Theriogenology
(Andrology, Gynecology & Obstetrics)

session-2

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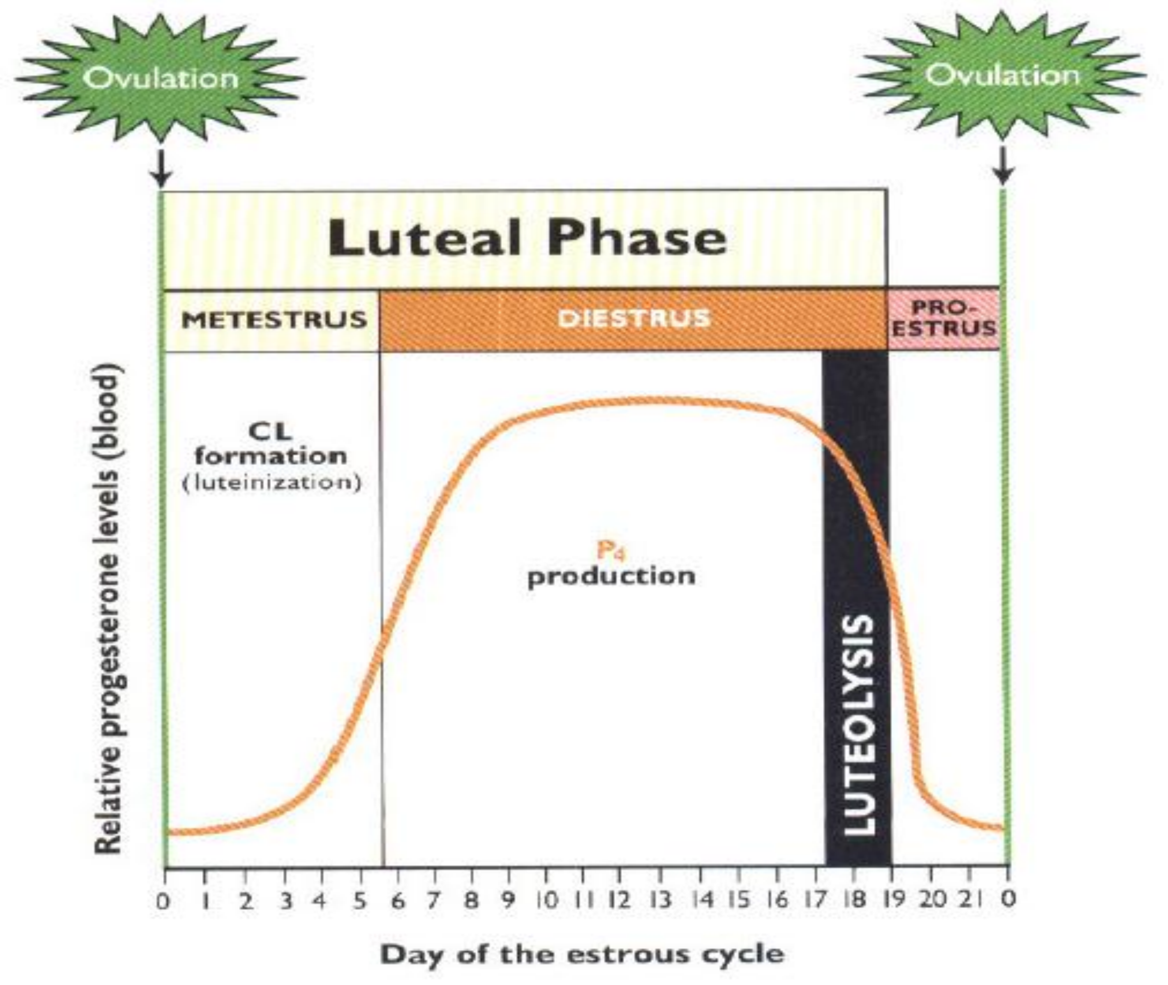
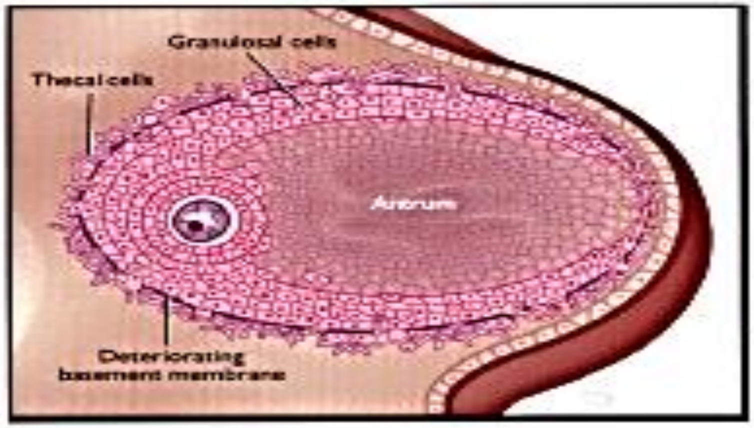
Reproductive Cyclicity

The Luteal Phase

The luteal phase consists of:

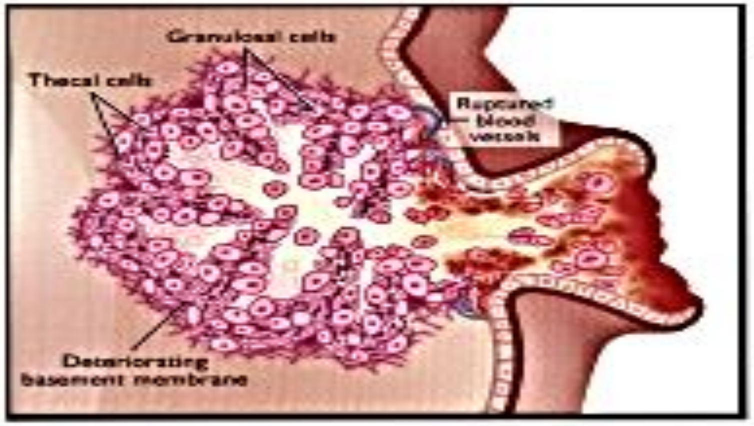
- ***corpora lutea formation (luteinization)***
- ***production of progesterone***
- ***luteolysis***





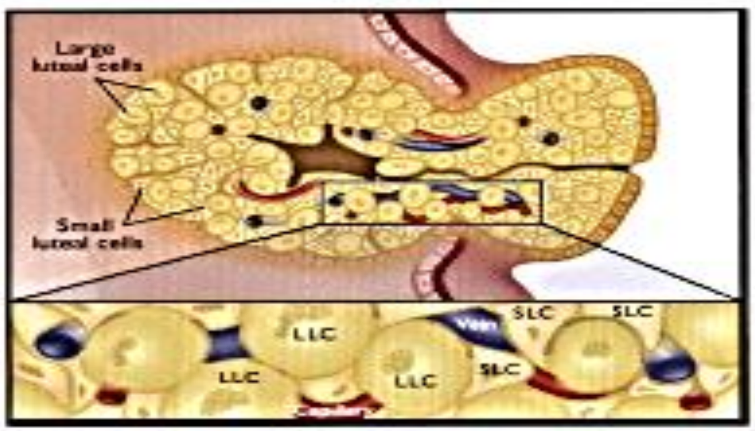
Preovulatory Follicle

The preovulatory follicle consists of granulosal cells that line the antrum. The basement membrane, separating the granulosal cells from the cells of the theca interna begins to deteriorate prior to ovulation because of the action of collagenase. Complete separation between the granulosal cells and the theca interna no longer exists and the cells can begin to intermingle.



Corpus Hemorrhagicum

During ovulation, many small blood vessels rupture causing local hemorrhage. This hemorrhage appears as a blood clot on the surface of the ovary that sometimes penetrates into the center of the follicle after ovulation (See Figures 9-3, 1A and B and 9-4, 1A and B). During ovulation the follicle implodes and is "thrown" into folds. The cells of the theca interna and the granulosa begin to mix. The basement membrane forms the connective tissue substructure of the corpus luteum.



Functional Corpus Luteum

The corpus luteum is now a mixture of large luteal cells, LLC (formerly granulosal cells) and many small luteal cells, SLC (formerly thecal cells). In some cases, there is a remnant of the follicular antrum that forms a small cavity in the center of the corpus luteum (See Figures 9-3, 3B and 9-4, 2B; 9-6, 3B).

The corpus luteum originates from the ovulatory follicle.

Luteal tissue consists of large and small luteal cells:

- *large cells originate from the granulosal cells*
- *small cells originate from the cells of the theca interna*

The Basement membrane (follicle) forms the connective tissue (CL) .

- Large Luteal Cells produce Oxytocine & Progesterone during estrous cycle and Relaxin during pregnancy.

- Small Luteal Cells only produce Progesterone.

large luteal cell undergo hypertrophy, while small luteal cells undergo hyperplasia as the CL develops.

The functional capability of the newly developed CL may also depend on the degree of vascularity in the cellular layers of follicle.

Figure 9-3. Luteal Anatomy in Relation to Progesterone Secretion During the Estrous Cycle in the Cow

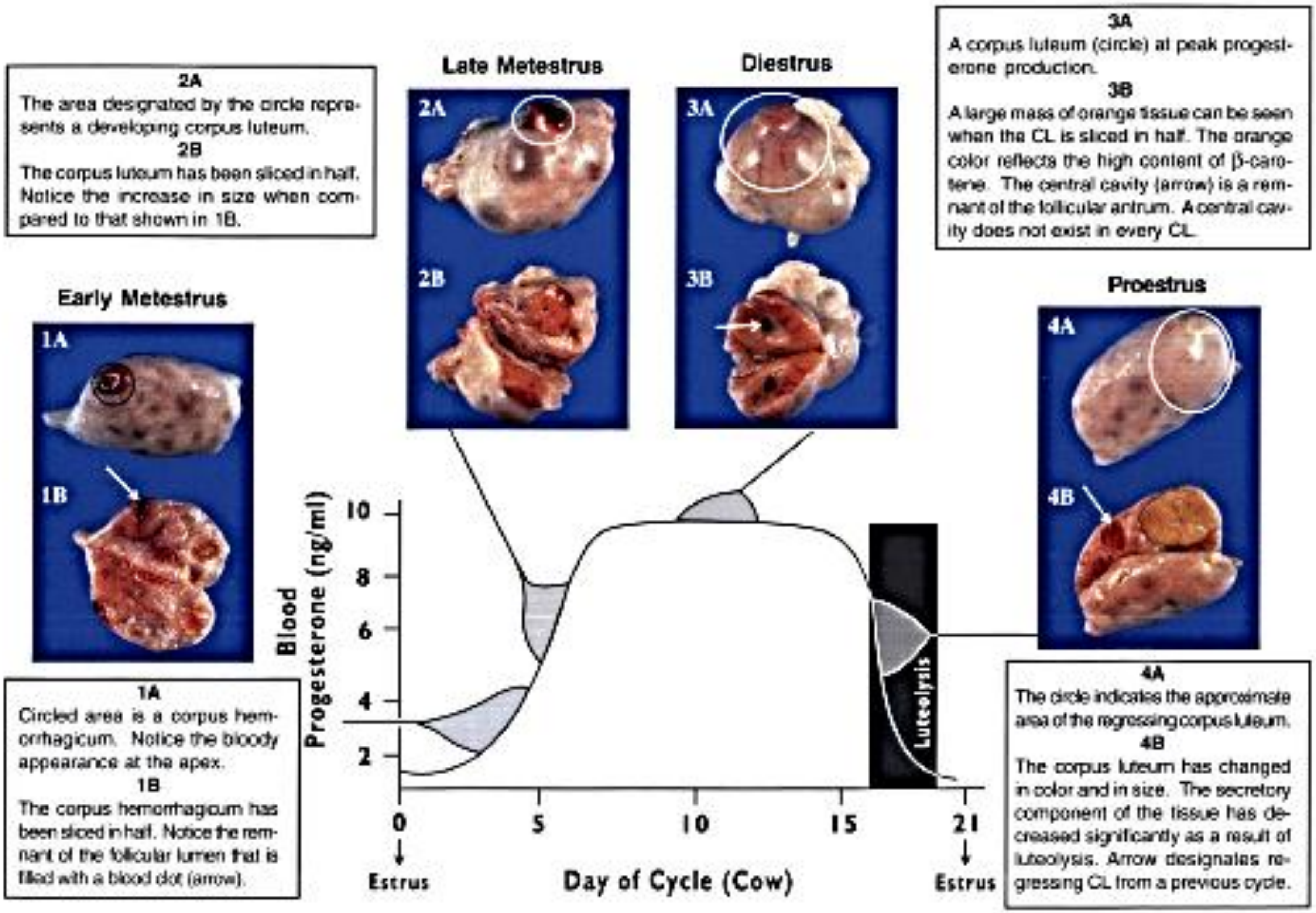
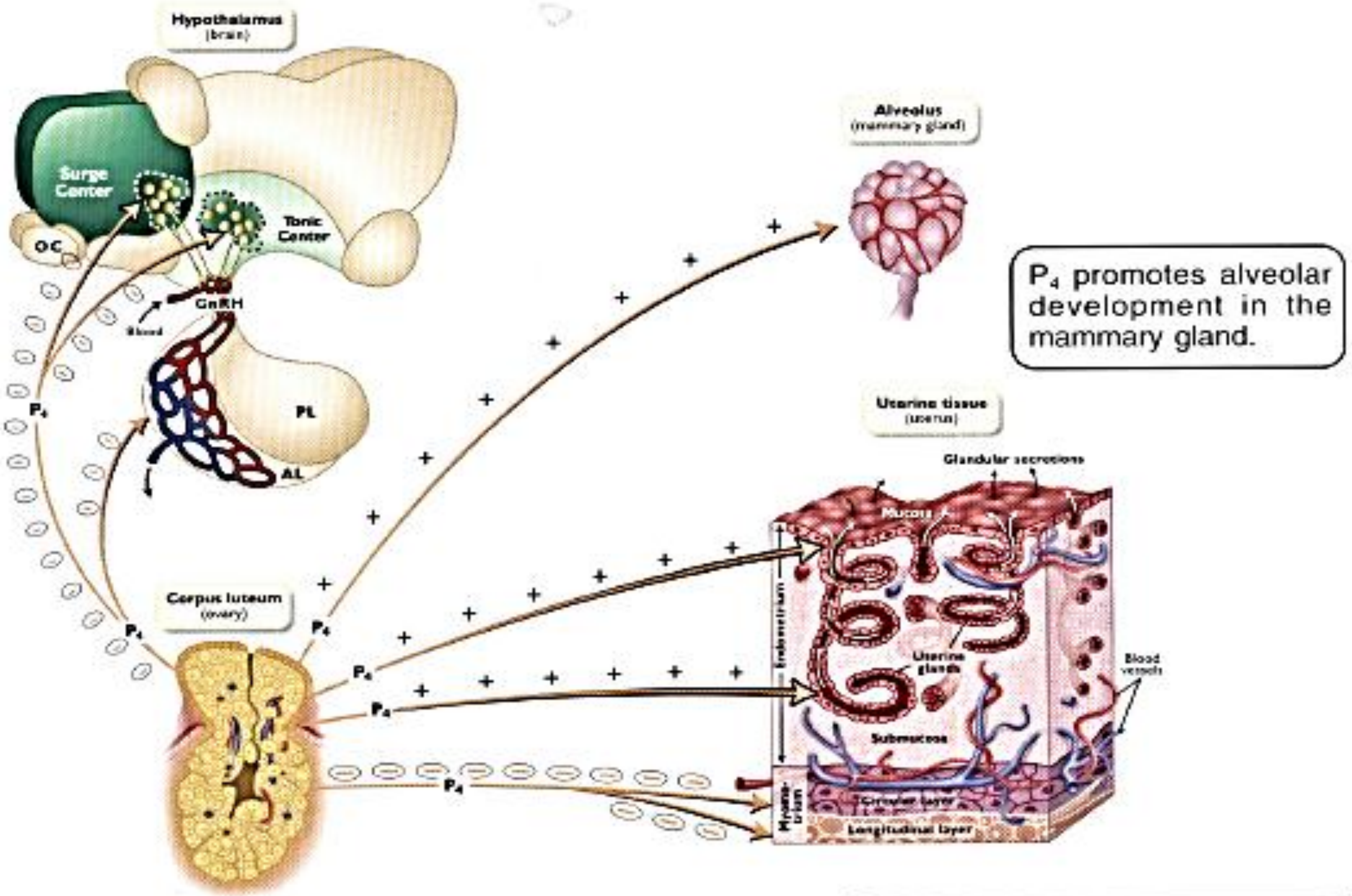


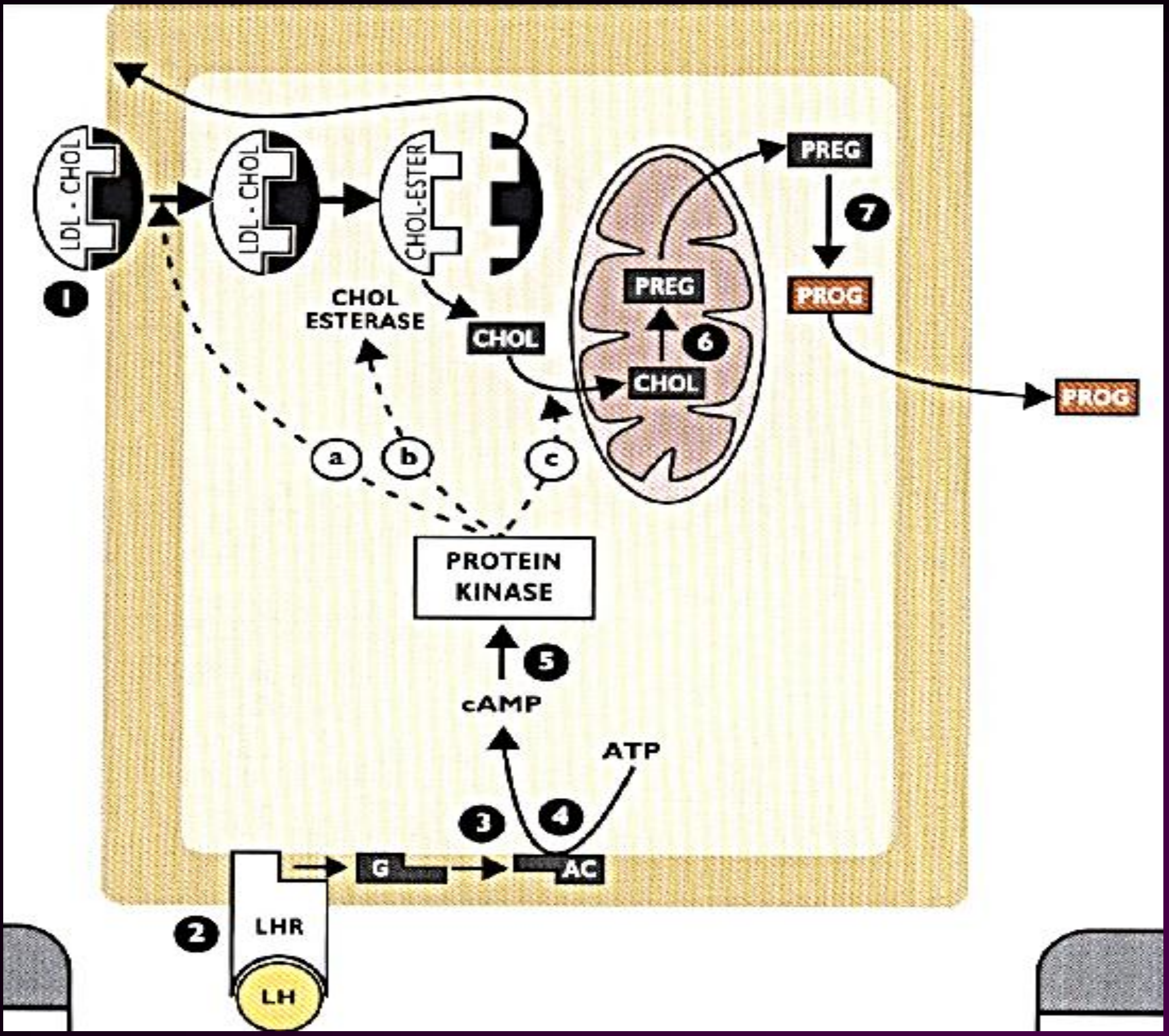
Figure 9-8. Progesterone (P_4) has Many Physiological Effects



P_4 produced by the CL exerts a negative (-) feedback on the GnRH neurons of the hypothalamus. Therefore, GnRH, LH and FSH are suppressed and little estrogen is produced. Progesterone is thought to decrease the number of GnRH receptors on the anterior pituitary.

P_4 exerts a strong positive (+) influence on the endometrium of the uterus. Under the influence of P_4, the uterine glands secrete materials into the uterine lumen. Progesterone inhibits the myometrium and thus reduces its contractility and tone.

The presence of basal (tonic) LH and cholesterol is necessary for progesterone to be produced by luteal cells.



LDL · CHOL
LDL · CHOL
CHOL-ESTER
PREG
7
PREG
PROG
1
CHOL ESTERASE
CHOL
6
CHOL
PROG
a
b
c
PROTEIN KINASE
5
cAMP
ATP
3
4
G
AC
2
LHR
LH

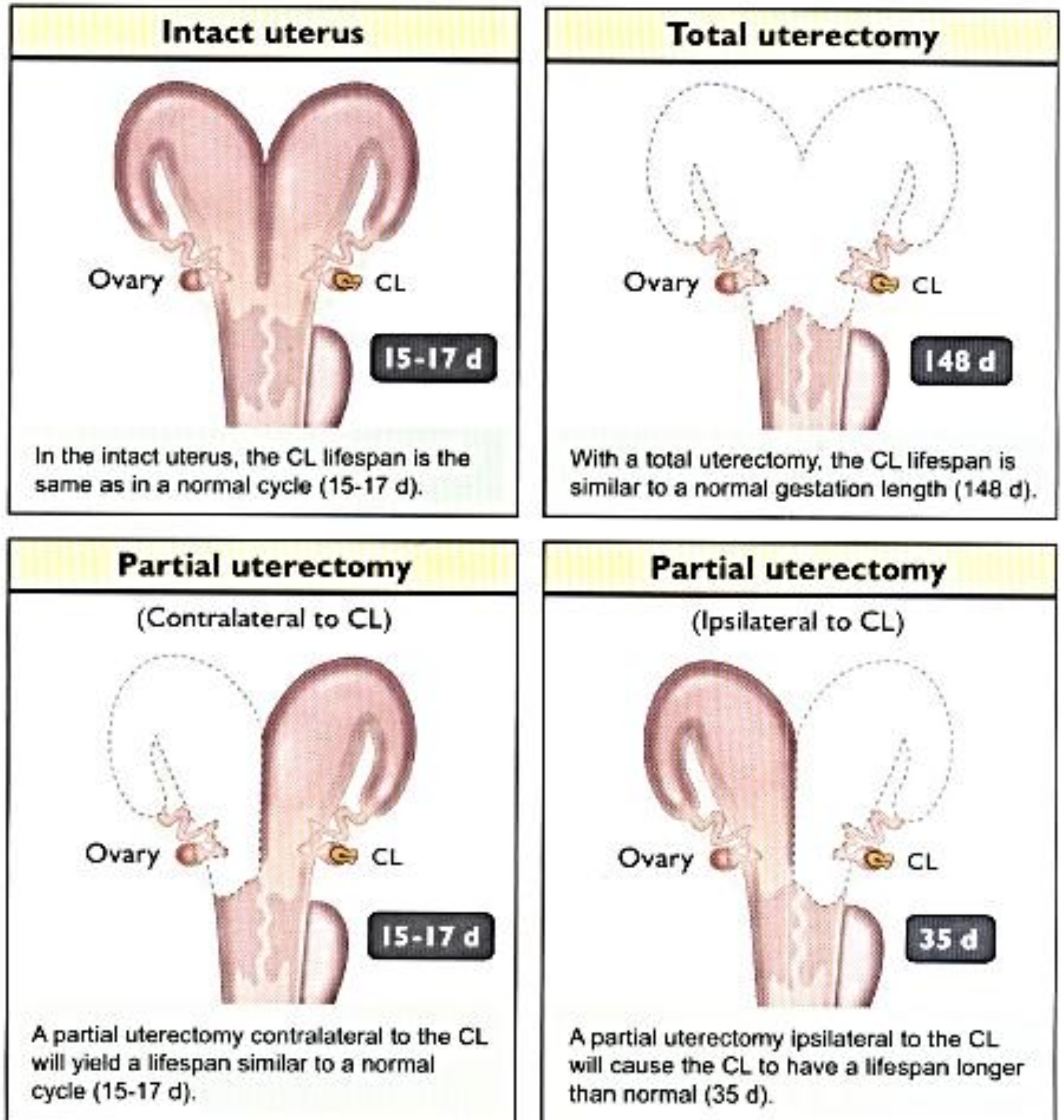
Progesterone is an inhibitor because it:

- ***reduces basal GnRH amplitude and frequency***
- ***prevents behavioral estrus***
- ***stops the preovulatory LH surge***
- ***reduces myometrial tone***

The uterus is required for successful luteolysis in many species.

A vascular countercurrent diffusion system insures that $PGF_{2\alpha}$ will reach the ovary in sufficient quantities to cause luteolysis in the ewe, cow and sow.

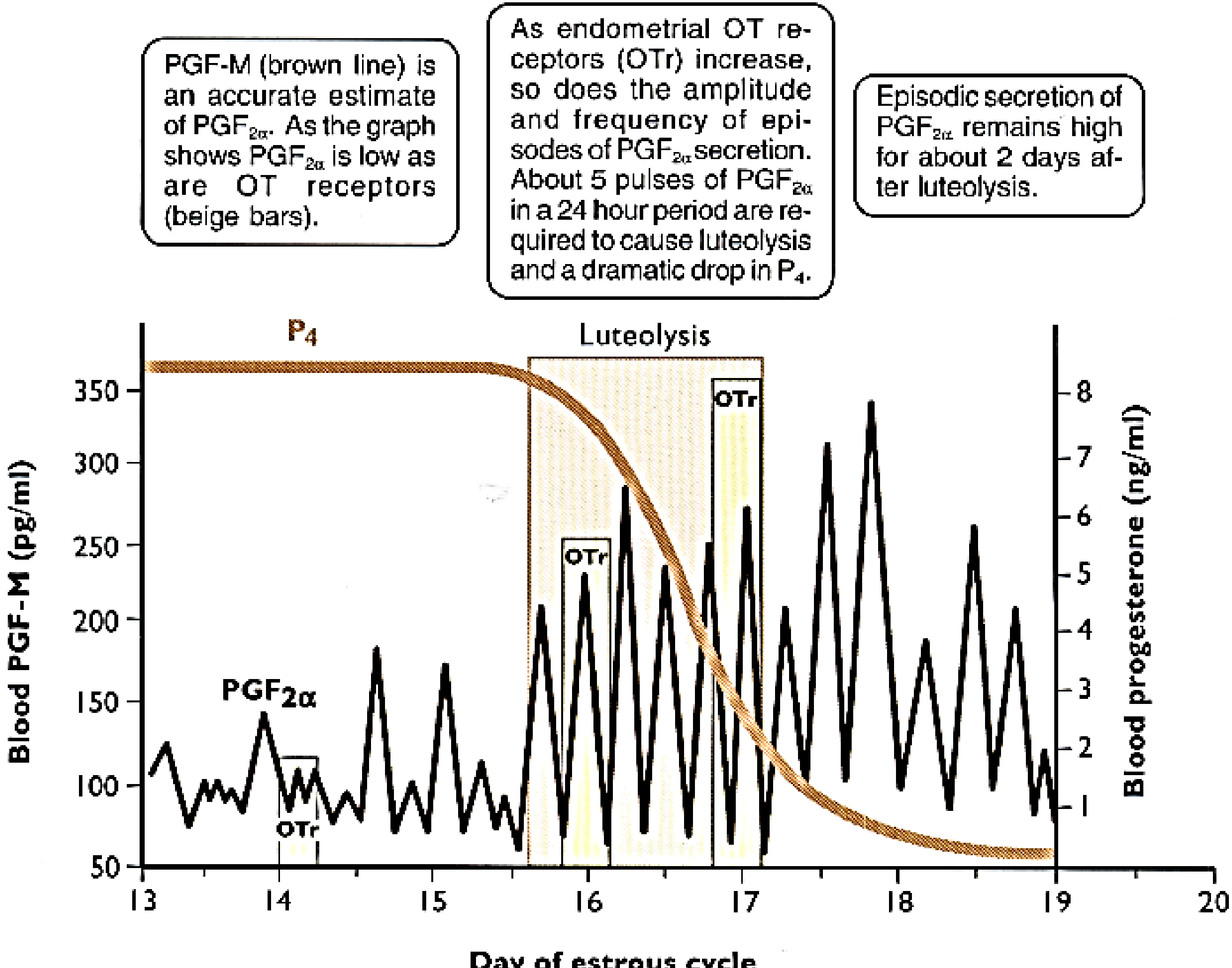
Figure 9-10. Effect of Uterectomy upon Estrous Cycle Duration in the Ewe



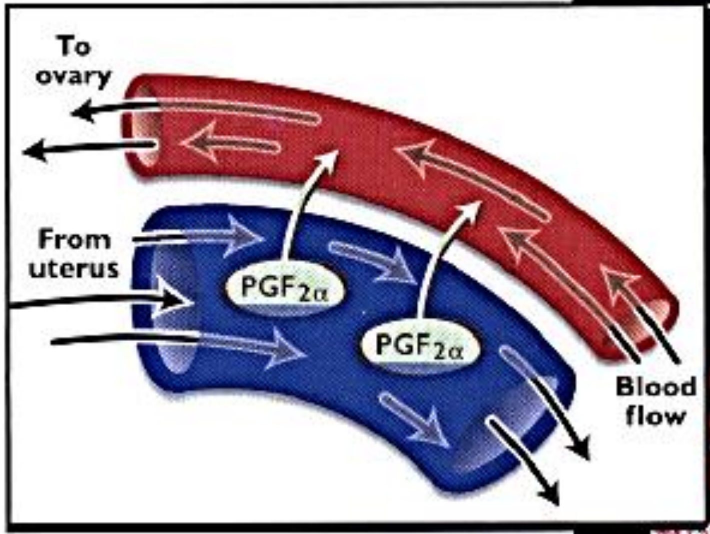
The requirements for luteolysis (in subprimate mammals) are:

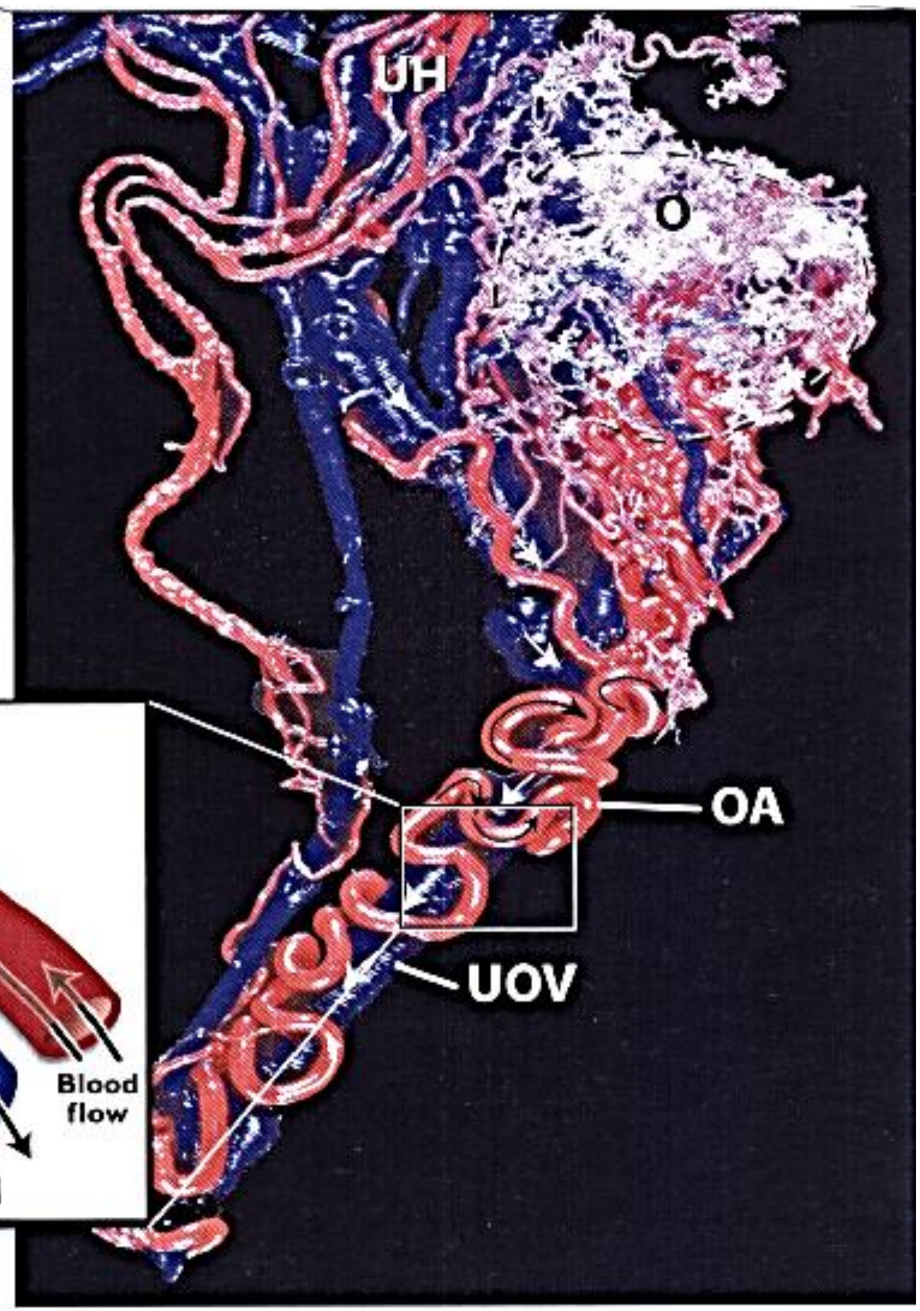
- *presence of oxytocin receptors on endometrial cells*
- *presence of a critical level of oxytocin*
- *$PGF_{2\alpha}$ synthesis by endometrium*

Figure 9-12. Changes in PGF Metabolite (PGF-M), Oxytocin (OT) and Oxtocin Receptors (OTr) During Late Diestrus and Proestrus



Schematic illustration of the countercurrent diffusion system in the cow, sow and ewe. A portion of uterine $PGF_{2\alpha}$ diffuses directly from the utero-ovarian vein into the ovarian artery where it has a direct lytic effect on the corpus luteum.





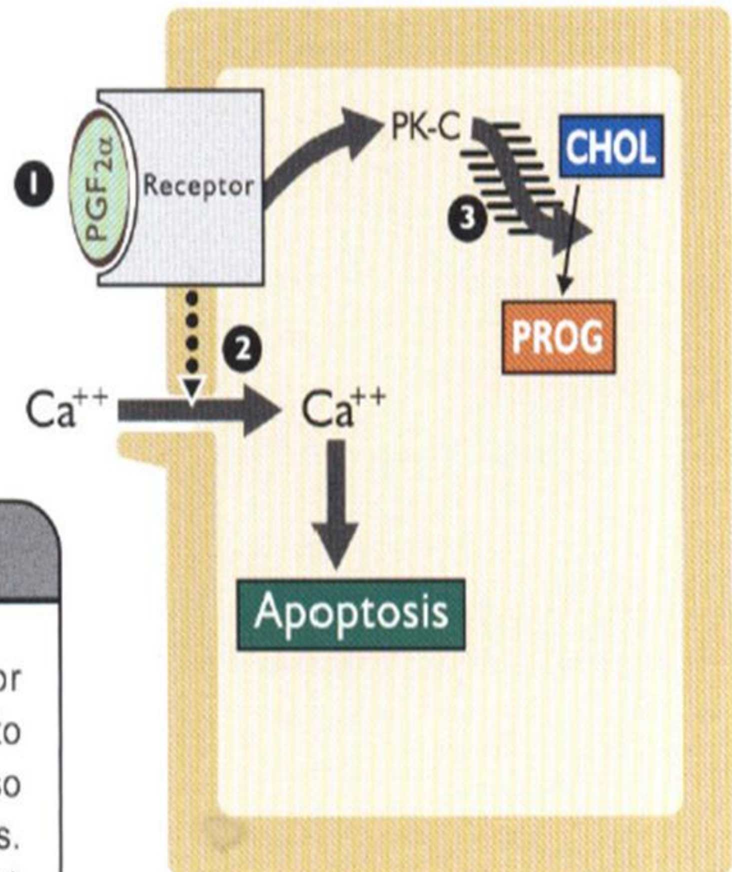
Luteolysis results in:

- *cessation of progesterone production*
- *structural regression to form a corpus albicans*
- *follicular development and entrance into a new follicular phase*

Figure 9-13. Proposed Steps Resulting in the Loss of Progesterone Production from Steroidogenic Cells

1

$PGF_{2\alpha}$ binds to specific receptors on the plasma membrane of the large luteal cells.



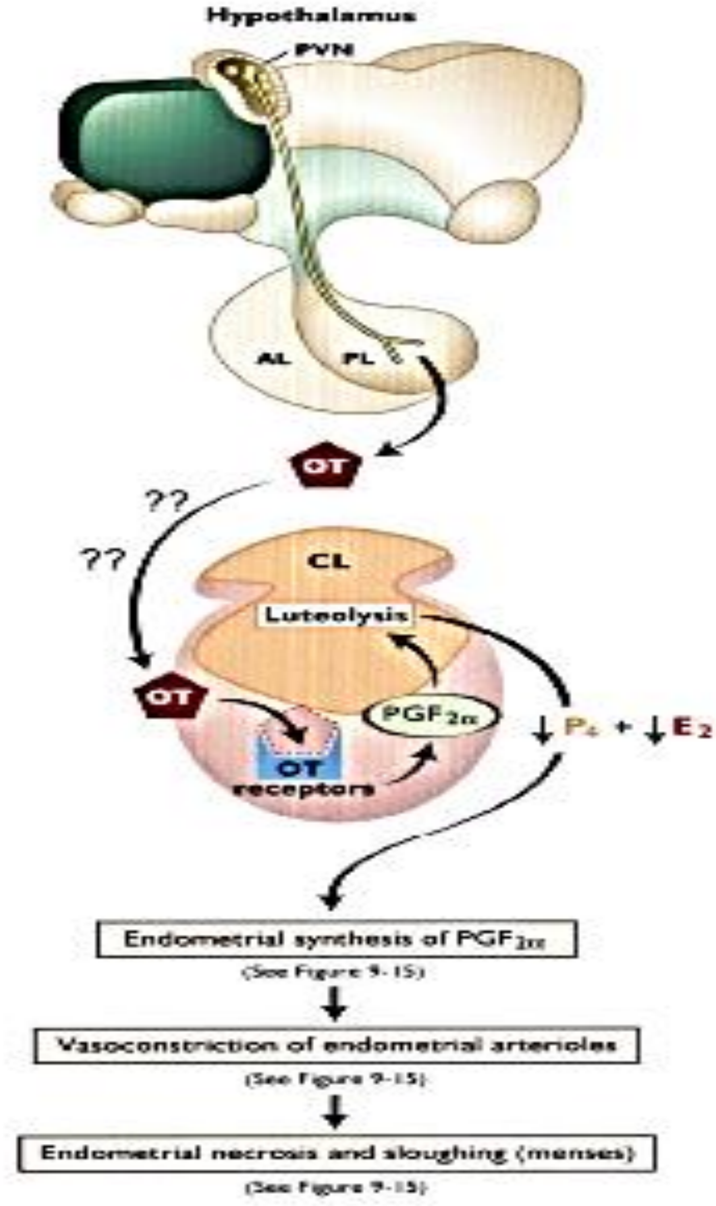
2

The $PGF_{2\alpha}$ receptor complex is believed to open Ca^{++} channels so that Ca^{++} influx occurs. High intracellular Ca^{++} is thought to cause apoptotic effects.

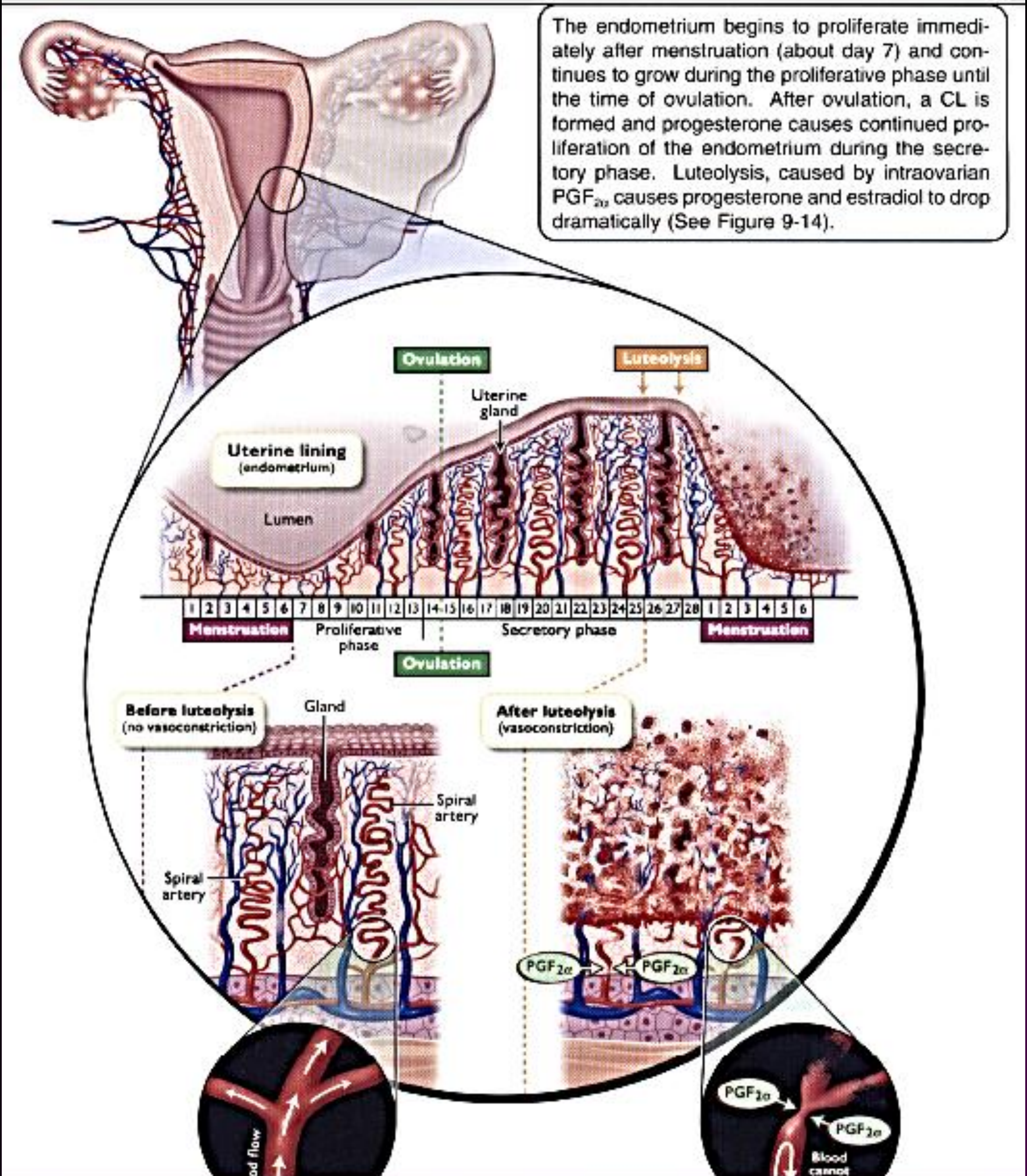
3

The $PGF_{2\alpha}$ receptor complex also activates protein kinase-C (PK-C) which inhibits progesterone synthesis.

Figure 9-14. Proposed Mechanism of Luteolysis in Primates



The endometrium begins to proliferate immediately after menstruation (about day 7) and continues to grow during the proliferative phase until the time of ovulation. After ovulation, a CL is formed and progesterone causes continued proliferation of the endometrium during the secretory phase. Luteolysis, caused by intraovarian $PGF_{2\alpha}$ causes progesterone and estradiol to drop dramatically (See Figure 9-14).



Endocrinology of the Male and Spermatogenesis

Before Spermatozoa can be produced, certain endocrine requirements must be met:

1- Secretion of GnRH from the hypothalamus

2- FSH and LH secretion from the anterior lobe of pituitary

3- Secretion of gonadal steroids

Note! The hypothalamus in the male does not develop a surge center.

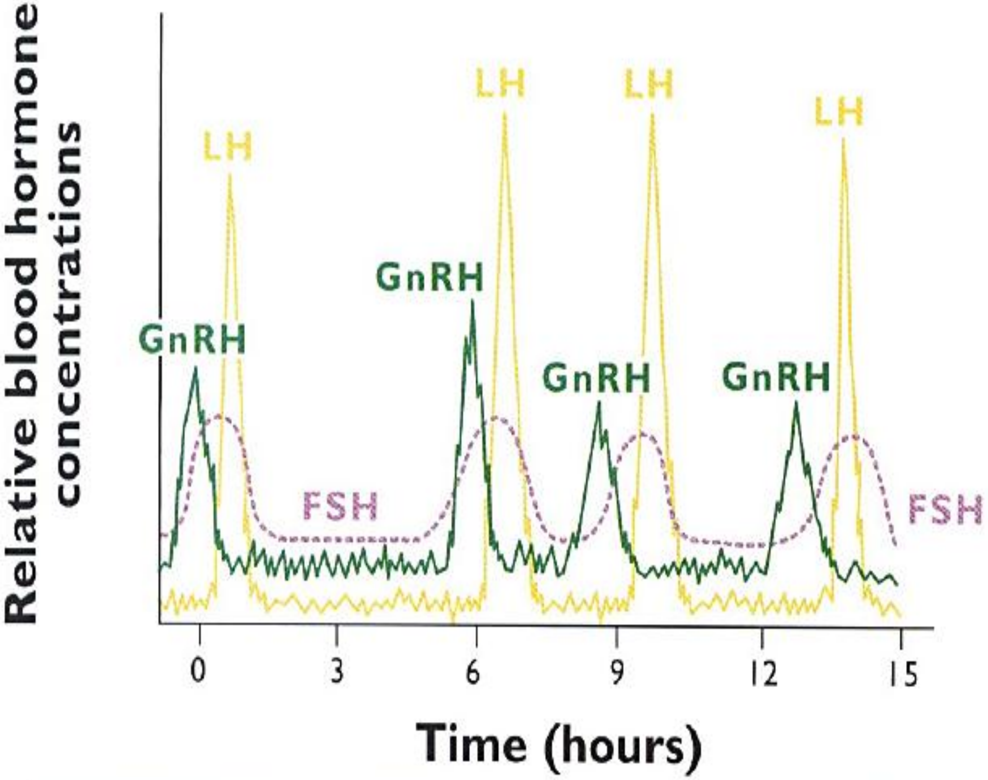
Production of normal numbers of fertile spermatozoa requires:

- *endocrine regulation of the testis*
- *mitotic divisions of spermatogonia*
- *meiotic divisions resulting in haploid spermatids*
- *morphologic transformation of spermatids into spermatozoa*

Leydig cells are the male equivalent of the follicular theca interna cells.

Sertoli cells are the male equivalent of the follicular granulosal cells.

Figure 10-1. Relationship Between GnRH, LH and FSH in the Male



GnRH causes the release of LH and FSH. Episodes of all three hormones occur between 4 and 8 times in 24 hours. The lower FSH profile, when compared to LH, is due to inhibin secretion by Sertoli cells. Also, the greater duration of the FSH episode is probably due to its longer half-life (100 min) when compared to LH (30 min).

Pulsatile discharge of LH is important for 2 reasons:

1- high concentrations of testosterone within seminiferous tubule are essentiaol for spermatogenesis (intratesticular levels of testosterone are 100-500 times higher than blood)

2- Leydig cells become refractory to sustained high levels of LH.

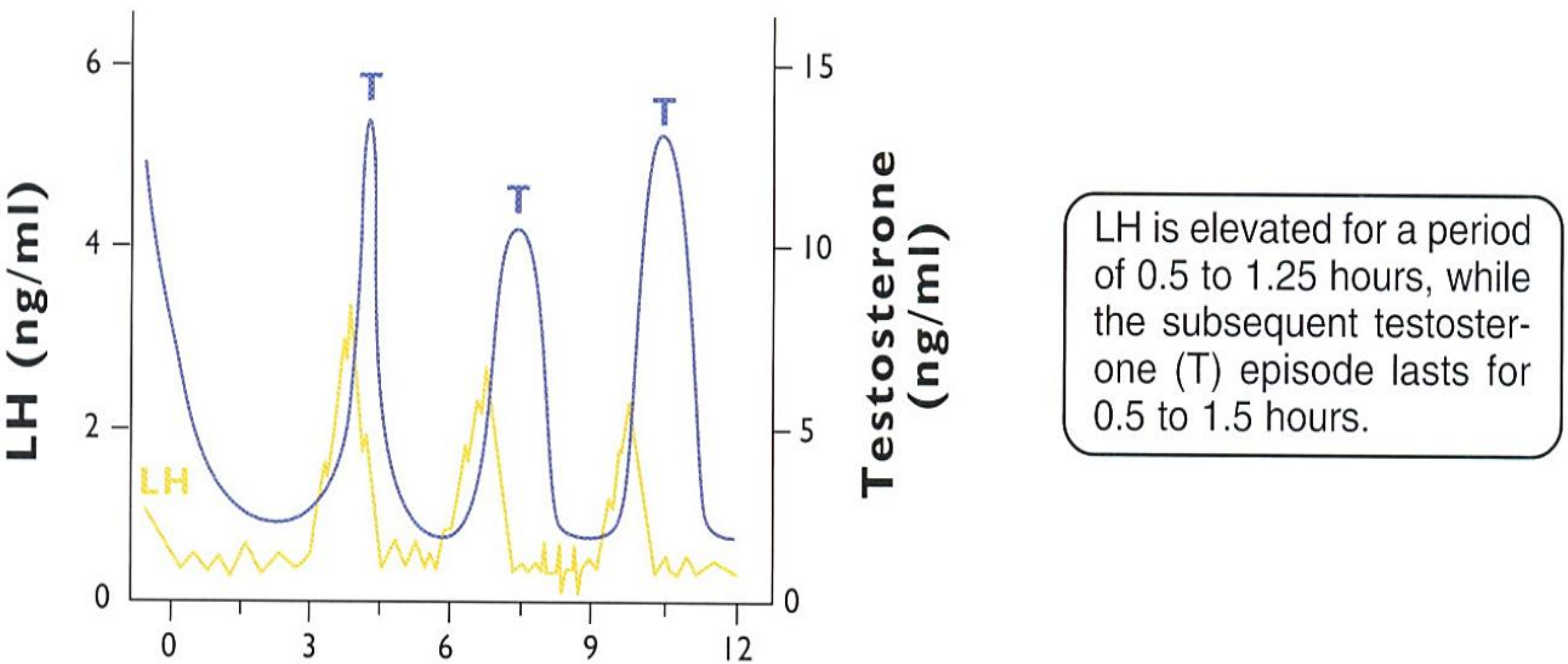
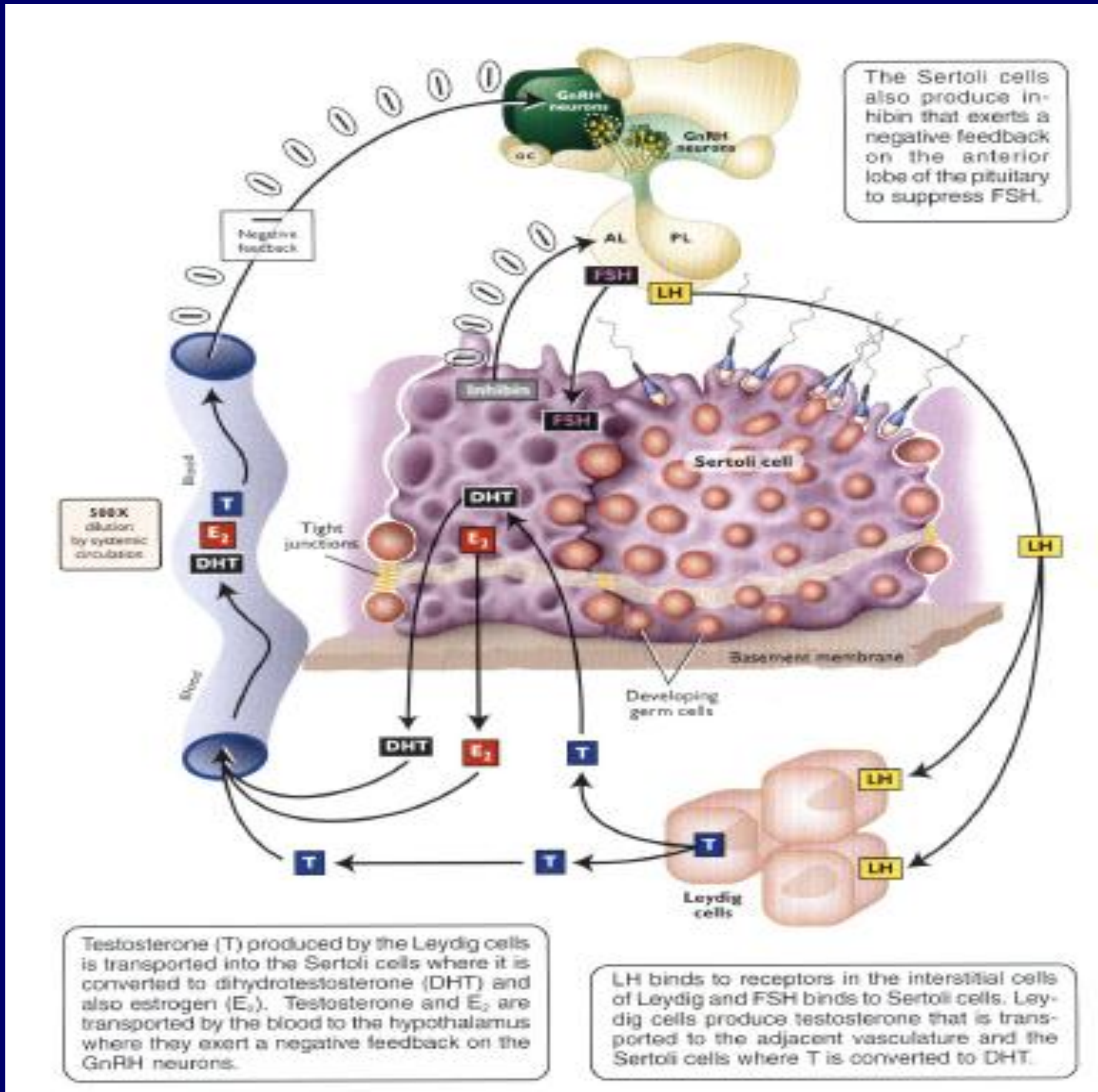


Figure 10-2. Typical Peripheral Concentrations of Blood LH and Testosterone (T) in the Male


The Sertoli cells also produce inhibin that exerts a negative feedback on the anterior lobe of the pituitary to suppress FSH.
GnRH neurons
GnRH neurons
OC
AL
PL
Negative feedback
FSH
LH
Inhibin
FSH
Sertoli cell
DHT
Tight junctions
E2
500X dilution by systemic circulation
T
E2
DHT
Blood
Blood
LH
Basement membrane
Developing germ cells
DHT
E2
T
LH
T
LH
T
T
Leydig cells
Testosterone (T) produced by the Leydig cells is transported into the Sertoli cells where it is converted to dihydrotestosterone (DHT) and also estrogen (E2). Testosterone and E2 are transported by the blood to the hypothalamus where they exert a negative feedback on the GnRH neurons.
LH binds to receptors in the interstitial cells of Leydig and FSH binds to Sertoli cells. Leydig cells produce testosterone that is transported to the adjacent vasculature and the Sertoli cells where T is converted to DHT.

Note!

It is believed that chronically high systemic concentration of testosterone suppress FSH secretion.

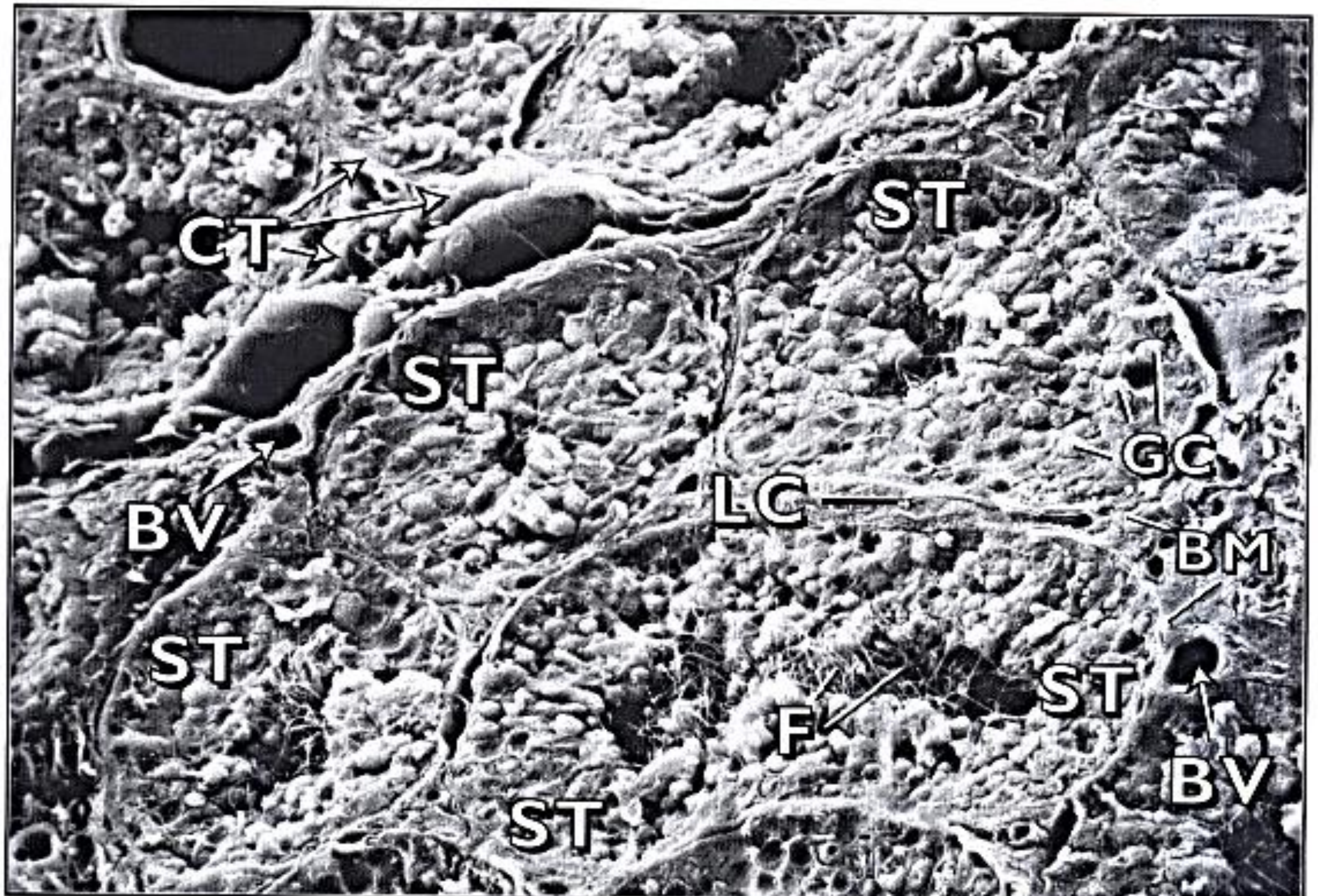
Spermatogenesis is the process of producing spermatozoa

The goals of spermatogenesis are to:

- *provide a male with a continual supply of male gametes (up to decades) through stem cell renewal*
- *provide genetic diversity*
- *provide billions of sperm each day (domestic animals) to maximize reproduction by both natural service and artificial insemination*
- *provide an immunologically privileged site where germ cells are not destroyed by the male's immune system*

Figure 10-4. Scanning Electron Micrograph of Testicular Parenchyma in the Stallion

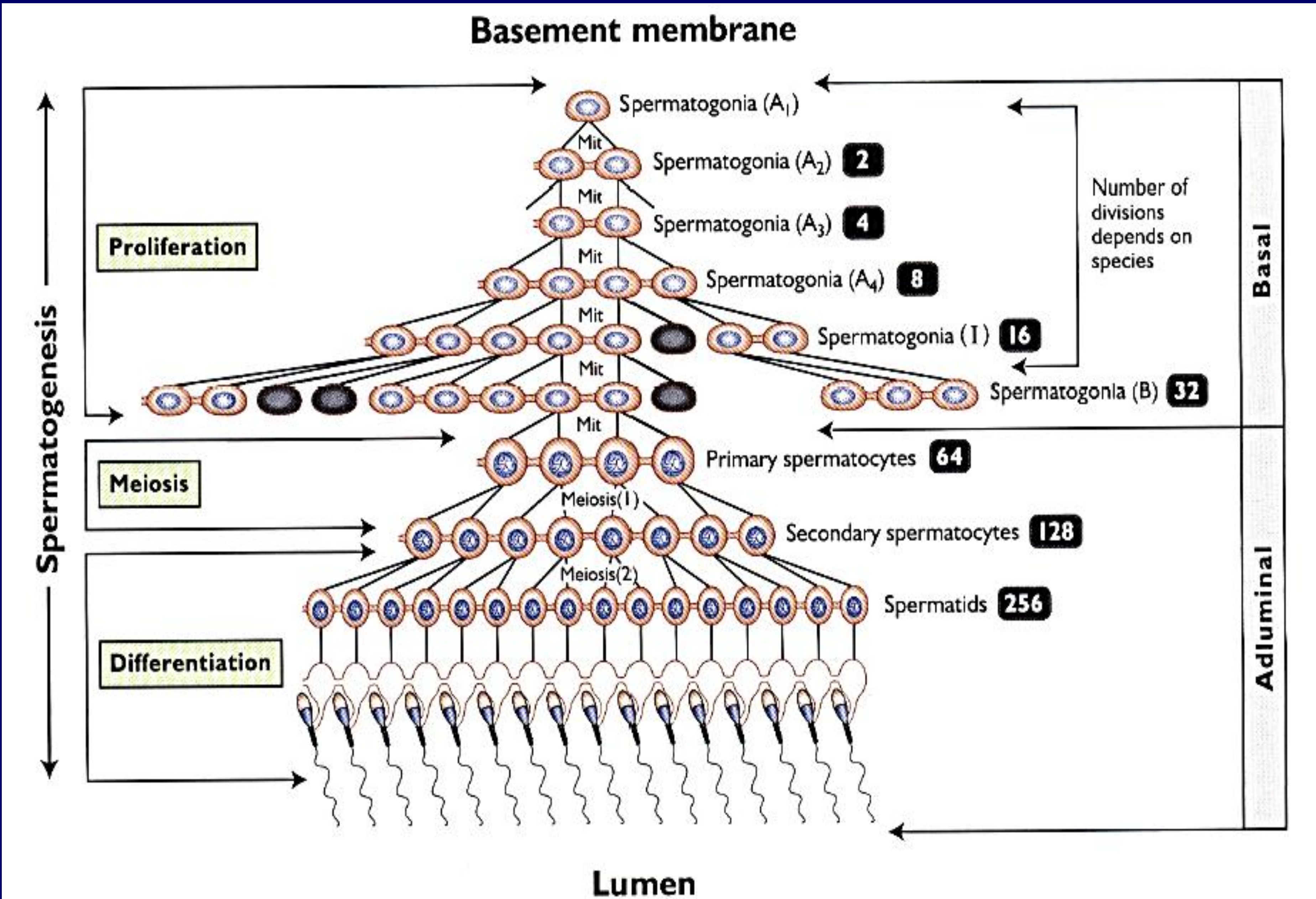
(Courtesy of Dr. Larry Johnson, Texas A&M University, The American Society for Reproductive Medicine. *Fertil. and Steril.*, 1978. 29:208-215)



Seminiferous tubules (ST) containing developing germ cells (GC) are surrounded by a basement membrane (BM). Flagella (F) from developing spermatids can be observed protruding into the lumen of some tubules. The interstitial compartment contains Leydig cells (LC), blood vessels (BV) and connective tissue (CT).

Spermatogenesis = proliferation + meiosis + differentiation

Spermatogenesis: Spermatocytogenesis+Spermiogenesis

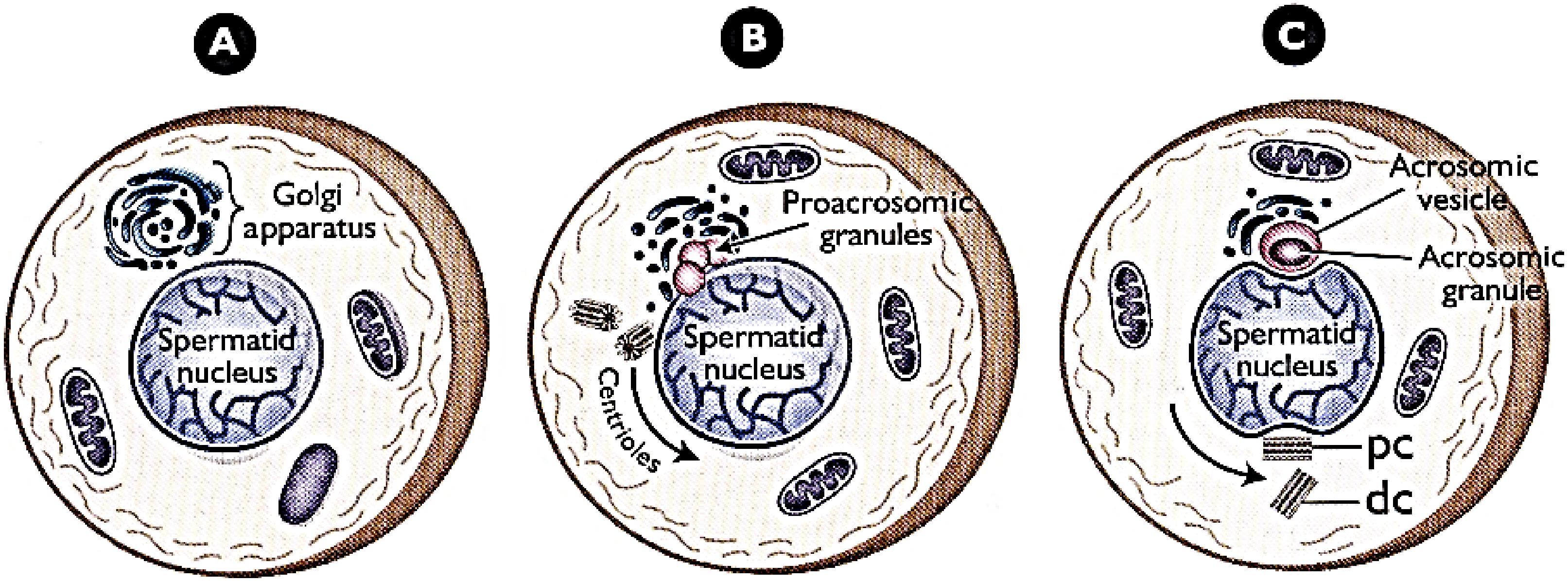


Basement membrane
Spermatogonia (A1)
Mit
Spermatogonia (A2) 2
Mit
Spermatogonia (A3) 4
Mit
Spermatogonia (A4) 8
Mit
Spermatogonia (I) 16
Mit
Spermatogonia (B) 32
Mit
Primary spermatocytes 64
Meiosis(1)
Secondary spermatocytes 128
Meiosis(2)
Spermatids 256
Number of divisions depends on species
Proliferation
Meiosis
Differentiation
Spermatogenesis
Basal
Adluminal
Lumen

Differentiation consists of the:

- ***Golgi phase***
- ***cap phase***
- ***acrosomal phase***
- ***maturation phase***

The Golgi phase = acrosomic vesicle formation

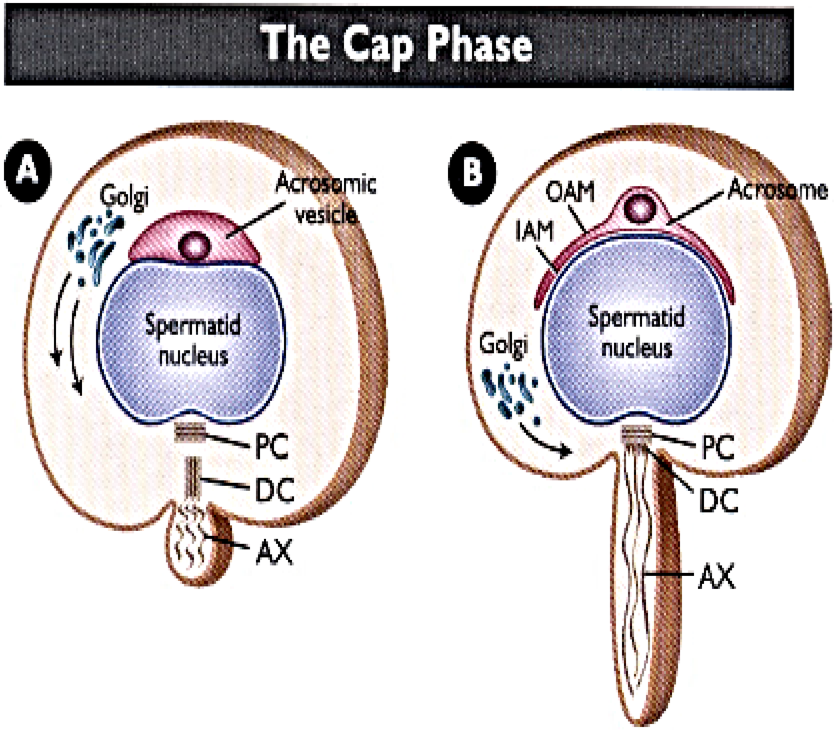


The newly formed spermatid is almost perfectly spherical and has a well developed Golgi apparatus.

Small vesicles of the Golgi fuse, giving rise to larger secretory granules called pro-acrosomic granules. The centrioles start to migrate to a position beneath the nucleus that is opposite the acrosomic vesicle.

Vesicle fusion continues until a large acrosomic vesicle is formed that contains a dense acrosomic granule. The proximal centriole (PC) will give rise to the attachment point of the tail. The distal centriole (DC) will give rise to the developing axoneme (central portion of the tail) inside the cytoplasm of the spermatid.

The cap phase = acrosomic vesicle spreading over the nucleus



A

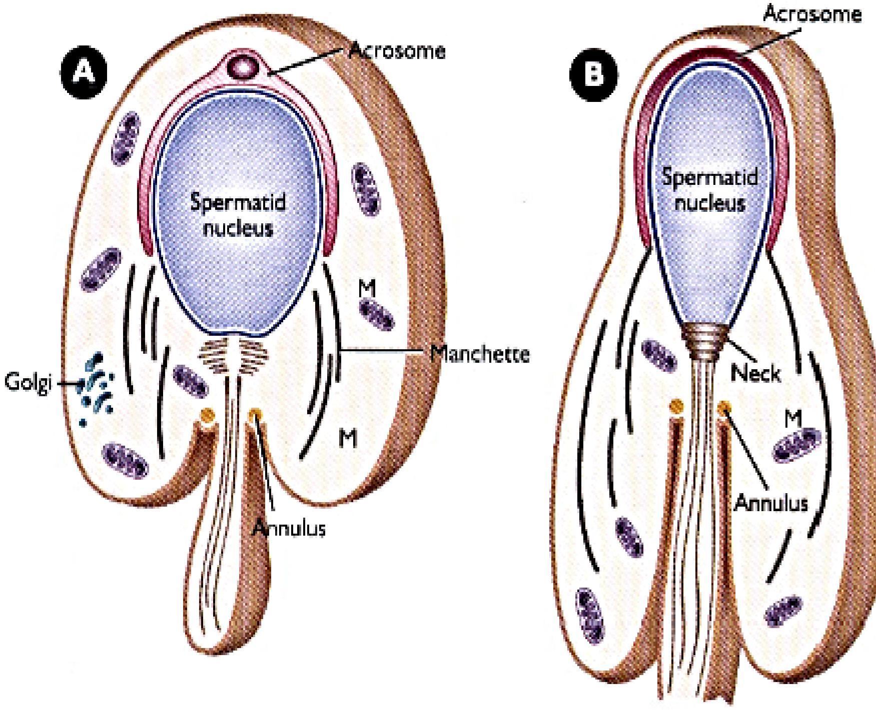
The Golgi migrates toward the caudal part of the cell. The distal centriole (DC) forms the axoneme (AX) or flagellum that projects away from the nucleus toward the lumen of the seminiferous tubule.

B

The acrosomic vesicle flattens and begins to form a distinct cap consisting of an outer acrosomal membrane (OAM), an inner acrosomal membrane (IAM) and the acrosomal contents (enzymes).

The acrosomal phase = nuclear and cytoplasmic elongation

The Acrosomal Phase



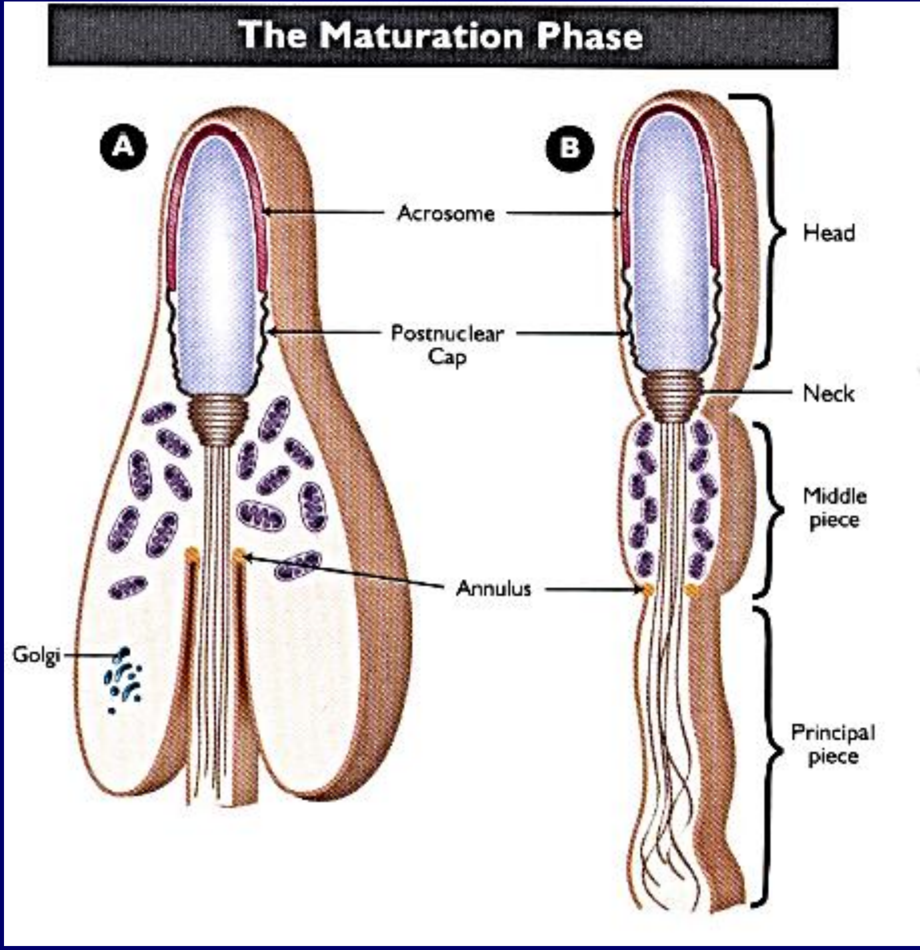
A

The spermatid nucleus begins to elongate and the acrosome eventually covers the majority of the anterior nucleus. The manchette forms in the region of the caudal half of the nucleus and extends down toward the developing flagellum.

B

The neck and the annulus are formed and the later will become the juncture between the middle piece and the principal piece. Notice that all components of the developing spermatid are completely surrounded by a plasma membrane. M = mitochondria.

The maturation phase = final assembly that forms a spermatozoon



A and B

Mitochondria form a spiral assembly around the flagellum that defines the middle piece. The postnuclear cap is formed from the manchette microtubules. The annulus forms the juncture between the middle piece and the principal piece.

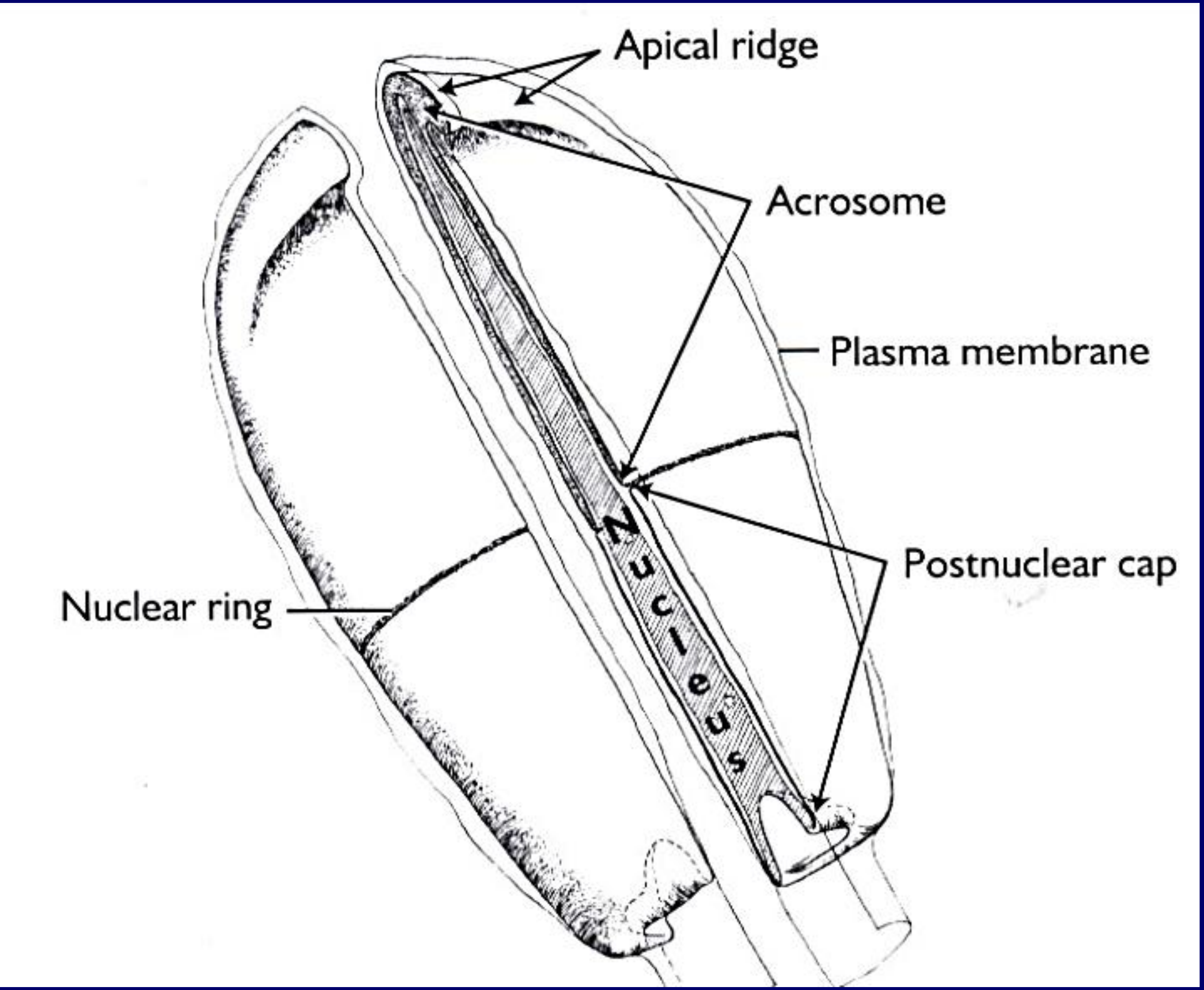
Spermatozoa = head + tail

Head = nucleus + acrosome + post-nuclear cap

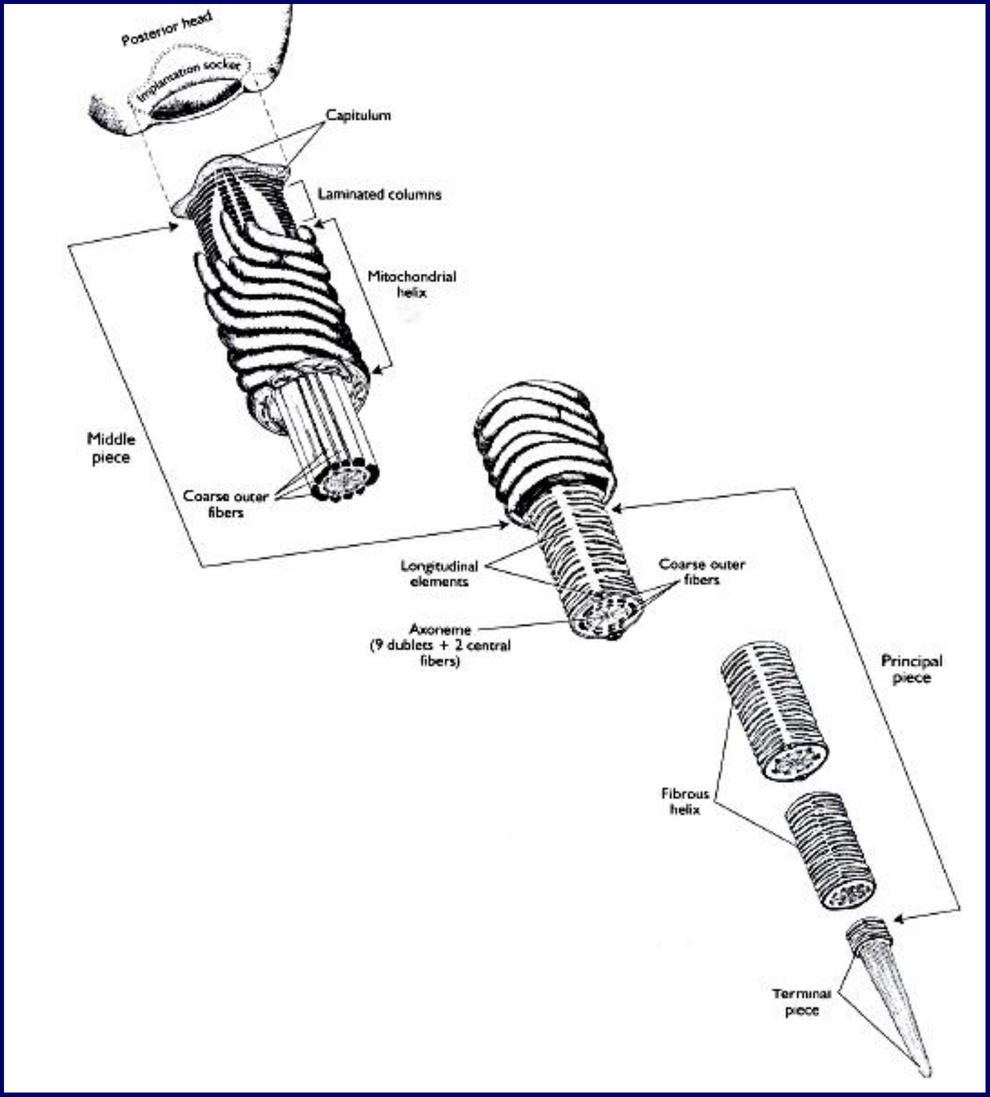
Tail = middle piece + principal piece + terminal piece

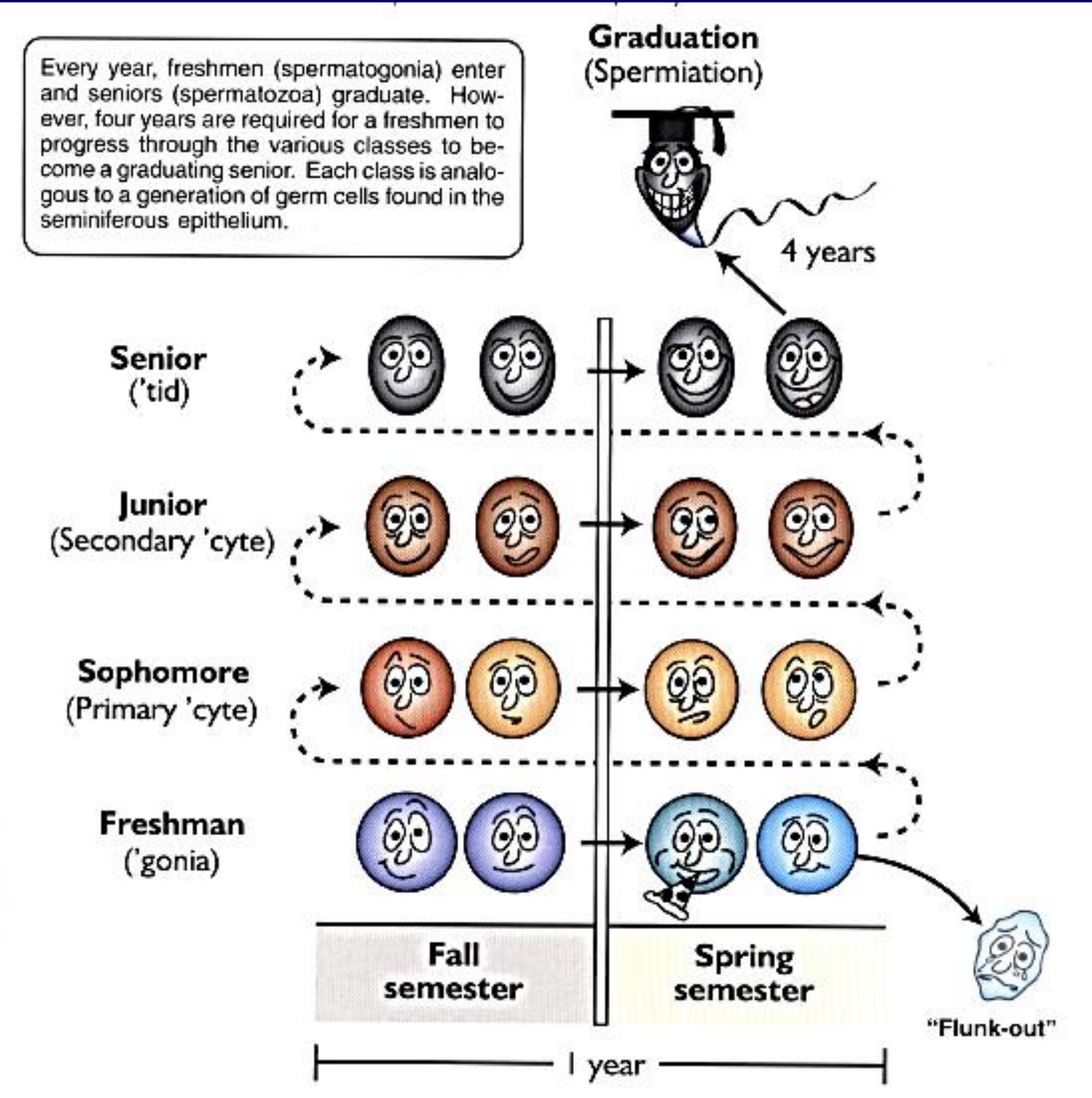
The anterior two-third of the nucleus is covered by the acrosome.

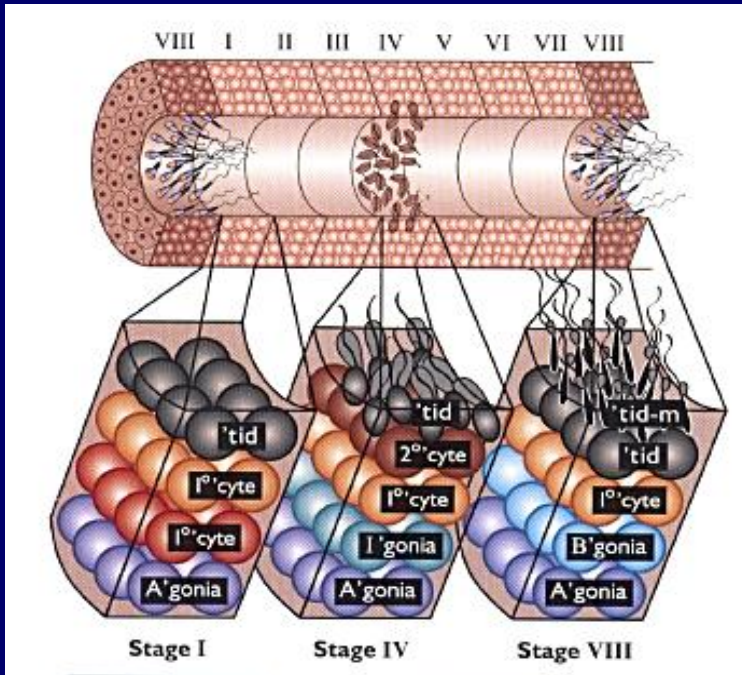
Acrosome contains hydrolytic enzymes(acrosin, hyaluronidase, zona lysin, esterase, acid hydrolases)



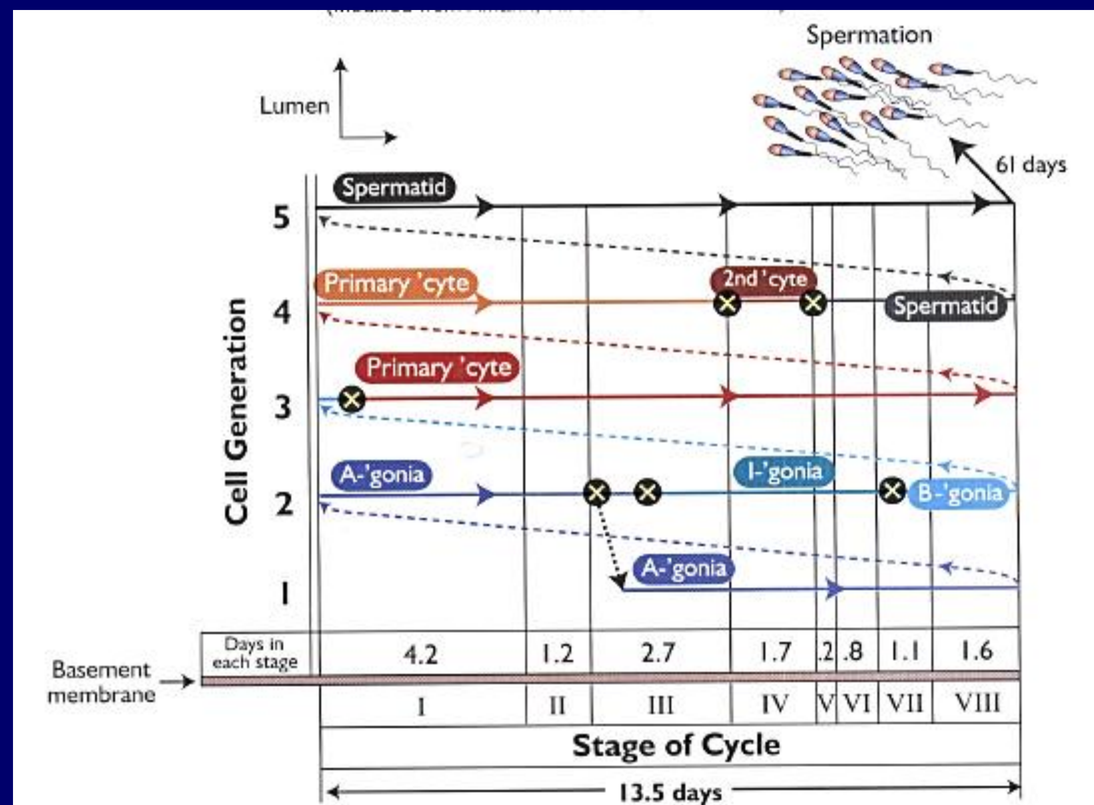
The sperm tail is a self-powered flagellum.




Every year, freshmen (spermatogonia) enter and seniors (spermatozoa) graduate. However, four years are required for a freshmen to progress through the various classes to become a graduating senior. Each class is analogous to a generation of germ cells found in the seminiferous epithelium.
Graduation
(Spermiation)
4 years
Senior
('tid)
Junior
(Secondary 'cyte)
Sophomore
(Primary 'cyte)
Freshman
('gonia)
Fall
semester
Spring
semester
"Flunk-out"
1 year



VIII I II III IV V VI VII VIII
'tid
I°'cyte
I°'cyte
A'gonia
'tid
2°'cyte
I°'cyte
I'gonia
A'gonia
'tid-m
'tid
I°'cyte
B'gonia
A'gonia
Stage I
Stage IV
Stage VIII



Spermation
Lumen
61 days
Spermatid
Primary 'cyte
2nd 'cyte
Spermatid
Primary 'cyte
A-'gonia
I-'gonia
B-'gonia
A-'gonia
Cell Generation
5
4
3
2
1
Days in each stage
4.2
1.2
2.7
1.7
.2
.8
1.1
1.6
Basement membrane
I
II
III
IV
V
VI
VII
VIII
Stage of Cycle
13.5 days

Table 10-1. Duration of the Stages of the Cycle of the Seminiferous Epithelium in Various Species

Stage	Bull	Ram	Boar	Stallion	Rabbit
I	4.2	2.2	1.1	2.0	3.1
II	1.2	1.1	1.4	1.8	1.5
III	2.7	1.9	0.4	0.4	0.8
IV	1.7	1.1	1.2	1.9	1.2
V	0.2	0.4	0.8	0.9	0.5
VI	0.8	1.3	1.6	1.7	1.7
VII	1.1	1.1	1.0	1.6	1.3
VIII	1.6	1.0	0.8	1.9	0.9
TOTAL[A]	13.5	10.1	8.3	12.2	11.0
SPERMATOGENESIS[B]	61	47	39	55	48

[A]Total days required for 1 cycle of the seminiferous epithelium

[B]Approximate days to complete spermatogenesis (spermatogonia to spermatozoa)

Table 10-2. Testicular Characteristics and Sperm Production Estimates of Sexually Mature Mammals

Species	Gross weight of paired testes (grams)	Sperm produced per gram of testicular parenchyma	Daily spermatozoal production
Beef Bull	650	11×10^6	6×10^9
Boar	750	23×10^6	16×10^9
Cat	21	16×10^6	32×10^6
Dairy Bull	725	12×10^6	7.5×10^9
Dog (16 kg body weight)	31	17×10^6	0.50×10^9
Man	35	4×10^6	0.13×10^9
Rabbit	6	25×10^6	0.20×10^9
Ram*	550	21×10^6	10×10^9
Rooster***	25	100×10^6	2.5×10^9
Stallion**	340	16×10^6	5×10^9

*in breeding season (shortening-day length), ** in breeding season (increasing day length), ***varies greatly with management and strain

Note!

Testicular Size is a Good Estimator of Sperm Producing Ability


PIX 2 PIX
.ORG