthesis)

**Placenta** – thick disk made by decidua and chorionic villi (formed at the start of month 4)



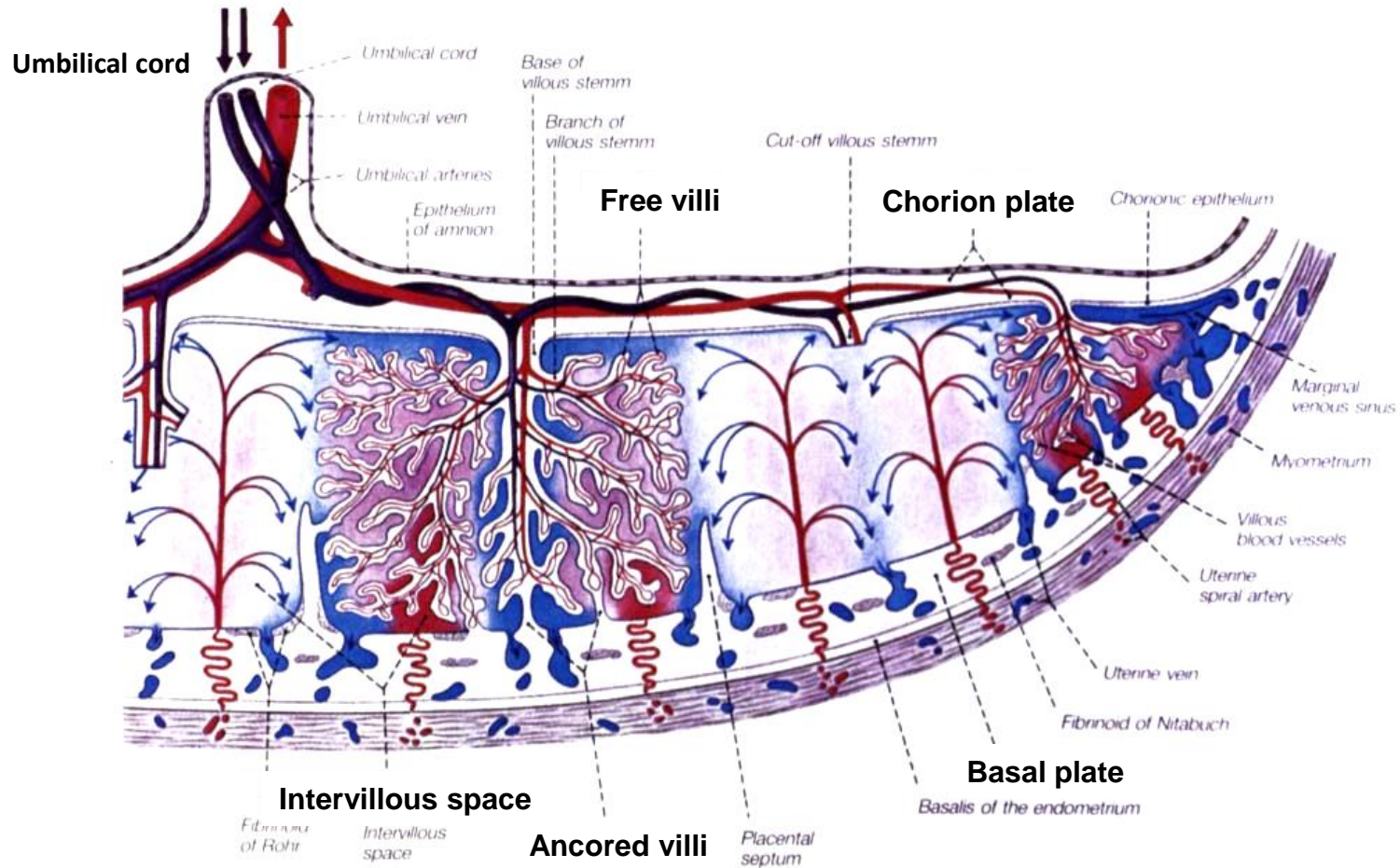
discoid  
15 – 20 cm  
400 – 600 g

# Placenta



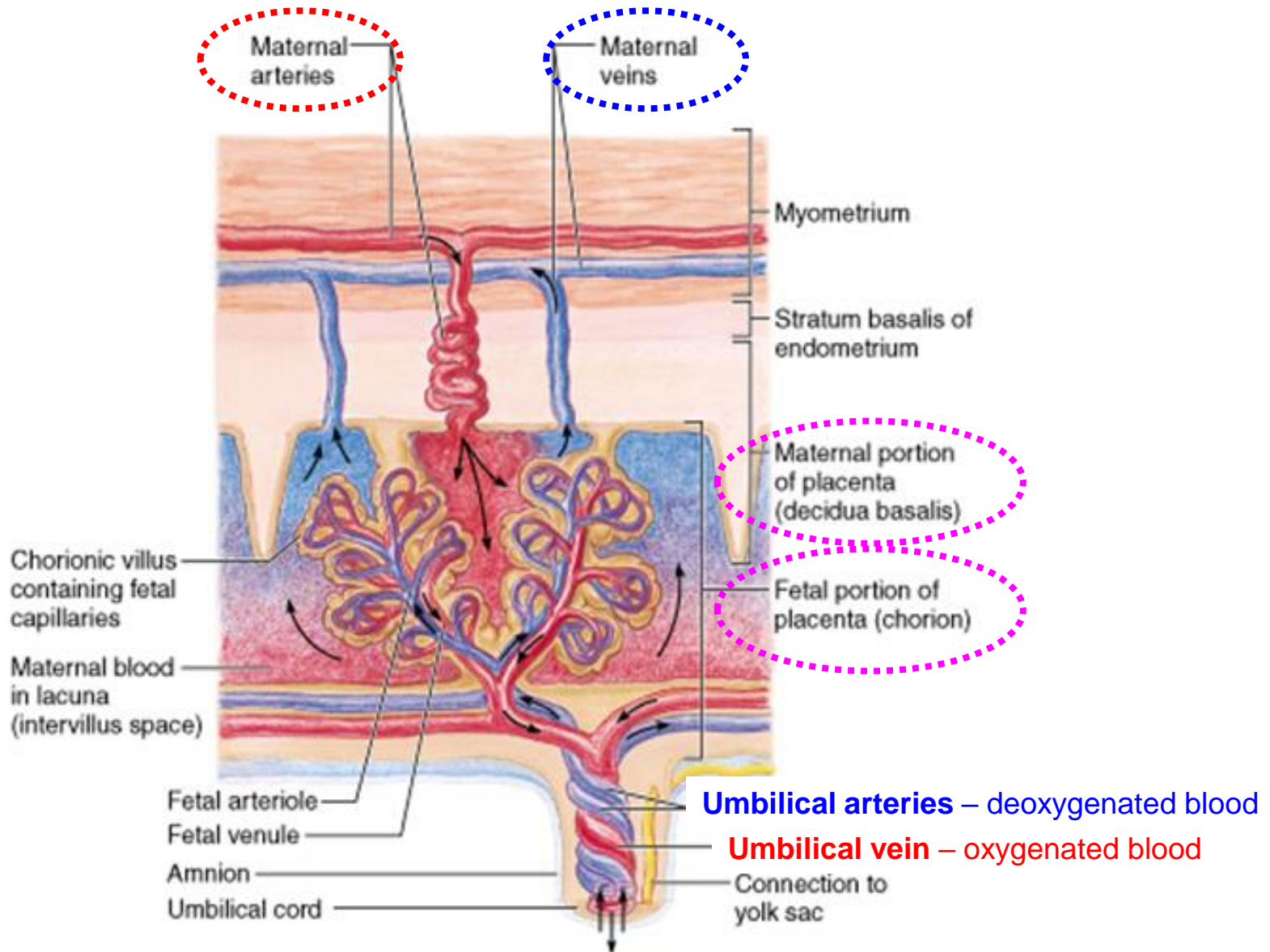
## Discoidalis + Hemochorialis

- **pars fetalis** – chorion plate, chorion villi (anchored, free = terminal)
- **pars materna** – decidua basalis
- **intervillous spaces** – develop from lacunes

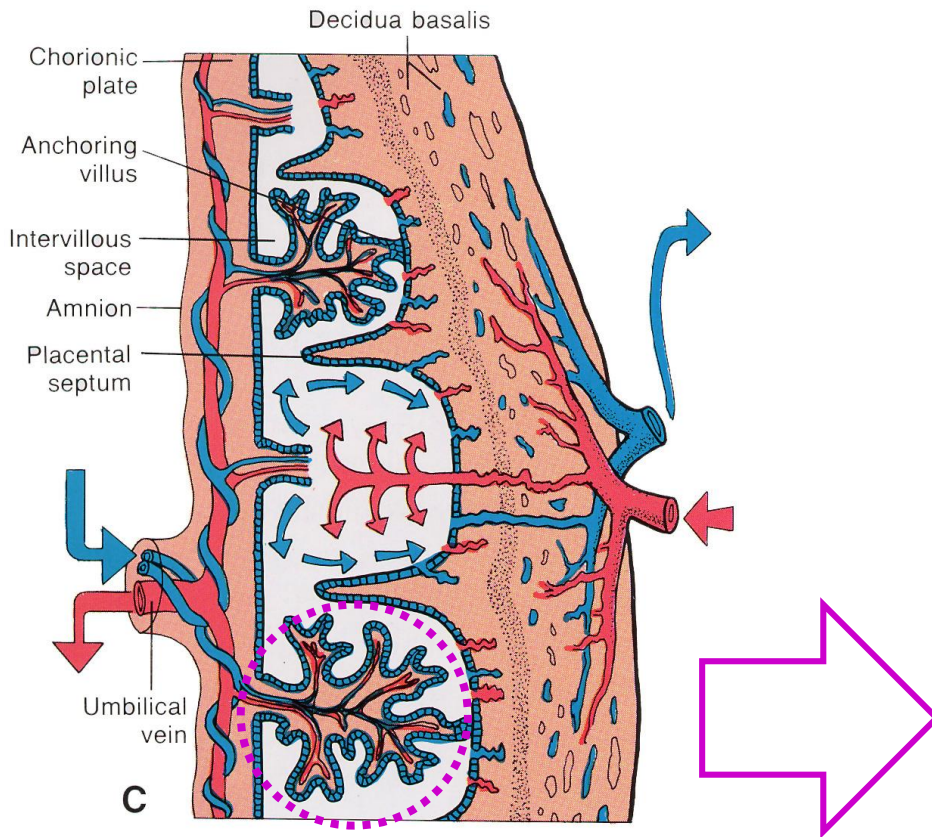




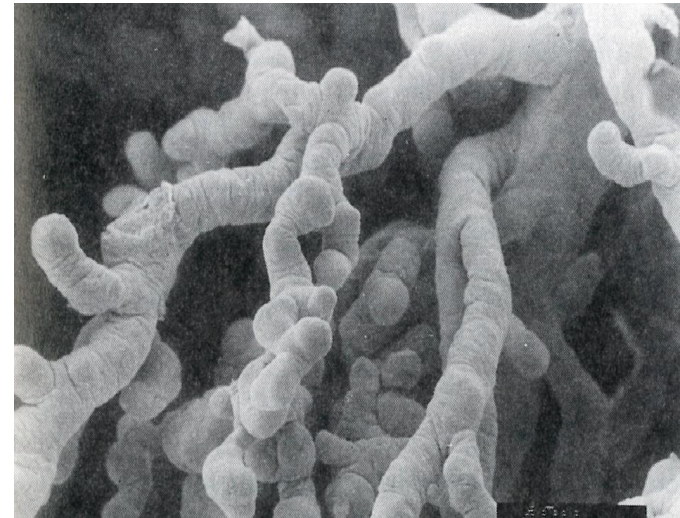
# Placenta – blood circulation



# Placenta – terminal villi

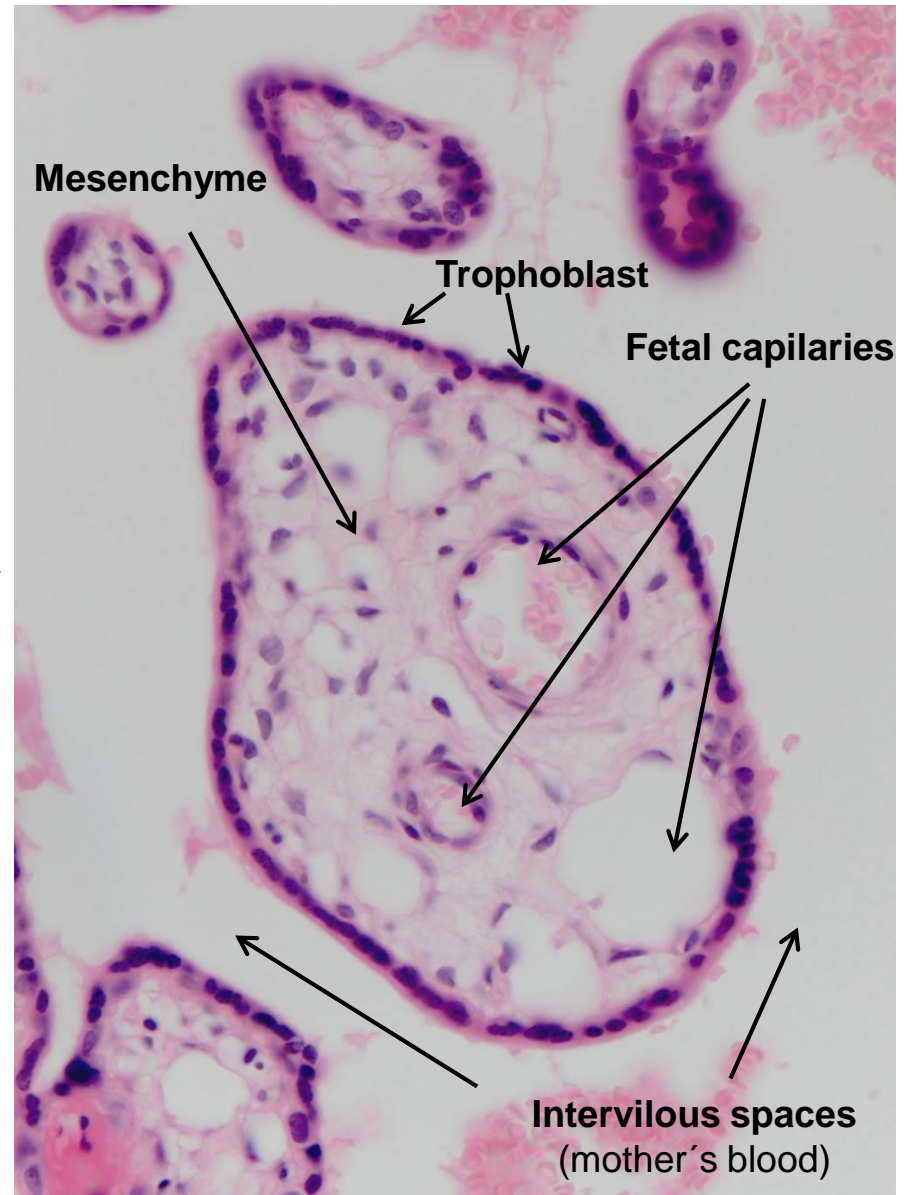
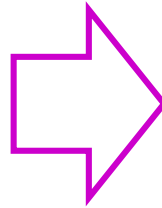
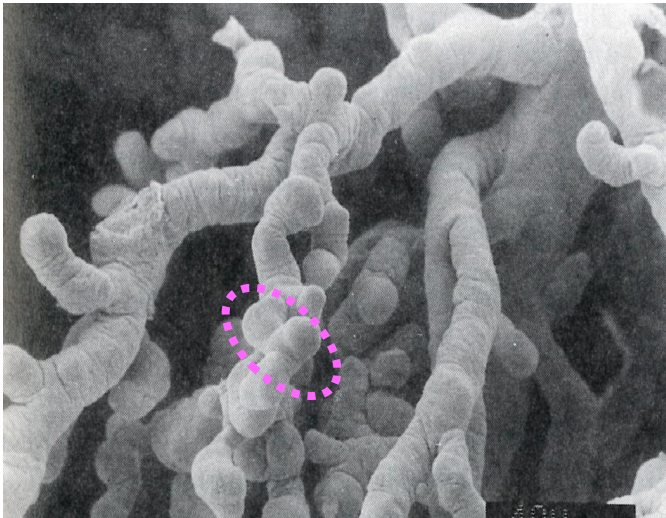


**Terminal villi – human – end of pregnancy**



# Placenta - fetomaternal barrier 1

Terminal villi – human – end of pregnancy



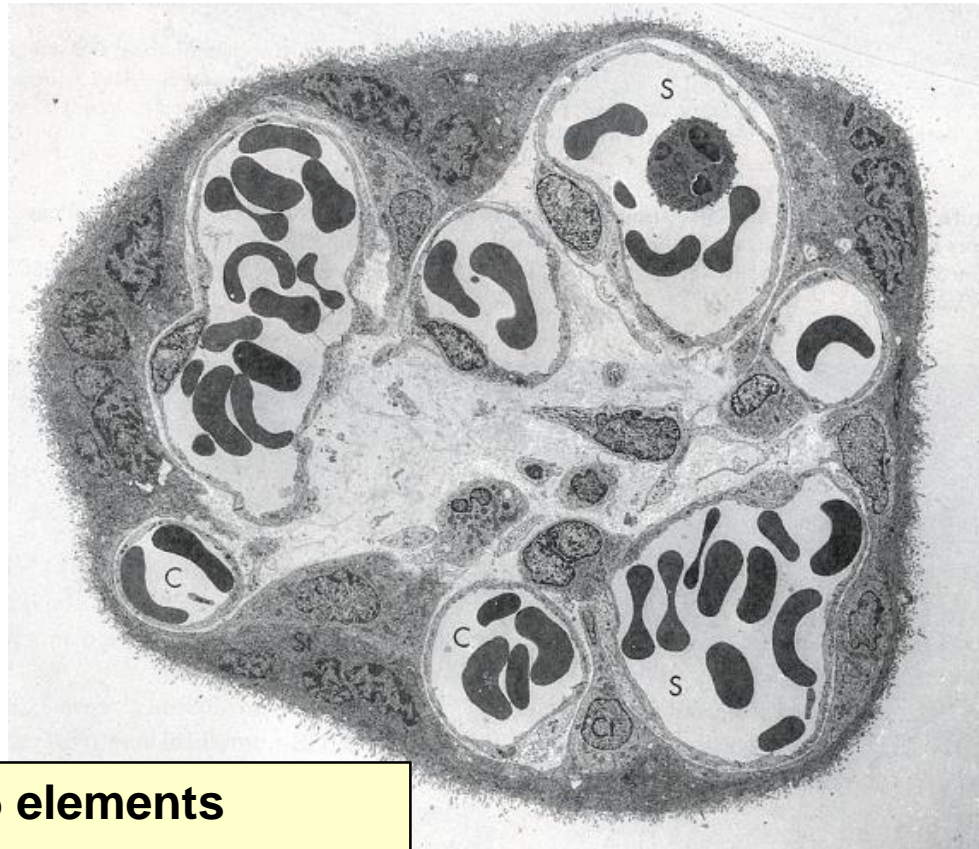
## Barrier = 5 elements

- Endothelium of fetal capillaries
- Basal membrane of endothelium
- *Mesenchyme of villi* (extraembr. mesoderm)
- Basal membrane of trophoblast cells
- Cells of cyto- a syncytio-trophoblast

since month 5 cytotrophoblast loses its continuity



## Placenta - fetomaternal barrier 2

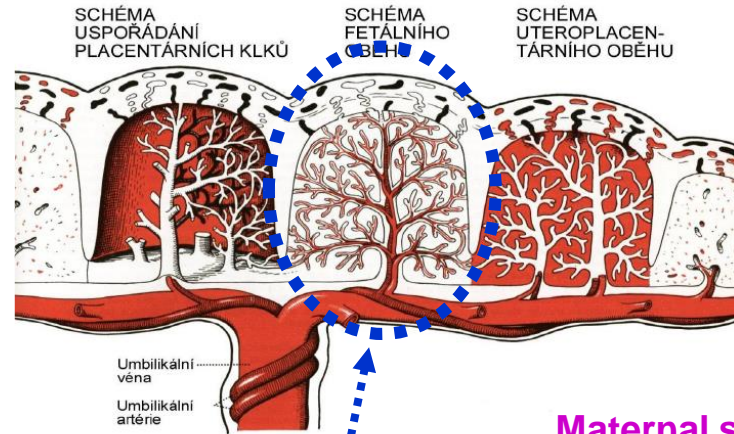


### Barrier = 5 elements

- Endothelium of fetal capillaries
- Basal membrane of endothelium
- *Mesenchyme of villi* (extraembr. mesoderm)
- Basal membrane of trophoblast cells
- Cells of cyto- a syncyio-trophoblast

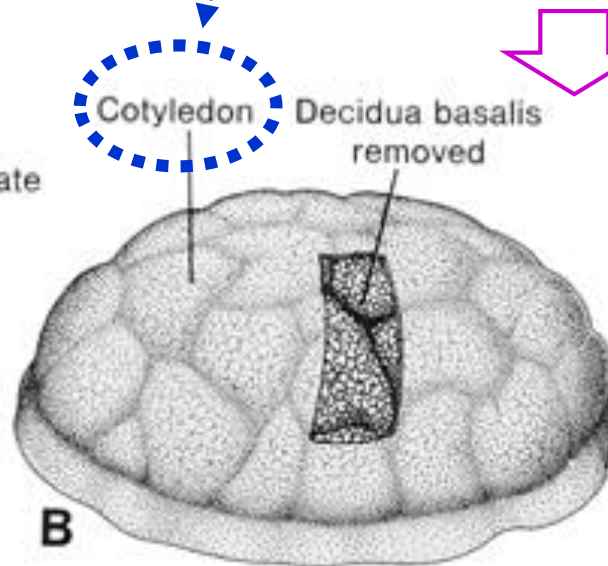
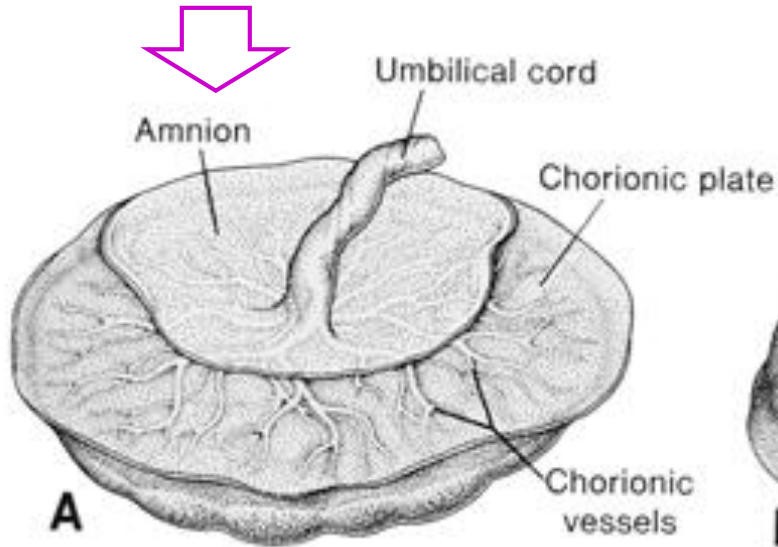
since month 5 cytotrophoblast loses its continuity

# Placenta - cotyledons

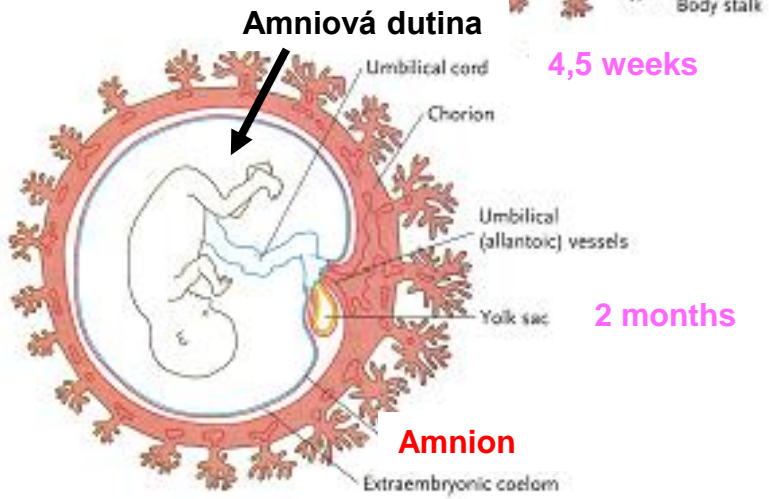
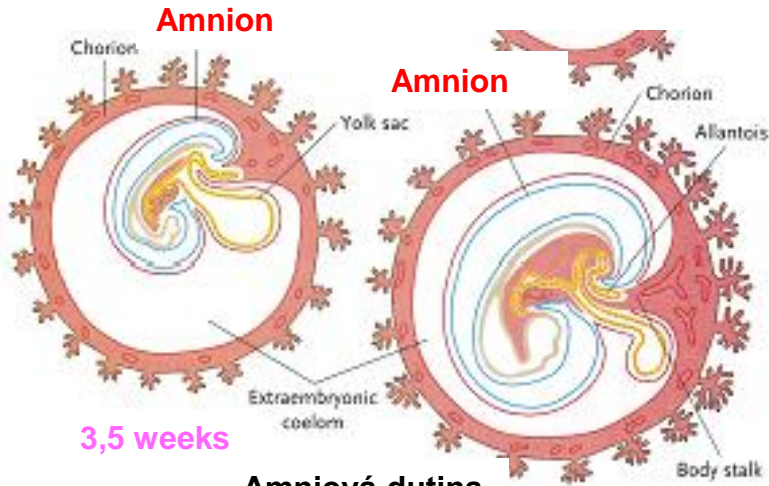


Fetal surface

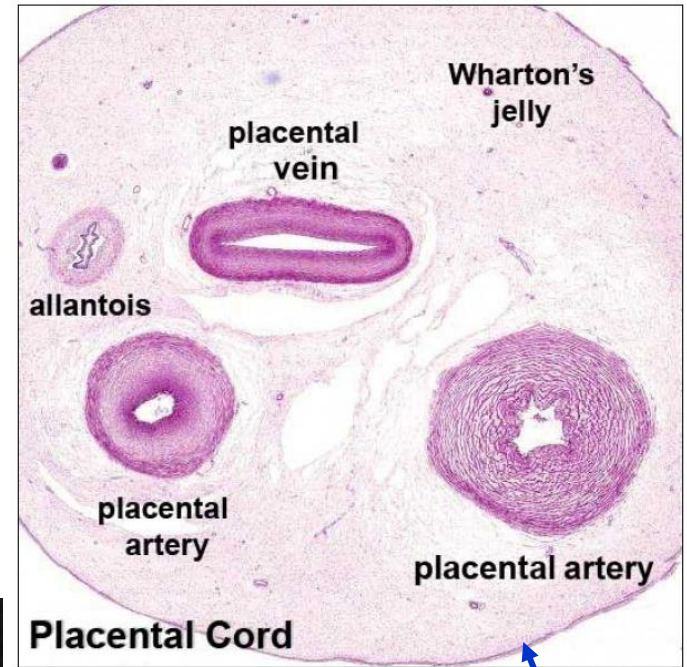
Maternal surface



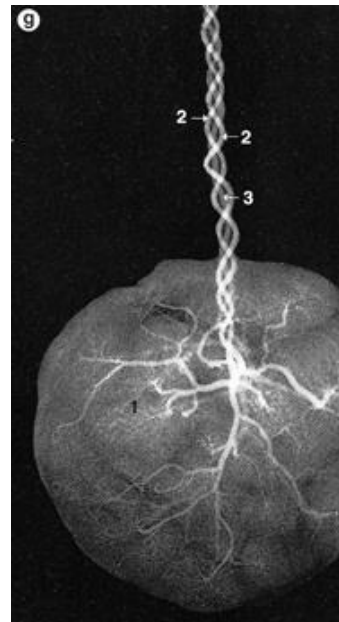
# Placenta – umbilical cord 1



**Amniotic cavity**  
(amniotic fluid)



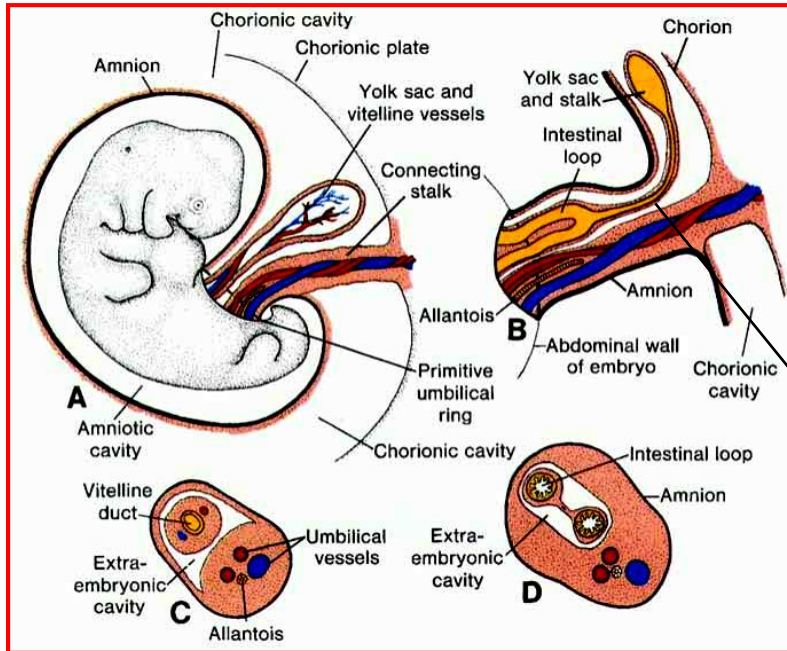
**Amnionic ectoderm**



- diameter 1,5 to 2 cm
- length 50 to 60 cm
- 1x vein + 2x artery (spiral organization)
- Wharton's jelly – loose connective tissue



# Placenta – umbilical cord 2



## 1-Connecting stalk:

### Allantois

Umbilical vessels (two arteries & one vein), they all embedded in

Wharton's jelly (extra embryonic mesoderm)

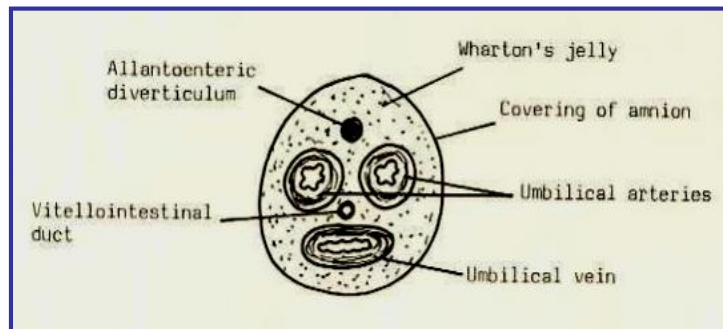
## 2-Yolk stalk (Vitello-intestinal duct):

*(Ductus omphaloentericus)*

A narrow, elongated duct which connects gut to yolk sac

It contains **Vitelline Vessels**

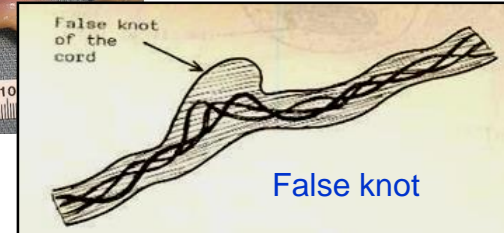
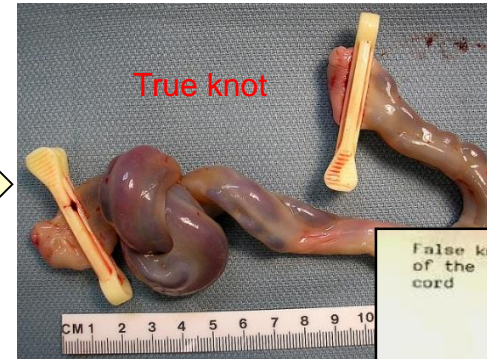
(Later on , it is obliterated and the vitelline vessels disappear).



# Umbilical cord - anomalies

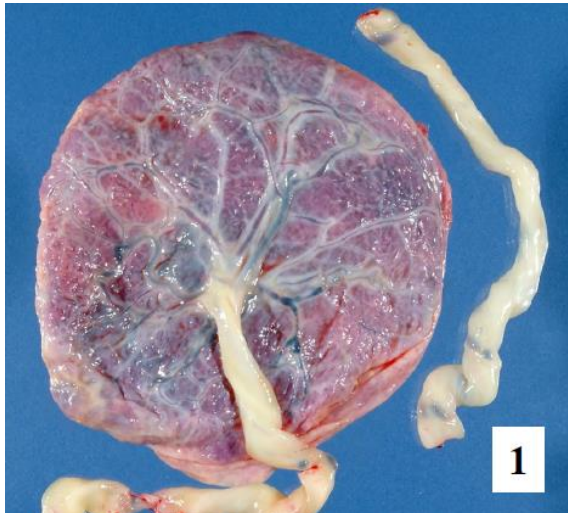
- Short umb. cord < 40 cm
- Long umb. cord > 60 cm
- Absence of one artery – fetal hypotrophy

**True knot**  
Fetal strangulation  
Umbilical prolapsus

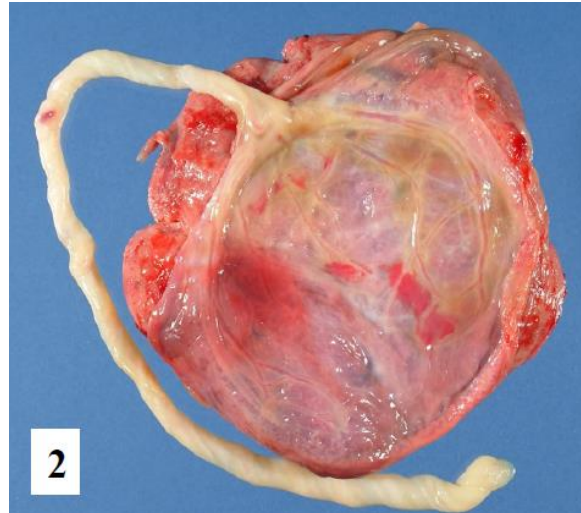


## Attachment of umbilical cord to placenta

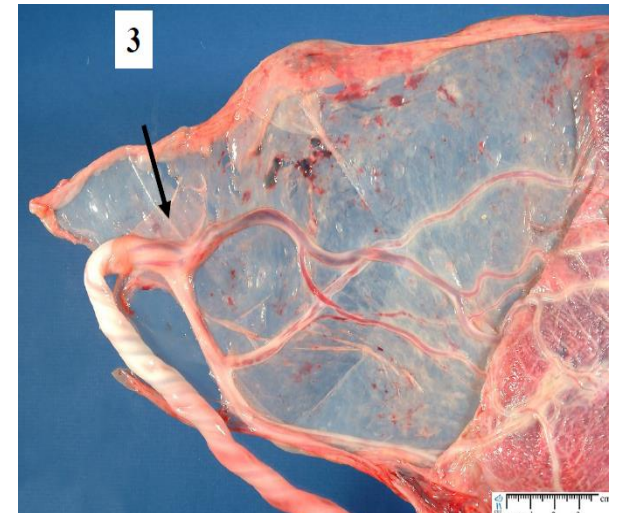
*Insertio centralis* (norm)



*Insertio marginalis*



*Insertio veluminosa* (to chorion laeve)

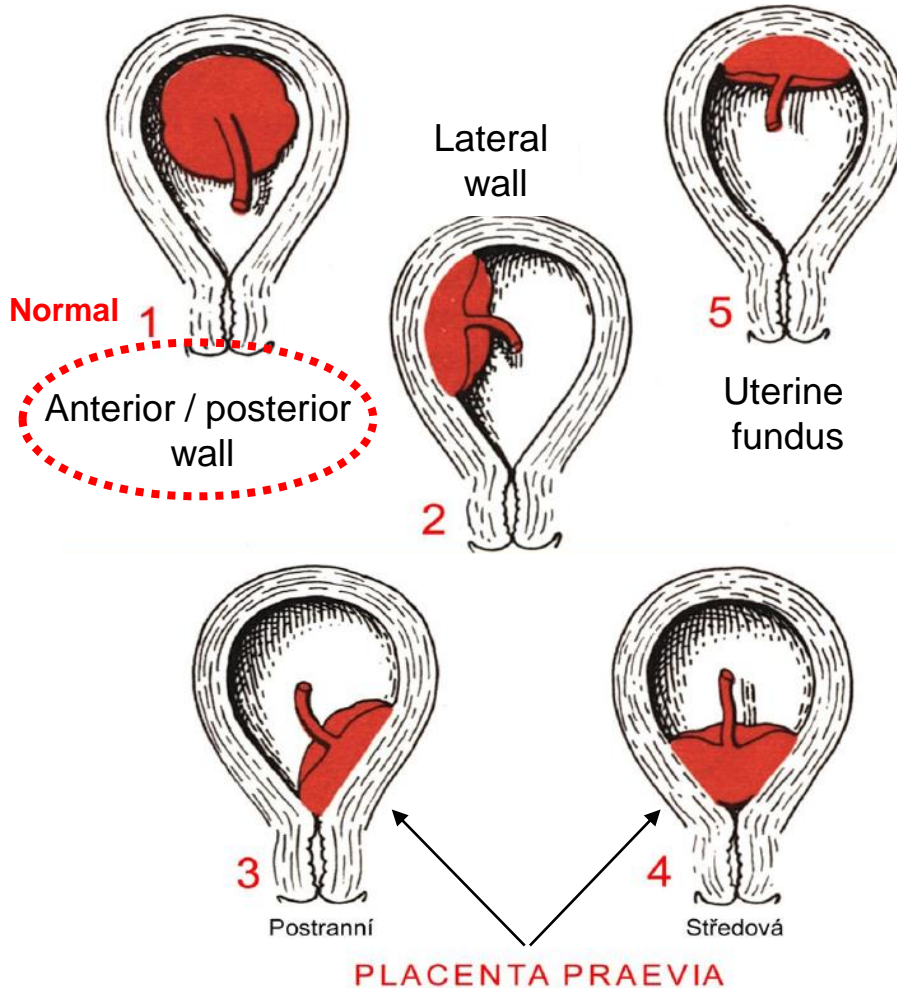




# Placenta – anomalies 1

## Location of placenta in uterus

(1 to 5 according to frequency)



## Attachment of placenta

(related to myometrium)

- **Placenta accreta**

attached to myometrium

- **Placenta increta**

grown into myometrium

- **Placenta percreta**

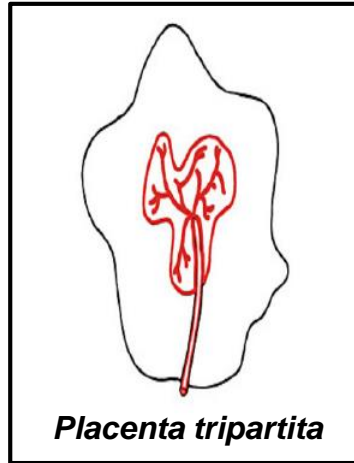
grown through myometrium

# Placenta – anomalies 2

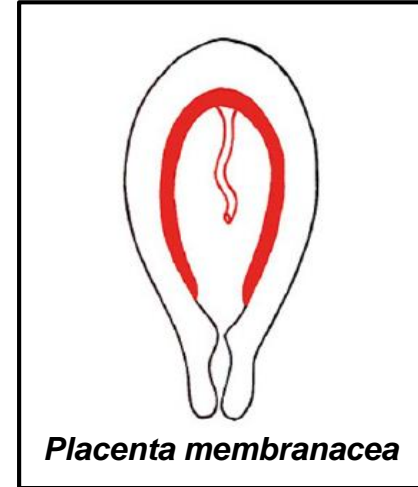
## Shape and formation of placenta



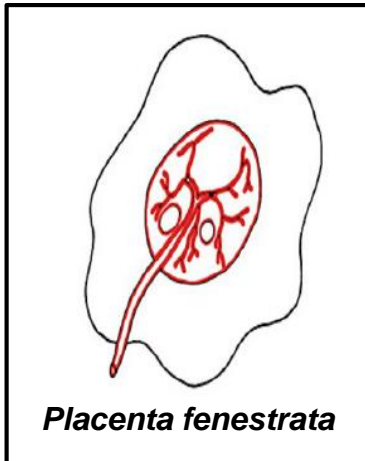
Normal placenta



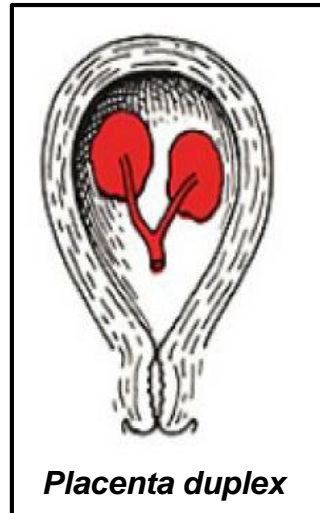
*Placenta tripartita*



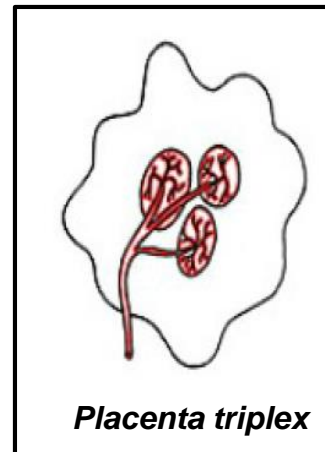
*Placenta membranacea*



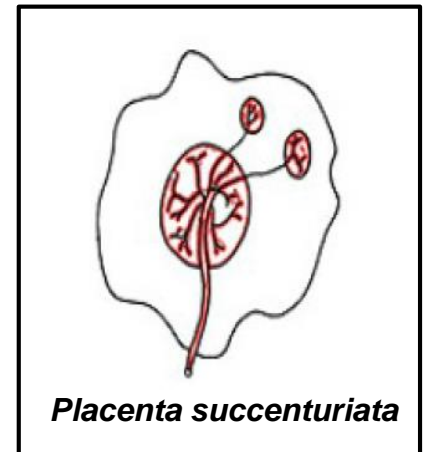
*Placenta fenestrata*



*Placenta duplex*



*Placenta triplex*



*Placenta succenturiata*

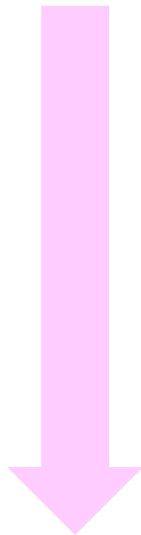
# Placenta – multiparous pregnancy 1

## TWINS

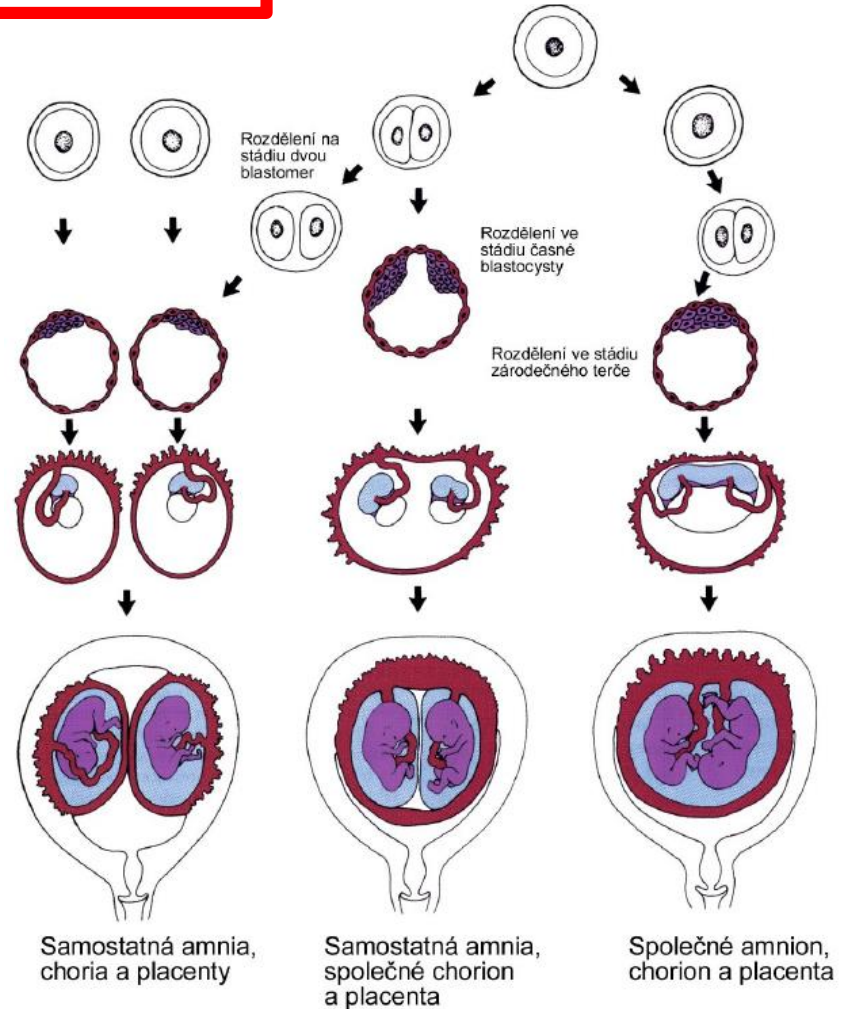
**Dizygotic**

**Monozygotic**

2 oocytes + 2 sperms

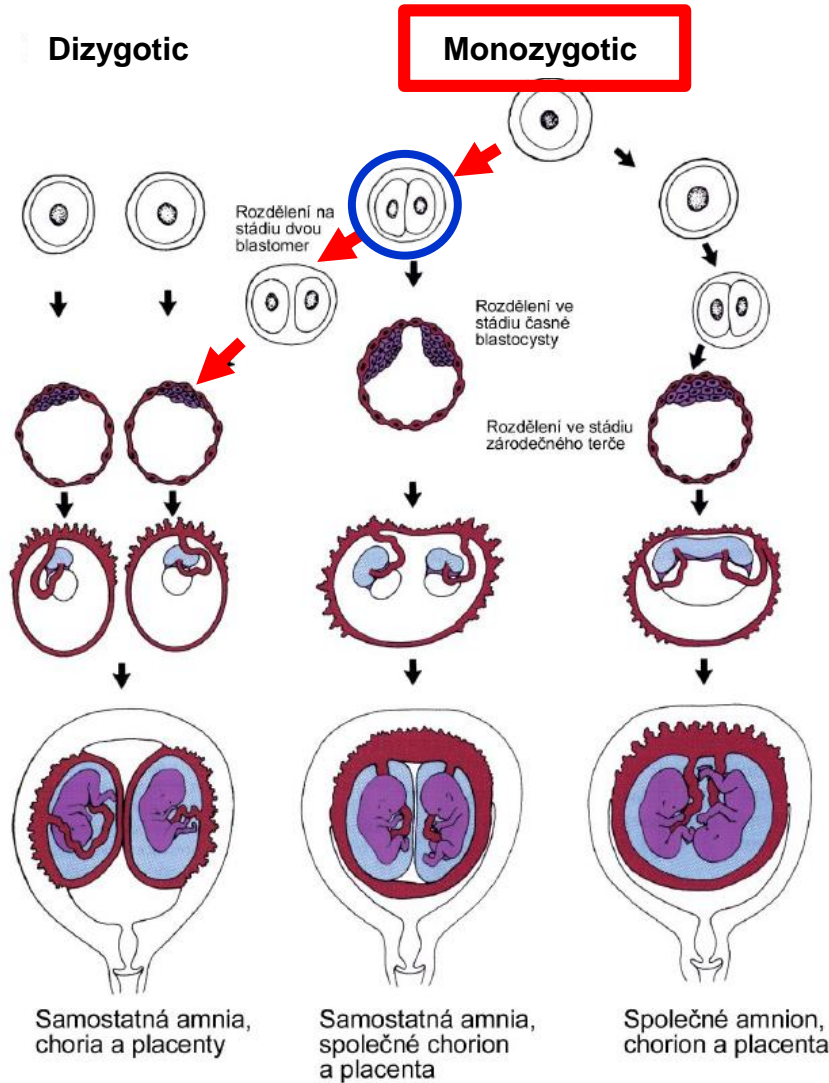


2x amnion + 2x chorion + 2x placenta



# Placenta – multiparous pregnancy 2

## TWINS



1 oocyte + 1 sperm

Embryo splits at 2-cell stage

2x amnion + 2x chorion + 2x placenta

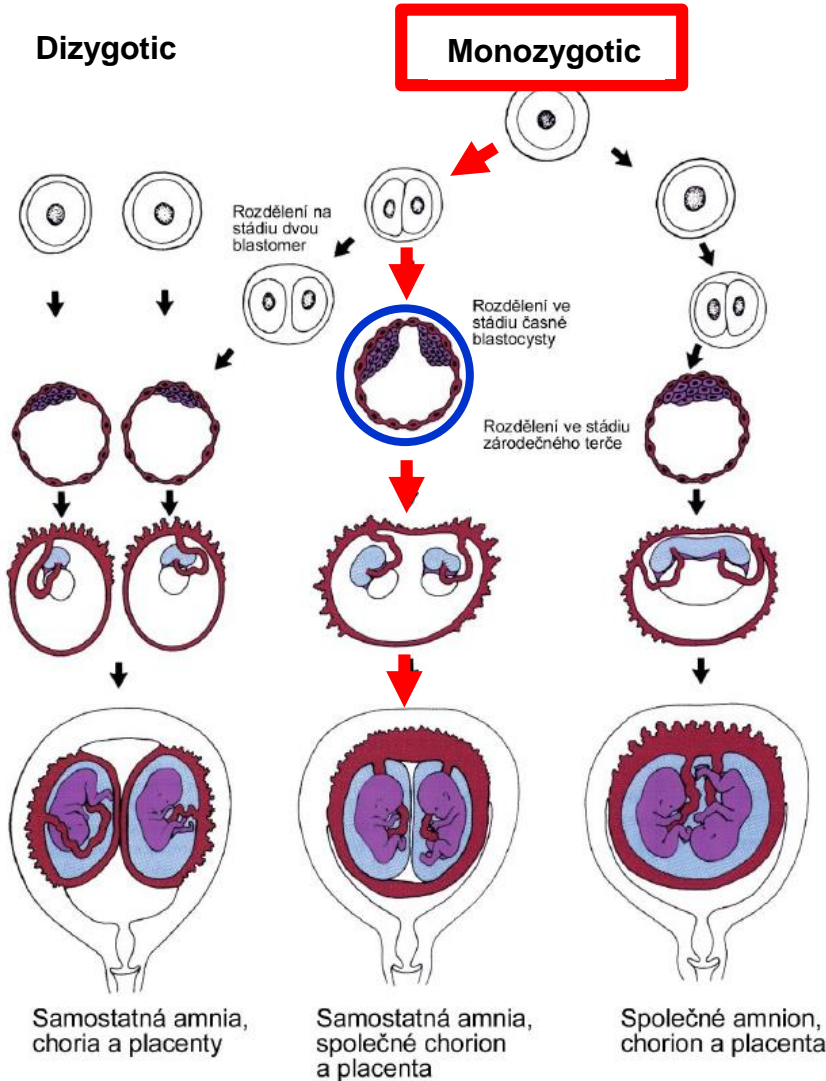
(as dizygotic twins)



# Placenta – multiparous pregnancy 3

## TWINS

Most frequent twins – 65 %



1 oocyte + 1 sperm

Embryo splits at blastocyst stage

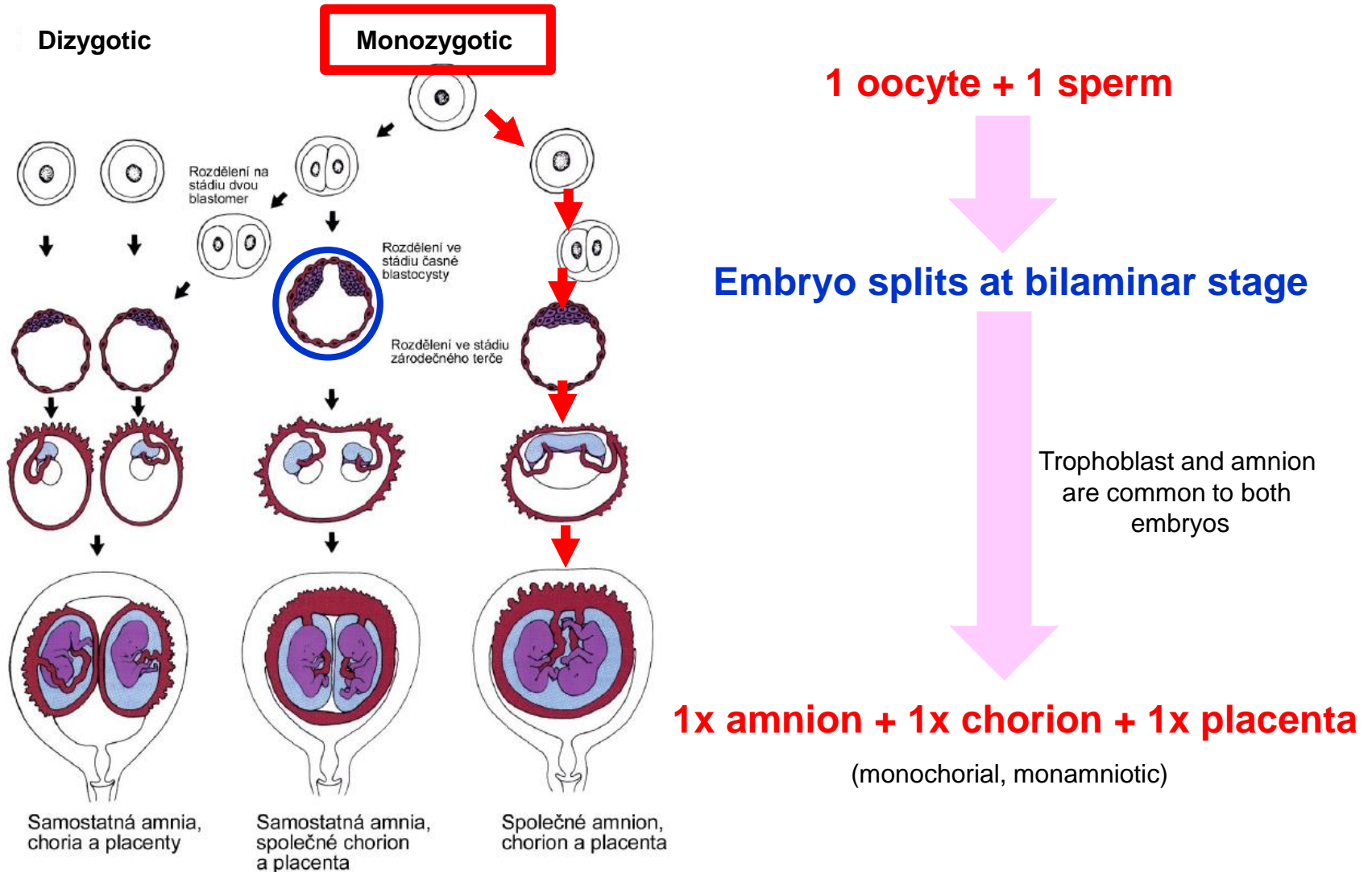
Trophoblast common to both embryos

2x amnion + 1x chorion + 1x placenta

(monochorial, diamniotic)

# Placenta – multiparous pregnancy 4

## TWINS



**Thank you for your attention !**

**Questions and comments at:  
ahampl@med.muni.cz**