

Laboratory Animal Production & Management

Notes:

Krishna Sharma

Assistant Professor

Department of LPM

FAVF, AFU

Laboratory animals

A laboratory animal is a small, cooperative, and cost effective species with physiological and anatomical characteristics that closely resemble humans, making it suitable for biological and medical research.

Importance: They are used to study disease mechanisms, test new drugs and treatments, study genetics and inheritance, and explore toxicological and pharmacological effects of chemicals.

Types: Mice, Rats, Hamsters, Ferrets, Guinea Pigs, Rabbits, Monkeys are popular Laboratory animals.

Classification of Laboratory animals:

- ❖ **Conventional animals:** Animals derived and grown in a general environment but free from diseases communicable to humans, like mycobacteria, Dermatophytic fungi, Pasteurella, and Sarcoptesscabiei.
- ❖ **Gnotobiotic animals:** - Animals with known microbes

such as viruses, bacteria, fungi, and protozoa.

❖ **Specific pathogen-free animals:** - An animal that is made free from a specific or particular microbe is known as SPF animals.

❖ **Germ-free animals:** An animal which don't have any demonstrable microbe is known as a germ-free animal.

Ethical considerations

Strict ethical guidelines govern research using animals. Institutions should follow the 3Rs principle:

Replacement – Use alternatives whenever possible.

Reduction – Use the minimum number of animals needed for valid results.

Refinement – Minimize pain and distress, and improve animal welfare.

Mice (*Mus musculus*): -

- They are smaller in size, have early puberty, high fecundity, short gestation, high position in the evolutionary scale. Due to these specialties, about 60-70% of laboratory animals are used as mice.
 - Uses: Genetic, cancer, immunology, infectious diseases. •
- The common strains are **Swiss Albino, Kausali, NIN, CBA, C-57, Bulb-S, CH-3** etc. **Swiss Albino** is the most popular.



Leeboot effect:-

When a large group of females is housed together, they go into anestrus due to pseudopregnancy, which is known as the Lee boot effect. Pheromonal signals from other females lead to anestrus or prolonged diestrus.

Whitten effect:-

When such females are housed again with males, within 72 hours they will exhibit regular estrus, which is called as Whitten effect. Androgen-dependent pheromones in male urine stimulate the onset of estrus in females.

Significance: Social environment affects reproductive physiology.

Mice and Rat:

Nature:- Prolific breeder

Puberty: - 4 to 6 weeks

Mating:- 8 to 9 weeks, early mating

reduces fertility. Estrous duration: - 4 to 5 days.

Gestation period:- 20 to 22 days.

Birth weight of young ones:- 1 to

1.5gm Litter size: - 10 to 12

Weaning weight:- 10 to 12gm and age 21 days.

Rat(*Rattus norvegicus*) :-

Nearly 2-3 times bigger in size than mice,
intelligent and social animal, short gestation,
prolific breeder, early puberty and high fecundity.

Uses: Neuroscience, behavior, cardiovascular
research and toxicology



Rat...

❖ The rat has a blocky body with comparatively rough hair, long pointed snout, long ears and sharp teeth.

❖ The body length is around 14-16cm, tail is 16-20cm. ❖ Adult weight 100-250gm.

❖ The common strains are- **Wister,**
Sprague-Dawley, Charles Foster, and Long Evans.

❖ **Detection of successful mating:**

Presence of sperm in vagina smear or copulatory

plug (solidified semen) in vagina.

Fur of rat is coarse.	Fur is Smooth
Tail of rat is thicker and well serrated (notches on edge)	Thinner and non-serrated
Skin of rat is thick	Thin
Rat is bigger in size and heavy in weight	Mice is smaller and light in weight

Guinea Pig (*Cavia porcellus*): -

● It is a prolific breeder, easy to rear and breed in captivity. ● Gestation period is comparatively little bit longer than that of rats & Mice.

● It needs supplementation of vitamins.C in the diet and ● More susceptible to T.B.& Anaphylactic shock.





- Gestation: - 60 to 75 days.
- Birth weight:- 45 to 50gm.
- Litter size:- 5 to 6.
- Weaning weight:- 160 to 230gm
- Puberty: - 4 to 5 weeks. Mating age: - 9 to 11 weeks.
- Weight at mating: - Male- 900 to 1500 gm
Female- 700 to 1300 gm.
- Estrous sign: - Hipswaying, mounting activity, unsteady

movements.

- Mating:- Polygamous
- Furrowing: - Communal, i.e., a group of females farrows in a common pen.

Guinea Pig...

- ❖ **Peruvian, English and Abyssinian** are popular strains.
- ❖ They are short rodents without tail. Head is blunt and rectangular. The neck is thick and so short merged with body.
- ❖ The hind legs are longer than forelegs.
- ❖ The weight of male is 800-1600gm and in females 700-1300gm.

Hamster: -

● Syrian and Chinese are two popular varieties of hamster used in various research, specifically for Riboflavin and Vitamin. E research. ● Chinese hamsters are

commonly used in research on diabetes due to the high incidence of diabetes mellitus in them.

● It has a low (22) chromosomal number, due to which they are useful in cytological study, tissue culture, genetics & radiation research.





Hamster...

Puberty: - 4 to 6 weeks.

Mating age: - 8 to 12 weeks.

Weight at mating: - Syrian- 80 to 90gm
Chinese- 35 to 40 gm

Estrus duration: - 4 to 5 days.

Mating: - Hand mating is followed

Gestation period: 16 to 18 days.

Litter size: - 5 to 7

Weaning age: - 20 to 25 days and weight 15gm.

Males and females are housed in separate quarters, brought together only at the time of mating and separated again immediately after mating; otherwise, females may cause serious injuries to the tail and testes of males.

Estrus sign: - Stringy, translucent mucus is extruded initially, turning into creamy white discharge in the later period with odour. A well-receptive female exhibits a typical lordosis posture (Crouching with hind quarters raised). Mating usually takes place on the 4th day.

❖ An adult Syrian hamster is 16cm in length and weighs 90-125gm. ❖ The Chinese hamster is only 9cm in length, weighing 55gm. Hence, also known as the Dwarf hamster.

❖ Chinese hamsters have a dorsally grey colour with a black strip in the centre.

❖ Syrian hamster are golden brown dorsally with little black

ventrally.

- ❖ **Breeds:** Syrian and Chinese are two popular strains. ❖ They have a chunky body with short legs, a fluffy tail, loose skin with dense, short, and soft fur.
- ❖ The front legs have four toes, whereas the hind legs possess five toes.



Ferrets (*Mustela putorius furo*)

- They are increasingly used in biomedical research due to their similar respiratory anatomy and physiology to higher mammals, making them an excellent model for studying respiratory diseases, especially viral infections like influenza and COVID-19.
- They show similar clinical symptoms (fever, sneezing,

nasal discharge) when infected with respiratory viruses.

- Ideal for pathogenesis, transmission, vaccine efficacy, and antiviral testing.

Characteristic Details

Lifespan 5–8 years (can be longer with care) Adult Weight

Males: 1.0–2.5 kg; Females: 0.5–1.0 kg Body Temperature

37.8–40.0°C

Heart Rate 200–250 bpm

Respiration Rate 33–36 breaths/min

Gestation Period ~42 days

Litter Size 6–8 kits (young ferrets)

Popular breeds Albino, Sable, Black Sable, Cinnamon,



Rabbits (*Oryctolagus cuniculus*) •

They are among the oldest and most commonly used non-rodent laboratory animals in biomedical research. •

Their moderate size, docile nature, and physiological characteristics make them ideal for many types of studies, especially in :

- Toxicology: Skin irritation, dermal toxicity, pyrogen testing
- Immunology: Polyclonal antibody production •

Ophthalmology: Eye irritation and corneal studies •

Cardiovascular studies: Atherosclerosis and cholesterol metabolism

Common breeds of Rabbits are: New Zealand White (NZW), Dutch Belted, Californian, Flemish Giant, Polish

Non-Human Primates

Non-human primates (NHPs) are members of the primate order, excluding humans. They include monkeys, apes, and prosimians. They are biologically and genetically close to humans, which makes them valuable models for studying human biology and diseases.

- Complex Behavior and Cognition
- Disease Models
 - They can model human infectious diseases (like HIV/AIDS, tuberculosis), neurological disorders, cardiovascular diseases, and vaccine development more accurately than rodents.
- Pharmacokinetics and Toxicology
- Genetic Similarity to Humans
 - NHPs share about 90–98% of their DNA with humans, making

their physiology, immune system, and brain functions very similar.

Parameter	Rhesus Macaque (<i>Macaca</i>	<i>mulatta</i>) Average birth Cynomolgus Monkey (<i>Macaca</i>	<i>fascicularis</i>) Baboon (<i>Papio spp.</i>)
weight	400–600 g	300–500 g	900–1,200 g
temperature	37.5–39.5°C	37–39°C	37.8–39.5°C
Typical litter size	1	1	1
Gestation period	~165 days	~160 days	~180 days
Age at sexual maturity		2.5–4 years (females)	3–4 years
Menstrual Cycle			4–6 years

length ~28 days ~28 days ~32–35 days **Lifespan in**

captivity 25–30 years 25–30 years 30–45 years

Biological & Physiological data of laboratory animals:

BirthWeight(gm)	1-1.5	6-7	70-100	20	50-70
Body weight (adult gm)	30-40	250	800	80-90/35-40	1500-5500
Daily feed consumpt.(gm)	5-6	15-20	45-50	10-15/10	200-250
Age offirst mating (M)	1.5-2	2.5-3	3	2-3/2-3	6-7
Age for experiment(M)	1.5	1.5	3	1/1	6

Heart rate (beat/min.)	330-750	310-500	250-400	315-410/310- 400	150-300
Respiration rate /min.	85-230	70-180	70-110	35-130/30-125	40-60

BloodVolume (% of BodyWt.)	7-10	5-7	6-12	6-9/6-10	5-8
Life span (Years)	1.5-2.5	2-3	3-5	2-3/2-3	5-6
Gestationperi od (Days)	20-21	21-22	65-67	16/21	31-32
Litter size	7-12	8-10	3-4	5-7/4-5	6-8
Age atWeaning (Weeks)	3	3	2-3	3½/3	7

Litter/year	8-10	7	4-5	10/7	4-5
Rest period for females in between 2 mating(days)	15	15	15	18/15	20
No.Of pairs of mammary gland	5	5	1	6-7/4	3-4

Care of Laboratory Animals

1. Environment

Temperature and Humidity:

- Maintain species-specific temperature ranges (e.g., 20–26°C for rodents).
- Humidity levels between 30% to 70% are ideal to prevent dehydration or respiratory issues.

Lighting:

- Follow a 12:12 light/dark cycle unless specific research protocols dictate otherwise.
- Use appropriate intensity to avoid stress or eye damage.

Species-Specific Ambient Temperature

Range_{Species} Temperature

Range (°C) Notes

Mouse 20–26 °C Sensitive to cold; nest building behavior increases in cold environments

Rat 20–26 °C Susceptible to thermal stress at extremes Guinea Pig

18–24 °C Heat stress >28 °C; good ventilation essential

Hamster 20–24 °C Overheat easily above >26 °C Ferret 15–24 °C ^{Prone}
to heat stress >26 °C; Humidity should be 40-55%

Rabbit 16–21 °C ^{Very susceptible to heat stress >28 °C due}
to poor sweating ability

NHP 18–29 °C ^{Depends on species; tropical NHPs need}
stable warmth and higher humidity

Environment..

Ventilation:

Provide 10–15 hourly air changes to ensure fresh air and reduce ammonia buildup.

Avoid drafts and ensure even airflow.

Noise Control:

Minimize exposure to loud or high-frequency noises to reduce stress.

Use soundproofing measures where needed.

2. Caging

Design:

- Ensure cages meet species-specific size and design standards.
- Provide adequate space for movement and natural behaviors.

Materials:

- Use durable, non-toxic materials like stainless steel or high-quality plastics.
- Materials should be easy to clean and resistant to

corrosion.

Safety:

Ensure cages have smooth edges to prevent injury.

Avoid overcrowding to reduce stress and aggression.

3. Bedding

Purpose: Absorbs moisture and odor, provides comfort, and supports natural behaviors like burrowing. **Types of Bedding:**

- Paper-based: Soft, absorbent, dust-free, and eco friendly.
- Wood shavings: Aspen shavings are preferred; avoid aromatic woods like cedar, which can cause respiratory issues.
- Corn cob: Good absorbent, but less

comfortable. **Maintenance:**

- Regularly replace bedding to maintain hygiene.
- Monitor for dust levels to prevent respiratory irritation.

Preferred bedding materials...

1. Mice and Rats: Wood shavings (Aspen), Paper-based 2.

Rabbits: Wood shavings, hay or straw, paper-based
Bedding should allow for burrowing and nesting to reduce stress

3. Ferrets: Paper bedding, fabric bedding.

They are sensitive to dust need low-dust bedding

4. Hamsters: Wood shavings, paper-based

It should be deep enough to allow burrowing 5.

Guinea pigs: Paper-based, Aspen shavings Sensitive respiratory systems; low-dust bedding is critical

6. NHP: Paper-based (for smaller NHP), Wood chips, Mulch, Soil (to encourage natural behaviours)

4. Cage and Equipment Sanitation

Cleaning Frequency:

Cages: clean weekly or as needed to prevent waste buildup.

Water bottles and feeding equipment: clean daily or during cage changes.

Disinfection:

Use appropriate disinfectants (e.g., diluted bleach, quaternary ammonium compounds).

Rinse thoroughly to remove chemical residues. **Sterilization:**

Autoclave cages and equipment used for immunocompromised animals.

Waste Management:

Dispose of soiled bedding and waste in designated biohazard containers.

Follow local waste disposal regulations.

5. Environmental Enrichment

Purpose: Enhance the well-being of animals by providing mental stimulation and physical activity.

Types of Enrichment:

- Physical: Tunnels, wheels, climbing structures, nesting materials.
- Sensory: Varied textures, novel scents, and auditory stimuli.
- Social: Pair or group housing for social species.
- Feeding: Use puzzle feeders or scatter food to

encourage foraging.

Types of Caging for Laboratory Animals

Caging systems for laboratory animals are designed to meet the specific needs of the species while ensuring animal welfare, research integrity, and ease of management. Here are the common types of caging:

1. Standard Cages

Description: Simple cages with solid or grid floors made of durable materials like plastic, stainless steel, or polycarbonate. **Usage:**

Common for rodents like mice, rats, and hamsters.

Typically used for basic housing and research.

Advantages: Cost-effective and easy to clean.

Limitations: Limited enrichment options.

2. Individually Ventilated Cages (IVC)



Specialized cages with built-in ventilation systems to provide filtered air and prevent cross-contamination.

Usage:

Frequently used in pathogen-free or immunocompromised animal housing.

Advantages:

Improves air quality and minimizes exposure to allergens. Maintains a controlled microenvironment.

Limitations: Higher cost and requires additional maintenance.



3. Metabolic Cages

- Designed to separate urine, feces, and food for precise measurement of metabolic output.
- Used in research involving metabolic studies (e.g., drug excretion, diet analysis).
- Advantages: Allows detailed collection of biological data.
- Limitations: Limited enrichment and space for movement.



4. Breeding Cages

- Larger cages with nesting material, designed to house breeding pairs or groups.
- Use: For breeding programs involving rodents, rabbits, and other small animals.
- Accommodates reproductive and maternal behaviors.
- Requires careful monitoring to avoid overcrowding.



5. Shoebox Cages: Rectangular, box-like cages typically made of polycarbonate with a solid bottom.

Use: Standard for housing small rodents in research facilities.

Compact design, easy to stack and transport.

Limited space for enrichment or natural behaviors.



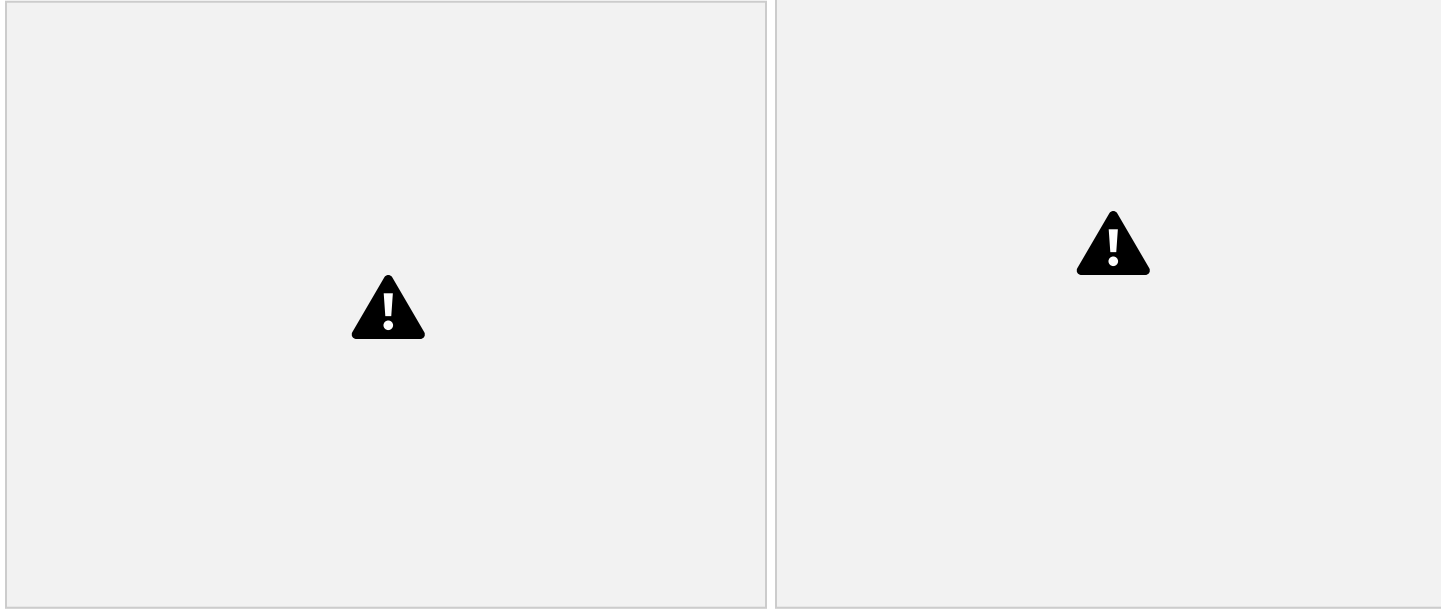
6. Suspended Cages: Cages suspended from a rack, often with wire-grid flooring to collect waste in trays below.

- Common for housing rabbits and larger rodents.
- Efficient for waste removal.
- Wire floors may cause discomfort or injury.



7. Enrichment Cages:

- Helps animals to engage in their environment.
- Cages with added items like tunnels, climbing structures, and nesting materials to promote natural behaviors.
- May require more frequent cleaning.
- Improves animal welfare and reduces stress.



8. Primates Cages:

- Large, robust enclosures with climbing structures and enrichment materials.
- Made of stainless steel or strong metal mesh to withstand and prevent escape.
- For housing non-human primates.

- Meets the physical and psychological needs of primates, as they are highly intelligent animals.
- High cost and space requirements.

9. Isolation cages: Cages designed to prevent contamination or exposure to pathogens, often with HEPA filters.

Use: For immunocompromised animals or quarantine purposes. Reduce the spread of diseases.

Provides a sterile and safe environment.

Expensive and requires special equipment.



Housing

- Laboratory animals require elaborate housing with adequate ventilation and space to move.
- The house should protect from weather, predators, mosquitoes, and insects.
- Laboratory animals can be housed on the floor or in

cages in groups in the house.

Table 1: Housing Space Requirements for Small Rodents (Published by the National Research Council (NRC), USA)

Mouse

Weight (grams)	Min. Floor Area	Min. Cage Height
<10 g	12 in ²	5 inches
10–15 g	15 in ²	5 inches
>25 g	22 in ²	5 inches

Rat

Weight (grams)	Min. Floor Area	Min. Cage Height
<200 g	70 in ²	7 inches
200–300 g	90 in ²	7 inches
300–500 g	120 in ²	7–8 inches
>500 g	193 in ²	8 inches

Hamsters

Weight (approx.)	Min. Floor Area	Min. Cage Height
~100–150 g	23 in ²	6 inches

Guinea Pigs

Weight (approx.) Min. Floor Area Min. Cage Height
~500–1000 g 144 in² 7 inches

Rabbits

Weight (kg) Min. Floor Area Min. Cage Height 2–5 kg
700–775 in² 14–18 inches

Nonhuman Primates

Macaques and Similar-Sized Primates

Weight (kg) Min. Floor Area Min. Cage Height 3–10 kg 576 in²

(4 ft²) 32–36 inches

Large Primates (e.g., Baboons)

Weight (kg) Min. Floor Area Min. Cage Height >15 kg 2,448
in² (17 ft²) 60–84 inches

Recommended space for cats, and dogs

Animals	Weight (kg)	Floor Area/Animal (feet ²)	Height (inches)
Cats	<4	3.0	24
	>4	>4.0	24

Dogs	<15	8.0	Comfortable to stand
	Up to 30	12.0	—
	>30	>24.0	—

Site of housing: -

- ❖ Higher elevation
- ❖ Minimum access for visitors
- ❖ Minimum noise disturbance
- ❖ Separate from the main biological/Immunological production unit
- ❖ Away from a polluted area.
- ❖ The house must be free from rodents and insects, such as mosquitoes, cockroaches, bugs, and flies.

Types of housing: -

1. Non-air-conditioned (Most common type)

2. Air conditioned

Non-air conditioned: - The air flow should not directly enter the entrance of the house. The house should have 2 corridors.

a. Clean corridor: - This is a place in front of the animal room and is used-

□ To bring in sterile food/feed.

□ To take in autoclaved cages.

□ To take bedding materials into the house.

• A foot dip with antiseptic/disinfectant solution be provided at the entry of the clean corridor.

b. Dirty corridor: - This is placed at the back of the animal house and used:

▪ To remove the garbage by having a washing room. ▪ To

check contamination of incoming food/feed by removing garbage by separate outlet.

- To prevent direct cold draft or entry of sunlight in animal rooms.



Feeding of laboratory animals:

Feed/food for the lab. Animals should not be stored for a longer duration because they lose their nutritive value and

leading to reduced acceptability.

Feeding raw materials:

- **Rodents** are naturally grain eaters, hence their feed formulation should consist of many grains, seeds, and nuts.
- Usually, more than one grain is mixed to compensate deficiency in one grain by another.
- Common raw materials used for preparing feed are crushed maize, whole wheat, barley, oat, sunflower seed, powdered millets, gram flour, salt, vitamin and mineral mixture, etc., can be used.
- Guinea pig and rabbits: Greens like succulent grasses, Lucerne or berseem along with vegetables are fed

Feed form:

- Pellets are preferred instead of mash because mash gets soiled faster with difficulty of storing as well as it is

messy, unhygienic , and laborious to prepare.

- Pellets are commercially available for feeding lab. Animals.

Feeding of mice and rat:

- These animals take small bites of food or pellets.
- They pull the pellets through wire mesh, hold them in their front legs, and then eat.
- Their food should contain 16 to 20 % protein, 4 to 5% fat, 4 to 6% fiber, and 45 to 50% carbohydrate.
- Daily requirement: Rat 15gm and Mice 6gm.

Wheat flour	75.0
Gramflour	10.0
Skimmed milk powder	10.0
Refined oil	10.0
Salt	1.0

Vitamins and Minerals	3.5
Total	100.0

Feeding of Hamsters:

- Hamsters have cheek pouch. They eat faster and store in the pouch.
- Then they go to corner and chew the stored food slowly.
- Their feed consists of 16 to 20% protein, 4 to 5% fat and 45 to 50% carbohydrate.
- Daily feed requirement- 15gm.

Feeding of Guinea Pig:

- Green grasses, leaves and vegetables chewing is main features of guinea pig feeding.

Wheat bran	20.0
Whole grams	10.0
Carrots or Cabbage	10.0
Lucerne	60.0
Total	100.0

Nutrient composition:

- 16 to 25% protein, 4 to 5% fat, 45 to 50% carbohydrate and 10% fibre.
- Specially required nutrient is Vitamin C. Hence it should be supplement either through feed or water @ 250mg/Liter. ●
- Daily feed requirement- 60 to 70gm.

Rabbits:

- Need 12-14% protein, 18-20% fiber, 40-50% carbs, 2-3% fat, 0.5-1% calcium.
- Pelleted Feed: Commercial rabbit pellets with 12–14% protein and 18% fiber.
- Hay: Unlimited grass hay (e.g., Timothy hay).
- Fiber is the most critical component of a rabbit's diet, essential for:
Gut motility, Prevention of hairball buildup, Healthy cecal fermentation
- Fresh Vegetables: Leafy greens (e.g., kale, parsley) and occasional treats like carrots.

Non-Human Primates

- Nutritional requirement (%DM): Protein (15-20%), Fiber (5-15%), Fat (5-10%), Carbs (45-60%), water (Ad libitum)
- Feeding schedule: Twice daily
- Commercial Primate Diet: Specially formulated biscuits with balanced nutrients.
- Fruits and Vegetables: Fresh produce as a supplement.
- Enrichment: Nuts, seeds, and leafy greens for foraging behavior.
- Provide Vitamin D for indoor-housed primates to prevent deficiency.
- Avoid foods high in sugar and fat to prevent obesity.

Identification of laboratory animals:

1. **Staining:** Painting the animal with dyes like crystal violet and carbol fuchsin can be conveniently used for marking. Staining

on the skin coat in a small area will be easily visible.
Temporary method.

2. **Tagging/Branding:** - Rabbits and Guinea pigs can be ear tagged. Aluminium tags with embedded numbers are normally used.

3. **Tattooing:** - usually carried on the ears, feet, shoulders, or hip for identification. It can be done in rabbits, rats, and guinea pigs.

4. **Notching:** - Cutting ears in a specific shape at a particular place
in

the form of a notch is also used as identification marks in rats and mice.

General Principles of Animal Handling

Minimize stress: Handle animals gently and confidently to avoid unnecessary stress.

Safety First: Wear personal protective equipment (PPE) as required (e.g., gloves, lab coats).

Species-Specific Knowledge: Understand the behavior and needs of the species being handled.

Regular Training: Personnel should be trained in proper handling techniques to ensure proficiency and compliance with ethical guidelines.

Use of Equipment: When necessary, use appropriate tools (e.g., restraint devices, transport containers) to handle animals safely and humanely.

Health Monitoring: Observe animals for signs of illness, injury, or stress during handling.

Laboratory animals are mostly smaller in size and more delicate, so handling is very important. Gentle handling is the best way to obtain real and correct experimental results.

Purpose of handling:

1. Housing

2. Weighing
3. Inoculation & sample collection
4. Inspection to note body changes
5. Marketing and transport.

Handling technique by species

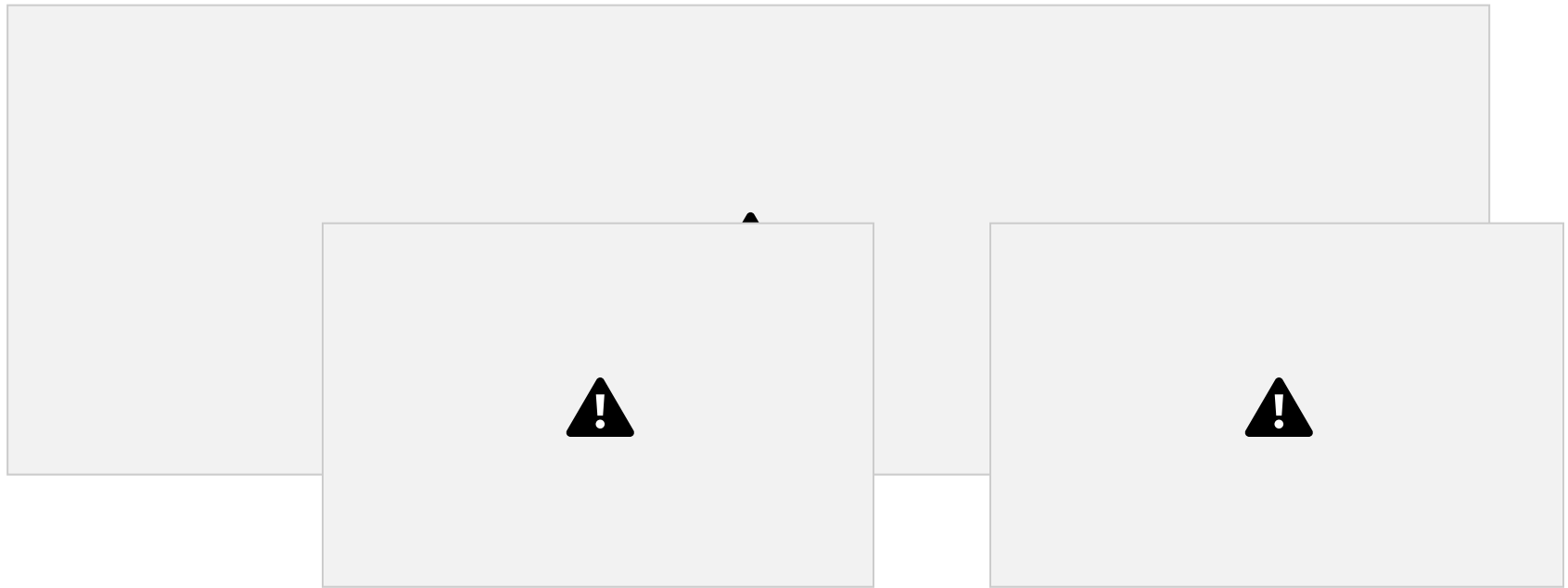
1. Mice

- Grasp the base of the tail gently but firmly. • For closer examination, restraint by scruffing (hold the loose skin at the back of the neck).
- Avoid lifting by the tip of the tail to prevent injury. • Minimize time in restraint to reduce stress.

Rats

- Support the body by placing one hand under the chest and the other under the hindquarters. • For restraint,

hold around the shoulders with one hand while supporting the body with the other. • Be cautious of biting, especially with stressed or unfamiliar rats.



3. Guinea Pigs

- Scoop up with both hands, supporting the chest and

hindquarters.

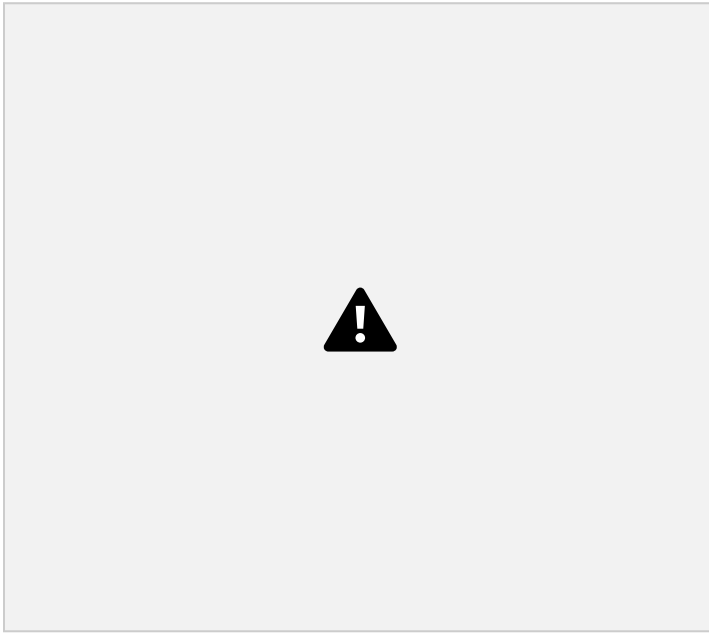
- Use a calm approach to avoid startling them.
- Avoid squeezing the thorax, as it may interfere with breathing.

4. Rabbits

- Support the hindquarters firmly to prevent spinal injuries.
- Hold close to the body to make the rabbit feel secure.
- Never lift a rabbit by its ears.
- Ensure a quiet environment to reduce fear responses.

6. Non-Human Primates

- Use protective equipment and, if necessary, chemical restraint for safety.
- Handle in pairs or groups of trained personnel.
- Monitor for zoonotic disease risks.

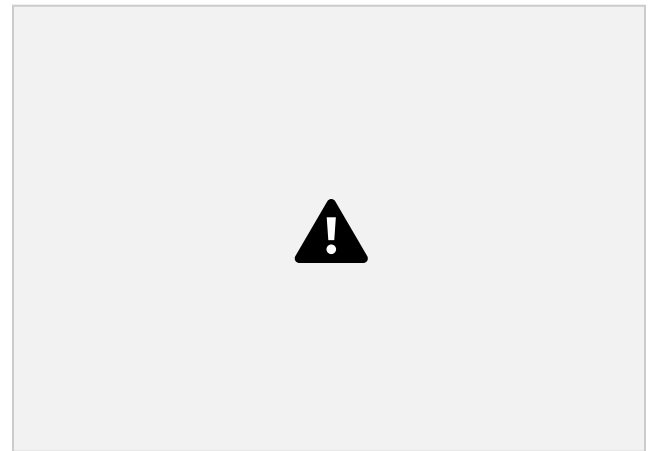


Mouse handling

1. Tail Handling (Traditional Method)

❖❖ Procedure:

- Grasp the base of the tail (not the tip) using your thumb and index finger.



- Lift the mouse slightly and place it on a stable surface.
- Allow the mouse to grip the surface with its front paws.
- If needed, provide additional support by placing a hand under the body.

2. Tunnel Handling (Refined Method)

❖❖ Procedure:

- Place a plastic tunnel inside the cage.
- Allow the mouse to enter the tunnel voluntarily.
- Cover one end with your hand and lift the tunnel.



Sexing of Mice

Observe the distance between the anus and genital opening.

Females: Shorter anogenital distance; small-visible nipples in the ventral region.

Males: Longer anogenital distance; Testes are visible and

located under the skin near the base of the tail.



Mouse restraint technique

1. Scruffing

- Grasp the base of the tail and place the mouse on a stable surface.
- Use your thumb and index finger to pinch the loose skin at the back of the neck.



- Gently lift the mouse while supporting its back with your palm. • Hold securely but avoid excessive pressure to prevent breathing issues.

2. Mechanical restraint (Plastic restrainer) • Select the appropriate size restrainer. Ensure that the restrainer is clean and has ventilation holes. • Gently place the mouse inside the tube with its head facing outward for the procedure.

- Close the end of the tube lightly, leaving the mouse's head exposed. Keep the body secure to avoid unnecessary movement.
- Proceed with the required medical or experimental procedure. Ensure the mouse is calm and well supported.

- Gently remove the mouse from the restrainer and return it to its cage. Observe the mouse for signs of distress or injury.





Ear punching for identification

Restrain the Mouse: Gently handle the mouse by holding its body securely while supporting the head to avoid sudden movements.

Positioning: Hold the mouse firmly but gently.

Clean the Ear: Use an antiseptic solution to clean the area of the ear where the punch will be made.

Punch the Ear: Place the ear punch tool at the selected site. Make sure to punch a portion of the ear that is far enough from the ear canal to avoid any complications. Gently apply pressure to remove a small piece of the ear. The punch should be quick and precise to minimize the time the mouse is under stress.



Rat handling

- First, assess the rats in their cage for normal behavior. • Gently pat and touch the animals. Try to calm them and let them sniff.
- Grasp the rat around the thorax with your thumb and forefinger under each of its front legs.
- Lift the rat out of the cage and place it in a new cage or on a firm surface.
- For aggressive rats, pick them up by grasping them by the base of the tail, close to the body.
- Do not suspend the rat by its tail or its upper body for a prolonged time.

- Do not let the rats hold on to the cage top.

Sexing of Rats

- To sex a rat, lift the base of the tail and hindquarters, and examine.
- The sex is determined by comparing the anogenital distance in adult males (the distance is longer than in females).
- The testes may also be visible in the scrotum, but can often be retracted.

Rat restraint technique for technical manipulation

1. Manual restraint

- Hold the rat by the base of the tail and grasp the rat by placing one hand firmly over the animal's

back and rib cage. Gently press the forelegs toward the head and restrain the head with your thumb and index finger.

- Do not attempt to scruff rats unless you are very experienced; unlike mice, they object strongly to being scruffed.
- To restrain an aggressive rat, wear a cloth glove when restraining.



2. Mechanical restraint of rats



- A Plexiglas restraint box is a transparent, ventilated enclosure designed to hold a rat in place while allowing access to specific body parts for procedures.
- Gently place the rat inside through the entry slot. • Adjust the rat's position so the required body part (tail, head, or abdomen) aligns with the access holes.
- Secure the sliding door or lid to prevent escape. • Monitor the rat continuously to avoid distress.

Ear punching for identification

- Restrain the rat.

- Immerse the ear punch in alcohol to disinfect it
- Place the ear punch on the outer edge of the ear where you want to mark the animal for identification
- Firmly and quickly punch the ear to avoid an incomplete cut.

Guinea pig handling and sexing

- Assess the guinea pigs in their cage for normal behavior.
- Grasp the guinea pig around the thorax, placing its front leg between the index and middle fingers for added support.
- Check the sex of the guinea pigs by applying gentle pressure above the genitalia.
- The penis of the male will protrude, making sexing easier.



Guinea pig restraint for technical manipulation

- Guinea pigs are quiet docile animals.
- Adequate restraint is usually achieved by placing the animal on a tabletop and supporting it with one hand at the head and the other hand around the

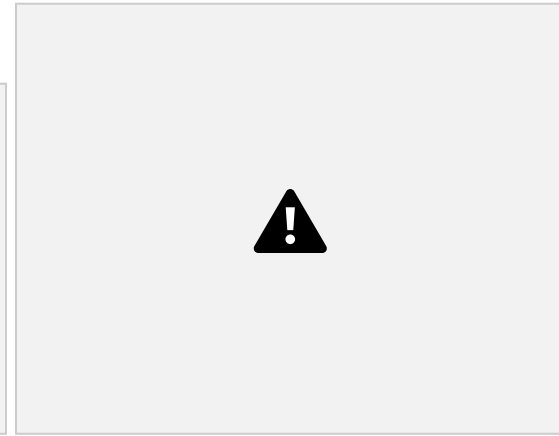
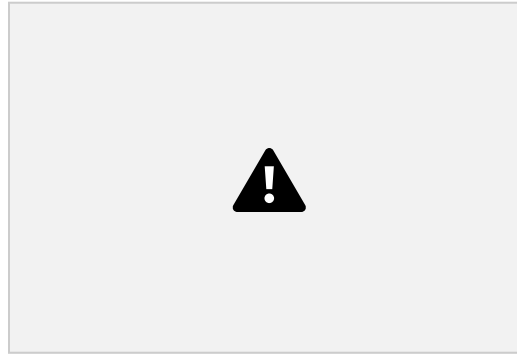
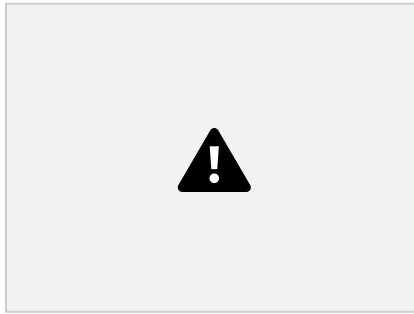
rump.

- It may be necessary for an assistant to help hold the guinea pig in place whilst the other person performs the procedures.
- Alternatively, the guinea pig can be anesthetized.

Rabbit handling and sexing

- Always check the condition of the rabbit before removing it from the cage.
- Grasp the rabbit firmly by the nape of its neck. Place one hand on the rump of the rabbit and lift it from the cage.
- Support its hind legs with the opposite hand. Tuck its head between its arm and body.
- Check the sex of the rabbit by applying gentle pressure above the genitalia. The penis of the

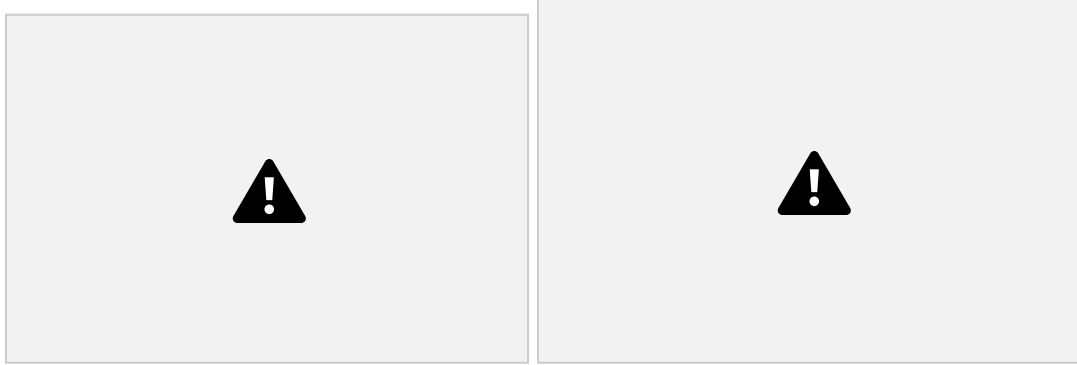
male will protrude, making sexing easier.



Rabbit restraint technique

- For manual restraint, an assistant can hold the nape of the rabbit's neck and place a gentle but firm hand on the back of the rabbit. Whilst in this normal seated position, the rabbit may be shaved for injections or given IM injections with the relative ease.
- If working alone, you should use a plastic restrainer, cat bag, or towel wrap, as it is safer.

- The towel wrap is by far the easiest technique and does not require any special equipment.



Rabbit restraining...

- Open the towel onto a nonslip surface. Place the rabbit in the center of towel and fold one side of the towel over, ensuring that the rabbit's head and ears are not covered, but that the feet and body are covered.
- Tighten your grip if rabbit struggles.



Nonhuman primates (NHPs)

- Examples:** Rhesus macaques, cynomolgus monkeys, baboons, squirrel monkeys, chimpanzees.
- Uses in research:** Neuroscience, behavior studies, infectious diseases, vaccine development.
- Legal and ethical concerns:** Highly regulated due to advanced cognitive and social abilities.

