Sex Determination

• Most animal species are *dioecious*
  – 2 sexes with different gonads
    • Females: produce eggs in ovaries
    • Males: produce sperm in testes

• Exception
  • Hermaphrodites: have both types of gonads

• Many animals also differ in secondary traits
What Determines Sex?

• Individual differentiates into male or female
• Causes
  – Genetic factors (sex chromosomes)
    – occur at fertilization
  – Environmental factors
    – occur after fertilization
How Do Vertebrate Gonads Develop?

- Gonad differentiation
  - first morphological difference between males and females

- Gonads develop from intermediate mesoderm

- Paired structures
What is a Bipotential Gonad?

- Indifferent gonad develops
  - 4-6 wks in human = “bipotential stage”
  - genital ridge forms next to developing kidney (mesonephric ridge)
Structure of the Indifferent Gonad

- Sex cords form
  - Columns of epithelial cells penetrate mesenchyme
  - Primordial germ cells migrate from posterior endoderm
  - Become surrounded by sex cords
What is the Fate of the Sex Cords?

- Initially in central area (medulla, medullary)
  - Will develop in male
  - Proliferate
- In outer area (cortex, cortical)
  - Develop in female
- Normally binary choice
Differentiation of the Gonad

• Into testes or ovaries
  – primary sex determination
  – does not involve hormones

network of internal sex cords (at puberty: --> seminiferous tubules, Sertoli cells)
new cortical sex cords cluster around each germ cell
Male Differentiation

- Male sex cords or testis cords proliferate and cortex becomes thick layer of extracellular matrix
- Male germ cells inhibited from entering meiosis
- Secrete factors causing cord cells to become Sertoli cells
Female Differentiation

- Female germ cells in cortex enter meiosis
- New sex cords form there and form clusters
- Germ cell becomes egg
- Cord cells become granulosa cells
- Surrounding mesenchyme cells become thecal cells
- Thecal + granulosa = follicle
Male Ducts

- Provide exit for gametes
  - In males remnants of mesonephric kidney called Wolffian ducts
  - Differentiates into *vas deferens* and *epididymis*

- Supported by testosterone
  - Made by Leydig cells
  - Leydig cells come from mesenchyme
Female Ducts

- Also provide exit for gametes
  - In females, Mullerian duct part of gonad
  - Differentiates into uterus, oviducts and upper vagina
- Supported by estrogen
  - Destroyed in males by anti-Mullerian duct hormone
vas deferens
oviduct
kidney
hormones
Secondary sex determination
Secondary Sex Determination

• Determination of *non-gonadal* differences
  – Females: oviduct, uterus, cervix, mammary glands
  – Males: vas deferens, seminal vesicle, penis, prostate glands
  – Also, non reproductive traits such as body size, vocal cords, musculature, etc.

• Usually depends on primary sex determination
Secondary Sex Determination

- Experiment:
- Remove gonad of young rabbit fetus before differentiation

- XX Rabbit   ------------> appears as female
- XY Rabbit   ------------> appears as female

- Female = default path of secondary sex determination
How is Primary Sex Determination Triggered?

- Chromosomes: XX or XY?
  - What are the genes involved?
  - Compare to model systems (*Drosophila, C.elegans*)
Testis Determining Factor

- **XX** and **XXX** are female
- **XY** and **XXY** are male
  - Must be a “testis determining factor on the Y chromosome”
  - Search for TDF
- **Sex-reversed Females**: **XY**
  - deletion of short arm of Y
- **Sex-reversed Males**: **XX**
  - addition of small piece from Y
SRY

• SRY identified
  – SRY = sex-determining region of the Y chromosome
  – Encodes a 223 a.a. protein
  – Transcriptional regulator of testes-specific genes
    • possible splicing factor
  – Where is SRY expressed?
**SRY Directs Male Development**

- **Experiments with mice and SRY:**
  - Mouse Sry is expressed in the bipotential gonad of males
    - Disappears after testes differentiate
  - Transgenic mice
    - inject Sry into pronuclei of newly fertilized mice
    - find some “male” looking mice that are actually XX
  - Other genes are necessary for testes determining ability: even genes on autosomes!
Male Development Also Requires *Sox9*

- *Sox9* = autosomal gene required for testicular development
- Expressed in genital ridges of males only, and in Sertoli cells throughout life
- Conserved throughout vertebrates
- Sex-reversed males:
  - XX lacking SRY
    - have extra copy of Sox9
  - XX transgenic for Sox9 form testis
- Transcriptional regulator
  - e.g. activates AMH gene (anti-Mullerian hormone)
- Splicing regulator also
Male Development Also Requires SF1

- SF1 (steroidogenic factor 1)
  - Transcription factor
  - Necessary for bipotential gonad
  - Expression drops in XX gonad but persists in XY gonad

- Activates genes necessary for testosterone production (in Leydig cells) and AMH (in Sertoli cell)
**Dax1 on X Antagonizes SRY and Sox9**

- Expressed in genital ridges (in both sexes)
  - Stays on in developing ovary
  - Turned off in developing testes
- Gonadal dysgenesis female
  - XY but no deletion of Y
  - Instead an extra copy of Dax1
    Dax1 acts as anti-testis gene in females
**$Wnt4$ May Be an Ovary-Determining Gene**

- Expressed in bipotential genital ridge
  - Expression lost in XY gonads, maintained in XX gonads
  - Activates $Wnt4$
- XX $wnt4^{-/-}$ mice fail to develop ovaries and instead gonads make AMH
- XY humans with double $wnt4$ make extra DAX1 and ovary
Partial Model of Vertebrate Sex Determination

- If XY: SRY → SOX9 → SF1 → Testis
- If XX: WNT4 → DAX1, Other genes → Ovary
- WT1, LHX9, LIM1, etc.