
UNIT 20 WELFARE ISSUES IN LABORATORY ANIMALS

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20.1 LEARNING OUTCOMES

- a) Knowledge and Understanding:** After studying this Unit, you will be able to:
- Understand the meaning and purpose of laboratory animals.
 - Classify the laboratory animals.
 - Explain the 3 R's principles, and role of lab animal veterinarians.

b) Practical and Professional Skills: After studying this Unit, you will be able to:

- Describe the important laboratory animals and their use in laboratory research.
- Discuss the specific animal welfare issues of laboratory animals.

20.2 INTRODUCTION

Laboratory animals in short lab animals are another important group of animals that are managed by humans, mostly in captivity. Animals have been used in medical research from as far back as 129-199 A.D. when Galen, a Greek medical scientist, used a pig for his experiments. In the sixteenth and seventeenth centuries, anatomical dissections were carried out on animals; Galvani used frogs in 1791 for his experiments and the Russian physiologist, Pavlov, carried out his famous dog experiments in the early 1900s. Since then, large numbers of animals have been used in biomedical and other research with serious implications to their welfare. The guidelines, rules, regulations, Acts and international agreements have also been developed simultaneously to foresee humane methods of animal experimentation and testing.

This unit introduces you to the meaning, history, uses and types of lab animals and common animal welfare issues in the lab animals.

20.2.1 Meaning of Laboratory Animals

Laboratory animals are those which can be bred and reared / maintained in the laboratory under suitable conditions and intended for use in research, testing, or teaching. The common laboratory animals are: rat, mice, guinea pig, rabbit and hamster. The other animals used for experimental purpose are cat, dog, frog, monkey, pigeon etc.

Following are the definitions of laboratory animals:

- Laboratory animals are any vertebrate animals produced for or used in research, testing or teaching - **NRC, 2011.**
- Laboratory animal is an animal intended for use in research - **OIE, 2012.**
- Laboratory animal is any live vertebrate animal (and any other animal designated by applicable legislation) used or intended for use in research, testing, or teaching. For accreditation purposes, the definition is not limited by the type of housing enclosure (e.g., cage, pen, paddock, pasture, tank, raceway, etc.) - **AAALAC International** (*Association for Assessment and Accreditation of Laboratory Animal Care International*)

In most cases the lab animals are bred purposively to have a defined physiological, metabolic, genetic or pathogen free status. More than 115 million animals are used for animal testing around the world on an annual basis. These animals experience stress, pain and suffering before, during and after experimentation. In most cases, animals are euthanized during or after the experiment. At present, in India, there are 1747 animal facilities registered with Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA) for conducting experiments using animals.

20.2.2 History

Since the second century, animals have been used in bio-medical research to understand various diseases and discovery of medicines. Historically, research on animals played a key role in understanding the anatomy and physiology of the human body as well.

- *Example 1:* Discovery of the blood circulation (William Harvey, 1628)
- *Example 2:* Function of the lungs (Leonardo da Vinci, late 15th century)

More recently, the development of important therapies and preventative treatments, including antibiotics, hormones, vaccines, organ transplantation and modern medicines, have involved animal research and testing. Animals are used widely for safety assessment and efficacy testing of nearly all new chemicals, pesticides, consumer products, drugs, medical devices, vaccines and many other products. This is not only the requirement of regulatory agencies of respective countries, but also to protect and advance the health of people, animals and environment.

Laboratory based research is essential to uncover biological processes and pre-clinical testing. The greatest drug discoveries in 19th and 20th centuries were possible due to the use of animals. Over the last century, almost every Nobel Prize winning medical research was dependent upon the use of animals in research. The major areas where the lab animals are used is summarised in Box 20.1.

Box 20.1: Major areas where the laboratory animals are used

- *Drug development:* For development of new drugs for both human and animals.
- *Toxicity (safety) testing:* To test the safety of new product i.e., drugs, food additives.
- *Basic research:* It is a curiosity driven research i.e., new models of human diseases.
- *Education:* To train biology students (including veterinary students) new researchers and surgeons.

In late 19th century, India experienced institutionalization of bacteriology laboratories:

- Imperial Bacteriology Laboratory at Pune (1890)
- Bacteriology Laboratory at Agra (1892)
- Plague Research Laboratory in Mumbai (1896)
- Pasteur Institute of India at Kasuli (1900), Coonoor (1907), Rangoon (1916), Shillong (1917) and Kolkatta (1924)
- Central Research Institute (CRI) at Kasuli (1905).

With the establishment of these institutes, the laboratory research and ethical treatment of lab animals assumed critical dimension as these institutes used massive animal resources (Chakrabarti, 2010).

- *Example:* The production of a single vaccine in one Pasteur institute required 6000 rabbits annually.

As we go through the history, the animal experiments in India were initiated in 1860s, as the then British Govt. attempted to introduce various chemical drugs in India which necessitated experiments on animals become necessary.

- *Example:* Use of chloroform as an anesthetic in experimental animals.

Box 20.2: IAEC
 Institutional Animal Ethics Committee (IAEC) is a local body, approved by CPCSEA, which has been empowered to permit experiments on small animals through an amendment in Rules for Breeding and Experiments on Animals (Control and Supervision) in the year 2006.

Debate about experimental lab animals is not new. However, the usefulness and relevance of the different kinds of research on animals need to be looked, for degree of pain and suffering which they may experience in research. To ensure good scientific investigation, the lab animal should have a normal physiology and behavior, apart from specific adverse effects under investigation. On scientific as well as ethical grounds, therefore, the psychological wellbeing of laboratory animals should be an important concern for lab animal veterinarians, animal technicians and scientists (Poole 1996).

Education: All over the world animals are being used for dissection in the field of education. You must have noticed the use of frogs for dissection in the biology classes. Similarly, the dogs and calves are commonly used in training students pursuing veterinary and animal husbandry courses. With the increase in the voice for the concern for animal welfare, rules and regulations have come into force to minimize the use of animals required for dissection and encourage the use of videos, simulations etc. in place of live dissections. The University Grants Commission issued guidelines in 2011 for discontinuation of dissection and animal experimentation in Zoology / Life Sciences in a phased manner (https://www.ugc.ac.in/pdfnews/6686154_guideline.pdf)

On par with international developments in welfare of lab animals, in India also several measures, rules and regulations came into existence to safeguard the welfare of lab animals.

- *Example:* CPCSEA - Committee for the Purpose of Control and Supervision of Experiments on Animals and *IAEC* - Institutional Animals Ethics Committee (Box 20.2)

(Please refer MAW-003 for more details on CPCSEA & IAEC)

Check Your Progress 1

Note: a) Use the spaces given below for your answers.

b) Check your answer with those given at the end of the unit.

- 1) Define laboratory animal.

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2) Name the major research areas where the laboratory animals are used.

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3) What is the role of Institutional Animal Ethics Committee (IAEC)?

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20.3 THE 3 R's AND LAB ANIMAL VETERINARIANS

20.3.1 Three R's Concept

The Three R's concept (reduction, refinement and replacement) was developed by Russell and Burch (1959) to provide a framework for improving the conduct and ethical acceptability of experimental techniques on animals. Since then it has become a common focus for both the animal welfare and the scientific communities. This approach to the humane use of animals is commonly referred as 3R's of alternatives, and many countries now have laws requiring that the 3 R's should be applied to minimise harm to animals – for example, that anaesthetic should be used to prevent pain – and demanding careful justification for any unavoidable harm (Box 20.3).

Box 20.3: Three R's in Laboratory Animal Research

- **Reduction:** Any strategy that will reduce number of animals being used in laboratory research.
- **Refinement:** Modify experimental procedure to minimise pain and increase quality.
- **Replacement:** Methods which avoid or replace the use of animals in laboratory research with other options like 3D models.

(Also refer MAW – 004 on 'alternatives to animal usage' for discussion on 3R's)

The concept of 4th R, Rehabilitation of laboratory animals refers to aftercare rendered to animals that have been:

- Bred for the purpose of experimentation
- Subject to any form of experimentation
- Retained in animal / breeding houses for further experiments or for education

The sole purpose of rehabilitation is alleviating the pain or suffering due to experimentation and to prolong the life of the animals until the point of natural death. Today CPCSEA has also made it a national policy that persons using laboratory animals have a moral responsibility towards these animals after the experimental use. Costs of rehabilitation of animals after the experiments are to be part of research costs.

Due to strict guidelines set by CPCSEA, the ethical concern about the use of animals in research is gaining momentum in India. The concept of the 3R's is only recently slowly attaining recognition among Indian scientists who use animals in their research. However, the general public is mostly unaware of the 3Rs as a fundamental set of principles for the use of animals in research, testing, and teaching (Bayne et al., 2015). Successful implementation of them requires that all personnel involved in handling and supervising animal experiments be suitably trained to scientifically and ethically evaluate the use of laboratory animals and to perform animal experiments that meet the highest animal welfare standards (Latha and Thomas, 2014).

20.3.2 Laboratory Animal Veterinarians

Please recollect that in MAW-001, we discussed the role of veterinarians in animal welfare and ethics. The implementation of 4Rs (reduction, refinement, replacement and rehabilitation) is the major responsibility of lab animal veterinarian. The specific responsibilities of laboratory animal veterinarians are summarised in Box 20.4.

Box 20.4: Specific Responsibilities of Laboratory Animal Veterinarians

- Daily inspections, supervision and welfare of lab animals.
- Participate in the ethical review and implementation of laboratory animal projects.
- Planning the use of lab animals in research.
- Monitoring, prevention and treatment of diseases and vaccination.
- Management and use of controlled drugs.
- Conducting post-mortem examinations and the production of post-mortem reports.
- Advising on measures to prevent and control of zoonoses, identifying the source of disease in order to minimize risks.
- Using isolation or segregation to prevent disease outbreaks when needed.
- Advise on quarantine programs when new animals arrive at the facilities and carry out the appropriate inspections.
- Advising and overseeing surgery and post-surgical care
- Professional guidance and performing the anesthesia, analgesia, and euthanasia.

(Please refer MAW-001, Unit 22 on 'role of veterinarians in animal welfare and ethics' for more details).

20.4 CLASSIFICATION OF LABORATORY ANIMALS

The lab animals are classified on the basis of level of sentience (Box 20.5), housing and the research protocol approving authority as:

- i) Small laboratory animals
- ii) Large laboratory animals

Box 20.5: Laboratory Animals vs. Sentience

The laboratory animals can be classified based on their level of sentience. A sentient being is one that has some ability to:

- Evaluate the actions of others in relation to itself and third parties
- Remember some of its own actions and their consequences
- Assess risks and benefits
- Have some feelings, and
- Have some degree of awareness

The level of sentience increases from invertebrates to birds, rodents, canines, felines, to bovines/equines to primates.

(Please refer MAW-001, Unit 19 for detailed discussion on 'animal ethics and sentience')

20.4.1 Small Laboratory Animals

Laboratory animals that can be housed in cages are called small laboratory animals.

- *Example:* Mouse, rat, guinea pig and rabbit

A rat is a medium-sized to large rodent, with a pointed muzzle, long slender and hairless tail, and dexterous forepaws. Mice are comparatively small in size weighing around 22-35 gm. Mice have large ear and long tail compared to body (Fig. 20.1). The Institutional Animal Ethics Committee (IAEC) approves the research protocol to use them in research.



Fig. 20.1: Small laboratory animals

20.4.2 Large Laboratory Animals

Laboratory animals that can't be housed in cages are categorized as large laboratory animals.

- *Example:* Dogs and Monkeys

They can only be housed in open run spaces. The protocols used in research which include these animals are approved by CPCSEA, where as the IAEC is only a recommending authority.

Laboratory animals can also be classified as:

- Rodents i.e. rats, mice, gerbils, hamsters, guinea pigs etc.
- Non-rodents i.e. lagomorphs, non-human primates, cats, dogs, sheep, goat, cattle, poultry, birds, fishes, amphibians etc.

20.5 IMPORTANT LABORATORY ANIMALS

20.5.1 Zebra Fish

There are characteristic zebra stripes running along the body and the fins, hence the name (Fig. 20.2). The lifespan of zebra fish is around 2-3 years. It is small in size (up to 6.4 cm) and hence culturing is easy. Zebra fish is developed from a fertilized egg to an adult outside the female in a transparent egg. This makes it possible to see changes in developing embryos.



Fig. 20.2: Zebra Fish

Research Use

- Gene expression patterns and gene functions can be studied by using zebra fish embryos. Their embryos are transparent and we can see dynamic gene expression in various tissues and organs *in vivo* without killing.
- Used for studying development of organs like heart, muscle, ocular etc. and nerve development.
- Used as a promising animal model for human diseases like cardiovascular diseases, cancer and behavioral studies.
- Used as biosensor for environmental toxicants.

20.5.2 Mouse

Mouse is the most widely used lab animal with more than 85% of the total biomedical research is conducted using mouse. There are more than 400 genetically defined inbred strains and many transgenic strains. However, most frequently used strain is albino with white coat color and pink eyes (Fig. 20.3).



Fig. 20.3: Mice

The presence of distinctive characteristics among inbred strains makes them suitable as lab animals. Mouse is a social animal, therefore it is housed in a small group of their own types. Mice are nocturnal, burrowing and climbing animal. They mark their territories with urine. The average life span of mouse is 1.5-2.5 years. Female mice has a gestation period of 19-21 days producing average litter size varying from 6-12 in number. Mouse like nesting so providing good nesting material is a good option to address the welfare issues.

Research Use

- Both inbred and out-bred strains are used widely in biomedical research because of its short gestation period, large litter size, great genetic diversity and genetically more closer to humans.
- Generally used in research for immunological drug screening, genotoxicity, infectious diseases like malaria, CNS (Central Nervous System) disorders, anti-prothrombin study, antiviral study, anxiety, TB testing, Alzheimer's disease, atherosclerosis, metabolic disorders (diabetes, obesity) etc.

20.5.3 Rat

The laboratory rats are docile social animals and are most commonly used next to the mouse in laboratories (Fig. 20.4). They are most comfortable if housed in groups to meet the welfare needs of company of the animal's own kind. They have highly developed sense organs and can sense human contact (touch), voice, smell and are responsive. They like digging, piling, borrowing, raring, gnawing, foraging etc. in the cage. Like the mice, they are also nocturnal animals and mark their territories with urine. They have average life span of 2-3 years. Female rats undergo 21-23 days of gestation period producing average litter size varying from 8-10 in number.

The rat is preferred for experimentation because of the following advantages:

- Small size compared to other animals so drugs are required in small quantity.
- Vomiting center is absent and so drugs can be administered orally.
- Because of the absence of gall bladder in rat there is continuous flow of bile into intestine. This facilitates the study of drugs acting on bile, cholesterol re-absorption etc.
- Pancreas is diffused, therefore, difficult to conduct pancreatectomy.
- In stomach, fundus and pyloric parts have clear lining and the gastric acid secretion is continuous.



Fig. 20.4: Rats

Research Use

- Used for regulatory toxicology and pharmacological studies, osteoporosis, infertility, hypertension, congestive heart failure, teratology, nephropathy, cancer and ageing, autoimmune diseases, diabetes etc.

20.5.4 Guinea Pig

The guinea pig is docile, short community animal that live together amicably thorough life (Fig.20.5). They develop rigid habits and dislike changes i.e. any change in food or water may cause them to stop eating. Guinea pigs are highly susceptible to tuberculosis and penicillin. It requires exogenous ascorbic acid in diet. Guinea pigs do not build nests but like to stay in nest made by others. They have an average life span of 3-5 years. Female counterparts has a gestation period of 68 days producing average litter size of 3-4 pups.



Fig. 20.5: Guinea Pig

Research Use

- Guinea pigs are primarily used in studies involving immunology, infectious diseases, audiology, nutritional and toxicological studies electrophysiology, antihistaminic activity, and TB research.
- They are well-established model for anaphylaxis (allergic reaction).
- Also used in bioassay of digitalis, evaluation of local anesthetics, study of histamine and antihistamines, hearing experiments because of sensitive cochlea and studies on isolated tissues especially, ileum, tracheal chain, vas-deferens etc.

20.5.5 Hamster

Hamsters are mouse like rodents and they are the third most widely used research model next to mice and rats (Fig. 20.6). Body is stout and compact with small furry ears; it is short with very small tail; has stocky legs, wide feet with thick set body, and prominent cheek pouches to transport food and nesting materials. Average life span is 2-3 years. Gestation period and average litter size are 16-21 days and 4-7 pups, respectively. They show special behaviour of hibernation (long physiological profound sleep).



Fig.20.6: Hamsters

Research Use

- Hamster is mostly used in biotransformation studies as it is susceptible for Leishmaniasis infection and metabolic diseases like diabetes mellitus.
- Research related to the immunology, implantation studies, cancer toxicology studies, virology and in bioassay of prostaglandin.
- They are also used to study the effect of hypothermia.

20.5.6 Rabbit

Rabbits are docile and timid animals with large ears (Fig. 20.7). The enzyme, atropine esterase is present in rabbit liver and plasma, so it can tolerate large dose of belladonna (atropine). Rabbit urine is normally thick and cloudy, with crystalline material. Coprophagy is a species-specific behavior of rabbits which ingest their own feces. The average lifespan of rabbit is around 4-6 years with a gestation period of 31 days and average litter size of 6-8 in number.



Fig. 20.7: Rabbits

Research Use

- Rabbits are very useful models in many biomedical research areas viz., embryology, toxicology, virology, etc.
- They are widely used in toxicity studies and safety testing (pyrogen, teratogenicity etc.).
- They are routinely used in serology to produce sera/antibodies in response to a wide variety of antigenic stimuli.
- They are also used in skin and eye irritation studies and also studies related to reproduction (anti-fertility agents) and local anesthetics.

20.5.7 Dogs

Beagle dogs are the most frequently used in biomedical research (Fig. 20.8). The mature, out-bred barrel-chested mongrels or hounds are also used as research animals in certain cardiovascular and skeletal studies. Dogs are always guided by their noses through their life, and they are never happier than when following an interesting scent. Most of the dog breeds are cheerful and fun loving, but the hounds may be inflexible and require patient and creative training techniques. Beagle was originally bred as scent hound to track small game, mostly rabbits and hares.



Fig.20.8: Beagle dogs

Research Use

- Dogs are commonly used as second (non-rodent) species in toxicity testing.
- Because of their distinct genetic, physiological and behavioral characteristics, they are used as models for behavioral analysis and genetic research.
- Dogs, like purpose-bred beagles, are used in cardiovascular studies, heart and lung research, dental research, genetic studies, age-related research, renal function studies, pulmonary studies and also for the development of prosthetic devices for hip and knee replacements.
- They are used to study the plasma elimination kinetics and evaluation of toxicity of drugs.

20.5.8 Non-Human Primates

Non-human primate groups are Prosimians (including lemurs, lorises, galagos and tarsiers) and Simians (monkeys and apes) (Fig 20.9). Monkeys and apes are primates belong to the highest order of mammals. The anatomy of monkey and apes are similar as that of human being. The studies done on monkeys may be directly transferred to human beings. Considering the welfare aspects, tests in the primates should be done only in the last stage of evaluation of drugs before clinical trials.



Fig. 20.9: Monkey

Research Use

- Physical, physiological and psychological similarity to humans makes the non-human primates most suitable pre-clinical test model before proceeding to clinical trials in human beings.
- Non-human primates are commonly used in research on developing vaccines, infectious, cardiovascular and neurological diseases, aging, reproductive biology, gene therapy, drug addiction, xeno-transplantation (transplantation of tissues from one species to the other) , toxicity testing and research on AIDS.
- Two most common primate species used in research are the Rhesus and Cynomolgus monkeys —also known as crab-eating macaques.
- Amongst the Great Apes the chimpanzees are the only species used in some specific biomedical research programmes.

Before we proceed, please complete activity 1.

Activity 1 (Visit and Observation Study): Visit any nearby research institute with lab animal facility. Discuss with the facility in charge on the type, number of animals and their purpose of keeping lab animals. Write the outcome of the discussion along with your observations.

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Check Your Progress 2

- Note:** a) Use the spaces given below for your answers.
b) Check your answer with those given at the end of the unit.

- 1) What is 3 R's concept?
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2) How do you classify the lab animals?

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3) What kinds of animals are used for research?

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4) Who approves the research protocol for using small and large laboratory animals?

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20.6 WELFARE ISSUES IN LABORATORY ANIMALS

Laboratory animal welfare is the totality of the concept of ensuring the health, psychological well-being, and the quality of life of laboratory animals, as well as the provisions made to achieve it. The welfare as applicable to laboratory animal is its state to cope with its environment. This includes both the extent of failure to cope and the ease or difficulty in coping. Coping is having control of mental and bodily stability. It is all about the treatment the lab animal receives i.e. animal care, animal husbandry and humane treatment. Practically the animal welfare is to reduce the negative experiences like pain, suffering and discomfort to the animal and improve the positive environment of the animal i.e. improved basic needs, enrichment, proper handling etc. Welfare is generally a relative term with respect to its need, health of animal, distress, adaptation (coping) and naturalness i.e. animal must live a reasonably natural life.

The major welfare issues in lab animals kept under confinement are:

- 1) Species differences and inadequate knowledge on the needs
- 2) Laboratory animal's environment
- 3) Housing conditions
- 4) Stress

- 5) Painful procedures
- 6) Use of animals for drug development
- 7) Using large sample of lab animals
- 8) Humane endpoint

20.6.1 Species Difference and Inadequate Knowledge on the Needs

Lack of understanding about “lab animal species difference” is cause of welfare concern. Mice, rat, rabbit, and guinea pigs are commonly used laboratory animals but they are very different from each other. Despite being close to humans in terms of genetic disposition, they might express difference in terms of their pathological conditions, physiological needs, and behavioral patterns with implications to welfare during their handling. Species difference is due to evolution, habitat, environmental conditions, geography, and behavior. Hence, researchers should be aware of these differences, so that pain and suffering can be minimized during experimentation.

Like in any other animals, the welfare of lab animals also comprises of three states - mental, physical and behavioural.

- a) **Mental Health:** Pain, fear and distress all compromise mental health and the emotional wellbeing of lab animals. Abnormal repetitive behaviours, stereotypy and compulsive behaviours are indicators of compromised mental health.
- b) **Physical Health:** It is the physical wellbeing of lab animals. Injuries, diseases and pain may all compromise physical health.
- c) **Behavioural Health:** It is the lab animals’ ability to demonstrate a normal behavioural range and to respond normally to novel stimuli.

The above three are essential components of lab animal welfare. In order to address these components, animal facility in research institutes has to be managed in a scientific manner. The lab animals housed in the facility have diverse and complex needs. The ability of animal facilities to meet these needs is slowed down by inadequate knowledge on management of lab animals in captive environment.

20.6.2 Laboratory Animals’ Environment

The lab animals’ environment must be optimally maintained throughout the breeding-holding phase and the experimental phase. The lab animals used for experiments need a well-controlled environment to keep them healthy. The design of animal accommodations must take into account the physical, nutritional, and general biological components in addition to the species maintained and the differing ages of the animals.

20.6.3 Housing Conditions

Housing conditions not only affect the behavior of the animals but also the experimental results. Adequate temperature, humidity, and air flow have to be maintained for all the animals in the first place. In animal house facilities, basic

requirements are provided but specific needs of each species of lab animal are hardly taken care of. Enrichment and refinement procedures can help in reducing the stress of animals in a particular environment (Zutphen et al., 2001; Balcombe et al., 2004). Enrichment procedures, aimed at providing the animals with an environment which meets their needs, provide them with opportunities to perform their species-specific repertoire and hence cause less stress in the animals which will affect their behavior in a positive way and can be considered a good option.

20.6.4 Stress

In addition to the physical pain, lab animals undergo severe stress during common laboratory procedures.

- *Example:* Elevations in pulse, blood pressure and stress hormones release that persist for some time after the procedure.

Stress responses in animals are also seen during caging, isolation, handling and blood collection. This not only compromises research results but also graphically illustrates the trauma that animals endure in laboratories.

20.6.5 Painful Procedures

Lab animals are routinely subjected to painful procedures. They are forcibly restrained, isolated, starved, addicted to drugs, subjected to painful procedures etc. They are usually killed afterwards. No procedure, no matter how painful, redundant or pointless, is prohibited by law.

Lab animal pain is a clinically important condition that adversely affects its quality of life. Drugs, techniques, or husbandry methods should be used to prevent, minimize, and relieve pain in animals experiencing or expected to experience pain. Protocols must be tailored to individual animals and should be based, in part, on the species, sex, breed, age, procedure performed, degree of tissue trauma, individual behavioral characteristics, assessment of the degree of pain, and health status of the animal (*Source: AVMA*).

20.6.6 Use of Animals for Drug Development

Administering drugs in the of trials on healthy lab animal, artificially inducing a condition that the lab animal would never normally contract, keeping the lab animals in an unnatural and stressful environment and trying to apply the results to naturally occurring diseases in human beings is dubious at best. Animals in laboratories also typically display behaviour indicating extreme psychological distress, and experimenters acknowledge that the use of these stressed-out animals jeopardizes the validity of the data produced (*Source: Chaitanya Koduri, PETA*).

20.6.7 Using Large Sample of Lab Animals

One of the principles under 3R's is reduction and any strategy that will reduce number of animals being used in laboratory research is good from welfare point of view. Research insists on large sample size to avoid errors from testing small sample size (atypical samples). However, most of the animal experiments are designed hazily on the basis of the literature available without any effort to calculate the sample size. Reduction in the number of experimental animals can be done by taking some precautions in the experimental design. They are:

- Preferring continuous measurements over categorical measurements
- Acquiring paired data wherever possible
- Performing one-tailed tests
- Precise measurements which reduce standard deviation and
- Using inbred strain of animals for the experiment

By taking care of the above mentioned points while designing the experiments and calculating the sample size, one can optimize the use of animals in the biomedical research.

20.6.8 Humane Endpoint

Humane endpoint is a refinement procedure, which avoid, alleviate or minimize the potential pain, distress or other adverse effects suffered by the lab animals involved, or which enhance animal well-being (Morton, 1998). This definition endeavors the practice of humane endpoints and justify their use in experimental design effectively. A humane endpoint is the point at which an experimental animal's pain and/or distress can be terminated, minimized, or reduced by actions such as killing the animal humanely, terminating a painful procedure, or providing treatment to relieve pain and/or distress (CCAC,1998). Defining the early endpoints can be a part of good experimental design and planning. Most research proposals submitted to the respective IAEC under CPCSEA guidelines in India does not include a description of humane endpoints. This leads to unjustified animal suffering when animals reach severe stages and are allowed to die from experimental disease. Experiments proposed should hence include humane endpoints, decided as the level of pain or suffering to which animals should not be allowed to exceed. Moreover, experimenting on a suffering or moribund animal will not generate valid experimental results. Researchers should thus emphasize the establishment of humane endpoints while designing the experiment for better outcomes and ethical study design overall. This refinement can thus not only improve the welfare of the animals but might also improve the experimental outcomes.

Before we proceed, please complete activity 2.

Activity 2 (Interaction): Visit a nearby research institute with laboratory animal facility. Discuss with the scientists and animal house in charge on how they are following the 3R's as fundamental welfare principles for the use of animals in research. Find out how they are practicing the 4th R i.e. Rehabilitation of laboratory animals. Write the outcome of the discussion with your observations.

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Check Your Progress 3

Note: a) Use the spaces given below for your answers.

b) Check your answer with those given at the end of the unit.

1) How the welfare of lab animals is linked to mental, physical and behavior states?

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2) How the lab animals' sample size in experimentation can be minimized?

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20.7 LET US SUM UP

- In this unit, you were introduced to the meaning and history of laboratory animals, the 3 R's and role of lab animal veterinarians, classification of laboratory animals, important laboratory animals and major welfare issues in laboratory animals.
- Laboratory animals are any vertebrate animals produced for or used in research, testing or teaching
- The major areas where the laboratory animals used are in drug development, toxicity (safety) testing, basic research and education.
- The Three R's concept (reduction, refinement and replacement) provide a framework for improving the conduct and ethical acceptability of experimental techniques on animals.
- The lab animals are classified based on their level of sentience, housing and the research protocol approving authority as small and large laboratory animals.
- Both small and large animals like mice, rats, guinea pigs, dogs, monkeys, poultry and farm animals are used for research.
- The IAEC approves the research protocol to use the small laboratory animals in research. For the large animals' research protocol, the CPCSEA is the approval authority, whereas the IAEC is only a recommending authority.

- One of the principles under 3R's is reduction and any strategy that will reduce the number of animals being used in laboratory research is good from welfare point of view.
- The important laboratory animals are zebra fish, mouse, rat, guinea pig, hamster, rabbit, dog and non-human primates.
- The major welfare issues in laboratory animals discussed are species differences and inadequate knowledge on the needs, laboratory animals' environment, housing conditions, stress, painful procedures, use of animals for drug development, using large sample of lab animals and humane endpoint.

20.8 KEYWORDS

Animal Ethics: Animal ethics is the term used to describe human-animal relationship and how animals ought to be treated.

Animal Welfare: Animal welfare means how an animal is coping with the conditions in which it lives.

Care of Laboratory Animals: The effective care given to laboratory animals by humans to safeguard their welfare and interests and to avoid causing them unnecessary harm.

Coprophagy: It is a species-specific behavior of rabbits to ingest their own feces.

Laboratory Animal Ethics: Compliance with the standards and principles of societal morality in the treatment of laboratory animals and the conduct of animal experiments.

Laboratory Animal Welfare: The totality of the concept of ensuring the health, psychological wellbeing and the quality of life of laboratory animals, as well as the provisions made to achieve it.

Laboratory Animal: An animal used in research, teaching, breeding, testing, and other scientific experiments.

Reduction: Methods which minimize the number of animals used per experiment to obtain the necessary data that support the scientific objectives.

Refinement: When laboratory animals must be used, the minimization as far as possible of the frequency of use of inhumane methods or of the degree of harm caused.

Replacement: Methods which replace animals of a higher life form with those of a lower life form, or avoid the use of animals and employ alternative methods to achieve the same objectives as experiments which use animals.

20.9 BIBLIOGRAPHY AND FURTHER READING

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20.10 SELF ASSESSMENT EXERCISES

- 1) Historically, research on animals played a key role in understanding various diseases and discovery of medicines. Illustrate with examples.
- 2) Discuss the role of the lab animal veterinarian in implementation of 4R's (reduction, refinement, replacement and rehabilitation) in research involving lab animals.
- 3) Name the important lab animals. Describe any three of them with their use in experimental research.
- 4) What are the major welfare issues in lab animals? Discuss any three of them with examples.

20.11 ANSWERS / HINTS TO CHECK YOUR PROGRESS

Check Your Progress 1

- 1) Laboratory animal is any vertebrate animal produced for or used in research, testing or teaching (NRC, 2011). Laboratory animal is an animal intended for use in research (OIE, 2012).
- 2) The major areas where the laboratory animals used are in drug development, toxicity (safety) testing, basic research, and education.

- 3) The IAEC is a local body, approved by CPCSEA, which has been empowered to permit experiments on small animals through an amendment in Rules for Breeding and Experiments on Animals (Control and Supervision) in the year 2006.

Check Your Progress 2

- 1) The Three R's concept (reduction, refinement and replacement) provide a framework for improving the conduct and ethical acceptability of experimental techniques on animals.
- 2) The lab animals are classified based on their level of sentience, housing and the research protocol approving authority as small and large laboratory animals.
- 3) Both small and large animal like mice, rats, guinea pig, dogs, monkeys, poultry and farm animals are used for research.
- 4) The Institutional Animal Ethics Committee (IAEC) approves the research protocol to use the small laboratory animals in research. For the large animals' research protocol, the CPCSEA is the approval authority, whereas the IAEC is only a recommending authority.

Check Your Progress 3

- 1) Pain, fear and distress all compromise mental health and the emotional wellbeing of lab animals. Abnormal repetitive behaviours, stereotypy and compulsive behaviours are indicators of compromised mental health. Physical health is the physical wellbeing of lab animals. Injuries, diseases and pain may all compromise physical health. Behavioural health is the lab animals' ability to demonstrate a normal behavioural range and to respond normally to novel stimuli.
- 2) One of the principles under 3R's is reduction and any strategy that will reduce number of animals being used in laboratory research is good from welfare point of view. Reduction in the number of experimental animals can be done by taking some precautions in the experimental design viz., preferring continuous measurements over categorical measurements, acquiring paired data wherever possible, precise measurements and using inbred strain of animals for the experiment.