LABORATORY ANIMAL MEDICINE AND SCIENCE SERIES II

RATS AND MICE:
Biology

V-9041

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AUDIENCE Veterinary and biomedical students, animal care technicians, and research investigators.

GOAL To familiarize the viewer with the taxonomy, anatomy, physiology, nutrition, and behavior of laboratory rats and mice.

OBJECTIVES At the conclusion of this program, you should be able to:

1. name genus and species of the laboratory rat and the laboratory mouse.
2. list four reasons why rats and mice are commonly selected and used in research.
3. describe the distinctive anatomical features of rats and mice.
4. give the approximate life span of the rat and the mouse.
5. list two forms of rodent diet.
6. describe the methods available for evaluating the stages of the reproductive cycle and assuring that mating has taken place.
7. recognize common behavioral traits in laboratory rats and mice.

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2. Program title — RATS AND MICE: Biology

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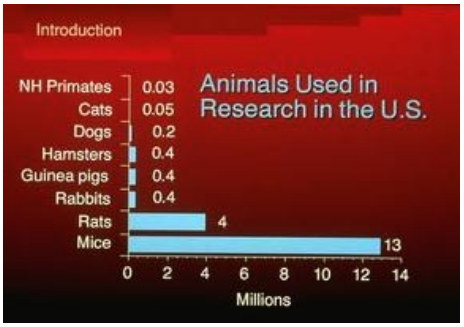
4. Section title — **INTRODUCTION**

5. Taxonomy — Rats and mice are grouped in the order *Rodentia*, suborder *Myomorpha,* family *Muridae*. The laboratory rat was derived from the wild Norway rat *Rattus norvegicus*. The laboratory mouse was developed mainly from the western European house mouse, and the taxonomic designation, *Mus musculus,* is a composite designation for several interbreeding species.

6. Numbers used



Rats and mice are the most commonly used animals in research and testing. This chart shows approximate numbers used in recent years (4). Rats and mice account for approximately 90% of all mammals used in scientific endeavors.

7. Attributes for research — The commercial availability of rats and mice, plus their small size, high reproductive rate, and minimal costs for purchase and maintenance, have made them the most studied and perhaps best understood laboratory animal species. In addition, they are also well understood and characterized anatomically, physiologically, and genetically.

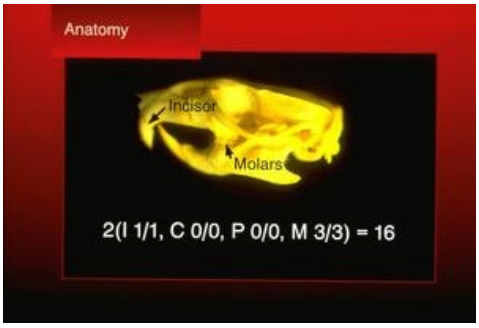
8. More attributes — Several stocks of rats and mice have withstood the process of inbreeding, allowing the commercial production of a large variety of inbred strains and providing the researcher with thousands of genetically similar individuals. A large number of mutant strains and stocks, with naturally occurring anatomical, physiological, or biochemical diseases, have been developed as animal models for similar conditions in humans and other animals. For a more detailed discussion of this topic, see *V-9039--Rats and Mice: Introduction and Use in Research, Part 1* (5).

9. Section title — **ANATOMY**

10. Physical appearance

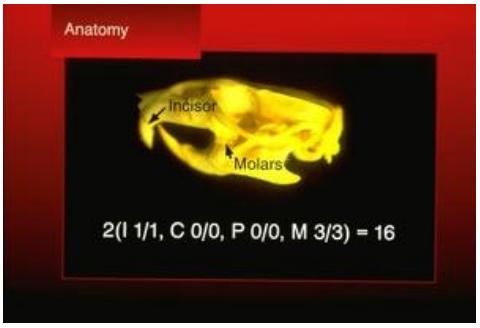
Rats and mice have short hair, long naked tails, erect rounded ears, protruding (exophthalmic) eyes, and pointed snouts with long whiskers (vibrissae). Both species have five toes on the fore- and hindfeet, but the first digit is reduced in size on the forefeet. A variety of coat colors are found in rats and mice, including albino, solid colors, and hooded patterns.

11. Skeletal system



The vertebral formula of laboratory rats and mice is identical and illustrated in this image of a rat skeleton. Bone maturation occurs slowly in these animals and ossification is not complete until near the end of the first year of life.

12. Oral cavity



This image of a rat skull illustrates the relative position of rodent teeth. Rats and mice have monophydont dentition, that is, they have only one set of permanent teeth. These teeth consist of two incisors and six molars in both the upper and lower dental arcades. The dental formula is: 2(I 1/1, C 0/0, P 0/0, M 3/3)= 16. The incisors are open rooted (hypsodontic) and erupt and grow continuously throughout life, while the molars are permanently rooted (brachiodontic) and do not continue to grow. Tonsils are not present in the oropharynx of rats.

13. Normal incisors

This image shows normal rat incisors. The incisors normally grow, calcify, and erupt throughout life. Their occlusal surfaces are continuously worn down by abrasion at the point of apposition of the upper and lower incisors. The tips of the lower incisors are positioned slightly posterior to the tips of the upper incisors, with each sharpened to a chisel-like configuration by their integrated occlusion.

14. Overgrown incisors

This image shows a rat with overgrown incisor teeth caused by malocclusion. With malocclusion, the points of apposition of the occlusal surfaces of the upper and lower incisors are malaligned, the teeth fail to wear down, and they overgrow. Malocclusion may follow the fracture or loss of an incisor, inappropriate nutrition or soft diets that do not contribute to normal tooth wear, genetic predisposition.

The overgrown teeth can be trimmed periodically to prevent damage to adjacent soft tissues but trimming may not correct the malocclusion. If genetic predisposition is suspected, the colony should be selectively bred to eliminate the trait.

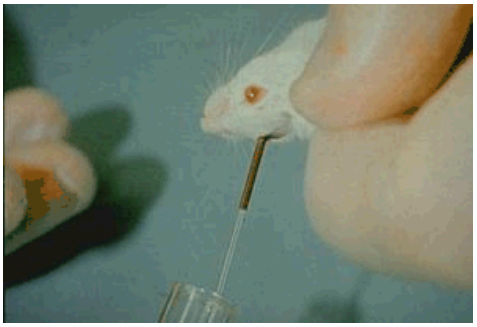
15. Nervous system

As shown in this image, the cerebral hemispheres of rats and mice are smooth, lacking the folds and grooves (gyri and sulci) seen in higher order species. The peripheral nervous system is composed of 34 pairs of spinal nerves: 8 pairs of cervical, 13 pairs of thoracic, 6 pairs of lumbar, 4 pairs of sacral, and 3 pairs of caudal nerves.

16. Eye

The eyes of both rats and mice protrude from the head, as illustrated by this normal rat. Consequently, the eyes are subject to injury and ulcers, scarring, or opacity of the cornea may be seen.

17. Vasculature of orbit

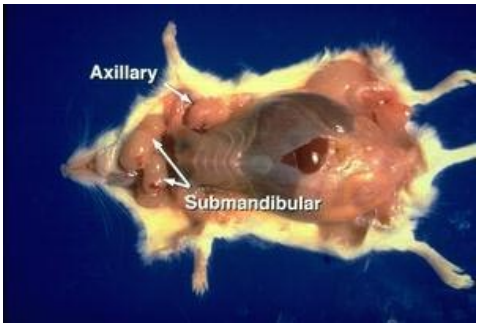


Mice have extensive periorbital venous sinuses behind the globe of the eye, while rats have a more discrete plexus of vessels (7). These vascular structures are useful for periorbital (orbital sinus) blood collection and pose less risk than cardiac blood collection in these species. Following anesthesia, a heparinized microhematocrit capillary tube can be inserted into the medial canthus of the eye until the bony orbit is contacted, then withdrawn slightly to allow the blood to flow through the tube and into a collection vessel.

18. Harderian gland

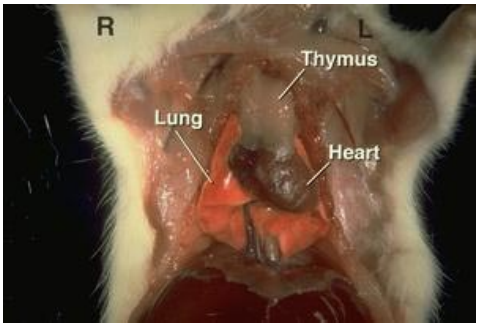
The horseshoe-shaped Harderian gland (arrow) is located within the bony orbit. Secretions from this gland include the reddish pigment porphyrin, and hypersecretion in stressed rats is often referred to as “red tears” (chromodacryorrhea). These secretions may be observed around the eyes, around the nares after passage through the nasolacrimal duct, or on the forelimbs after the animal has groomed its face. Stressors may include disease, environmental, social, or experimental factors.

19. Lymph nodes-



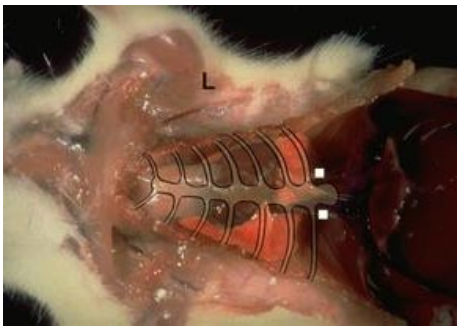
Lymph nodes are located superficially, subcutaneously or between muscles; and deep, within the abdominal or thoracic cavities. They may be difficult to palpate or to visualize in healthy animals, but may be enlarged in animals with localized or systemic infections or in neoplastic conditions, such as lymphosarcoma illustrated in this image.

20. Thoracic viscera



This image of an opened rat thoracic cavity shows the relative positions of the lungs, the heart, and the thymus. A knowledge of the location of thoracic visceral structures is valuable for accomplishing a variety of experimental techniques (i.e., thymectomy, pulmonary lobectomy, coronary arterial occlusion, and cardiac blood collection). The thymus has paired, triangular shaped lobes and is located ventral to the trachea at the thoracic inlet. The gland is large in young animals, reaches maximum size at sexual maturity, and then regresses in size.

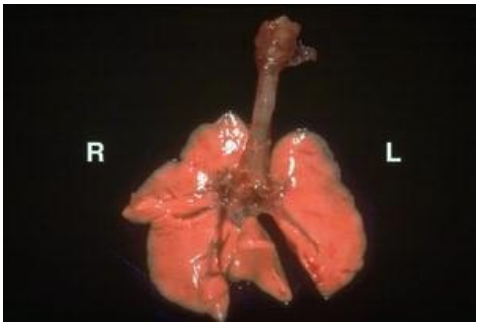
21. Cardiovascular system



As seen in this image of a rat, the heart is located along the midline of the thorax surrounded by the lobes of the lung. The apex lies close to the diaphragm (allowing cardiac puncture for blood collection, with the needle inserted cranially next to the xiphoid process (squares)). Cardiac puncture can also be performed by inserting the needle between ribs 3 to 5 on the animal's left side. The vasculature is comparable to that in other mammalian species with the following exceptions:

1) rats and mice have two anterior vena cavae;
2) the right precava in rats empties directly into the right atrium, while the left precava joins the azygous vein and ultimately empties into the caudal vena cava;
3) both anterior / superior vena cavae in mice empty into the right atrium.

22. Lungs

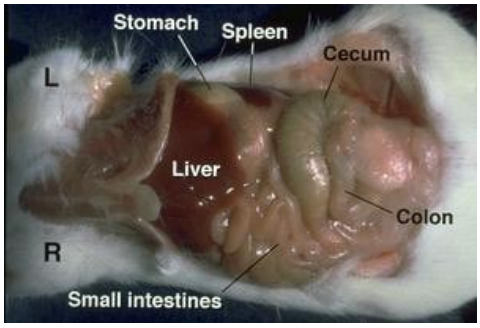


This ventral view of the lungs shows the arrangement of lung lobes in the rat and the mouse: there is a single left lobe, and four right lobes—the cranial, medial, caudal, and postcaval lobes. The trachea of both species is composed of incomplete,'C'-shaped, cartilaginous rings extending from the larynx into the thoracic cavity. The trachea branches to form the left and right bronchi, composed of completely circular cartilaginous rings.

23. Brown fat

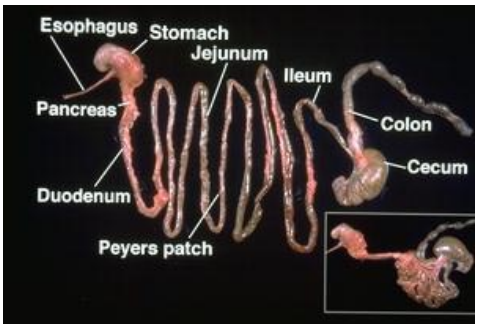
Brown fat, referred to as the 'hibernating gland' or multilocular adipose tissue, is present in rats and mice, despite the fact that they are not hibernating species. The greatest accumulation of this pigmented fat is found between the scapulae (arrows), but it can also be observed in the axillary and cervical regions, along the jugular veins and thoracic aorta, at the hilus of the kidney, and along the urethra. Also note that the pigmentation may appear light, as in this photo, or as a darker brown. Although rats and mice are not hibernating animals, this tissue may serve as an economizer of proteins by ensuring utilization of reserve carbohydrates and fats, and plays a major role in thermogenesis during cold exposure (1).

24. Abdominal viscera



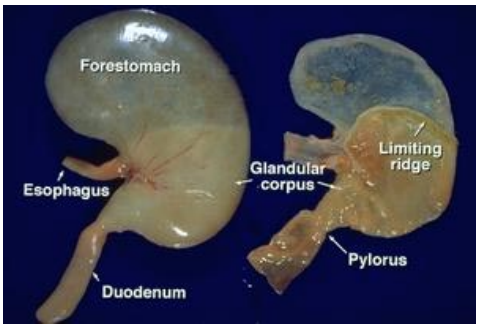
In this ventro-dorsal view of the abdominal viscera of a mouse, the liver can be seen as the organ adjacent to the diaphragm. The liver accounts for 4 to 5% of the total body weight in adults and is composed of four lobes in both rats and mice. The median lobe, which is not visible here, surrounds the gall bladder in mice. The rat has no gall bladder. The spleen lies along the greater curvature of the stomach, and the small intestine is seen as a folded mass on the right side. The cecum originates at the junction of the ileum and colon and is usually seen as an ingesta-filled blind pouch extending across to the left side of the abdominal cavity.

25. Gross



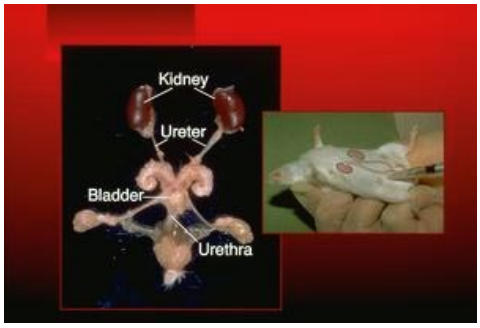
In this image, the gastrointestinal tract of a rat has been removed from the abdomen, the mesentery cut away, and the individual structures labeled. The smaller view (inset) shows the more natural arrangement. The cecum has a rumen-like function, with the resident microbes aiding in digestion of cellulose and other plant materials. In germ-free animals, the cecum will become enlarged and distended with ingesta. Though rat and mouse cecae lack a vermiform appendix, they both have a focal accumulation of lymphoid tissue near the apex of the cecum, analogous to the appendix in humans. Focal lymphoid aggregates, Peyer's patches, can be found dispersed along the length of the small intestine.

26. Dissected stomach



This image presents gross dissections of a rat stomach with portions of the esophagus and duodenum attached. The one on the right is filled with ingesta; the one on the left has been opened and emptied. The esophagus in rodents is lined with keratinized epithelium and is attached at the lesser curvature of the stomach, near the 'limiting ridge.' The cardiac sphincter is located at the terminus of the esophagus. The stomach of rats and mice is divided into two regions, both grossly and histologically. The nonglandular forestomach is lighter in color and thin-walled, compared with the pinker, thicker-walled glandular corpus that empties into the duodenum. A 'limiting ridge' separates the two regions and prevents vomiting in rats but does not prevent ingesta from moving freely from one side to the other. The pyloric sphincter is located at the junction of the stomach and the duodenum

27. Urinary system

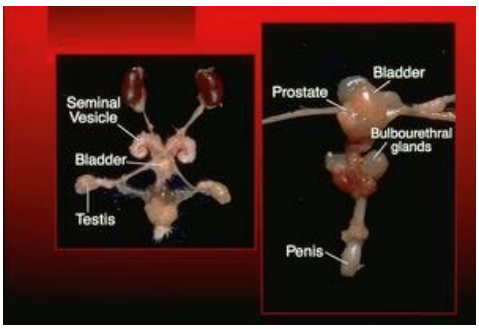


The urinary system is composed of the paired kidneys and ureters, the urinary bladder, and the urethra. The right kidney and adjacent adrenal gland are located more cranial than the left kidney. The kidney is classified as unipapillate, having one papilla and one calyx, which enter the ureter directly. The urinary bladder is located in the midline in the caudal portion of the abdomen and must be avoided during intraperitoneal injections (inset). The urethra opens in the penis in males and in the genital papilla near the base of the clitoris in females (rather than in the vagina or vestibule as is the case in domestic animals and humans).

28. Male genitalia

This view of the ventral surface of a male mouse shows the scrotum and genital papilla with relation to the anus. The testes lie in the scrotal sac on both sides of the ventral midline between the opening of the prepuce and the anus. The inguinal rings are open throughout life, allowing the testicles to be withdrawn into the abdomen. The penis is located within the genital papilla (prepuce), and contains a bony process, the os penis.

29. Male accessory sex glands

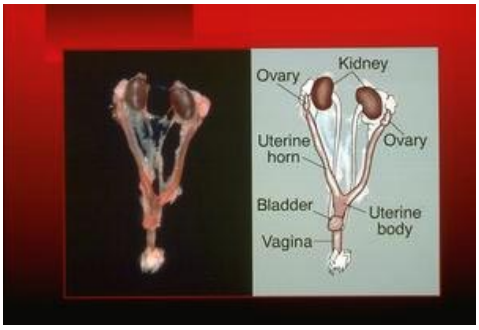


The accessory sex glands and urogenital system of a male mouse are shown in this image. The largest accessory male sex glands are the paired, elongated, curved vesicular glands, often referred to as the seminal vesicles, which empty into the urethra near the neck of the bladder. The coagulating glands are located within the lesser curvature of the seminal vesicles. The view on the right shows the prostate gland which has two pairs of lobes, dorsal and ventral, and the paired bulbourethral glands. The prostate gland is located along the body of the penis. The mixture of fluids from these glands forms the vaginal (sperm) plug. Rats and mice may be castrated for experimental endocrine and other studies.

30. Female genitalia

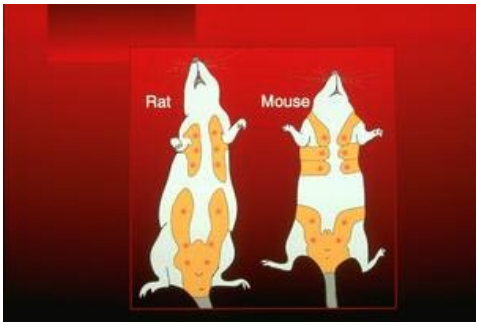
This image illustrates the external genitalia of female rats and mice, consisting of the genital papilla, containing the clitoris and urethral orifice, and the vulva, containing the vaginal opening. A vaginal closure membrane is usually present in immature females and disappears prior to sexual maturity. The anogenital distance (from the genital papilla to the anus) is shorter in females than in males.

31. Female reproductive system



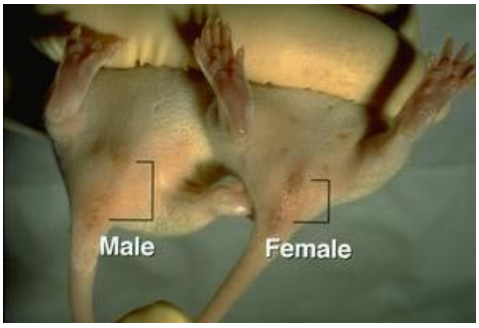
This dissected specimen of the female urogenital system of a rat demonstrates that the ovaries are close and slightly caudal to the kidneys and usually embedded in fat. The uterus is bicornuate, that is, it has two horns that merge into the body of the uterus. The rat has two cervices, one for each horn, while the mouse has only a single cervix at the junction of the body of the uterus and the vagina. The uterus and ovaries lie close to the dorsal body wall, allowing a dorsolateral approach for removal of the ovaries or uterus (ovariectomies and ovariohysterectomies) for a variety of research purposes. The vagina is a short canal leading to an opening caudal to the genital papilla and cranial to the anus. The types of cells present in a vaginal smear can help identify the current stage of the estrous cycle of the female. The large size of the pronucleus in the ovum of the mouse facilitates microinjection procedures for transgenic mouse production.

32. Mammary tissue



As these illustrations show, rats have six pairs of nipples; mice have five pairs. Mammary tissue is widely distributed in both species and may extend up onto the lateral and dorsal areas of the abdomen and thorax. It is, therefore, possible to find mammary tumors on the back of a female.

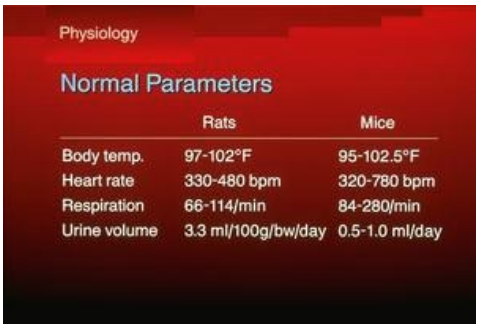
33. Gender identification of neonates



As seen in this image, one can identify the gender of neonates by comparing the relative distance between the genital papilla and the anus, referred to as the anogenital distance, which is greater in males.

34. Section title **PHYSIOLOGY**

35. Normative data



This image shows the physiologic parameters for adult rats and mice. (Refer to Appendix I for additional values.) Some variations among these values may occur between groups of rodents due to stock or strain differences, source of the animals, and research conditions.

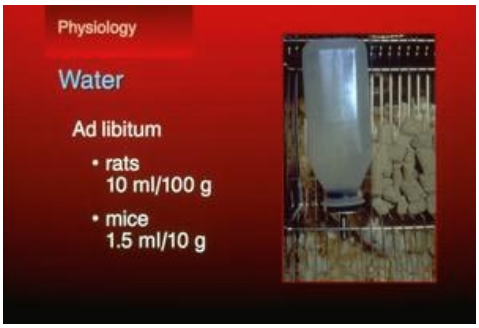
36. Diet

The composition of the diet fed to laboratory animals is of major importance in maintaining them in good health and yielding consistent data. In experimental studies of nutrient excess and deficiencies, rats have been used much more frequently than mice and much more is known about their nutrient requirements. Experience has proved, however, that both rats and mice can be maintained satisfactorily on a low fiber diet (5 %) and are typically fed a commercially pelleted diet referred to as “rodent chow” (3). Rats consume approximately 5 g per 100 g of body weight in food daily and mice 12 g per 100 g of body weight per day.

37. Types of diet

Rodents can be fed a pelleted diet or a powdered (mash) diet. The pelleting process requires heat that might adversely affect chemicals being studied, but test chemicals can be mixed into mash diets for research purposes. Barrier conditions and maintenance of immunocompromized animals (i.e., SCID or nude mice, transgenic animals) may require that caging, bedding, water, and feed be autoclaved so that animals are not exposed to infectious agents. Autoclaving can also affect levels of nutrients within the diet, therefore, commercially available autoclavable chow is formulated with higher levels of vitamins to ensure that an adequate level of vitamins will remain after autoclaving. It is beneficial to purchase rodent chow from feed manufacturers producing “open-formula diets” composed of natural products but with specification of the exact ingredients (2). The nutritional consistency of these diets over time eliminates one variable in the research process.

38. Water



Fresh, clean water free from bacterial or chemical contamination should be available ad libitum. Rats consume approximately 10 ml/100 g of body weight per day; mice consume 1.5 ml/10 g of body weight per day. Water can be delivered to animals via water bottles or through an automatic watering system.

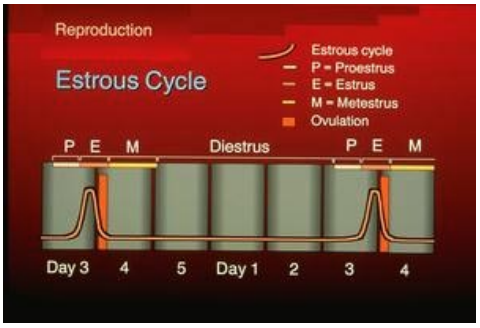
39. Life span and

Rats generally live for two to three years, but some that were fed calorically restricted body weight diets lived longer (8). The life span of mice is usually two years. Mice maintained in a pathogen-free environment may outlive others raised and housed in conventional environments because they are not exposed to the detrimental effects of subclinical diseases. Adult male rats weigh 300 to 500 g and females weigh 250 to 300 g. Male mice will weigh 20 to 40 g at maturity and females 25 to 30 g. (See Appendix 2 for additional parameters.)

40. Sexual maturity — The age at puberty varies between the species, strain or stock of rats and mice. In general, rats reach puberty around 50 to 60 days of age, while mice reach puberty around 28 to 49 days of age. The first fertile mating in mice generally occurs around 7 to 10 weeks of age, while rats are usually bred after 9 weeks of age.

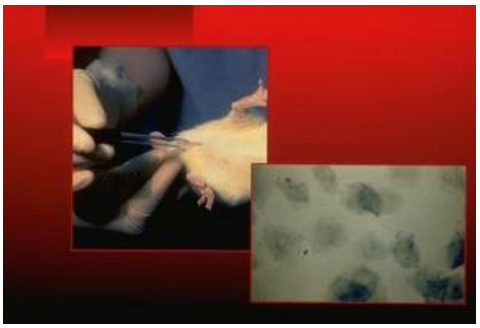
41. Section title — **REPRODUCTION**

42. Estrous cycle



Rat and mouse females are polyestrus, breeding throughout the year. They exhibit regular estrous cycles, lasting approximately 5 days in the rat, and 4 to 5 days in the mouse. Estrus or ”heat,” the period of sexual receptivity of the female, usually occurs spontaneously at night after day 3 of the cycle and lasts for 12 to 14 hours. Ovulation occurs spontaneously within 8 to 12 hours after the onset of estrus.

43. Cell monitoring of estrous cycle



The stages of the estrous cycle can be determined by performing daily vaginal lavage using sterile saline(shown) and microscopically examining the cell types during the cycle (staining with methylene blue will facilitate identification of cornified epithelial cells present during estrus). This procedure facilitates breeding of animals on the day of ovulation and producing timed-pregnant animals of known gestational age of the fetuses.

44. Mating

When indicators of ovulation are detected by vaginal cytologic examination, females are placed with males for breeding. Receptive females exhibit lordosis, a rigid posture with the hindquarters elevated when downward pressure is applied to the pelvis (as demonstrated by the female at the top). Mating usually occurs during the dark phase of the photoperiod. Increasing the light phase of the photoperiod to 14 hours per day enhances mating and reproductive rates. Females can also be bred during a postpartum estrus that occurs between 14 and 28 hours after parturition.

45. Vaginal plug

Successful mating in rats and mice is verified by the presence of a vaginal plug in the females. The vaginal plug is formed from the mixing of vesicular and coagulating gland secretions at the time of ejaculation. Vaginal plugs may be retained in mice for 16 to 48 hours, but often fall out of rats within hours after breeding. Direct observation of the vaginal plug in the vagina or its presence in the waste pan beneath suspended wire caging is a good indication that mating took place. Another technique utilized to detect mating is vaginal lavage and microscopic examination for the presence of sperm. Presence of the plug does not guarantee that an animal is pregnant, thus mated animals should be observed and later palpated to confirm pregnancy.

46. Gestation / parturition

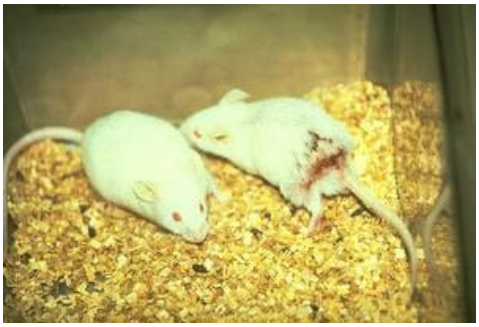
The length of the gestation period may vary with the strain or stock of animal. The range in rats is from 21 to 23 days (usually averaging 21 days) and 18 to 21 days in mice (19 day average). Periodic stretching and extension of the hindlegs signals impending parturition; for delivery, females stand in a semi-crouched position. Fetuses are delivered with either head or breech presentation. The female usually eats the placenta, then turns her attention to the pups. Delivery of all pups may take 1 to 4 hours.

47. Section title **BEHAVIOR**

48. Handling

Rats and mice are nocturnal and, if disturbed during the daytime, may bite the handler. Both rats and mice can be "gentled" by careful handling when they are very young. Once gentled, they are easier to handle during husbandry and research procedures and less stressed by the handling. Animals familiarized with handling or experimental procedures have a higher pain tolerance threshold, and are less stressed by experimental conditions. The *Guide* discourages long term restraint of animals because it is stressful. Biotelemetry systems and tether systems can be utilized to allow greater freedom of movement for animals with minimal distress.

49. Fight wounds



Male mice housed together may fight and may be injured or killed. If the aggressor is removed and housed individually, fighting is reduced or stopped. Male rats are less inclined to fight when grouped together.

50. Behavior

Some dominant females may groom their cage mates excessively or nibble their hair, referred to as barbering (shown in this image). This form of hair loss must be differentiated from endocrine or parasitic causes of hair loss. Both rats and mice are sensitive to pheromonal changes within their environments, and changing bedding or introducing new animals into the room may affect the behavior and physiology of the inhabitants. Other physical, biological, and social factors can influence experimental integrity by affecting food and water consumption, reproductive performance, drug metabolism and other altered physiologic parameters (6).

51. Conclusion

This concludes this autotutorial session on the biology and use in research of rats and mice. There are six programs in this series:

- V-9040 RATS AND MICE: Use in Research
- *V-9041 RATS AND MICE: Biology*
- V-9042 RATS AND MICE: Care and Management
- V-9043 RATS AND MICE: Bacterial and Mycotic Diseases
- V-9044 RATS AND MICE: Viral Diseases
- V-9045 RATS AND MICE: Parasitic Diseases

52. ACLAM credits

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APPENDICES

APPENDIX 1

NORMAL PARAMETERS

	Rats	Mice
Lifespan	2-3 years	2 years
Body Temperature	97-102°F (99.5°F)	95-102.5°F (97.5°F
Heart rate	330-480 bpm	320-780 bpm
Respiration	66-114/min (85/min)	84-280/min (163/min)
Urine volume	3.3/100g body wt/day	0.5-1.0 ml/ day
Total BTU/hour/animal	4.3	0.6
Body weight	250-500 g	25-40 g

APPENDIX 2

NORMAL ADULT PARAMETERS

	Rats	Mice
Age at puberty	50-60 d	35d
Minimum breeding age	55-90 d	M~60 d; F~50-60d
Food consumption	5g/100g bwt/d	12g/100g bwt/d
Water consumption	10ml/100g bwt/d	1.5ml/10g bwt/d
Mature body weight	M~300-500g	M~20-40g
	F~250-300g	F~25-30g

NOTES