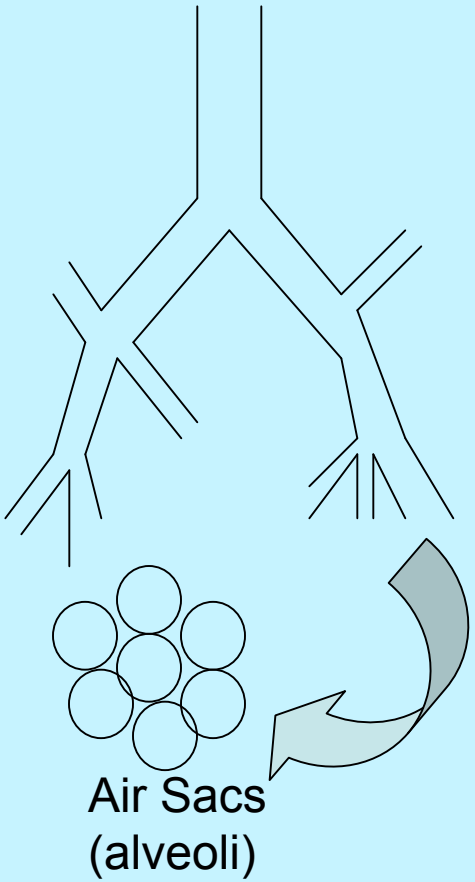
Function of Breathing

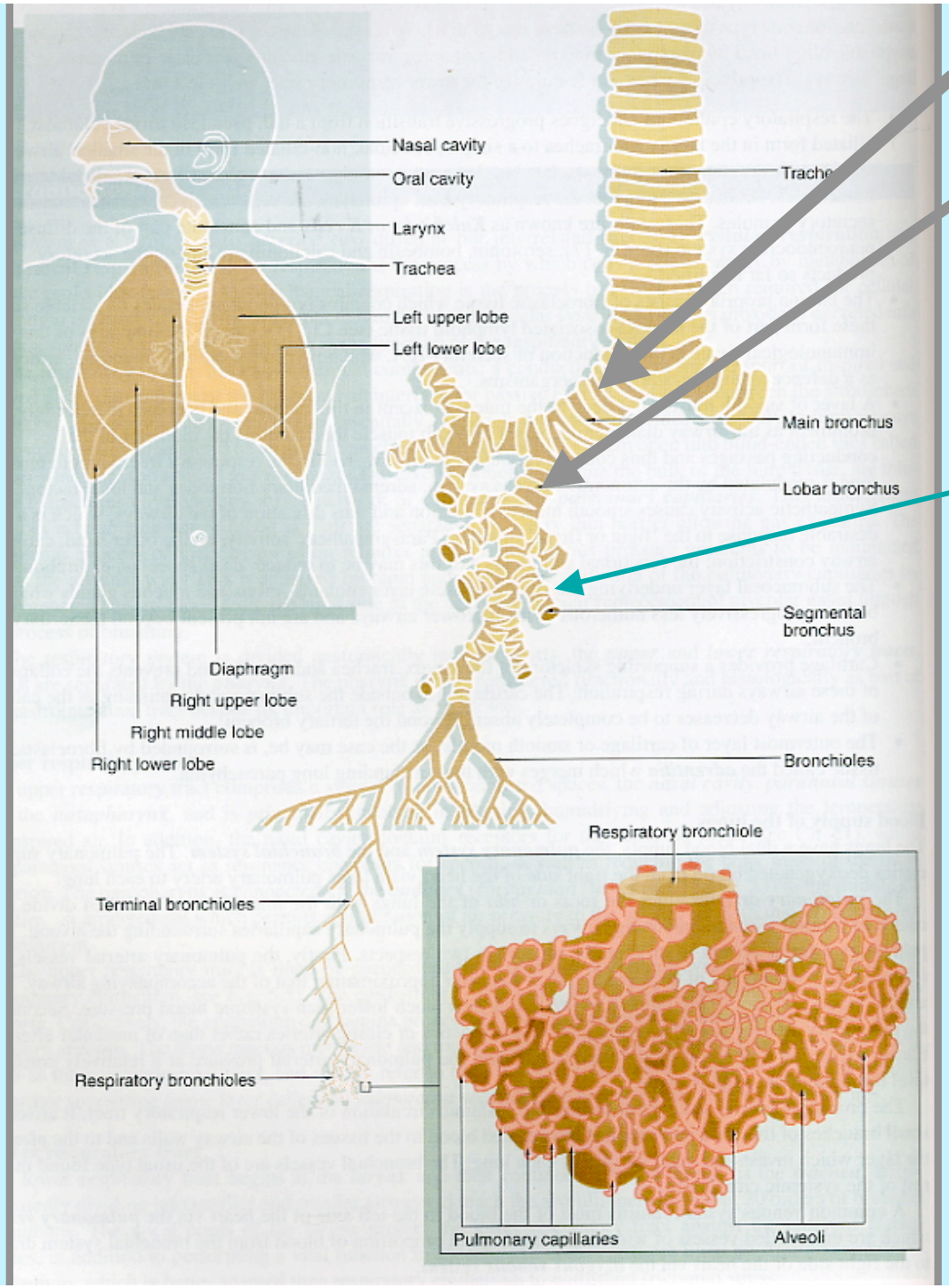


Ventilation-air conduction
- Moving gas in and out of the chest

Gas Exchange
- Moving gas Oxygen and carbon dioxide in and out of the blood
  - oxygen from air to blood
  - carbon dioxide from blood to air

Critical to the Development of the Lung

- Need a branched respiratory tree with a mucociliary cleaning mechanism
- A complex gas-exchange region with efficient diffusion and short diffusion distance
- Network of capillaries in close contact with the airspaces
- A surface film to reduce the surface tension of the alveoli and prevent collapse



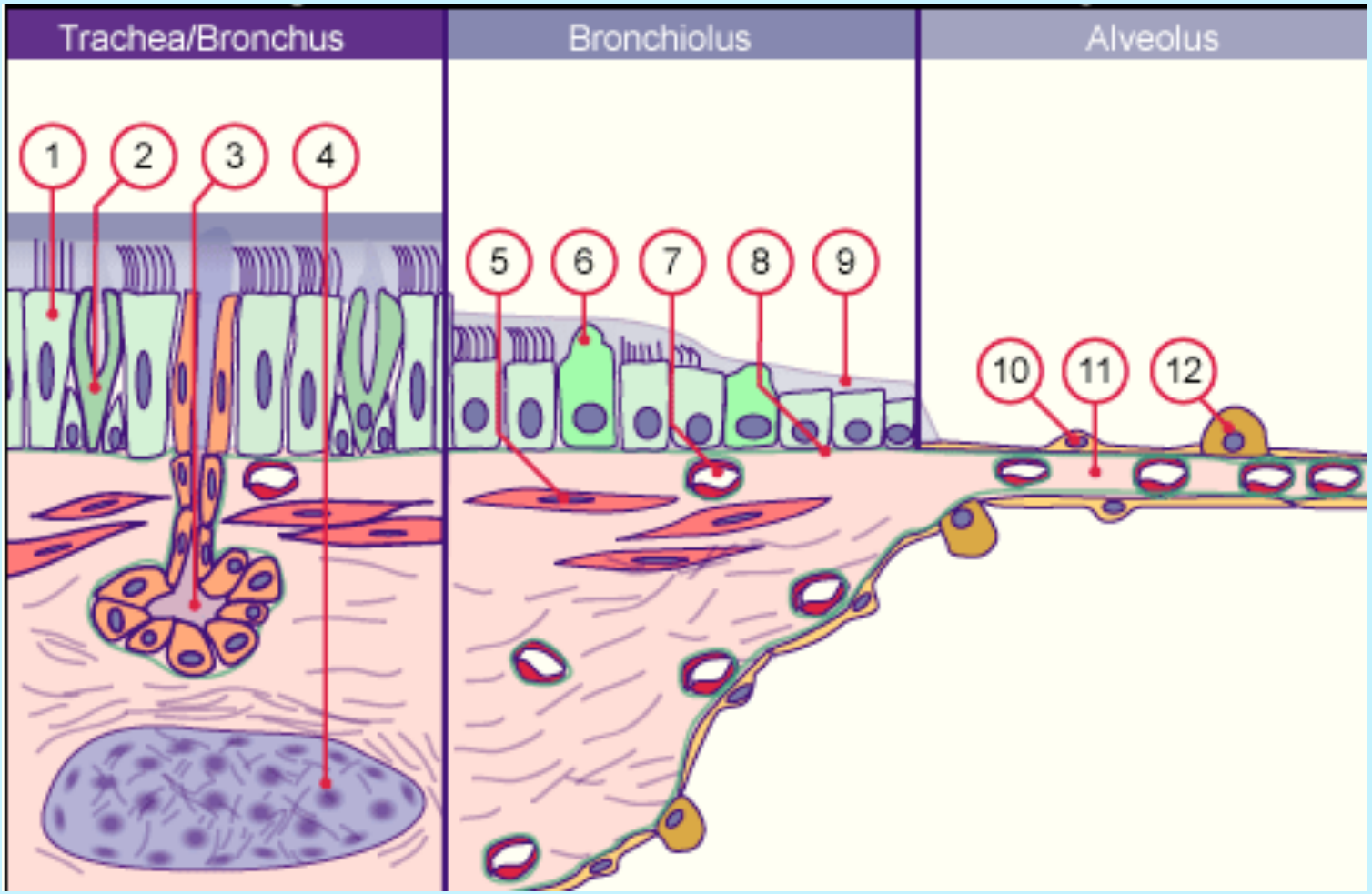
Main stem bronchus

Lobar bronchus (5 lung lobes)

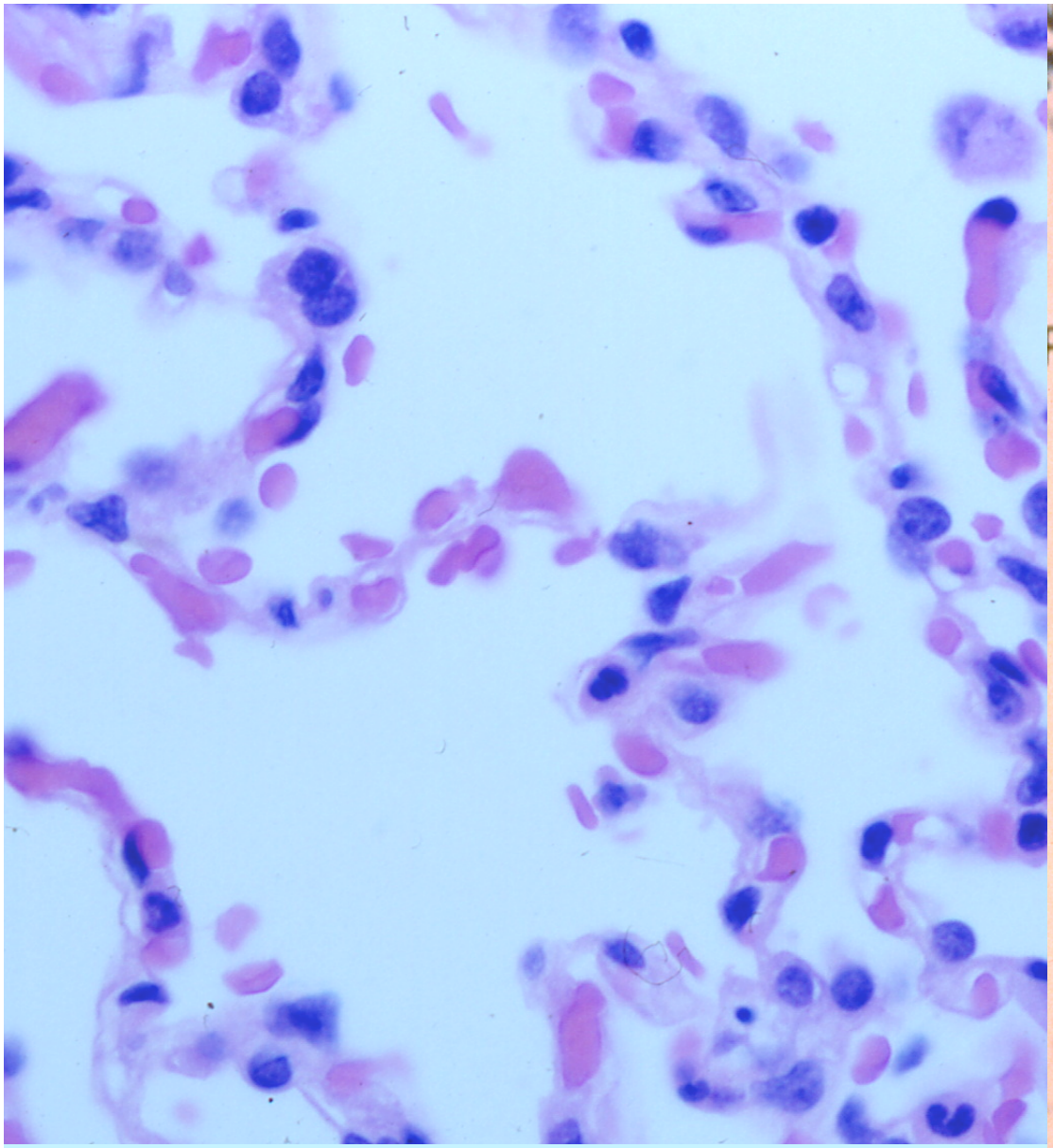
Segmental bronchus (10 bronchopulmonary segments on right, 9 on left

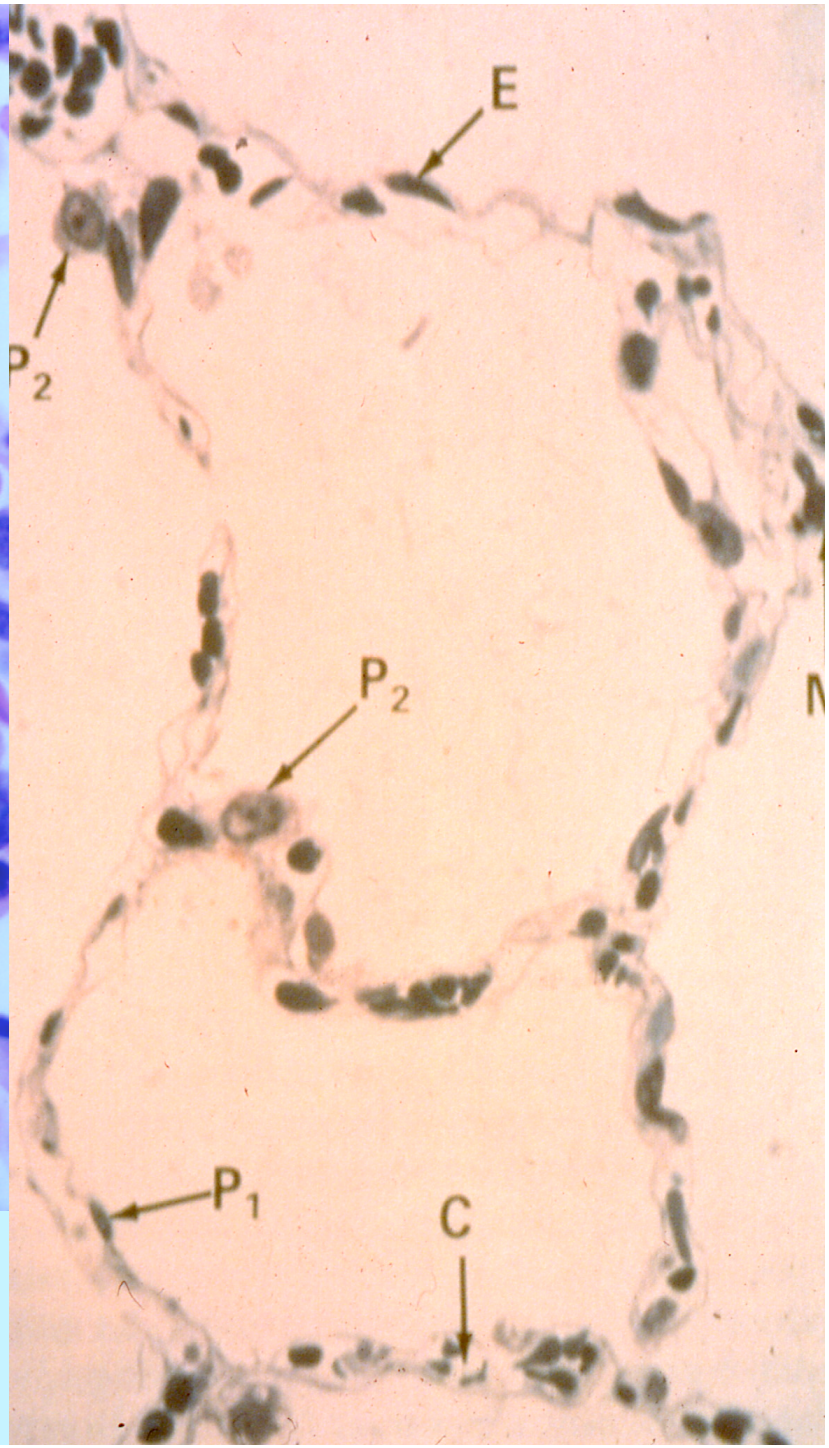
Branching continues as airways become bronchioles, then at terminal bronchioles airways transition into respiratory bronchioles

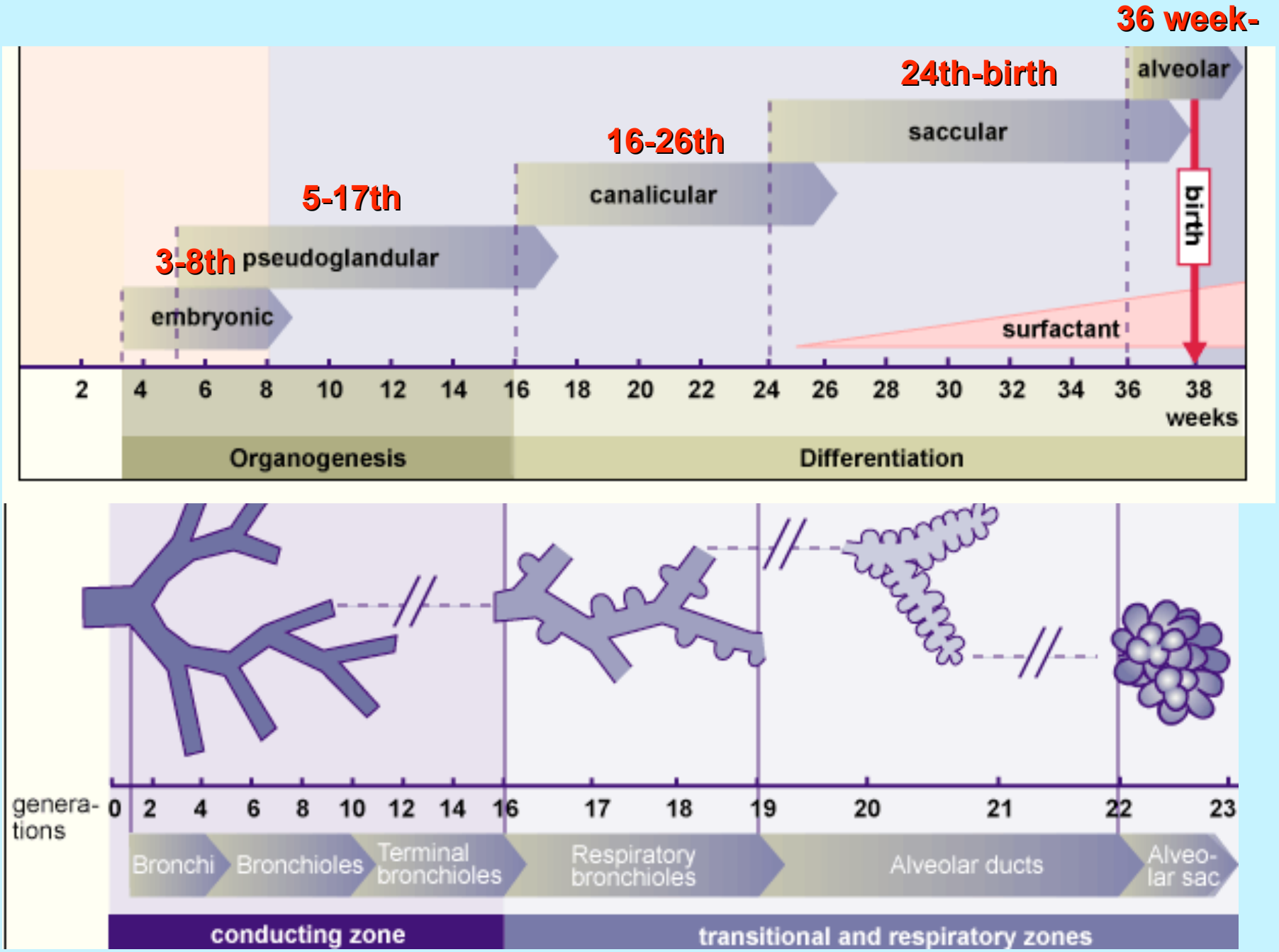
About 20 branch generations from beginning to end



1. Ciliated Epithelium
2. Goblet Cell
3. Gland
4. Cartilage
5. Smooth Muscle Cell
6. Clara Cell
7. Capillary
8. Basal Membrane
9. Surfactant
10. Type I pneumocyte
11. Alveolar Septum
12. Type II pneumocyte




E
P2
P2
P1
C


36 week-
24th-birth
alveolar
saccular
16-26th
canalicular
5-17th
3-8th pseudoglandular
embryonic
birth
surfactant
2 4 6 8 10 12 14 16 18 20 22 24 26 28 30 32 34 36 38
weeks
Organogenesis
Differentiation
genera-
tions
0 2 4 6 8 10 12 14 16 17 18 19 20 21 22 23
Bronchi
Bronchioles
Terminal bronchioles
Respiratory bronchioles
Alveolar ducts
Alveo-lar sac
conducting zone
transitional and respiratory zones

Embryonic Phase

3-7 Weeks

Initial budding and branching of lung buds from primitive foregu

Laryngeal development

- Week 4
  - Respiratory primordium arises from distal/caudal pharynx
    - Laryngo-tracheal groove
  - Endodermal derivative of epithelium of larynx trachea and bronchi
  - Connective tissue, smooth muscle and cartilage from splanchnic mesenchyme surrounding the foregut

Laryngeal development

- LT groove evaginates and forms LT diverticulum
- This becomes invested with splanchnic mesoderm to form lung bud
- This maintains a laryngeal inlet

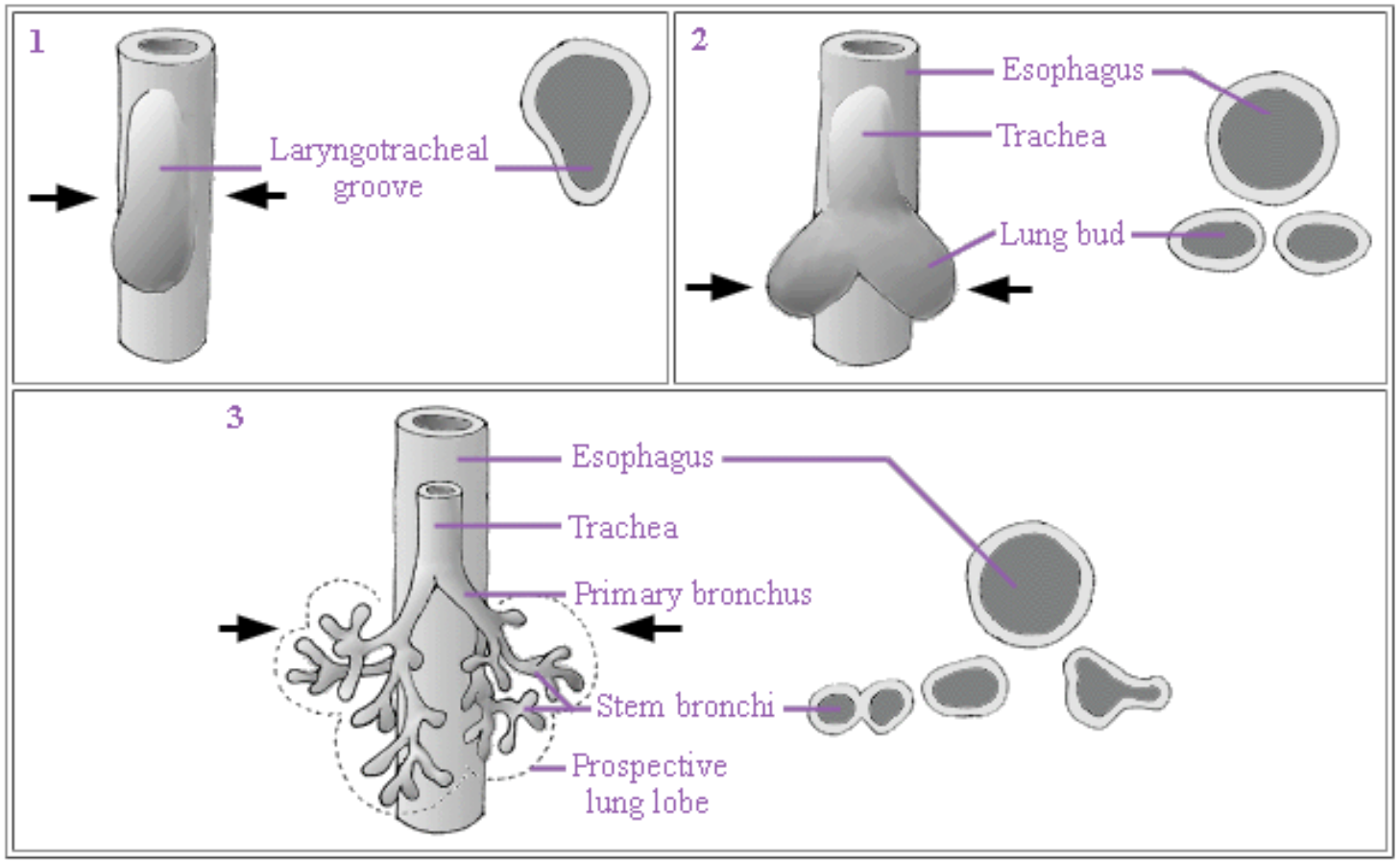
The septum that forms by folds and fusion keeps a septate inlet that becomes trachea and esophagus

Epithelium of the larynx

- Endoderm of proximal/cranial end of LT tube and cartilage from neural crest origin
- Formation of proximal larynx – cranial tube
  - Arytenoid swellings grow towards tongue
  - Airway gets closed off, eventually recanalizes
- Laryngeal webs – Incomplete recanalization
- Laryngeal atresia – ascites, hydrops and lungs do not properly form.

Trachea

- Endoderm of distal LT tube
  - Epithelium of trachea and lung
- Splanchnic mesenchyme
  - Connective tissue
- 4th week
  - If esopahgeal separation from LT tube is incomplete, develops into TE fistula



This page shows ventral views of the esophagus and developing lungs, accompanied by cross-sectional views through the area between the black arrows. Note how the lung starts as an evagination, from the esophogeal endoderm, called the larygotracheal groove (1). As the the larygotracheal groove grows, it develops two outcroppings at its caudal end, the lung buds (2). As the lung buds grow, they branch repeatedly forming the primary bronchi and stem bronchi (3) which branch further to form bronchioles, which will eventually develop terminal air sacs (alveoli) to complete the adult lung. Also, note how the trachea, once attached as a ventral groove on the esophagus, has separated to become a distinct tube (3).

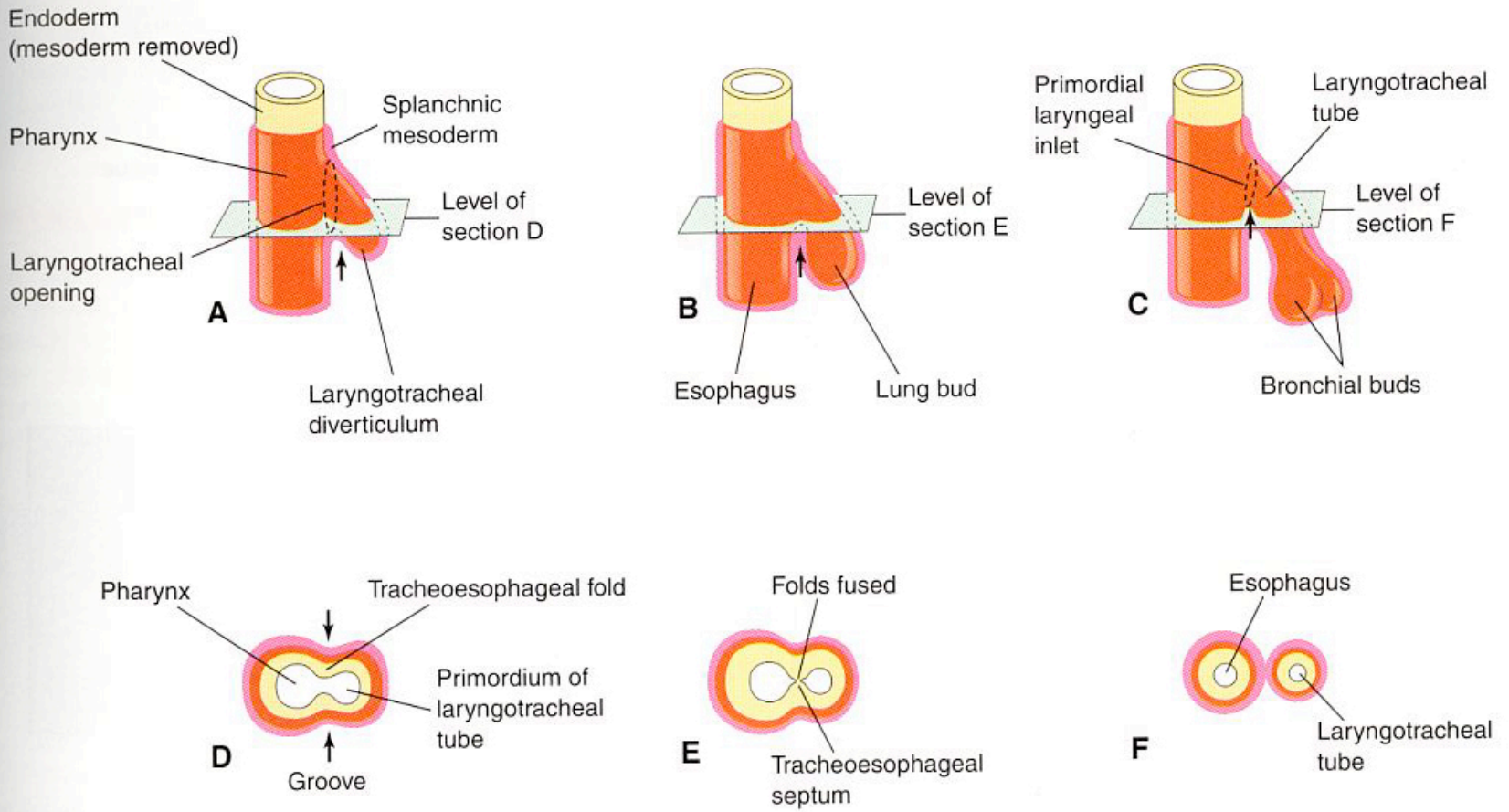
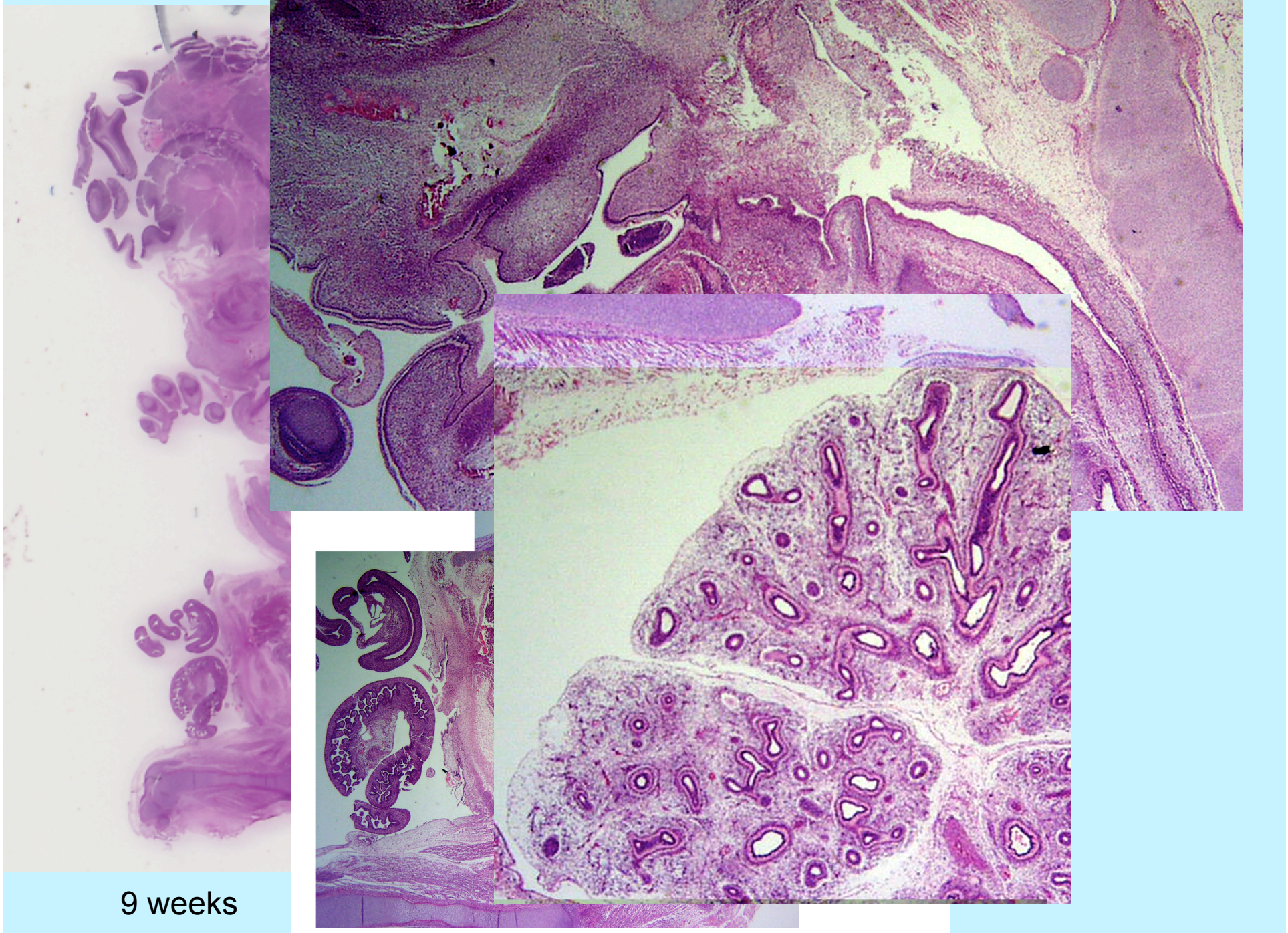
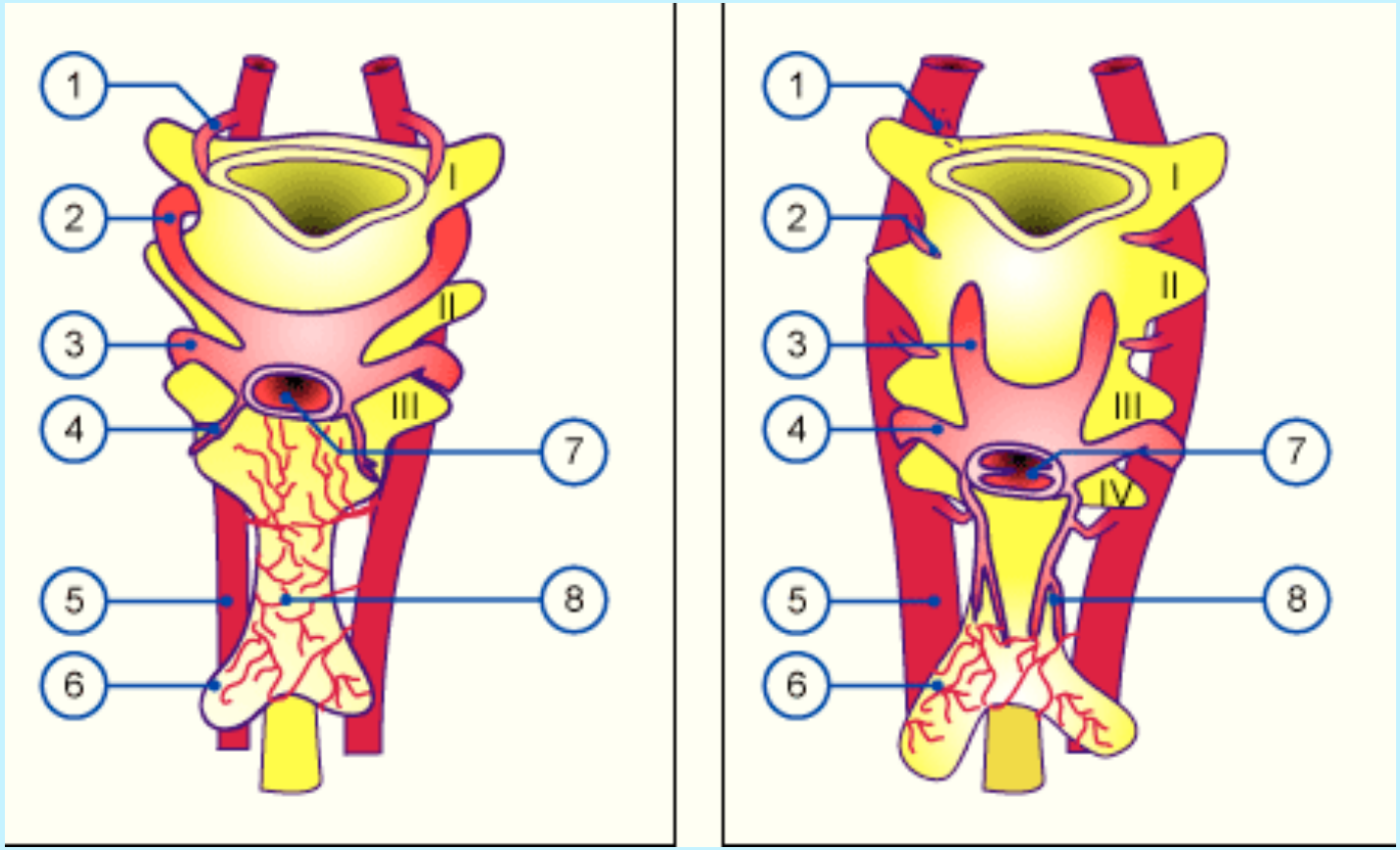


Figure 11–2. Drawings illustrating successive stages in the development of the tracheoesophageal septum during the fourth and fifth weeks. *A*, *B*, and *C*, Lateral views of the caudal part of the primordial pharynx showing the laryngotracheal diverticulum and partitioning of the foregut into the esophagus and laryngotracheal tube. *D*, *E*, and *F*, Transverse sections illustrating formation of the tracheoesophageal septum and showing how it separates the foregut into the laryngotracheal tube and esophagus.



9 weeks

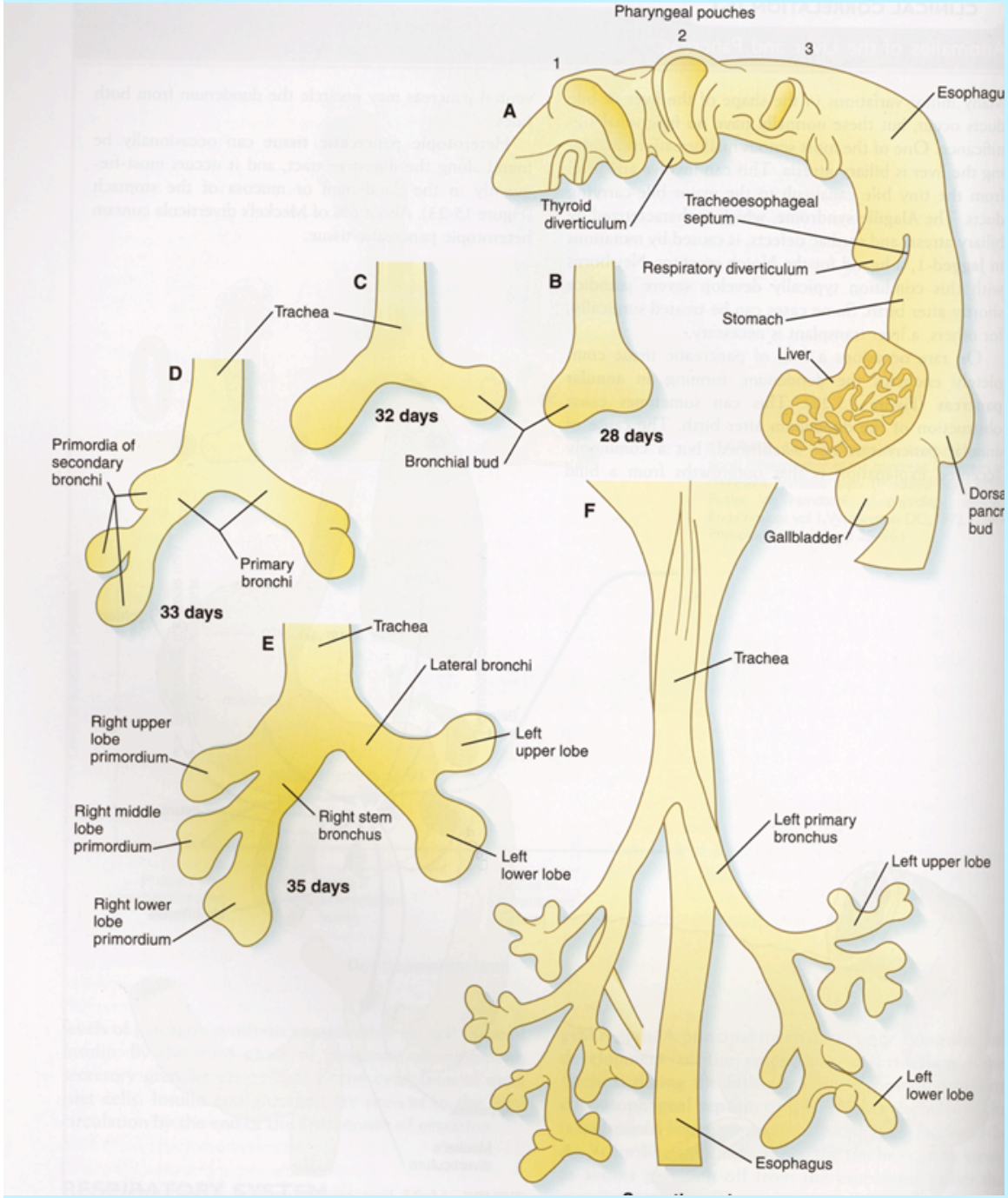
Pulmonary Circulation



1. First aortic arch
2. Second aortic arch
3. Third aortic arch
4. Fourth aortic arch
5. Dorsal Aorta
6. Lung buds
7. Aortic Sac
8. Pulmonary Plexus

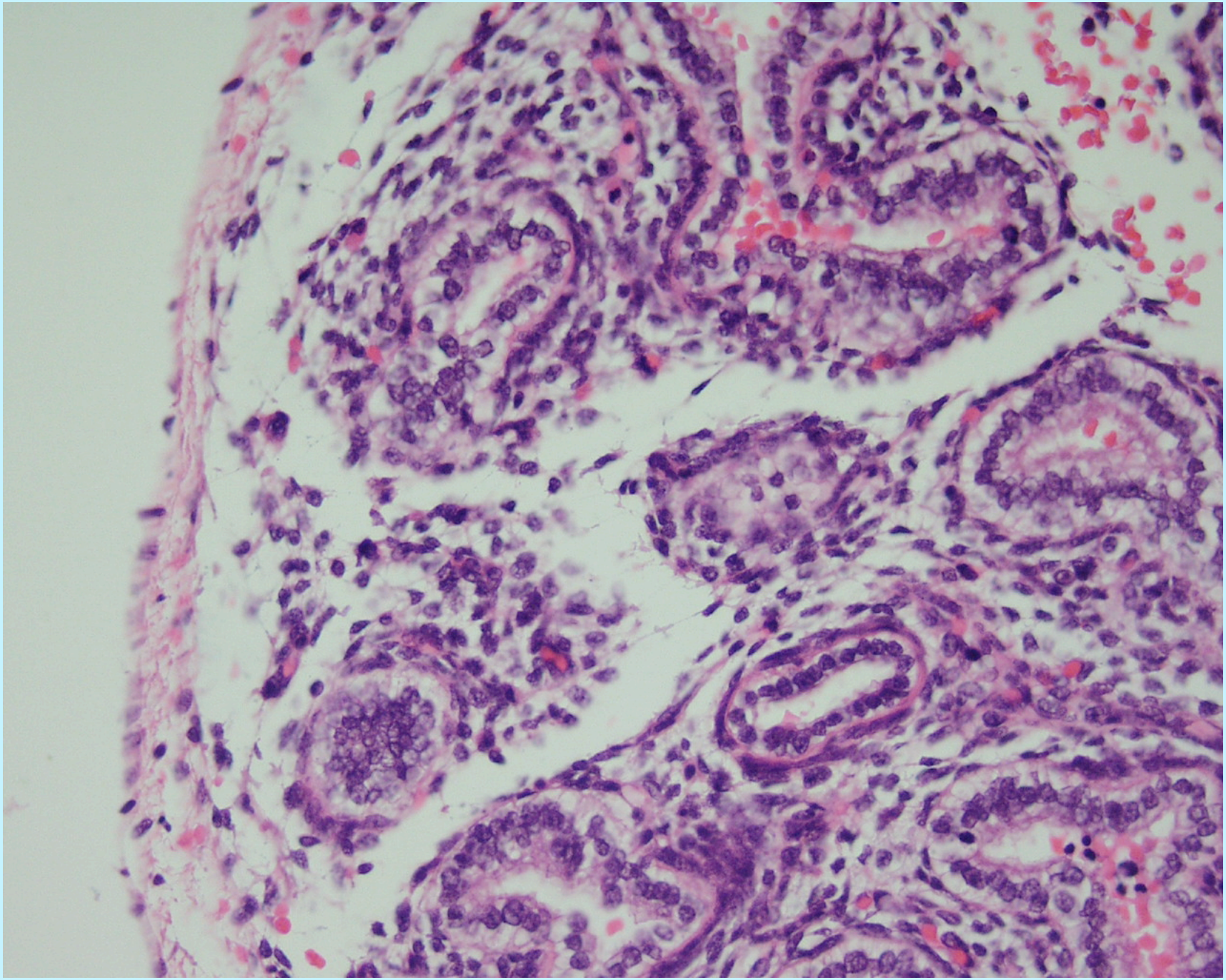
Bronchi/lungs

- By 28 days – endodermal buds grow along with splanchnic mesenchyme
- By 35 days – Second degree bronchi, upper middle and lower on right, upper and lower on left
- By 42 days – Tertiary bronchopulmonary segments, 10 on the right and 8-9 on the left.


Pharyngeal pouches
1
2
3
A
Esophagu
Thyroid diverticulum
Tracheoesophageal septum
Respiratory diverticulum
Stomach
Liver
Dorsa pancr bud
Gallbladder
B
C
Trachea
32 days
28 days
Bronchial bud
D
Primordia of secondary bronchi
Primary bronchi
33 days
E
Trachea
Lateral bronchi
Right upper lobe primordium
Left upper lobe
Right middle lobe primordium
Right stem bronchus
Left lower lobe
35 days
Right lower lobe primordium
F
Trachea
Left primary bronchus
Left upper lobe
Left lower lobe
Esophagus

Branching morphogenesis

- By 24 weeks, 17 orders of bronchi and respiratory bronchioles (7 more after birth)
- Lungs grow to pleura – visceral pleura from splanchnic mesenchyme and parietal pleura from somatic mesoderm.

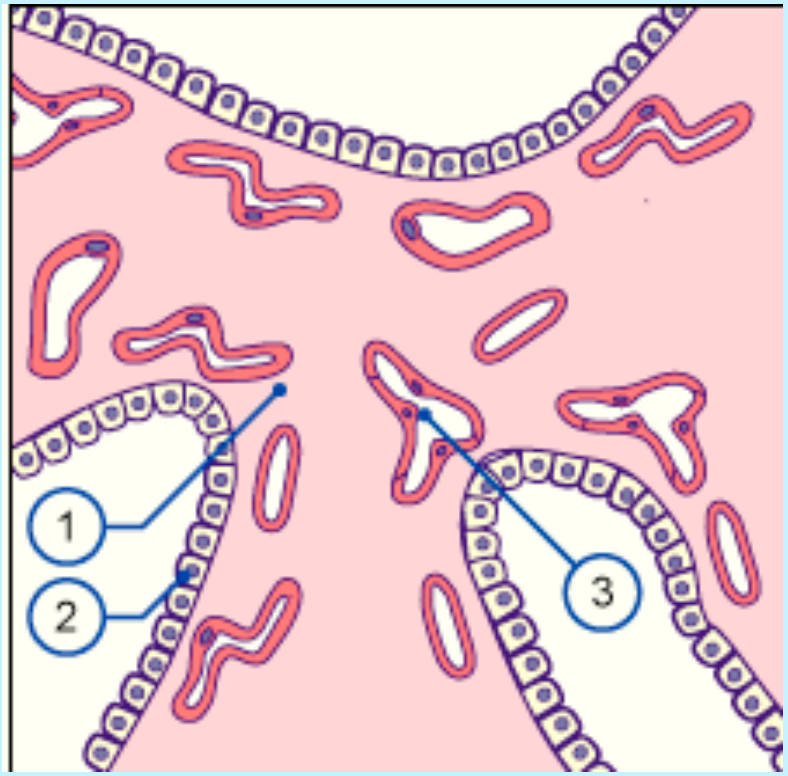


Pseudoglandular

5-17 Weeks

Formation of bronchial tree up to a preacinar level

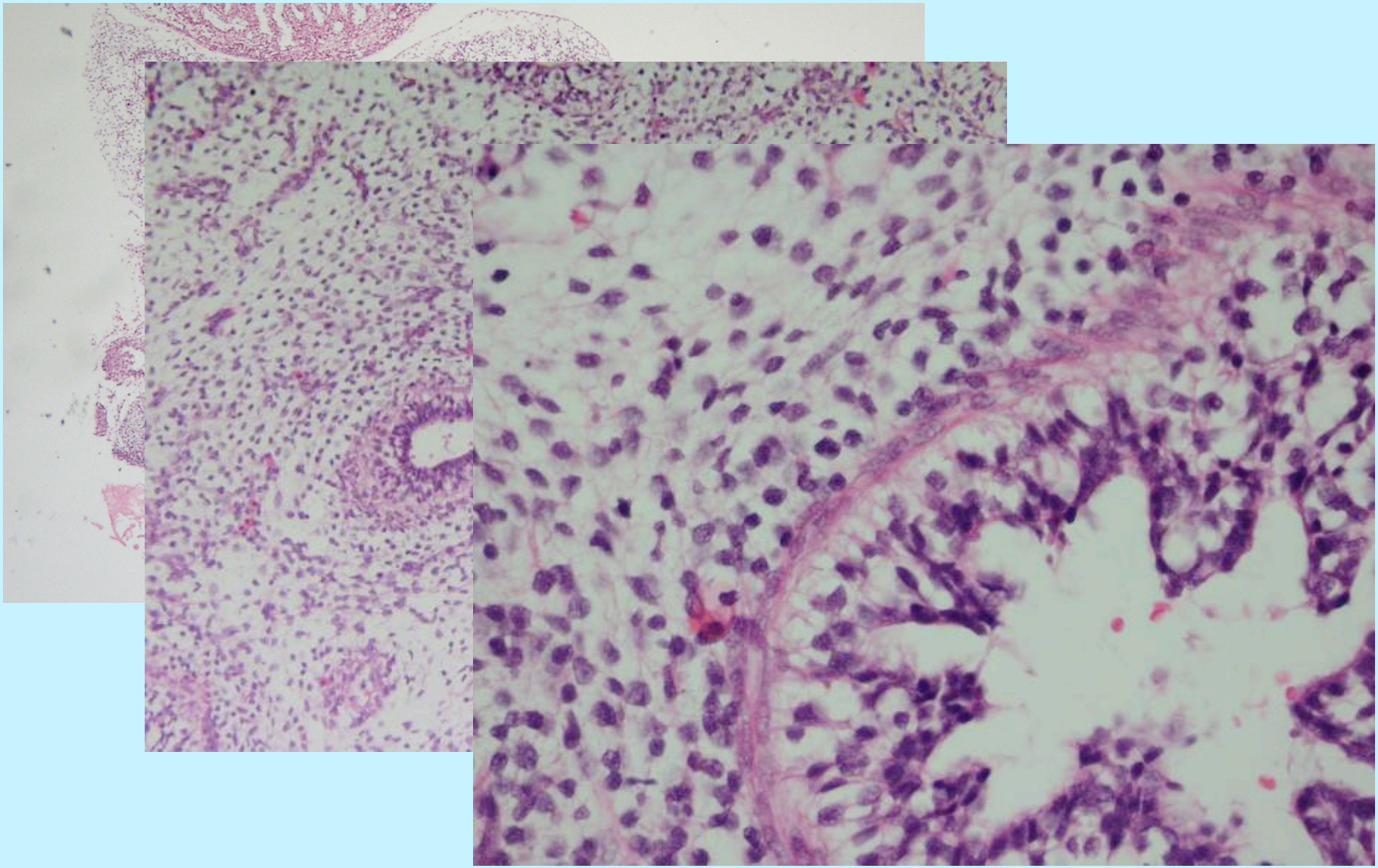
Pseudoglandular



In the pseudoglandular phase the lungs resemble a gland. At the end of this phase the precursors of the pneumocytes can be discerned in the respiratory sections as cubic epithelium

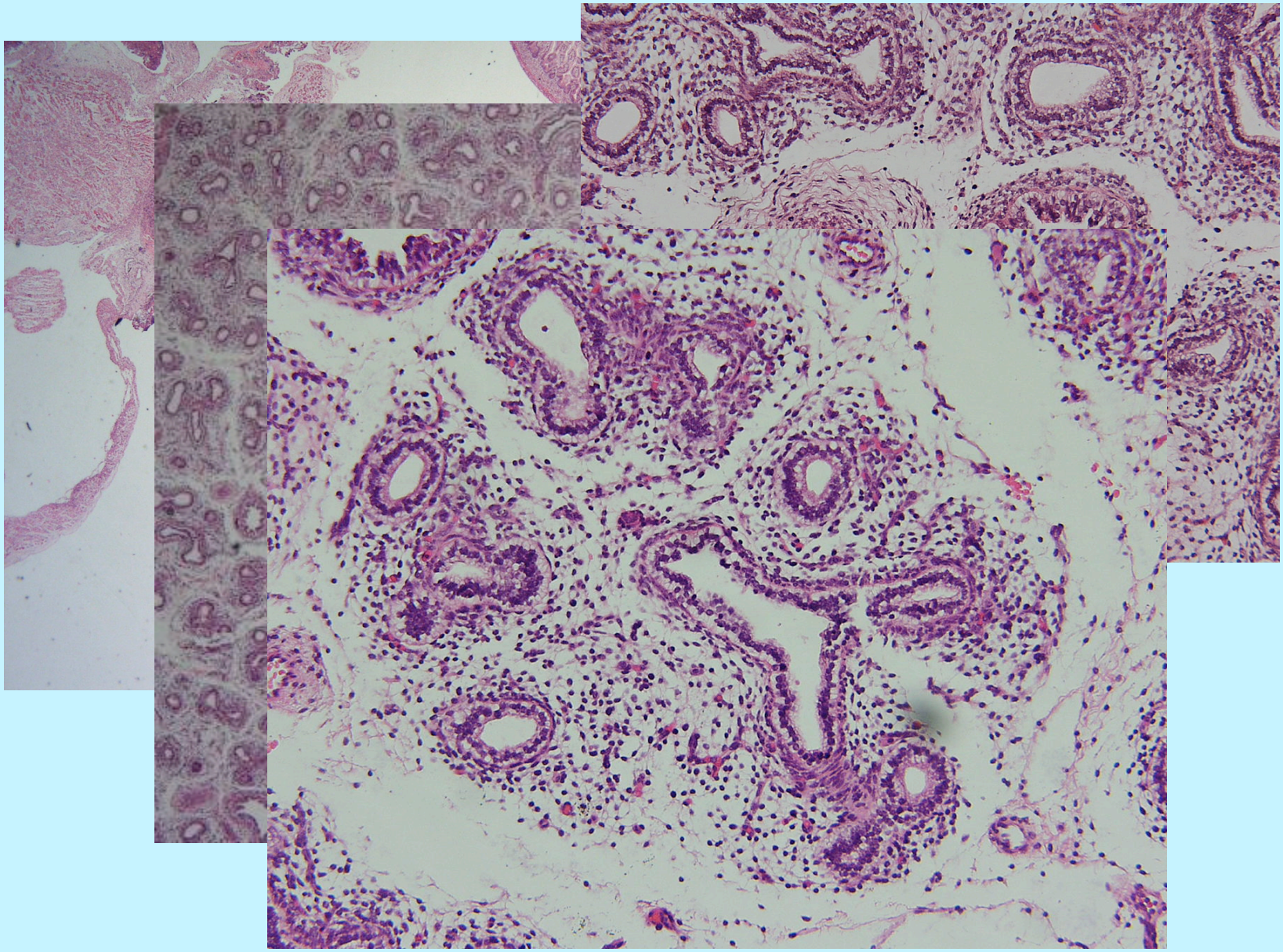
1. Lung mesenchyme
2. Type II pneumocytes
3. Capillaries

Early pseudoglandular 8 wk

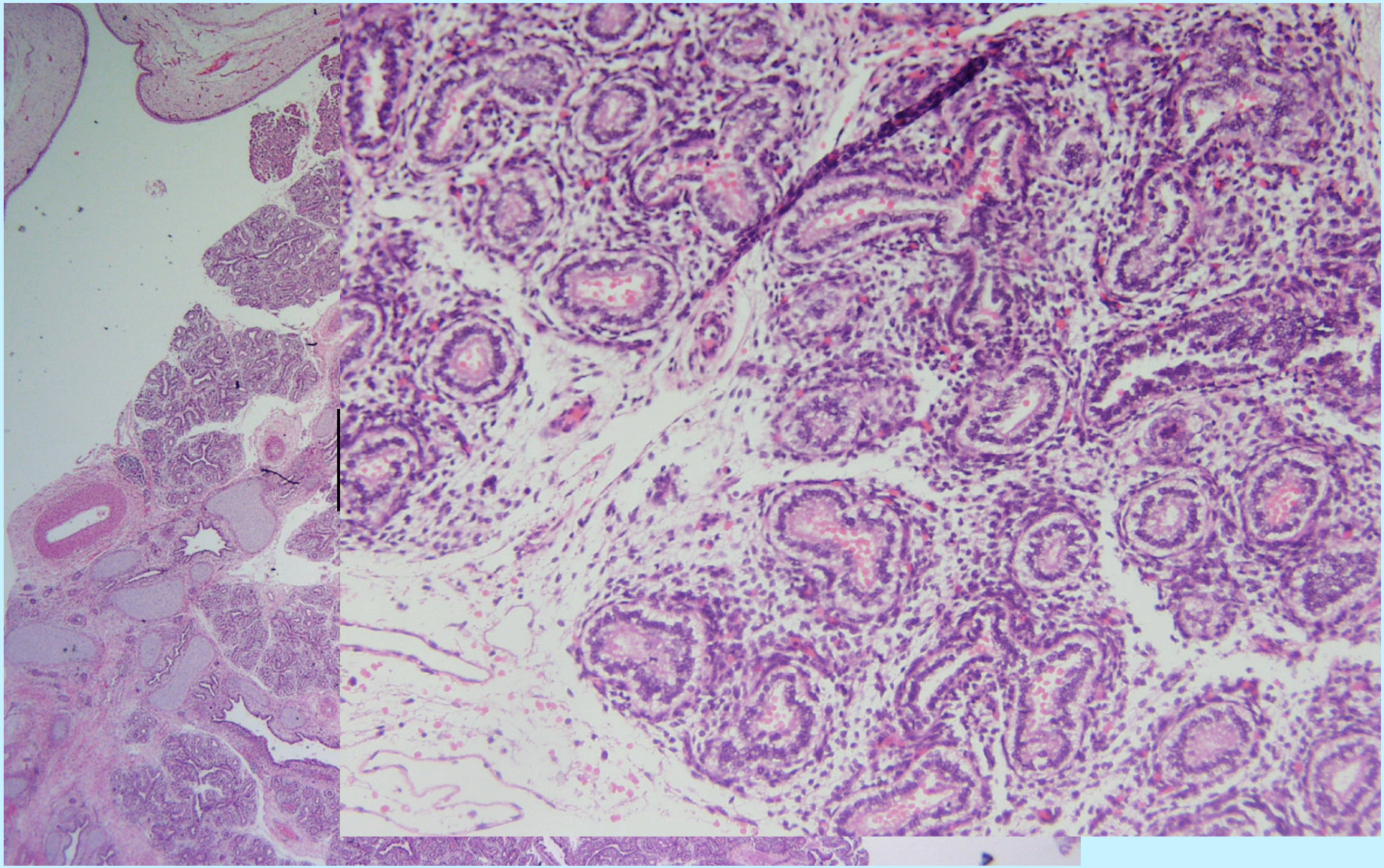


13 wk

Mid Pseudoglandular 13 wk



Late pseudoglandular-16 weeks

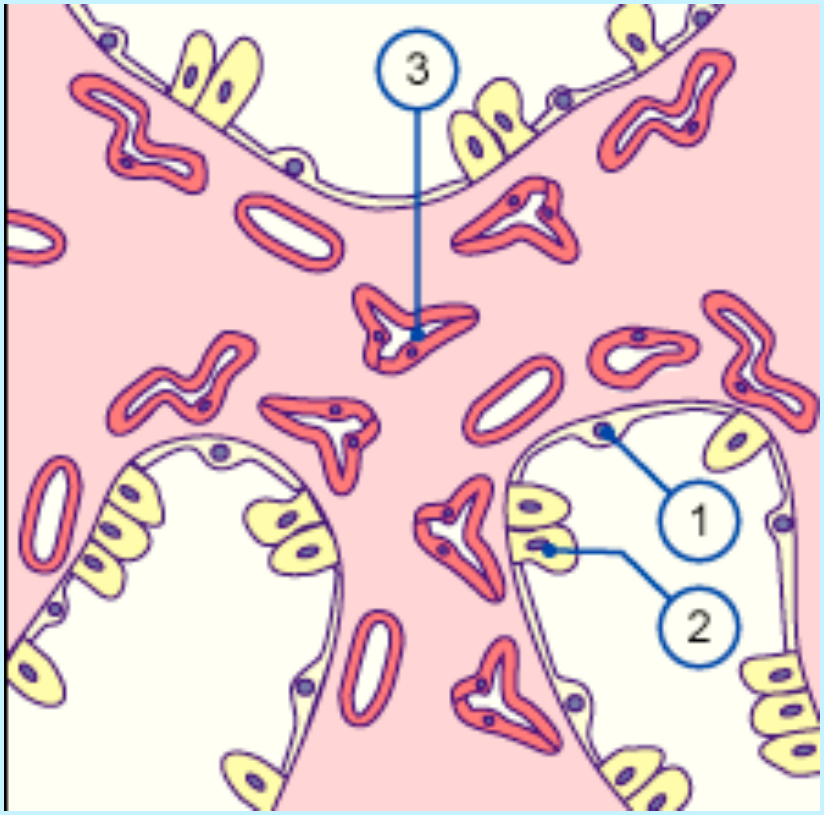


Canalicular

16-26 weeks

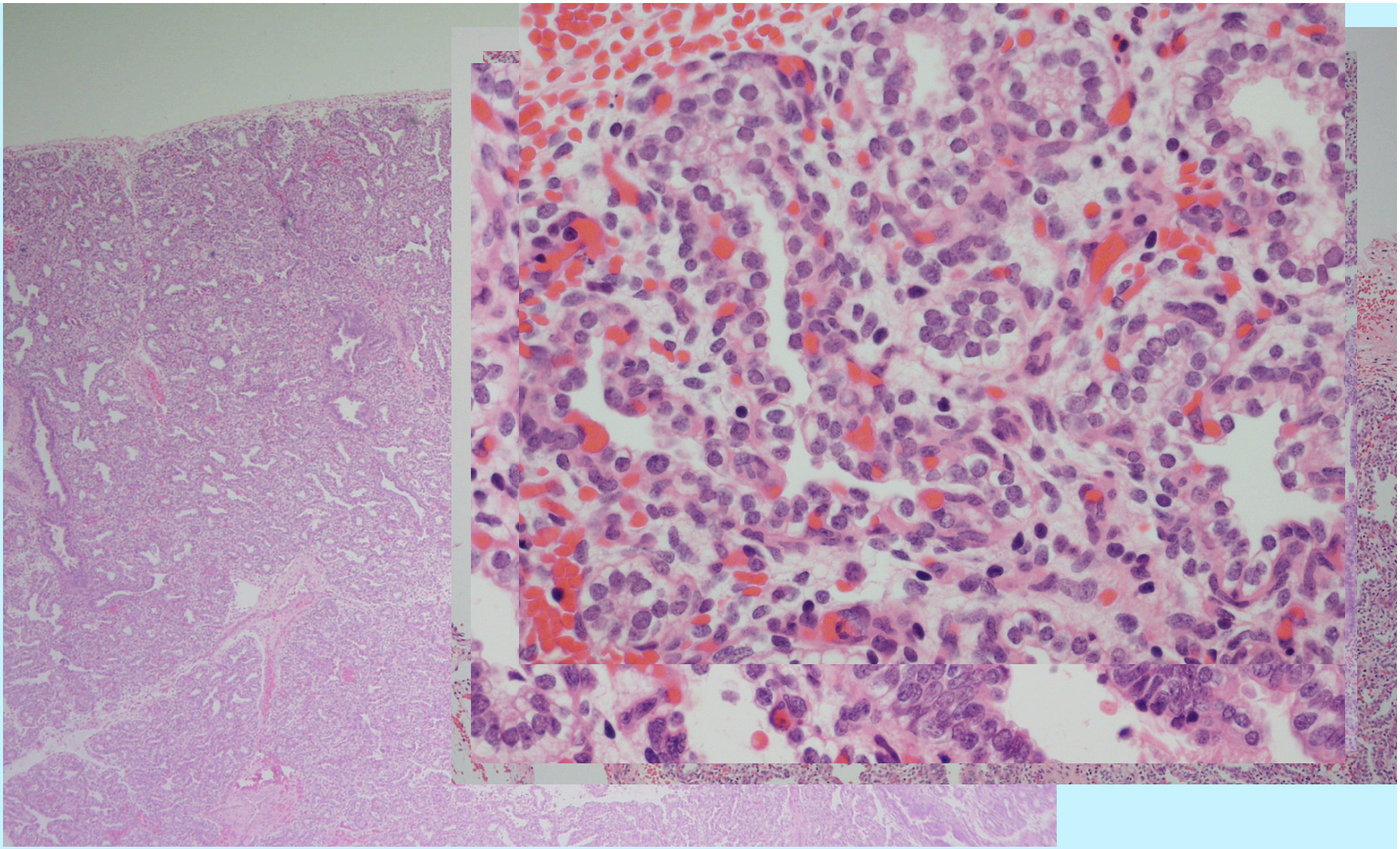
Formation of the pulmonary acinus and of the future air-blood barrier, increased capillary bed, epithelial differentiation and first appearance of surfactant

Canalicular



In the canalicular phase the type I pneumocytes differentiate out of the type II pneumocytes. The capillaries approach the walls of the acini.

1. Type I pneumocytes
2. Type II pneumocytes
3. Capillaries



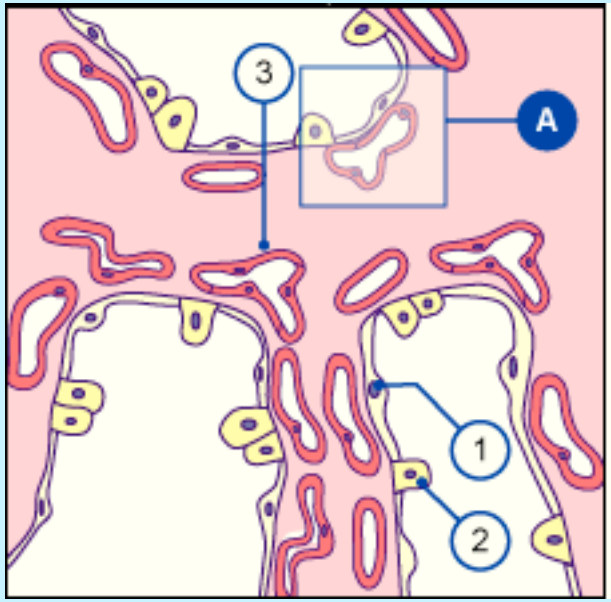
Mid-canalicular - 22 weeks

Saccular

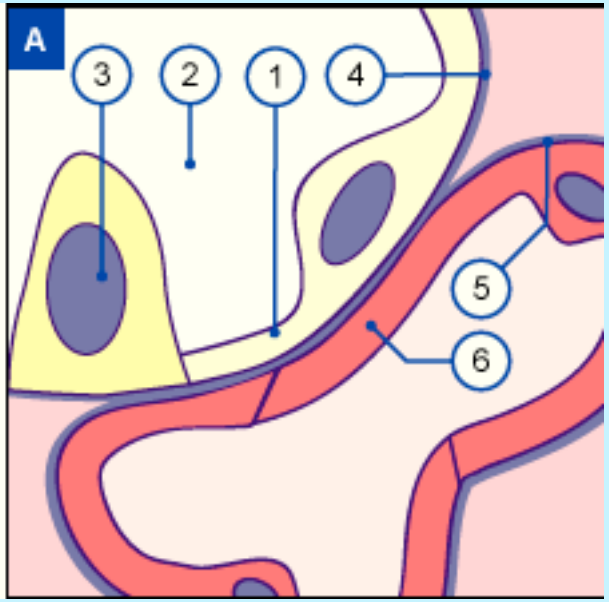
24-38 weeks

Formation of transitory air spaces

Saccular



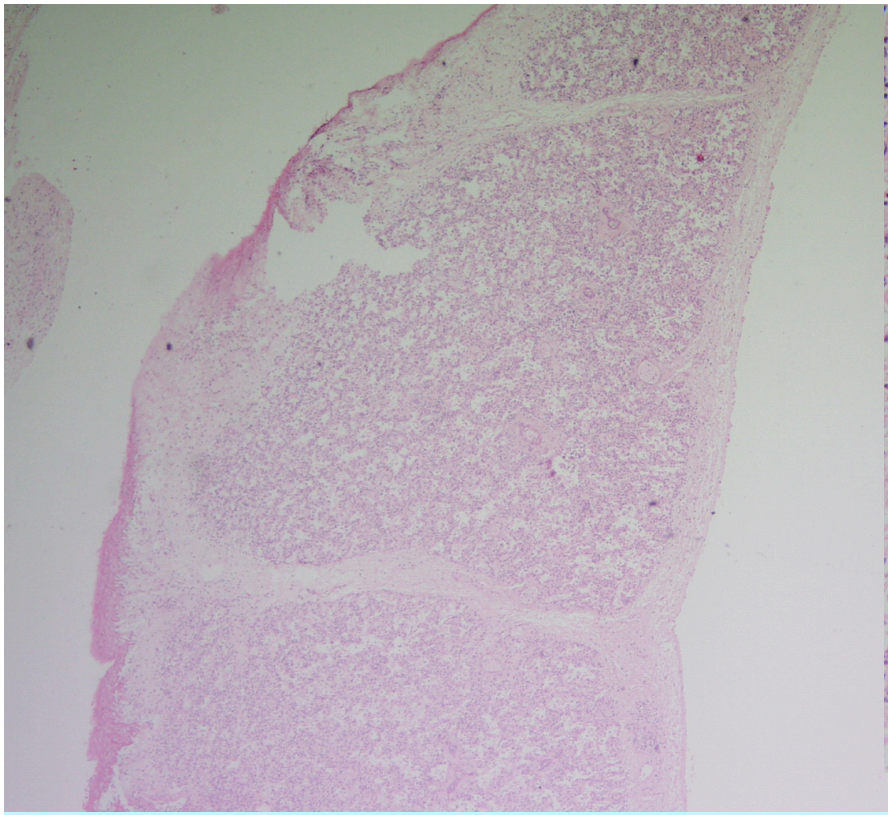
1. Type I pneumocyte
2. Type II pneumocyte
3. Capillaries

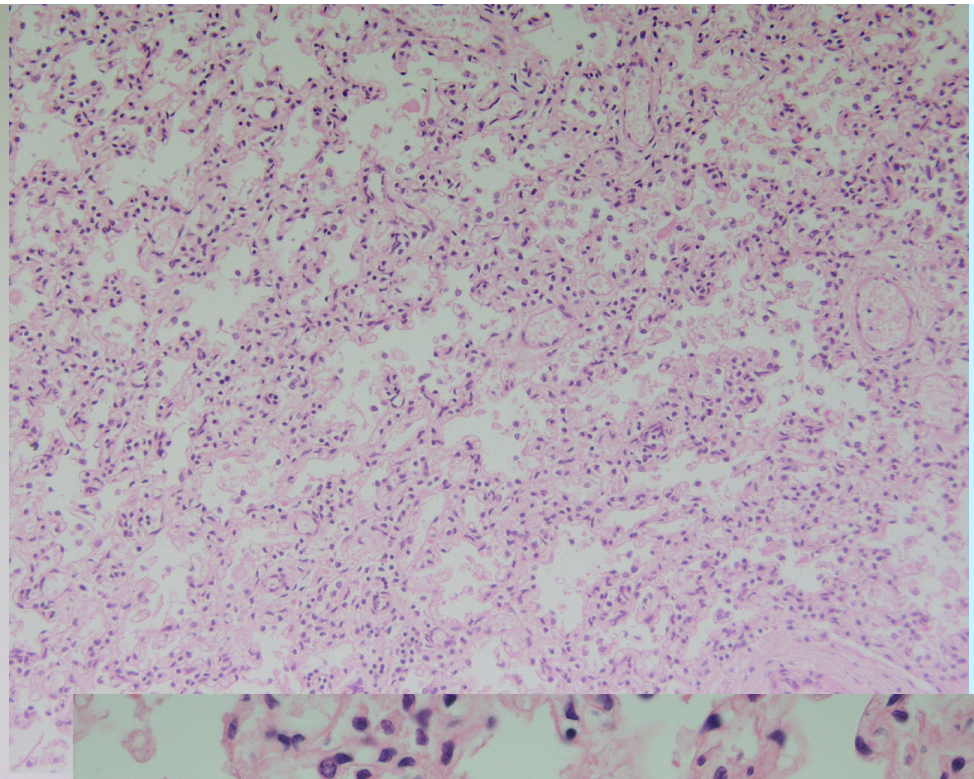


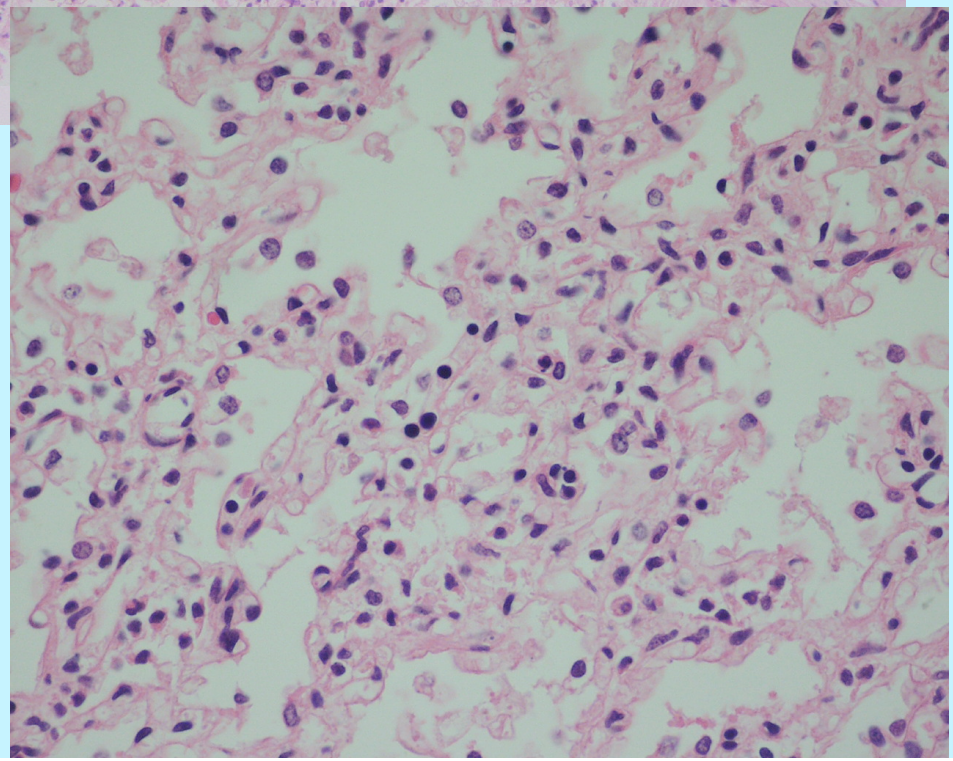
1. Type I pneumocyte
2. Saccular space
3. Type II pneumocyte
4. Basal membrane of the air passage
5. Basal membrane of the capillaries
6. Endothelium of the capillaries

The capillaries multiply around the acini and push close to the surface and form a common basal membrane with that of the epithelium

The blood-air barrier in the lungs is reduced to three, thin layers: type I pneumocyte, fusioned basal membrane, and endothelium of the capillary





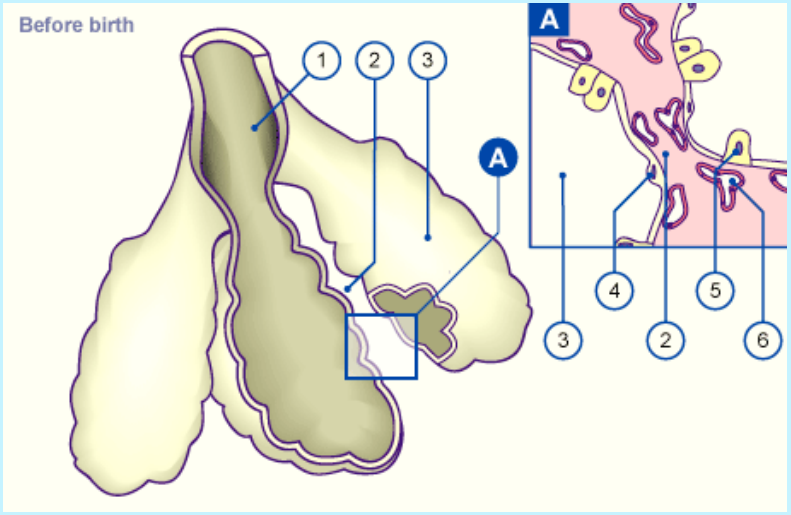


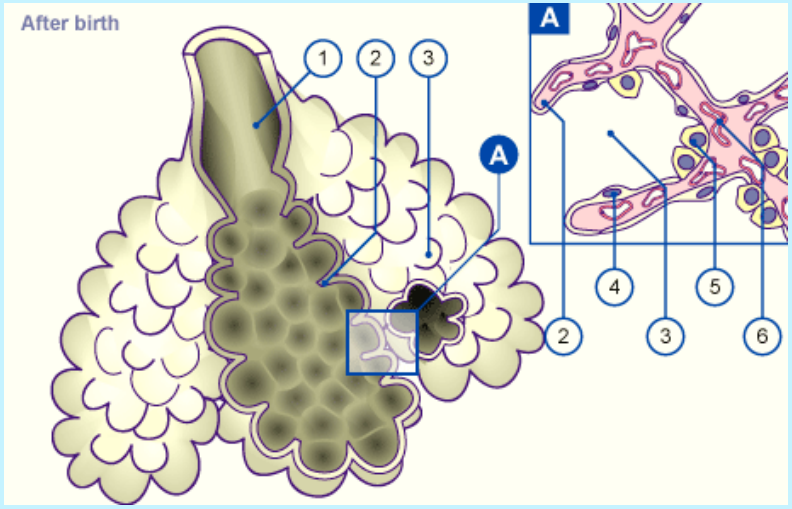
SACCULAR 31 weeks

Alveolar

36 weeks to 2 years postnatal

Alveolarization by forming of secondary septa



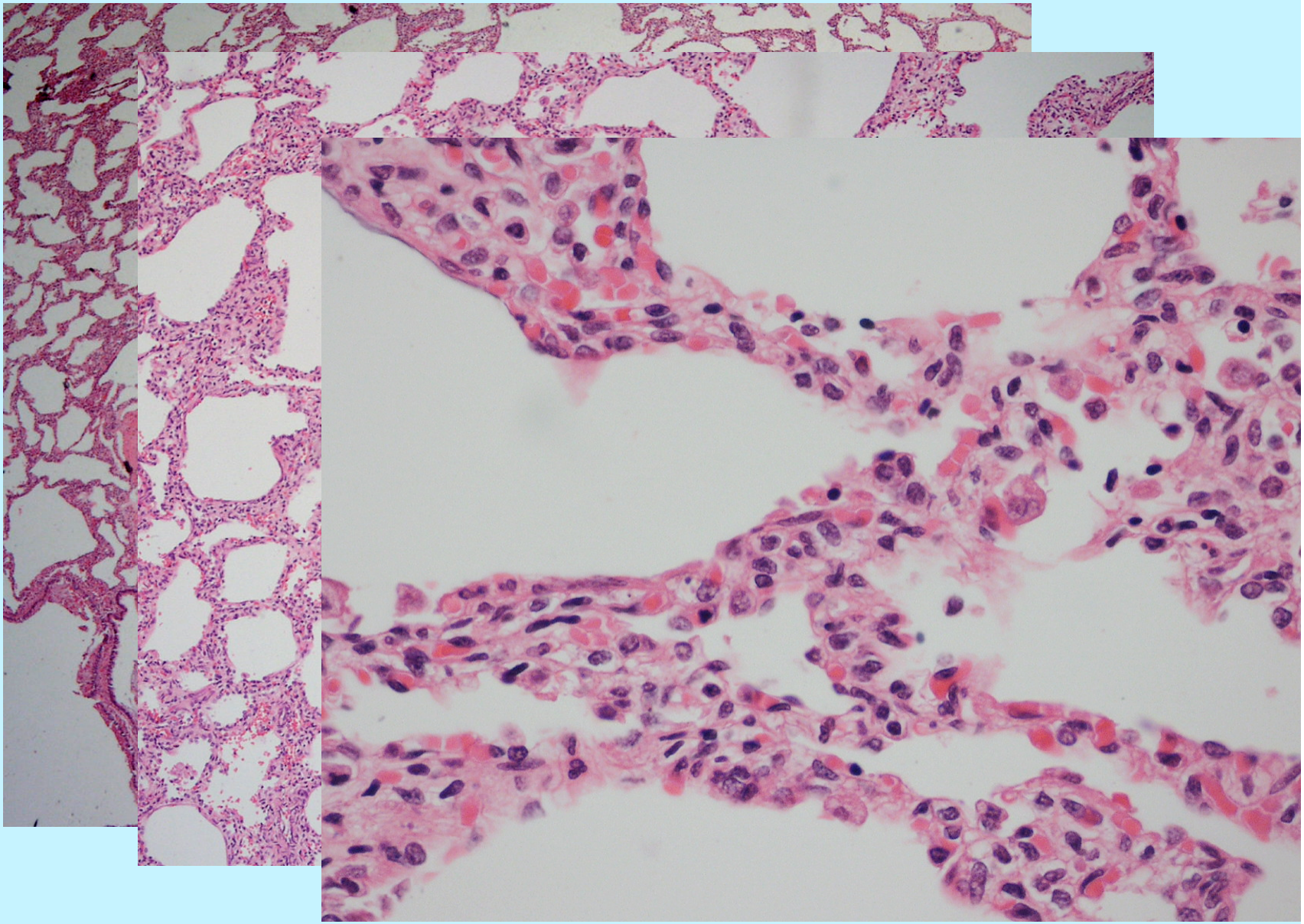


1. Alveolar duct
2. Primary septum
3. Alveolar sac
4. Type I pneumocyte
5. Type II pneumocyte
6. Capillaries

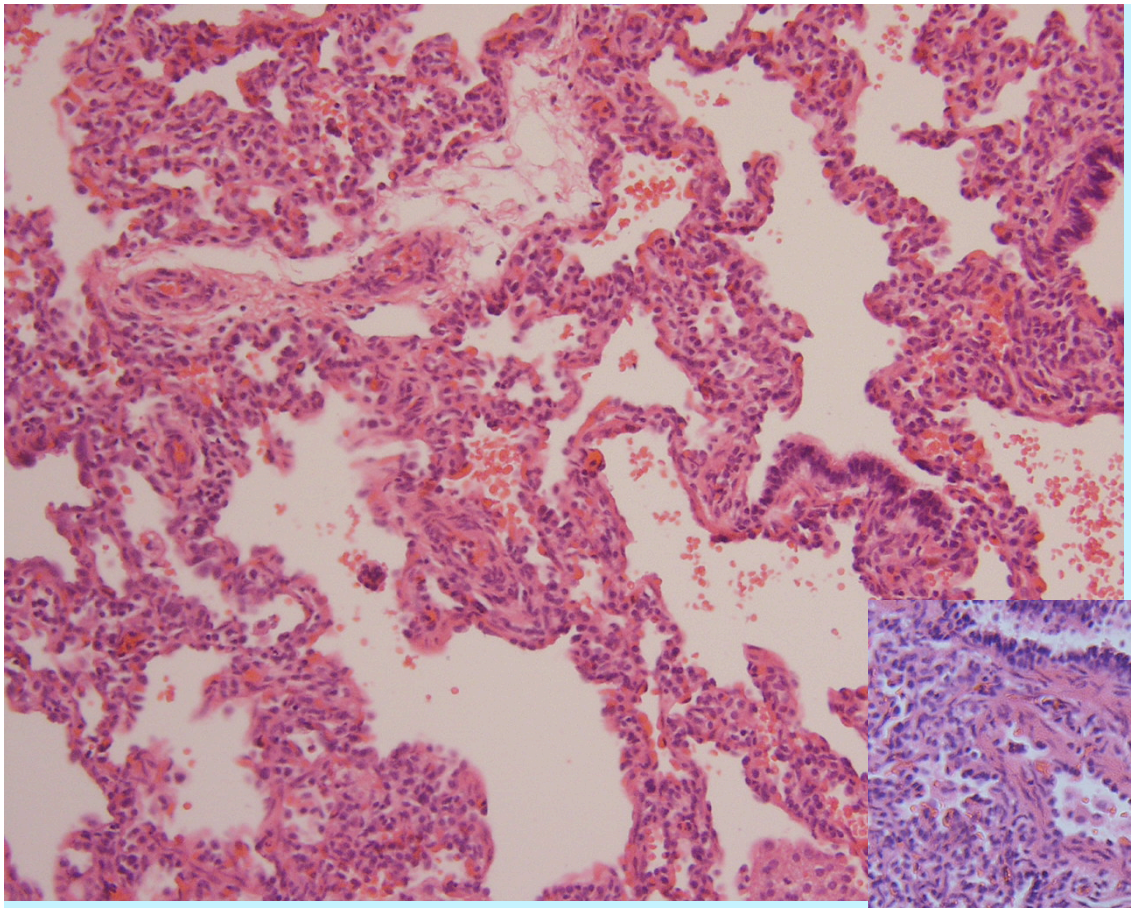
Alveolar

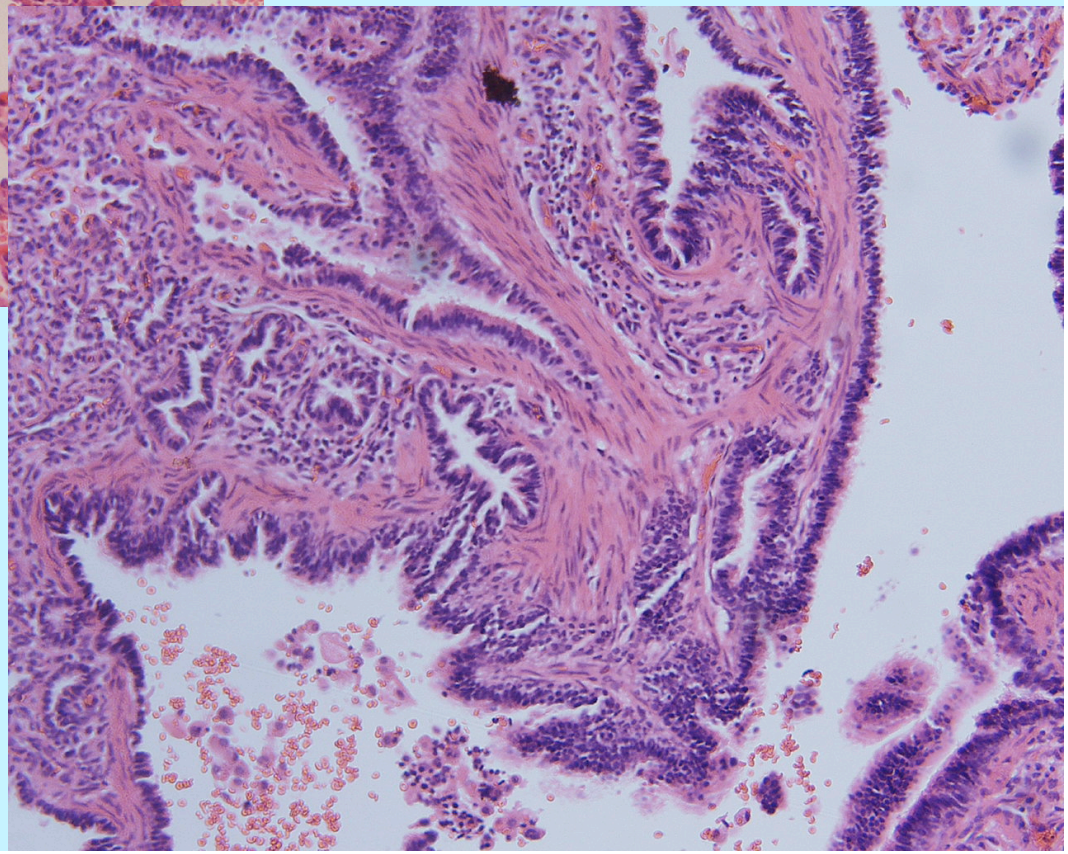
The alveoli form from the terminal endings of the alveolar sacculi and with time increase their diameter. After birth more and more alveoli form from the terminal endings of the alveolar and increase in diameter. They are delimited by secondary septa.

Term lungs

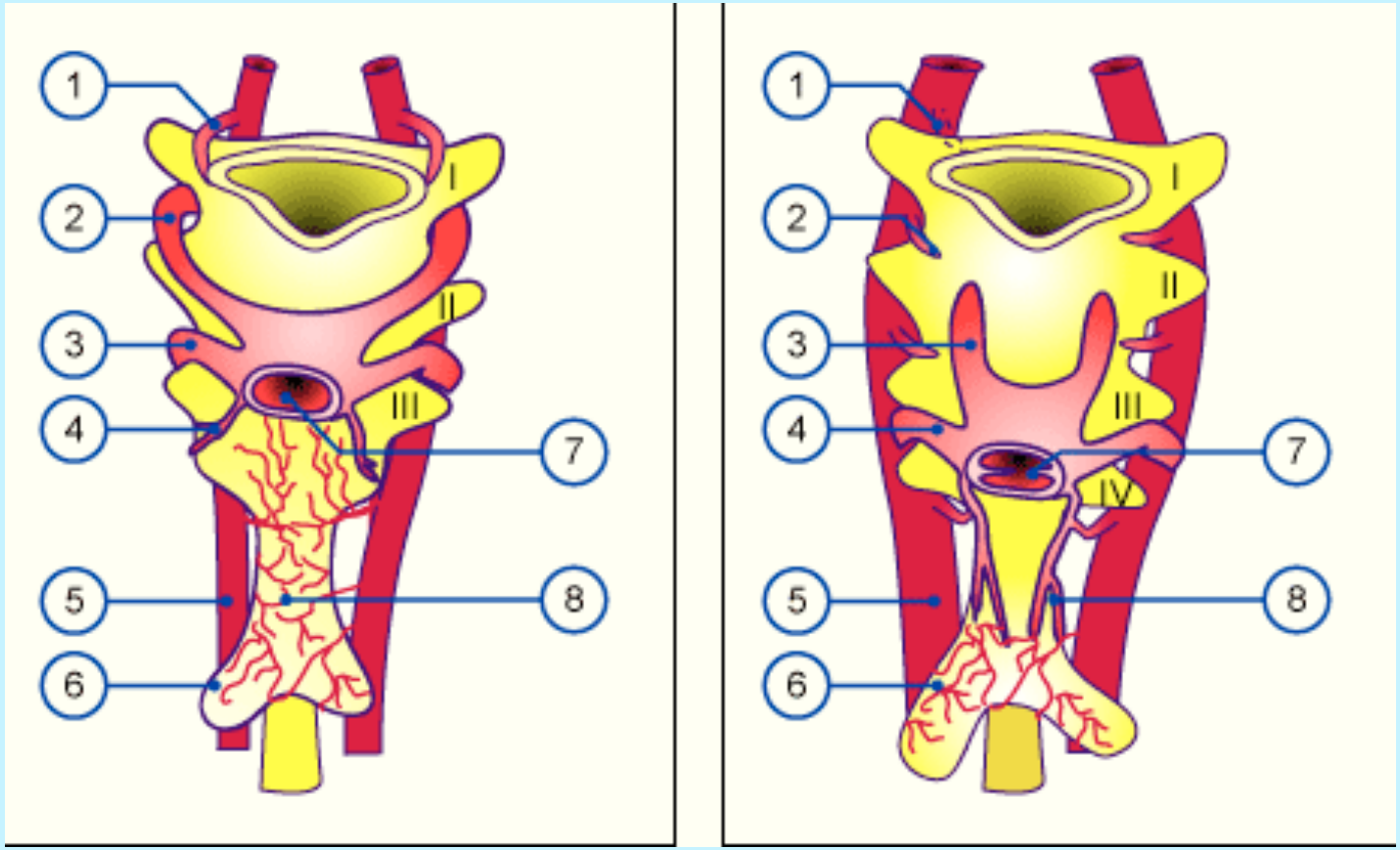


Term and CCAM





Pulmonary Circulation



1. First aortic arch
2. Second aortic arch
3. Third aortic arch
4. Fourth aortic arch
5. Dorsal Aorta
6. Lung buds
7. Aortic Sac
8. Pulmonary Plexus

In the embryonic mouse lung, Fgf10 regulates the placement and expansion of the lung bud (Bellusci et al., 1997b). Mice homozygous for loss-of-function mutations of Fgf10 lack limbs and lungs, while endodermal expression of a dominant negative Fgf receptor (Fgfr2IIIb) causes mice to lack terminal buds in their lungs (Peters et al., 1994; Min et al., 1998; Sekine et al., 1999). Moreover, the addition of Fgf10 to 11.5d embryonic mouse lung rudiments in Matrigel causes extensive budding (Bellusci et al., 1997b). FGF10 is seen both in the mesenchyme around both the terminal and lateral branches.

The regulation of FGF10 appears to be controlled, at least in part, by Sonic hedgehog and BMP4 (Lebeche et al., 1999). Shh is expressed throughout the respiratory epithelium, with the highest expression being in the terminal buds (Bellusci et al., 1997a). In lung rudiments where Shh is overexpressed, Fgf10 transcription is reduced significantly (Bellusci et al., 1997b). In normal mouse lung development, the lateral buds become surrounded with Shh-expressing mesenchyme after they form (Figure 1).

One possible scenario is envisioned in Figure 1. (A) During bud outgrowth, Shh and Wnt7b from the epithelium induce FGF10 and cell proliferation of both the epithelium and mesenchyme cells. (B, C) As outgrowth progresses, the levels of BMP rise in the distal tip, and it reaches a level where it can inhibit FGF10. FGF10 expression is then seen more laterally, where it initiates the formation of new buds. (D) At the most distal region, a cleft appears, and extracellular matrix molecules stabilize this cleft.

Congenital malformations

- Cystic adenomatoid malformations
  - Maturation arrest in lung segments
- Azyous lobe
  - Superior apical bronchus grows medially instead of laterally; vein is at bottom of superior lobe fissure
- Sequestration –
  - Accessory piece of lung that becomes disconnected from tracheobronchial tree and parasitizes systemic circulation from diaphragm.

Causes of Lung hypoplasia – diminished lung development

- Oligohydramnios – insufficient amniotic fluid
- Compression
  - Congenital diaphragmatic hernia – intestinal contents compress left hemithorax (usually)
  - Intrathoracic fluid or thoracic wall abnormality

RDS-Respiratory distress syndrome

- Low surfactant – Respiratory distress syndrome – usually due to pre-maturity, rarely due to surfactant protein deficiency (genetic cause)
  - Surfactant is critical to reduce surface tension and allow lung expansion at the air fluid interface.
    - Inadequate surfactant leads to alveolar collapse on expiration of air, and difficulty re-inflating
    - Damage to the alveolus leads to cellular injury and exudation of proteins known as hyaline membranes (Hyaline membrane disease)
      - Continued injury from ventilation of immature lungs can lead to chronic injury known as bronchopulmonary dysplasia.
  - Steroids accelerate lung development and surfactant production
    - Surfactant can also be administered

Pulmonary vasculature

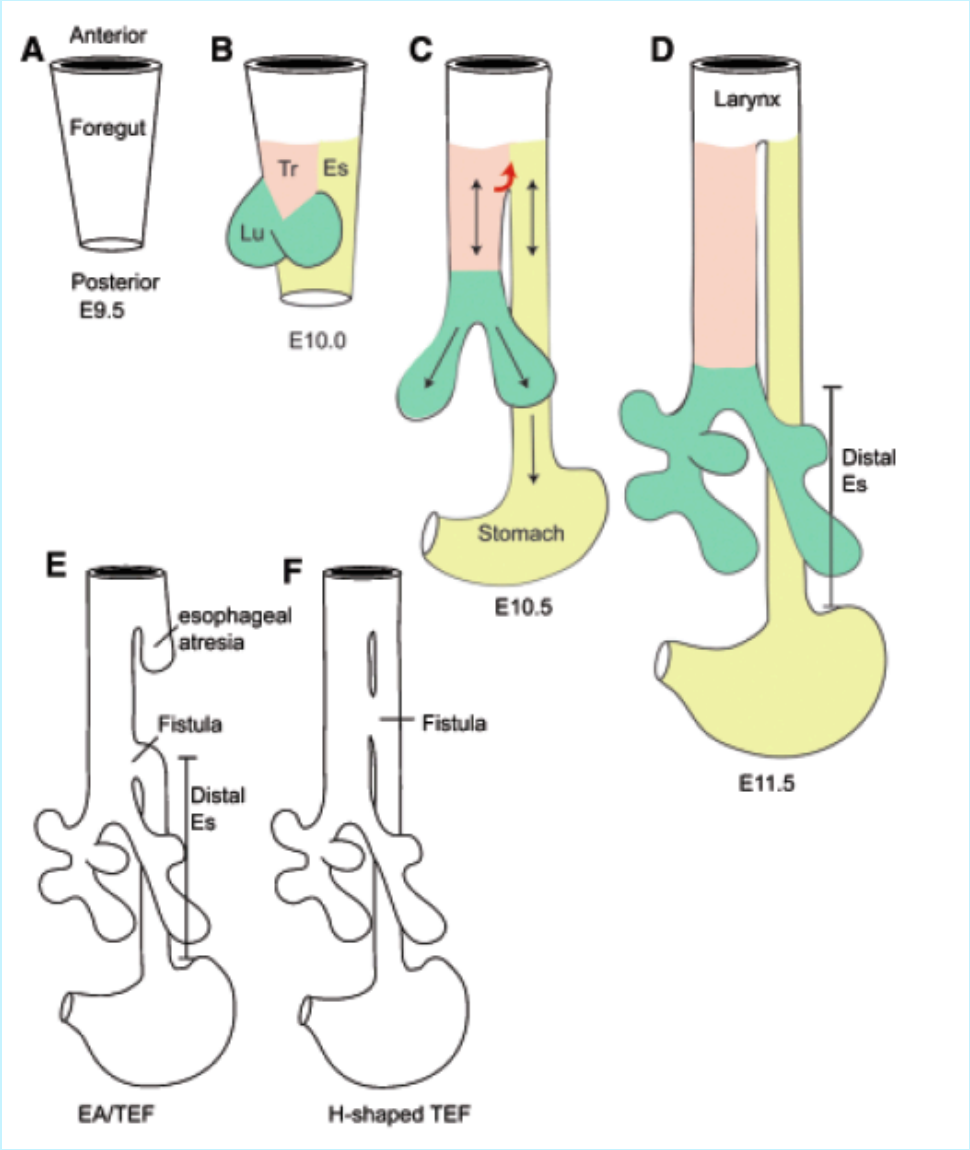
- At birth, fetal lung circulation is a high pressure that must convert to a low pressure circulation.
- As air enters the lung with the first breath, oxygen tension rises.
  - Increased nitric oxide production increases arterial vasodilation, reducing pulmonary arterial pressure.

Molecular Determinants of Lung Development

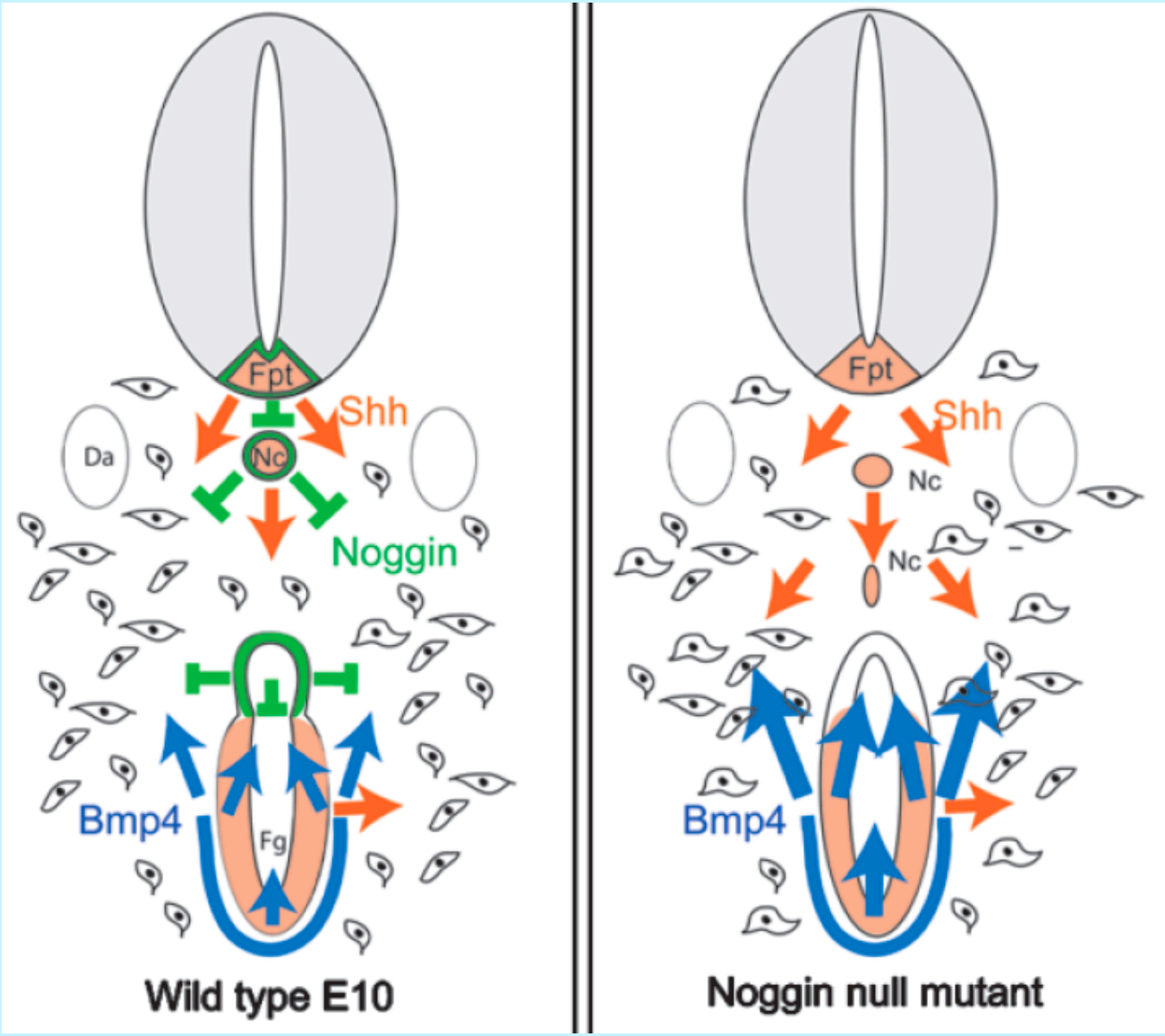
- Production of laryngotracheal groove correlates with the appearance of retinoic acid in the ventral mesoderm. If RA is blocked, foregut will not produce the lung bud (Desai, 2004).
- Regional specificity of the mesenchyme determines differentiation of the developing respiratory tube (Wessels, 1970)
  - Neck-grows straight forming trachea
  - Thorax-branches
- Retinoic acid induces formation of Fgf10 by activating Tbx4 in the splanchnic mesoderm adjacent to the ventral foregut (Sakiyama, 2003).

Defect in Embryonic Phase

- Shh null embryos delay lung bud emergence and no separation of a trachea and esophagus (Littingtung, 1998).
- Nkx2.1 null mice develop severe defects in separation of trachea and esophagus (Minoo, 1999).
- RAR a1-/-/b2-/- mutant embryos fail to separate. Nkx2.1 is regulated by RA signaling (Mendelsohn, 1994)
- Disruption of Bmp4-noggin antagonism (Que, et al, 2006)


A
Anterior
Foregut
Posterior
E9.5
B
Tr
Es
Lu
E10.0
C
Stomach
E10.5
D
Larynx
Distal
Es
E11.5
E
esophageal
atresia
Fistula
Distal
Es
EA/TEF
F
Fistula
H-shaped TEF

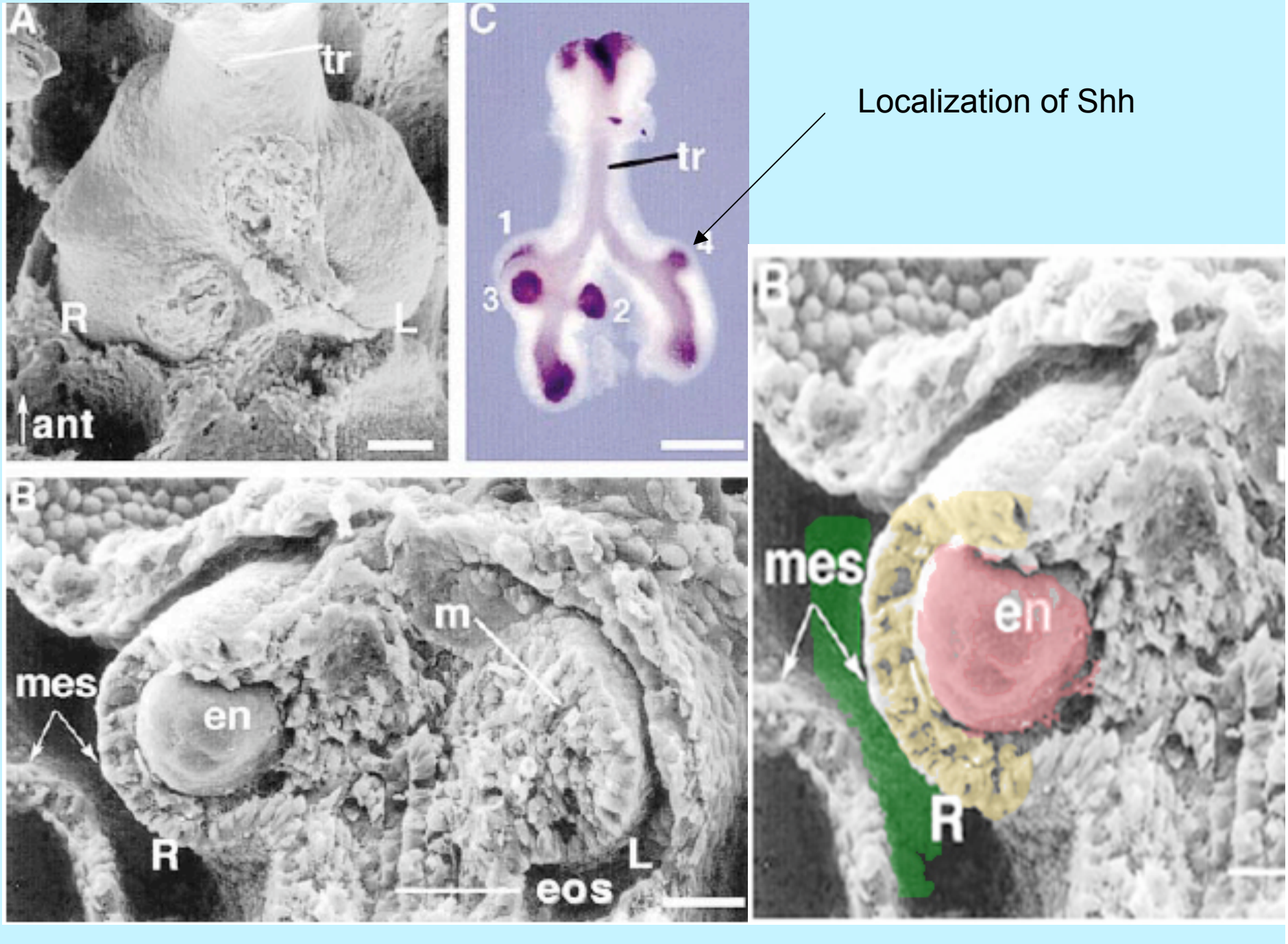
Mouse gene	Molecular function	Foregut phenotypes	References
Foxf1	Forkhead family transcription factor	Hets have narrow esophagus or TEF, abnormal lungs	Mahlapuu et al. (2001)
Gil2 &Gli3	Hedgehog pathway transcription factors	EA/TEF, abnormal lung development	Motoyama et al. (1998)
Nkx2.1	Homeodomain-containing transcription factor	Es and Tr do not separate, abnormal lung development	Minoo et al. (1999)
Noggin	Secreted BMP antogonist	EA/TEF in about 60% of heterozygotes	This work
RAR α & β2	Retinoic Acid Receptors, nuclear hormone receptor superfamily	Failure of foregut separation and abnormal cartilage development in some compound	Mendelsohn et al. (1994)
Shh	Secreted hedgehog family ligand	EA/TEF, abnormal lung development	Litingtung et al. (1998)

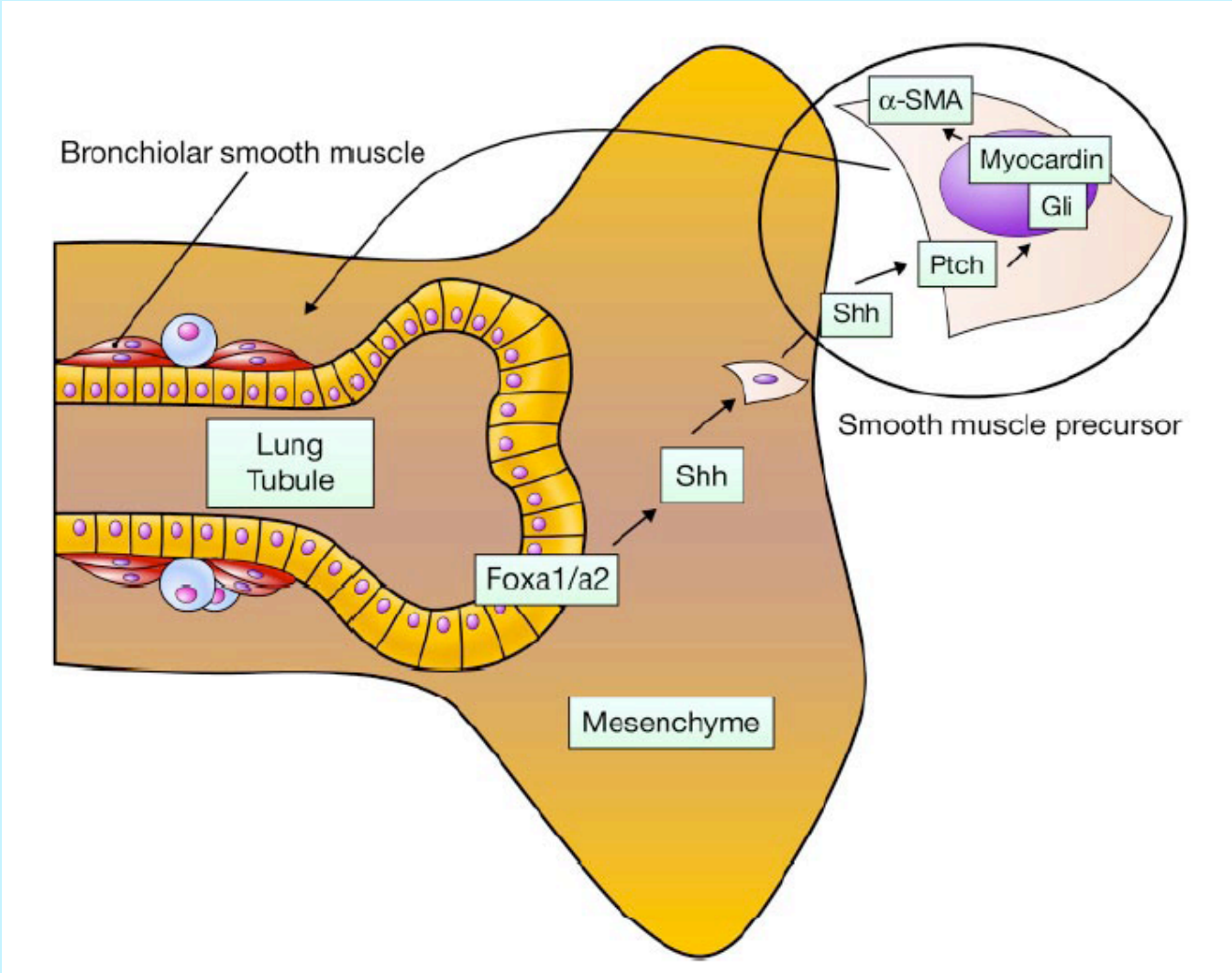


(Que, et al, Differentiation 2006)

Inhibitors to block branching process

- Competitive inhibitor of integrin receptors
  - Abnormal morphology of the primordium
- Monoclonal antibodies to laminin
  - Reduced terminal buds and segmental dilation of the explanted primordia
- Antisense oligonucleotides against TTF-1 (Thyroid Transcription Factor-1)
  - Reduction of terminal branches of the lung primordium
- Knockout mice experiments

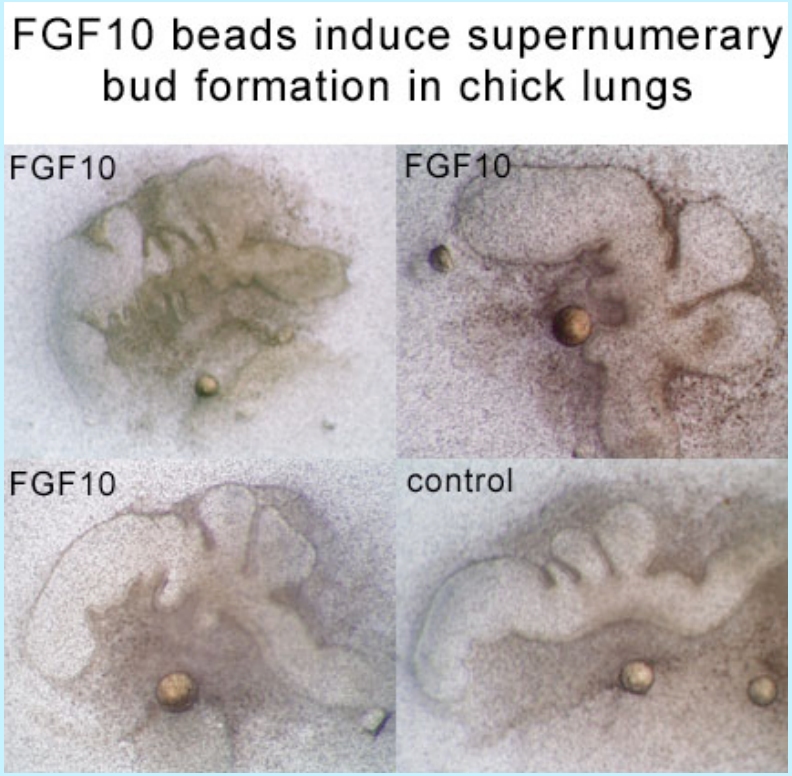

A
tr
R
L
ant
C
tr
1
4
3
2
Localization of Shh
B
mes
en
m
R
L
eos
B
mes
en
R

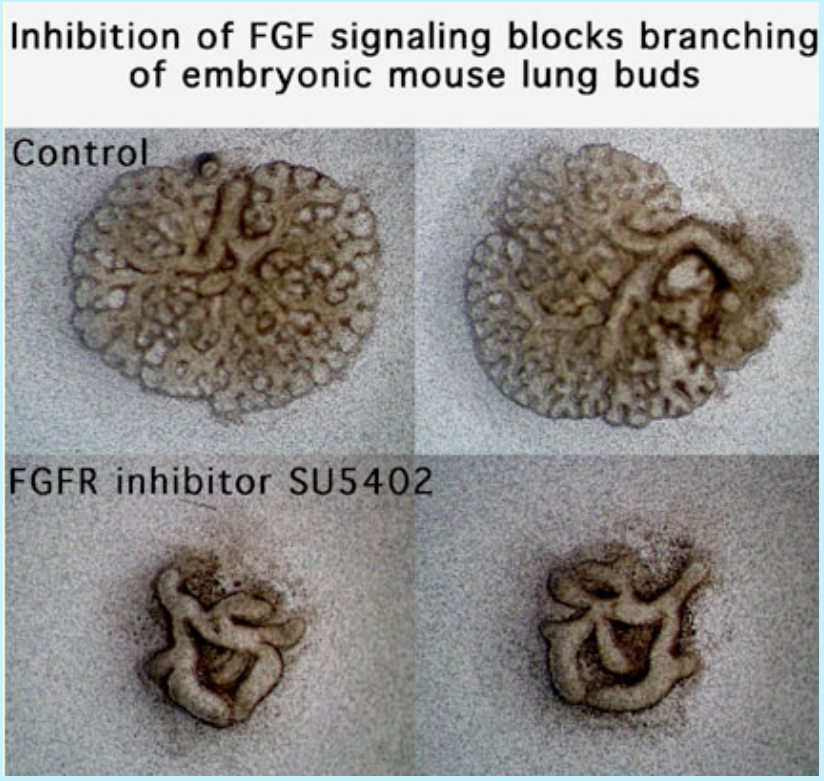


Maeda, et al 2007

FGF Ligands and Receptors Direct Epithelial Migration and Differentiation

- FGF10 promotes directed growth of the lung epithelium and induces both proliferation and chemotaxis of isolated endoderm.
- The chemotaxis response of the lung endoderm to FGF10 induces the coordinated movement of the entire epithelial tip towards an FGF10 source.





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