2 Selection of Biomedical Animal Models

MICHAEL S. RAND

ABSTRACT

Laboratory animals play a crucial role in research discovery and technological advances, and they will continue to take part in improving the lives of people and other animals. It is incumbent upon the researcher to know his subject well in order to provide relevant information to the scientific world. In an effort to assist the biomedical researcher in gaining this knowledge, this chapter provides the following key elements: definition of types of animal models, legislative and legal requirements, criteria for choosing a model, extrapolation validity recommendations, and descriptive features for publication.

Key Words: Animal models, Laboratory animal(s), Animal model types, Animal use criteria, Choosing animal model, Animal factors, Extrapolation, Animal description.

INTRODUCTION

Over the last one and one-half centuries, almost all medical knowledge, treatment regimes, and medical device development have involved research using animals. The key factor in using animals in research is in its extrapolatability of results to humans. Animals in research have been and still are essential in developing treatments for asthma, HIV/AIDS, cancer, birth defects, bioterrorism medical countermeasures, vaccines, antibiotics, high blood pressure, and much more. Additionally, they have been vital in the development of antibiotics, vaccines, and organ transplantation techniques.[1] As the rise in emerging infectious diseases (e.g., West Nile virus and avian influenza) continues, animals will be key and essential in the development of preventive and treatment modalities.

THE HISTORY OF ANIMAL USE IN RESEARCH

The use of animals to study human physiology and anatomy can be traced back to the second century AD in which Galen was a Greek physician and philosopher. His research was based almost exclusively on studies using apes and pigs. Unfortunately, this initiated many errors based on his accepted authority and the prohibition by the Church of using human cadavers for research purposes. Galen was later blamed for using incorrect methods in research when in truth it would be more accurate to say that he drew wrong conclusions based on uncritical interspecies extrapolation of data. That is, he assumed that all extracted information derived from his use of animals could be directly applied to humans. It was not until the late sixteenth century that this error began to be recognized.

Modern research principles can be attributed to three physiologists from the 1860s. In 1865, Claude Bernard, a French physiologist, published *An Introduction to the Study of Experimental Medicine*.[2] This book was intended to provide physicians with guidance in experimental research. It proposed the use of chemical and physical induction of disease in animals, thus becoming the first published book to advocate creating "induced animal models" for biomedical research. His peers of the time were Louis Pasteur in France and Robert Koch in Germany. Louis Pasteur and Robert Koch introduced the concept of specificity into medicine and the "germ theory of disease." The turning of the century saw the development and use of animal models for infectious diseases and screening and the evaluation of new antibacterial drugs based upon the work of these three researchers.

During the first quarter of the nineteenth century, animal studies were crucial for less than one-third of the major advances that occurred. With the contributions of Claude Bernard, Louis Pasteur, and Robert Koch, animal studies contributed to more than half of the significant discoveries made thereafter. Since 1901, two-thirds and 7 of the last 10 Nobel Prizes in medicine have relied at least in part on animal research.[3] Today, researchers rely on the identification and development of animal models to explore all avenues of medical science to include assessment of pathogenic mechanisms, diagnostic and therapeutic procedures, nutrition and metabolic diseases, and the efficacy of novel drug development.

THE CONCEPT OF ANIMAL MODELS

WHAT IS AN ANIMAL? Etymologically, the word "animal" derives from the Latin *animal* meaning soul/spirit, thus describing living organisms that are *animated.*

WHAT IS A MODEL? A model is an *object of imitation,* something that accurately resembles something else, a person or thing that is the likeness or *image* of another. The *Holy Bible* tells us that God said, "Let us make man in our image, in our likeness, so God created man in his own image, in the image of God he created him; male and female he created them." God created man out of the dust of the ground and then breathed into his nostrils the breath of life to animate him. Thus, humans are "animal models" of God.

Consequently, combining the two definitions, an "animal model" is an *animated object of imitation,* an "image of Man" (or other species), used to investigate a physiological or pathological circumstance in question.[4]

From: *Sourcebook of Models for Biomedical Research*
(P. M. Conn, ed.), © 2008 Humana Press Inc., Totowa, NJ.

WHAT IS AN ANIMAL MODEL? The U.S. National Research Committee on Animal Models for Research on Aging attempted to define the term "laboratory animal model" as "an animal in which normative biology or behavior can be studied, or in which a spontaneous or induced pathological process can be investigated, and in which the phenomenon in one or more respects resembles the same phenomenon in humans or other species of animal."

Using the term animal model can be confusing because what is often meant by the term "animal model" is actually studying human conditions. In other words, it is not the image of the preferred animal that is the focus of research but the analogy of the physiological behavior of this animal to our own (or another) species. It would, thus, be more correct to speak of "human models" in this context. Indeed, although using animals in research can benefit other animals, it is much more focused on improving the human condition.

TYPES OF ANIMAL MODELS

When animals are used in research to study biological and functional systems in humans, they are broken down into the following categories:

1. **Exploratory.** Animals used in this category are used to gain an understanding of fundamental biological mechanisms, whether normal or abnormal. An example would be the use of a novel animal model of aging, particularly for identifying genes and biochemical pathways regulating longevity.

2. **Explanatory.** Animals used in this category are used to gain an understanding of complex biological problems. An example would be the use of cognitive and psychosocial animal models to provide an etiology for anorexia nervosa.[5]

3. **Predictive.** Animals used in this category are used to discover and quantify the impact of investigative treatments whether for diseases or chemical toxicities. Predictive animal testing models are important in improving the success of a drug or medical device in clinical trials and for generating new data in support of the ongoing marketing of existing products.

When animals are used in disease research, they are broken down into the following categories:

1. **Induced (or Experimental).** Induced models are ones in which normal animals are experimentally created either through surgical modifications, genetic modifications, or chemical injections. An example would be a myocardial infarction induced by coronary artery surgical ligation.

2. **Spontaneous.** Spontaneous models are genetic variants, which mimic the human condition. The variance occurs naturally through mutation and not by experimental induction. The *nu* mutation was first reported in 1966 in a closed stock of mice in a laboratory in Glasgow, Scotland. It was not until 1968, however, that it was discovered that the homozygous nude mouse also lacked a functional thymus, i.e., it was *athymic*. The mutation produces a hairless state, generating the name "nude." The other, unique defect of nude mice is the failure of the thymus to develop normally to maturity. The thymus remains rudimentary and produces reduced numbers of mature T cells. This means nude homozygotes (animals with identical mutant genes at corresponding chromosome loci) do not reject allografts and often do not reject xenografts (tissue from another species). The discovery that human neoplasms (tumors) could be grown in nude mice was immediately recognized as an important research tool. Thus, the spontaneous mutation of *nu* among laboratory mice was a serendipitous development that led to the nude mouse becoming the first animal model of a severe immunodeficiency. In the decades since, the nude mouse has been widely utilized by researchers studying factors regulating transplantable human tumor growth and cancer metastasis.[6]

3. **Transgenic.** Transgenic models are induced models in which DNA is inserted into or deleted (knockout) from the genome of the animal. The term "transgenic" was coined in 1981 by Gordon and Ruddle to describe an animal in which an exogenous gene was introduced into its genome. In the late 1980s, the term transgenic was extended to gene-targeting experimentation and the production of chimeric or "knockout" mice in which a gene (or genes) has been selectively removed from the host genome. Today, a transgenic animal can be defined as one having any specific, targeted genetic modification. Transgenic animals are most commonly produced through (1) germline modifications of gametes, (2) microinjection of DNA or gene constructs into zygotes (unicellular embryos), or (3) incorporating modified cells, including embryonic stem (ES) cells, into later stage embryos. After gamete or embryo modifications, the resultant embryos are matured to term in a recipient female.[7]

4. **Negative.** Negative models fail to react to a disease or chemical stimulus. Thus, their main use in biomedical research is for studies on the mechanism of disease resistance. A classic example is the failure of gonococcal infection to develop in rabbits after an experimental treatment that induces the disease in other animals. Negative animal models have become increasingly important with the advent of transgenic technology. For example, a novel transgenic mouse was created to study the lack of development of autoimmune thyroiditis with the injection of self-thyroglobulin. This strain of mice lacked certain surface epitopes to account for this negative reaction.[8]

5. **Orphan.** Orphan models are the opposite of negative models. Orphan models are animals in which a disease occurs but there is not a corresponding disease in humans. Orphan models may become induced models when a similar disease is recognized in humans later on. Historically, scrapie in sheep was such a model, but now is useful as a model for the human spongiform encephalopathies that are of so much concern (e.g., BSE, "mad cow disease," and CWD, chronic wasting disease in deer).

All categories above may be further subcategorized with the following divisions:

1. **Fidelity.** The extent a biological structure in an animal resembles that of a human. Thus, a high fidelity animal model gives a highly relevant biological closeness to the human structure. Model fidelity is best conceptualized as a continuous spectrum, ranging from low to high fidelity. Examples of low-fidelity models include bench models made of simple materials that often have little anatomical resemblance to reality. However, these models incorporate some of the key constructs of the simulated tasks. At the other end of the spectrum are high-fidelity models such as human or animal cadavers or the new array of virtual reality simulators. These simulators usually incorporate highly realistic visual and tactile cues in the midst of a highly interactive model. In between these two extremes, almost any kind of intermediate fidelity can exist.

2. **Homologous.** The symptoms shown in the animal are identical to those shown in the human. For instance, the recent discoveries of swine hepatitis E virus (HEV) from pigs and avian HEV from chickens afforded an opportunity to develop small homologous animal models for HEV.[9]

3. **Isomorphic.** The animal's symptoms or anatomy are similar to those in the human but the etiology or genetic character is different. For example, there is a set isomorphism between the human and mouse heart at the organ level and also at the organ part level: each species has a heart and a corresponding set of cardiac chambers (right and left atrium, right and left ventricle) and the wall of each chamber has a corresponding set of layers (epicardium, myocardium, endocardium).

4. **Partial.** These models do not mimic the entire human disease but enough similarities exist to allow their use in studying aspects of the disease or treatments. For instance, animal models of Alzheimer's disease can be created based on the accumulation of increased levels of amyloid-ß peptide in the brain and have many amyloid plaque deposits; however, they have only subtle behavioral and electrophysiological deficits, thus providing only a partial model of the human condition.[10]

5. **Face validity.** The degree to which there is a similar phenotypic display between the disease in the animal and the corresponding disease in the human. For example, it could be argued that the demonstration of drug effects in an animal model for depression after a period of chronic administration is important for establishing its face validity, but is not relevant to the model's predictiveness and therefore to its ability to serve as a screening test for treatments for the modeled disease.[11]

6. **Construct validity.** The degree to which there is a similar genetic display between the disease in the animal and the corresponding disease in the human. As an example of high construct validity, research was performed on three candidate dopaminergic genes (DRD2, DRD4, and DAT-1) that were sequenced in spontaneous hypertensive (SHR) and Wistar Kyoto (WKY) rats. No differences were found in DRD2 or DRD4 genes, but several variations were found in the DAT-1 gene that are of significance because several ADHD families show linkage to DAT-1. It also strengthened the validity of using WKY as a control for SHR, because their behavioral characteristics are similar to those of other rat strains.[12]

LEGISLATIVE AND LEGAL REQUIREMENTS FOR USING ANIMALS IN RESEARCH

Biomedical research is among the most regulated industries in the world. A comprehensive overview of global requirements can be found in the *Handbook of Laboratory Animal Science*, 2nd edition, Chapter 3.[13] Failure to comply with regulatory requirements can result in fines levied against the institution, suspension of authority to operate, permanent revocation of the facility's license, and withdrawal of public funding.

One newly regulated aspect of biomedical research not covered in this chapter occurred after the terrorist attack on September 11, 2001. The attack increased concerns in the United States for the possibility of bioterrorism using agents that would destroy human, animal, and plant life. This concern escalated the need for research that involved the development of therapeutic and preventive measures against such agents. In response, congress passed and President Bush signed into law the "Public Health Security and Bioterrorism Preparedness and Response Act of 2002" (Public Law 107–188) on June 12. The purpose of the act was to improve the capacity of the United States to prevent, prepare for, and respond to bioterrorism and other public health emergencies and to enhance the control of dangerous biological agents and toxins. The Centers for Disease Control and Prevention (CDC) is the agency with the primary responsibility for implementing the provisions of the Act with regard to human pathogens and toxins and the United States Department of Agriculture (USDA) with regard to animal and plant pathogens and toxins. The regulation provides for expanded regulatory oversight of select agents and toxins, and a process for limiting access to persons who have a legitimate need to possess, use, or store these agents. The regulation also establishes a requirement for a security risk assessment performed by the Federal Bureau of Investigation for those persons needing access to select agents and toxins. It also establishes and enforces safety and security procedures, including measures to ensure proper training and appropriate skills to handle agents and toxins; a requirement to designate an institutional Responsible Official to ensure compliance with the regulations; and a requirement to obtain a certificate of registration when there is a need to possess, use, or transfer select agents and toxins. Infectious agents labeled as "select" as determined by the CDC and USDA, registration forms, and other information concerning the Select Agents Program may be found at http://www.selectagents.gov.[14]

CHOOSING THE RIGHT MODEL

To quote the philosopher, Bernard Rollin, "The most brilliant design, the most elegant procedures, the purest reagents, along with investigator talent, public money, and animal life are all wasted if the choice of animal is incorrect." Once it has been determined that the use of laboratory animals is necessary, the most appropriate species, breed, and strain with the closest homology to humans must be chosen in order to give the research face and/or construct validity. Because new animal models are continually being identified and characterized and the field of biomedical research has become global in nature, the search for the appropriate animal model should start with a thorough literature search and a check of appropriate web sites (see Chapter 7). The Institute for Laboratory Animal Research maintains a very practical and useful search engine for this purpose.[15]

Selection of a species should not be based solely on availability, familiarity, or cost. Animals that meet these criteria may not provide the genetic, physiological, or psychological facets needed or wanted for the proposed project. It is almost impossible to give specific rules for the choice of the best animal model, because the many considerations that have to be made before an experiment can take place differ with each research project and its objectives. Nevertheless, some general rules can be given.

RESEARCH FACTORS

- Appropriateness as an analogue. Ensuring that the part or organ being studied has a function similar to the target species is vital in applying research-derived data from the chosen model.
- Transferability of information. The usual goal of research using an animal model is to define a process in a system with the hope of transferring the data gained to a more complex system. Traditionally, one-to-one modeling is

sought: modeling in one group of organisms that can be transferred to another group that has several analogous features of interest. This is especially helpful in modeling disease states. However, in modern research, many-to-many models are mainly used. This technique begins by analyzing the component parts of a process or disease, and then finding for each component analogous models in many taxa of living species.[16] This is especially helpful when a plurispecies approach is needed to gain approval for new medications or medical devices.

- Generalizability of the results. The ability to generalize results to the target species is important. Federal regulations prohibiting the unnecessary duplication of previous research highlights the importance of choosing an animal model in which testing results can be easily repeatable and verifiable on which to build new research. In May, 2006, the world was shocked when famed South Korean cloning scientist Dr. Hwang Woo-suk was charged with fraud and embezzlement when scientists could not verify his published data. In addition, if the ultimate target species is human, it is well known that this species is genetically highly variable, with cultural, dietary, and environmental differences. This may be of lesser importance in disease modeling since most diseases do not choose its victim based on genetic variability. However, this is now well known to be of importance in pharmacological and toxicological modeling and has opened up the new field of research in pharmacogenetics.
- Ethical implications. Certainly research must start with justification for using an animal at all. Federal regulations require the use of alternate methods if feasible. Alternate methods could consist of using cell lines, bacteria, computer models, or even human volunteers. The three Rs of Russell and Burch (replacement of existing experiments with animal-free alternatives, or reduction in the number of animals used, or refined methods to reduce animal suffering) help to meet the ethical concerns.[17]
- Numbers needed. Certainly consultation with a biostatistical analyst prior to submitting a proposal is highly recommended. Numbers needed to provide scientific validity, especially for publication, will impact many other factors such as cost and housing availability.
- Customary practice within a particular discipline. Caution must be displayed when using this criterion. Customary practices may not always mean that the most appropriate animal model has been used. The "customary" animal may not represent the most accurate genetic, microbiological, physiological, or psychological facets needed for the study. Historical evidence has revealed that using animal models just because others have has led to substandard results. However, customary practices when justified and supported by the other criteria listed can be a satisfactory and faster route of choosing the animal model needed.
- Existing body of knowledge of the problem under consideration. This criterion again emphasizes the need for a thorough literature search before forming the basis for the research project. The literature search will emphasize what is already known to prevent accidental duplication, but will also reveal what is not known. It will also make known published authorities in the discipline that may serve as a consultation source to prevent unnecessary and competitive research projects.
- Natural versus experimentally produced models. Unavailability of natural models will require the use of experimentally produced models. Depending on the objectives of the study, both may be needed.

ANIMAL CARE FACTORS

- Cost and availability. Certainly cost and availability are important factors when choosing an animal model, but they can be disastrous if the decision is based solely on cost and not the other listed factors. Cost also includes ongoing care not only for husbandry but also from experimental manipulations. Certainly, the best animal model can be in short supply as illustrated by the CNN news report on August 9, 2003. This report emphasized the increased demands in research due to public health crises such as AIDS and the threat of bioterrorism. The increased demands have led to a national shortage of rhesus macaques. In addition, the shortage has skyrocketed the cost per monkey.
- Housing availability. Another practical consideration in choosing the animal model is the accessibility of housing. Research animal housing requirements are stringent and may lessen the availability according to the species chosen. For instance, choosing a nonhuman primate may require the purchase of new caging and the hiring of additional personnel to provide specialized husbandry care, as opposed to choosing mice, which can be placed several to a cage and hundreds in a room.
- Husbandry expertise. Some models require not only special housing, but also special care.
- Stress factors. Stress sources from many different causes can affect the animal's physiology, biochemistry, and behavior. Sources of stress can be transportation, handling and manipulations, overcrowding, lack of environmental enrichment, and the research project itself.

PHYSICAL AND ENVIRONMENTAL FACTORS

- Ecological consequences. While the best animal model may be available only by capturing in the wild, ecological consequences must be considered in its removal. In addition, safety measures must be in place to prevent accidental escape from the research facility. A prime example is the *Xenopus* spp. frog. If it escapes, it can overrun local ponds and rivers endangering natural amphibian populations. Furthermore, care must be taken not to violate the Endangered Species Act (http://www.fws.gov/endangered/wildlife.html) or the Convention on International Trade in Endangered Species of Wild Fauna and Flora (http://www.cites.org).
- Hazardous components. Many research projects entail the use of chemicals, infectious agents, and radioisotopes. The uses of these components are highly regulated and the appropriate proper authority (Institutional Biosafety Committee, Radiation Safety Committee, Occupational Health and Safety Committee, Environmental Health and Safety Committee, and Institutional Animal Care and Use Committee) within each institution must give approval for

its use. In addition, all those who will be exposed, whether from the research side or the animal care side, must be notified.

- Environmental influences. Environmental aspects may be important to a particular animal species.[18] Environmental factors that fit into the broad categories of physical, chemical, biological, and social may impact the physiological and behavioral responses of animals. These factors include humidity, ventilation, light cycle and quality, noise, cage size and bedding materials, diet and water, and room temperature.[19] As an example, high temperature and humidity have been proven to impair memory in mice.[20]

ANIMAL-RELATED FACTORS

- Genetic aspects. Uniformity of organisms may be necessary where applicable. "This insidious evolution of the inbred genotype is known as genetic drift. It is capable of subverting the conclusions reached about comparable research results coming from different laboratories when each uses its own subline of the same inbred strain (Bailey DW, 1977)."[21] The importance and methods of preventing genetic drift in biomedical research can be found at http://jaxmice.jax.org/geneticquality/drift.html. In addition, it is important to remember that to be in compliance with the *National Institutes of Health's Guidelines*,[22] work with transgenic animals requires the approval of the Institutional Biosafety Committee as well as the Institutional Animal Care and Use Committee.
- Background knowledge of biological properties. Knowledge of biological properties such as generalized and specialized function of body components is needed in order to validate interspecies transfer of information. Certainly, a rat would not be the best choice in biliary studies due to the absence of a gallbladder. Knowing the biological properties also aids in the decision of whether the animal is a spontaneous model or must be experimentally induced.
- Ease of and adaptability to experimental manipulation. This is unquestionably a practical matter. Guinea pigs have highly inaccessible blood vessels and would be impractical in studies requiring repeated blood sampling. Prairie dogs and woodchucks can be vicious to handle; therefore, knowing the response to experimental manipulation may also influence the choice.
- Size of the animal. This item is important from several different aspects. The size of the animal impacts housing and husbandry availability. However, size is also important to consider when tissue sampling or blood collection is necessary. For instance, many proposals are rejected because the researcher failed to abide by published guidelines for removal of blood.[23] In addition, it is also important to incorporate the size of the animal into the decision-making apparatus when physiological or morphological properties such as joint strain or organ size must be identical to that of a human, especially when developing medical devices.
- Life span and age. Studies requiring components at different stages of life can certainly impact the species chosen. The average lifespan of a rat is 2.5–3.5 years, whereas it can be over 30 years for a rhesus monkey.
- Sex. The alternating cycle of hormonal production in the female gender and its influence on the data outcome must be considered when planning for the research project.
- Progeny needed. Female mice and rats can produce 5–10 progeny per month, whereas the rhesus monkeys only one or two per year. *Xenopus* sp. frogs produce thousands of ova during their lifetime, whereas mammals produce only dozens.
- Diseases or conditions that might complicate results. An excellent historical review on the struggle against pathogens in laboratory rodents can be found in Weisbroth.[24] The effects on research can be found in Baker.[25] Both publications emphasize the need for disease-free animals in research to prevent adverse effects on resultant data. Just as in human AIDS, the realm of disease-causing organisms changed with the advent of immune deficient models. Special caging and care procedures are fundamental in minimizing such infections.
- Special features of the animal such as unique responses or microflora. It is important to be familiar with unique anatomical or physiological features of the species you will be working with. The results could be quite unexpected otherwise. For example, in rabbits, the terminal portion of the ileum empties into an enlarged rounded viscus called the sacculus rotundus and not the colon as in humans. This unique feature of the rabbit is important to know when designing gastrointestinal studies.

Forming the above standards into a checklist will help to fulfill the criteria needed to choose the best model for the proposed research project. Model selection is the privilege of individual researchers, but they must be very cautious in their selection because in the end, it is up to them to convince the rest of the scientific community that they made the right choice.

Before choosing, consultations should occur with scientists who have already used the animal model. Just as with equipment purchase, communicating with previous users can be very helpful in learning unique features of the selected species, breed, and strain. Not all attributes (especially negative ones) are published, making it even more important to contact those who have experience with the animal model you choose.

Preparatory consultation should also occur with those who will be responsible for housing and maintaining the animals, as they will be the most familiar with the care of the animal and its physical and environmental needs. Preparatory consultation with the laboratory animal veterinary practitioner should also occur to discuss the animal-related factors.

EXTRAPOLATION FROM ANIMALS TO HUMANS

Extrapolation from animals to humans does not necessarily mean that biomedical research data obtained from using animals are then used to find a corollary in a human. Rather, a hypothesis is formed first based on human relevancy, and then tested on an animal. Answers are obtained, analyzed, and published based on the hypothesis. Although true in many cases, caution must be exerted in assuming that a close phylogenetic relationship or anatomical similarity guarantees an identical biochemical or physiological response in the animal. In addition, it must be realized that humans to whom the results are being extrapolated are genetically highly variable due to cultural, dietary, and environ-

mental differences. This is of minor importance when developing disease models but is highly important for pharmacological and toxicological models.

So, how can the validity of extrapolation be verified? Complete reliability cannot be guaranteed; however, following the following vital requirements will help to avoid several of the mistakes of the past and overcome problems of the future:

- *Taking a plurispecies approach.* Most of the regulating authorities require two species in toxicology screening, one of which has to be nonrodent. This does not necessarily imply that excessive numbers of animals will be used. The uncritical use of one-species models can mean that experimental data retrospectively turn out to be invalid for extrapolation, representing real and complete waste of animals. Using more than one species is, of course, no guarantee for successful extrapolation either.
- *Metabolic patterns and speed and body size must match between species.* The use of laboratory animals as models for humans is often based on the premise that animals are more or less similar with respect to many biological characteristics and thus can be compared with humans. However, there is one striking difference between mouse and human, and that is body size. In proportion to their body size, mammals generally have very similar organ sizes expressed as percentage of body weight. Take the heart for instance, which often constitutes 5 or 6 g per kilogram of body weight, or blood, which is often approximately 7% of total body weight. It is well known that the metabolic rate of small animals is much higher than that of large animals and, thus, provisions must be made to adjust the study accordingly. Drugs and toxins exert their effect on an organism not per se but because of the way that they are metabolized, the way that they and their metabolites are distributed and bound in the body tissues, and how and when they are finally excreted. Adjusted doses should include the following provisions:
  - If the object is to achieve equal concentrations of a substance in the body fluids of animals of different body size, then the doses should be calculated in simple proportion to the animals' body weights.
  - If the object is to achieve a given concentration in a particular organ over a certain time period, the calculation of dosage becomes more complicated, and other factors, including the physicochemical properties of the drug, become important.
  - Metabolism or detoxification and excretion of a drug are not directly correlated with body size but, more accurately, to the metabolic rate of the animal.
  - Some species react with particular sensitivity toward certain drugs, and marked variations in the reaction of animals within a species occur with respect to strain, pigmentation, nutritional state, stress level, type of bedding, ambient temperature, age, sex, route or time of administration and sampling, diurnal variation, and season of the year. As much as possible, these items must be controlled.[4]
- *Experimental design and the life situation of the target species must correspond.* A model cannot be separated from the experimental design itself. If the design inadequately represents the "normal" life conditions of the target species, inaccurate conclusions may be drawn, regardless of the value of the model itself.

DESCRIPTION OF ANIMAL MODELS[16]

Unlike the old days when the researcher could write in the materials and methods section "black mice were used in the study," modern obligations require an exact description of the model. The description should include the following.

- Genetic strain and substrain using correct international nomenclature.[26,27]
- Special genetic features.
- Microbial status of the animal.
- Age.
- Housing standards.
- Maintenance procedures.
- Diet.
- If used in infectious disease studies, the description should also include
  - Strain of the organism.
  - Method of inoculum preparation.
  - Route of inoculation.

CONCLUSIONS

Laboratory animals play a crucial role in research discovery and technological advances, and they will continue to take part in improving the lives of people and other animals. It is incumbent upon the researcher to know the subject well in order to provide relevant information to the scientific world. The final judgment in the choice of the animal model will always be in its ability to elucidate and predict the observed effects in the target species.

REFERENCES

1. Americans for Medical Progress. Everyday wonders. (Accessed August 16, 2006, at http://www.amprogress.org.)
2. Bernard C. *Introduction to the Study of Experimental Medicine*. Mineola, NY: Dover Publications, 1957.
3. Foundation for Biomedical Research. Nobel prizes: The payoff from animal research. (Accessed on August 30, 2006, at http://www.fbresearch.org/education/nobels.htm.)
4. Hau J. Animal models. In: Hau J, Van Hoosier GL, Eds. *Handbook of Laboratory Animal Science*, 2nd ed., Vol. II. Boca Raton, FL: CRC Press, 2003:1–9.
5. Connan F, Campbell IC, Katzman M, Lightman SL, Treasure J. A neurodevelopmental model for anorexia nervosa. *Physiol Behav* 2003;79:13–24.
6. Immunodeficient rodents opening new doors for investigators. *Res Anim Rev* 1996;1(2):1–8.
7. Pinkert CA. The history and theory of transgenic animals. *Lab Anim* 1997;26:29–34.
8. Yan Y, Panos JC, McCormick DJ, *et al.* Characterization of a novel H2A(−)E+ transgenic model susceptible to heterologous but not self thyroglobulin in autoimmune thyroiditis: Thyroiditis transfer with Vbeta8+ T cells. *Cell Immunol* 2001;212(1):63–70.
9. Meng XJ. *Discoveries of Animal Strains of Hepatitis E Virus: Implications for Animal Models and Zoonosis.* Ithaca, NY: American College of Veterinary Pathologists and American Society for Veterinary Clinical Pathology, 2004. (Accessed at http://www.ivis.org.)
10. St. George-Hyslop PH, Westaway DA. Antibody clears senile plaque. *Nature* 1999;400:116–117.
11. Willner P. The validity of animal models of depression. *Psychopharmacology* 1984;83:1–16.

12. Sagvolden T, Russel VA, Aase H, Johansen EB, Farshbaf M. Rodent models of attention-deficit/hyperactivity disorder. *Biol Psychiatry* 2005;57:1239–1247.
13. Bayne K, deGreeve P. An overview of global legislation, regulation, and policies on the use of animals for scientific research, testing, or education. In: Hau J, Van Hoosier GL, Eds. *Handbook of Laboratory Animal Science*, 2nd ed., Vol. I. Boca Raton, FL: CRC Press, 2003:31–50.
14. Fleischauer AT, Ellis B, Ostorff S, Hemphill MS. The select agents and toxins interim final regulations: Applications to the small animal facility. *Contemp Top Lab Anim Sci* 2003;42(4):126–140.
15. Animal models and strains search engine. Washington, DC: Institute for Laboratory Animal Research. (Accessed August 16, 2006, at http://dels.nas.edu/ilar_n/ilarhome/search_amsst.shtml.)
16. Davidson MK, Lindsey JR, Davis JK. Requirements and selection of an animal model. *Isr J Med Sci* 1987;23:551–555.
17. Russell WMS, Burch RL. *The Principles of Humane Experimental Technique*. London: Methuen, 1959.
18. Environmental control for animals and plants. In: *ASHRAE Handbook*. Atlanta, GA: American Society of Heating, Refrigerating & Air Conditioning Engineers, Inc., 2005:Chapter 10.
19. Rose, MA. Environmental factors likely to impact on an animal's well-being-an overview. In: Baker RA, Jenkin G, Mellor DJ, Eds. *Improving the Well-Being of Animals in the Research Environment*. Adelaide, Australia: ANZCCART, 1994:284–301.
20. Inan SY, Aksu F. Amnesic effects of relative humidity and temperature in mice. *Lab Animal* 2002;31(2):40–48.
21. Bailey DW. Genetic drift: The problem and its possible solution by frozen-embryo storage. *Ciba Found Symp* 1977;291–303.
22. NIH guidelines for research involving recombinant DNA molecules. Bethesda, MD: National Institutes of Health, 2002.
23. Morton DB, Abbot D, Barclay R. Removal of blood from laboratory mammals and birds. *Lab Anim* 1993;27:1–22.
24. Weisbroth SH. Post-indigenous disease: Changing concepts of disease in laboratory rodents. *Lab Anim* 1996;25:25–33.
25. Baker DG. Natural pathogens of laboratory mice, rats, and rabbits and their effects on research. *Clin Microbiol Rev* 1998;11(2): 231–266.
26. Mouse Genome Informatics. Rules for nomenclature of mouse and rat strains, 2005. (Accessed August 30, 2006, at http://www.informatics.jax.org/mgihome/nomen/strains.shtml.)
27. Mouse Genome Informatics. Rules for nomenclature of genes, genetic markers, alleles, and mutations in mouse and rat, 2005. (Accessed August 30, 2006, at http://www.informatics.jax.org/mgihome/nomen/gene.shtml.)