



Husbandry and Enrichment

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What is husbandry?

The routine **care and management of laboratory animals** to ensure their health, comfort, and welfare.

It includes:

- **Housing and caging** including containers and utensils for food and water
- **Bedding and nesting material** to support comfort and natural behaviors
- **Environmental conditions** (temperature, humidity, light, noise) and **cage hygiene**
- **Identification methods**
- **Handling and sex determination**
- **Transportation** (within the facility or between locations, ensuring minimal stress)

What is enrichment?

The **enhancement of the animal's environment** through social, physical, sensory, cognitive, or nutritional additions that encourage **natural species-specific behaviors**, reduce stress, and improve **psychological well-being**.

Why husbandry and enrichment matters?

Animal Welfare and 3Rs

Good husbandry and enrichment are refinements that improve animal well-being. Poor conditions cause stress and abnormal behavior (stereotypies, aggression)

Scientific validity

Poor husbandry leads to stress, which alters animal physiology, behavior, and immune responses- introducing unwanted variability into data and compromising reliability and reproducibility.

Regulatory Compliance

EU Directive 2010/63/EU mandates high standards of care and housing – poor husbandry can lead to legal non-compliance and jeopardize project approvals.

Staff Safety & Efficiency

Well-managed environments reduce animal aggression and improve working conditions

Husbandry

Types of Rodent Cages

Conventional cages

- Open-top cages
- Filter top cages

Individual ventilated cages (IVC)

- Forced air
- Motor free
- Disposable single-use

Special Purpose

- Metabolic cages
- Transport cages

Open-top cages



Advantages

- ✓ **Lower acquisition cost** compared to ventilated systems
- ✓ **Simple design** with no integrated containment features, making them easy to set up.
- ✓ **No need for specialized equipment** - Handling can be done without additional containment measures.
- ✓ **Quick setup** – Suitable for short-term or basic housing needs.

Disadvantages

- ✗ **Poor environmental control**- No built-in ventilation or barrier system; relies on room air.
- ✗ **Higher contamination risk** – Pathogens and allergens can spread between cages via airborne particles.
- ✗ **Greater exposure to allergens for staff**- Requires strict use of personal protective equipment (PPE).
- ✗ **Dependence on PPE**
- ✗ **Not suitable for SPF or immunocompromised animals** - Inadequate protection from environmental microbes.
- ✗ **Require 15–20 air exchanges per hour**

Filter-top cages



Advantages

- ✓ **Lower acquisition cost** compared to ventilated systems (but higher than open-top)
- ✓ **Improved containment** of infectious agents and reduction of **cross-contamination** risk between cages due to the barrier provided by the filter lid.
- ✓ **Simple to use and maintain**, requiring no complex ventilation setup.
- ✓ **Partial reduction of allergen dispersion** into the room environment, offering some protection for animal care staff.

Disadvantages

- ✗ **Passive ventilation only** – no active air exchange within the cage, which can lead to **stale air and ammonia build-up** if not changed frequently.
- ✗ **Increased humidity** inside the cage relative to the macroenvironment, particularly with high bedding moisture or large animal groups.
- ✗ **Lower housing density** – larger cage volume and manual handling requirements reduce the number of cages per rack or room.
- ✗ Requires **15–20 air exchanges per hour**

Individual Ventilated Cages (IVCs)

A. Forced air



B. Motor free



Individual Ventilated Cages (IVCs)

Advantages

- ✓ **Precise control of cage microclimate** (temperature, humidity, airflow).
- ✓ **Stable, protective environment** ideal for breeding, long-term housing, and SPF colonies.
- ✓ **Allows co-housing of animals** with different microbiological or genetic backgrounds.
- ✓ **Minimizes pathogen spread** — each cage acts as an individual bio-barrier.
- ✓ **HEPA-filtered airflow** improves air quality and reduces ammonia and odors.
- ✓ **Suitable for quarantine or high-risk studies**, especially under negative pressure.
- ✓ Requires **10–12 air exchanges per hour**
- ✓ Maximizes housing density complying with EU guidelines
- ✓ **Reduces allergen exposure** for personnel.

Disadvantages

General

- ✗ High acquisition and maintenance cost
- ✗ Cage handling under a biosafety cabinet (class II) to maintain barrier integrity.
- ✗ Time-consuming cage changes

Forced-Air Systems

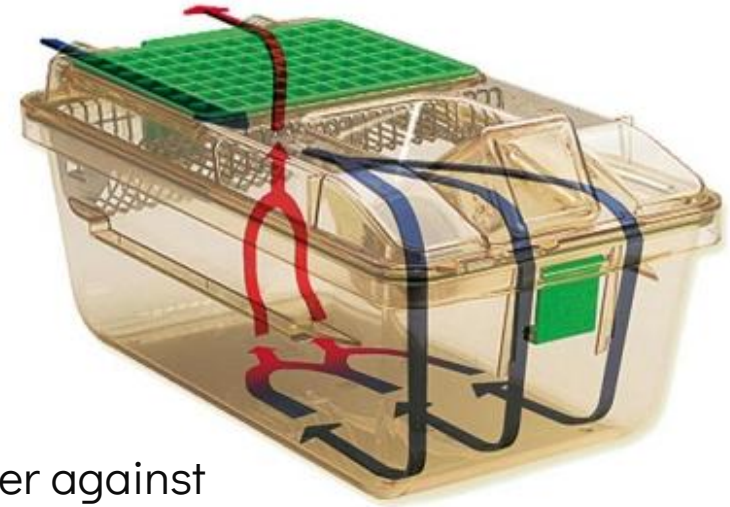
- ✗ Air velocity may cause mild stress in some animals especially if not properly adjusted.
- ✗ Fan or motor noise can contribute to chronic low-level stress or impact hearing-sensitive strains

Motor-Free Systems

- ✗ Fewer customization options for rack configuration or ventilation balancing.
- ✗ Challenging design for cleaning and autoclaving,



Airflow Dynamics in IVCs

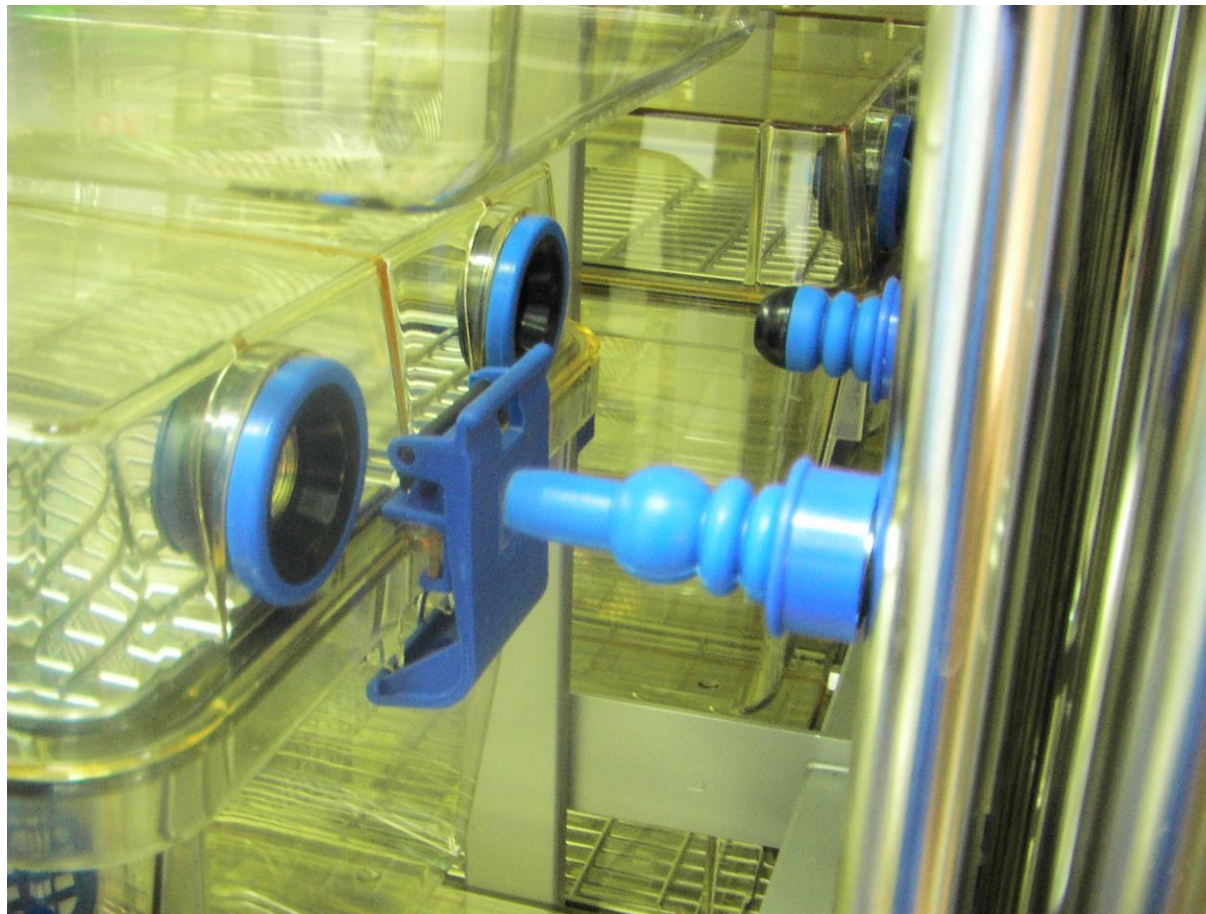


● Inflow (Clean Air):

- Show HEPA-filtered air entering the cage from the top.
- This air is typically supplied by a ventilation unit in the IVC rack.
- The air enters the cage under positive pressure, creating a protective barrier against contaminants from the room or adjacent cages.

● Outflow (Exhaust Air):

- Show used air exiting the cage.
- This air exits through a separate exhaust port, helping prevent the buildup of CO₂, ammonia, and humidity inside the cage.
- The exhausted air is either filtered again before being released into the room or sent to an external exhaust system, depending on the setup



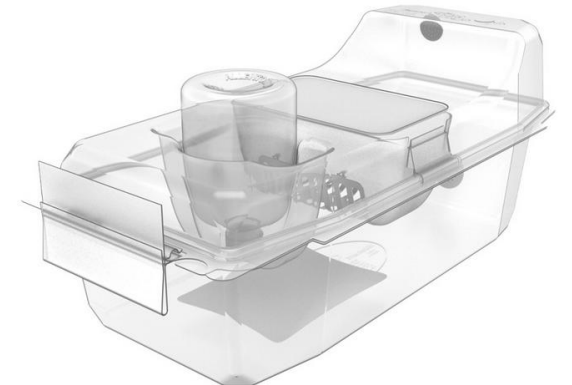
Disposable single use cages

Advantages

- ✓ Ideal for quarantine, transport, or biohazardous studies.
- ✓ Eliminates the need for washing, autoclaving, or decontamination.
- ✓ Reduces the risk of cross-contamination and pathogen spread.
- ✓ Time- and labor-saving in high-turnover or temporary housing situations.

Disadvantages

- ✗ Higher cost per use compared to reusable cages.
- ✗ Not environmentally sustainable due to plastic waste.
- ✗ Limited options for enrichment and long-term housing.
- ✗ May lack robust ventilation, depending on design



Metabolic cages

Specialized cages enable precise collection of physiological and biochemical data from laboratory animals, especially rodents, by separately collecting urine, feces, and sometimes exhaled gases.

Common Applications:

- Drug metabolism and pharmacokinetics (ADME)
- Nutritional studies (e.g., calorie intake)
- Renal function assessments
- Water and food consumption monitoring
- Toxicity studies (e.g., urinary excretion of toxins)
- Circadian rhythm and behavior studies (with specialized setups)



Metabolic cages

Advantages

- ✓ Accurate separation and collection of **urine and feces**
- ✓ Precise **monitoring** of **food and water** intake
- ✓ Ideal for **pharmacokinetic, toxicology, and metabolic studies**
- ✓ Supports **reproducible** and **quantitative data**
- ✓ Some systems allow **automation** and **continuous data recording**

Disadvantages

- ✗ **Restrictive housing**- may cause stress or altered behavior
- ✗ **Not suitable** for **group housing** or **long-term use**
- ✗ May require animal **acclimatization**
- ✗ **Expensive** equipment and maintenance
- ✗ Can affect **animal welfare** and **natural circadian patterns**

Transport cages

Designed for **safe and temporary movement** of laboratory animals

- Ensure adequate ventilation, protection from injury, and secure containment
- Must comply with animal welfare regulations (e.g., space, bedding, temperature)
- Often used for transfers between facilities, within buildings, or during experiments
- Available in various materials and sizes
- Should be sanitized between uses to prevent cross-contamination
- Minimize transport duration to reduce stress and maintain animal health



Golden Rules

Before deciding which caging system is best for your facility, consider:



Your research needs – What types of studies will be conducted?



Space and facility layout – Does the system fit your current infrastructure?



Flexibility – Can the system adapt to future changes or different protocols?



Cost and maintenance – What is the total cost of purchase, operation, and upkeep?

In any case, never forget regular monitoring..



Environmental conditions

Parameter	Recommended Range	Notes
Temperature	20-24°C	Maintain within ± 2 °C;
Relative humidity	40–60%	Avoid prolonged periods below 30% or above 70%
Ventilation	10–20 air changes/hour (room)	IVCs typically provide 50–75 air changes/hour at cage level.
Lighting	12 hours light:12 hours dark	Some facilities use red lighting during the dark phase if staff need to check animals
Noise	< 60 dB (preferably < 40 dB)	Sudden loud noises or high-frequency sounds (ultrasound) must be avoided

Cage hygiene

- Maintaining cage cleanliness is essential for animal health, comfort, and the prevention of disease.
- Soiled bedding leads to the accumulation of ammonia, moisture, and pathogens, increasing the risk of respiratory and skin issues.
- Cage changing frequency depends on species, housing density, bedding type, and ventilation (e.g. once per two weeks for IVCs, once per week for open-top cages).
- Regular monitoring of cage hygiene helps optimize change schedules.
- Proper hygiene supports both animal welfare and data reliability.

Cleanliness is often seen through a human-centered lens, focusing on odor and appearance rather than animal comfort—yet excessive cleaning can disrupt stability and elevate stress levels.



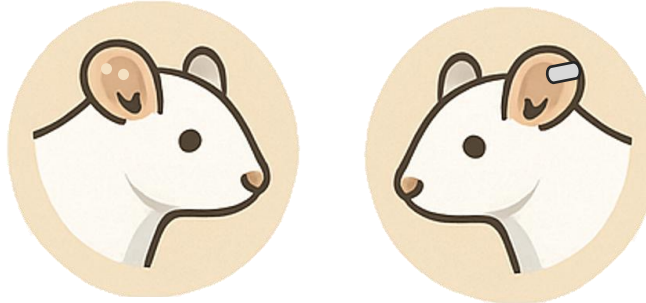
Identification methods

Temporary Methods



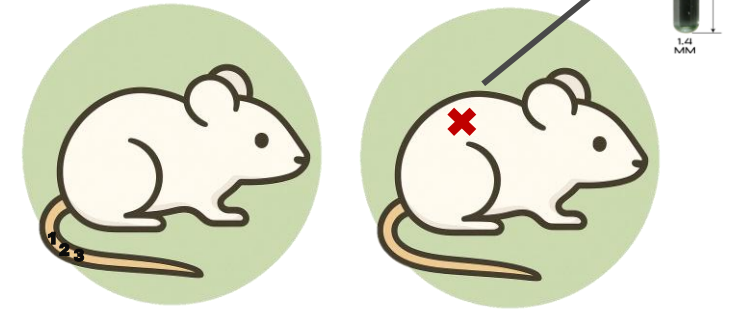
- **Coat marking** (e.g., non-toxic markers)
 - Easy to apply, for short-term studies
 - May fade or be groomed off.
- **Tail marking** (permanent marker)
 - Quick and minimally invasive.
 - Limited durability.

Semi-permanent Methods



- **Ear punching**
 - Involves removing small sections of the ear in a coded pattern.
 - Requires skill; may affect ear integrity or healing.
- **Ear tags**
 - Visible, relatively easy to apply.
 - Risk of loss or infection.

Permanent Methods



- **Microchip transponders (RFID)**
 - Injected subcutaneously
 - Offers unique, permanent ID.
 - Ideal for long-term studies.
 - Requires a reader.
- **Tattooing**
 - Ink applied to the tail, footpad, or ears using manual or automated tools.

Bedding and nesting material



Bedding

- Dust-free, absorbent bedding (e.g. corn cob, wood chip) to allow burrowing and odor absorption. Keep sleeping/resting areas clean and dry

Nesting & shelter

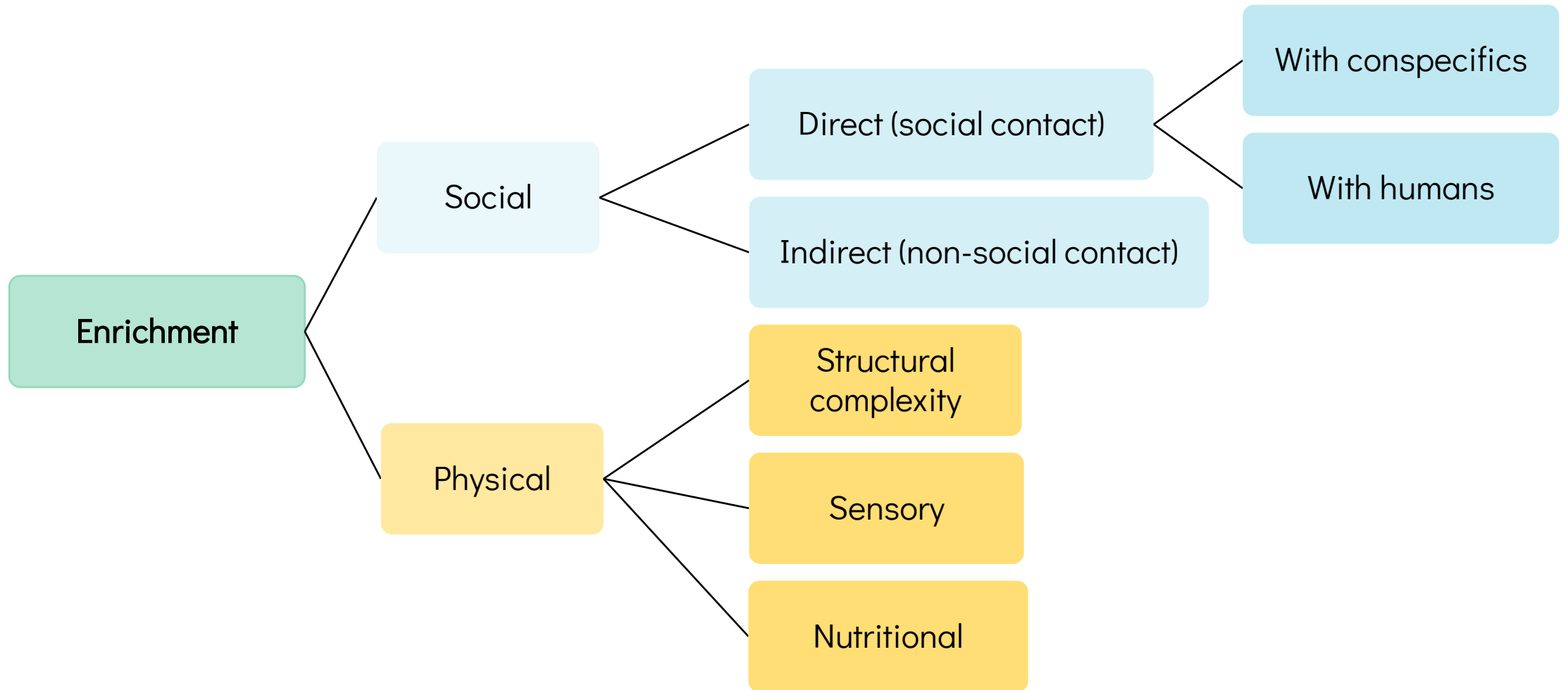
- Always provide nesting material (e.g. paper strips, cotton nestlets) especially for mice and breeding female
- Include shelters or hiding places (plastic huts, tubes) for security and thermoregulation.

Enrichment

What do we mean with the term “enrichment”

- Enrichment refers to any objects, practice or modification of the captive environment designed to enhance physical, cognitive, and social stimulation in animals.
- Effective enrichment promotes species-typical behaviors, introduces cognitive challenges, and offers opportunities for choice and environmental control, thereby improving their overall quality of life.

Types of enrichment



Social enrichment

A. With Conspecifics

- Gregarious species should be housed in pairs or groups wherever possible
- Social partners enhance alertness, promote exploratory behavior, and offer a sense of diversion and occupation
- Group housing has been shown to reduce the frequency and duration of procedure-induced stress responses

B. With Humans

- Positive human-animal interactions, including handling, training, and socialization
- Stimulating cognitive function and fostering positive relationships with caretakers, technicians, and researchers.

C. Non-contact enrichment

- Visual, auditory, and/or olfactory communication with conspecifics through barriers

Physical enrichment

A. Structural Complexity

- Shelters, nest boxes, tubes, and platforms support resting, hiding, and exploration
- All materials must be durable, safe, and easy to clean to ensure hygiene and animal safety
- To maintain novelty and engagement, items should be rotated periodically



Physical enrichment

B. Sensory enrichment

- Sensory enrichment includes visual, auditory, olfactory, tactile and taste stimuli.
- Auditory stimuli, such as soft background music or natural sounds, can be beneficial.
- While cage cleaning is essential, it is important to avoid removing olfactory cues entirely, as this can disrupt the social hierarchy within the cage, potentially leading to aggression.
- Providing different food items (e.g., seeds) provides taste stimulation and encourages foraging behaviors.



Physical enrichment

C. Nutritional enrichment

- Rodents tend to be highly motivated by food-based enrichment
- In nature, rodents spend a huge portion of their time foraging for food
- Enrichment items should be species-appropriate, non-toxic, and **not interfere with dietary control in experimental protocols.**
- Portion sizes and frequency must be managed to avoid overfeeding or nutritional imbalance.

Scatter some food in the bedding so the animals have to sniff and dig around to find it

Hide treats inside a cardboard tube or a puzzle feeder



Chocolate O's



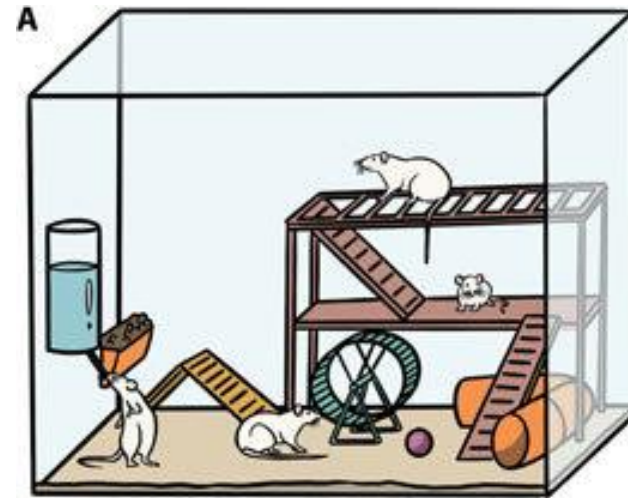
Mini yogurt drops



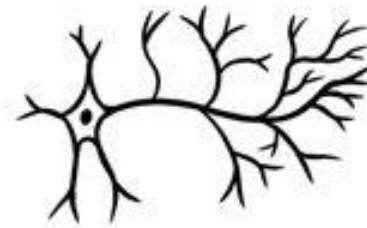
Banana Chips



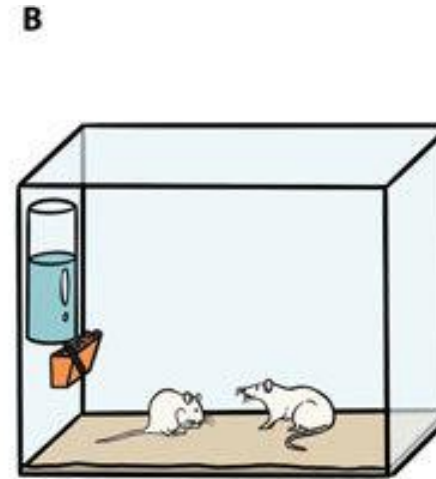
Dried Cranberries



Enriched environment



Appearance of nerve cells in the mouse brain



Standard environment



Appearance of nerve cells in the mouse brain

Vive, S., Af Geijerstam, J.-L., Kuhn, H. G., & Bunketorp-Käll, L. (2020). Enriched, task-specific therapy in the chronic phase after stroke: An exploratory study. *Journal of Neurologic Physical Therapy*, 44(2), 145–155. <https://doi.org/10.1097/npt.0000000000000309>

Choosing Environmental Enrichment

Animal related factors

- Species and strain
- Age and sex
- Health status
- Housing system
- Social dynamics

Study related factors

- Scientific goals
- Standardization vs variability
- Duration of the study
- Regulatory requirements

Practical considerations

- Ease of cleaning or replacing
- Compatibility with cage design & ventilation
- Safety and non-toxicity
- Cost and availability

So! Good enrichment should balance welfare, scientific integrity, and practical implementation

Preference tests



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Kostomitsopoulos, N. G., Paronis, E., Alexakos, P., Balafas, E., van Loo, P., & Baumans, V. (2007). The influence of the location of a nest box in an individually ventilated cage on the preference of mice to use it. *Journal of Applied Animal Welfare Science*, 10(2), 111–121. <https://doi.org/10.1080/10888700701313256>

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Thank you for your attention!



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