

SECTION 11 - REPRODUCTIVE SYSTEM

EXERCISE 11.1 OVARIAN CYCLE AS STUDIED BY A VAGINAL SMEAR OF A RAT

Approximate Time for Completion: 1 hour

Introduction

This exercise is designed to introduce students to the ovarian changes that occur during the mammalian reproductive cycle. This exercise can easily be combined with others in section 11 since 30 minutes out of the hour required for this exercise is free while the slides are in the stain. Since about six slides can be made using one rat and since the rats are not harmed by this procedure, the vaginal smear exercise requires only minimal vivarium facilities. If the lab instructor desires to change this exercise into a more long-term experiment, this can easily be done. For example, some rats can be previously ovariectomized, some can be ovariectomized and given daily estradiol replacements, and so on. It has been previously reported that a group of female rats will synchronize their cycles when placed near male rats (this is a pheromone effect). An experiment might be performed to test this observation.

Materials

1. Young female rats
2. Ether jar (large wide-mouthed jar with close-fitting lid), ether. This is optional.
3. Isotonic saline, cotton swabs (or, alternatively, eyedroppers may be used)
4. Giesma's stain (dilute concentrate 1:50) and absolute methyl alcohol in staining jars. Staining is optional.
5. Microscopes, clean microscope slides, coverslips

Textbook Correlations: Chapter 20 – Menstrual Cycle

Answers to Questions

1. estrus
2. estrogen
3. ovulatory
4. diestrus
5. menstrual
6. follicular
7. luteal
8. The estrous cycle is usually complete in 4 to 5 days. The cycle is roughly divided into four stages. (1) Proestrus – follicles mature under gonadotropic hormone stimulation; estrogen levels rise (corresponds to follicular phase). (2) Estrus – fluid enlargement of the uterus; estrogen secretion peaks as ovulation occurs (ovulatory phase). (3) Metestrus – corpora lutea secrete progesterone to prepare the uterus (luteal phase). (4) Diestrus – corpora lutea regress, estrogen and progesterone levels drop and the uterus regresses (menstrual phase).
9. (1) Menstrual phase – exfoliation of vaginal squamous cells and endometrium (follicular phase of the ovary).
(2) Proliferative phase – estrogen stimulates the growth of a new endometrium (follicular phase of ovary).
(3) Secretory phase – progesterone prepares the endometrium for implantation (luteal phase of the ovary).
10. The ovariectomized rat would be in continuous diestrus and its vaginal smear would consist entirely of leukocytes. This is because an ovariectomy removes the source of estrogen and progesterone. This condition would mimic the normal diestrus phase when the estrogen levels decline.

11. The vaginal smears would contain both leukocytes and cornified epithelial cells. This is because the estrogen and progesterone levels are now artificially high, as they are normally in the metestrus phase of the cycle when the corpus lutea secrete estrogen and progesterone.

EXERCISE 11.2 HUMAN CHORIONIC GONADOTROPIN AND THE PREGNANCY TEST

Approximate Time for Completion: 15 minutes

Introduction

This exercise is designed to introduce students to methods for determining pregnancy. Students actually perform a pregnancy test in the laboratory. This exercise could be combined with other exercises from section 11 for a complete laboratory period on the reproductive system.

Materials

1. Urine collection cup, proper urine disposal receptacle
2. Pregnancy kit, agglutination type (DAP test kit – Wampole, Qupid test – Stanbio, ICON II – Hybritech, or similar test)
3. Over-the-counter home pregnancy kit (e.p.t. or similar kit)

Textbook Correlations: Chapter 20 – Implantation of the Blastocyst and Formation of the Placenta

Answers to Questions

1. decrease
2. corpus luteum
3. human chorionic gonadotropin (hCG)
4. trophoblast cells of the blastocyst (embryo)
5. luteinizing hormone (LH)
6. LH is secreted from the anterior pituitary to form the corpus luteum from the empty ovulated follicle. Once formed, the corpus luteum secretes estradiol and progesterone during the luteal phase of the cycle. The estradiol and progesterone secreted during the luteal phase exert negative feedback inhibition of gonadotropin secretion from the anterior pituitary. As a consequence of the low LH, the corpus luteum dies at the end of an infertile menstrual cycle. If fertilization occurs the embryo secretes hCG that acts like LH on the corpus luteum and maintains it for the first ten weeks of pregnancy.
7. The pregnancy test is called an immunoassay because it is based on the combination (agglutination) of antibodies against hCG with hCG antigens. In the typical pregnancy test, particles of latex rubber (easily seen) were coated with hCG antibodies and mixed with samples of urine. If the urine contained the hCG antigen, the combination of hCG with the antibodies caused the particles of latex rubber to agglutinate and can be evaluated visually. Urine samples without hCG will not result in agglutinated latex particles.
8. Most pregnancy tests are not valid unless enough time has passed after conception because hCG is secreted in small amounts very shortly after conception. For these tests to be valid the levels of hCG in the blood must rise high enough for adequate amounts to appear in the urine and thus be within the sensitivity of the pregnancy test. Blood tests, however, are very sensitive and can detect extremely small amounts of hCG within 7-10 days after conception.
9. A man with a tumor that secretes hCG would have adequate blood levels of the hormone. He would give a positive result on a pregnancy test. Since hCG mimics luteinizing hormone (LH), the LH target cells in the interstitial cells of Leydig would be stimulated to release more testosterone. The rise in testosterone levels however, should exert negative feedback effects at the hypothalamus and anterior pituitary, thereby reducing the secretion of LH and FSH.

EXERCISE 11.3 PATTERNS OF HEREDITY

Approximate Time for Completion: 30 minutes

Introduction

This exercise is designed to introduce students to human genetics and heredity. In this exercise, students will test themselves for PTC taste, discuss patterns of inheritance of ABO and Rh blood typing antigens, discuss sickle cell anemia, and test for color blindness. All of these traits are heritable and students can use simple Punnett squares to determine their genotype for each of these traits.

Materials

1. Phenylthiocarbamide (PTC) paper (VWR Scientific Products, Ward's Biological)
2. Sickle cell turbidity test (Chembio Diagnostic Systems, Inc.); prepared slides of sickle cell anemia and normal blood, microscopes
3. Ishihara color blindness charts

Textbook Correlations: Chapter 3 – DNA Synthesis and Cell Division

Chapter 10 – Cones and Color Vision

Chapter 16 – Inherited Defects in Hemoglobin Structure and Function

Answers to Questions

1. autosomes
2. homozygous
3. heterozygous
4. sex-linked
5. phenotype
6. Sickle cell disease is inherited as an autosomal recessive trait. This means that those with the disease and display the sickle cell phenotype must be homozygous for the trait. The heterozygote does not display the disease but is a carrier of the trait. When two heterozygotes mate, one-fourth of the offspring will be homozygous and display the disease phenotype, one-half will be carriers, and one-fourth will be homozygous normal.
7. Color blindness is inherited as a sex-linked trait, whose gene is located on the X chromosome. Although recessive to the normal phenotype, a male will express color blindness if inherited on his only X chromosome. A female however, must be homozygous for the condition and have the trait on both X chromosomes to be colorblind. For this reason colorblindness is more common in men than in women. The normal female can be either homozygous normal or heterozygous (carrier).
8. a. 50%
b. 50%
9. The probability that their first child will have hemophilia is 25%; this is the same for the next child because the probability for each event is independent of the other events. The sex of the child is male. This is because hemophilia is inherited on the X chromosome and the male always gets his X chromosome from his mother (who in this case is a carrier for hemophilia). Females get one X chromosome from their mother and one from their father, who carries only the dominant normal gene.