UNIVERSITY OF GUELPH

Reproductive Pathology:

Male and female gonadal disease made easy

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VetReproPath.com

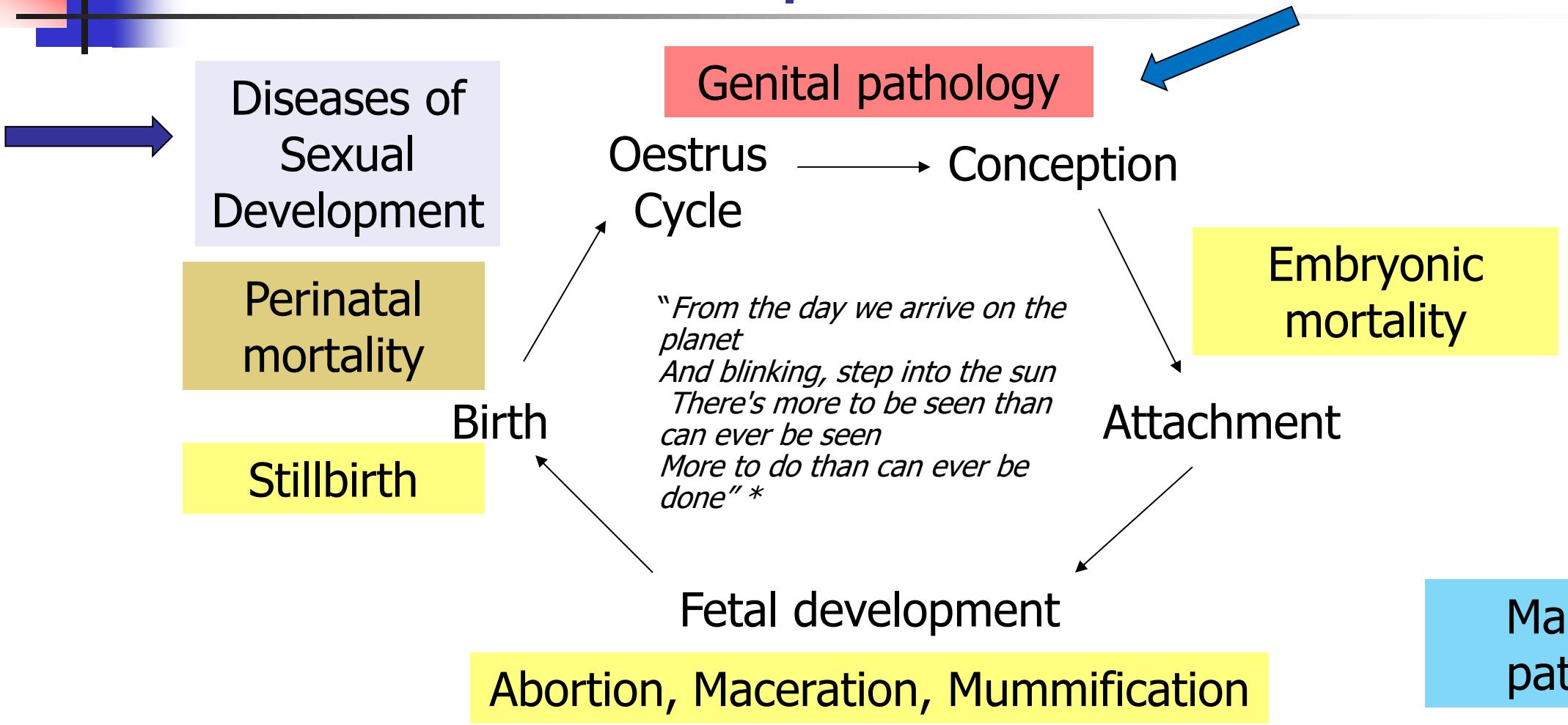
UNIVERSITY OF GUELPH | Ontario Veterinary College

Objectives

- Outline the embryology of sexual development.
- Use embryology to explain disorders of sexual development, gonadal neoplasia, and common tests applied.

- Highlight common and important diseases and conditions

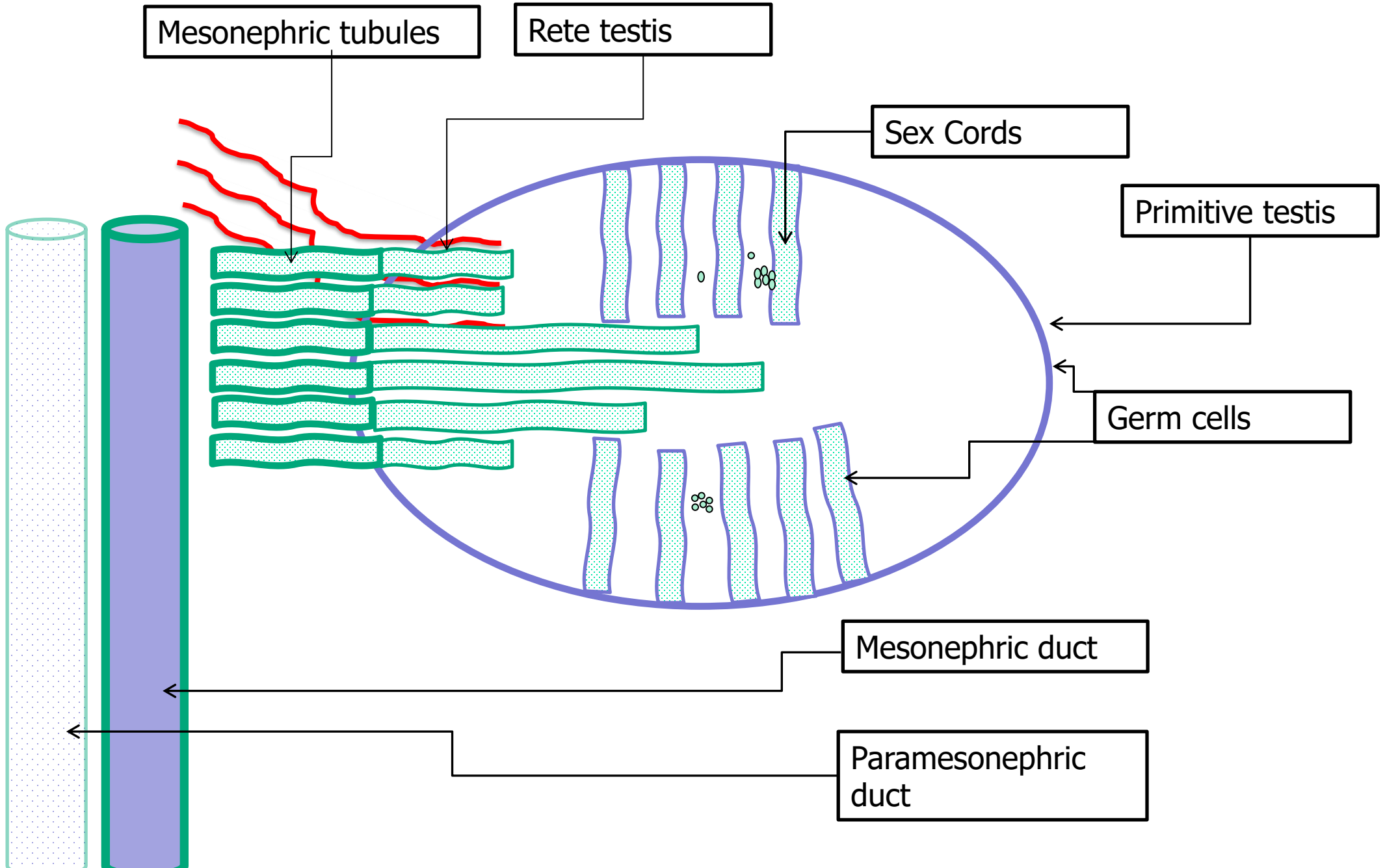
The Circle of Reproductive Life*



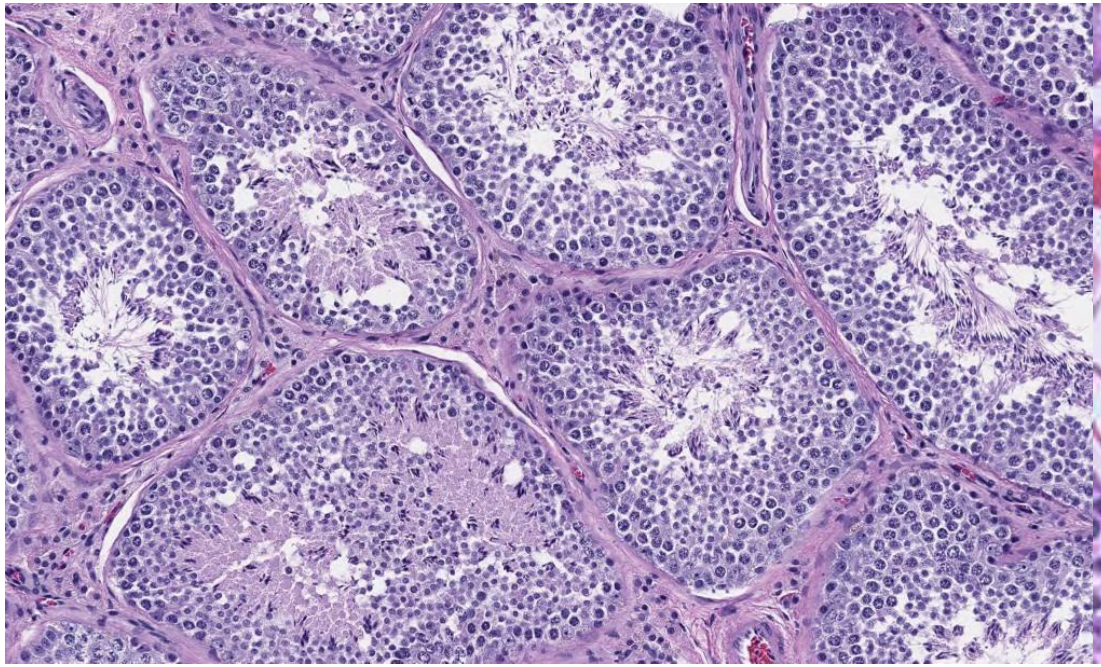
* "Circle of Life" Music by Elton John, lyrics by Tim Rice, Performed by Elton John

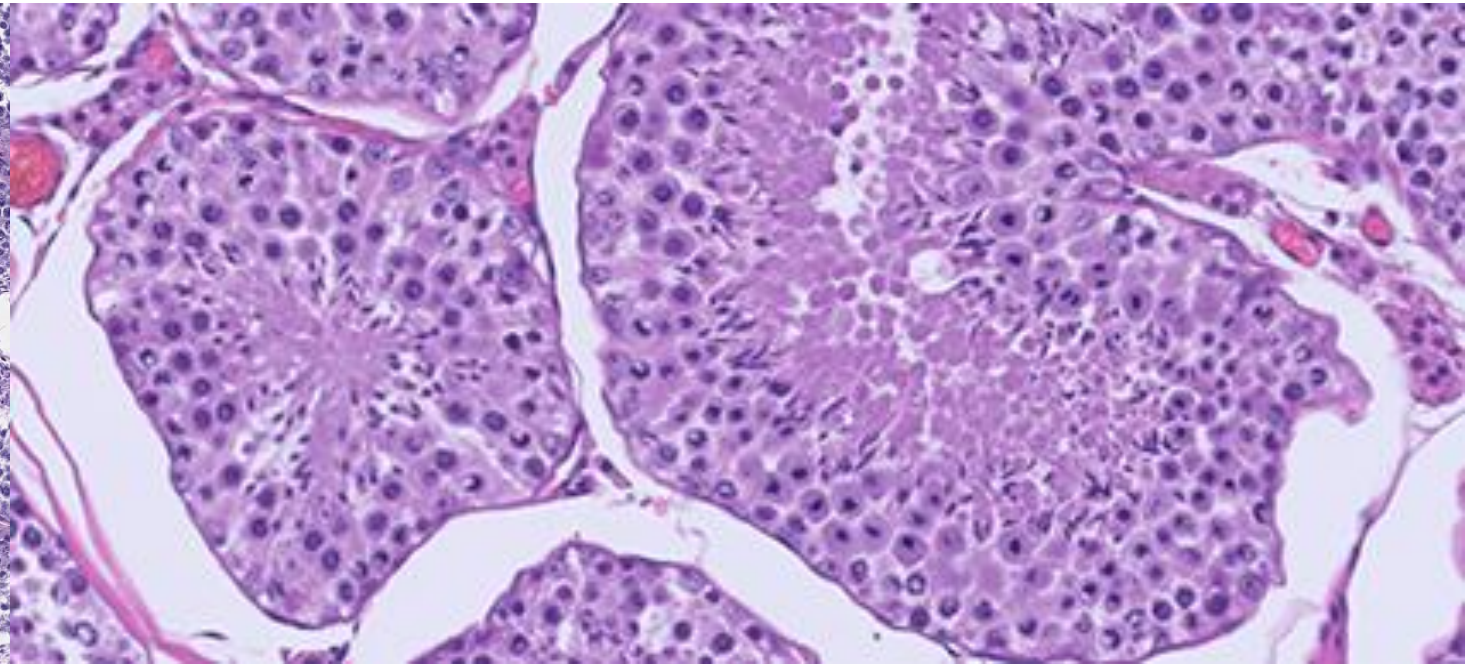
Male gonad

Embryology of the Male



Normal testis





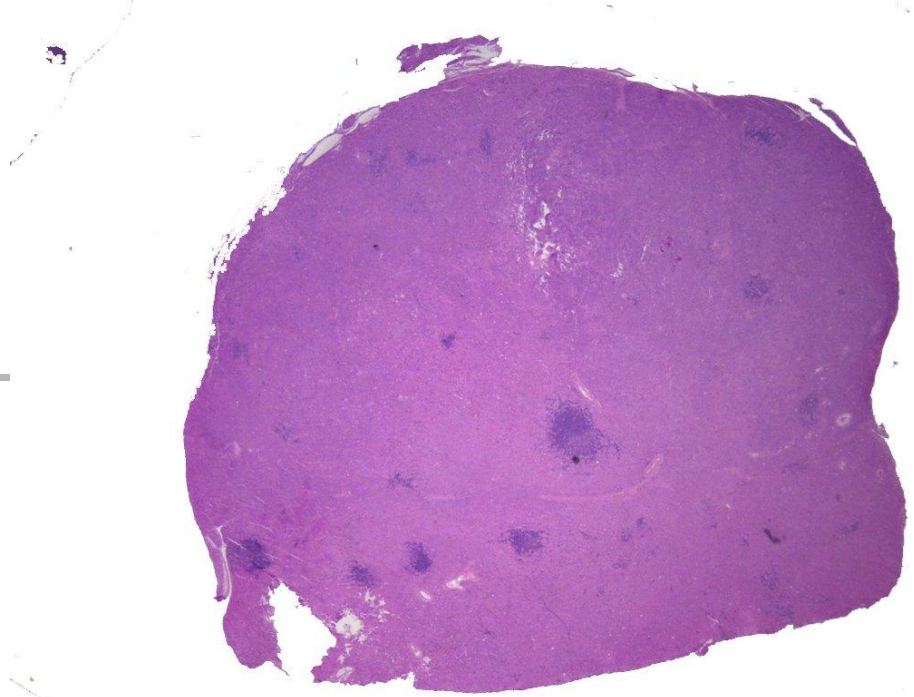
Male gonadal neoplasia

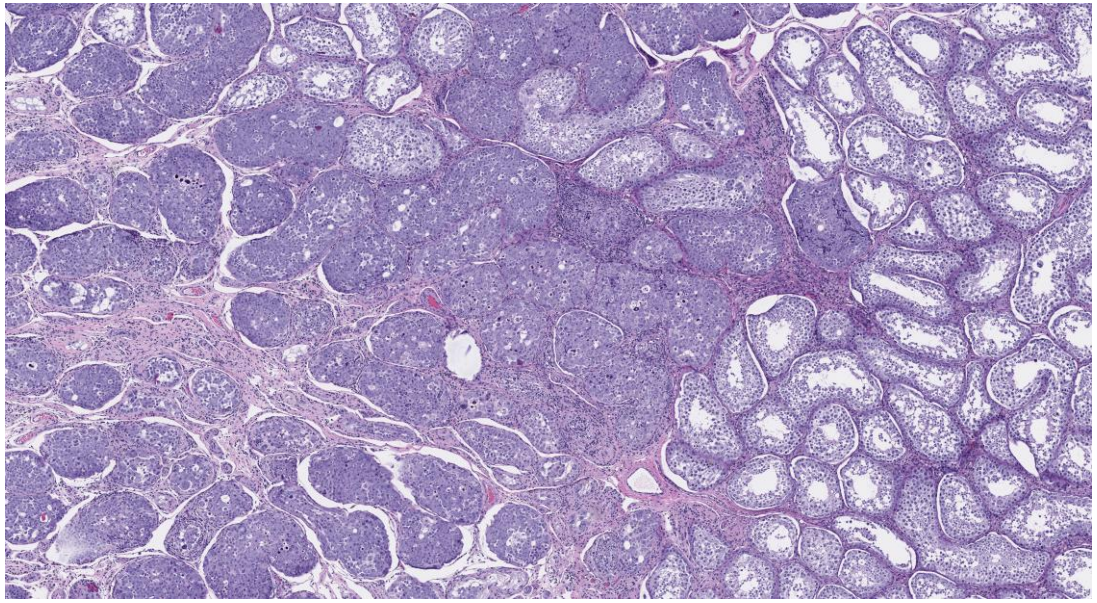
- Germ cell
  - Seminoma
- Sex cords and Stroma
  - Sustentacular cell tumor
  - Interstitial cell tumor
- Epithelium
  - Rete adenocarcinoma (too rare to consider)

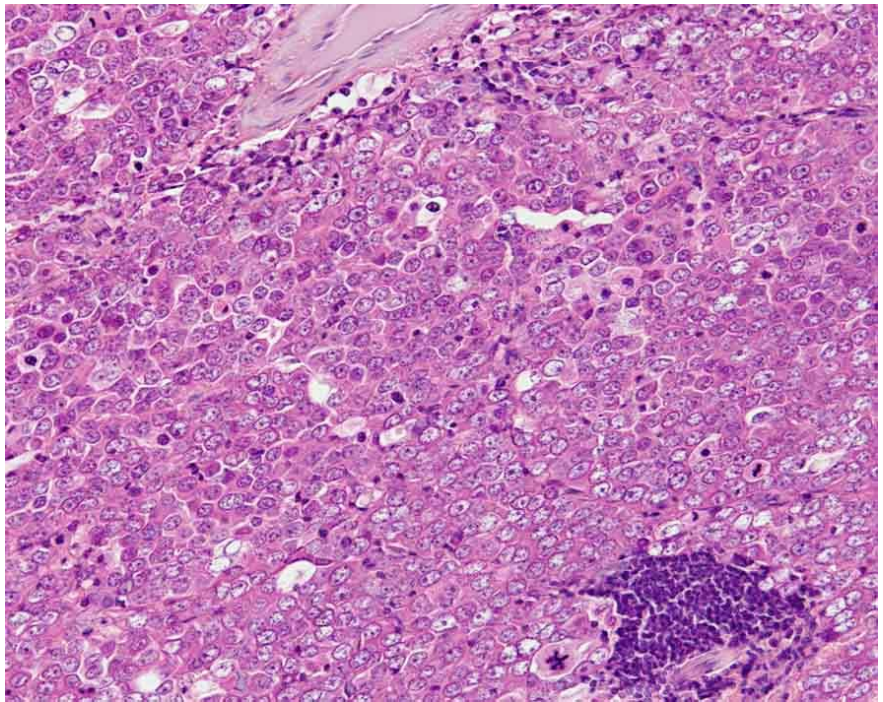
Seminoma

Intratubular

Diffuse

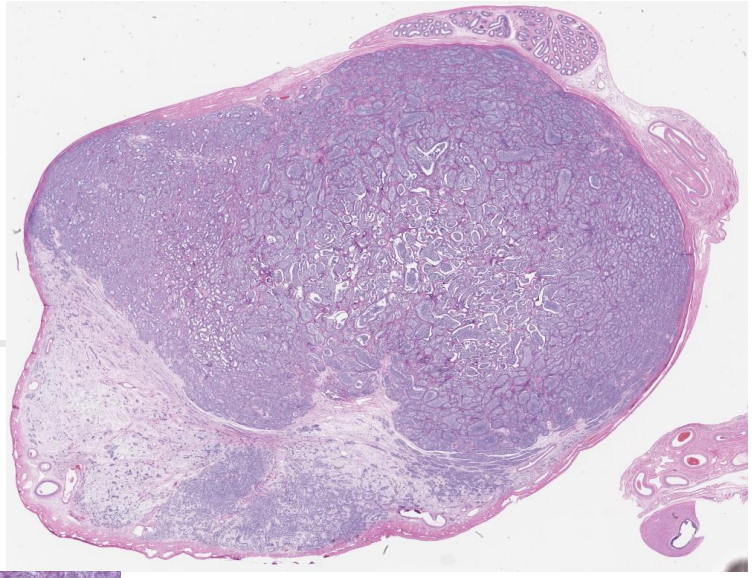


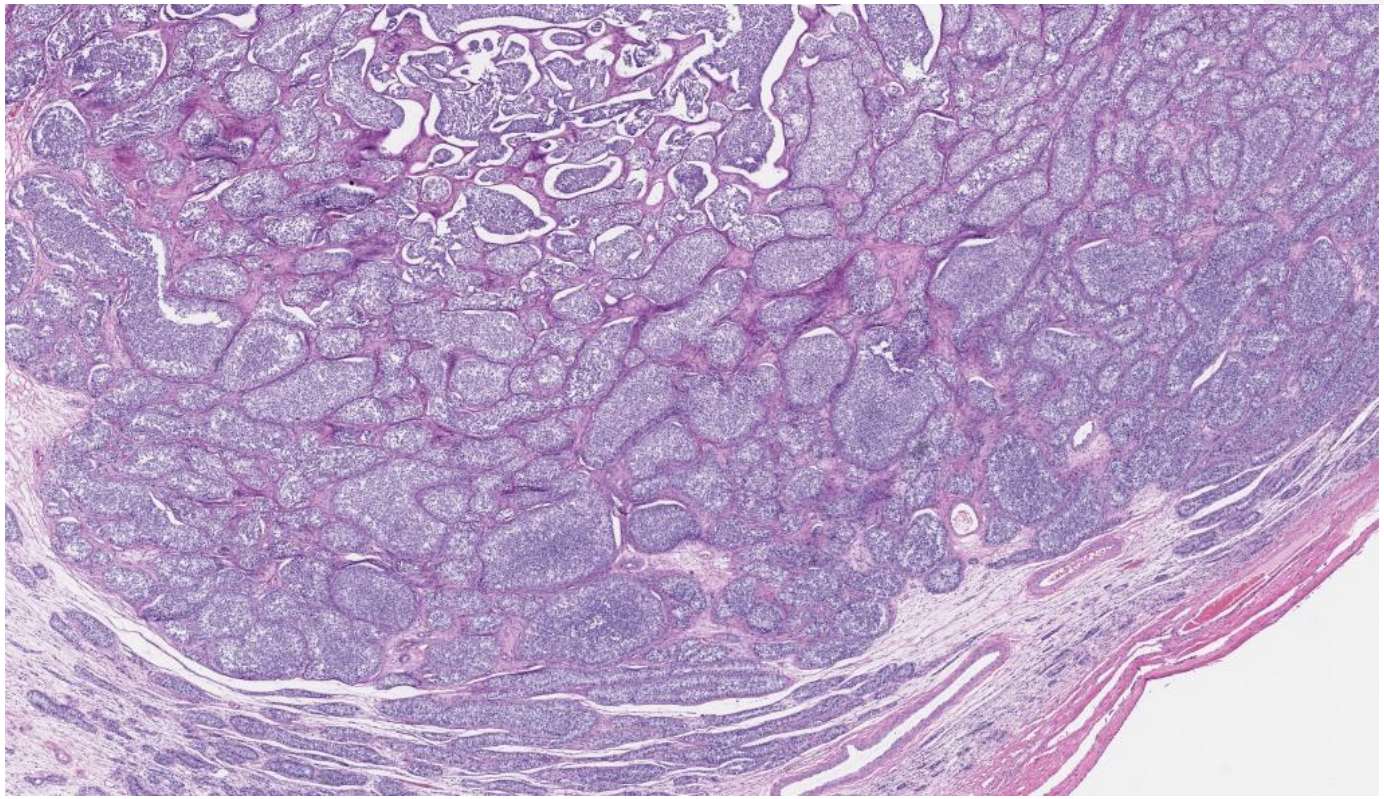




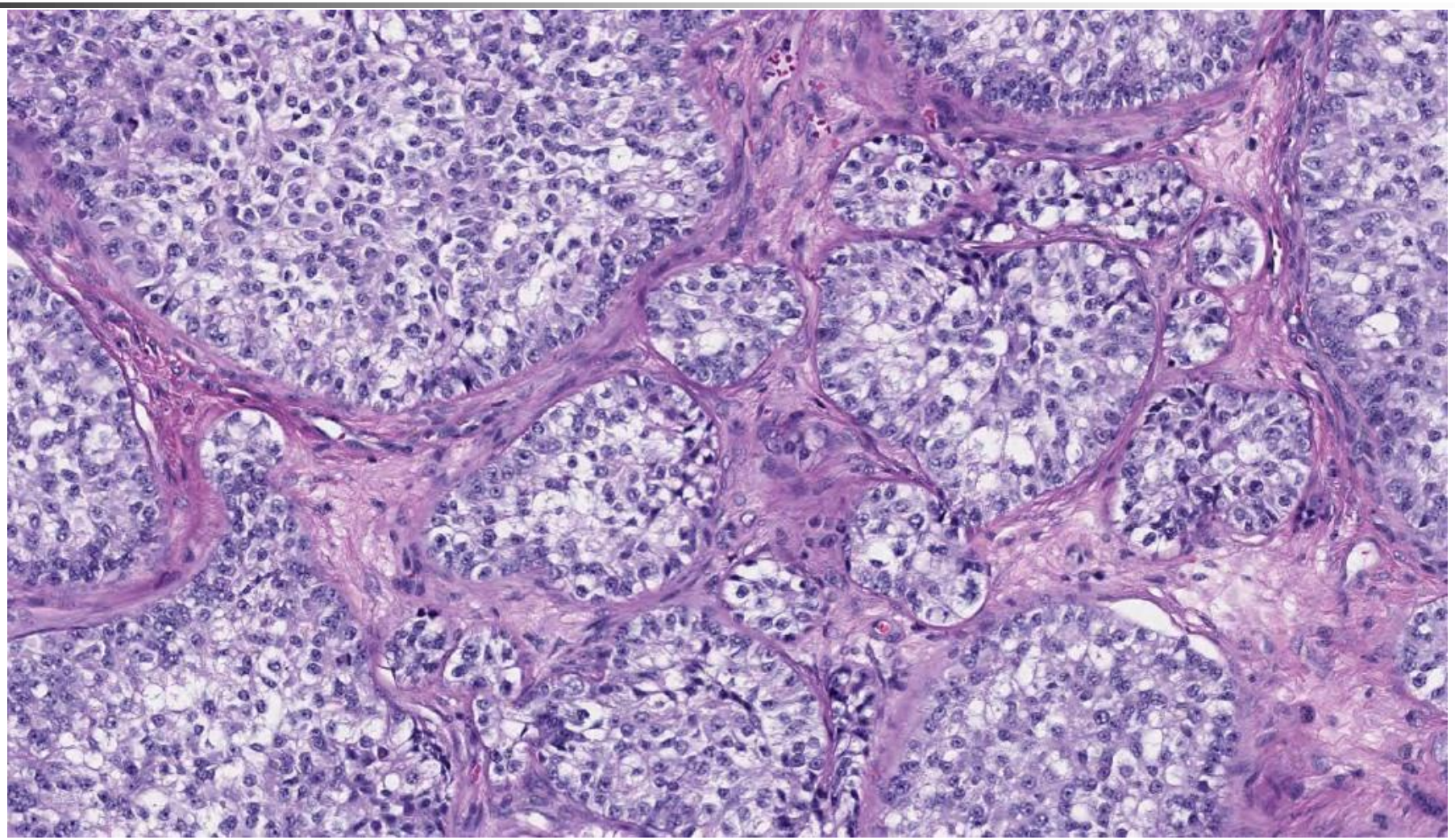
Sustentacular cell tumor

Tubular
Diffuse

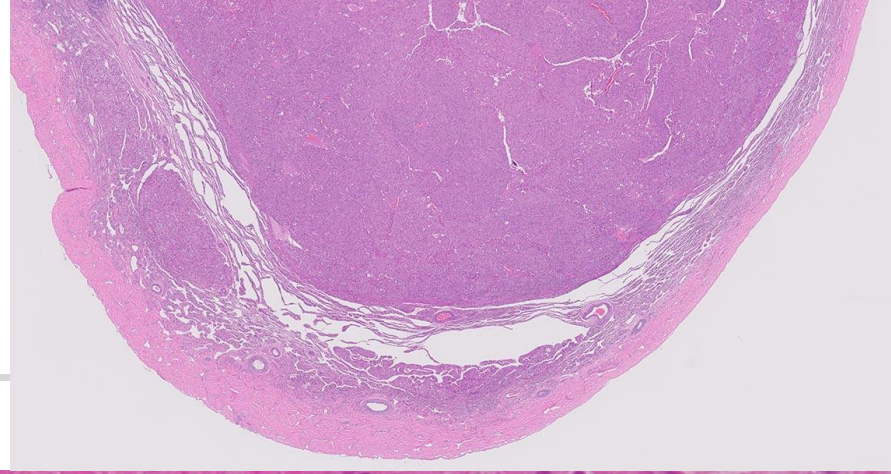


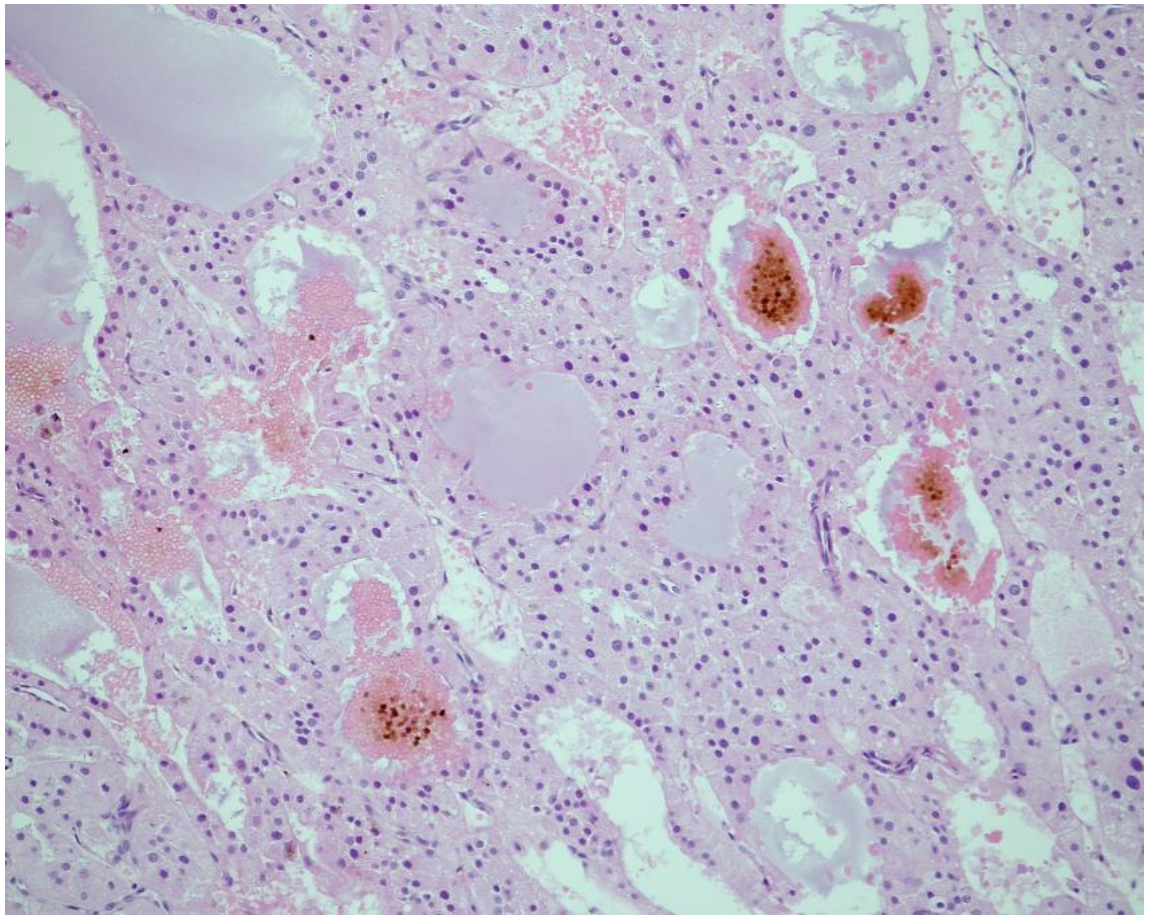


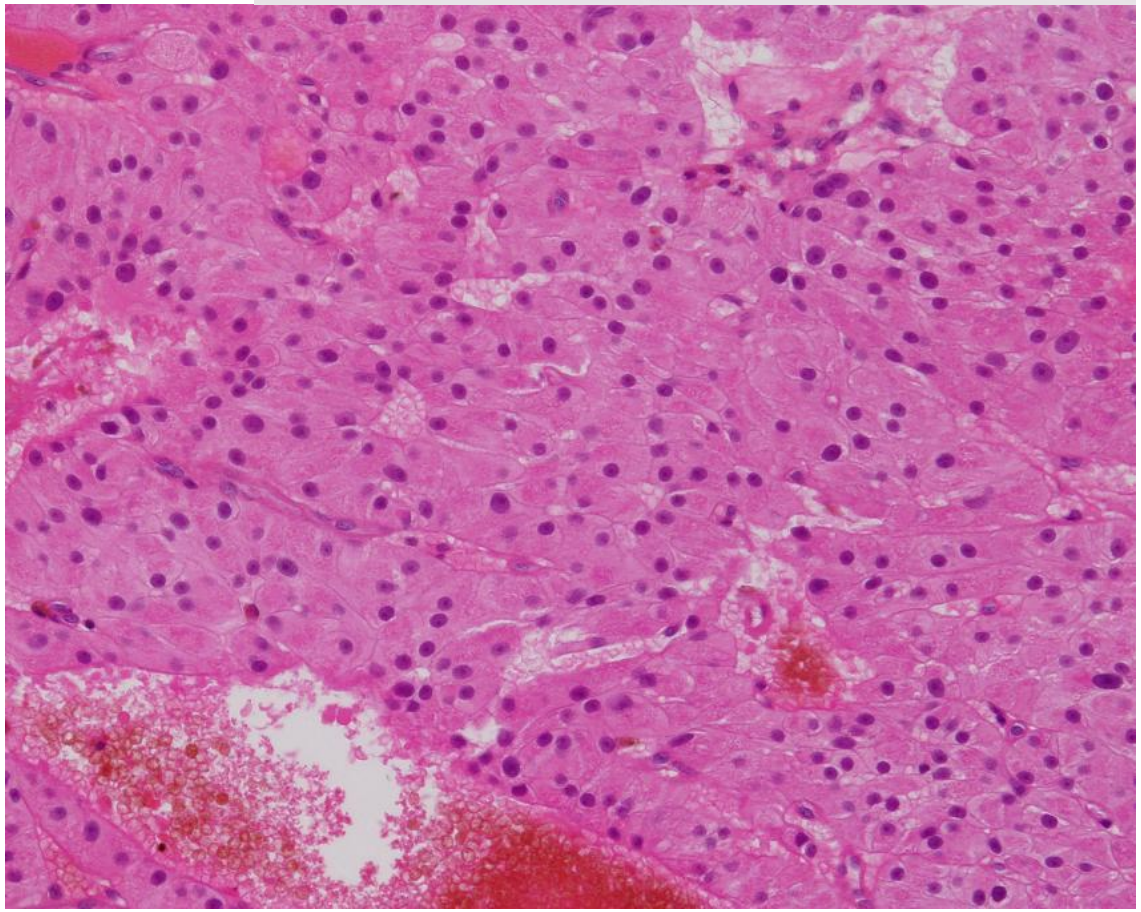
Sustentacular cell tumor - tubular



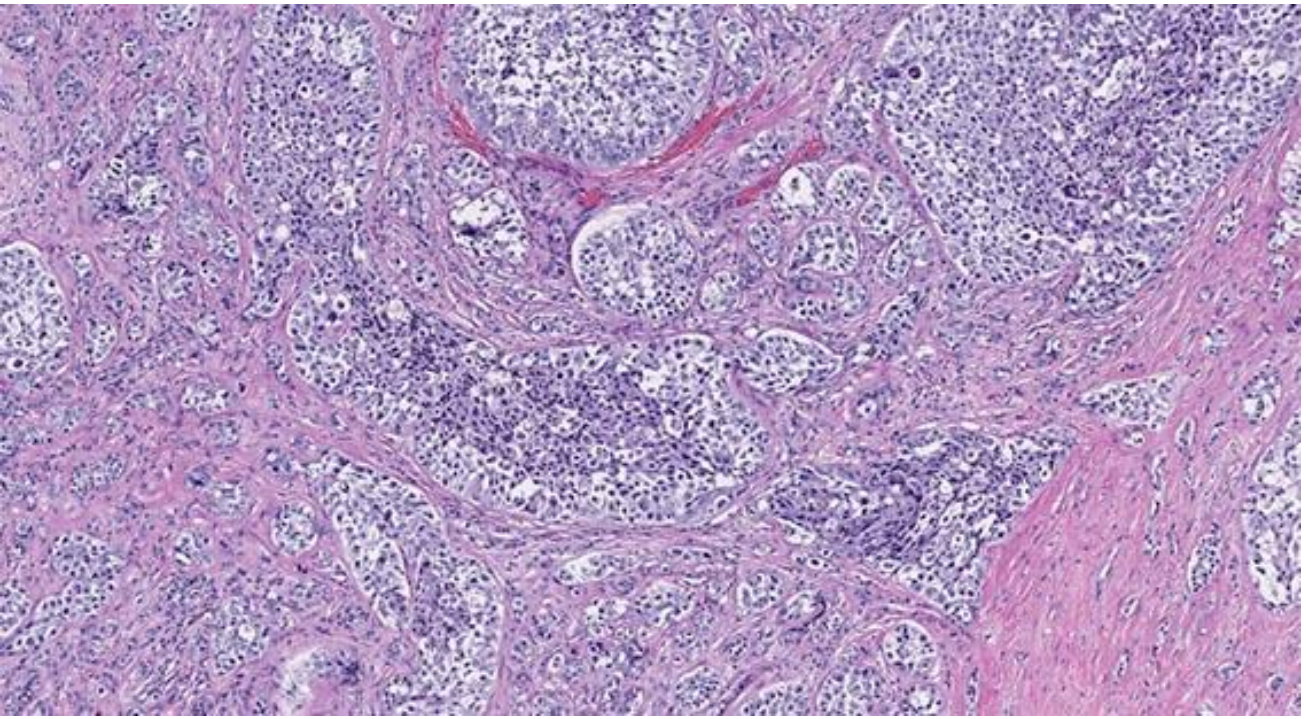
Interstitial cell tumor

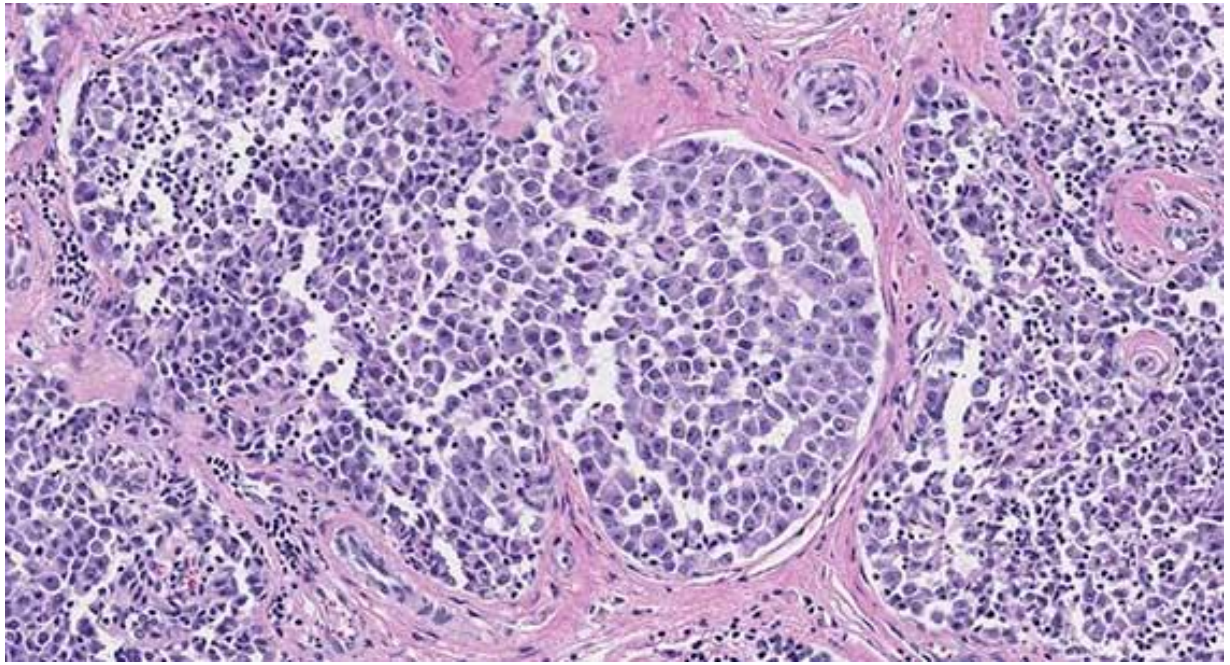






Mixed germ cell sex cord stromal tumor



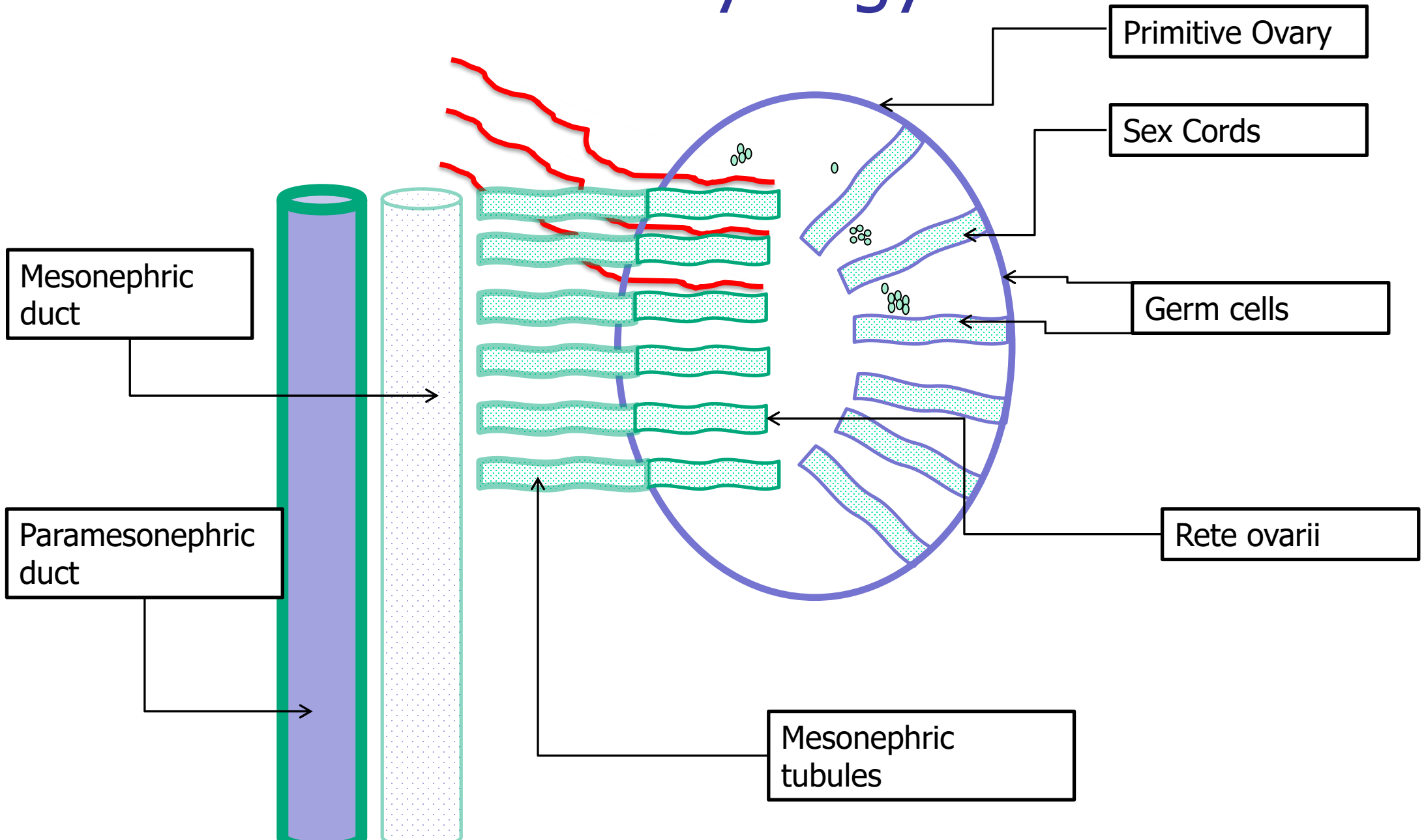


Testicular - Immunohistochemistry

- Germ cell tumors
  - CD117, Placental ALP
- Sex cord stromal tumors
  - Sustentacular cells
    - fetal: panCK, desmin, SOX-9, AMH
    - Adult: inhibin-α,
  - Interstitial endocrine cells – Inhibin-α, AMH (adult), CD117
- Epithelium
  - Pancytokeratin, CK7, vimentin

Female gonad

Normal female embryology



Normal female development

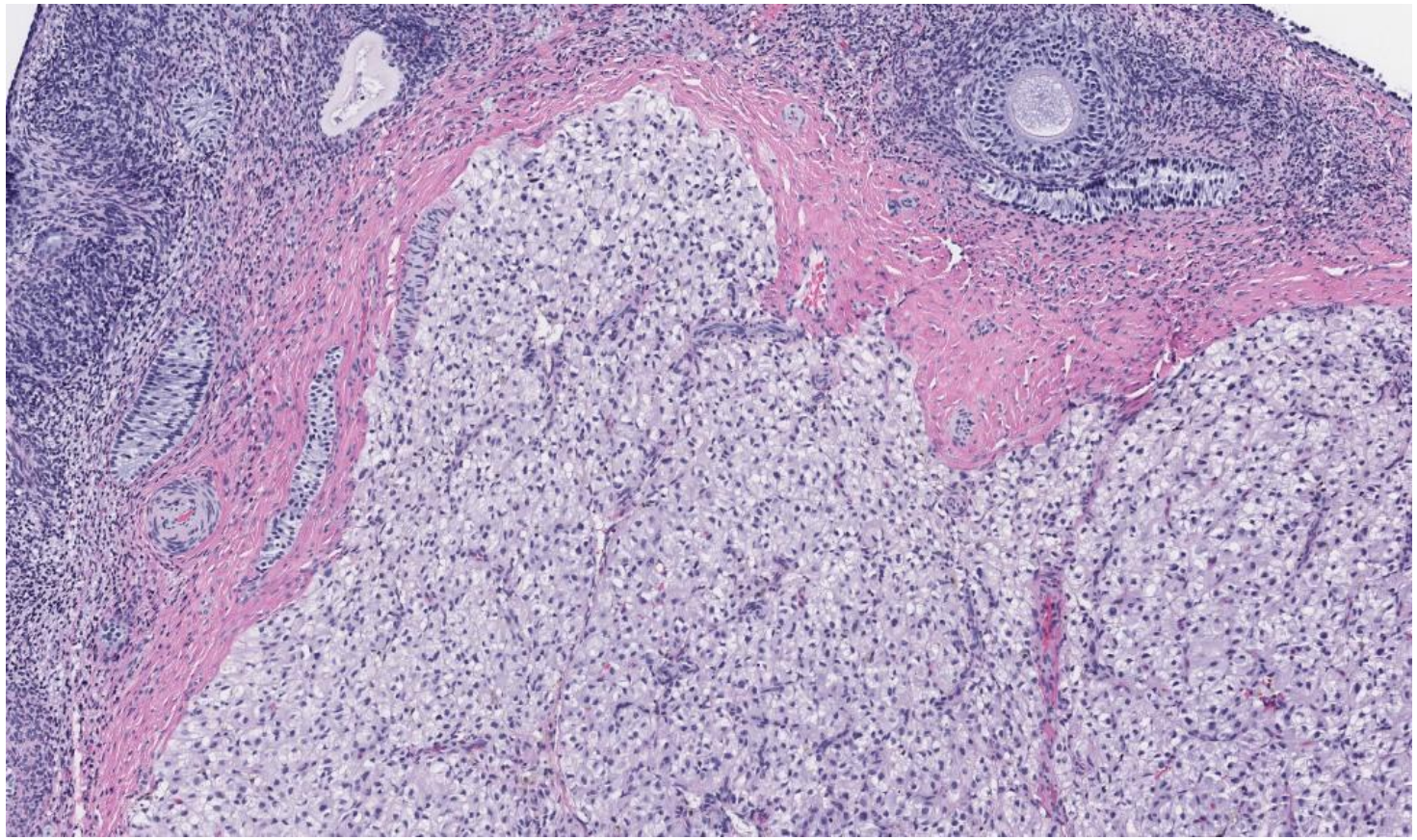
- Gonad - ovary
  - No specific ovary determining gene like *SRY* of testis
  - Pro-ovarian genes: *RSPO1-WNT4-FOXL2* pathways.
  - *DAX1* and *FOXL2* occurs early and or before *SRY* is expressed in males.
  - If activated in a male embryo, there is a male to female sex reversal.
- Tubular genitalia
  - If no AMH – uterine tube, uterus, cervix, cranial vagina don't develop.
- External genitalia – female phenotype
  - In no testosterone.

Reproductive embryology of ducts/tubules

Ovary	Origin	Testis
Rete ovarii	Mesonephric tubules	Rete testis
	Mesonephric duct	Epididymis Deferent duct
Uterine tube Uterus Cervix Cranial Vagina	Paramesonephric duct	
Caudal Vagina Vulva	Urogenital tubercle and sinus	Prostate, Bulbourethral gland Penis and prepuce

Ovarian development

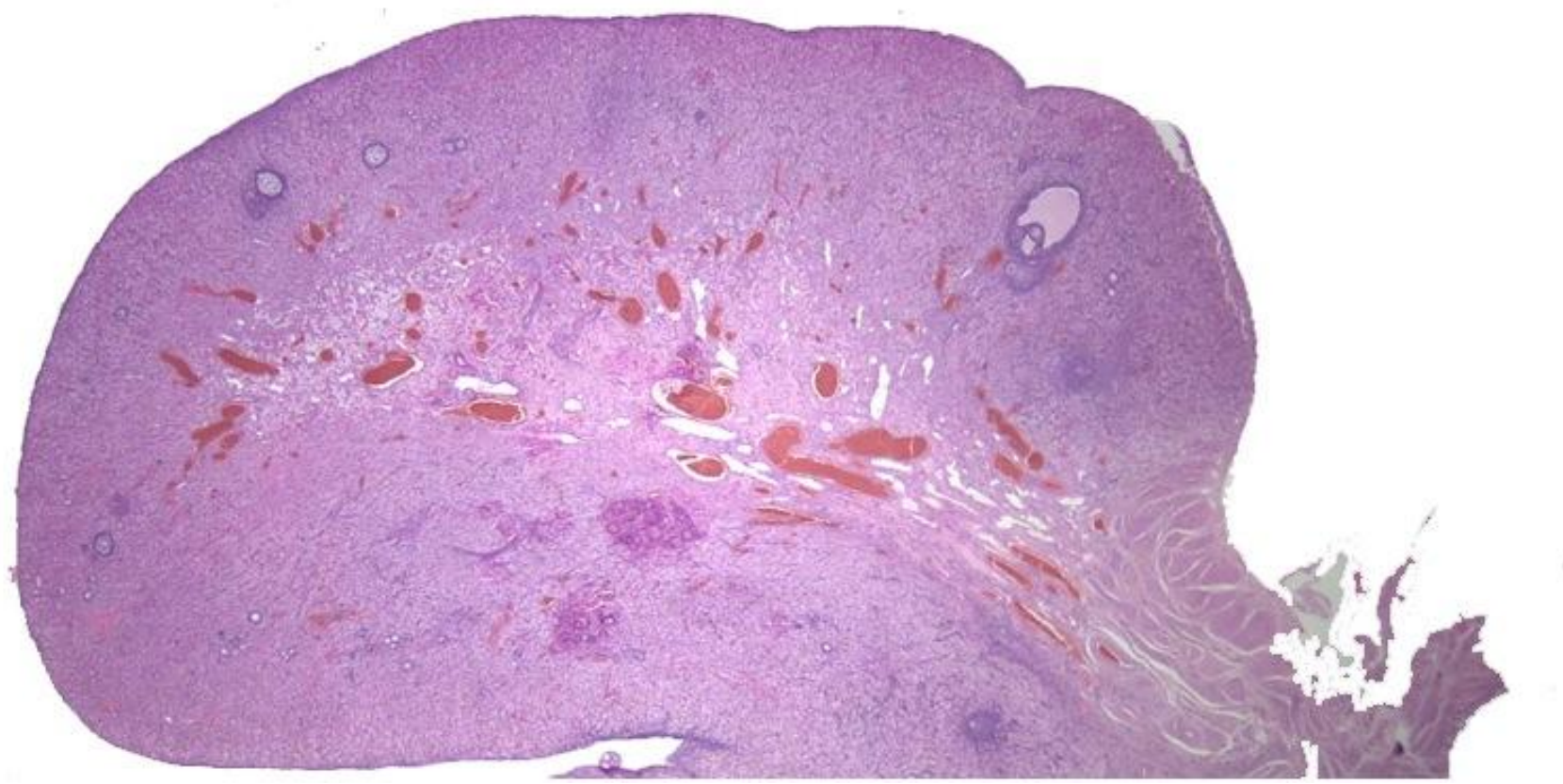
- Pro-ovarian genes
- Germ cell migration from Caudal epiblast and Yolk sac to sex cords to form primordial follicles
- Vascular migration from retroperitoneum
  - Interstitial endocrine cells similar origin to adrenal (pericytes – vascular smooth muscle cells)
  - Cats have lots of interstitial endocrine cells
  - Ectopic adrenal tissue especially in cats.



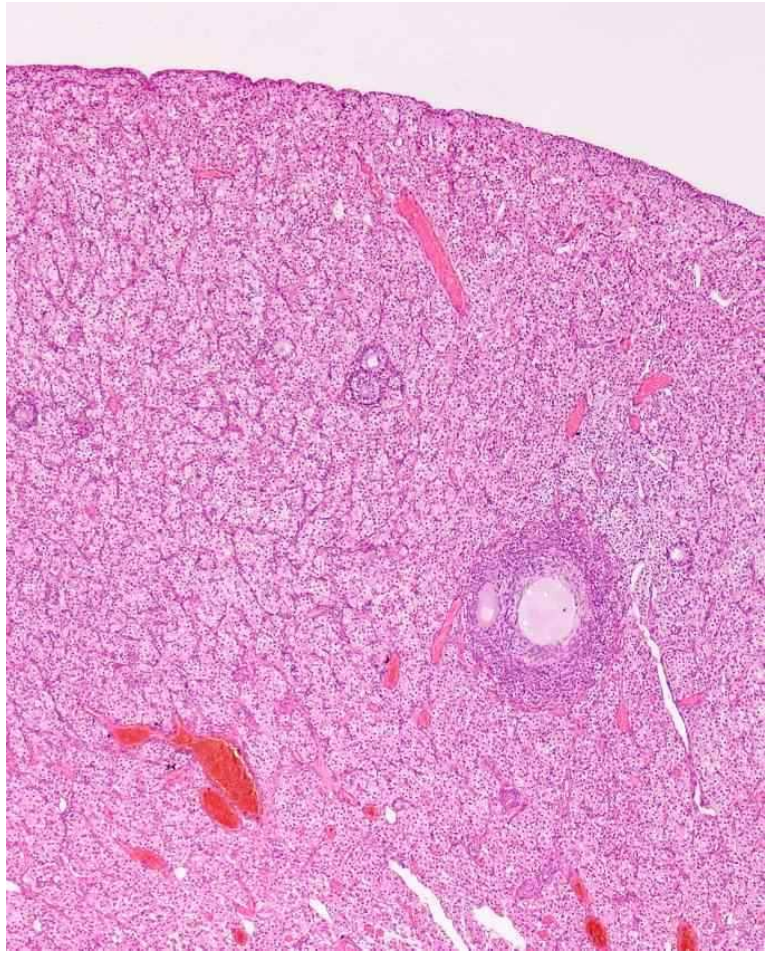
Ovarian – Normal immunohistochemistry

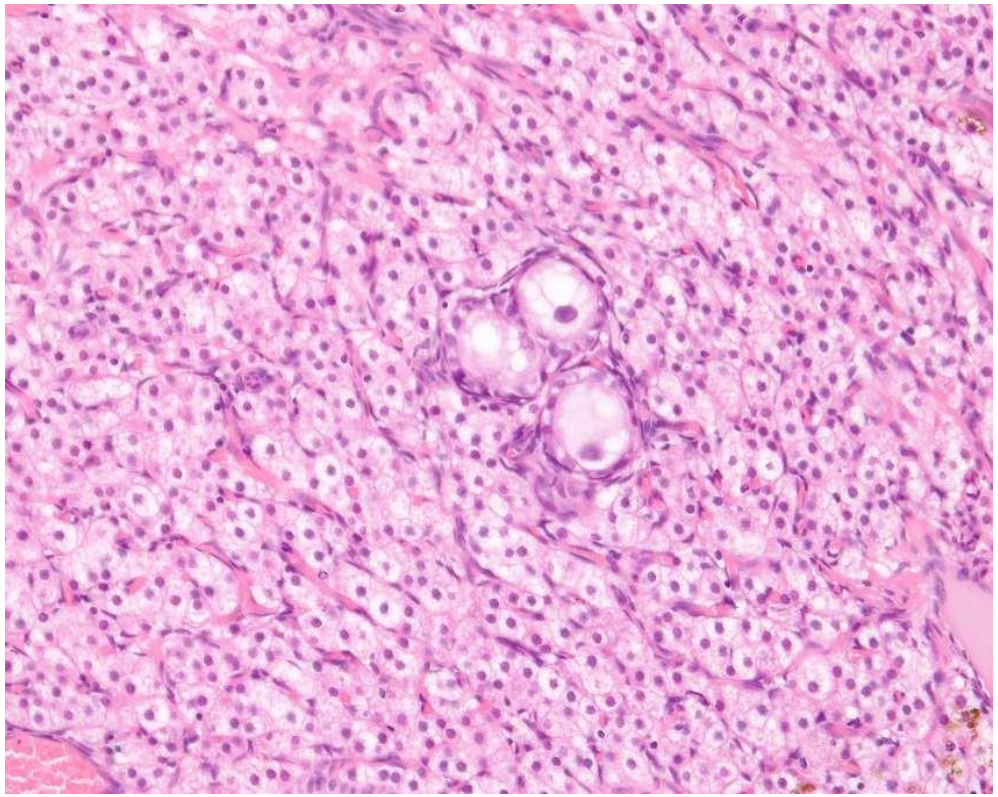
- Germ cells
  - CD117, PALP
- Sex cord stromal cells
  - Granulosa cells
    - fetal: panCK, desmin, SOX-9, AMH
    - Adult: inhibin-α
  - Interstitial endocrine cells – Inhibin-α, AMH (adult), CD117
- Epithelium
  - Pancytokeratin, CK7, vimentin

Feline ovarian enlargement



Interstitial endocrine cell hyperplasia



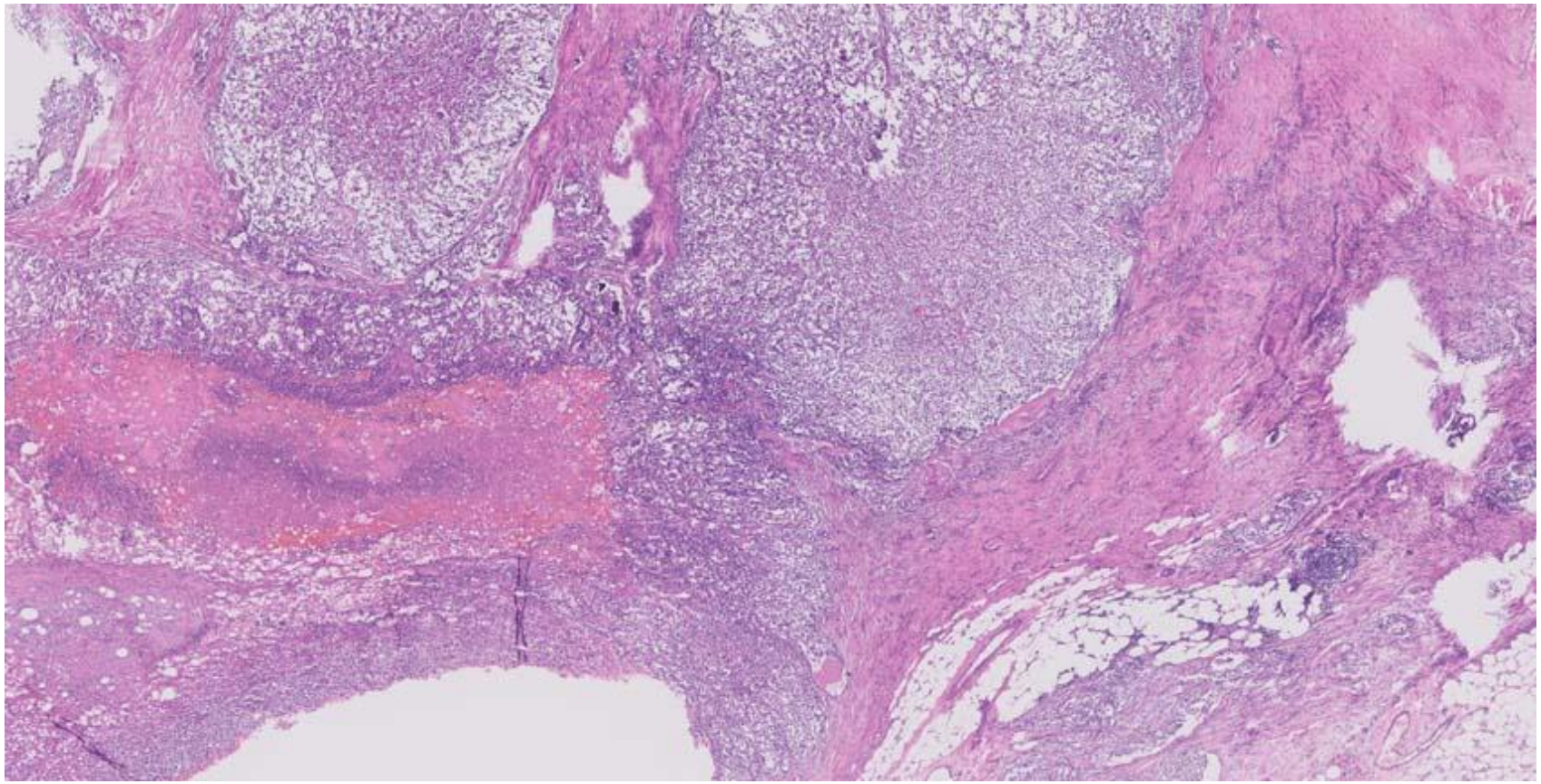


Sex cord stromal tumours

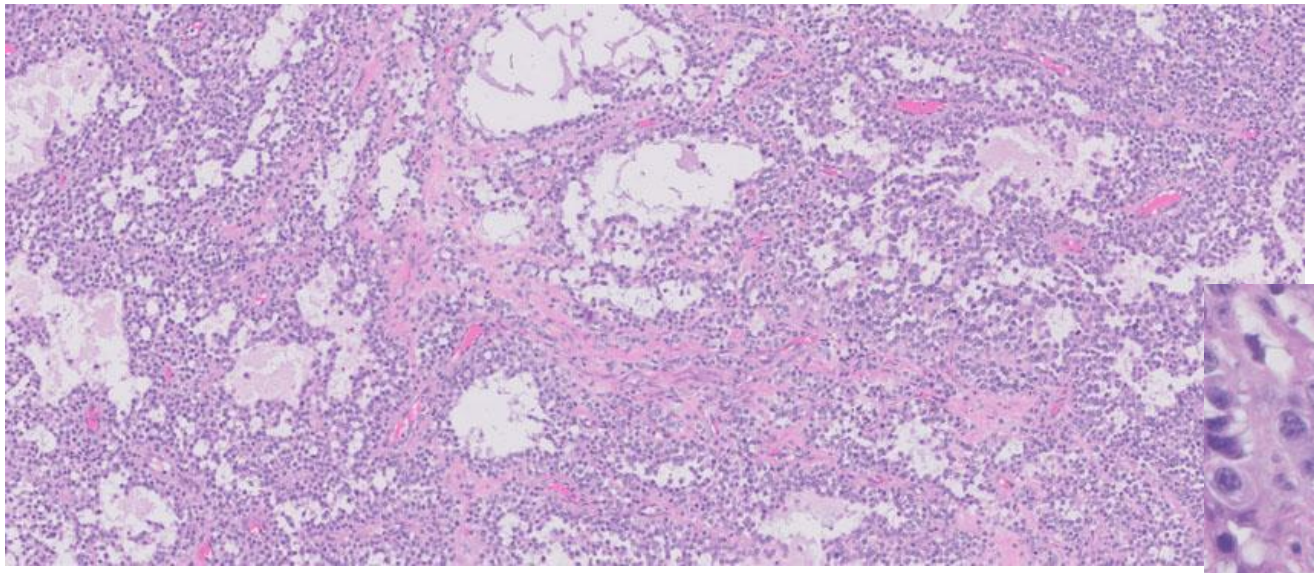
- Granulosa cell tumor
- Thecoma
- Luteoma

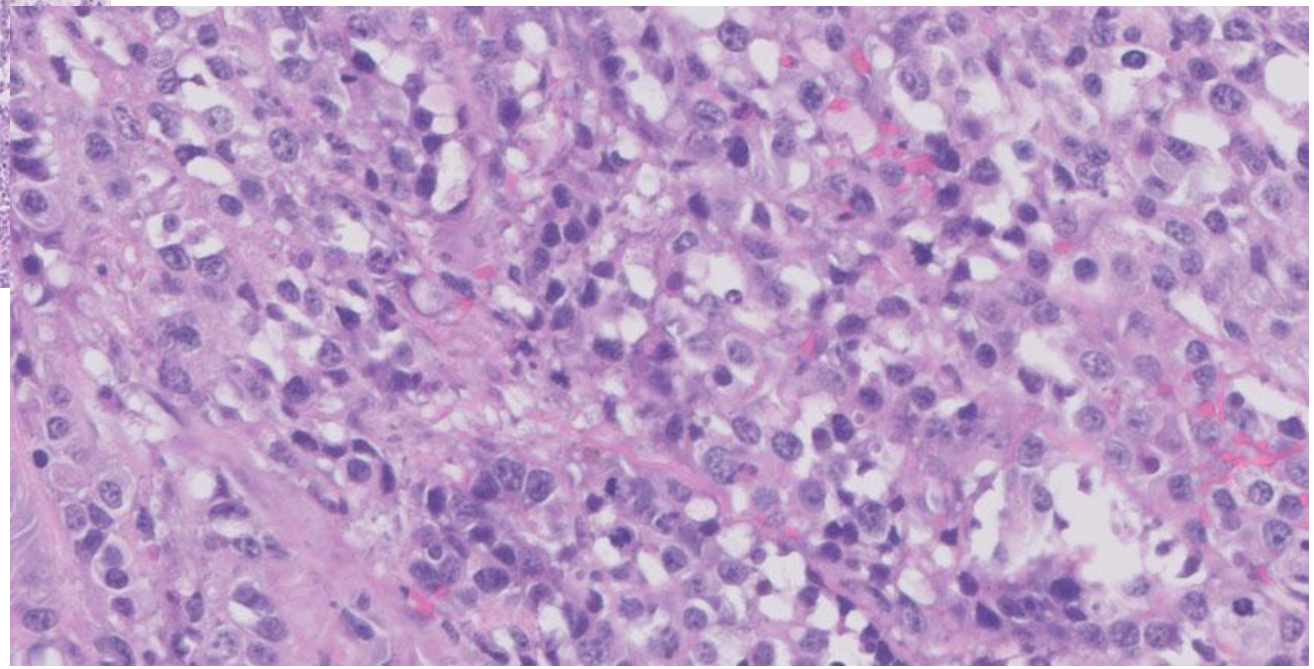
- Interstitial cell tumour

The many faces of Sex cord Stromal Tumors

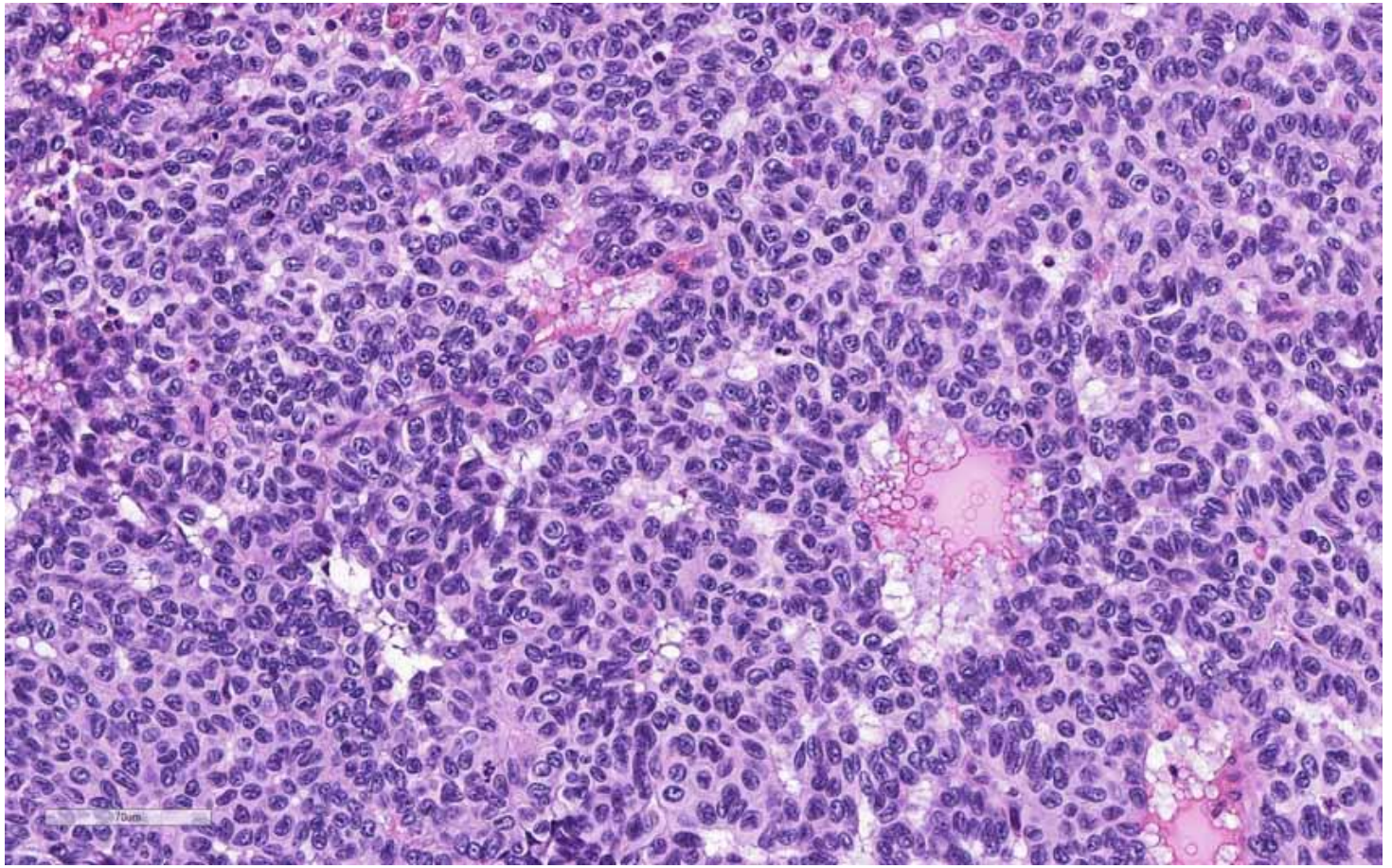


ScST – granulosa cell tumour

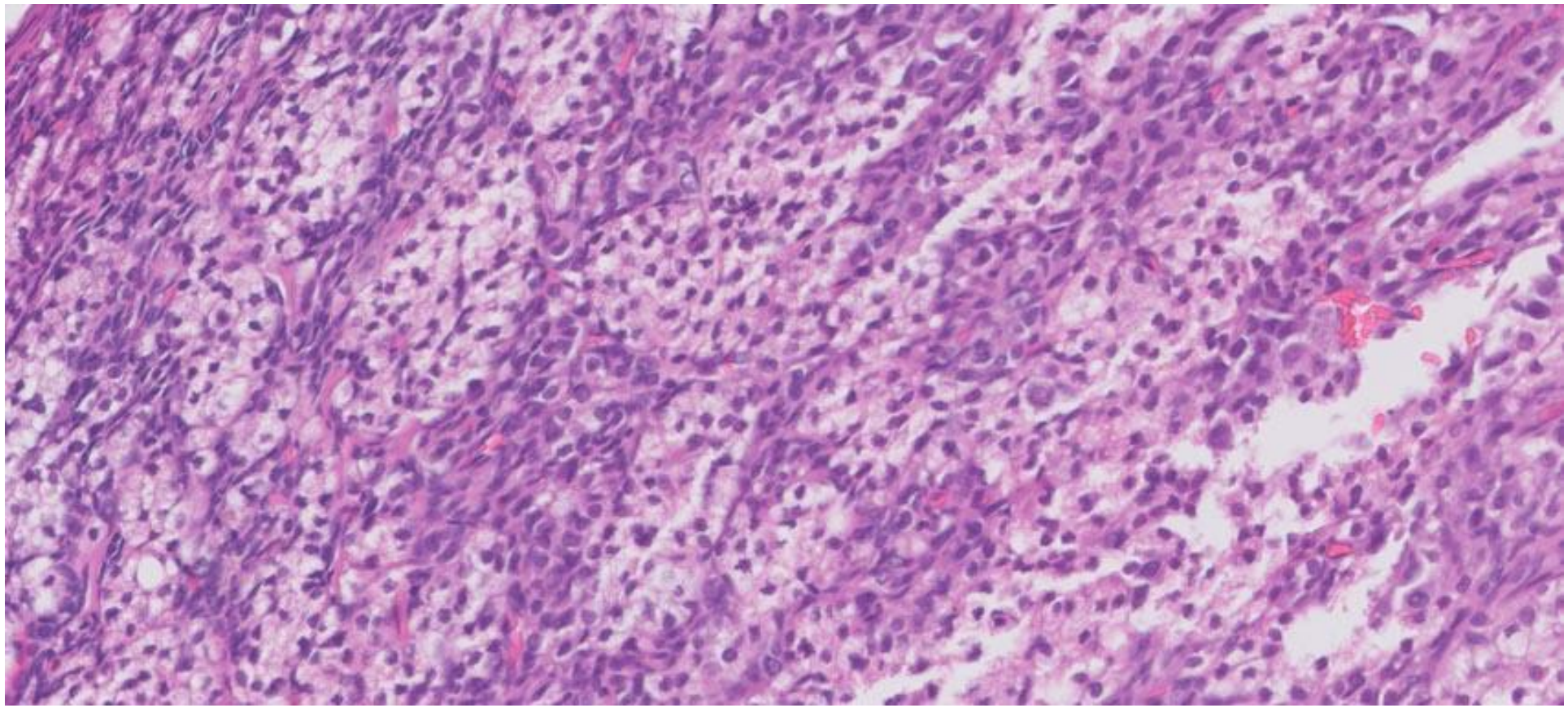




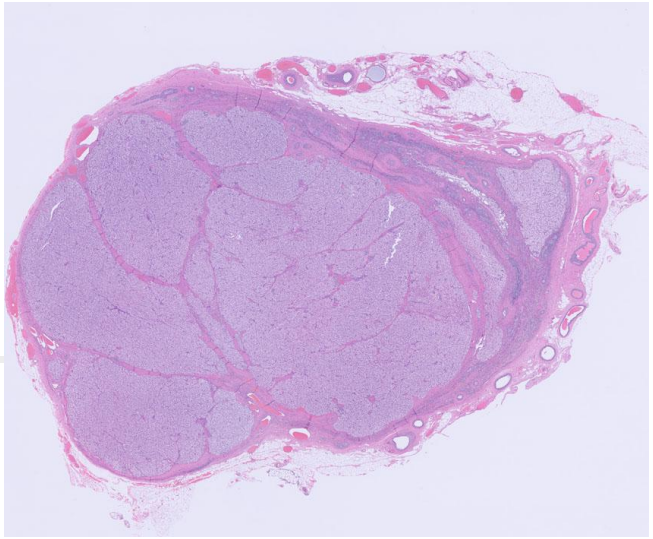
ScST – granulosa cell tumour

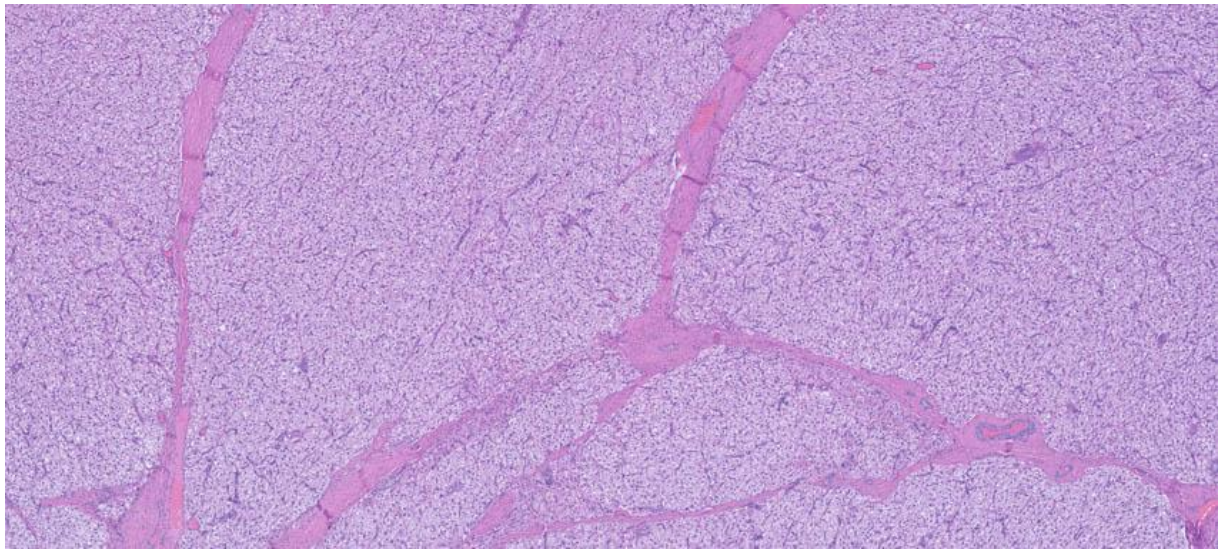


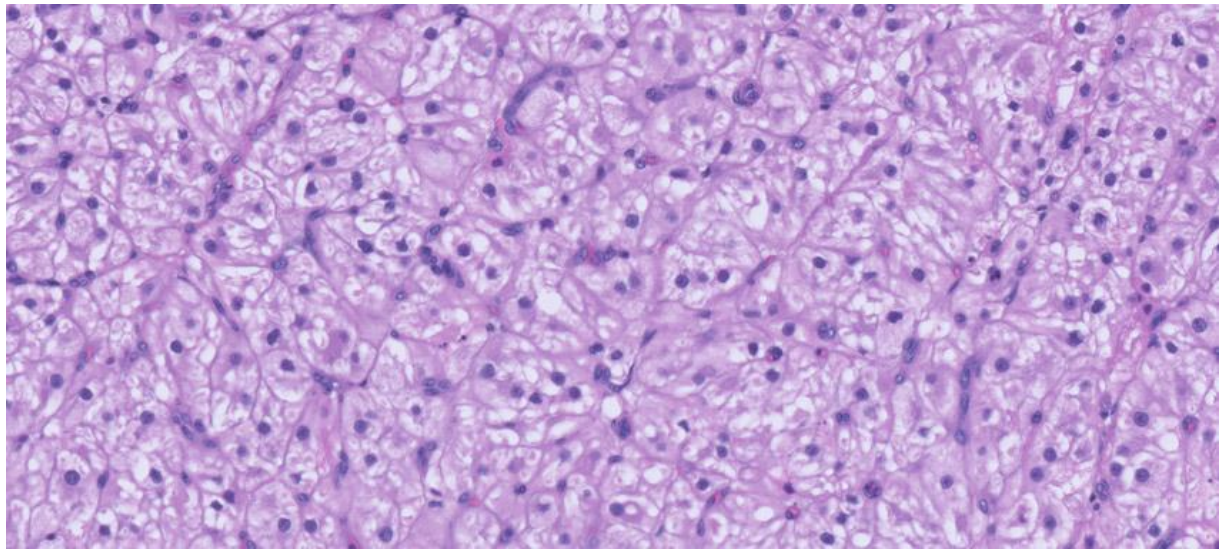
ScST – theca cell tumour



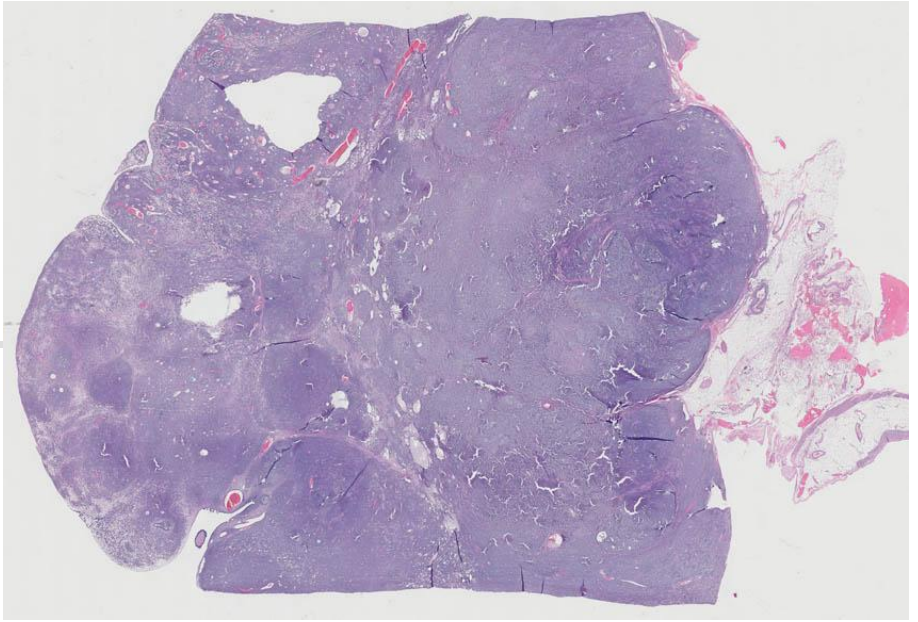
ScST – Luteoma

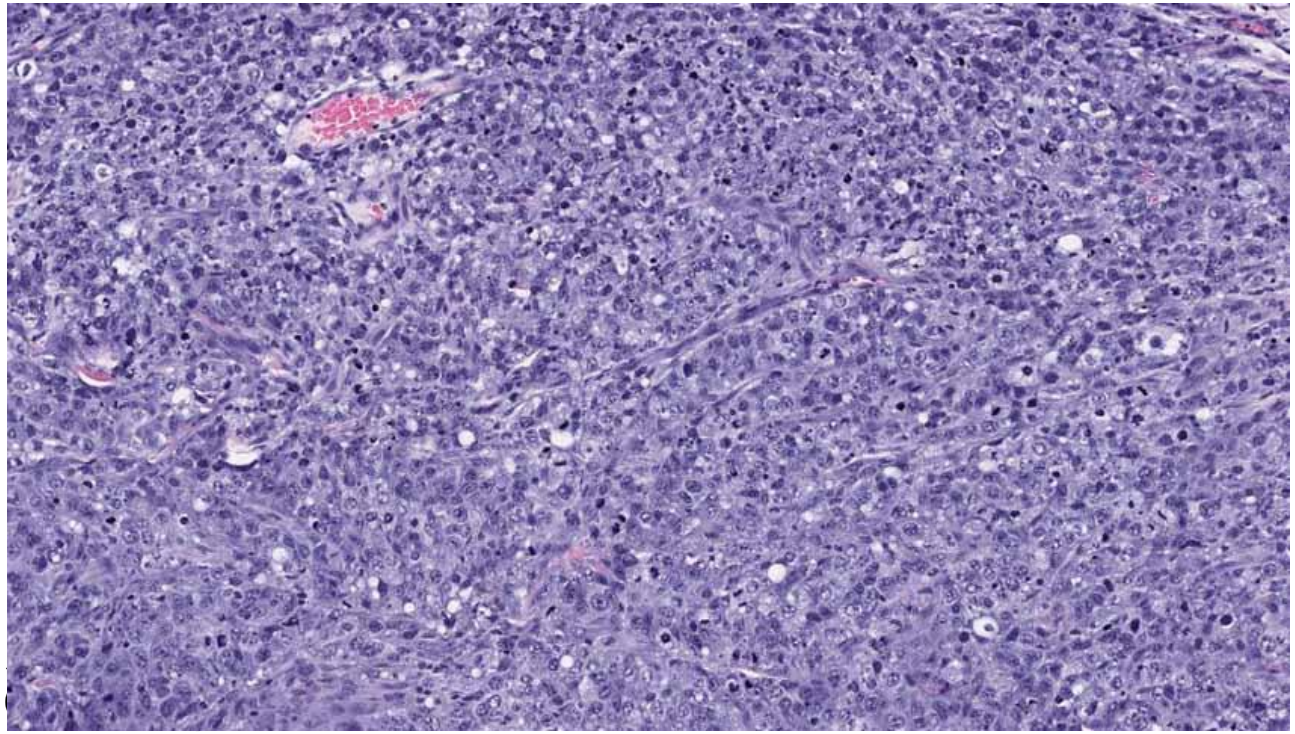


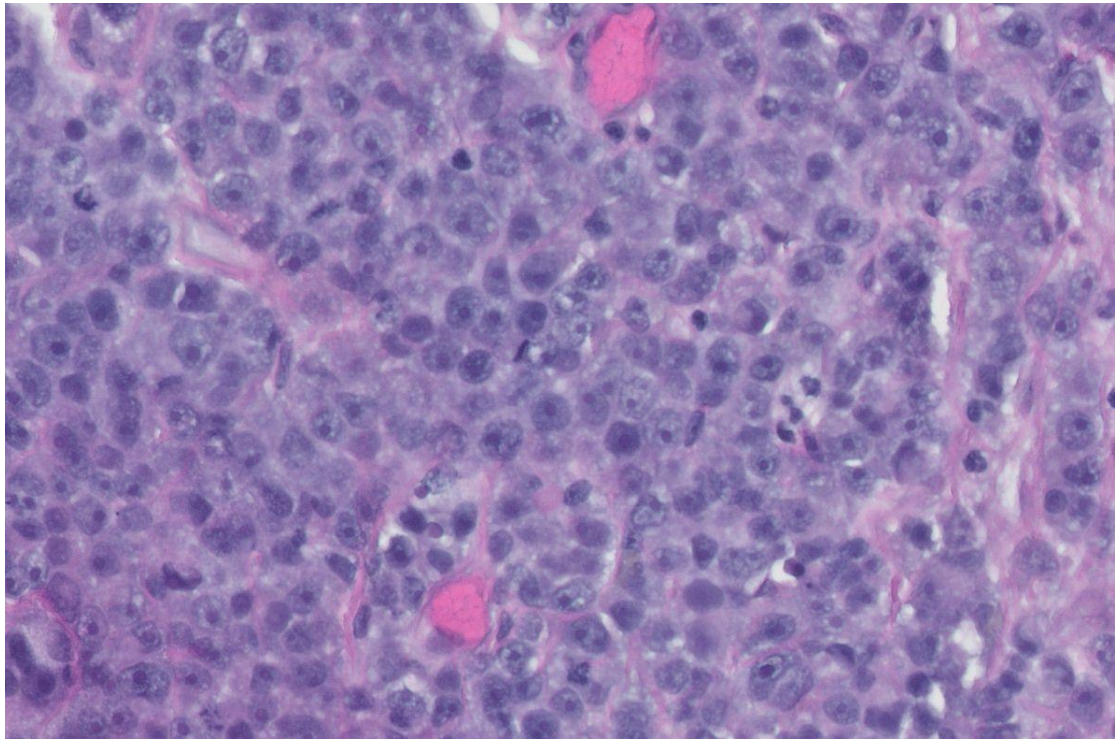




Germ cell tumor - dysgerminoma







Ovarian tumors – immunohistochemistry

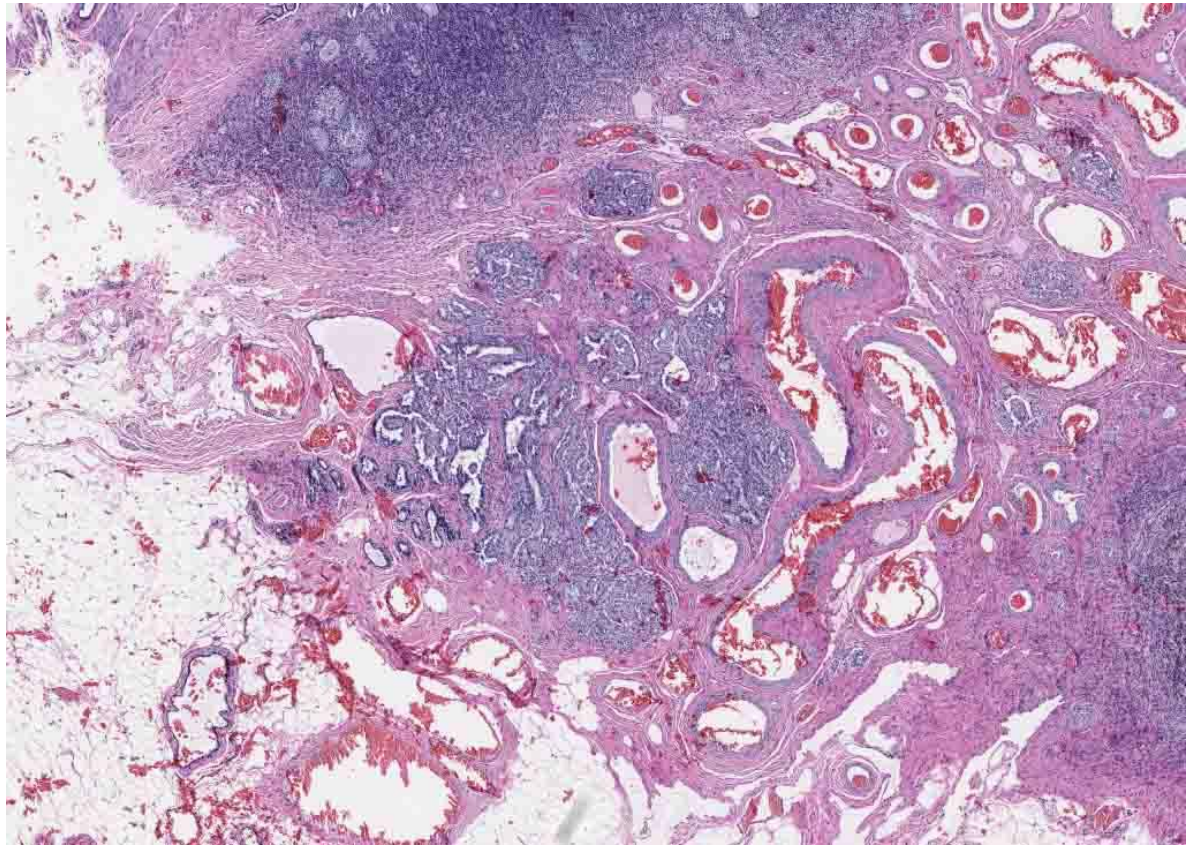
Tumor classification	Result
Epithelium	CK+, vimentin +, HBME-1+
Sex cord stromal	Inhibin-α, AMH
Germ cell	CD117+, PLAP+

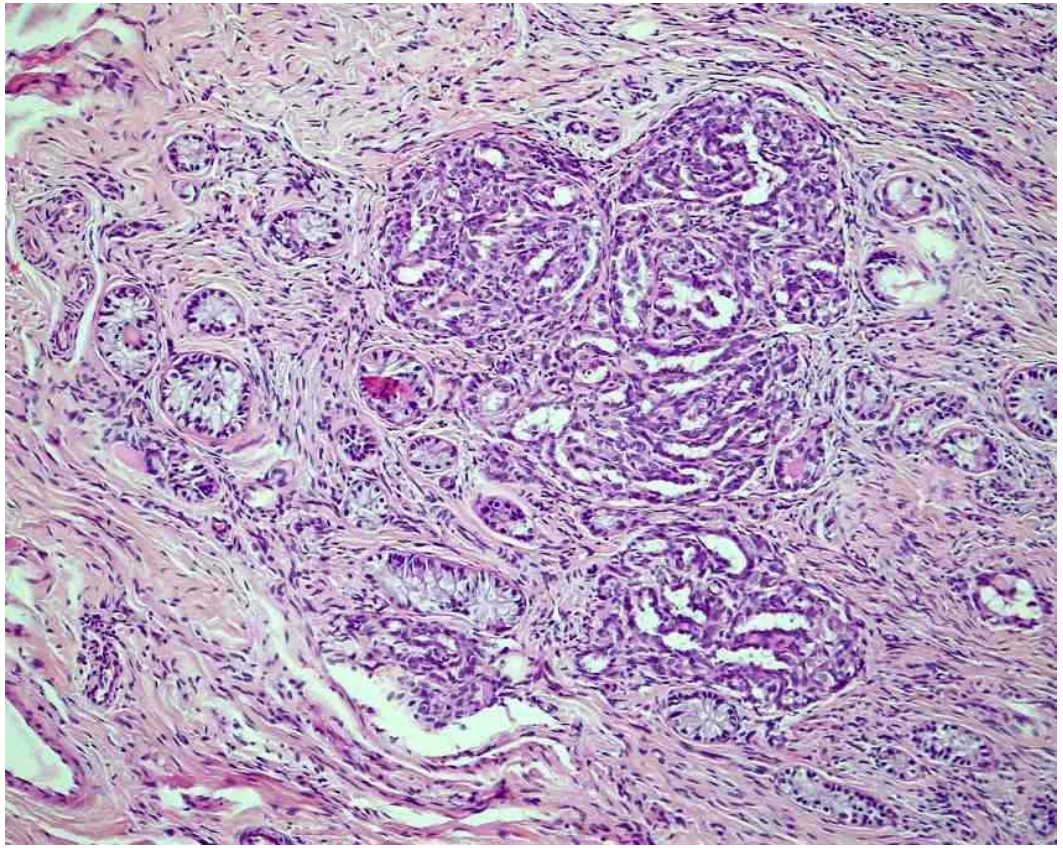
Epithelium

- Surface epithelium – mesothelium
- Rete ovarii

- Adenocarcinomas are in dogs mostly

Rete ovarii





Ovarian epithelial lesions

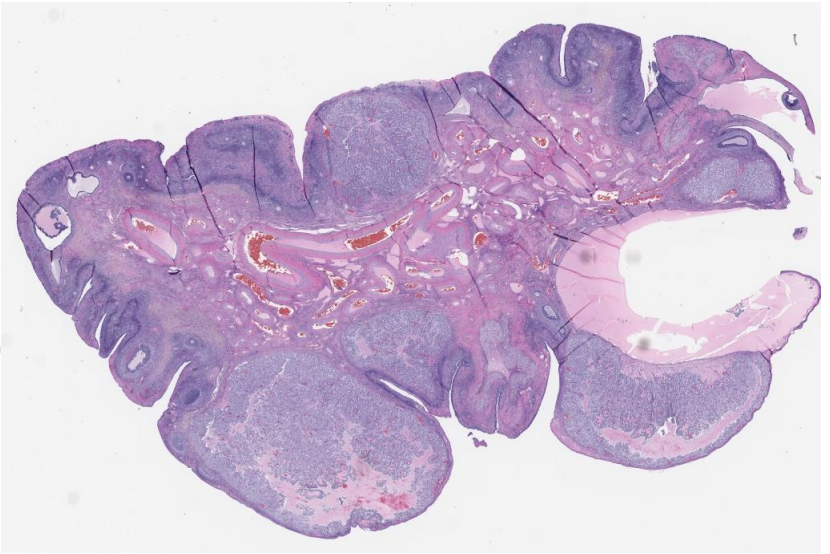
- Dog disease
  - Multiple in each ovary (thus bilateral)
  - Subsurface epithelial structures
  - Classification
    - Hyperplasia
    - Adenoma
      - Cystic and papillary
    - Adenocarcinoma
      - Papillary

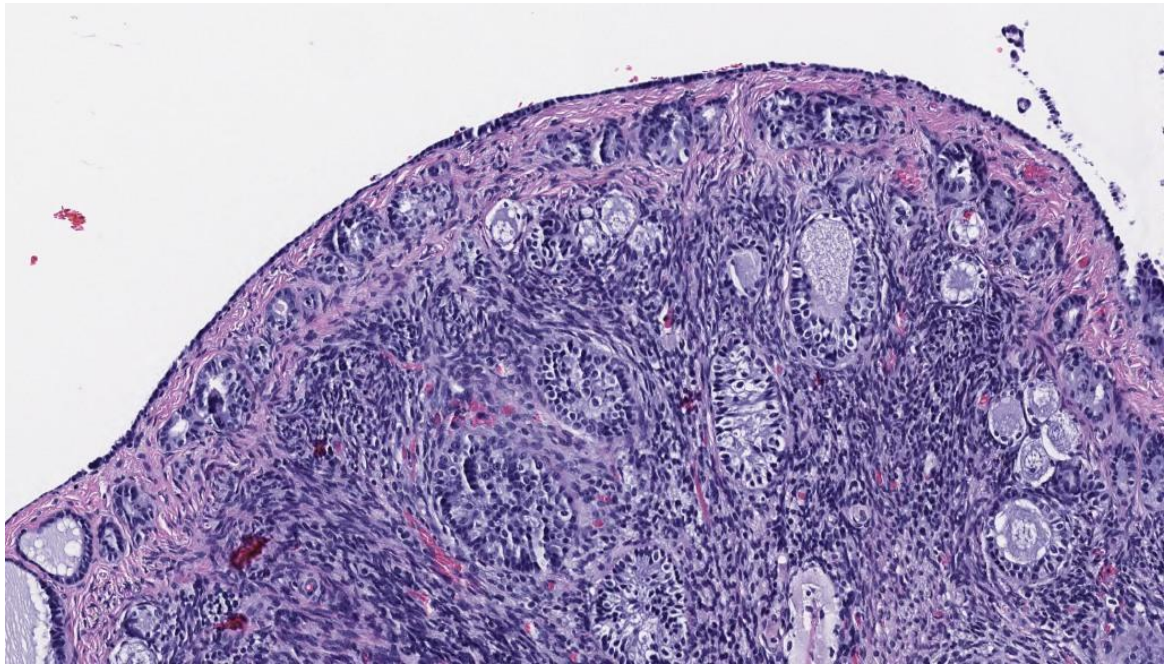
Classification

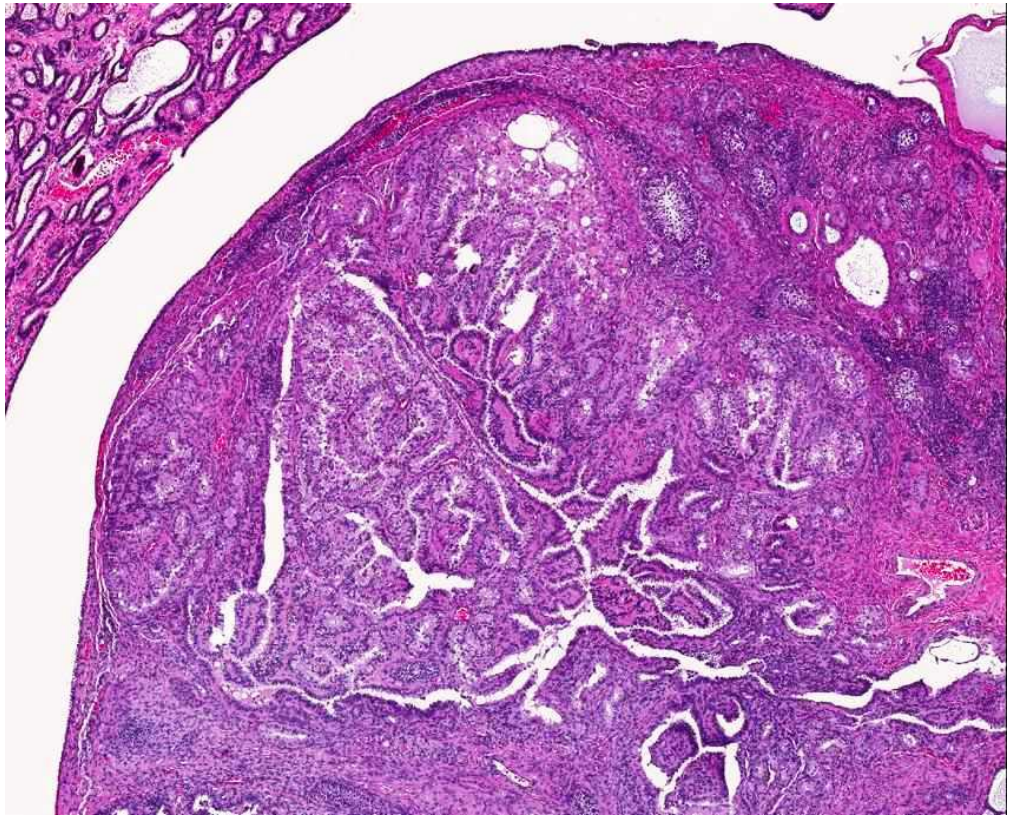
- Continuum; hyperplasia → adenoma → adenocarcinoma
- Not based on clinical outcome
  - Peritoneal carcinomatosis can lack mitotic figures, anisokaryosis, pleomorphism.
- Dolensek et al "effacement of the ovary, invasion of the ovarian stroma, or extension into the ovarian bursa. Mitotic counts ranged from 0 to 19 per HPF"

Dolenšek T, Knific T, Ramírez GA, Erles K, Mallon HE, Priestnall SL, Suárez-Bonnet A. Canine ovarian epithelial tumours: histopathological and immunohistochemical evaluation with proposed histopathological classification system. J Comp Pathol. 2024;212:42-50.

SES Neoplasia adenoma







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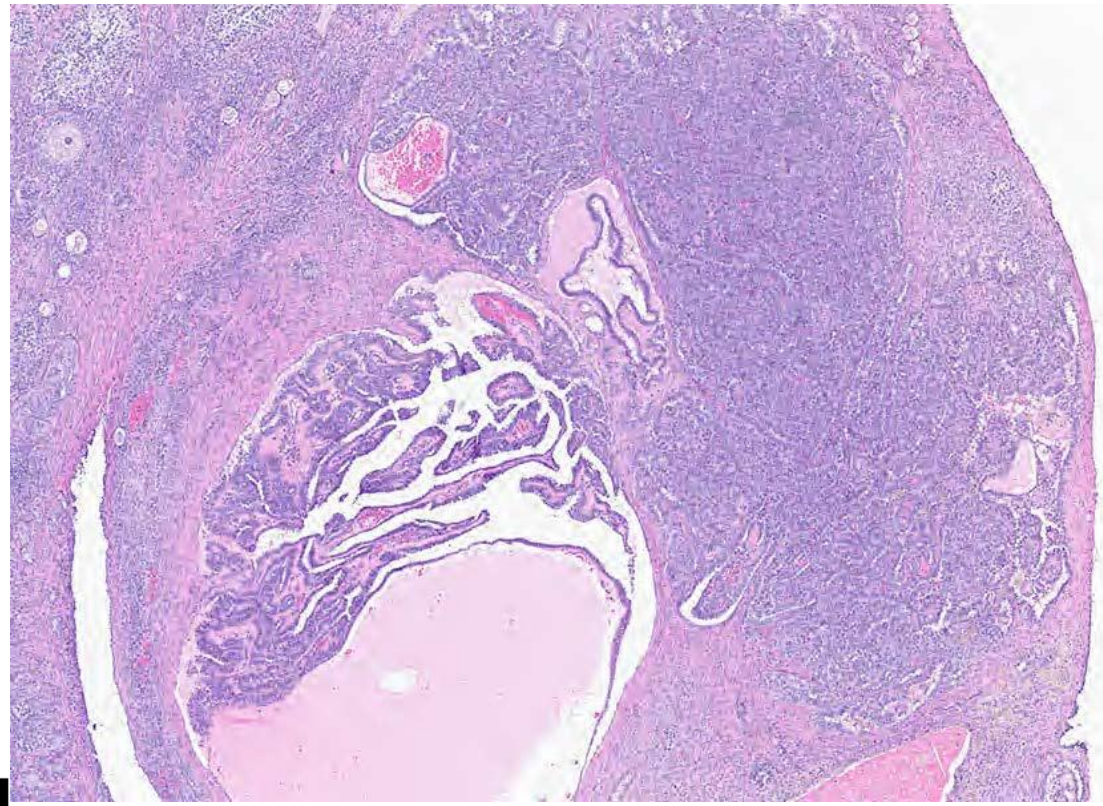
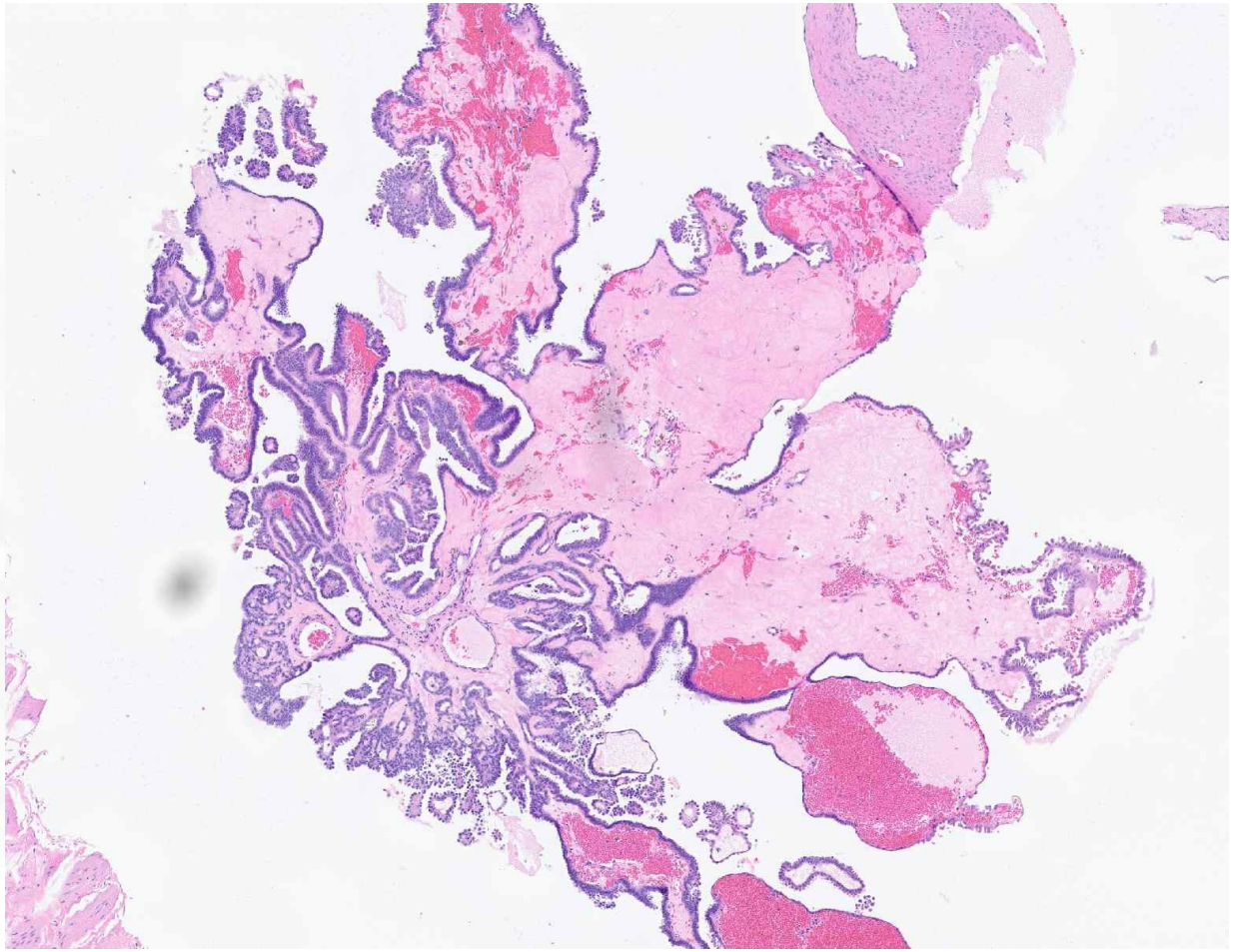
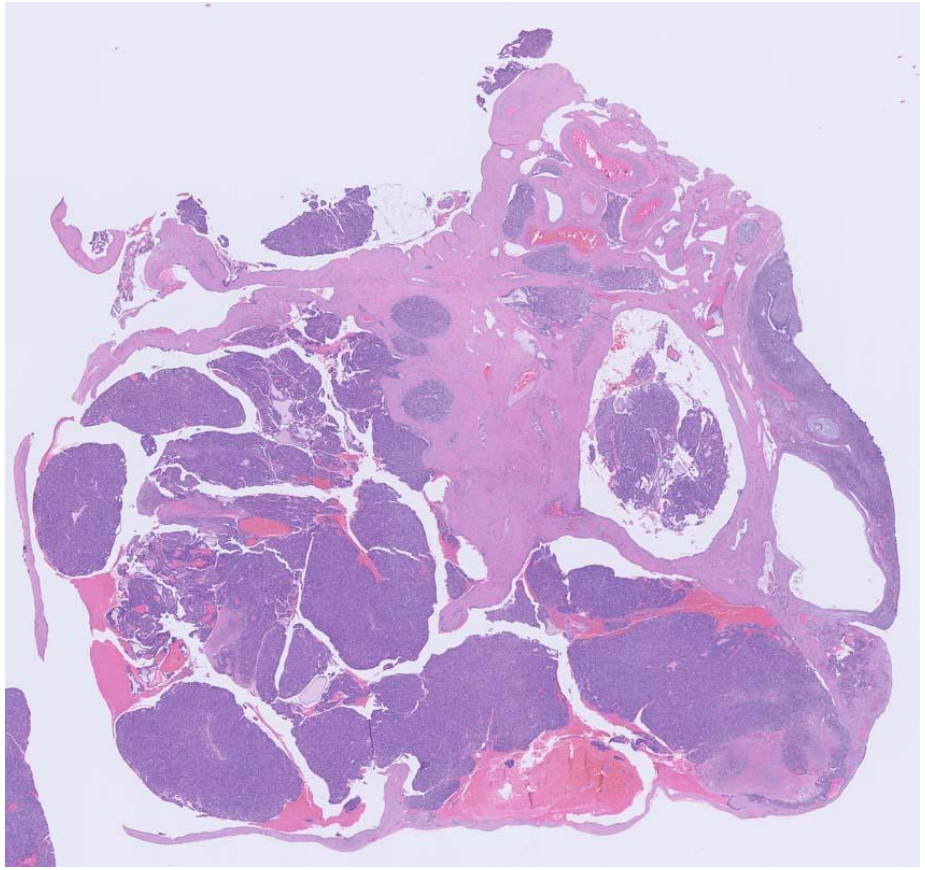
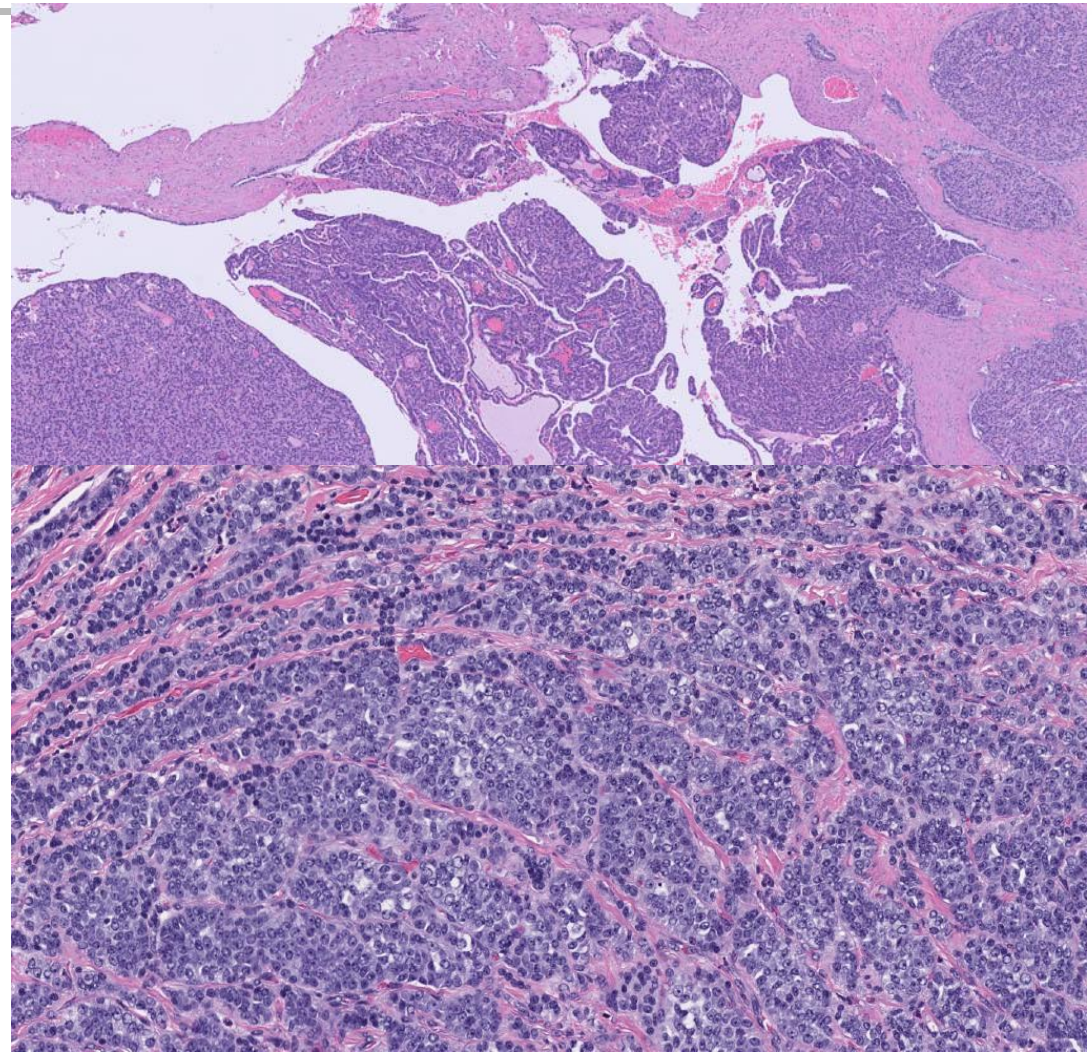
Ovarian adenoma





SES Neoplasia - adenocarcinoma





The end of neoplasia!

Disorders of Sexual Development

Chromosomes

Gonads

Tubular genitalia

External phenotype

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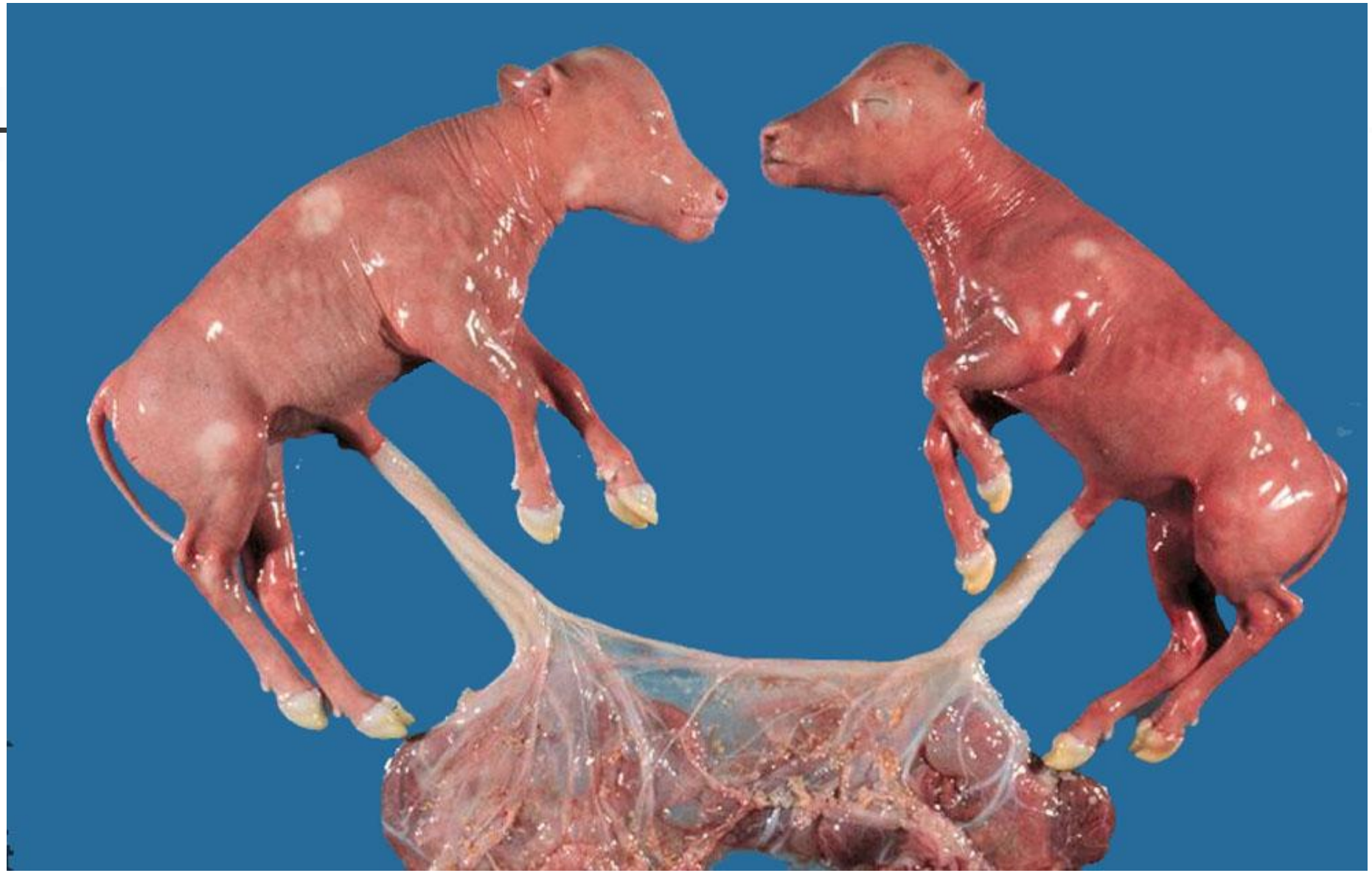
DSDs

- Chromosomes
  - Abnormal chromosomes – chromosomal DSD
  - XX DSD
  - XY DSD

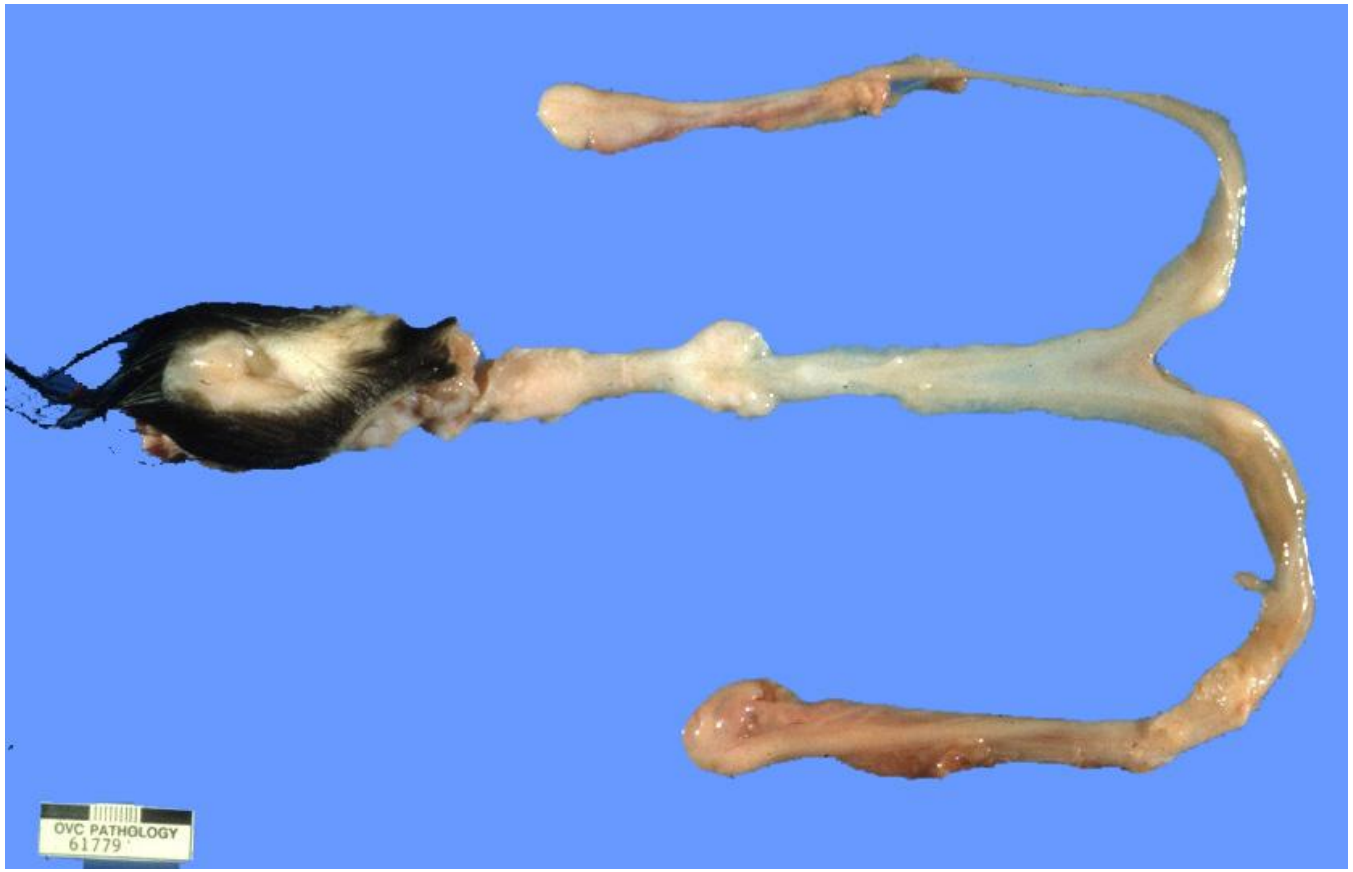
Chromosomal DSD

- Aneuploidy
  - X_ - Turner
  - XXX - Trisomy X
  - XXY - Klinefelter
- Mosaics and Chimera
  - Chimera (XX/XY) - cells of different zygotes fuse
    - Freemartin
  - Mosaic - nondisjunction in a single zygote
    - Calico or tortoise shell male cat
    - XXY, XXXY, XXXXY



Bovine: Chimerism; freemartin

Ovary
Epididymis
Segmental aplasia of uterus
Vesicular gland*
Bulbourethral gland
Vagina and vulva
Prominent clitoris



Freemartinism

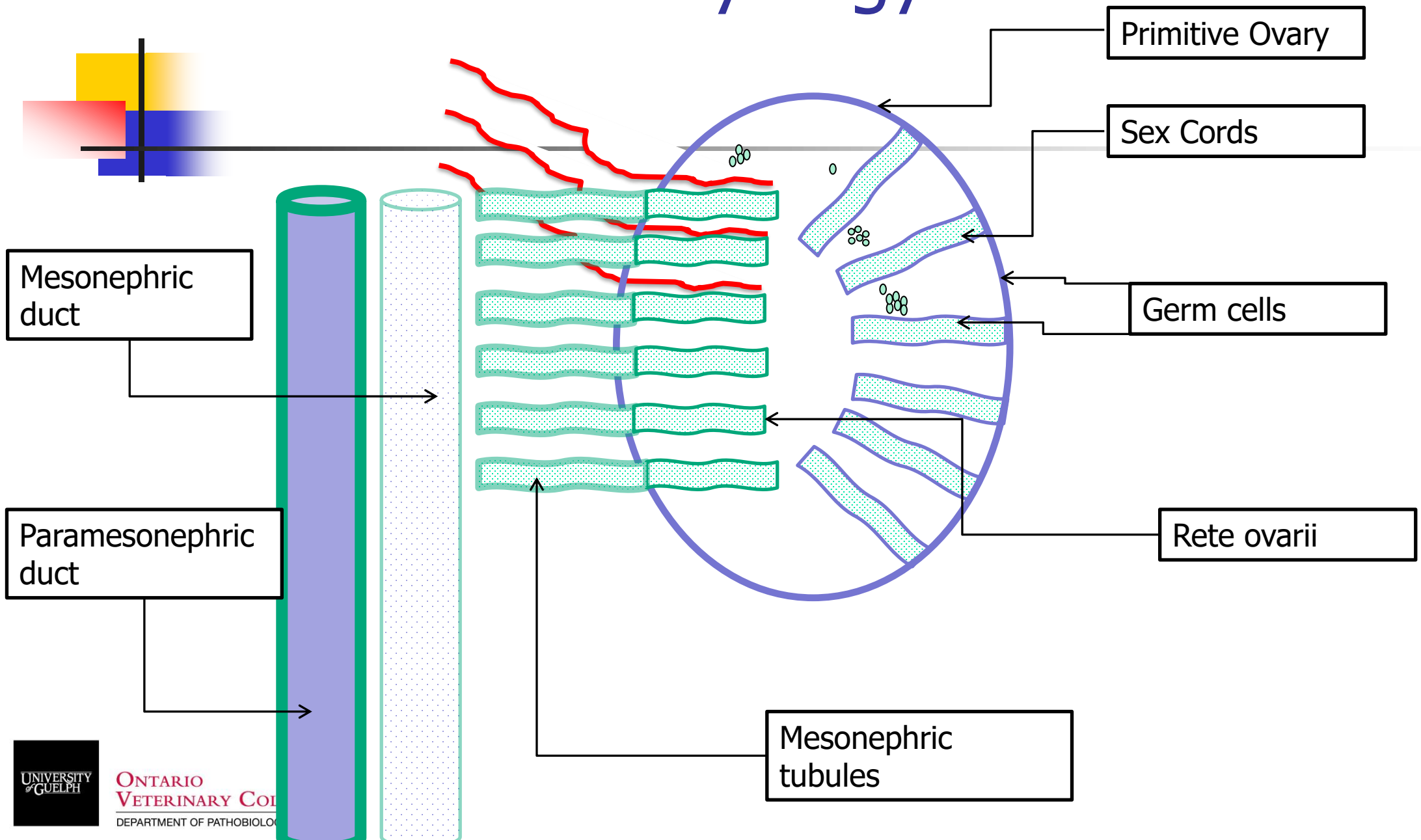
- Species?
- Twin and single born freemartins
- XX/XY chimera
- Ovary but paramesonephric ducts don’t develop.
- Masculinized external genitalia

A.M. Padula (2005) The freemartin syndrome: an update. Anim Reprod Sci (2005) 87: 93–109

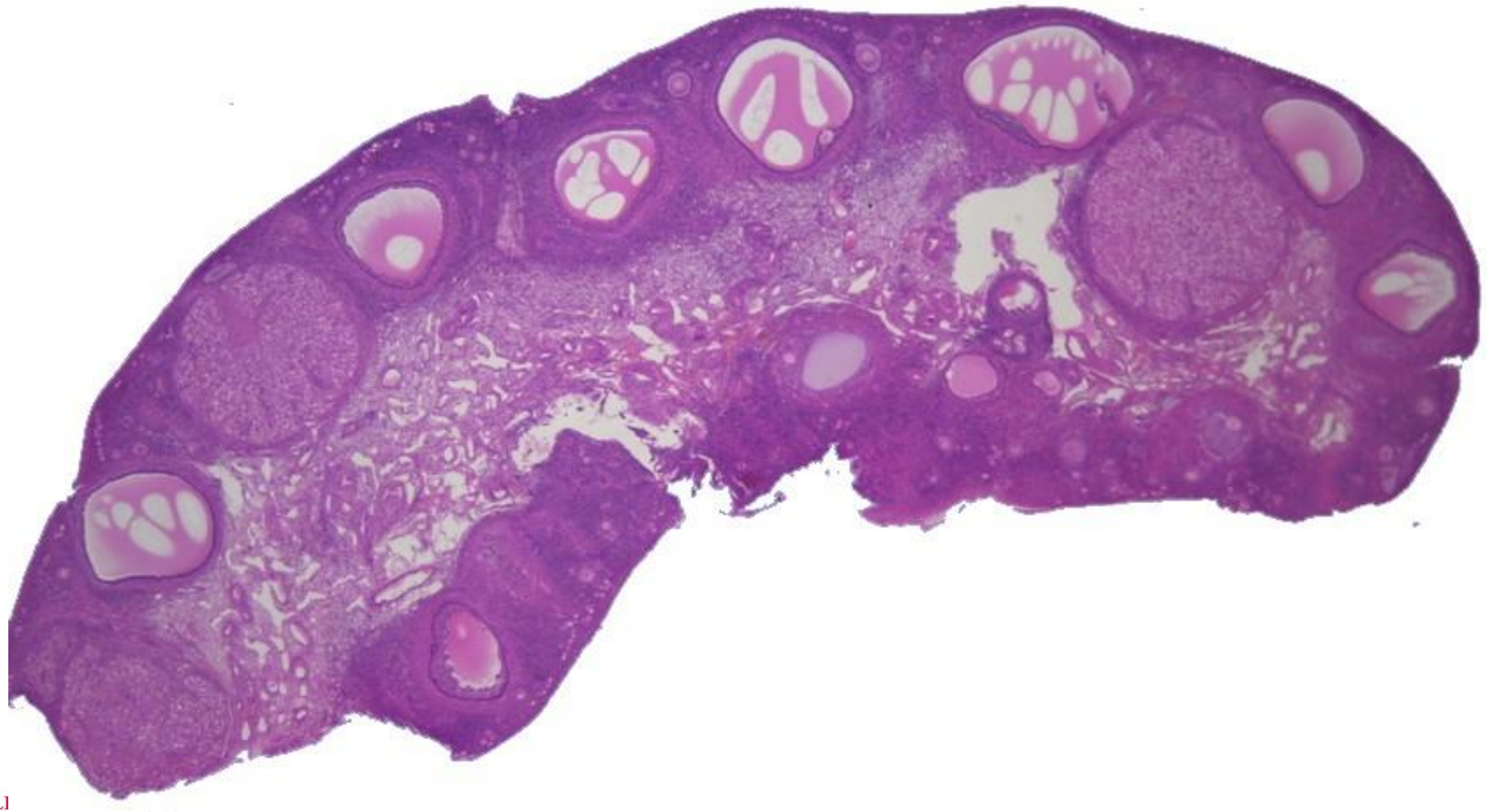
XX DSD

- Normal gonads
  - XX ovarian DSD
- Abnormal gonads
  - XX Testicular DSD
  - XX Ovotesticular DSD
  - XX Gonadal dysgenesis DSD

Normal female embryology



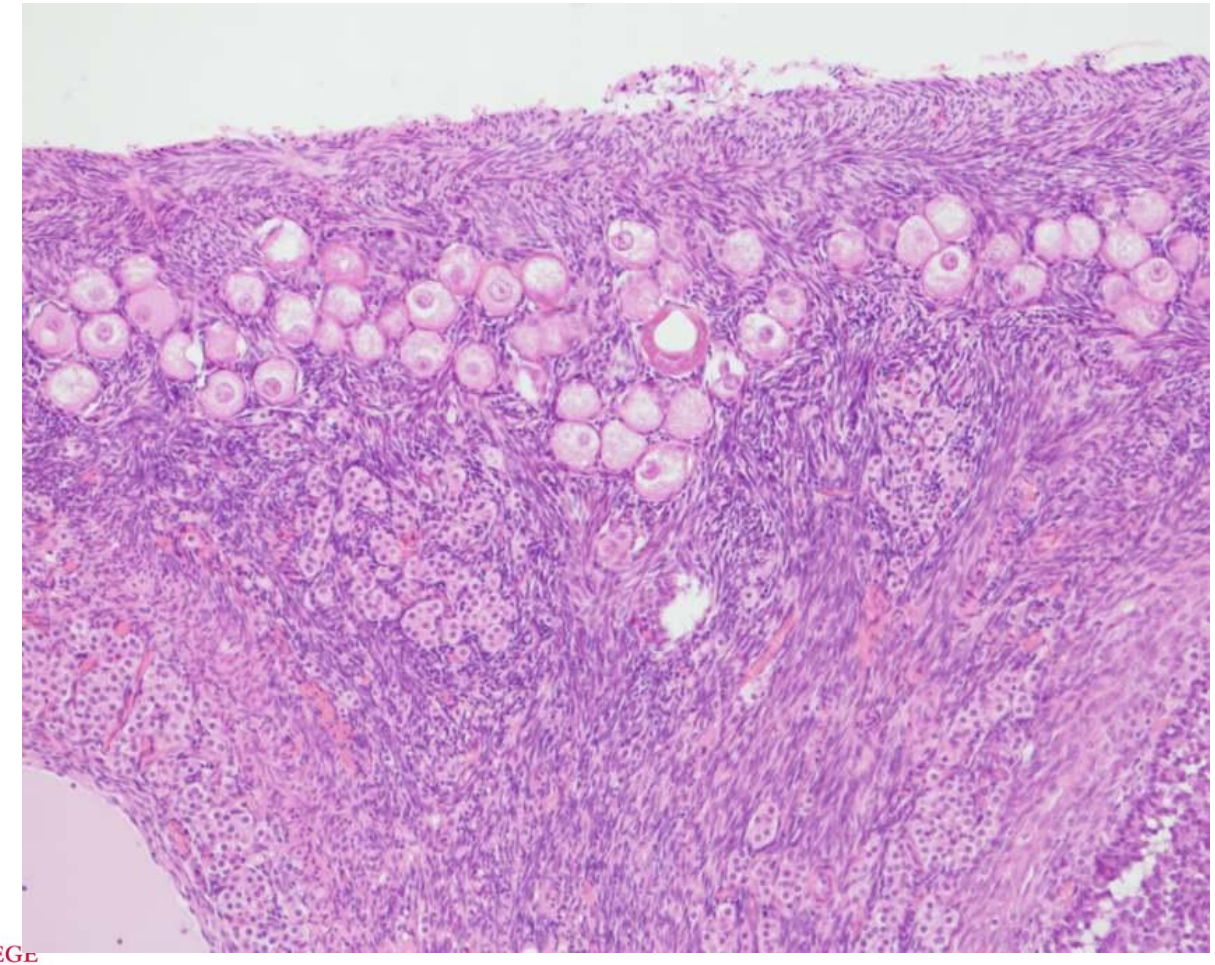
Feline ovary

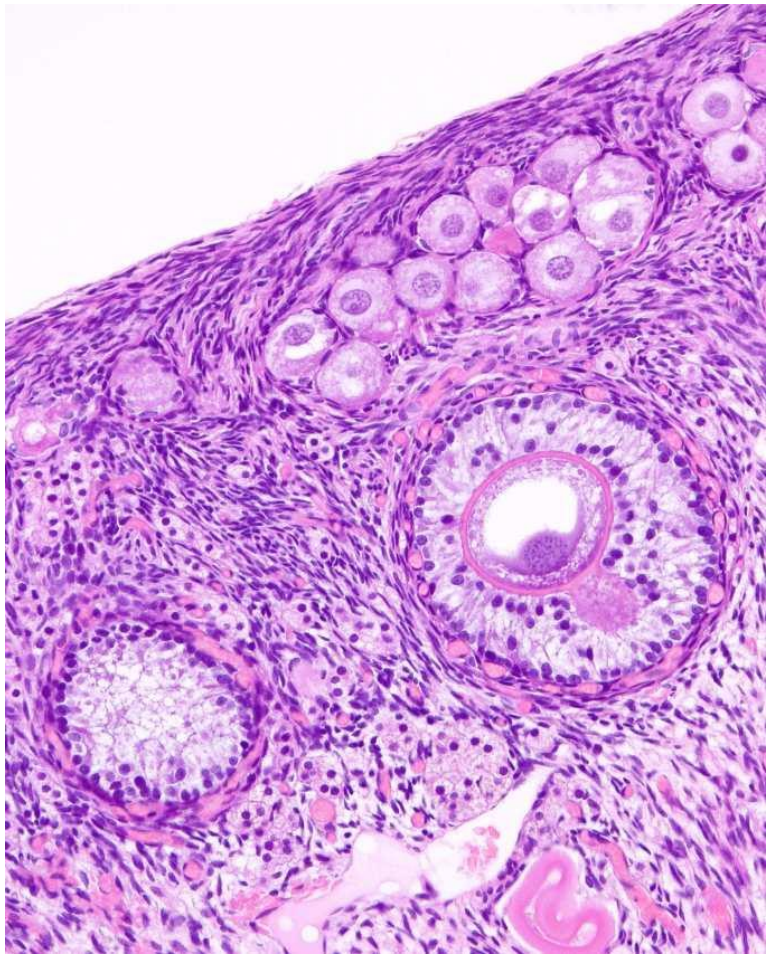


Normal prepubertal feline ovary

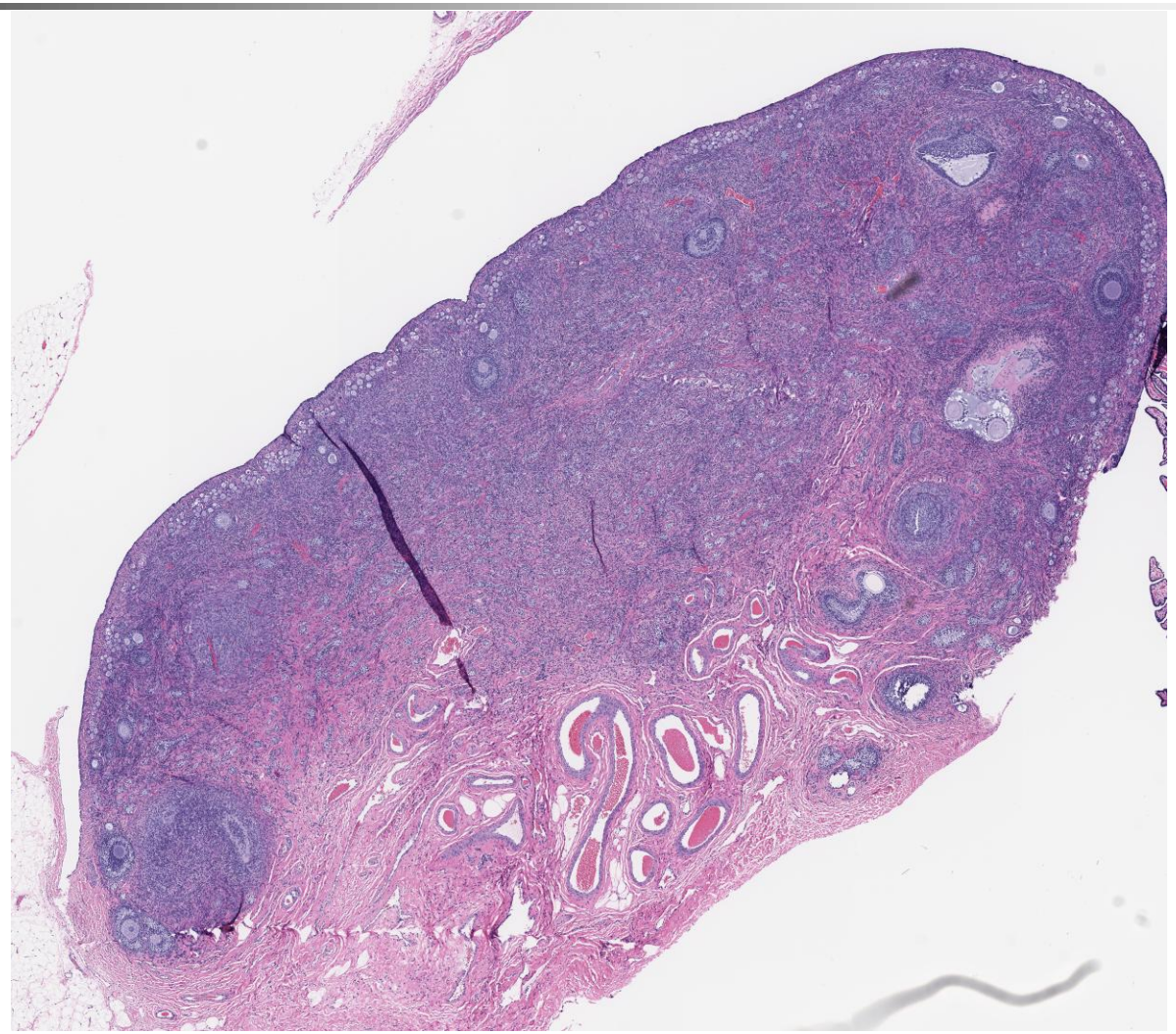
Follicles

Primordial
Primary
Secondary
Tertiary

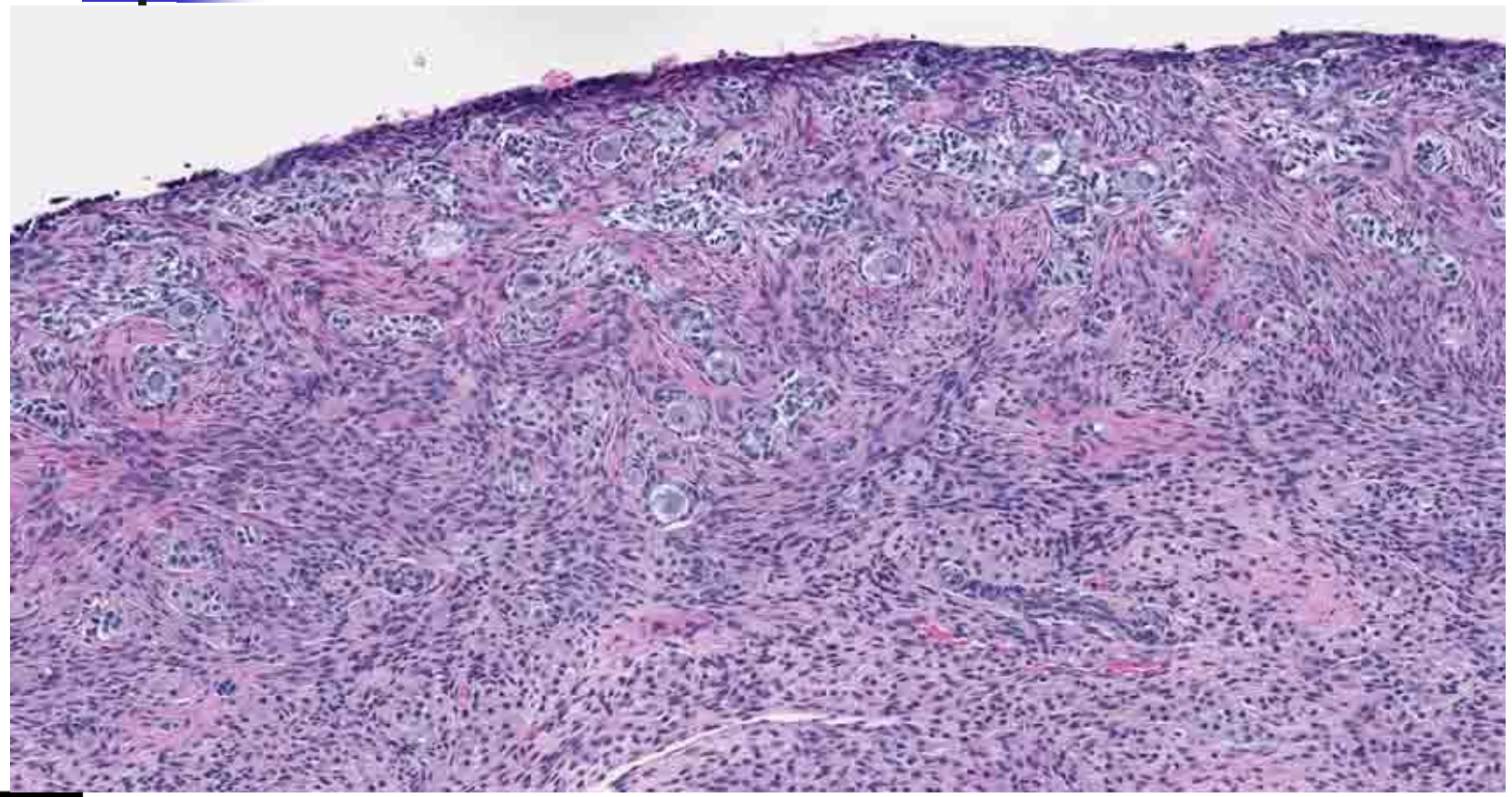


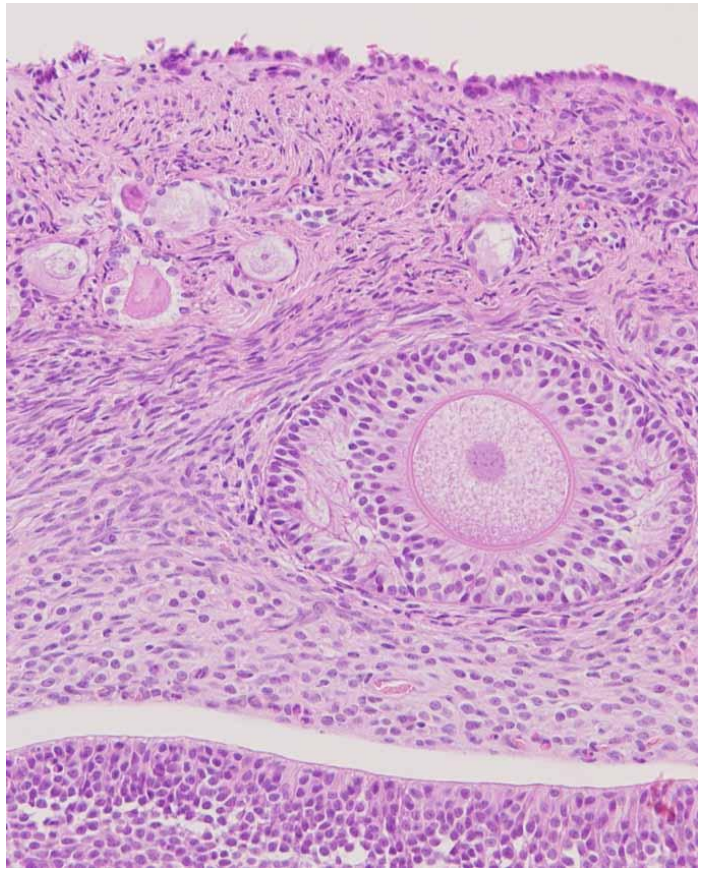


Canine ovary



Normal prepubertal canine ovary





Can you predict sex chromosome from gonad, tubular genitalia or phenotype

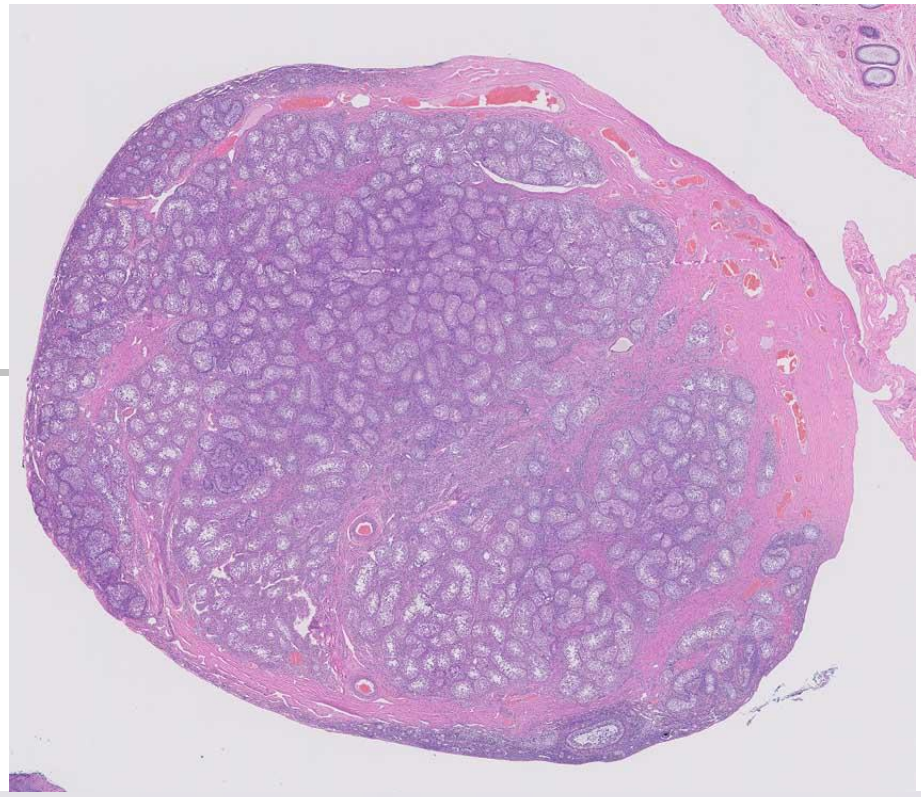
Answer – NO

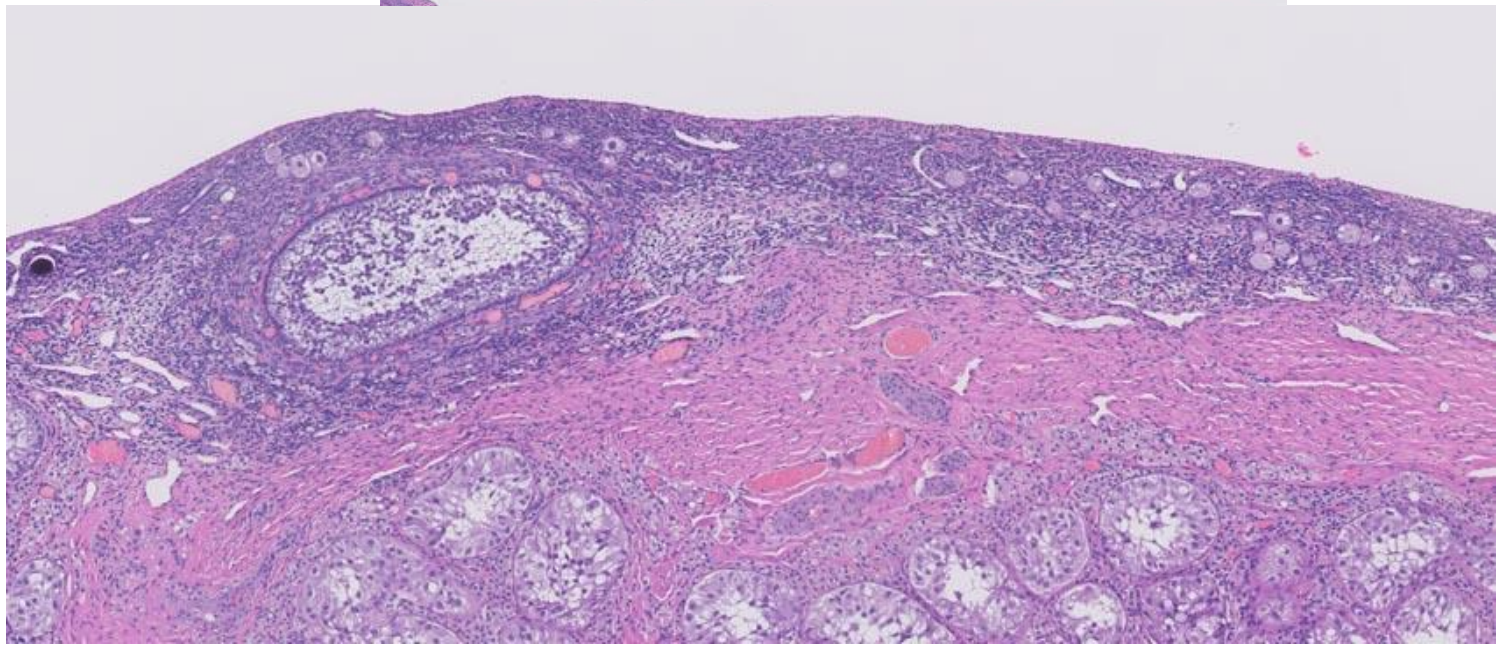
Pathologists are the only ones who can tell the gonadal type!

Disorders of Sexual Development

Gonads

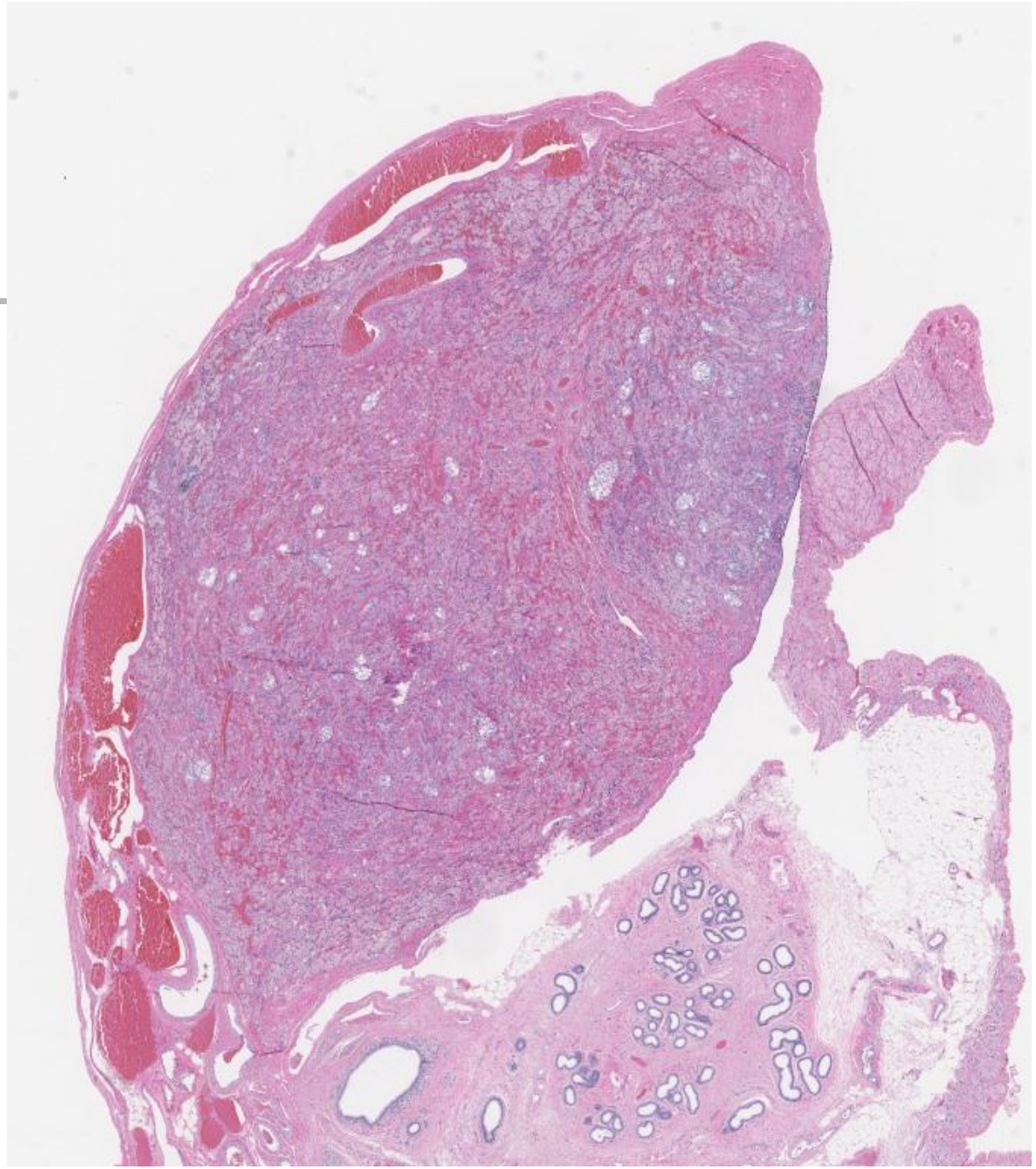
- Testes
- Ovaries
- **Ovotestes**
- Dysgenesis



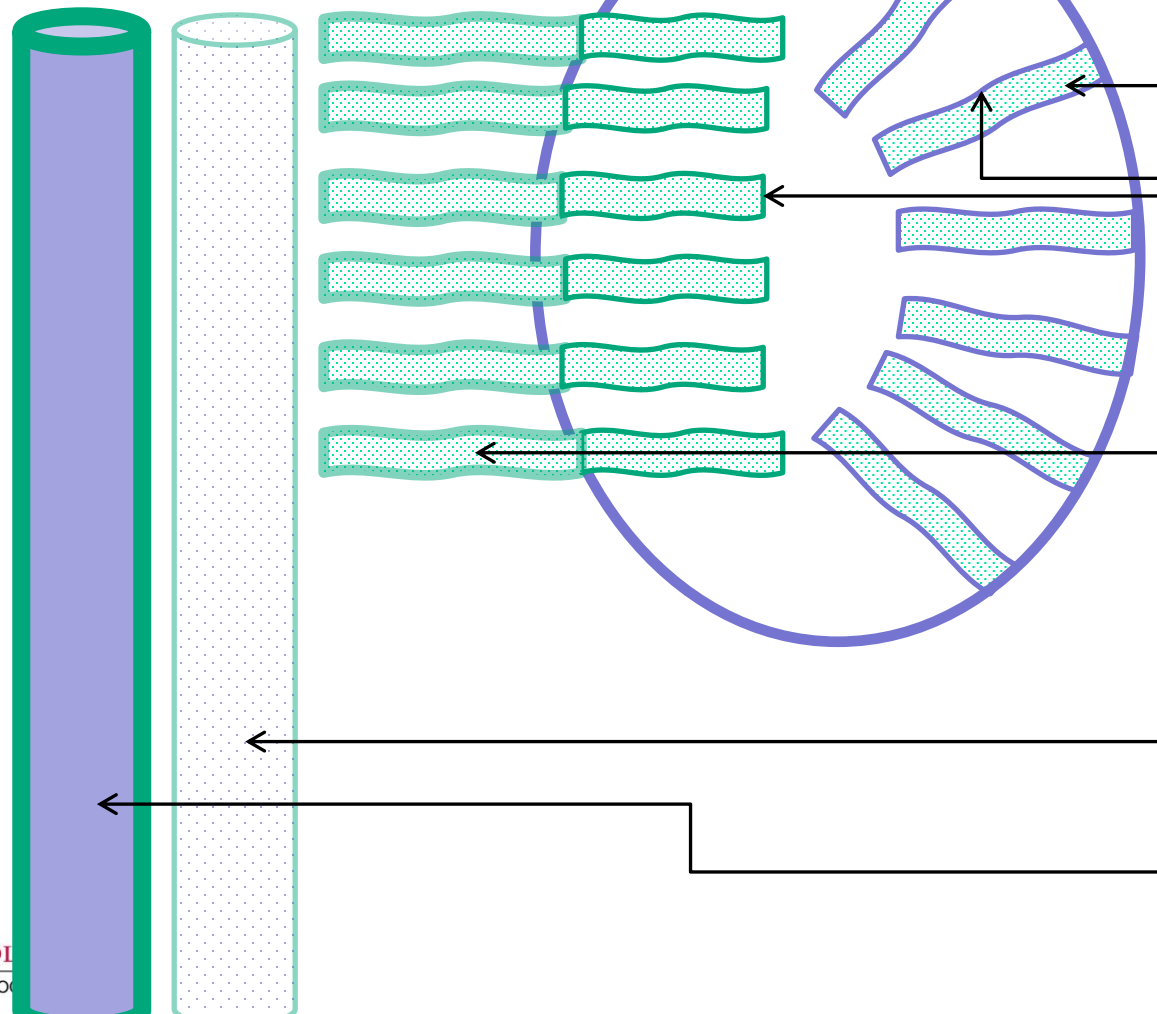


Gonadal type

- Testes
- Ovaries
- Ovotestes
- Dysgenesis



Normal female embryology



Primitive Ovary

Sex Cords

Germ cells

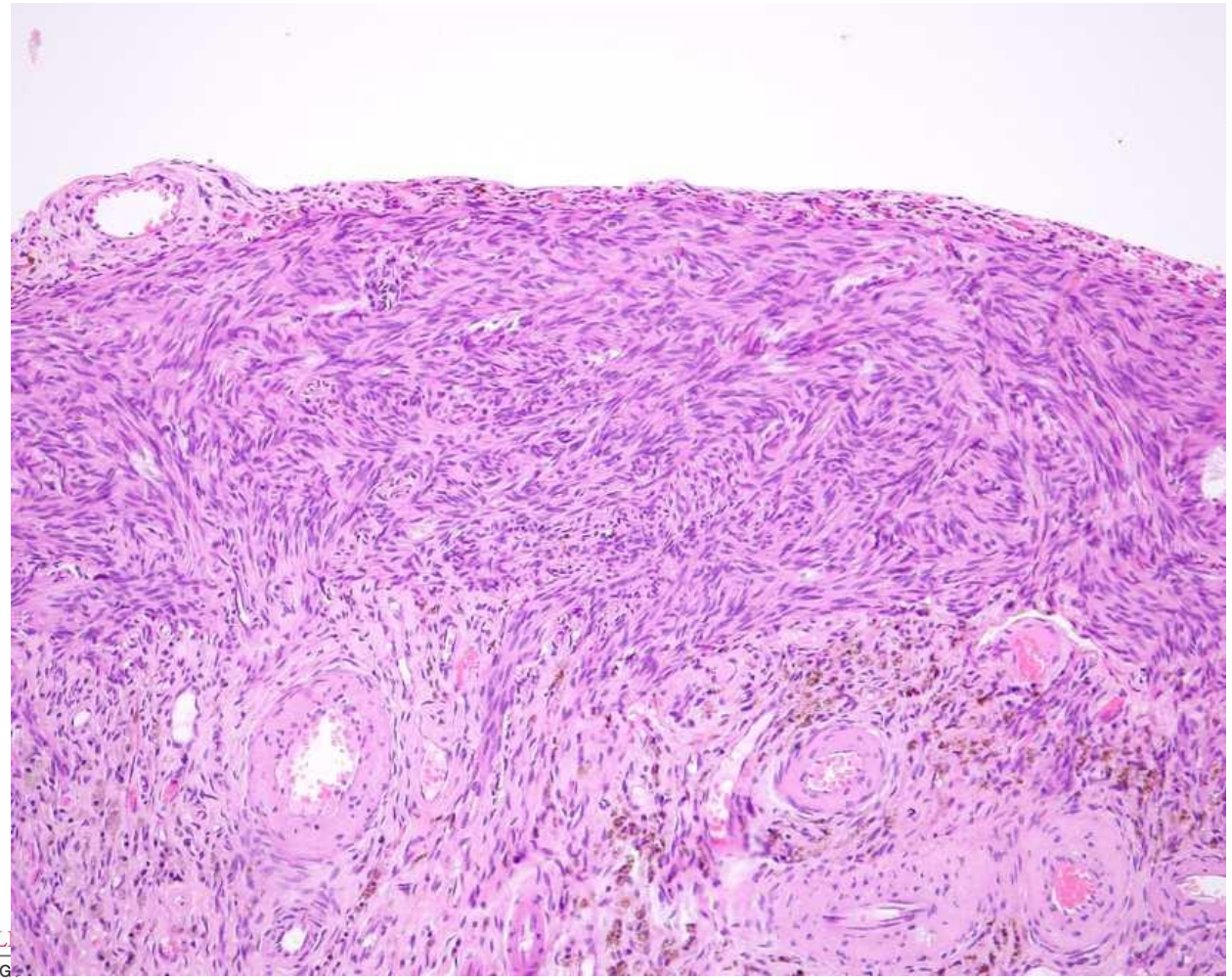
Rete ovarii

Mesonephric tubules

Mesonephric duct

Paramesonephric duct

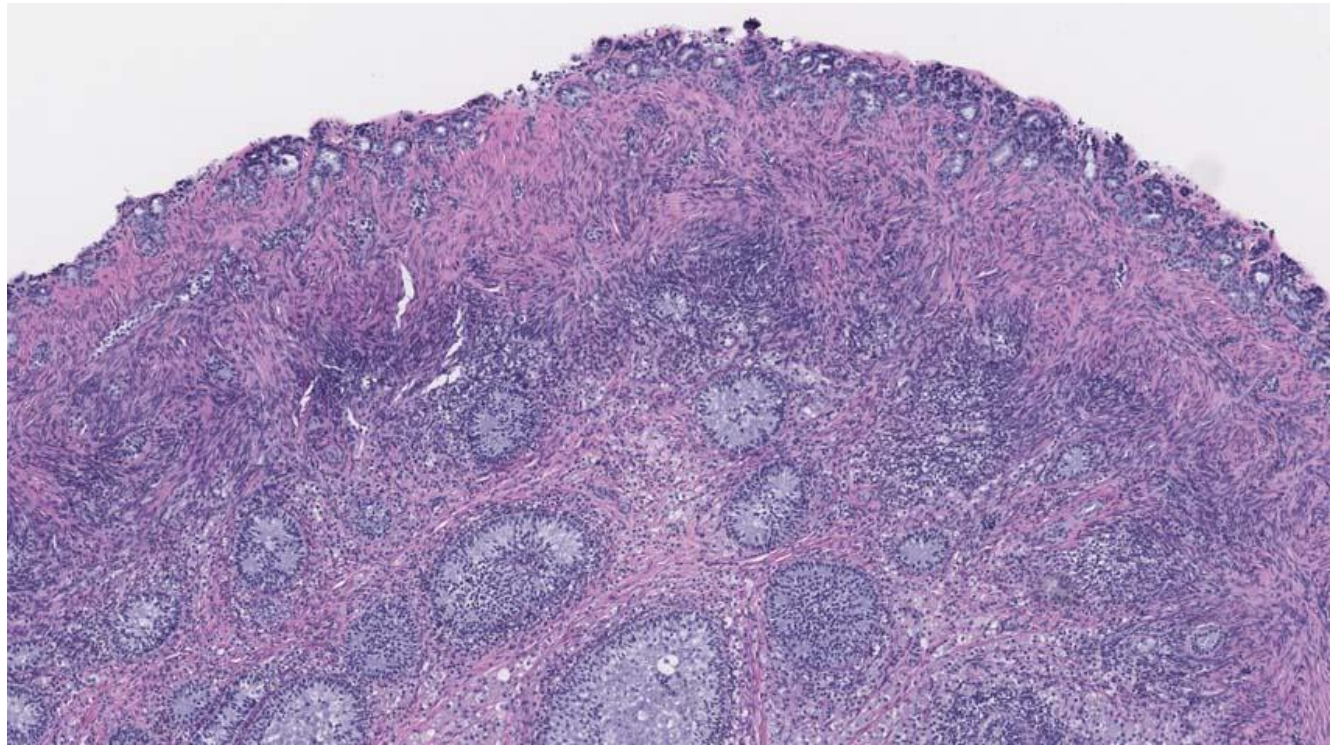
Feline ovarian hypoplasia/dysgenesis



Either

- No germ cell migration
- Germ cells died
- No sex cord development
- Sex cords died

Canine ovarian hypoplasia



- Can you tell if this animal has
  - a chromosomal abnormality causing germ cells to undergo cell death

  or
  - germ cells failed to migrate

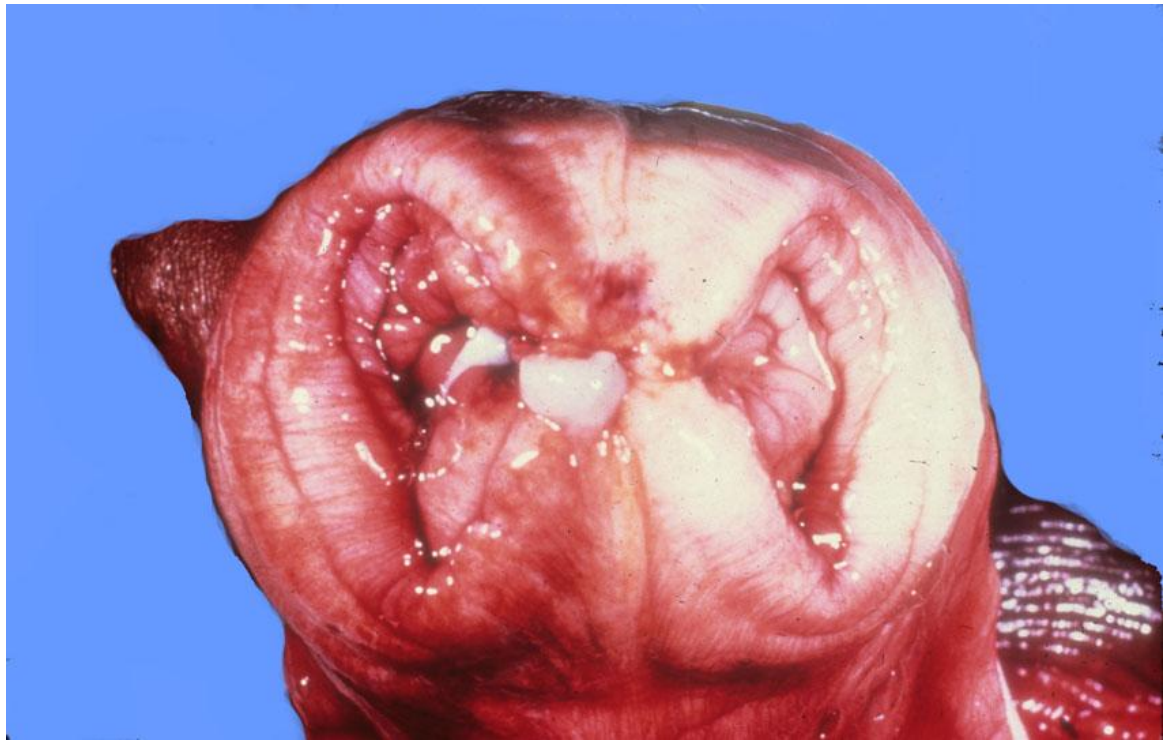
Epididymis in a female

- Must have sustentacular cells/androgens/luminocentric factors

- Look for ovotestis
- Freemartin?

Segmental aplasia (of paramesonephric duct)

Duplications

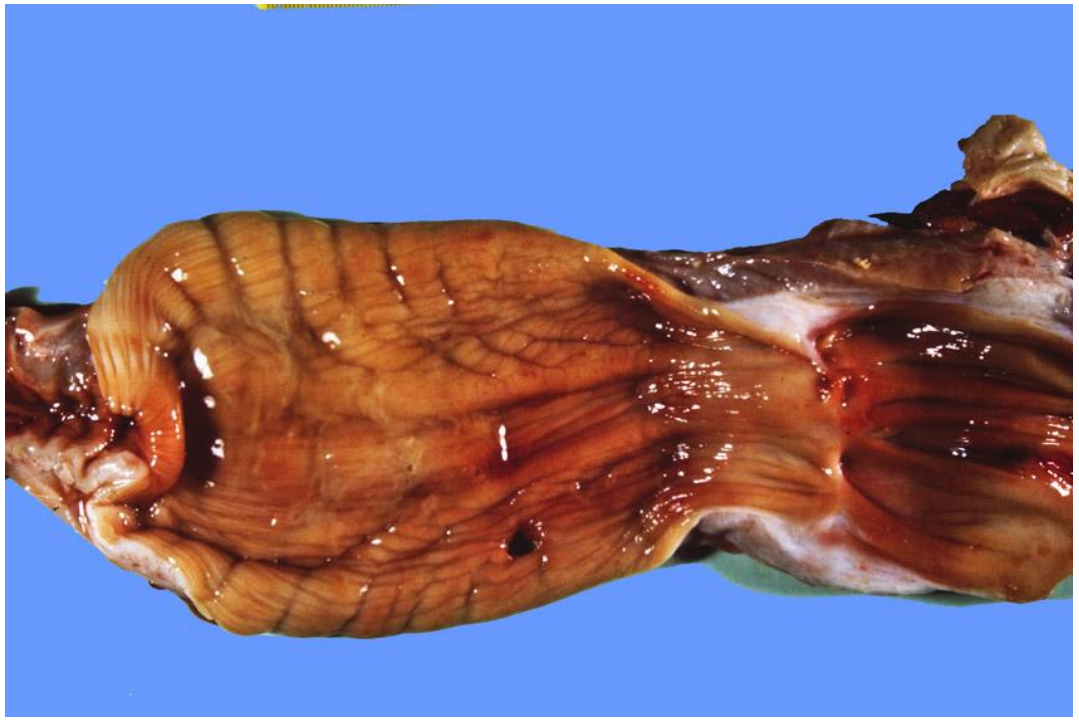


Bovine cervix
Photograph complements of
Dr Philip Ladds

Canine uterus

Incomplete junctions

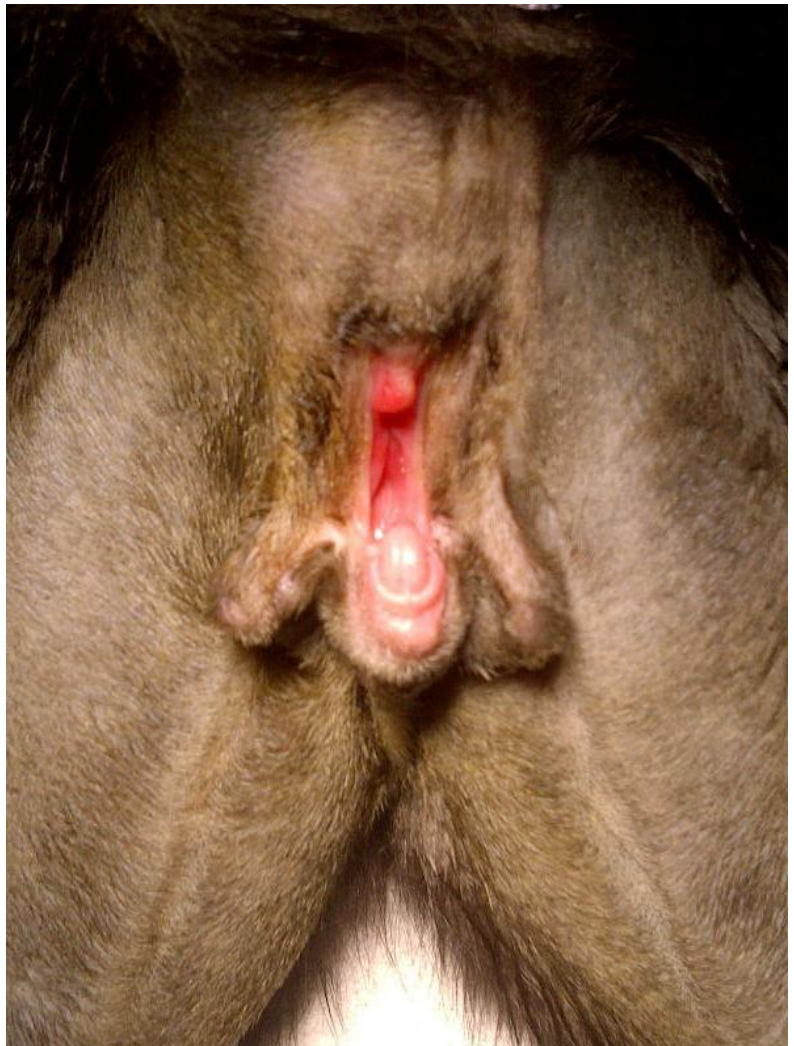
- Persistent hymen
- Bands



Photograph complements of
Dr Philip Ladds

External genitalia ambiguous or feminized

- Require testosterone/androgen to form male external genitalia
- No hormone or no receptor means female or feminization
- Enlarged clitoris
- Hypospadias
- Bifid scrotum



Photograph complements of Dr Jonathan Mittleman

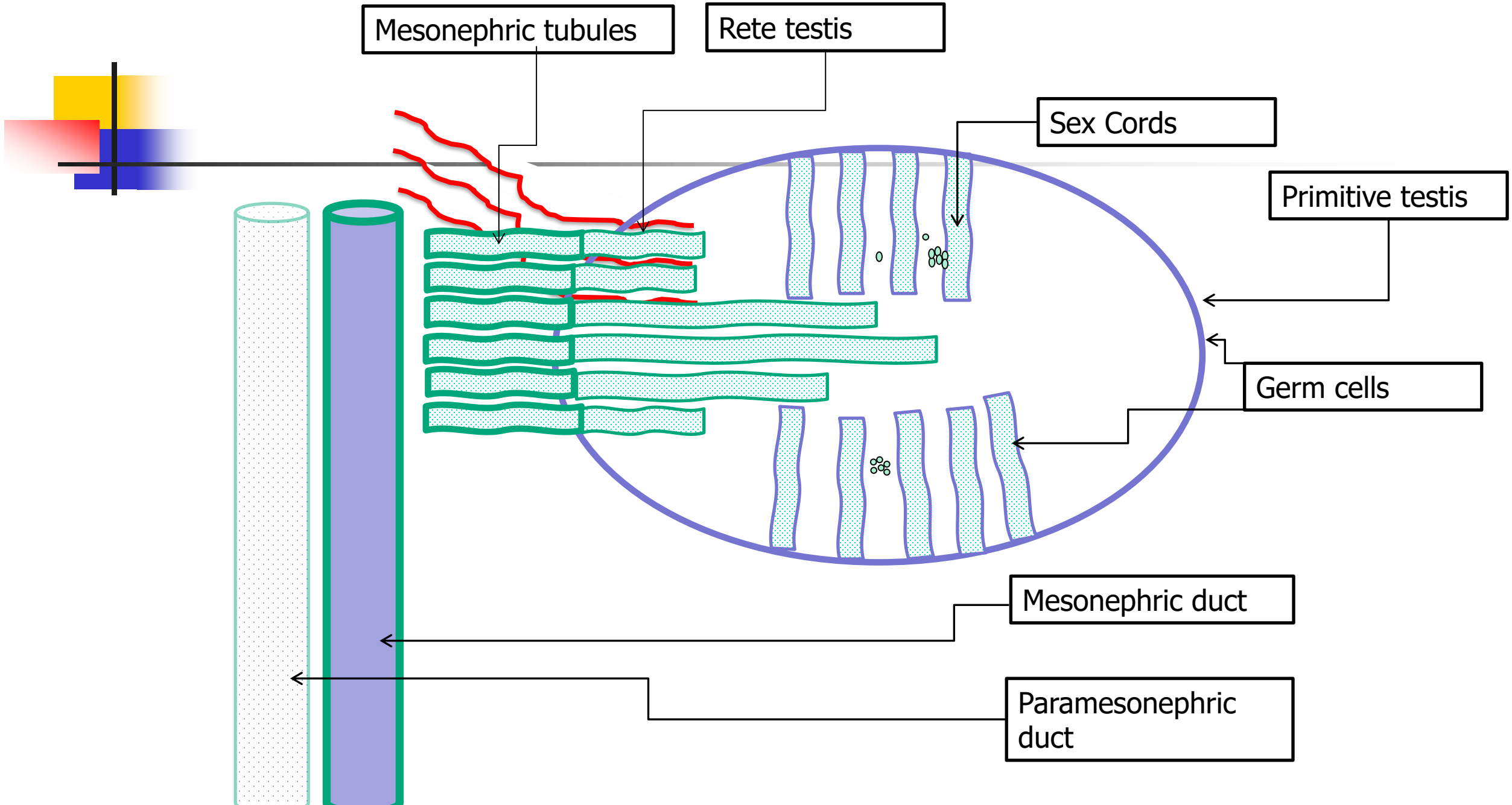
XY DSD

- Normal gonads
  - XY testicular DSD
- Abnormal gonads
  - XY ovotesticular DSD
  - XY ovarian DSD
  - XY gonadal dysgenesis DSD

Normal Male development

- Sex Chromosome – XY
  - *SRY* gene activates sustentacular (Sertoli) cell development
  - *SRY* gene inhibits female genes
- Gonad – Testis
  - Germ cell migration - spermatogonia
  - Vascular migration - interstitial endocrine cells
- Tubular genitalia - from mesonephric duct
  - Epididymis, deferent duct, ampulla, vesicular gland –
  - Sustentacular cells - Anti-Müllerian Hormone (AMH) inhibits paramesonephric duct development (uterus)
  - Sustentacular cells – luminocentric factors
- Urogenital sinus - Phenotype – penis and scrotum
  - Vascular pericytes - interstitial endocrine cells
  - Testosterone
  - Male external genitalia

Embryology of the Male

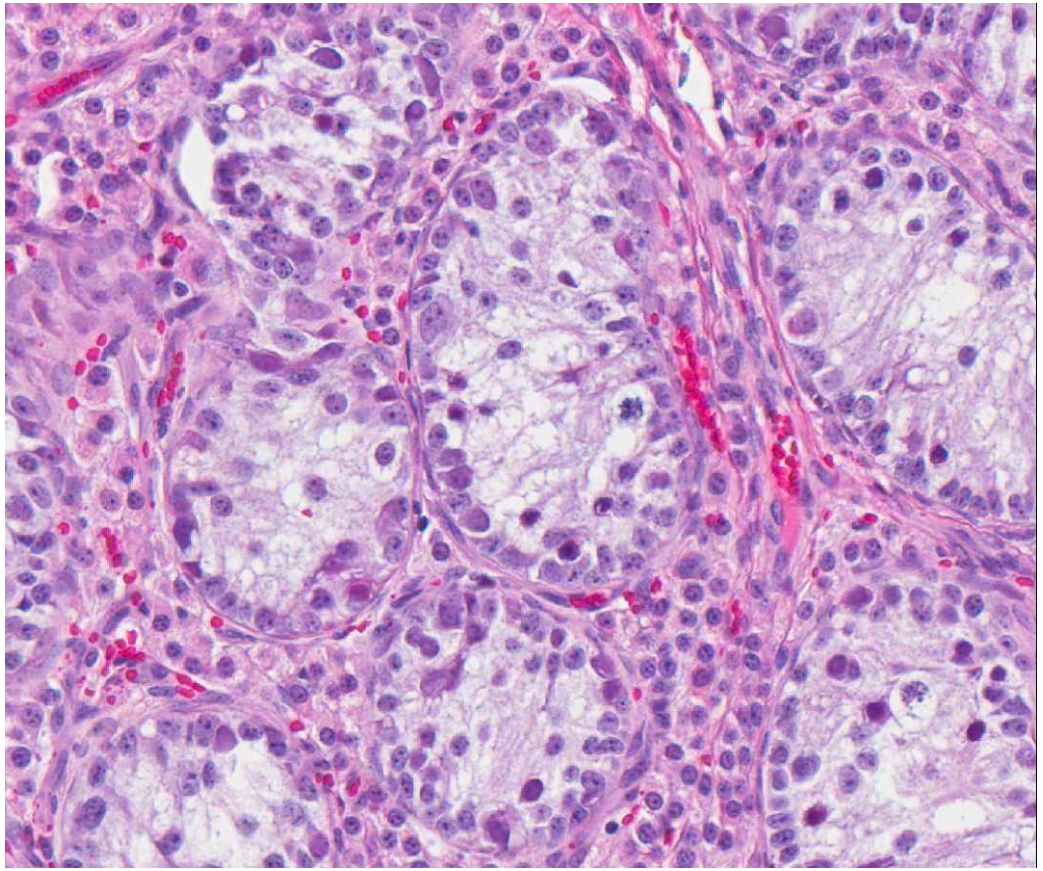


Testis: Small - Hypoplasia

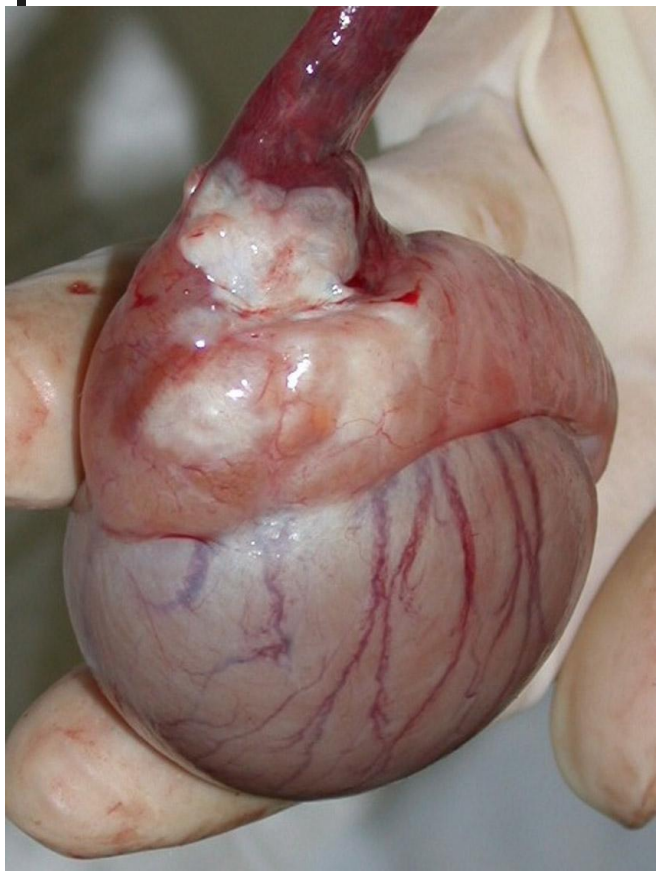
- Hypoplasia

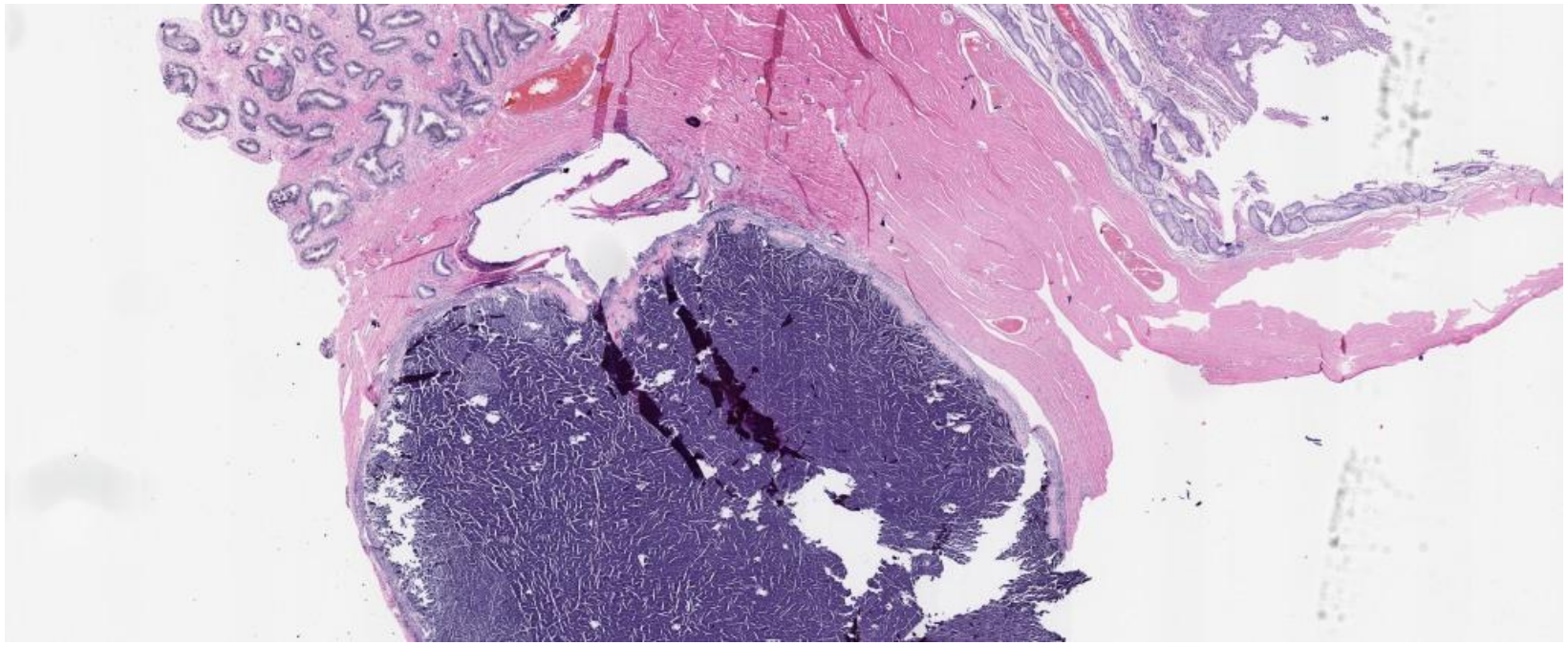
All these testes were from rams of the same age

Testicular hypoplasia



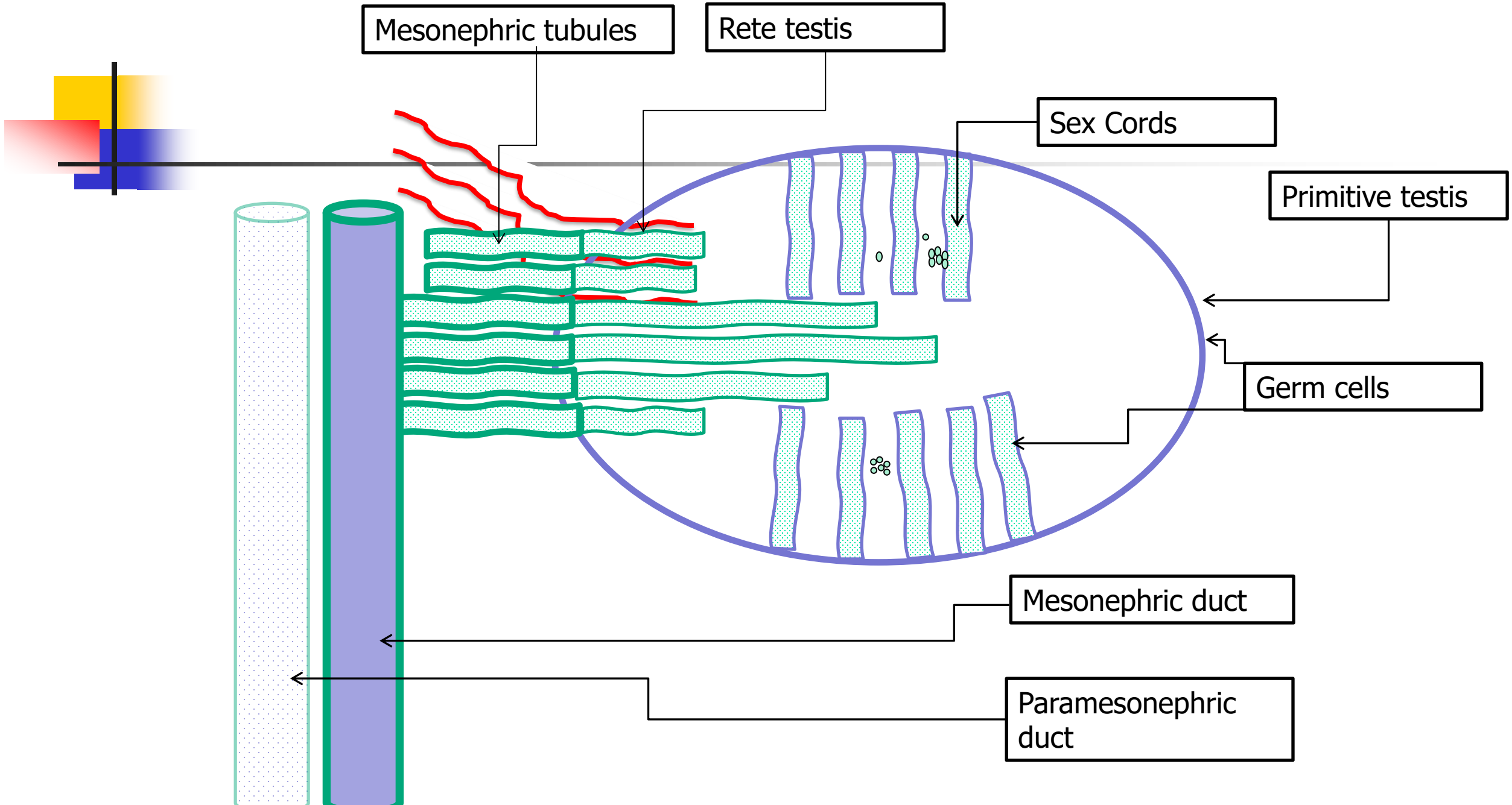
Segmental aplasia of epididymal head



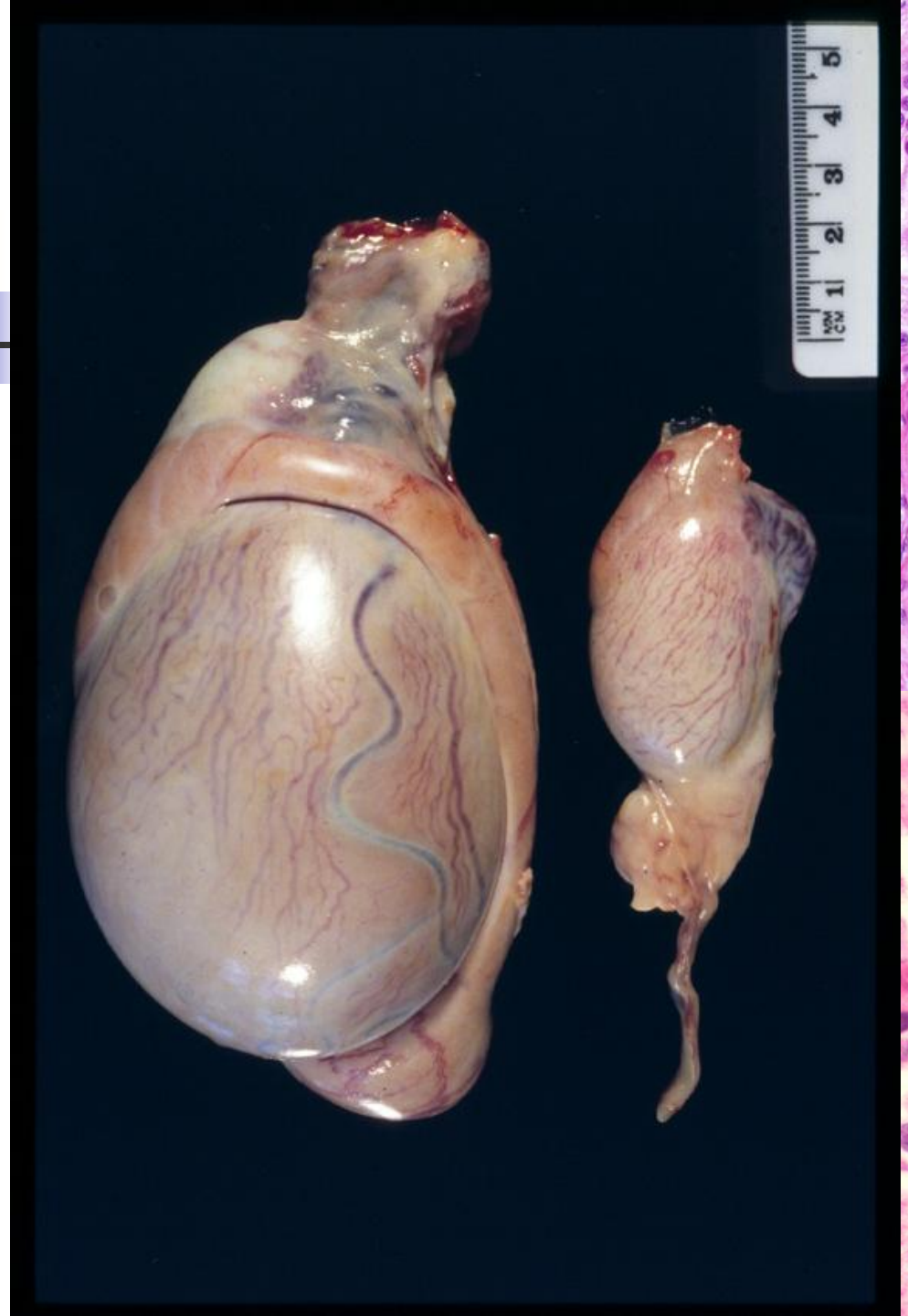


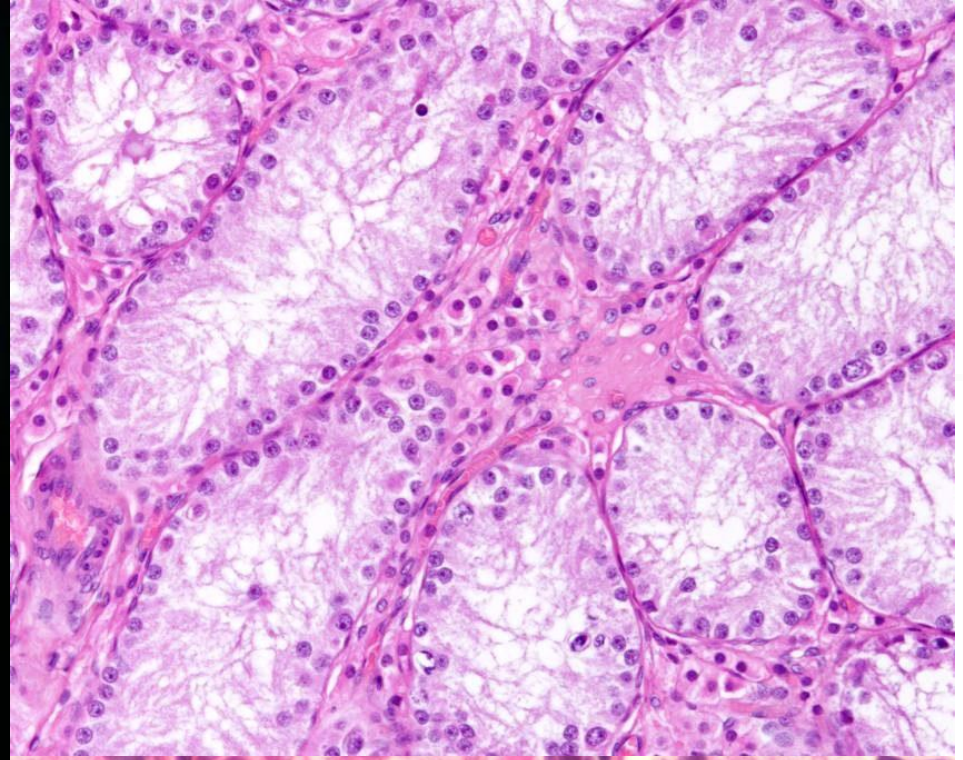
Photograph complements of Dr Cathy Gartley

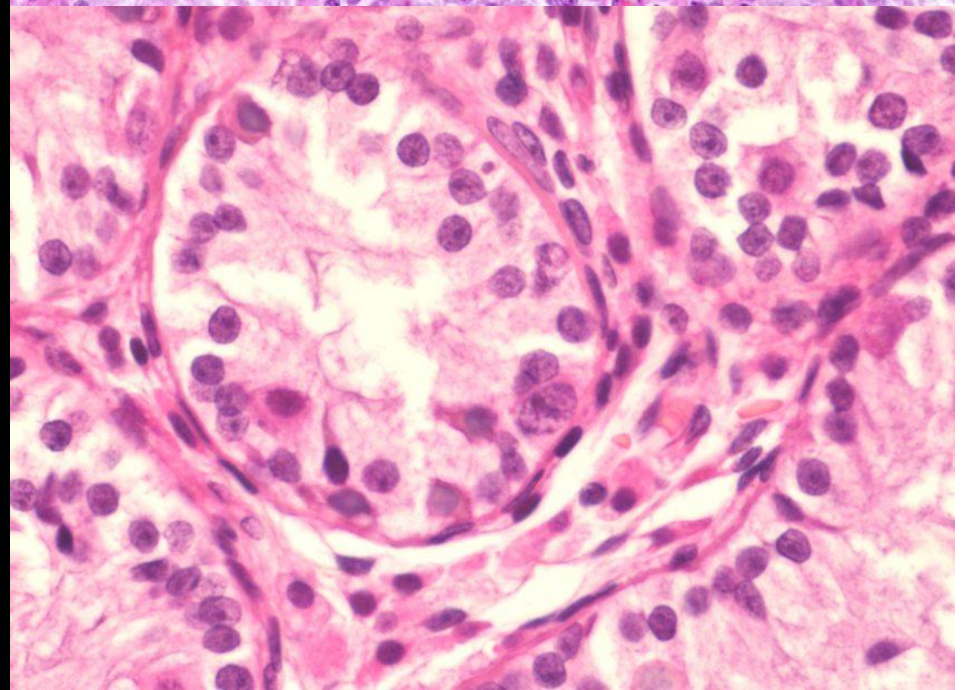
Spermatic granuloma of the epididymal head



Cryptorchid







Cryptorchidism

- Very common
- Little challenge – clinical diagnosis
- Hormonal profile in bilateral cryptorchidism?
  - Testosterone
  - GnRH stim
  - Inhibin
  - AMH

Cryptorchid and thickened deferent duct

Male dog with uterus/cystic endometrial hyperplasia and or pyometra

- Anti Müllerian hormone receptor defect.

- Persistent Müllerian duct syndrome of miniature Schnauzer (XY *SRY*+ testicular with uterus, cryptorchidism and phenotypic male.

Diagnosis

- XY, *SRY*+, testicular DSD, Uterus and deferent duct, Male, cryptorchid. Persistent Müllerian Duct Syndrome (PMDS)

- Autosomal recessive in Miniature Schnauzer.
- Mutation in Anti-Müllerian Hormone receptor gene
- Paramesonephric duct fails to regress
- Cryptorchidism from resistance to AMH in testicular descent

Epididymis:

- **Segmental Aplasia of Mesonephric Duct**
  - XY Testicular DSD
  - Don’t use for breeding
    - Hereditary
    - No spermatozoal transport – Infertility
    - Easily missed

Examples of XY DSD

- Testicular hypoplasia
- Cryptorchidism
- Persistent Müllerian duct syndrome (Miniature Schnauzer)
- Segmental aplasia of mesonephric duct

- Cystic remnants are rare.

Equine DSD

- XX *SRY*- ovotesticular DSD females
  - 6 cases
- XY *SRY*- gonadal dysgenesis females
  - 6 cases
- XY *SRY*+ testicular (normal) females
  - 5 cases
- No XX *SRY*+ ever been verified in horses

Villagómez DAF, Lear TL, Chenier T, Lee S, McGee RB, Cahill J, Foster RA, Reyes E, St John E, King WA. Equine Disorders of Sexual Development in 17 Mares Including XX, SRY -Negative, XY, SRY -Negative and XY, SRY -Positive Genotypes. Sexual Development 2010

Acknowledgements

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- Department of Pathobiology, Ontario Veterinary College
- Department of Population Medicine - Theriogenologists

- Pathologists who send me such cool cases.
- Graduate students of Department of Pathobiology and Population Medicine (Theriogenology)