Angiogenesis in Human Development

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BACKGROUND READING:

Vascular Development

"Signaling Vascular Morphogenesis and Maintenance"
Douglas Hanahan. *Science* 277: 48-50. in Perspectives. (1997)

VEGF Signaling

"VEGF Receptor Signaling – in control of vascular function"
Ollsson, Dimburg, Krueger, and Lena Claesson-Welsh. Nat. Rev. Mol. Cell Biol. 7: 359-71 (2006)

Notch and arterial specification

"Arterial Venous Specification During Development"
Matthew Swift and Brant M. Weinstein . *Circ. Res. 104*: 576-588 (2009)

Lymphatic Development

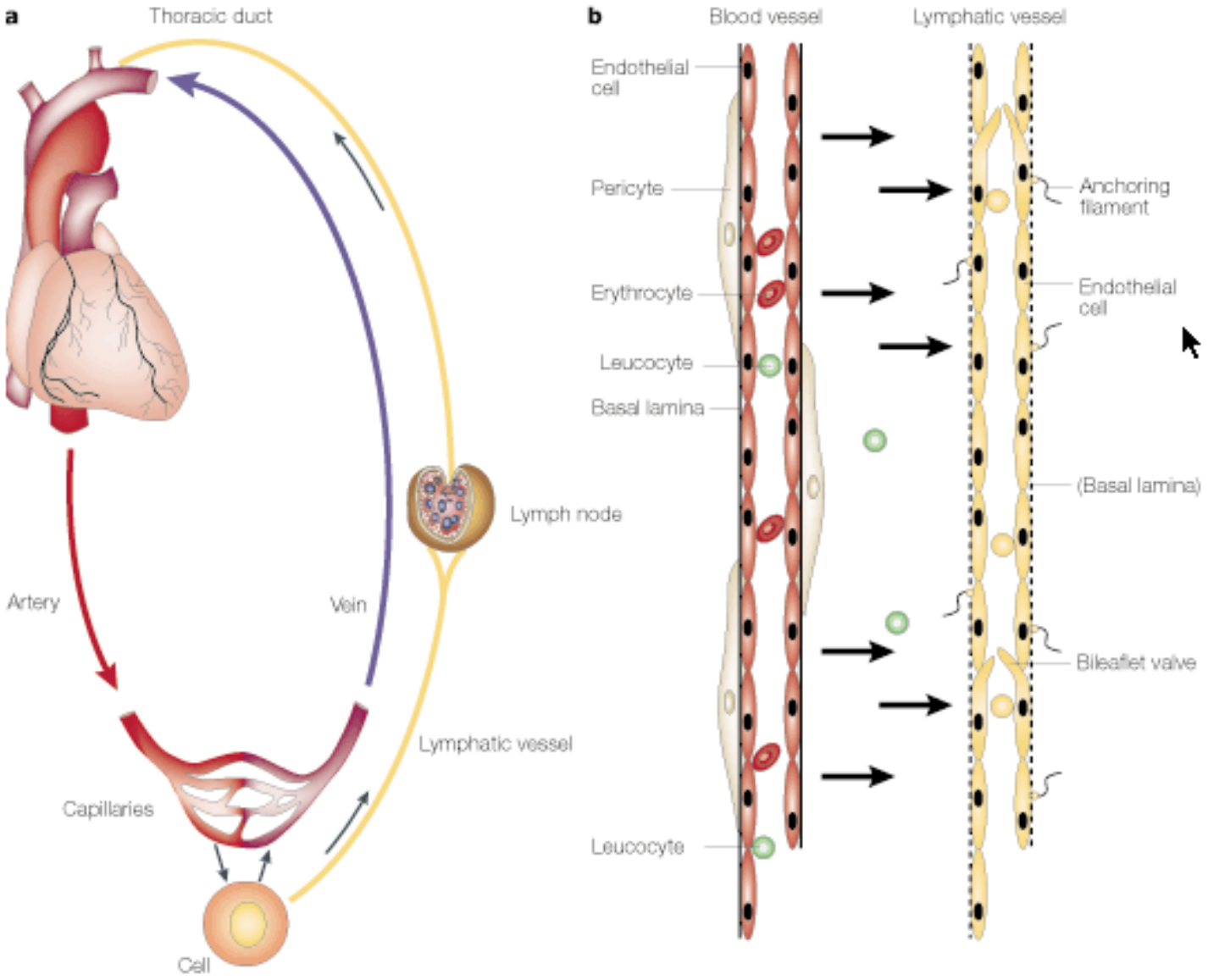
"Lymphatic Vascular Development"
Guillermo Oliver. *Nature Rev Immunology 4*: 35-45 (2004)

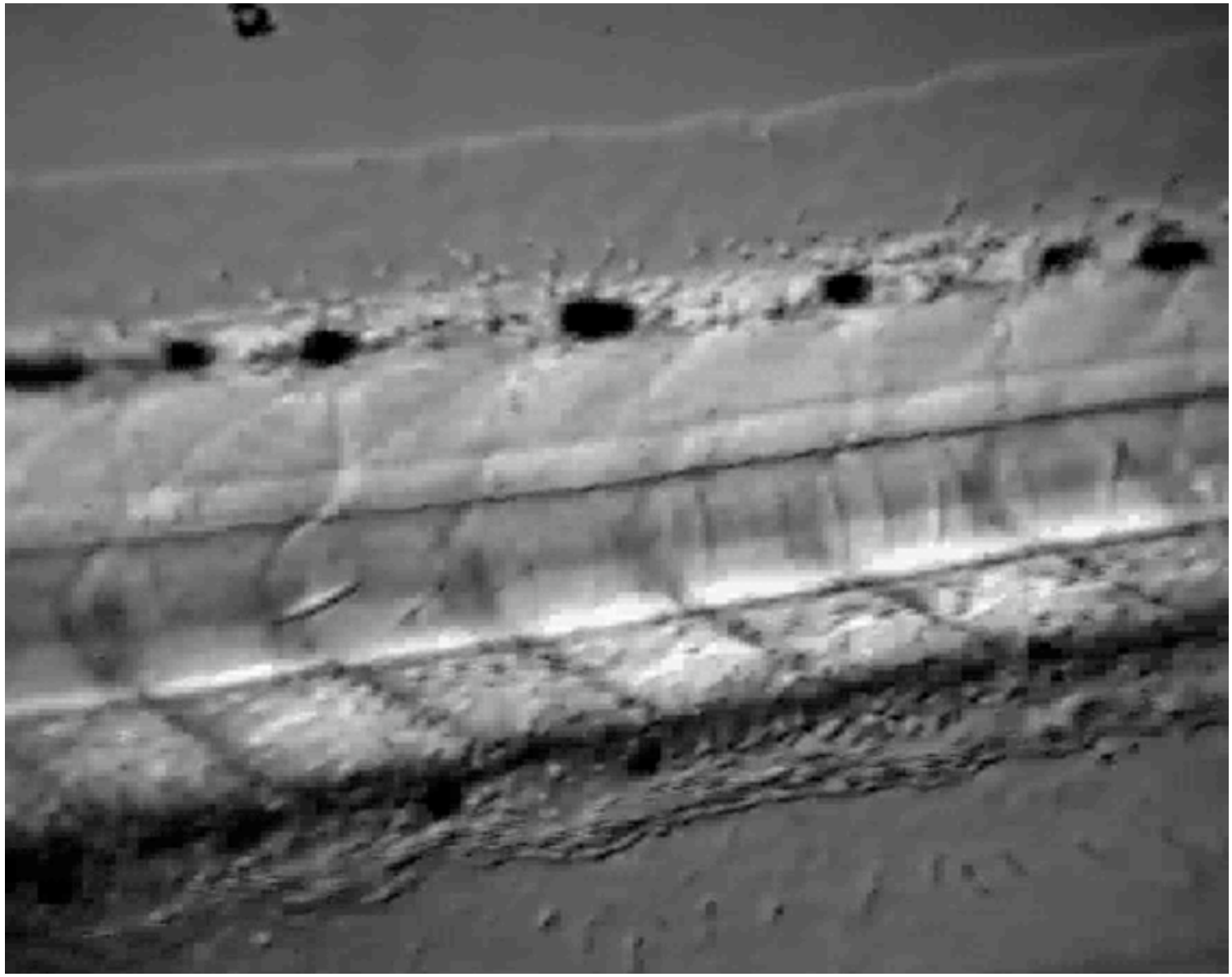
Wnts and retinal angiogenesis

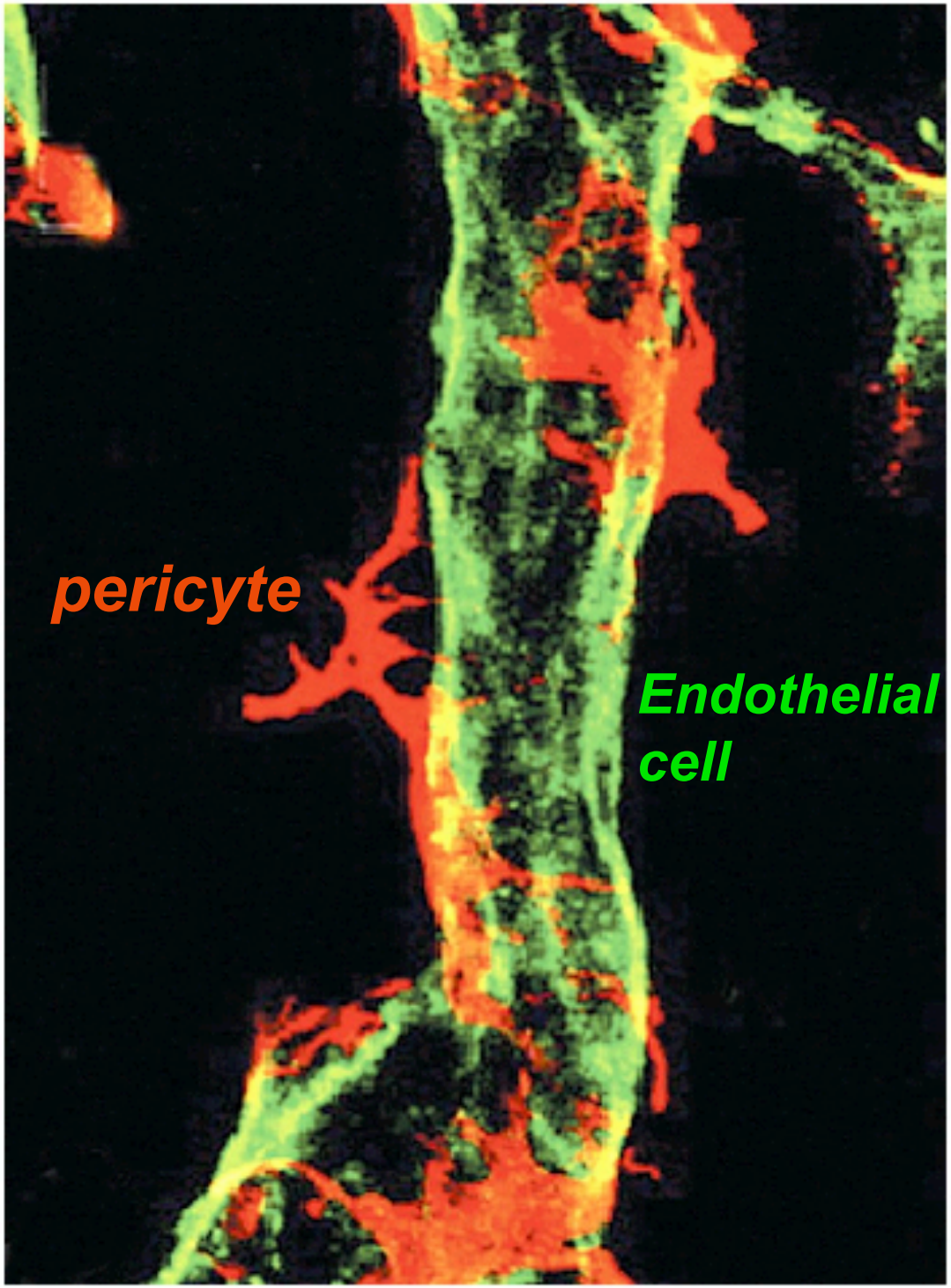
"Wnt/Frizzled Signaling in the Vasculature: New Angiogenic Factors in Sight"
Nestor Masckauchan and Jan Kitajewski. *Physiology 21*: 181-188 (2006)

Vascular Development

- Vasculogenesis = de novo tube formation
- Angiogenesis = sprouting of new tubes off of pre-existing tubes
- Cell types
  - Endothelial Cell = cell type that makes up and lines blood vessels
  - Mural Cells = specialized cells that surround blood vessels
    - Pericytes
    - Smooth muscle cells
- Angiogenic Factors
  - Vascular Endothelial Growth Factor (VEGF-A, VEGF-B, PlGF, VEGF-C…..)
  - Angiopoietins (Ang 1, Ang2, ……..)
  - Notch ligands (Jagged1, Delta4)


a
Thoracic duct
Lymph node
Artery
Vein
Lymphatic vessel
Capillaries
Cell
b
Blood vessel
Lymphatic vessel
Endothelial cell
Pericyte
Erythrocyte
Leucocyte
Basal lamina
Leucocyte
Anchoring filament
Endothelial cell
(Basal lamina)
Bileaflet valve





gure 13.33 *The Biology of Cancer* (© Garland Science 2007)

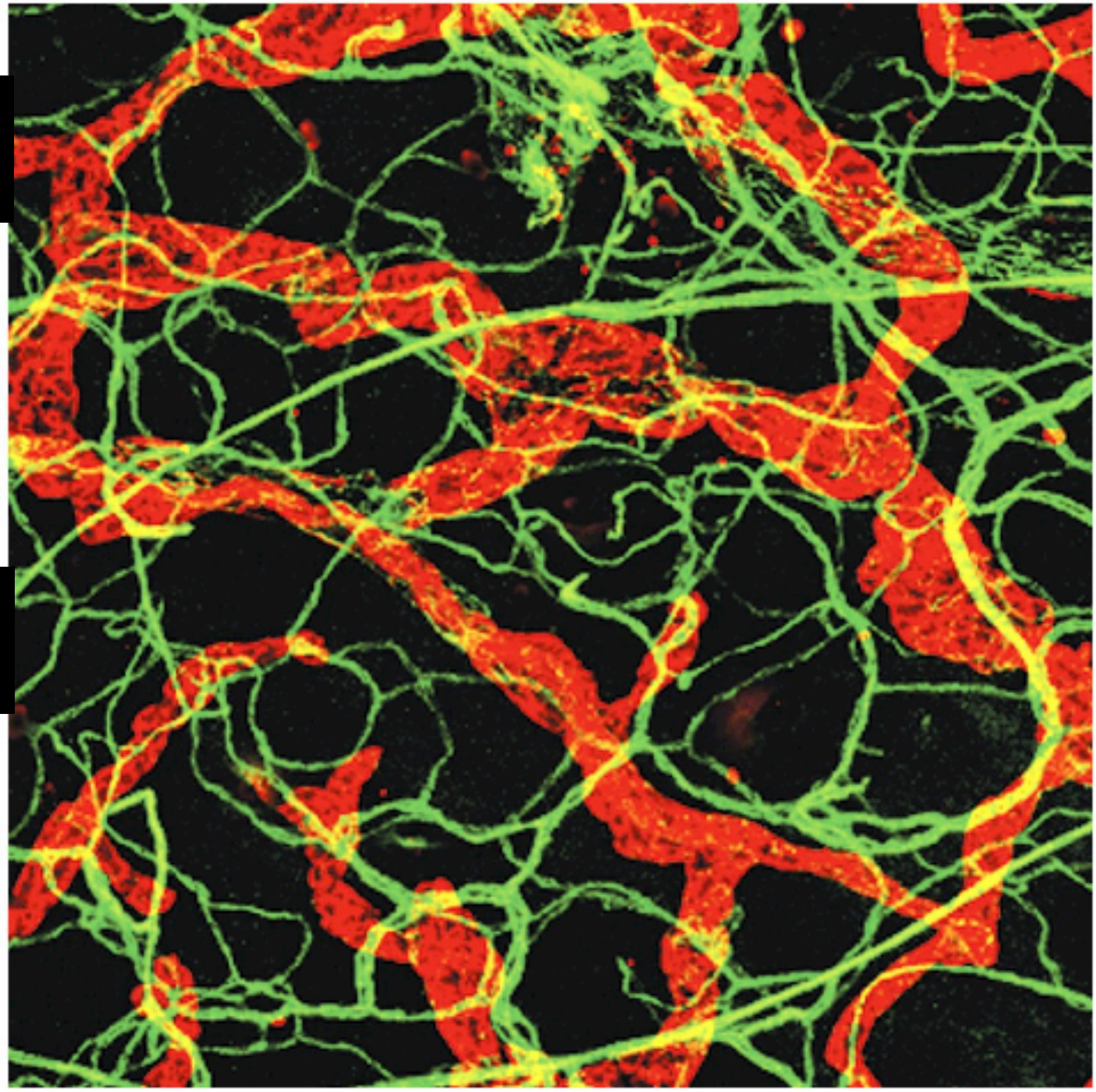
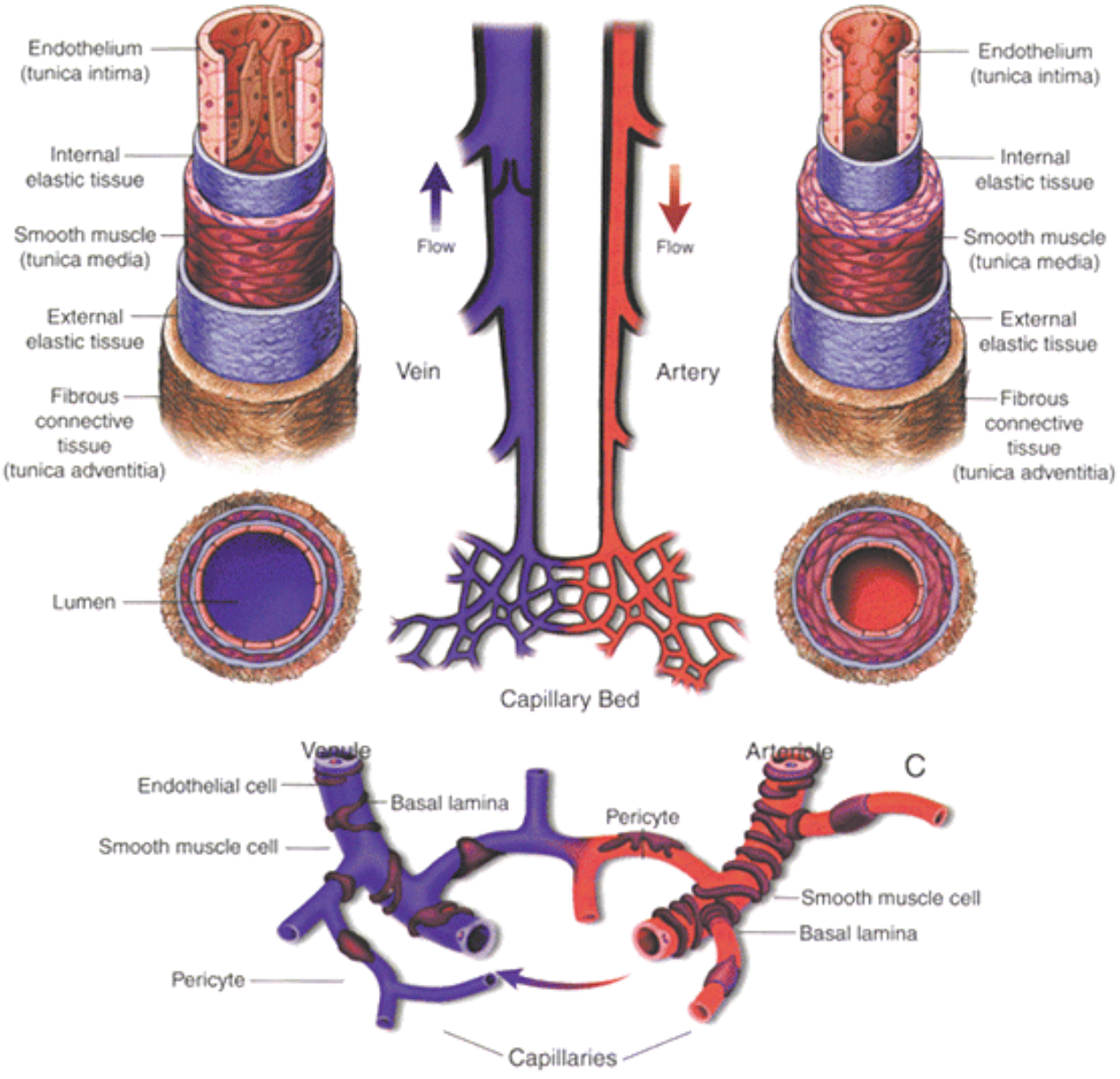
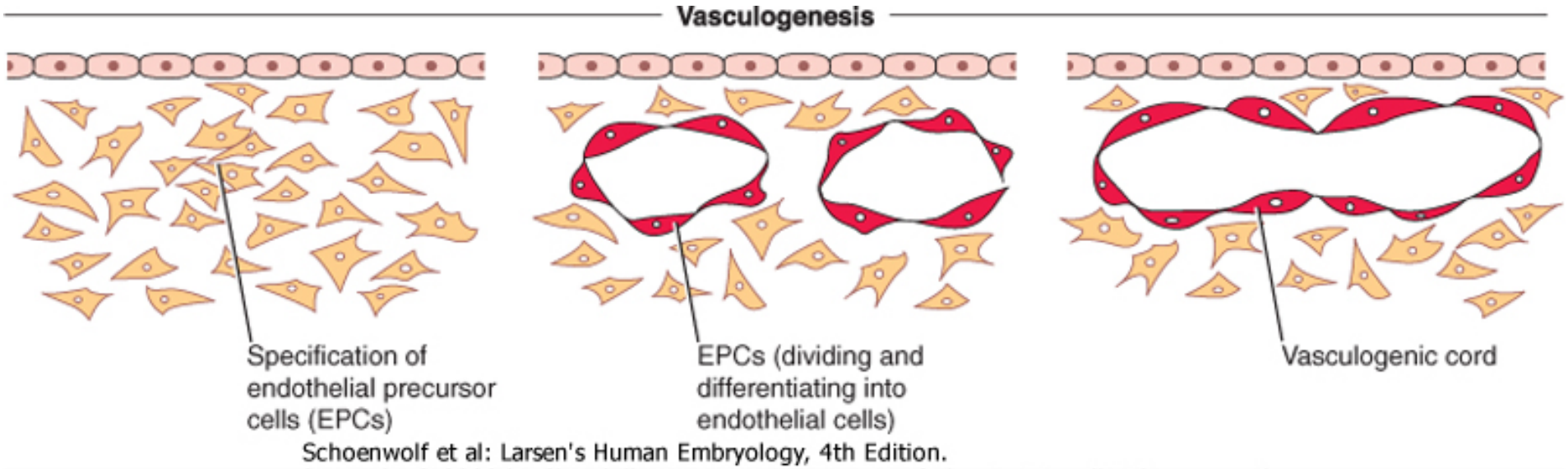


Figure 13.31 *The Biology of Cancer* (© Garland Science 2007)

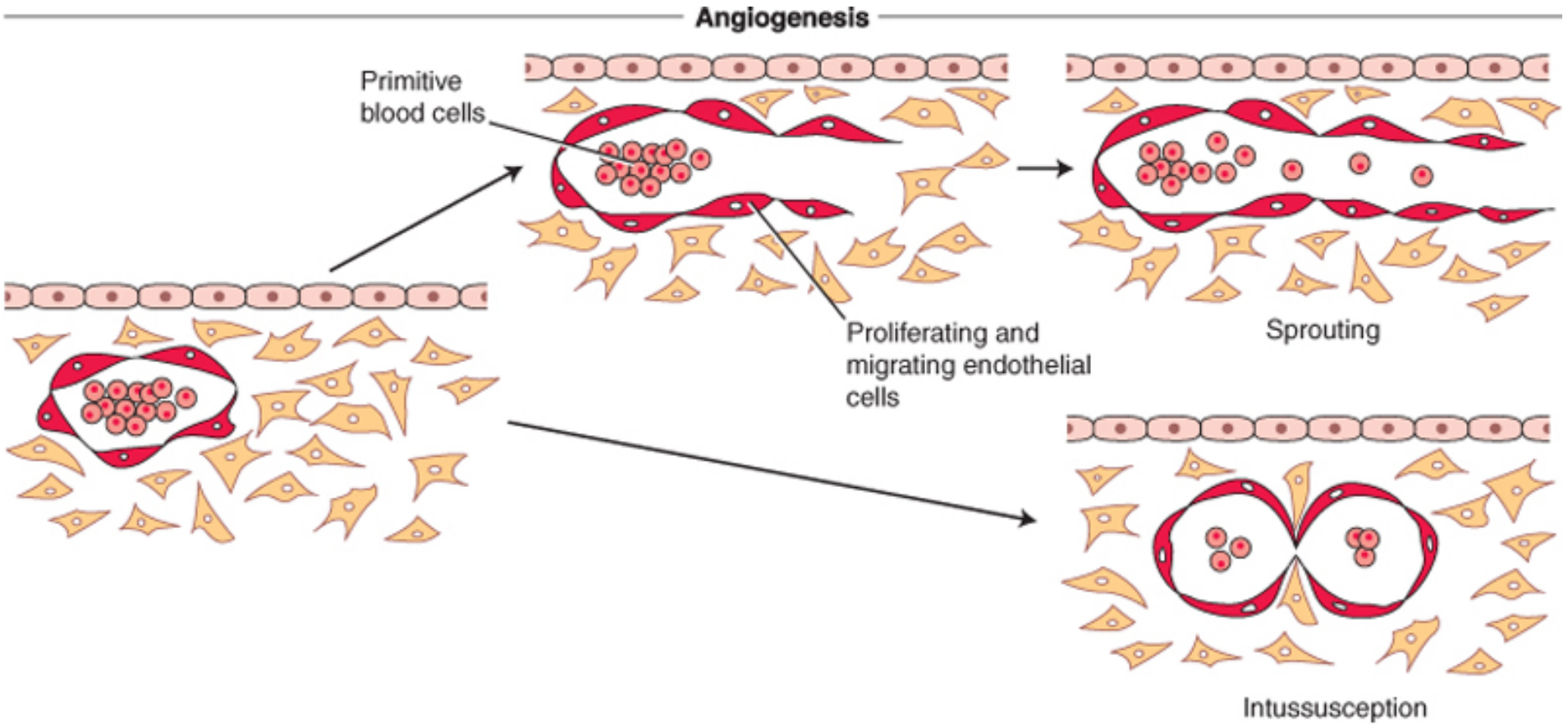

Endothelium (tunica intima)
Internal elastic tissue
Smooth muscle (tunica media)
External elastic tissue
Fibrous connective tissue (tunica adventitia)
Lumen
Flow
Flow
Vein
Artery
Capillary Bed
Endothelium (tunica intima)
Internal elastic tissue
Smooth muscle (tunica media)
External elastic tissue
Fibrous connective tissue (tunica adventitia)
Venule
Arteriole
C
Endothelial cell
Basal lamina
Pericyte
Smooth muscle cell
Smooth muscle cell
Basal lamina
Pericyte
Capillaries



Schoenwolf et al: Larsen's Human Embryology, 4th Edition.

Vascular Development

VASCULOGENESIS



Schoenwolf et al: Larsen's Human Embryology, 4th Edition.

Vascular Development

ANGIOGENESIS

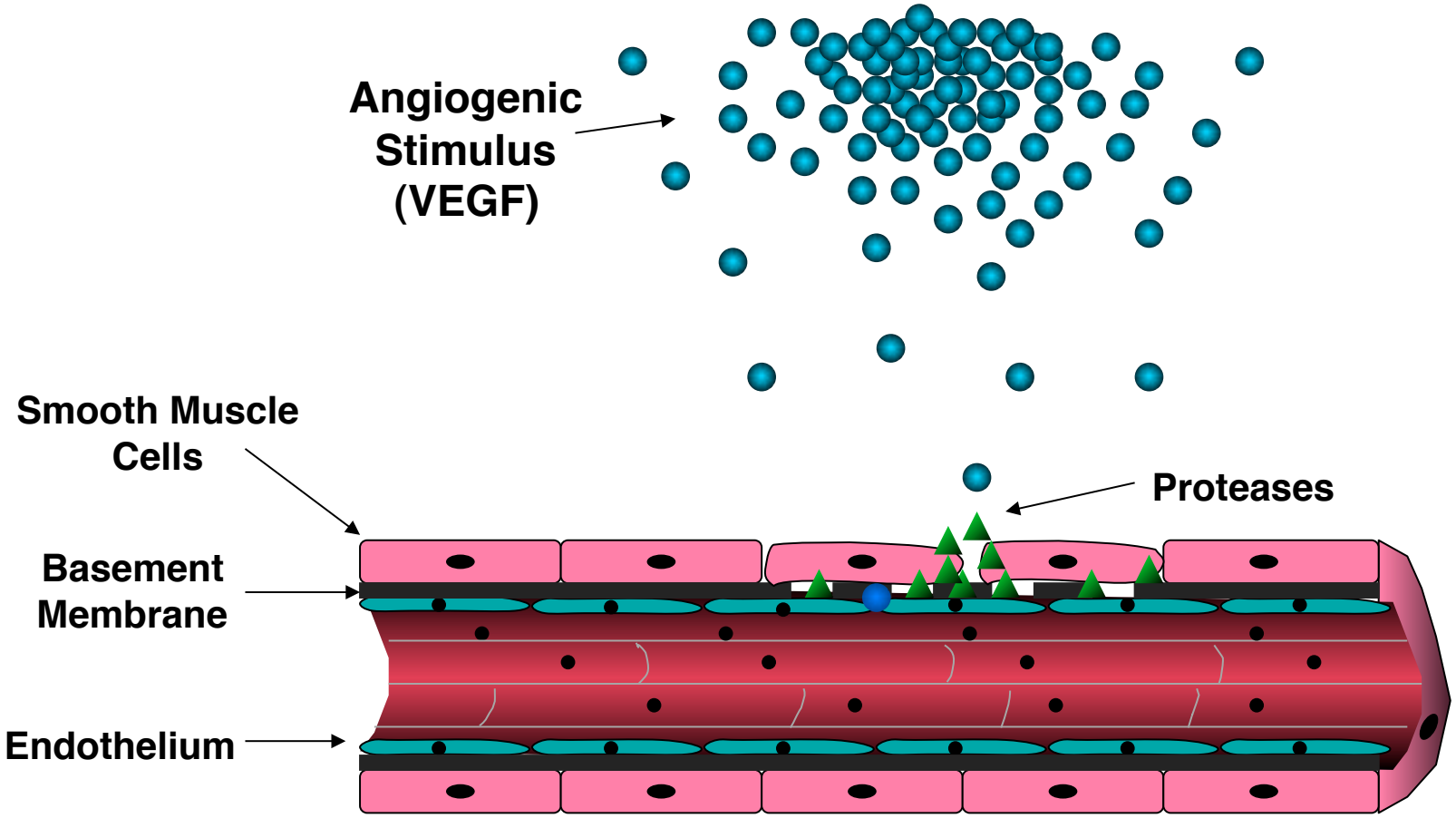
Vascular Development

ANGIOGENESIS

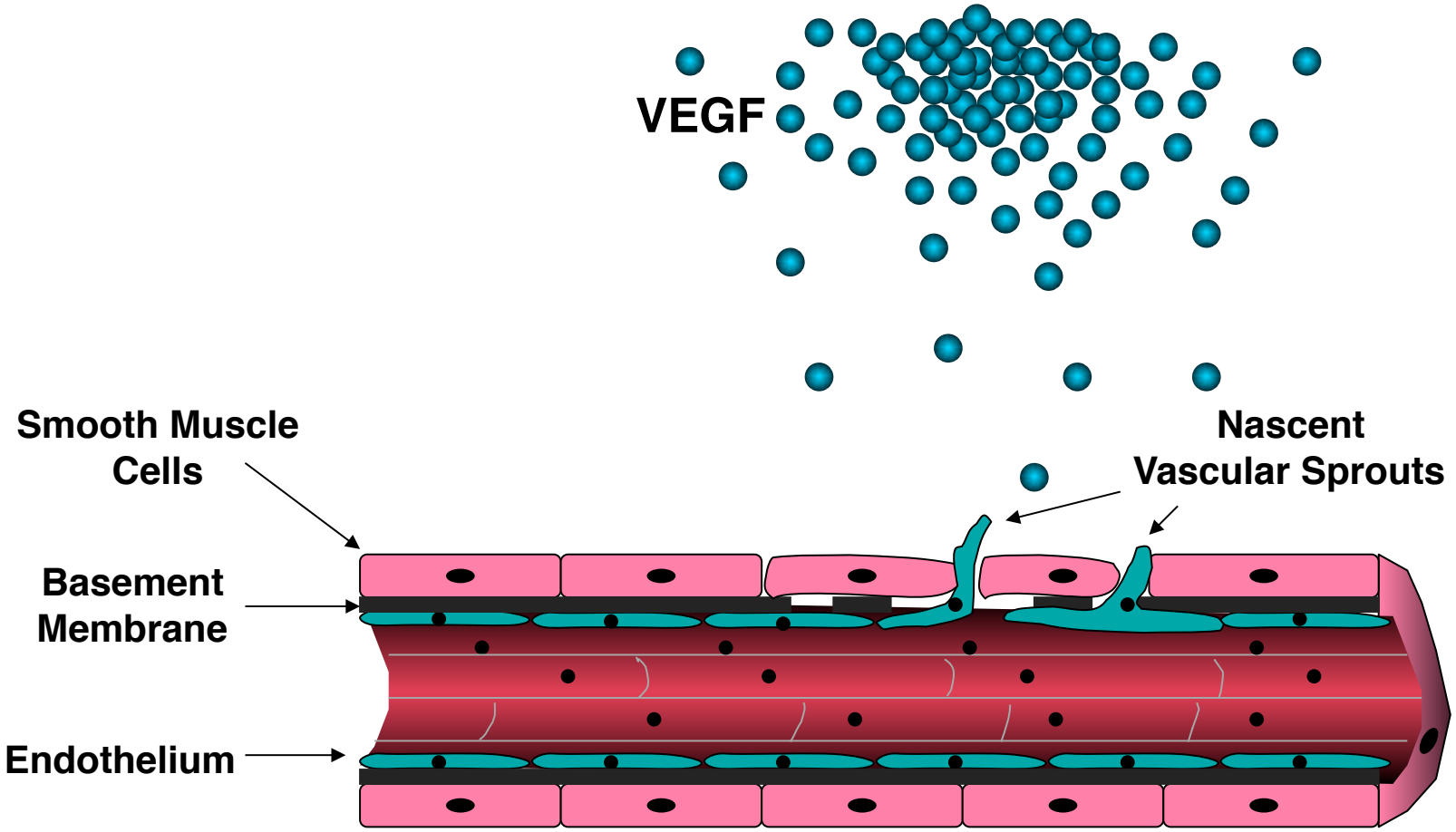
Cellular steps in Angiogenesis

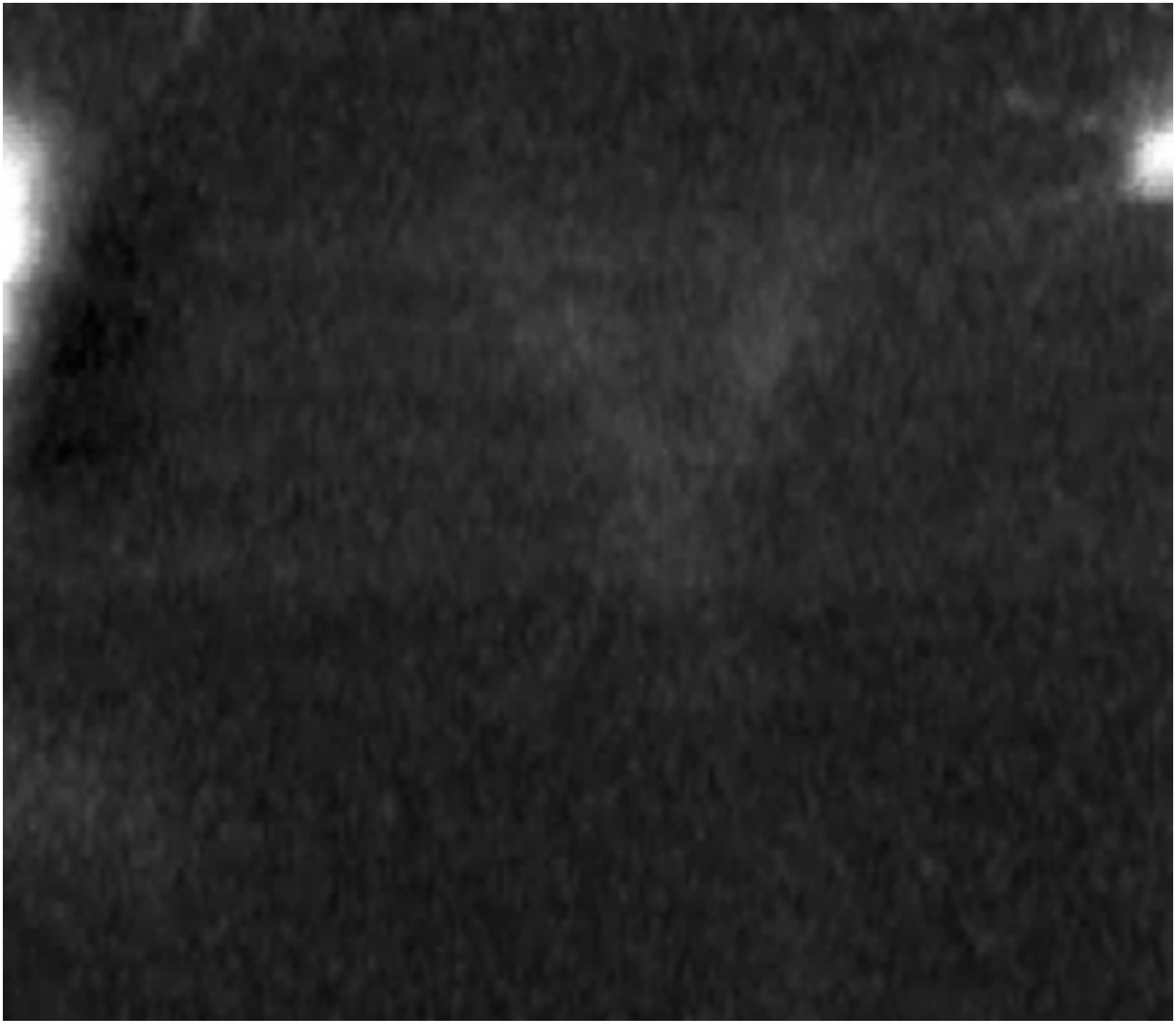
1) Biochemical Response and Preparation
2) Sprout initiation
3) Migration
4) Proliferation
5) Survival
6) Tube Formation
7) Maturation
8) Completion
9) Blood Flow

Angiogenesis - Basement Membrane Breakdown

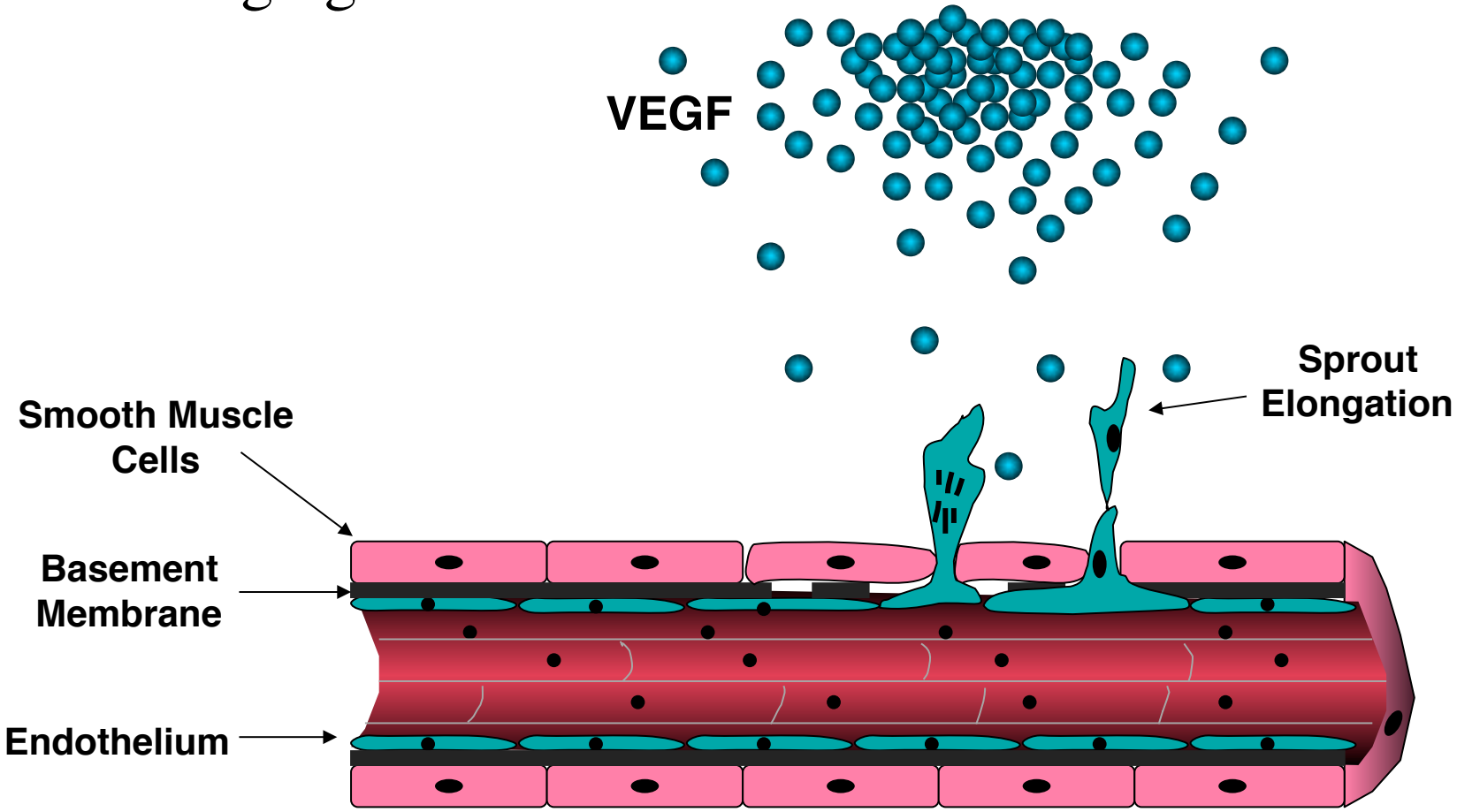


Angiogenesis - Endothelial Cell Migration

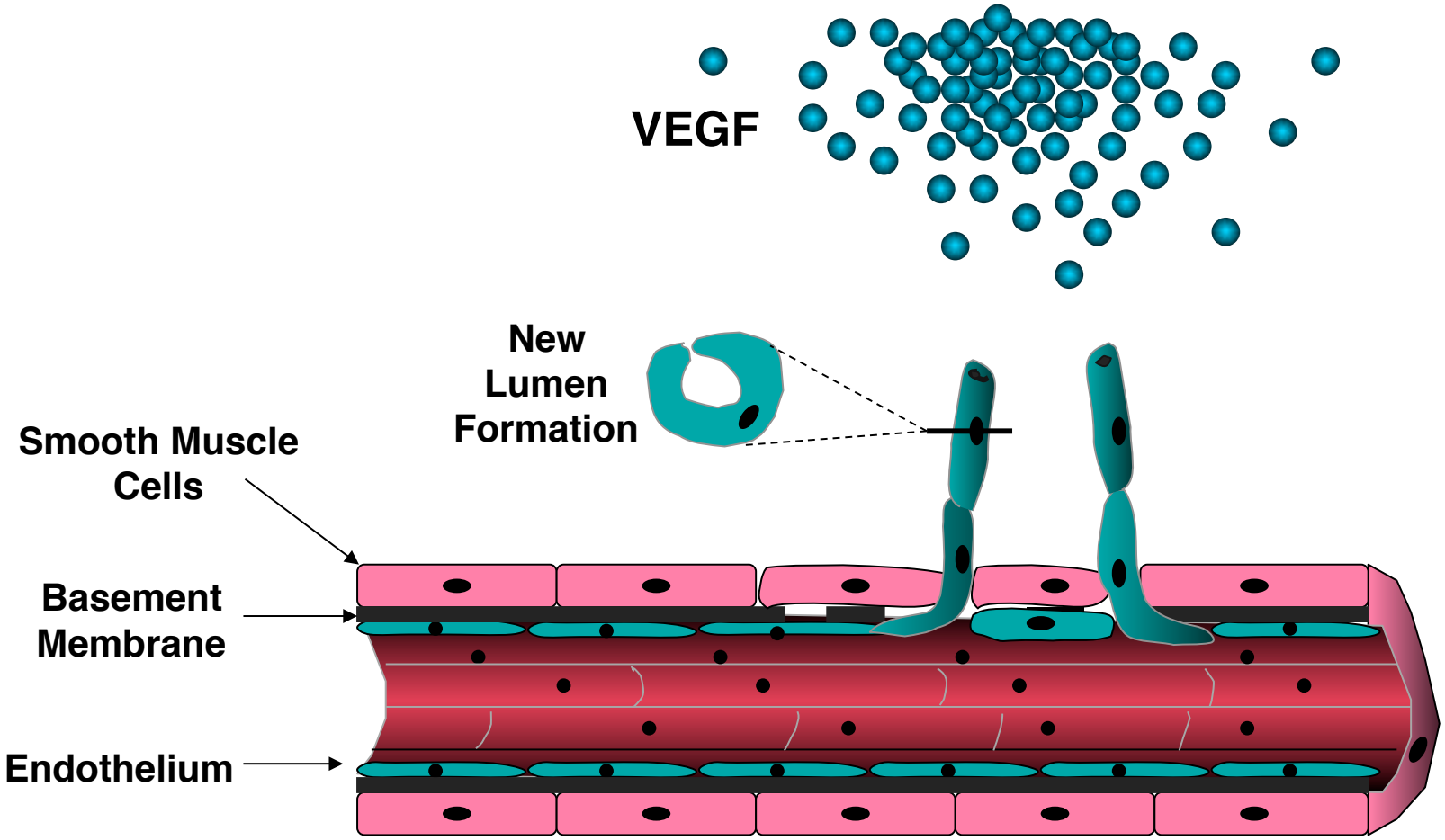


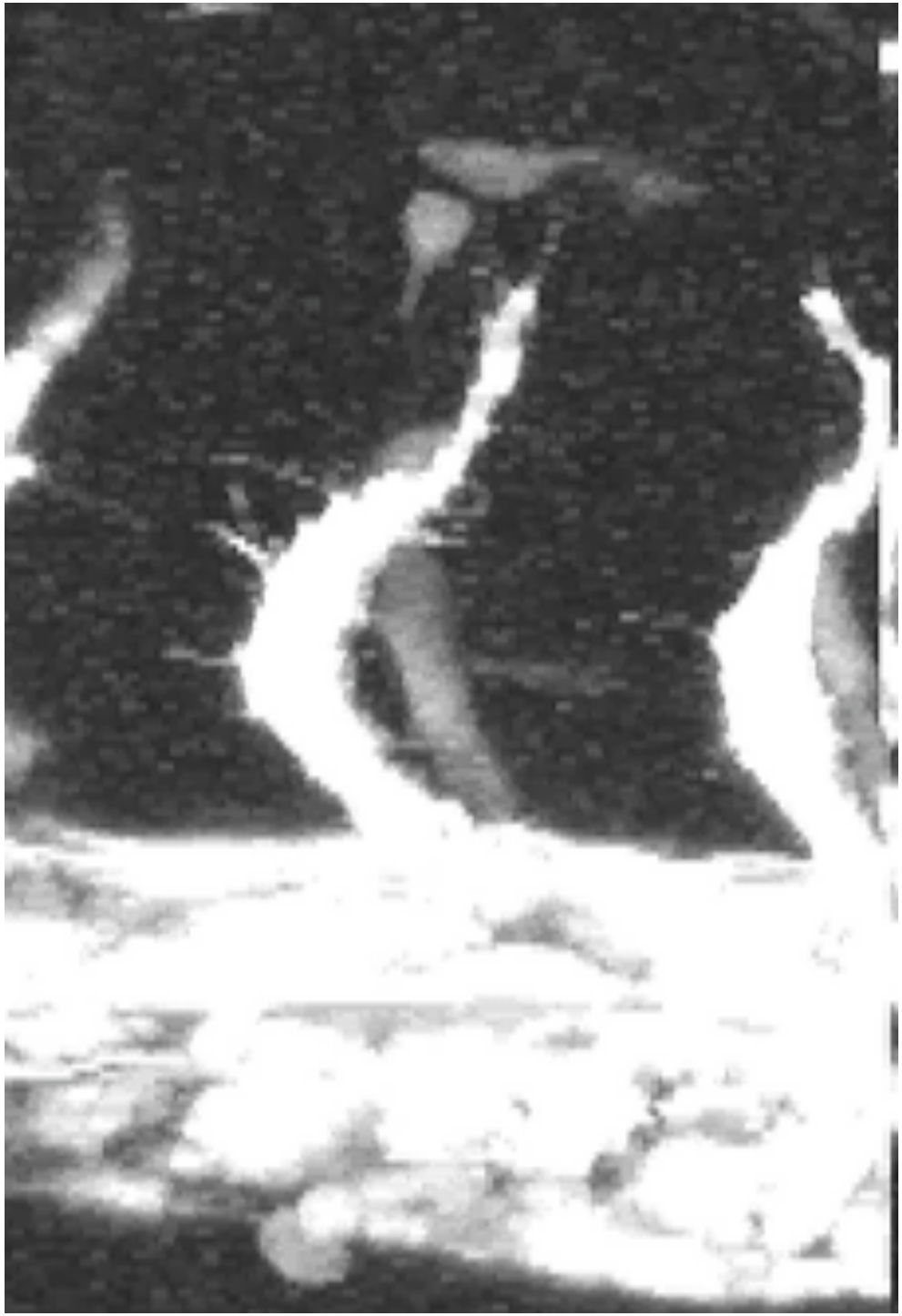


Angiogenesis - Endothelial Cell Proliferation

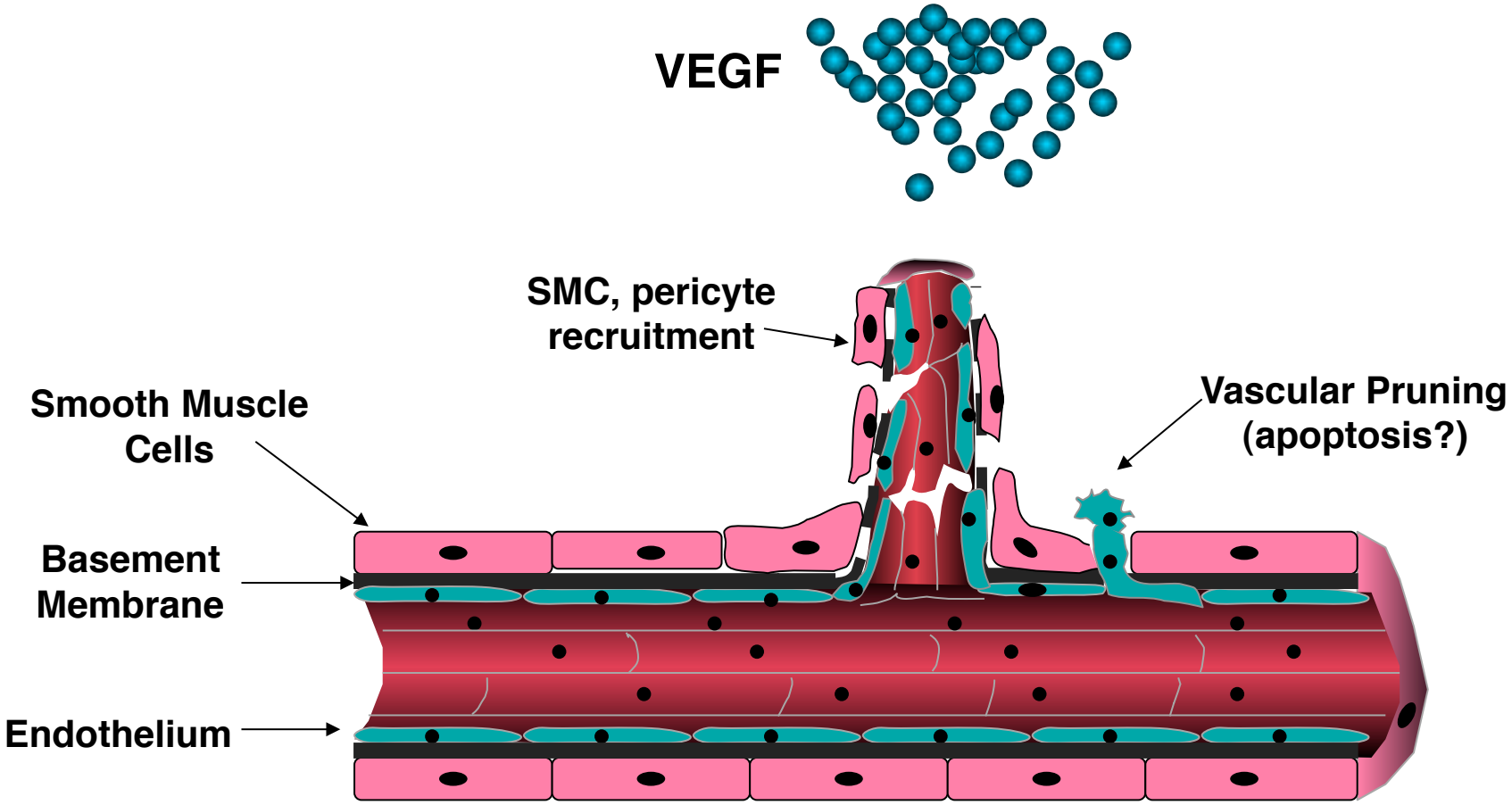


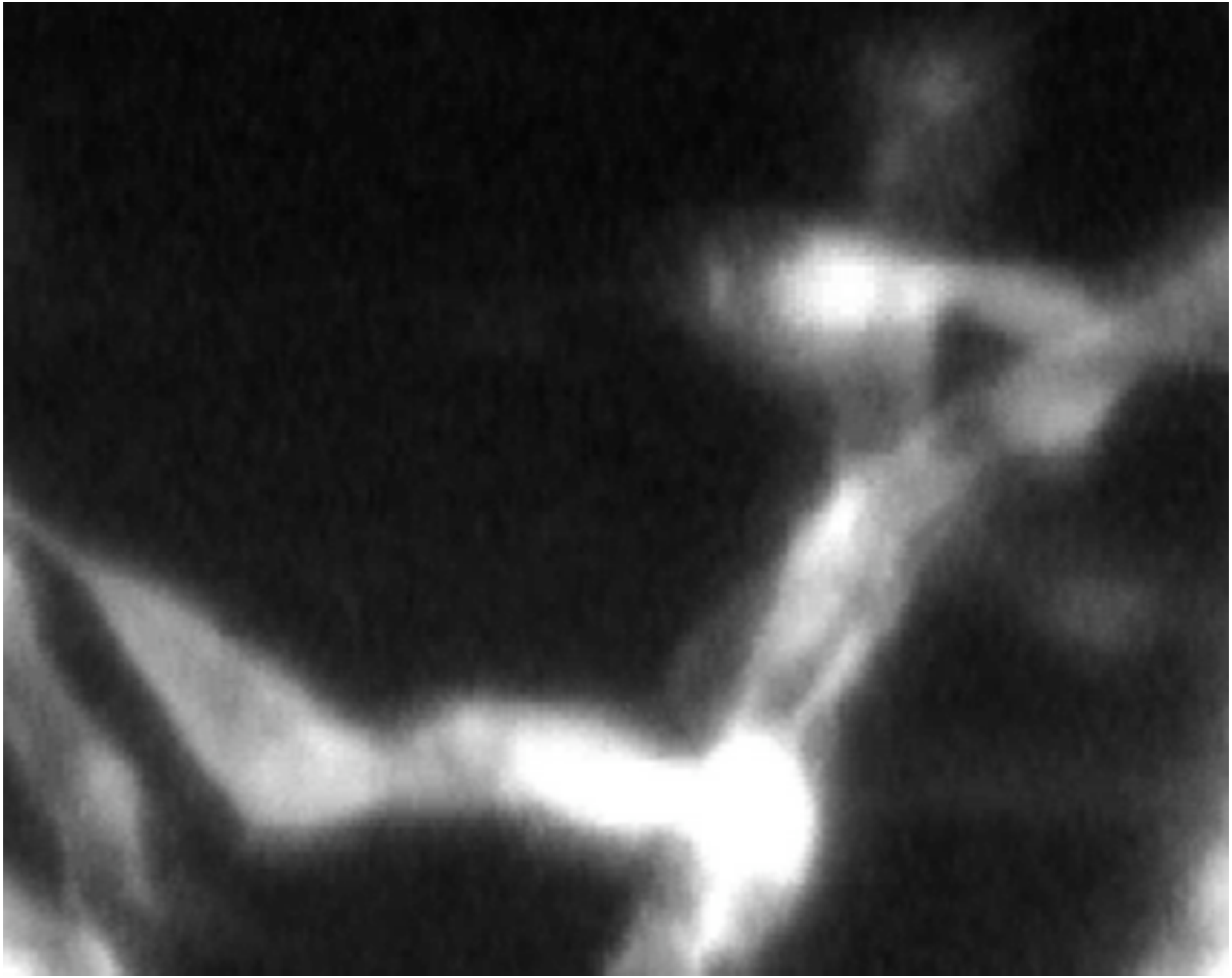
Angiogenesis - Capillary Morphogenesis



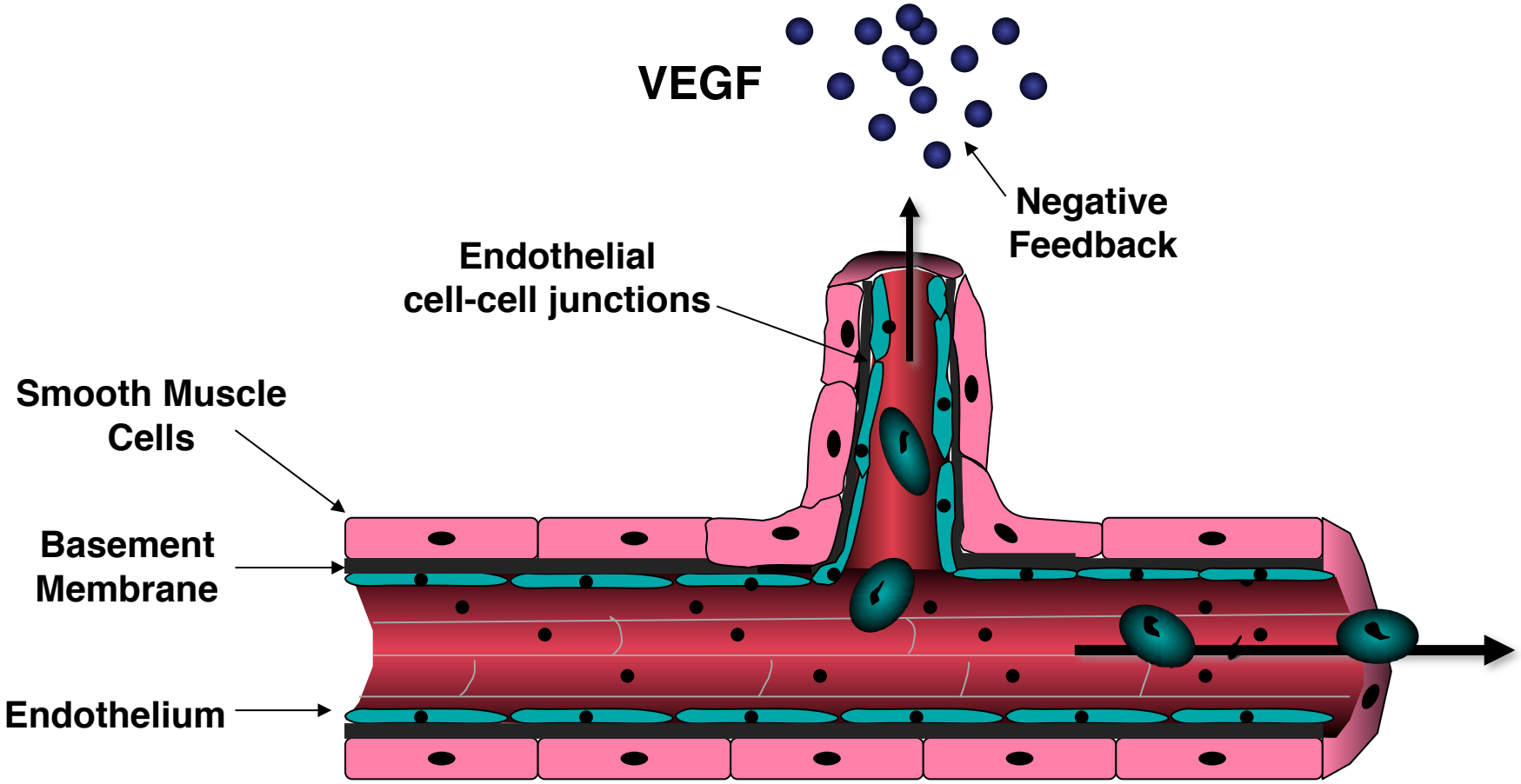


Angiogenesis - Vascular Maturation

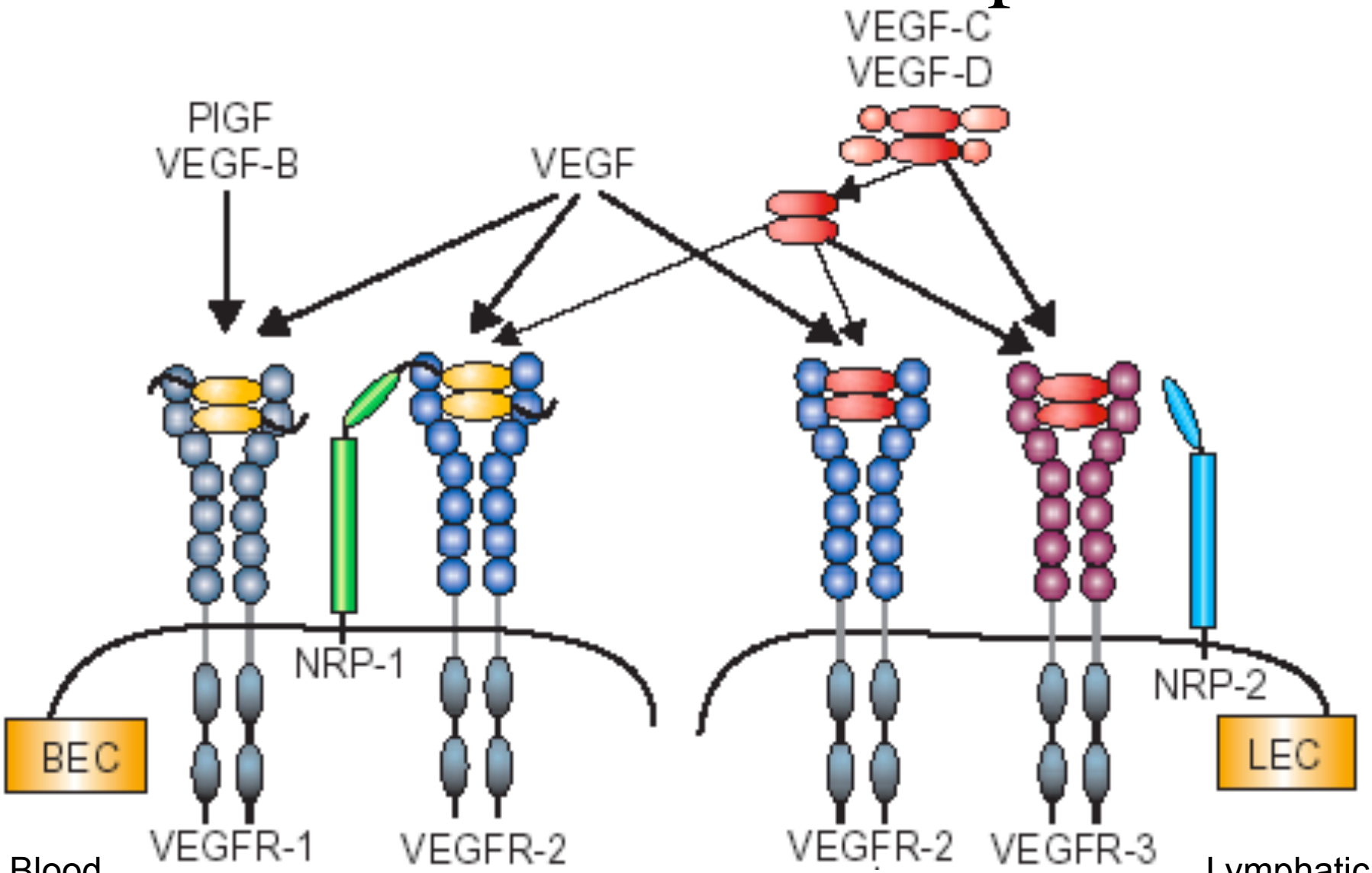




Angiogenesis - Vascular Maturation



VEGF and VEGF Receptors

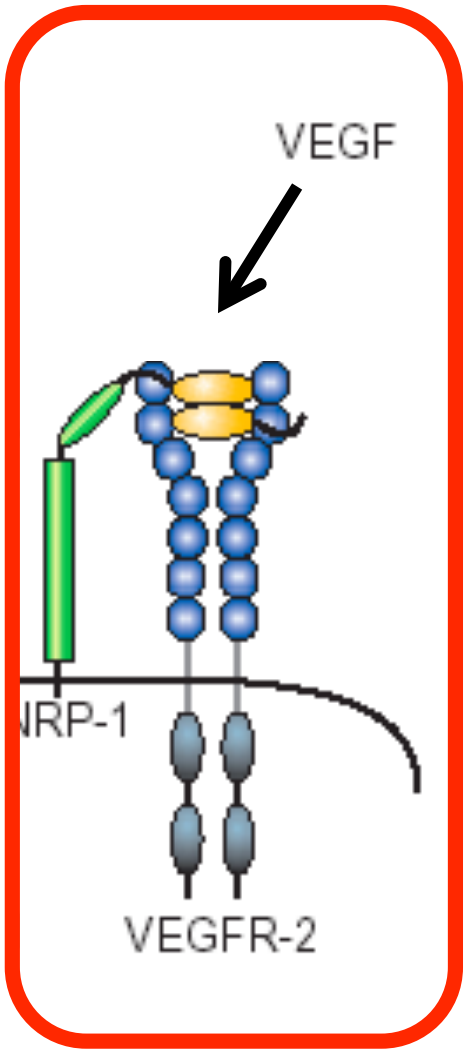


Blood Endothelial Cell

Lymphatic Endothelial Cell

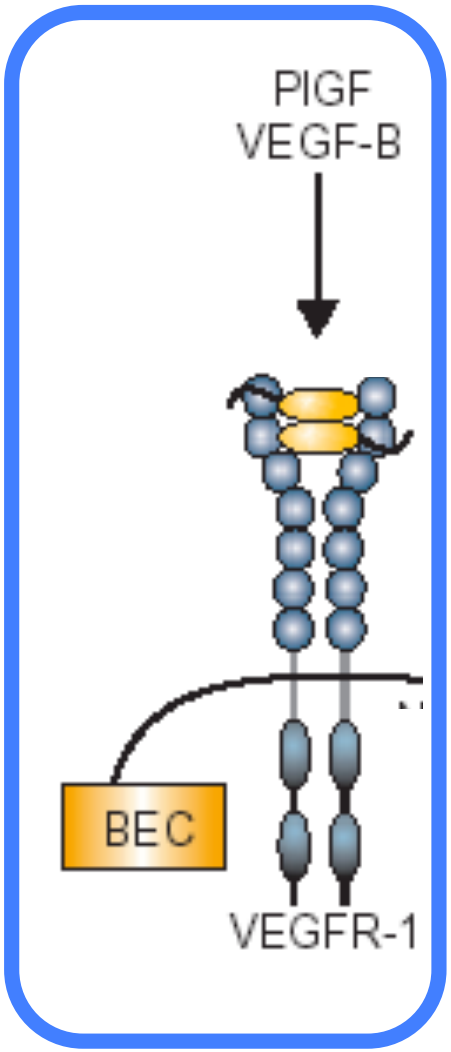
VEGF = VEGF-A angiogenic factor

VEGF Receptor-2 endothelial-specific signaling receptor



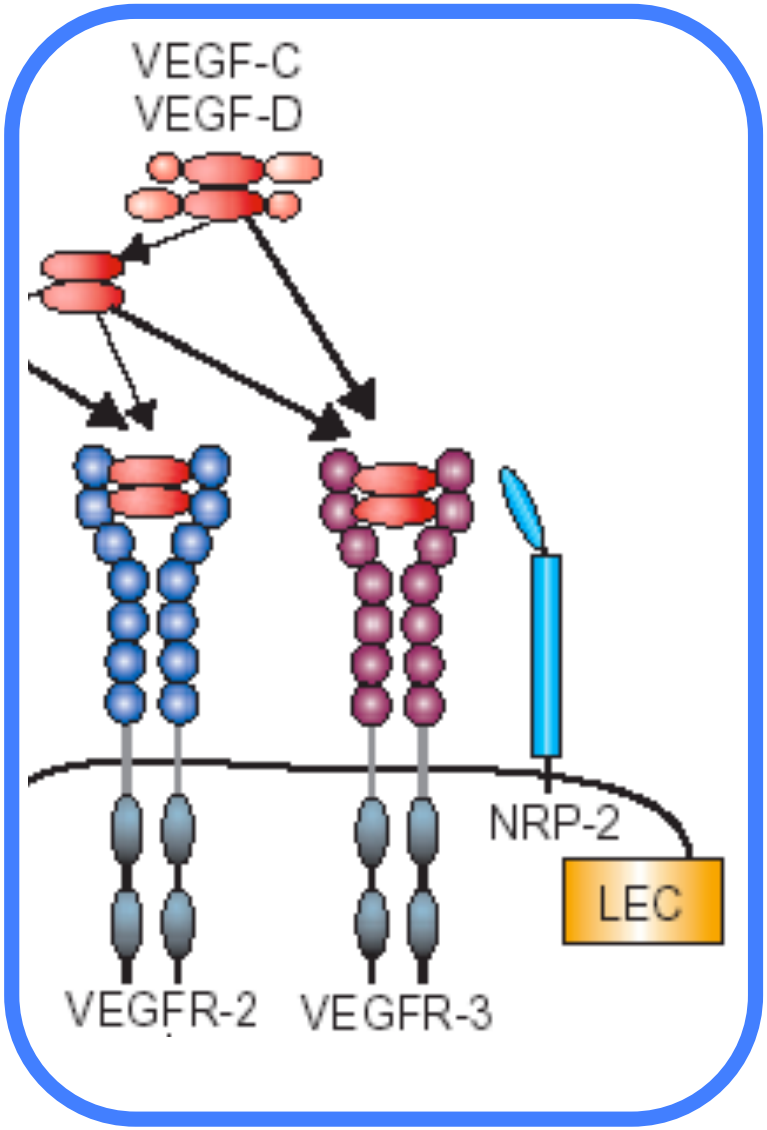
VEGF-B and PlGF
VEGF Receptor-1

specialized angiogenic factor
endothelial specific signaling



VEGF-C and VEGF-D lymphangiogenic factor

VEGF Receptor-3 lymphatic endothelial specific signaling



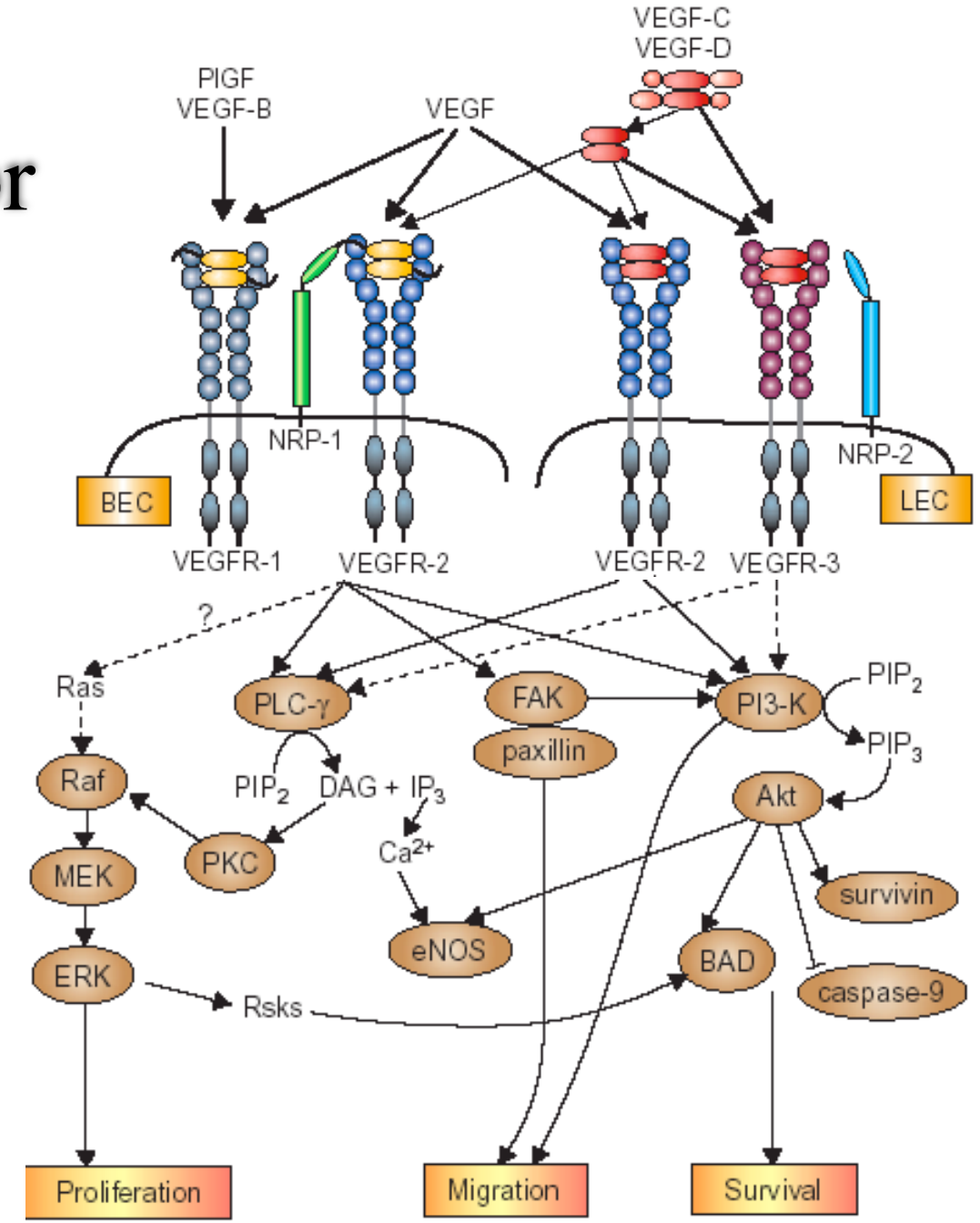
VEGF-receptor signaling

Promotes:

Proliferation

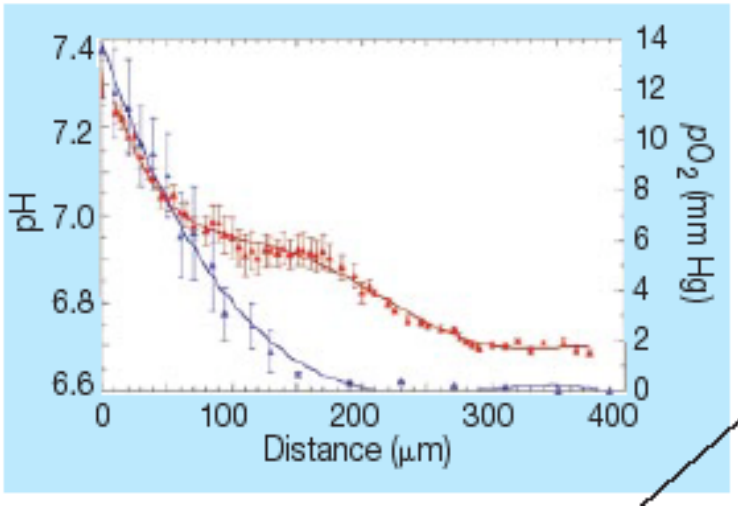
Migration

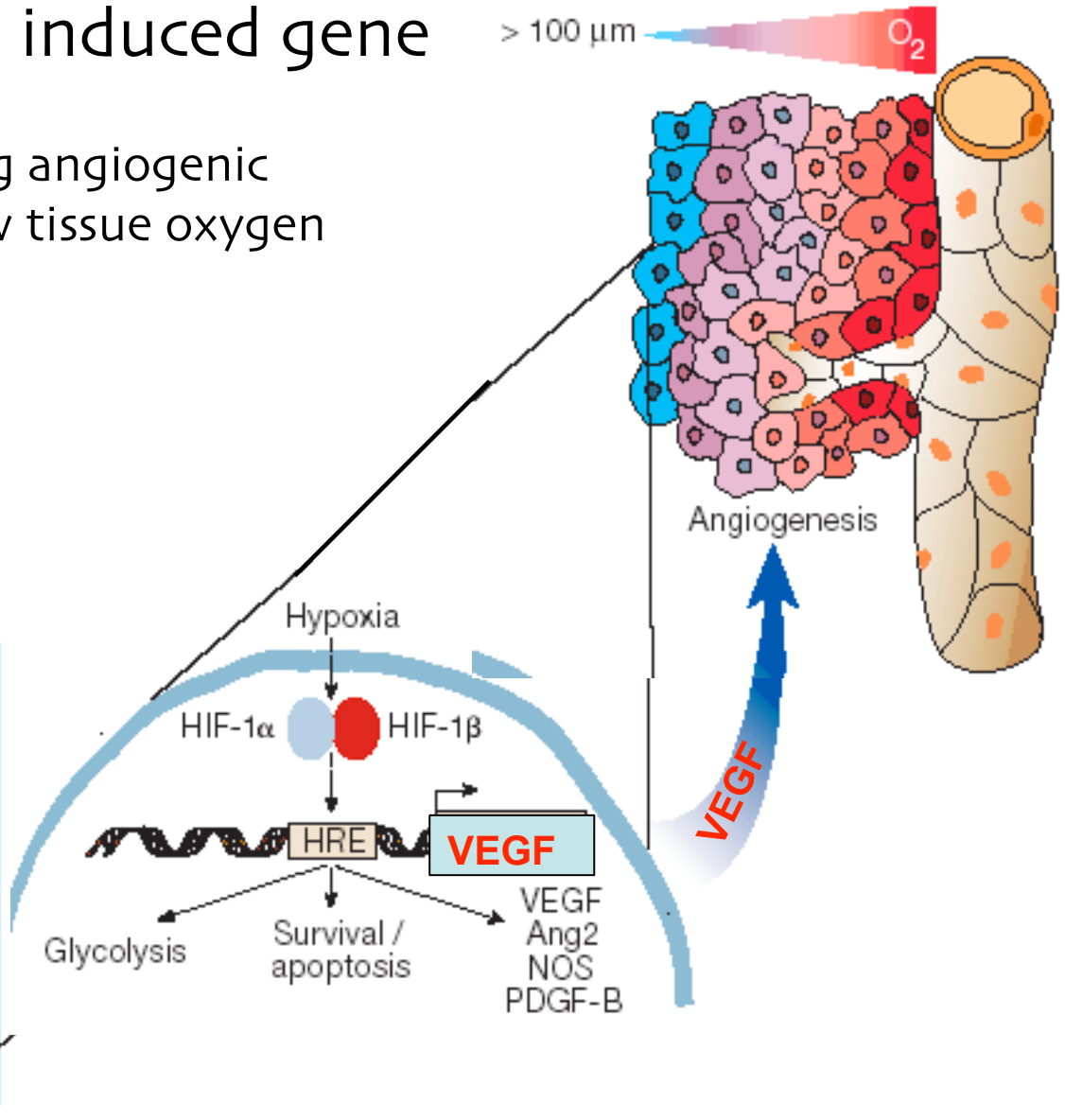
Survival

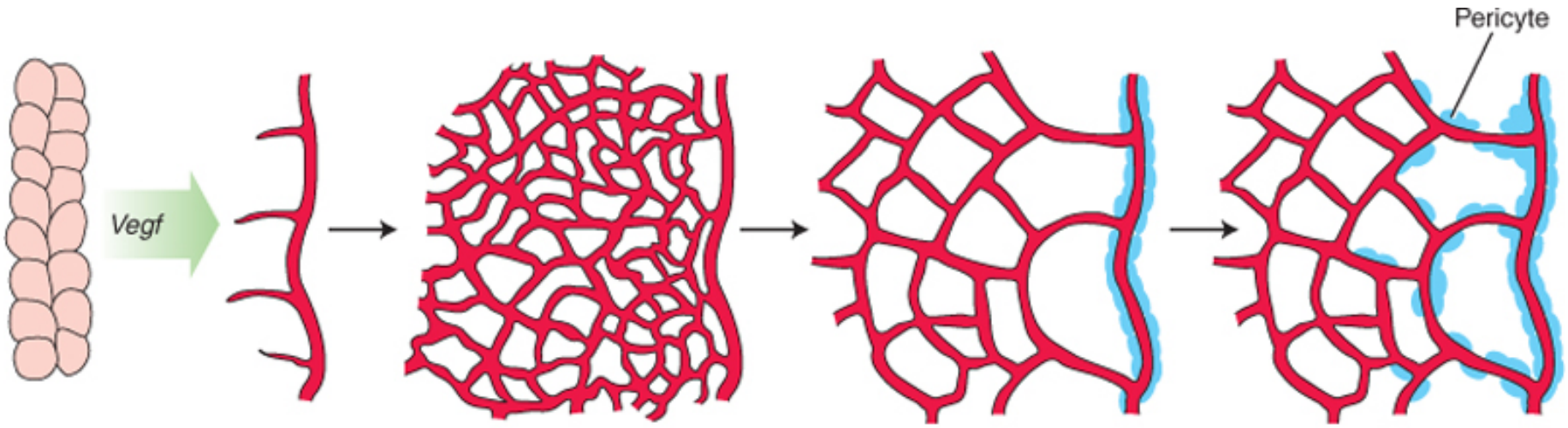


VEGF is a hypoxia induced gene

Mechanism for producing angiogenic factor in condtions of low tissue oxygen







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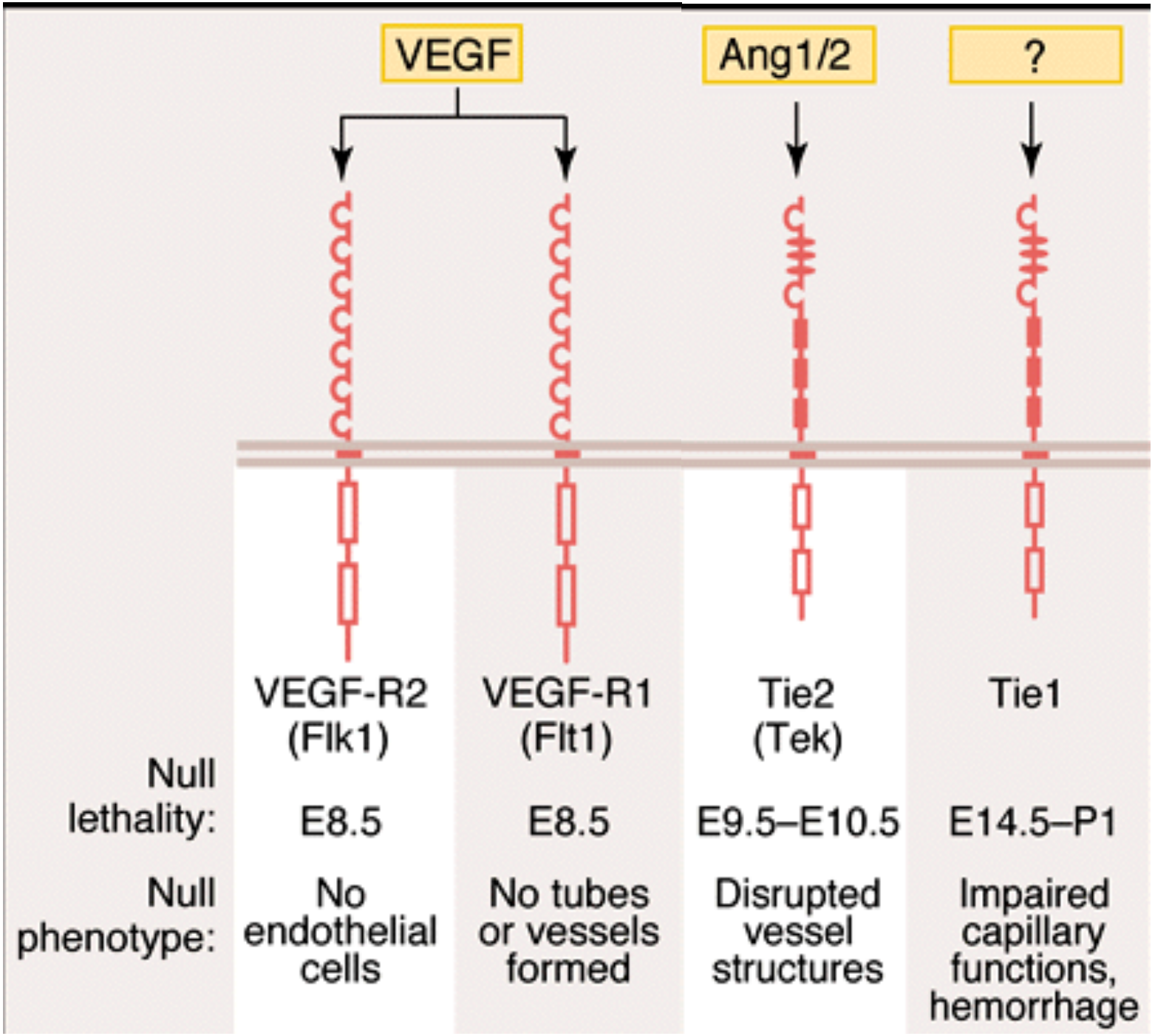
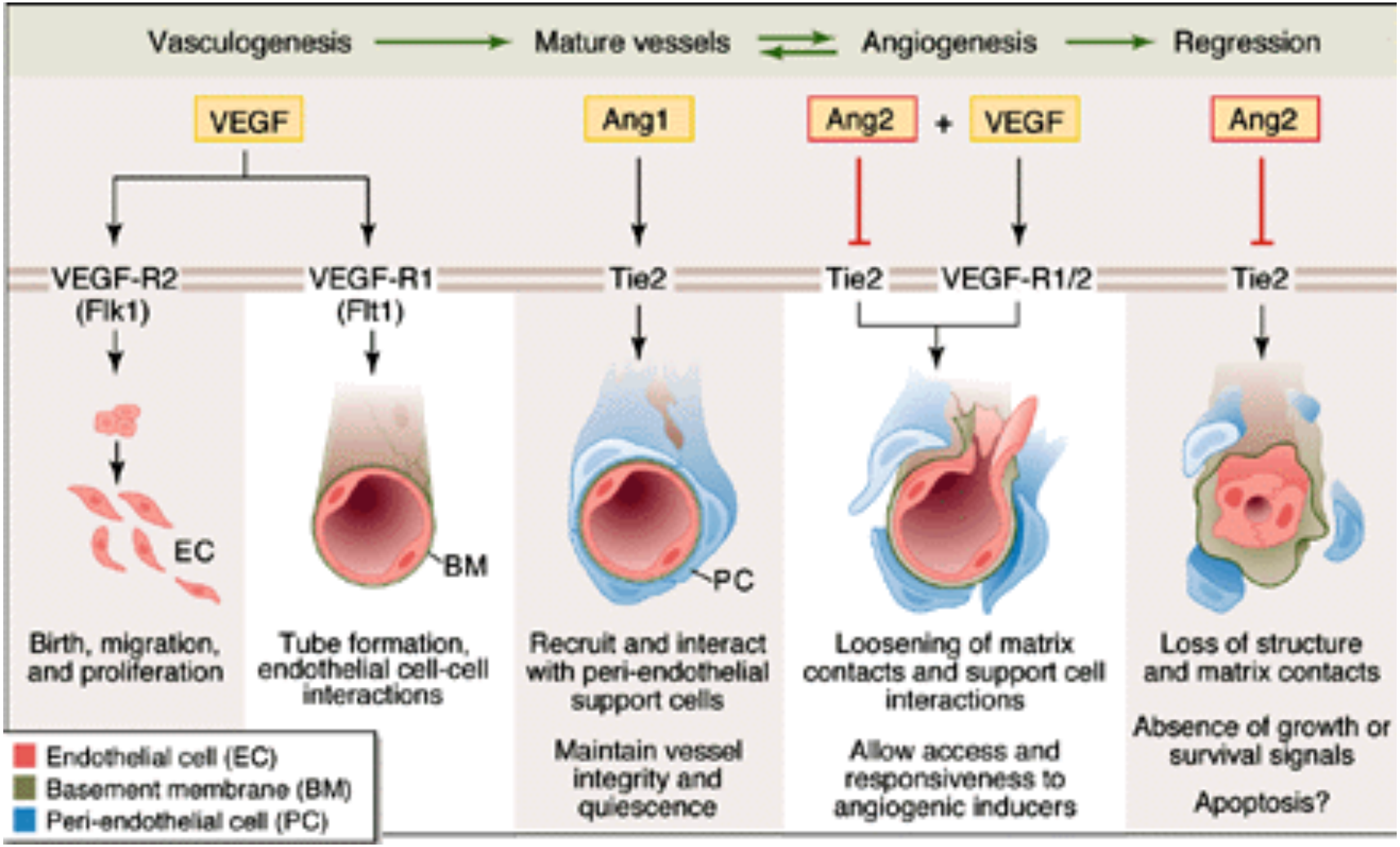

VEGF
Ang1/2
?
VEGF-R2 (Flk1)
VEGF-R1 (Flt1)
Tie2 (Tek)
Tie1
Null lethality:
E8.5
E8.5
E9.5–E10.5
E14.5–P1
Null phenotype:
No endothelial cells
No tubes or vessels formed
Disrupted vessel structures
Impaired capillary functions, hemorrhage


Vasculogenesis
Mature vessels
Angiogenesis
Regression
VEGF
Ang1
Ang2
+
VEGF
Ang2
VEGF-R2
(Flk1)
VEGF-R1
(Flt1)
Tie2
Tie2
VEGF-R1/2
Tie2
EC
BM
PC
Birth, migration, and proliferation
Tube formation, endothelial cell-cell interactions
Recruit and interact with peri-endothelial support cells
Maintain vessel integrity and quiescence
Loosening of matrix contacts and support cell interactions
Allow access and responsiveness to angiogenic inducers
Loss of structure and matrix contacts
Absence of growth or survival signals
Apoptosis?
Endothelial cell (EC)
Basement membrane (BM)
Peri-endothelial cell (PC)

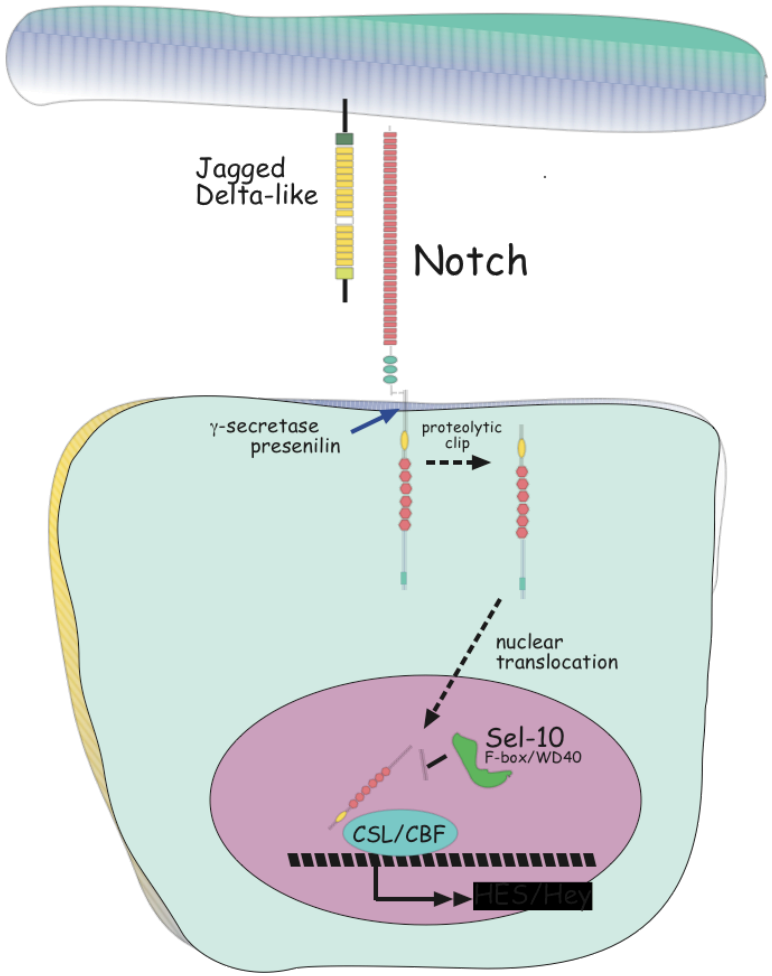
Notch drives cell fate determination

Notch/Notch ligand interaction:

-mechanism for setting and maintaining state of differentiation

-fates locked in via lateral inhibition

-



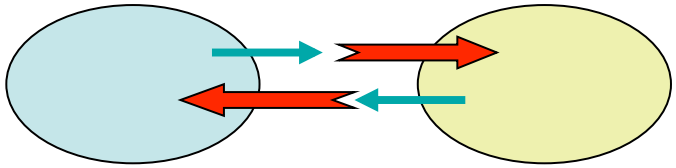
Notch drives cell fate differentiation

Notch/Notch ligand interaction:

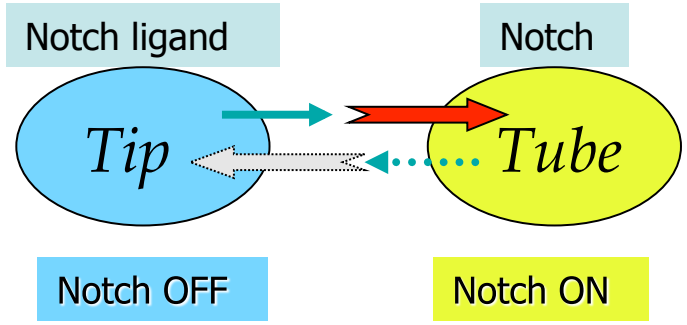
-mechanism for driving state of differentiation

-fates locked in via lateral inhibition

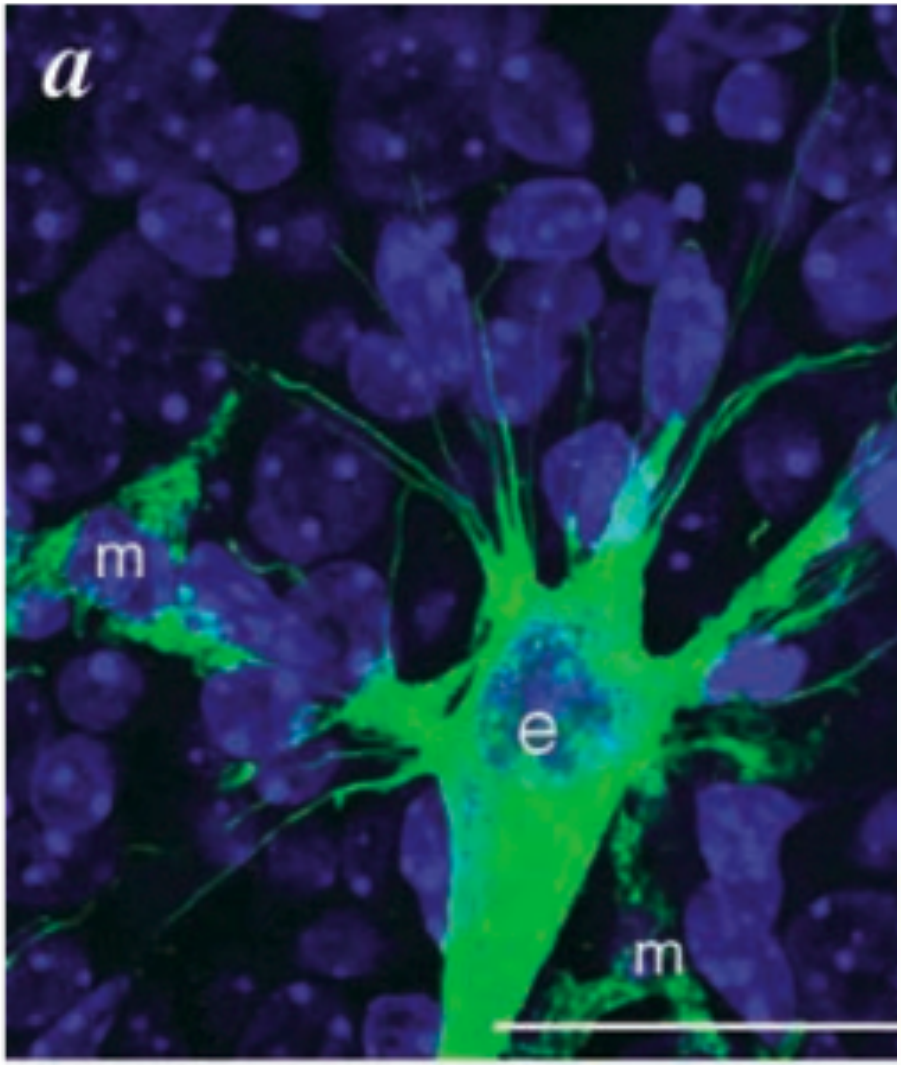
Notch drives cell fate differentiation

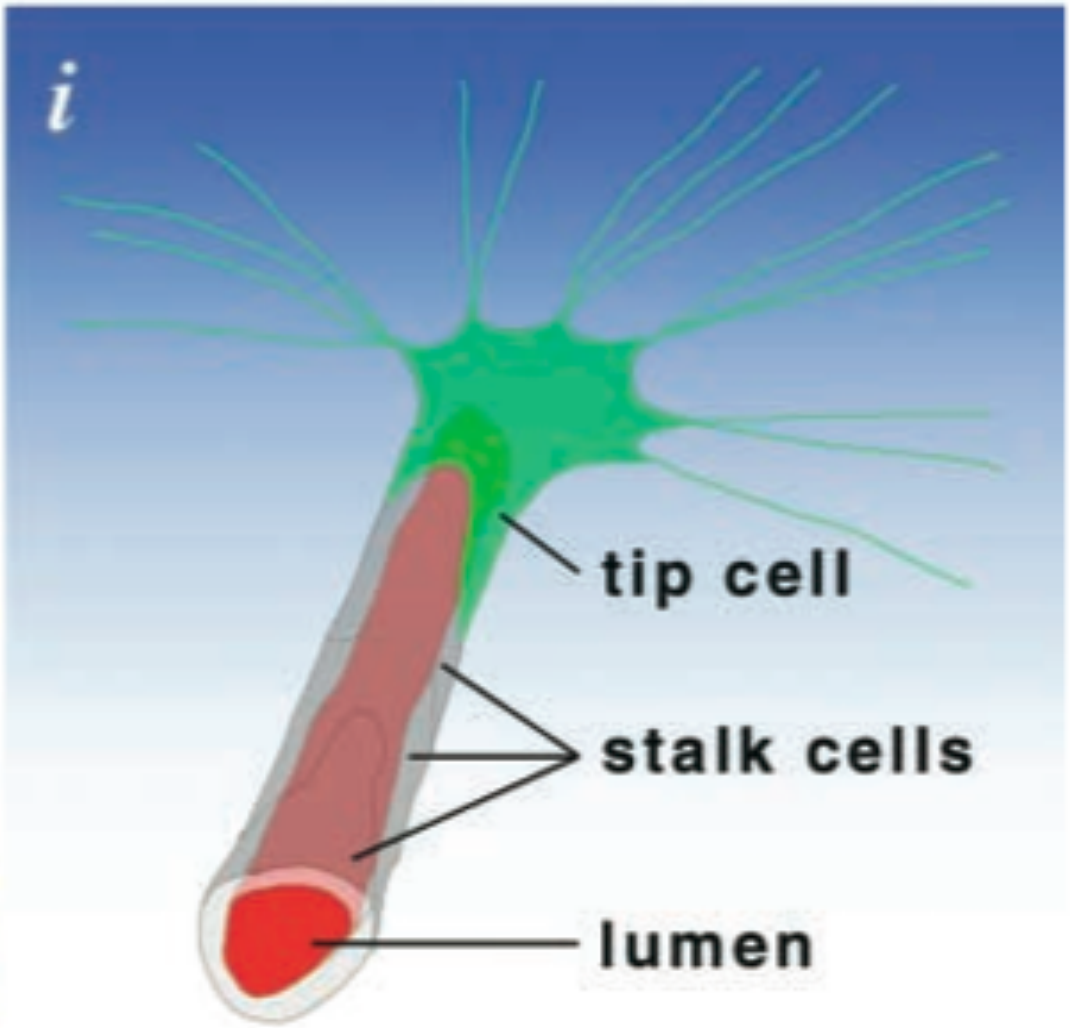


Notch drives cell fate differentiation

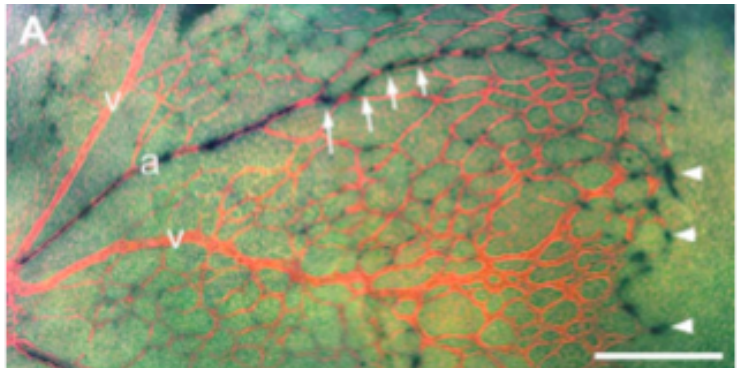


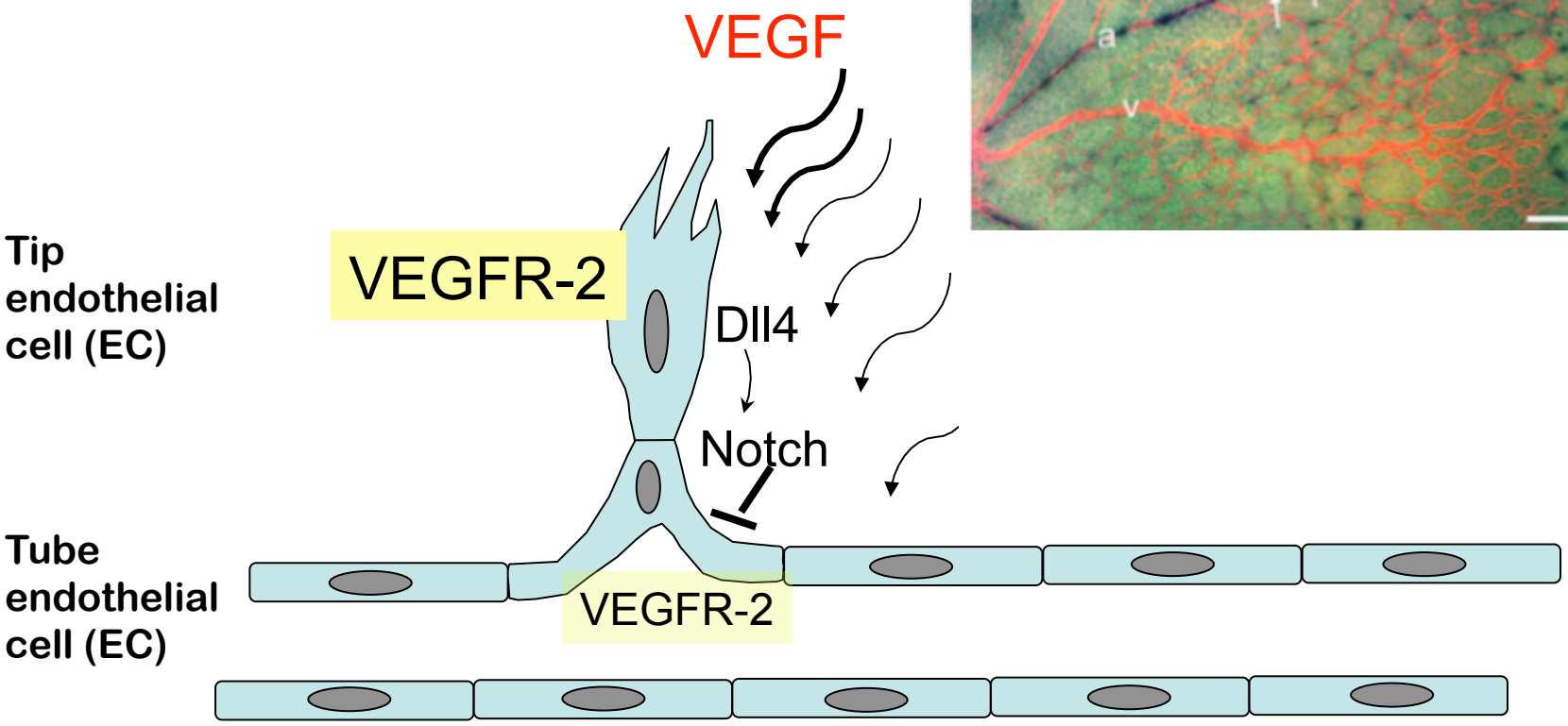
Notch restricts tip cell specification

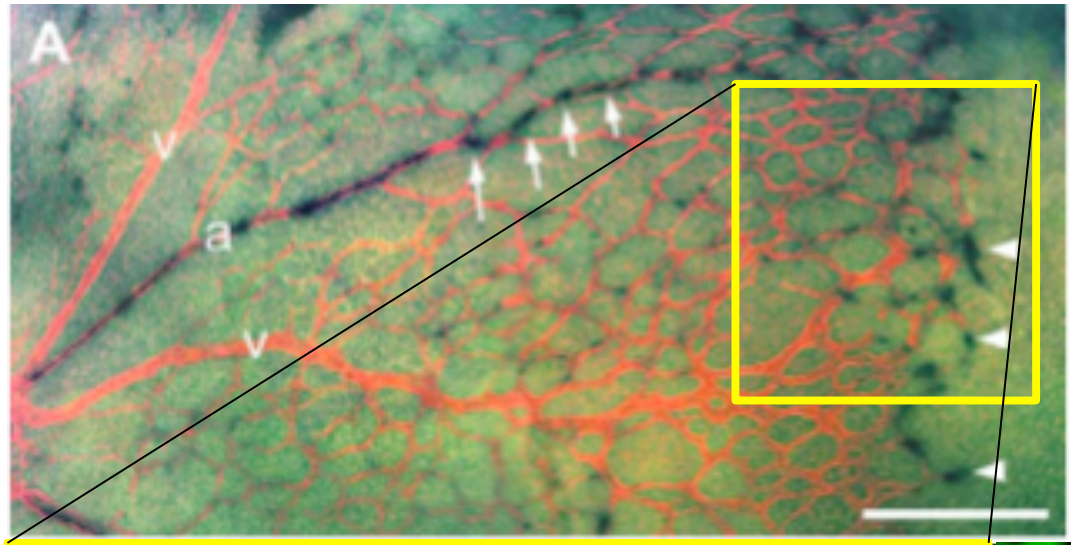

a
m
e
m

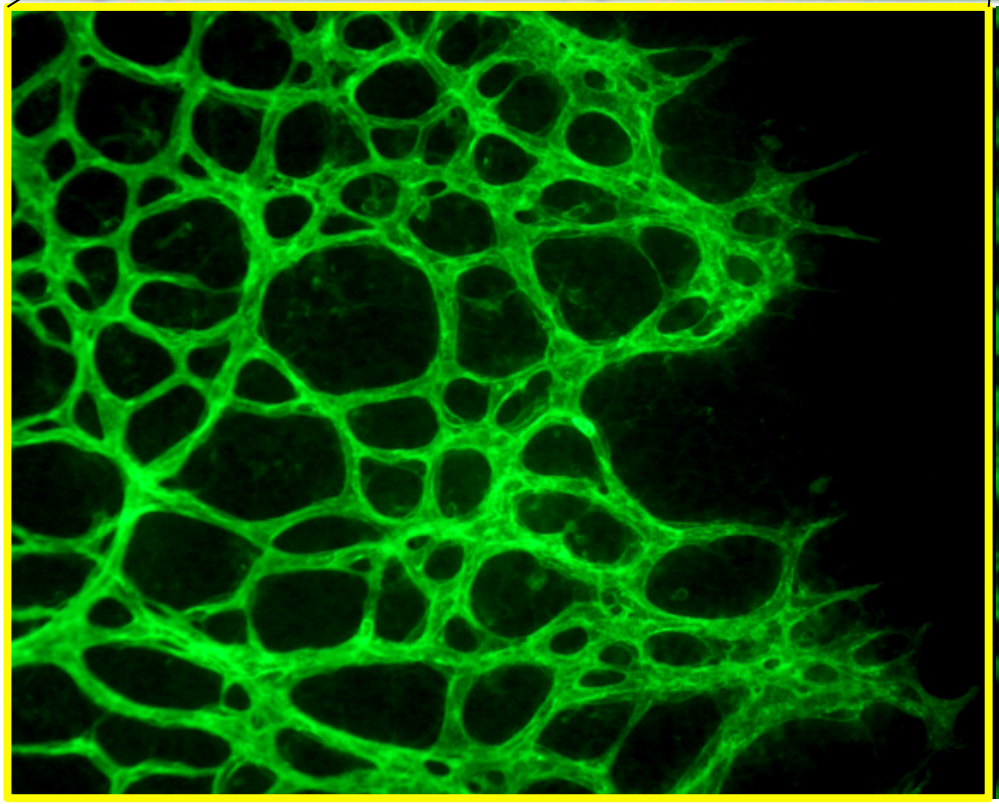

i
tip cell
stalk cells
lumen

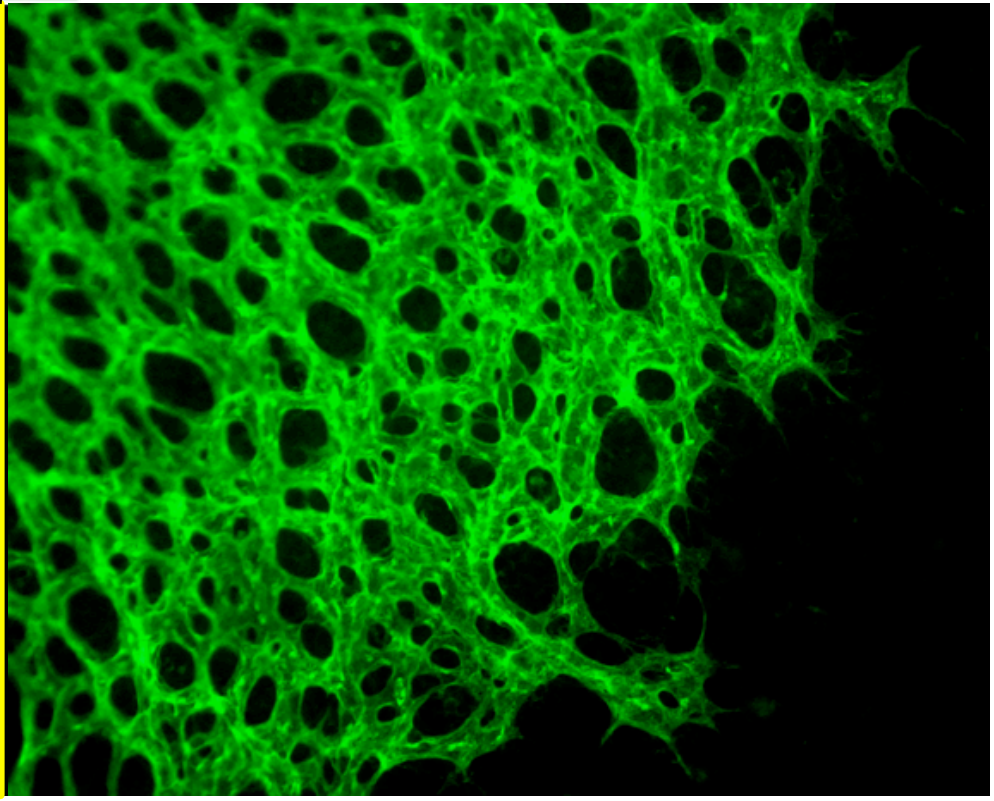
Notch restricts endothelial sprouting through feedback to VEGF signaling





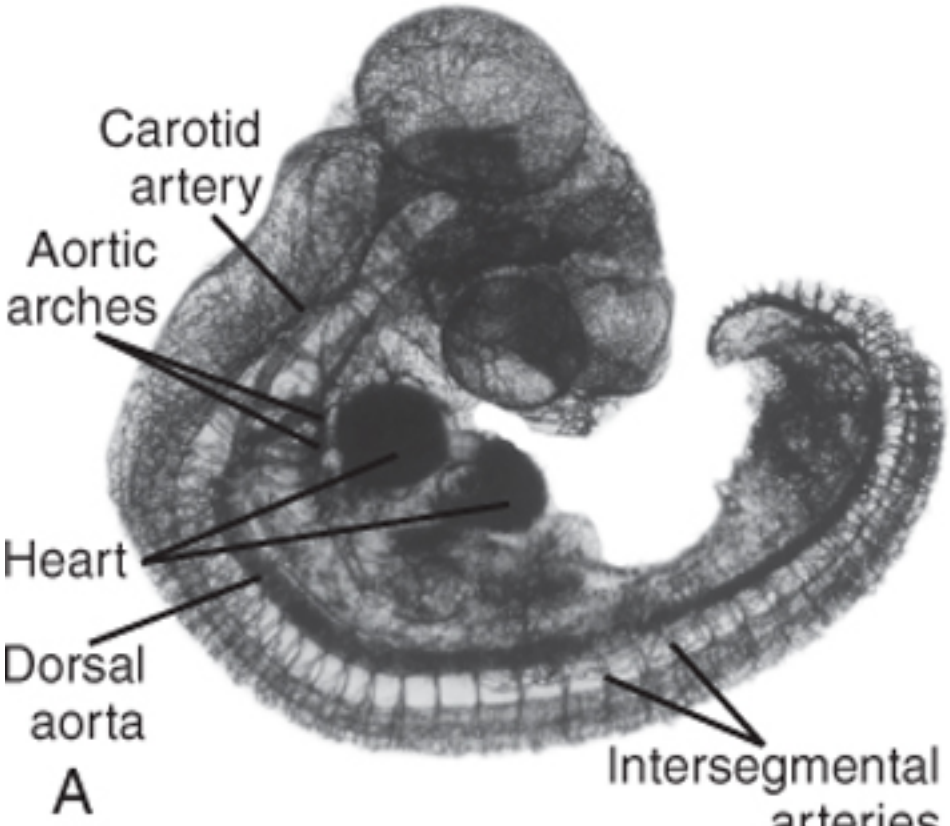


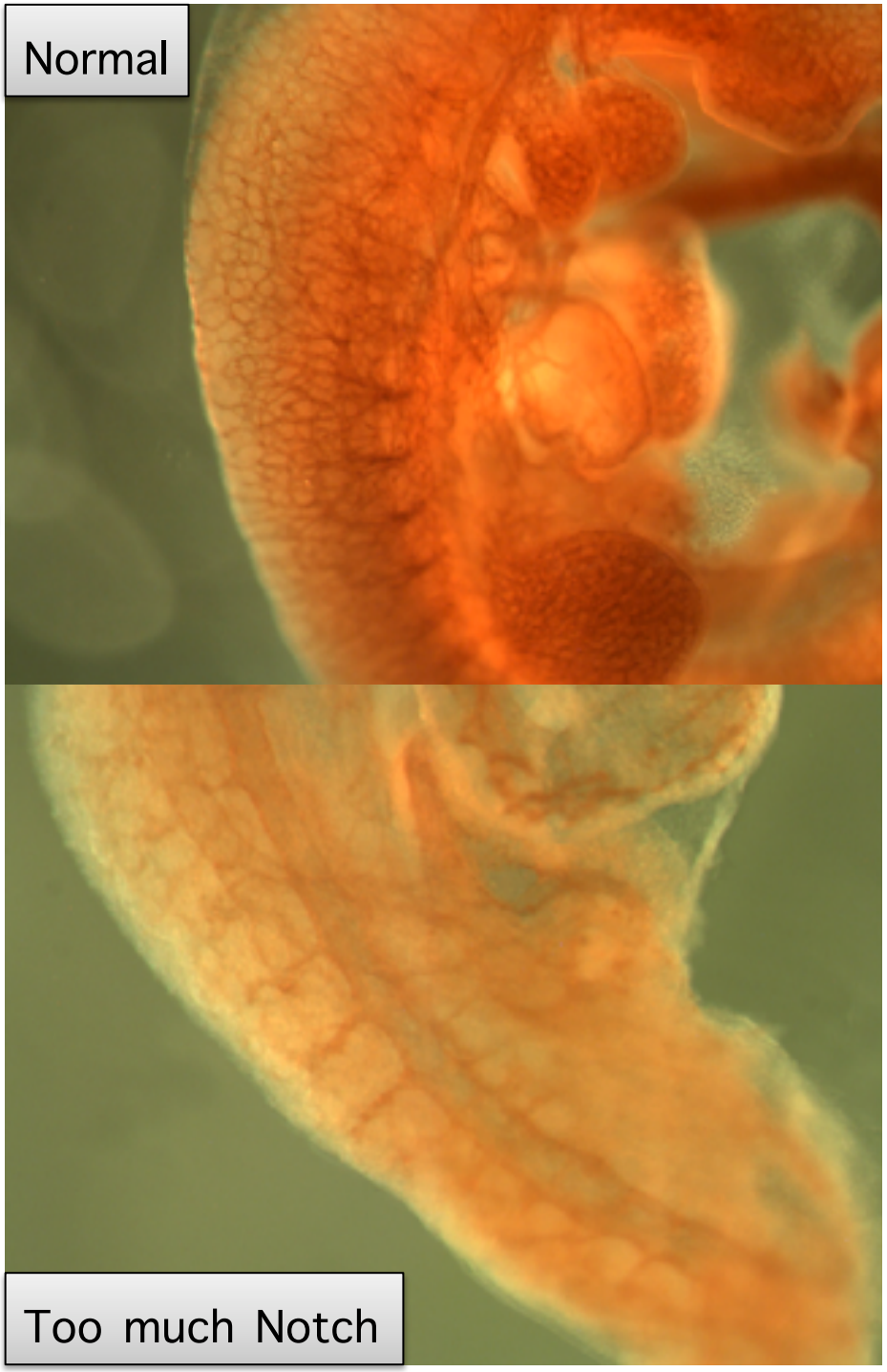




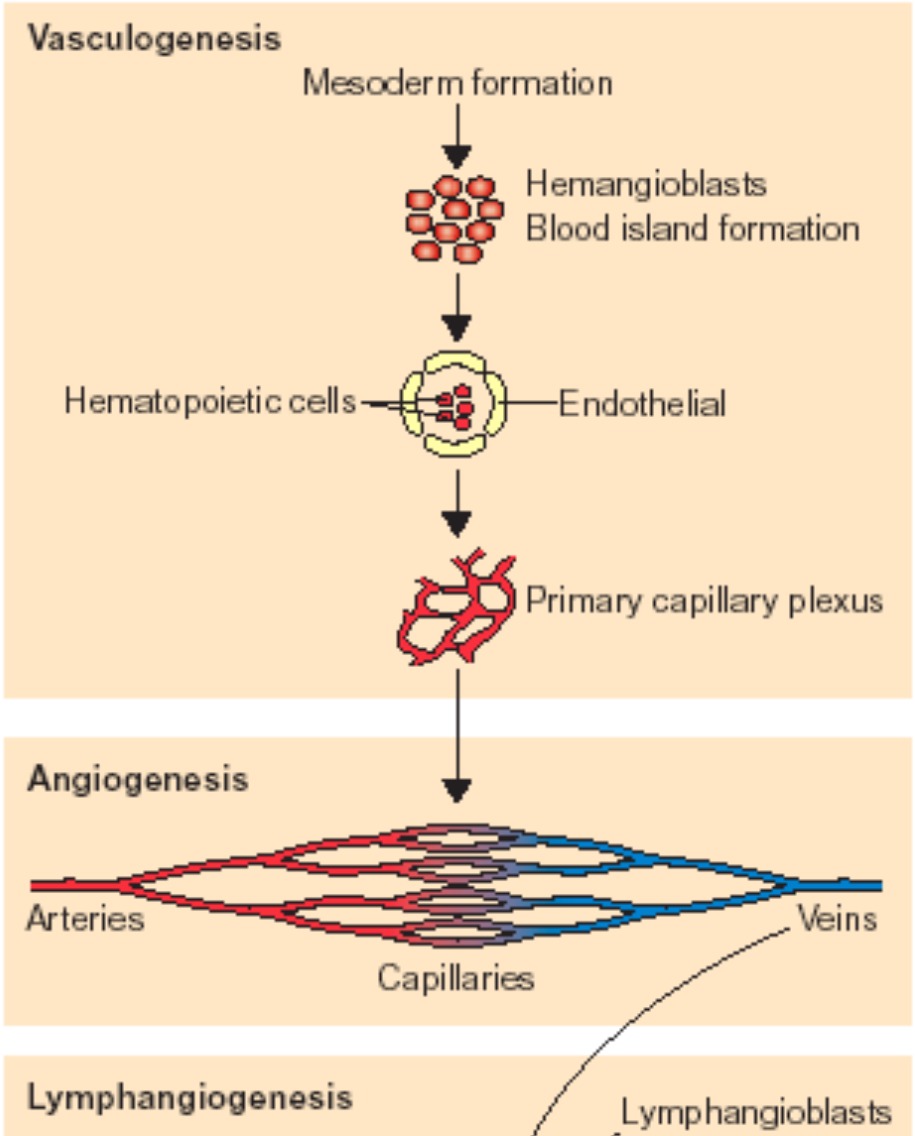
Wild-type

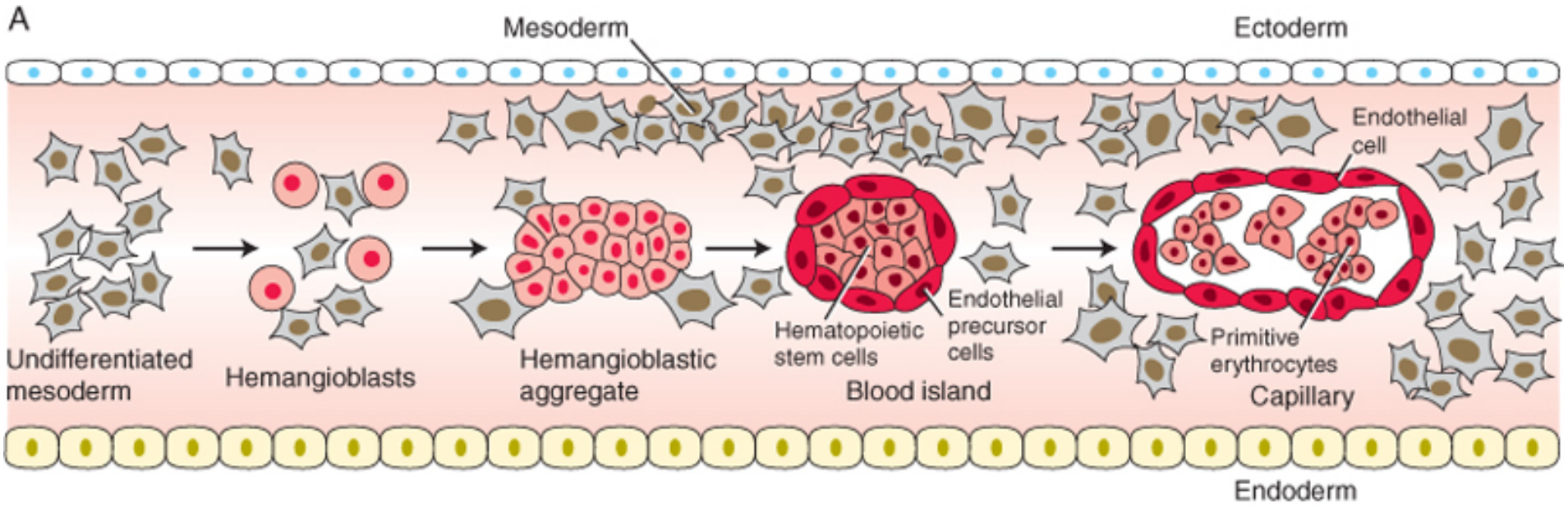
Notch Mutant

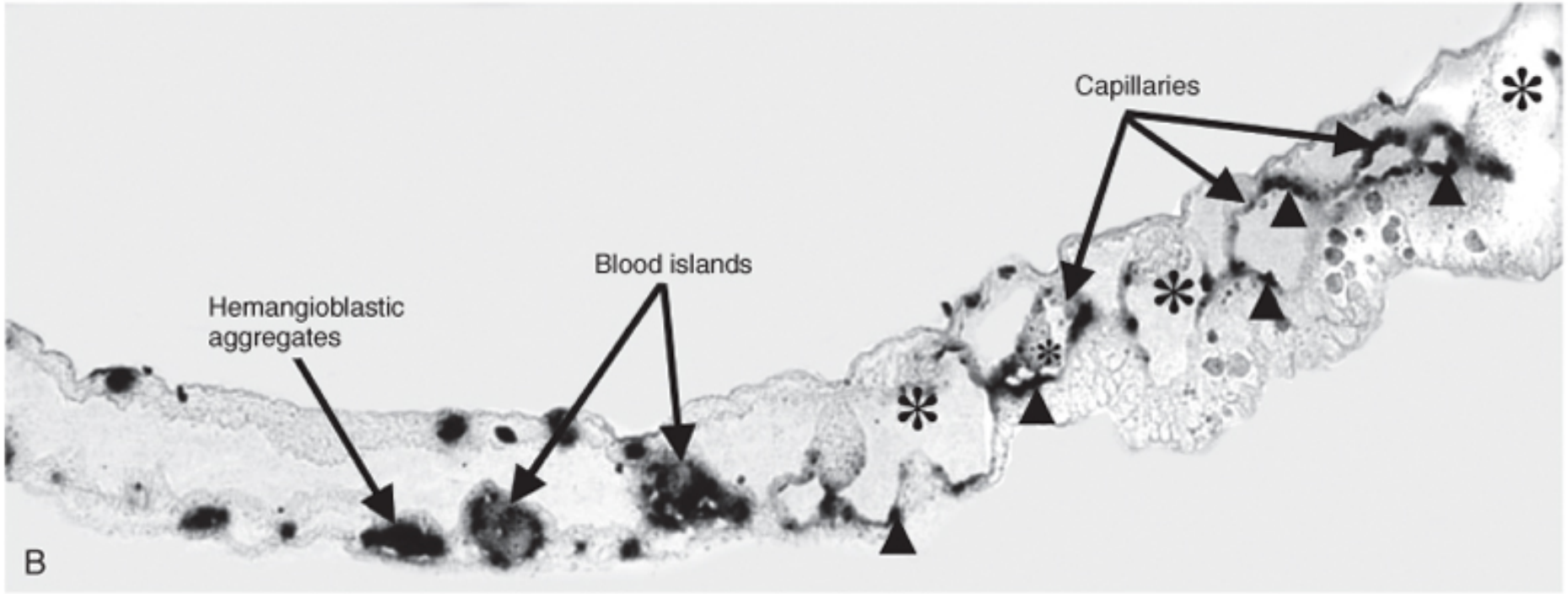

Carotid artery
Aortic arches
Heart
Dorsal aorta
Intersegmental arteries
A


Normal
Too much Notch

Endothelial Specification





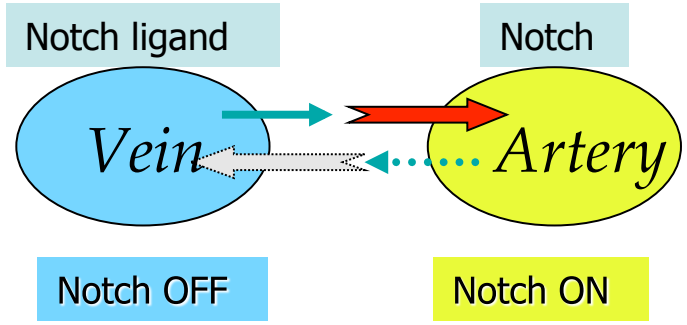


Vascular Specification

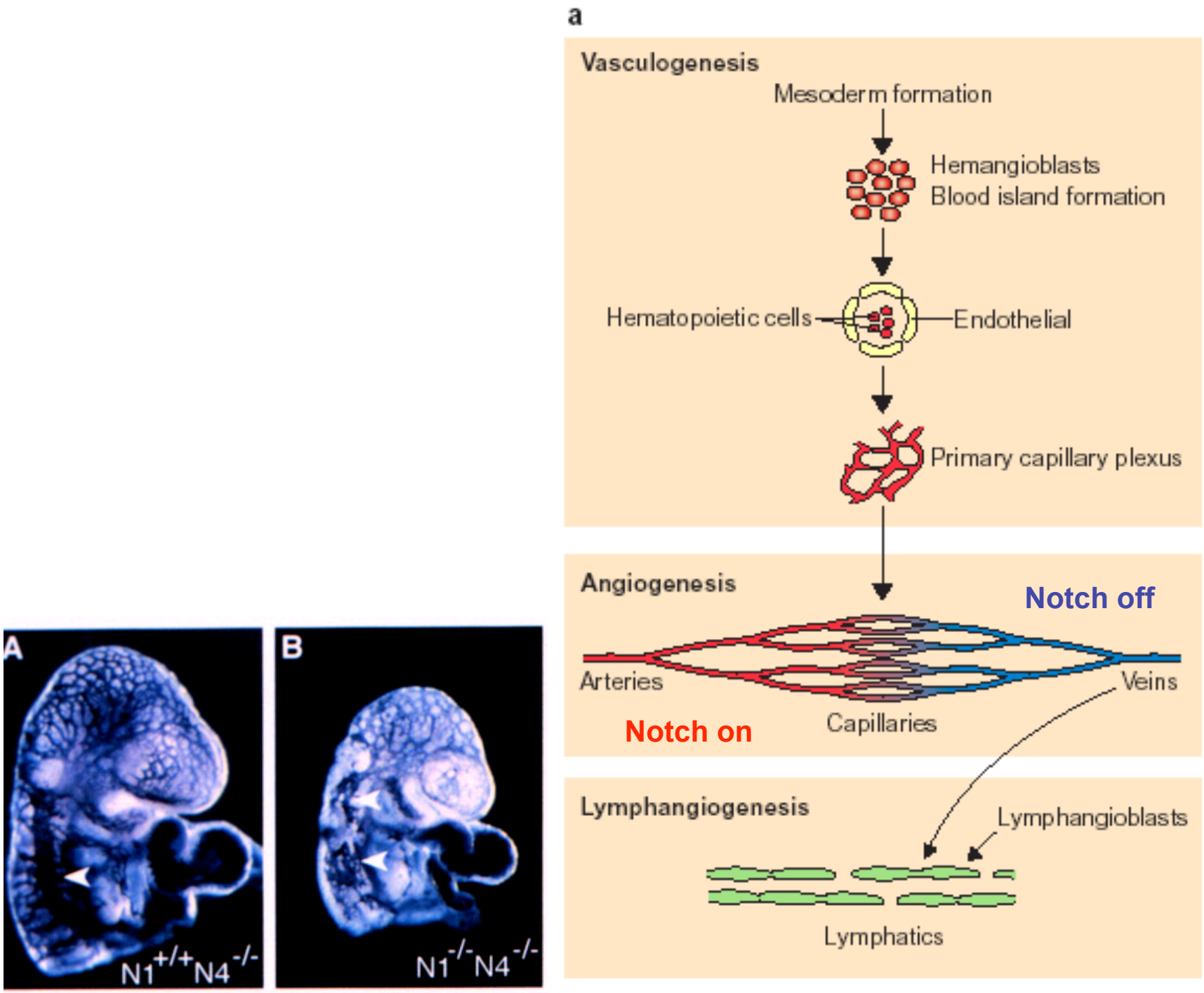
Vessels specialization during development

1) Arterial-Venous Vessels
2) Lymphatic Vessels
3) Tissue specific vessels

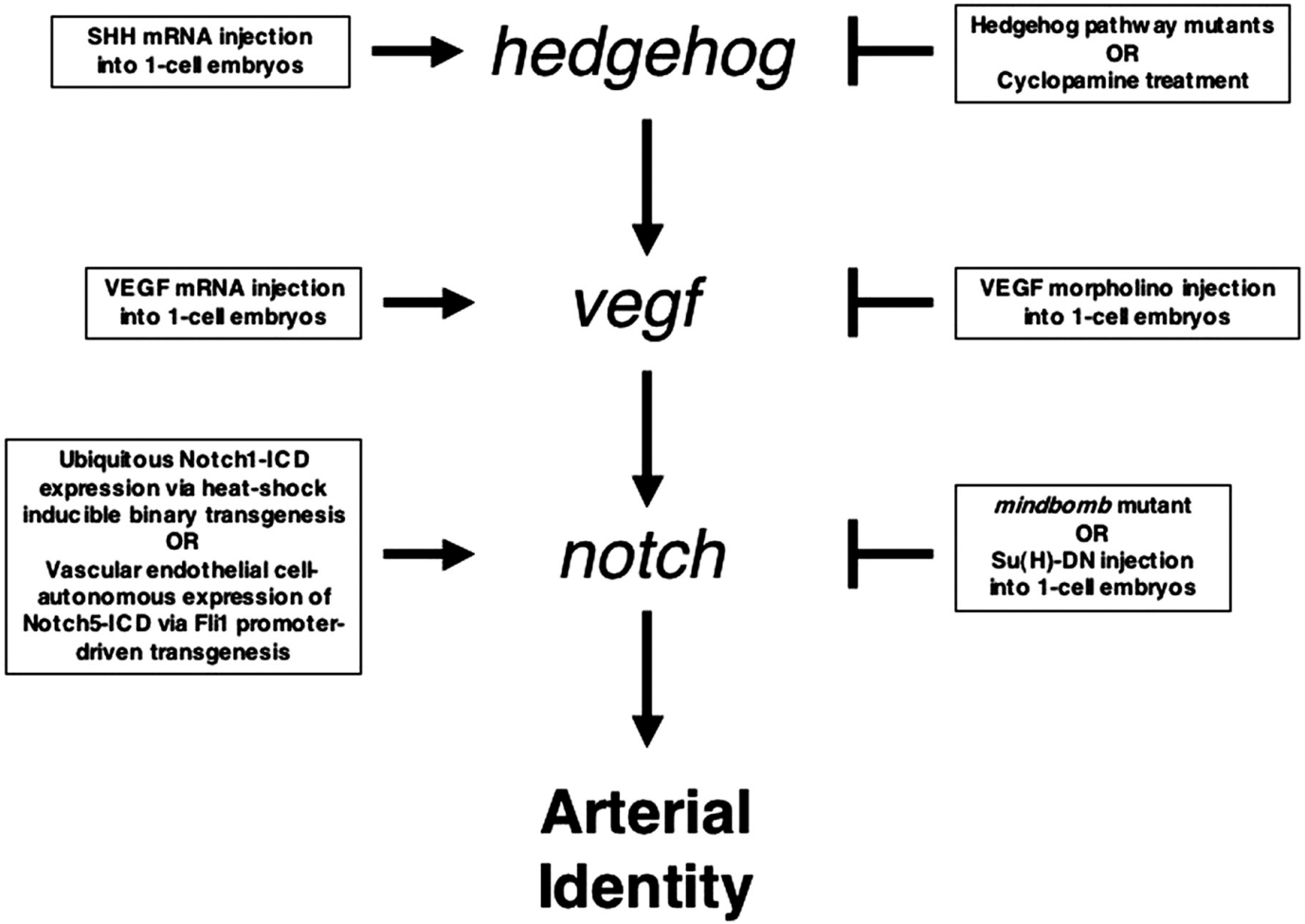
Notch drives cell fate differentiation



Notch promotes arterial specification

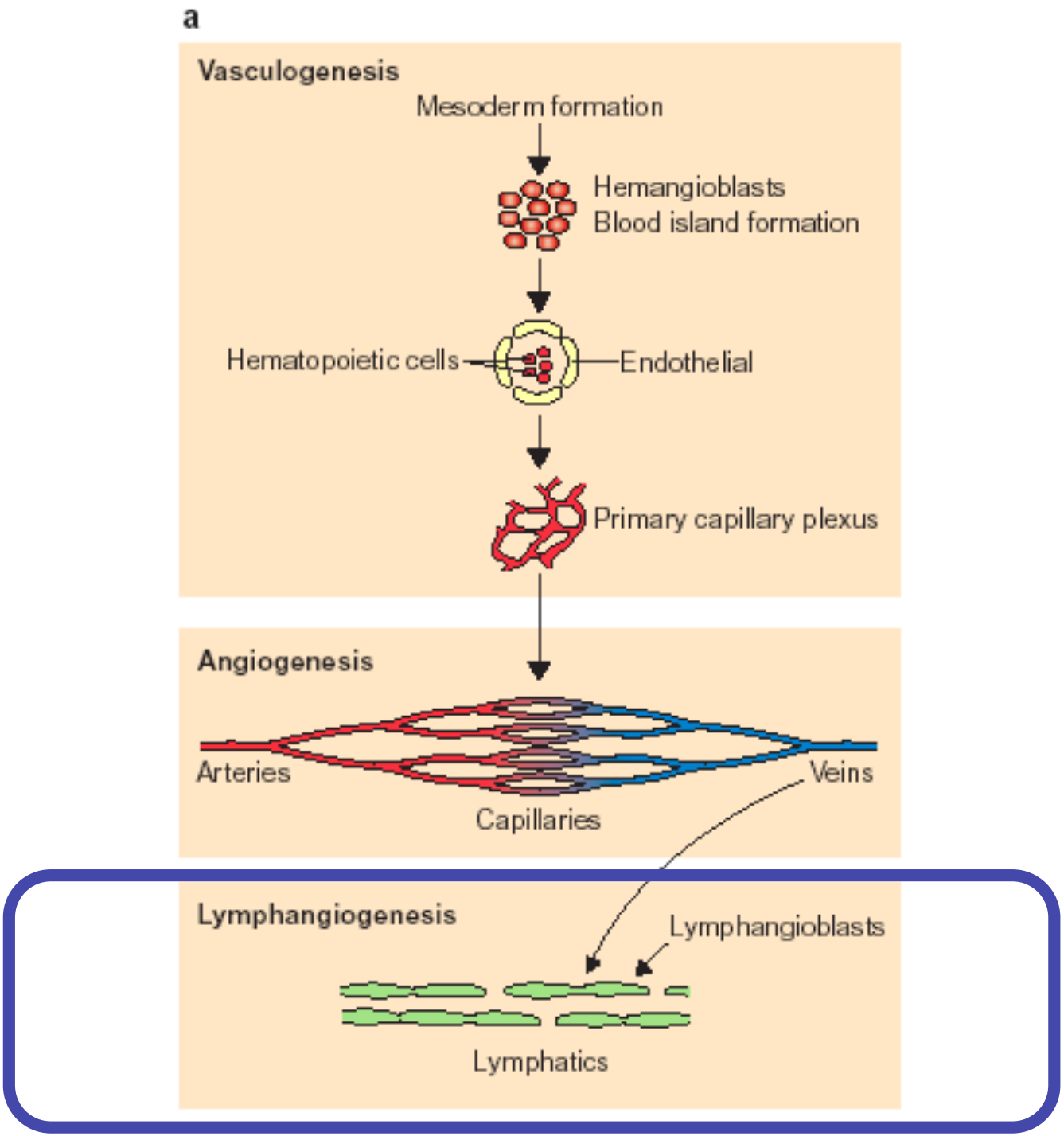

a
Vasculogenesis
Mesoderm formation
Hemangioblasts
Blood island formation
Hematopoietic cells
Endothelial
Primary capillary plexus
Angiogenesis
Notch off
Arteries
Veins
Capillaries
Notch on
Lymphangiogenesis
Lymphangioblasts
Lymphatics
A
N1+/+N4-/-
B
N1-/-N4-/-
D
E

A molecular pathway for arterial-venous fate determination

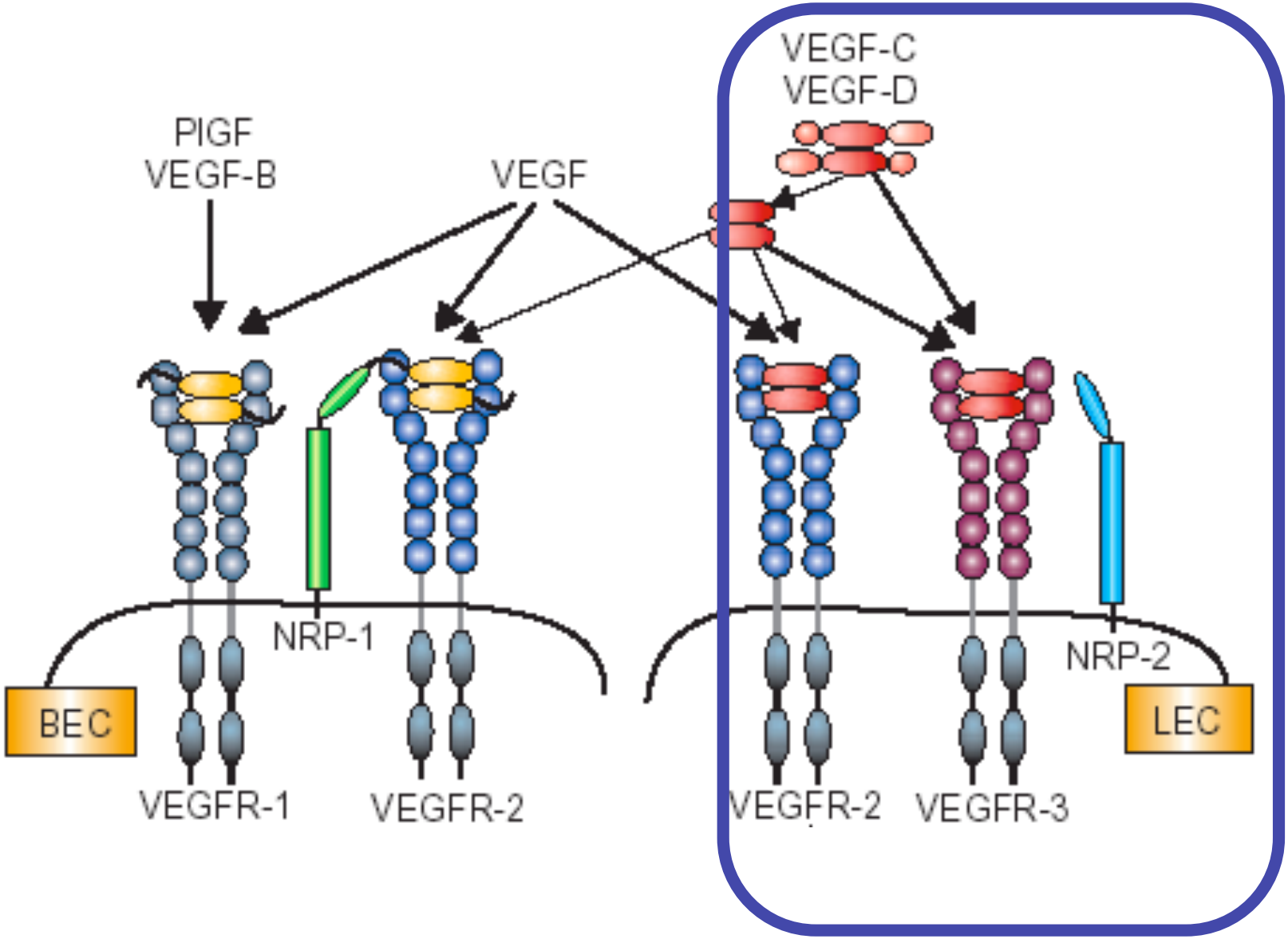


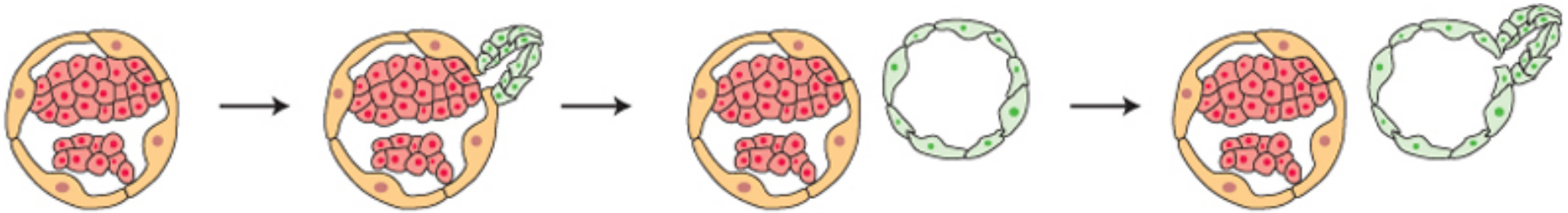
Swift, M. R. et al. Circ Res 2009;104:576-588

Circulation Research

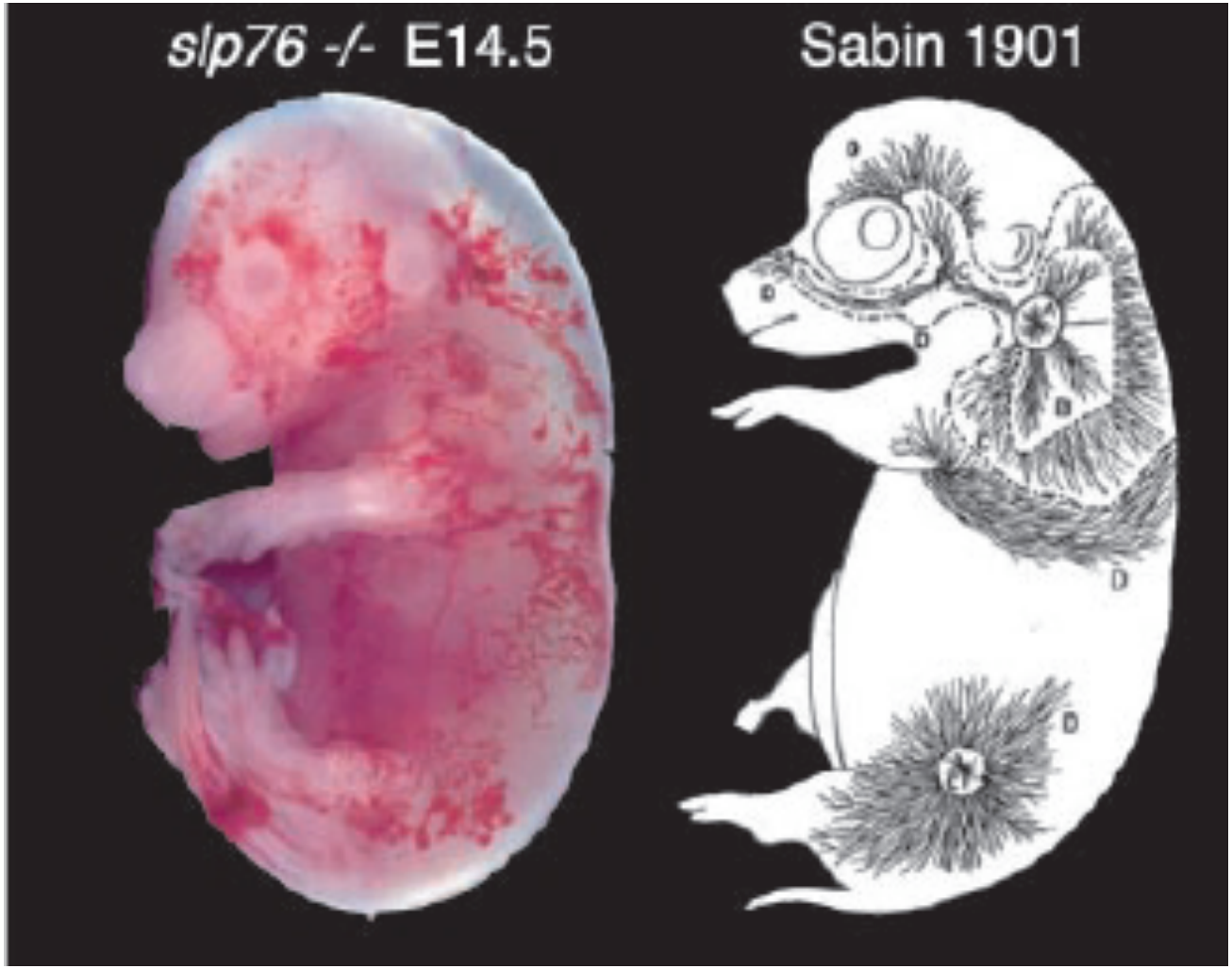

a
Vasculogenesis
Mesoderm formation
Hemangioblasts
Blood island formation
Hematopoietic cells
Endothelial
Primary capillary plexus
Angiogenesis
Arteries
Veins
Capillaries
Lymphangiogenesis
Lymphangioblasts
Lymphatics

VEGF and VEGF Receptors



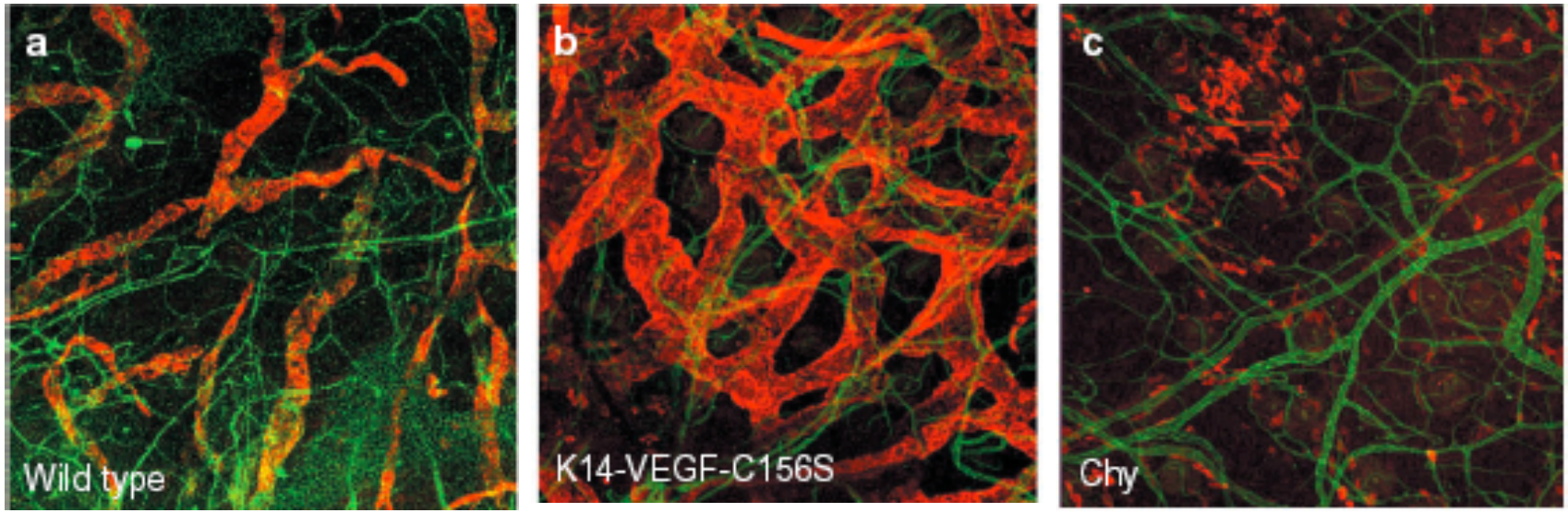


Schoenwolf et al: Larsen's Human Embryology, 4th Edition.


slp76 -/- E14.5
Sabin 1901

Structure of blood and lymphatic vasculature in dermis

normal + lymphatics - lymphatics

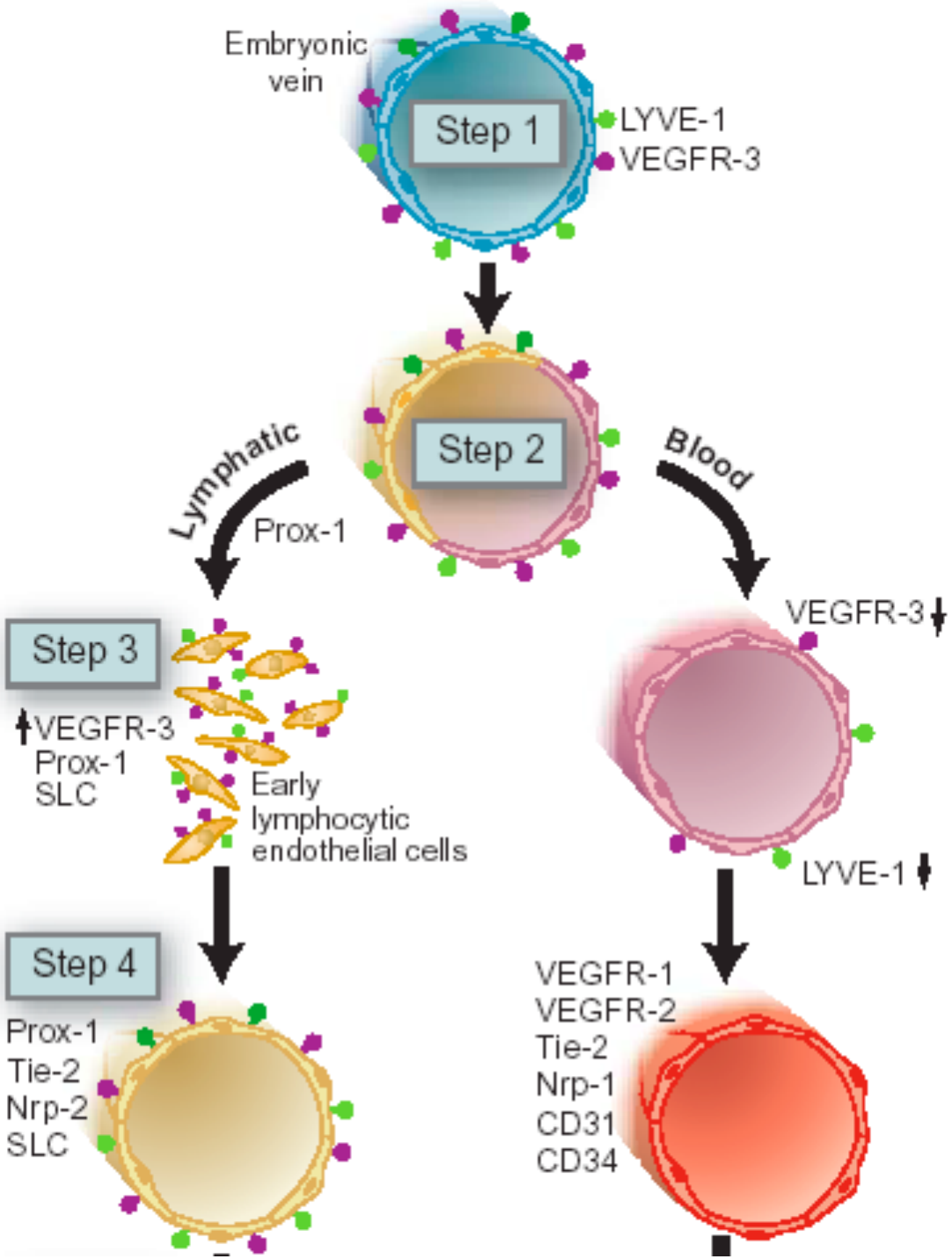


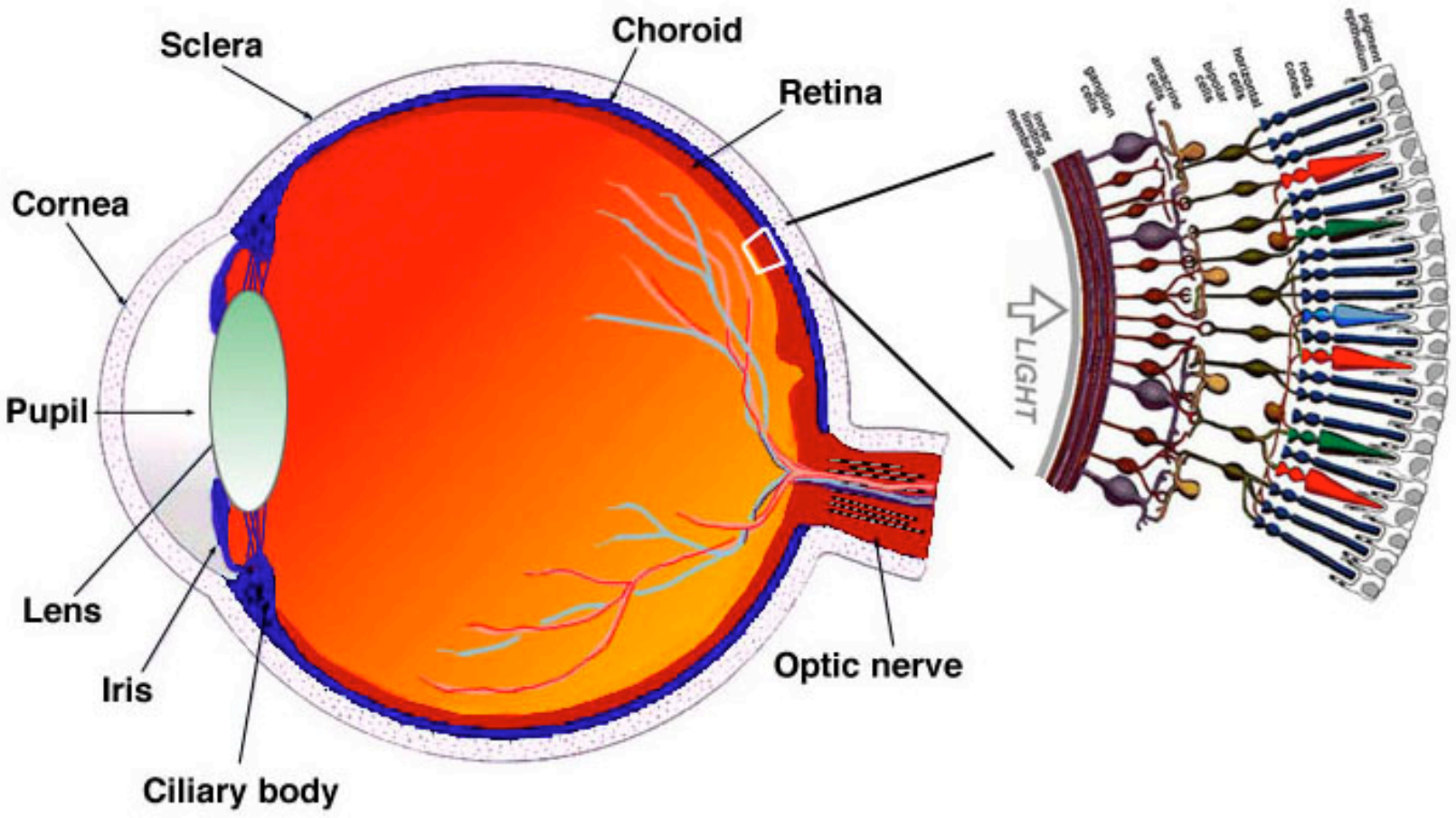
Mouse with mutation in VEGFR-3

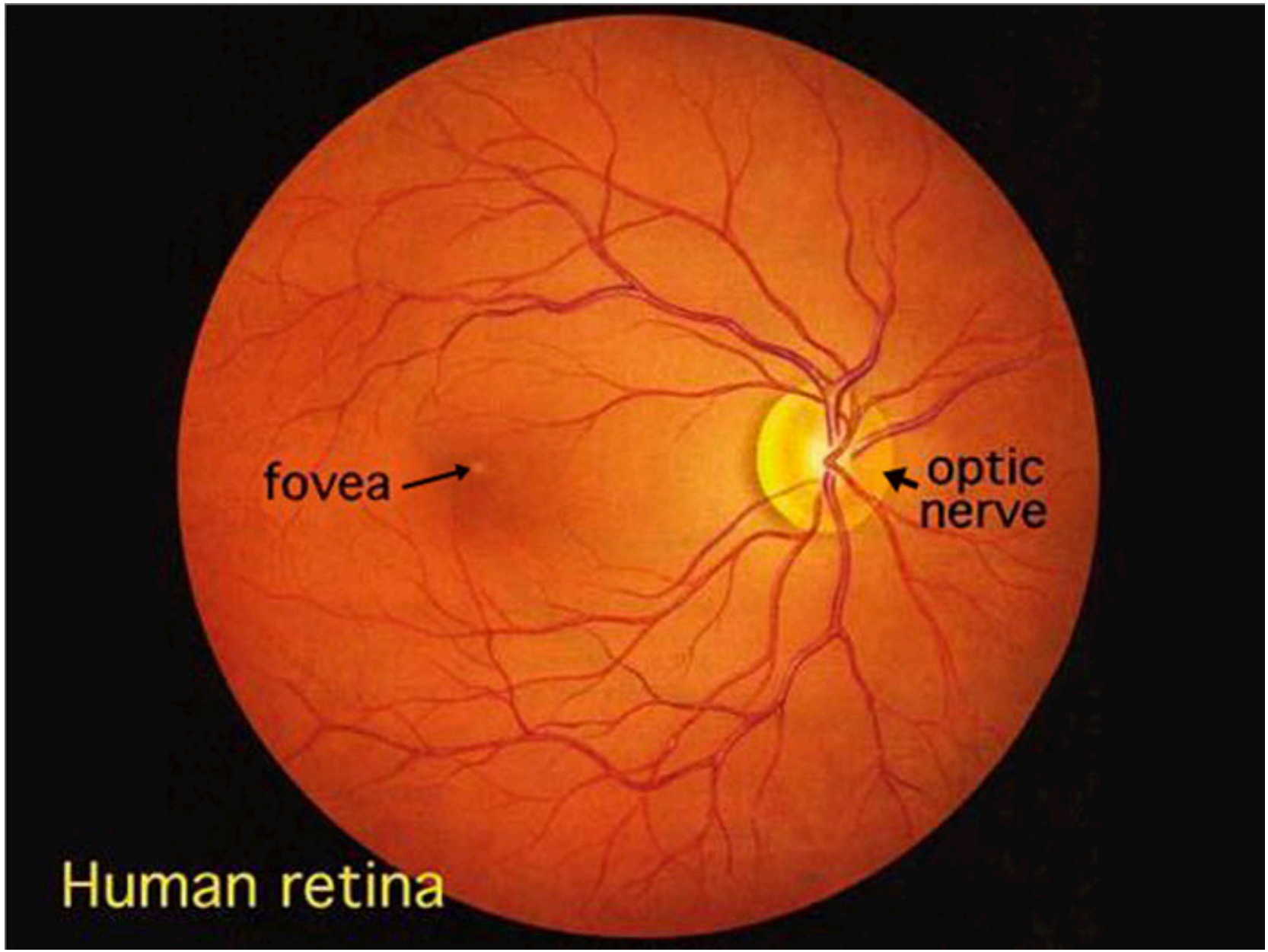
blood vessel

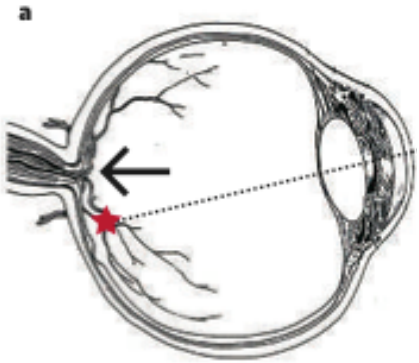
lymphatic vessel

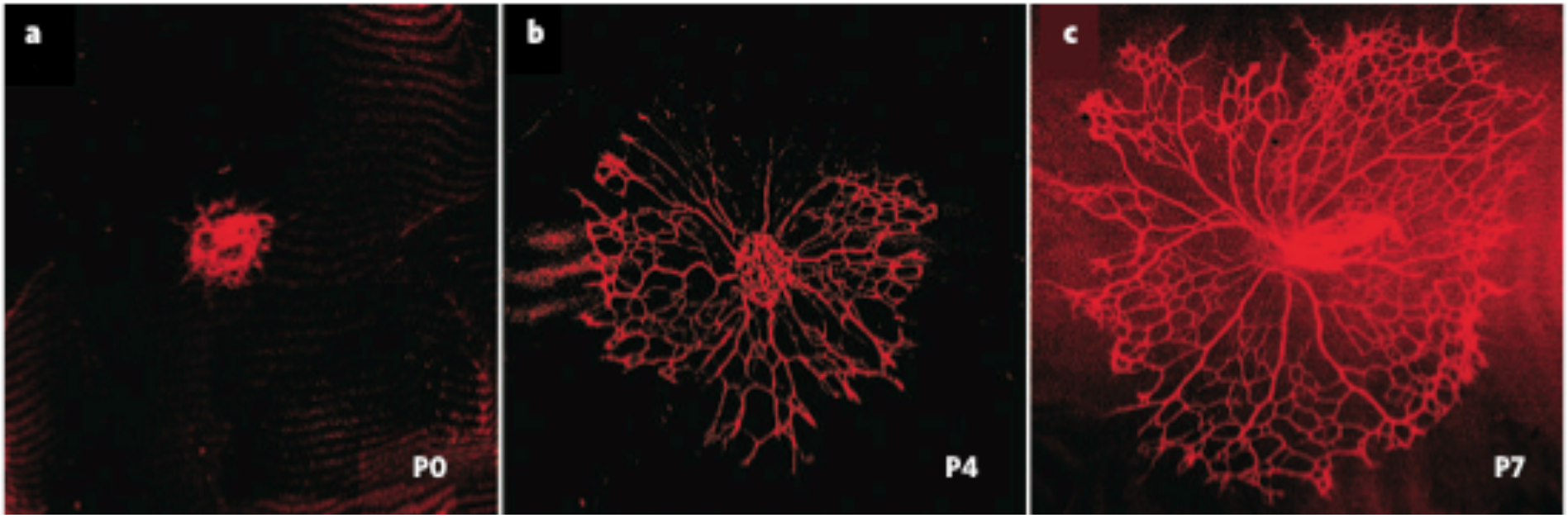
Figure 2 **Blood and lymphatic vessels in mouse ear.** a, **Whole mount immunohistochemical staining of lymphatic vessels (LYVE-1; red) and blood vessels (PECAM-1; green) in mouse ear.** b, **Transgenic overexpression of the VEGF-C mutant C156S in the skin leads to enlargement of the lymphatic vessels, whereas only a few lymphatic ECs are present in the ear of a Chy lymphoedema mouse[12]** (c). **We thank G. Thurston for the stainings and D. Jackson for the LYVE-1 antibodies.**


Embryonic vein
Step 1
LYVE-1
VEGFR-3
Step 2
Lymphatic
Prox-1
Blood
Step 3
VEGFR-3
Prox-1
SLC
Early lymphocytic endothelial cells
VEGFR-3
LYVE-1
Step 4
Prox-1
Tie-2
Nrp-2
SLC
VEGFR-1
VEGFR-2
Tie-2
Nrp-1
CD31
CD34


Sclera
Choroid
Retina
Cornea
Pupil
Lens
Iris
Ciliary body
Optic nerve
LIGHT


fovea
optic nerve
Human retina


a


a
P0
b
P4
c
P7

Normal intraretinal vasculature

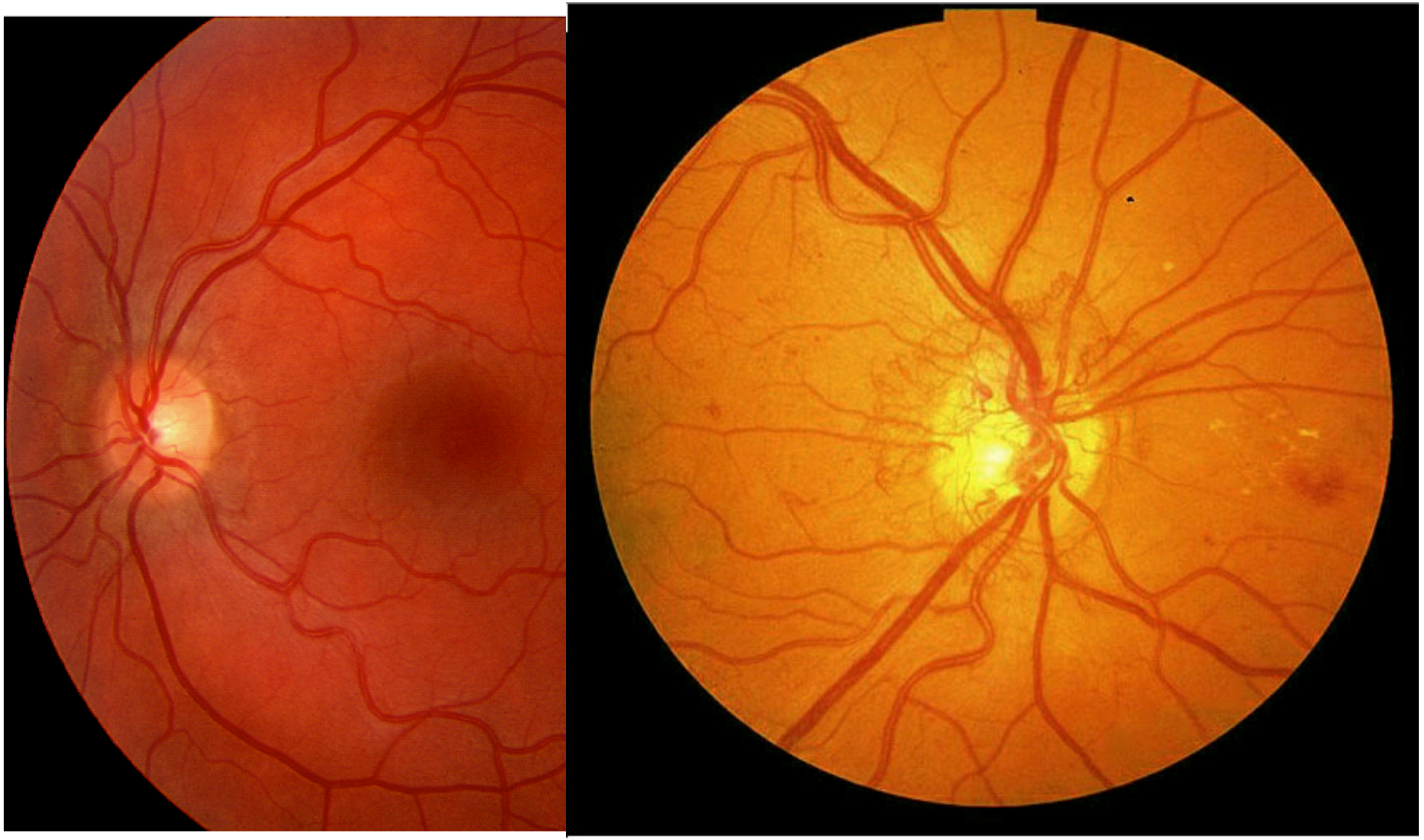
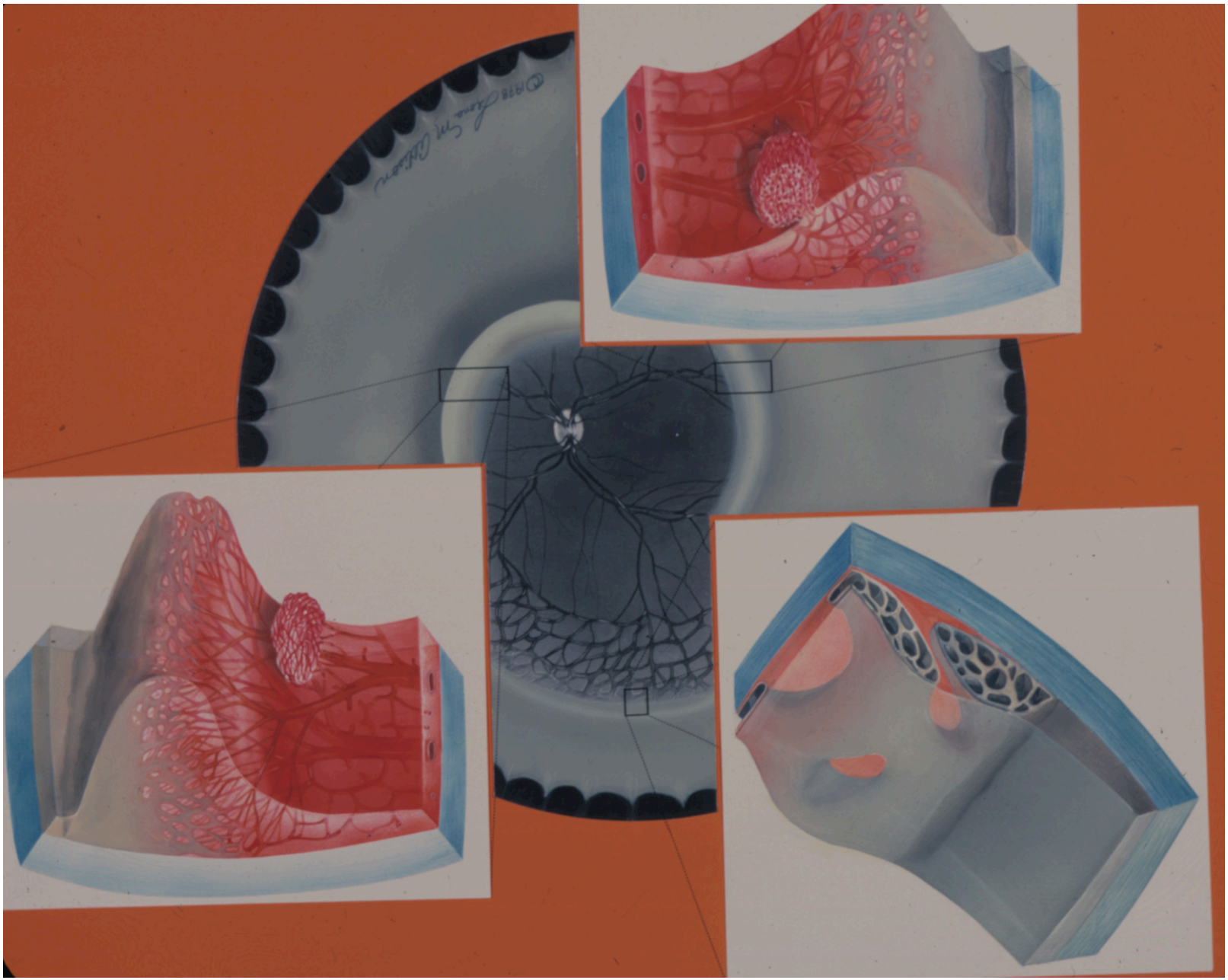


Fig. 25. A view of the fundus of the eye and of the retina in a patient who has advanced diabetic retinopathy.

Retinopathy of Prematurity



Retinopathy of Prematurity

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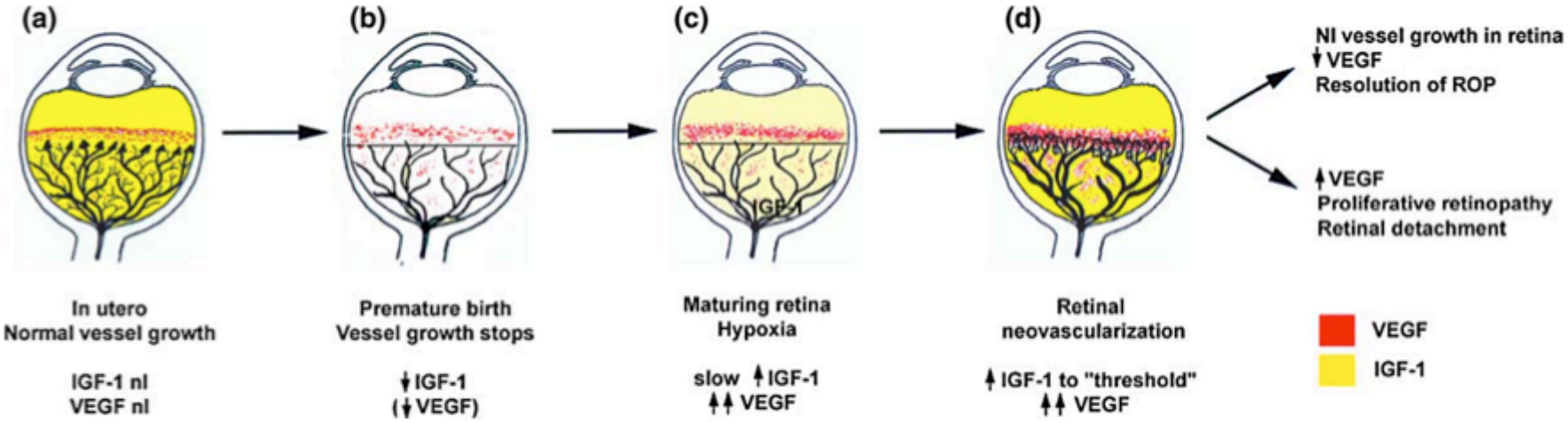
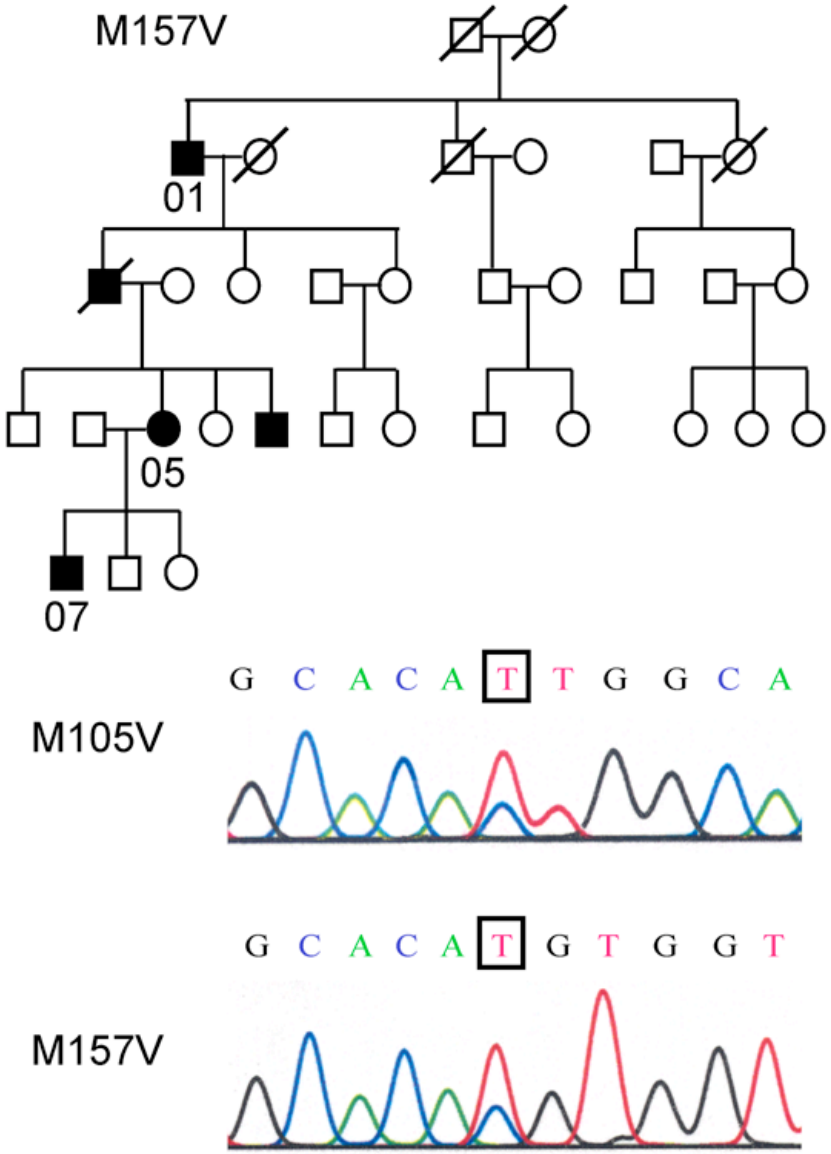


Fig. 1. Mechanism for retinopathy of prematurity (ROP) (Reprinted, with permission, from [37]).

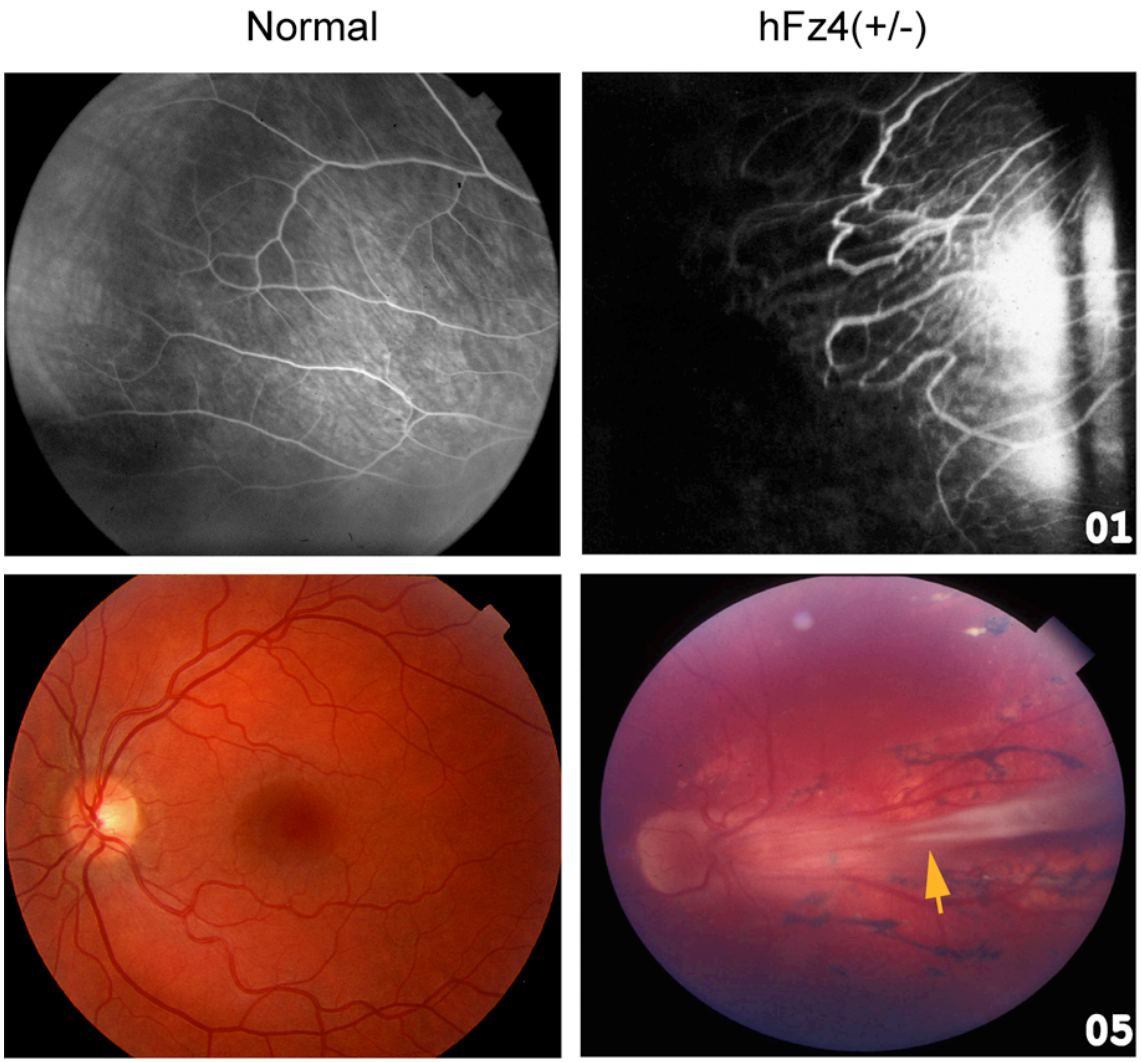
FEVR- Familial Exudative Vitreoretinopathy

- First described by Criswick and Schepens [Am. J. Ophthalmol. 68: 578-594 (1969)]
- Autosomal dominant, recessive, and X-linked forms; variable phenotype
- Clinical characteristics
  - mild to severe vision loss
    - retina: avascular peripheral retina, exudates, neovascularization, fibrovascular masses, traction or rhegmatogenous retinal detachment
    - vitreous: posterior vitreous detachment, fibrovascular membranes, hemorrhage
    - other: cataract, neovascular glaucoma

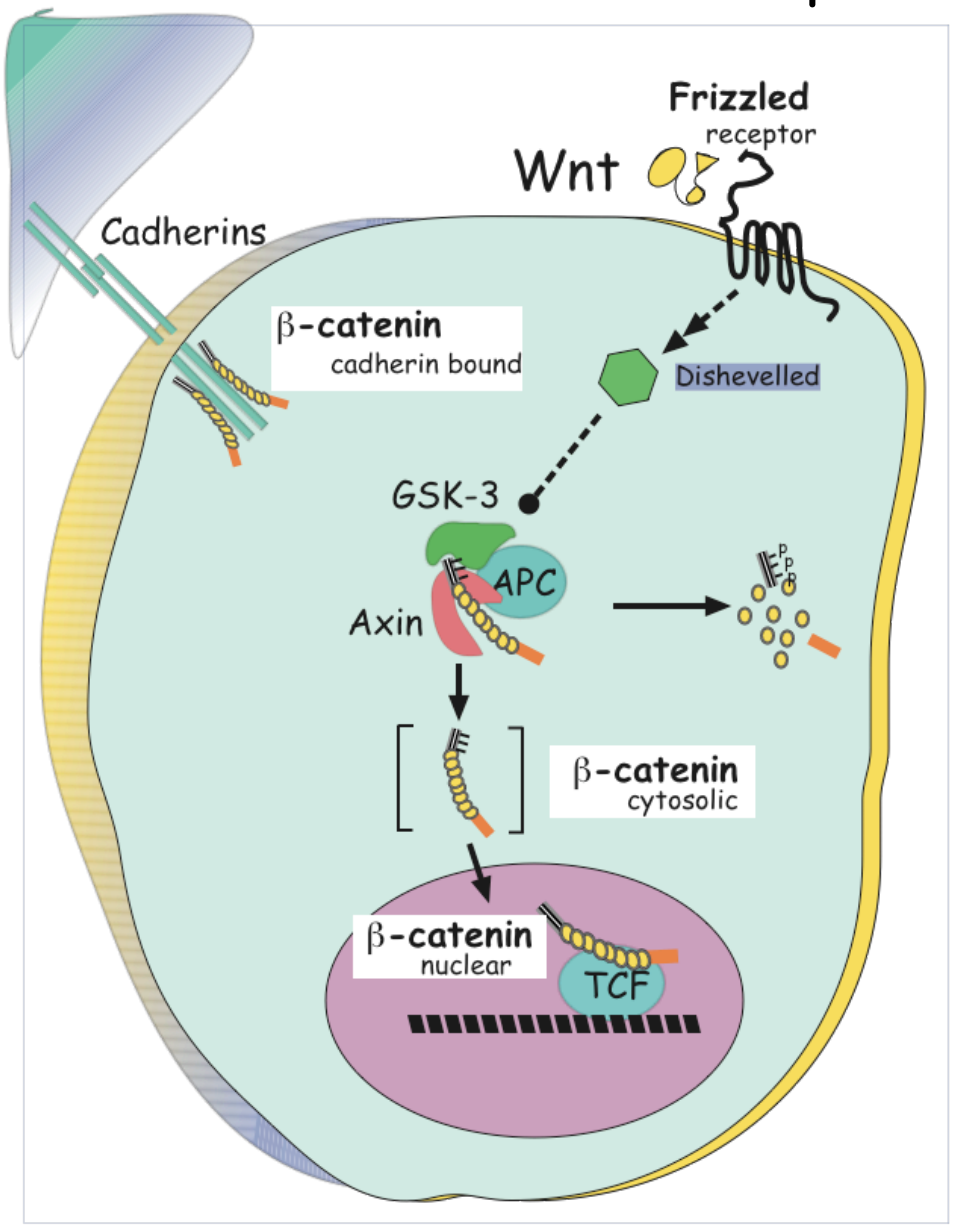
Autosomal dominant
FEVR mutations
In the cysteine-rich
domain (CRD) of Fz4



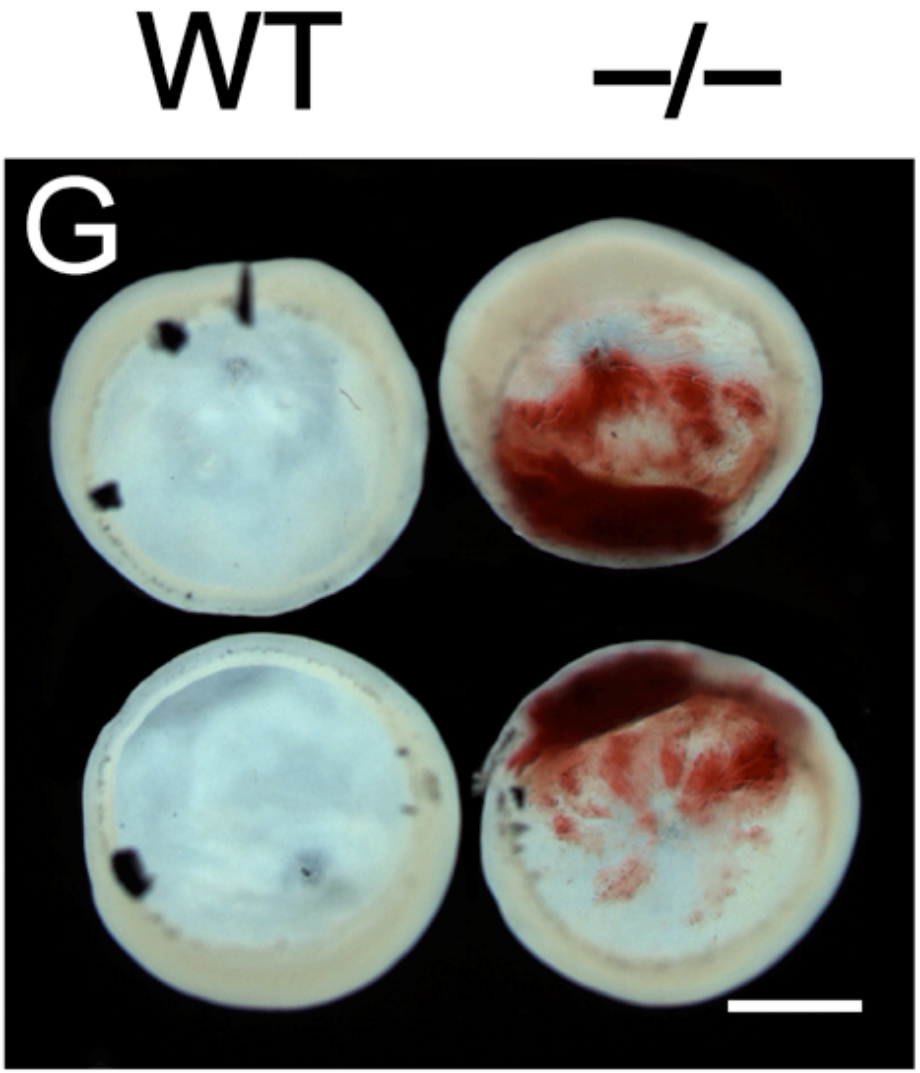
Retinal defects in FEVR patients heterozygous for Fz4 M157V



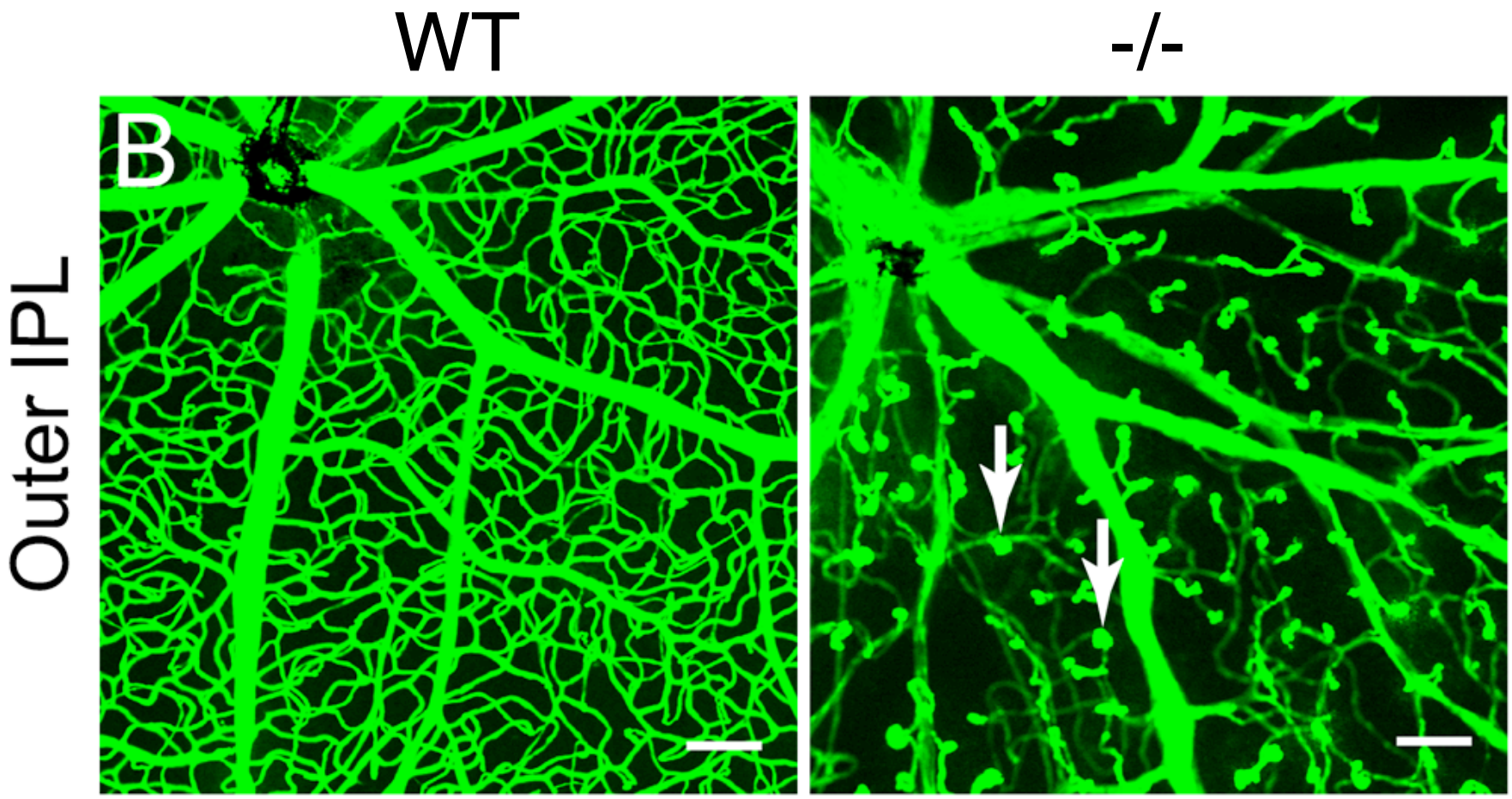
Frizzled-4 is a Wnt receptor



Intraocular hemorrhage in Fz4(-/-) mice



Absence of intra-retinal capillaries in Fz4(-/-) mice



Norrie Disease

- First described by Norrie (1927) and analyzed systematically by Mette Warburg [Acta Ophthalmologica 39: 757-772 (1961); 41: 134-146 (1963); 89: 1-147 (1966)]
- X-linked recessive with variable phenotype
- Clinical characteristics

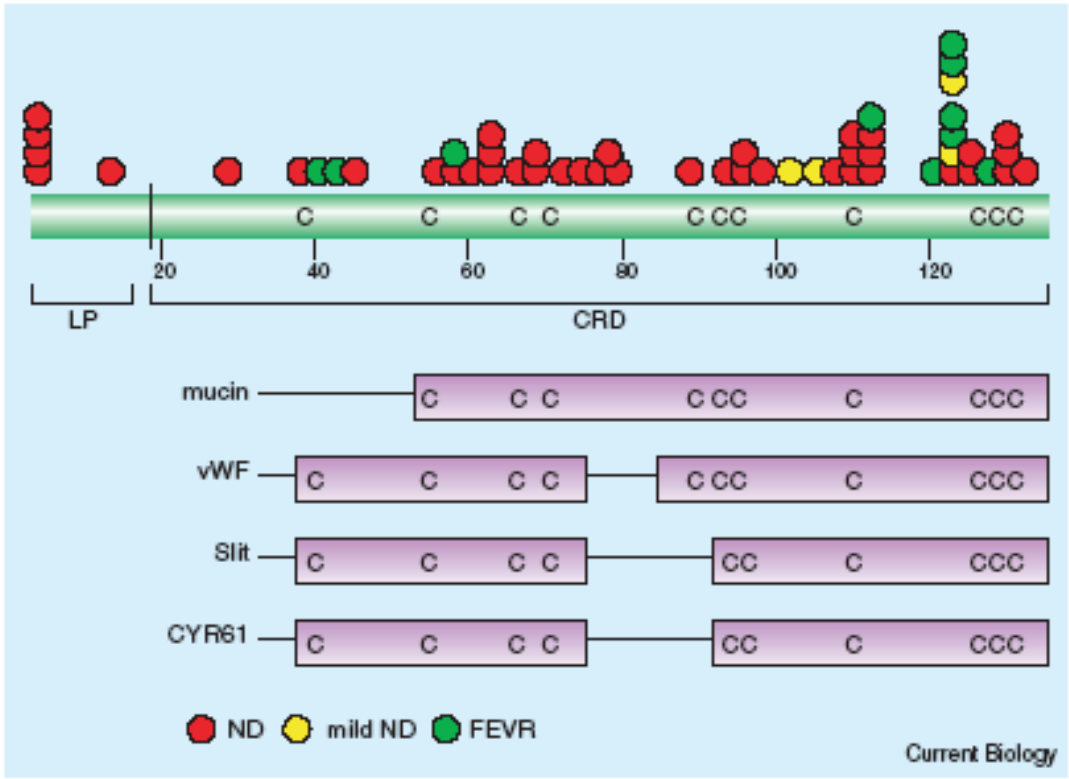
  moderate vision loss to congenital blindness

  retina: retinal folding and detachment, retinal degeneration, fibrovascualar masses, vitreoretinal hemorrhage

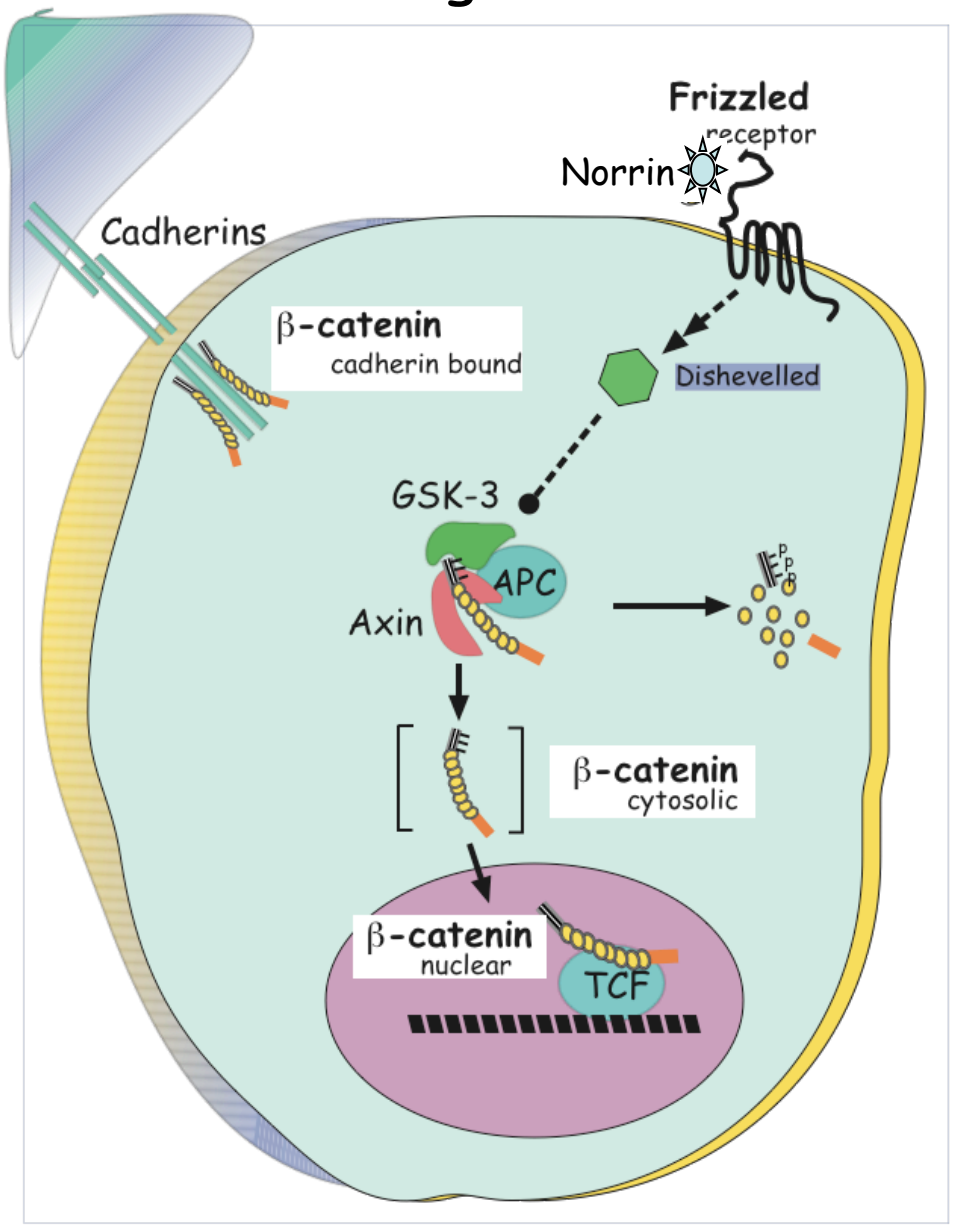
  vitreous: persistent primary vitreous

  other: progressive sensorineural deafness

Figure 1. The Norrin protein.
Missense and nonsense mutations in Norrin leading to classic or mild Norrie disease (ND) and to FEVR. Amino acid numbering is given on the scale below the protein. LP, leader peptide; CRD, cysteine-rich domain. The CRD is homologous to domains in the proteins indicated below (adapted from [1]).



Norrin is a ligand for Frizzled-4



Molecular genetics of Norrie Disease and FEVR

- FEVR

  One autosomal dominant FEVR gene identified by Robaitaille et al [Nature Genetics 32: 326-330 (2002)] encodes Frizzled4, a putative Wnt receptor. A second autosomal dominant FEVR locus encodes the Wnt co-receptor Lrp5 [Toomes et al [IOVS 45: 2083-2090 (2004)]; Jiao et al [Am J Hum Genet 75: 878-884 (2004)].

- Norrie disease

  Gene identified by Berger et al and Chen et al [Nature Genetics 1: 199-203 and 204-208 (1992)]

  The encoded protein is small (133 amino acids in length), has the same pattern of cysteines as seen in transforming growth factor beta, and begins with a signal sequence (i.e. it looks like a secreted protein). No known biochemical function.

Vessel component to human diseases

- Tumor angiogenesis *(treatment available ?)*
- Tumor lymphangiogenesis, metastasis
- Diabetic vascular complication
  - Diabetic retinopathy
  - Stroke
  - Ischemia
  - Wound repair
- Heart disease
  - Occluded vessels
  - Rebuilding vessels of damaged heart tissue
- Obesity
- Lymphedema
- Blindness
  - Wet Macular Degeneration *(treatment available)*
  - Retinopathy of Prematurity