

Bibliography For Members Of Institutional Animal Care And Use Committees, March 2008



*Animal Welfare Institute
Washington, DC*

This bibliography may serve as a guide to published and online material assisting non-affiliated and affiliated members of IACUCs in their commitment to ensure ethically and scientifically acceptable research protocols involving live animals. It is available on the web at: http://www.awionline.org/lab_animals/iacuc2.htm

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GENERAL

Animal Care Matters. Committee on Animal Care. 1993. Massachusetts Institute of Technology (MIT). Available from MIT, 37 Vassar Street 45-105, Cambridge, MA 02139.

This 25 minute-videotape is designed to aid institutions in providing effective education to animal research personnel. Included are constructive discussions of the ethical and moral issues concerning animal research, the role of laboratory personnel for ensuring humane treatment and species-adequate housing of research animals, applicable legislative and regulatory guidelines, the responsibility of IACUCs, and alternatives to animals in biomedical research.

Animals, Science, and Ethics. Donnelley S and Nolan K, eds. 1990. *The Hastings Center Report*, Supplement May/June.

Invaluable background information addressing ethical theory and the moral status of animals, animals in science, animal suffering and IACUCs.

Animal Welfare Information Center Bulletin. Animal Welfare Information Center, National Agricultural Library, USDA. 1031 Baltimore Blvd, Beltsville, MD.

<http://www.nal.usda.gov/awic/pubs/bulletin.shtml>

A quarterly publication providing "current information on animal welfare to investigators, technicians, administrators, exhibitors and the public."

Guide to the Care and Use of Experimental Animals, Volume 1 (2nd Edition). Olfert ED, Cross BM and McWilliam AA, eds. 1993. Canadian Council on Animal Care. Ottawa, Ontario.

http://www.ccac.ca/en/CCAC_Programs/Guidelines_Policies/GUIDES/ENGLISH/toc_v1.htm

A thorough manual with a clear discussion of relevant literature. Pertinent chapters: responsibility for the care and use of experimental animals; laboratory animal facilities; laboratory animal care; social and behavioral requirements of experimental animals; restraint and manipulations; standards for experimental animal surgery; control of animal pain in research, teaching and testing; anesthesia; euthanasia; use of animals in psychology; use of animals in neuroscience research; space requirements; categories of invasiveness in animal experiments; ethics of animal investigation.

In the Name of Science. Issues in Responsible Animal Experimentation. Orlans FB. 1993. Oxford University Press. New York, NY.

Comprehensive analysis of the social, political, and ethical conflicts surrounding the use of animals in scientific experiments. The author makes "recommendations for policy changes that are achievable within the foreseeable future and that would improve the lot of animals used for experimentation without hampering the scientific process... A reasonable way of looking at these issues is to ask the question. 'Can the harms be reduced?'"

Laboratory Animal Science. 1987. 37 (Special Issue).

"Case studies of ethical dilemmas." Orlans FB, 59-64.

Five cases based on actual situations are discussed in depth to illustrate opportunities for the IACUC to modify protocols to introduce more humane experimental design.

"Reducing pain in laboratory animals." Spinelli JS, 65-70.

Terminology of pain is summarized; simple but effective strategies for the control of pain are proposed.

"Assessment of animal pain in experimental animals." Soma LR, 71-77.

The signs and behavioral changes associated with acute and chronic pain in animals are clearly described.

Dr. Soma states, "When there is doubt, the bias should be in favor of the animal."

"Public concerns for animals in research." Clark J, 120-121.

The author highlights the fundamental dilemma of regulated animal welfare: "If we want to protect laboratory animals from neglect or abuse, we must insist on strong laws that can be enforced."

The Monkey Wars. Blum D. 1994. Oxford University Press. New York, NY.

A realistic picture of the scientific and ethical dilemmas that accrue from biomedical and psychological experimentation with animals, in particular with nonhuman primates. Succinct discussions include the standpoints of extremists and moderates and are based primarily on interviews with leading primatologists and animal advocates across the US.

INSTITUTIONAL ANIMAL CARE AND USE COMMITTEES (IACUCs)

“Such members shall possess sufficient ability to assess animal care, treatment, and practices in experimental research as determined by the needs of the research facility and shall represent society's concerns regarding the welfare of animal subjects used at such facility.” AWA Section 13 (b)(1).

“Appointing animal protectionists to Institutional Animal Care and Use Committees.” Levin LA and Stephens ML. 1994/95. *Animal Welfare Information Center Newsletter* 5(4), 1-2 & 8-9.

<http://www.nal.usda.gov/awic/newsletters/v5n4/5n4steph.htm>

Authors “propose that animal research facilities, as a show of good-will, voluntarily appoint individuals to their IACUC who are recognized in the local community as advocates for animals.” The advantages and potential pitfalls are discussed.

“The attending veterinarian as an ally and leader of the IACUC?” Silverman J. 2000. *Lab Animal* 29(10), 26-27.

The role of the attending veterinarian (AV) is clearly delineated as a moral and scientific leader of the IACUC who must take an active part in the committee’s work. “The veterinarian can take an assertive yet non-confrontational role in helping the IACUC ensure the proper care and use of research animals” in the spirit of the 3 Rs, “preferring not to use animals if efficacious alternatives are available.” The AV needs job security, i.e. tenure, since “fear of retribution can negatively affect the AV’s performance on the IACUC.”

“Best practices for animal care committees and animal use oversight.” DeHaven WR. 2002. [*Institute for Laboratory Animal Research*] *Journal* 43 (Supplement).

http://dels.nas.edu/ilar_n/ilarjournal/43_sup/v43supDehaven.shtml

There are many “best practices” that can help the animal care committees (ACC) promote institutional compliance and good animal welfare. These practices, although not universally appropriate for all institutions or activities, are examined.

“A current perspective on the role and needs of IACUC unaffiliated members.” Mondschein SG 2007. *Lab Animal* 36(6), 21-26.

“The unaffiliated member, whose role on the Committee is to represent the general public, is often a non-scientist with little or no previous exposure to the concepts described in the animal-use protocols he or she is charged with reviewing. The author, himself an unaffiliated IACUC member, provides advice and suggestions.”

“Community members on animal review committees.” Orlans FB. 1993. In *In the Name of Science. Issues in Responsible Animal Experimentation.* 99-117. Oxford University Press. New York, NY.

To be effective, community members “need to be able to withstand role ambiguity and to deal with group pressures. They need ... an ability to present a reasoned view with dignity and without hostility to persons who do not agree with them. They must be satisfied with having only moderate or minor impact on the committee and seeing only occasional disapprovals of protocols. Their overall impact of contributing balance and some measure of public accountability to the proceedings must suffice. For this, they must be prepared to devote a considerable amount of their time.”

“Contemporary Topics for Animal Care Committees.” 2007. *ILAR [Institute for Laboratory Animal Research] Journal* 48 (1) Whole issue. http://dels.nas.edu/ilar_n/ilarjournal/48_1/html/

Recent studies, new approaches, and ethical challenges in animal research are presented.

"Defining the animal care and use program." Sandgren EP. 2005. *Lab Animal* 34(10), 41-44.

"An effective Animal Care and Use program is critical to an institution's ability to ensure that animal research is conducted humanely and follows all applicable regulations and guidelines. The author provides a global view of the key programmatic components, which can be used to improve existing programs or implement new programs."

"Do pressure and prejudice influence the IACUC?" Silverman J. 1997. *Lab Animal* 26(5), 23-25.

"I believe that the disparity of IACUCs being less rigorous in their deliberations when approving a given number of mice versus the same number of dogs is the more significant dilemma" [than the influence of the perceived power of an investigator]. "IACUCs and laboratory animal specialists must overcome any of our own prejudices and take the lead in speaking out on behalf of animals – all animals."

"Engaging the IACUC through comprehensive training." Haywood JR, Greene M, James ML and Bayne, K. 2005. *Lab Animal* 34(10), 33-37.

"The IACUC is one of the most important committees at a research institution and plays a critical role in the success of an animal care and use program. It is the responsibility of the institution to provide IACUC members with adequate and appropriate training. The authors explore various IACUC training options."

"Ethics of animal welfare in research: The institution's attempt to achieve appropriate social balance." Prentice ED, Zucker IH and Jameton A. 1986. *The Physiologist* 29(2), 17 & 19-21.

http://www.unmc.edu/dept/iacuc/index.cfm?L1_ID=4&CONREF=26

Paper describes 14 ethical principles governing research involving animals adopted by the University of Nebraska Medical Center. These clearly stated principles serve as the protocol review criteria employed by the IACUC.

The IACUC Handbook. Silverman J, Suckow MA and Murthy S, eds. 2nd ed. 2006. CRC Press. Boca Raton, FL.

This practical guide for IACUC members uses a question-and-answer format to address the problems and concerns often confronting IACUCs. The book's chapters not only discuss the structure and responsibilities of the IACUC, they also include such issues as pain and distress, euthanasia, surgery, occupational health and safety, laboratory animal enrichment, and animal mistreatment and protocol noncompliance. The second edition features comprehensive updates for all pertinent federal laws, regulations, and policies and also contains an expanded survey of IACUC practices from institutions around the nation.

Information Resources for Institutional Animal Care and Use Committees 1985-1999. Allen T, ed., revised 2000. AWIC Resource Series No. 7. U.S. Department of Agriculture, National Agricultural Library, Animal Welfare Information Center. Beltsville, MD.

<http://www.nal.usda.gov/awic/pubs/IACUC/iacuc.htm>

Extensive resource. "This publication is divided into 10 sections: Introduction to Animal Care and Use Committees; U.S. Government Principles, regulations, policies and guidelines; agency directives for federal fund-holders; professional guidelines; world wide web resources; articles and bibliographies; primary references; selected software providers; organizations; and an appendix."

Institutional Animal Care and Use Committee Guidebook. Applied Research Ethics National Association (ARENA); Office of Laboratory Animal Welfare (OLAW). 2002. 2nd edition. National Institutes of Health. Bethesda, MD.

<ftp://ftp.grants.nih.gov/IACUC/GuideBook.pdf>

Information about authority, composition and functions of IACUCs, issues, criteria, oversight of animal care and use program, evaluation of animal welfare concerns, record keeping and reporting, and special considerations such as alternatives to the use of live animals, instructional use of animals, farm animals, and legal concerns. Subject areas covered in the second edition, but not the first, include the following: IACUC

operation and administration; training for IACUC members; oversight of the animal care and use program; behavioral management; emergency preparedness; breeding colonies and transgenic animals.

Institutional Animal Care and Use Committees: A Comprehensive Resource of Online Information. Web site. Duffee N, Barnett L, Cody C and Silver C. 2008*. AALAS. Memphis, TN.

<http://www.iacuc.org/>

Useful links archive to resources for IACUCs, organized by area of interest.

Public Health Service Policy on Humane Care and Use of Laboratory Animals. Office of Laboratory Animal Welfare (OLAW). 1985. Rev. 1986, amended 2002.

<http://grants.nih.gov/grants/olaw/references/phspol.htm>

Functions of IACUCs as defined by the Public Health Service policy are clearly outlined. Useful user-friendly tutorial on the PHS policy is offered at: <http://grants.nih.gov/grants/olaw/tutorial/index.htm>

Public Health Service Policy on Humane Care and Use of Laboratory Animals: Frequently Asked Questions. OLAW. Sept 2006.

<http://grants.nih.gov/grants/olaw/faqs.htm>

The purpose of the new page is to provide up-to-date guidance for institutions and IACUCs to use in implementing the Public Health Service Policy on Humane Care and Use of Laboratory Animals (PHS Policy). Many of the answers refer to guidance previously published as articles in journals and magazines. The new FAQs provide guidance on the Freedom of Information Act, post-approval monitoring, HVAC malfunctions and failures, rodent cage density, and other issues not previously addressed by OLAW.

"Reliability of protocol reviews for animal research." Plous S and Herzog H. 2001. *Science* 293 (July), 608-609.

<http://www.socialpsychology.org/articles/scipress.htm>

"A random sample of 50 Institutional Animal Care and Use Committees participated in a study of the protocol review process. Each committee submitted three animal behavior protocols it had recently reviewed, and these protocols were reviewed a second time by another participating committee. The result showed that approval decisions were statistically unrelated. In most cases, proposals that were disapproved by one committee were approved by the second committee. "All told, 61% of [150] protocols were judged as either "not very understandable" or "not understandable at all," as having "poor" research designs and procedures, or as justifying the type and number of animals in a way that was deemed "not very convincing" or "not convincing at all."

"Should IACUCs review scientific merit of animal research projects?" Mann MD and Prentice ED. 2004. *Lab Animal* 33(1), 26-31.

Whether IACUCs should review animal research protocols for scientific merit is not addressed in the federal regulations, resulting in ongoing confusion on the subject. The authors examine this issue, discuss the pros and cons, suggest how IACUCs can go about reviewing protocols for scientific merit, and question what effect recent changes in regulations will have on this issue. "Lawmakers and regulatory agencies expect the IACUC to serve as a 'gatekeeper' that ultimately ensures that research involving animals is justified and humanely conducted."

"A study of three IACUCs and their views of scientific merit and alternatives." Graham K. 2002. *Journal of Applied Animal Welfare Science* 5(1), 75-81.

Three IACUCs were evaluated using a 19-question survey. "Although 76% of members answered that scientific merit should be more diligently assessed if more than slight pain is caused, 14% believed that assessing scientific merit is not the role of the IACUC. Nearly 86% agreed that the search for alternatives should be more diligent for protocols that incur more than slight pain to the animals involved. Some members believed that alternatives were not actively enough sought after, while others believed no viable alternatives existed."

“Toward better unaffiliated members: Goal of two unaffiliated members per IACUC offers advantages.” Liss C. 2000. *Science and Animal Care* 11(1), 1-4.

“Outside members on IACUCs have a very tough job. These lone representatives, unaffiliated with the research institutions and unpaid for their services, carry the responsibility of representing the community’s concerns for the welfare of the animals used for experimentation, teaching and testing... Bringing in a second unaffiliated member (UM) on the IACUC would relieve some of the pressure. If one of the UMs is unable to attend a meeting, the other will be there thus ensuring that an outside member is always present for committee activities... In addition, each of the UMs should bring an unique perspective to the meetings.”

What Investigators Need to Know About the Use of Animals. OLAW. 2006.

<http://grants.nih.gov/grants/olaw/InvestigatorsNeed2Know.pdf>

This brochure educates investigators about their responsibilities under PHS Grants Policy and PHS *Policy*. It describes the expectations and requirements when using animals in research supported by the PHS.

“What’s wrong with the IACUC?” Opinion. 2000. *Lab Animal* 29(10), 28-29.

“IACUCs need an IACUC Chair and members who are not concerned about promotion and tenure issues being compromised by their IACUC role...IACUC members told me they would never take serious action against other faculty members because it would be taken against them during promotion and tenure.” Diane McClure.

LEGISLATION, REGULATIONS AND GUIDE

Animal Welfare Act as Amended. (7 USC, 2131-2156) Federal law.

<http://www.aphis.usda.gov/lpa/pubs/awact.html>

Chief federal animal protective law, adopted in 1966 and amended in 1970, 1976, 1985, 1991 and **2002**; sets minimum standards for the care and housing of animals used in research, exhibition and the wholesale pet trade; mandates principal investigators to consult with a veterinarian and to consider alternatives *before* conducting any procedure likely to produce pain or distress in an experimental animal; requires semi-annual inspections by the IACUC and at least one inspection per year by USDA; places the authority and control of animal usage with the IACUC. Contains the 2002 Farm Bill amendments to the Animal Welfare Act.

Code of Federal Regulations, Title 9, Chapter 1, Subchapter A - Animal Welfare. USDA. 2007.

http://www.access.gpo.gov/nara/cfr/waisidx_07/9cfrv1_07.html

Regulations developed by the USDA that specify how to comply with the Animal Welfare Act and its amendments, divided into 4 sections: definitions, regulations, standards and rules of practice. The bulk of the subchapter is the third section that provides **standards** for specific species or groups of species such as cats and dogs, guinea pigs and hamsters, rabbits, nonhuman primates, marine mammals, and the general category of "other warm-blooded animals." Standards include those for facilities and operations, health and husbandry systems, and transportation.

Animal and Plant Health Inspection Animal Care Policy Manual. USDA. 1999-2000.

http://www.aphis.usda.gov/animal_welfare/policy.shtml

“The Animal Care Policy Manual further clarifies the intent of the Animal Welfare Act.” Policy and enforcement guidelines that determine many of the actions that IACUCs must take as they inspect facilities and review protocols.

The manual includes:

“USDA’s AWA Policy #11 -- Policy about painful/distressful procedures” -- April 14, 1997.

“A painful procedure is defined as any procedure that would reasonably be expected to cause more than slight or momentary pain and/or distress in a human being to which that procedure is applied. The Institutional Animal Care and Use Committee (IACUC) is responsible for ensuring that investigators have

appropriately considered alternatives to any procedures that may cause more than slight or momentary pain or distress.” Examples of such procedures are given.

“USDA’s AWA Policy #12 -- Consideration of alternatives to painful/distressful procedures” – June 21, 2000.

“The Animal Welfare Act (AWA) regulations require principal investigators to consider alternatives to procedures that may cause more than momentary or slight pain or distress to the animals and **provide a written narrative** of the methods used and sources consulted to determine the availability of alternatives, including refinements, reductions, and replacements.” Gives guidance on the requirement to provide a written narrative, and a search for alternatives.

Guide for the Care and Use of Laboratory Animals. Institute of Laboratory Animal Resources, National Research Council. 1996. National Academy Press. Washington, DC

<http://www.nap.edu/books/0309053773/html/>

Updated basic reference on housing, handling and care of animals in scientific institutions and government agencies. Includes ***US Government Principles for the Utilization and Care of Vertebrate Animals Used in Testing, Research, and Training.*** <http://darwin.nap.edu/books/0309053773/html/116.html>

“Unless the contrary is established, investigators should consider that procedures that cause pain or distress in human beings may cause pain or distress in other animals.”

RELATED DOCUMENTS AND ARTICLES:

Animal Welfare Act 1966-1996: Historical Perspectives and Future Directions. Kreger M, Jensen D’A, and Allen T, eds. 1998. Proceedings of a symposium sponsored by the USDA, held on September 12, 1996 in Riverdale, Maryland. WARDS. Vienna, VA.

<http://www.nal.usda.gov/awic/pubs/96symp/awasymp.htm>

“This symposium takes a retrospective look at the development and effectiveness of the Federal animal welfare regulations since 1966. Leaders from government, including those directly involved in writing the regulations of the original act, industry, and humane groups offer their views of the history and impact of the act and their visions for its future.”

“Animal Welfare Act - Requirements for the minimization of pain and distress.” DeHaven WR. 1998. ***Pain Management and Humane Endpoints.*** Johns Hopkins Center for Alternatives to Animal Testing workshop.

<http://altweb.jhsph.edu/meetings/pain/dehaven.htm>

Clear explanation of policy No. 11 and 12 and the role of the IACUCs in implementing them.

ETHICS

“Ethics in our Western world has hitherto been largely limited to the relations of man to man. But that is a limited ethics. We need a boundless ethics which will include the animals also.” Albert Schweitzer

Animals, Nature and Albert Schweitzer. Free AC, ed. 1982. Animal Welfare Institute. Washington, DC. Available from Animal Welfare Institute, PO Box 3650. Washington, DC 20007

<http://www.awionline.org/schweitzer/as-idx.htm>

A brief outline of Albert Schweitzer's ethic of Reverence for Life. "Whenever an animal is somehow forced into the service of men, every one of us must be concerned for any suffering it bears on that account. No one of us may permit any preventable pain to be inflicted, even though the responsibility for the pain is not ours."

“Beyond ‘adequate veterinary care’.” Anchel M. 1976. *Journal of the American Veterinary Medical Association* 168, 513-517.

A very clear definition of the laboratory animal veterinarian's professional and ethical obligations. "The veterinarian must have the courage – and it will require courage – to insist on standards that are absolute, and not relative to the pressures within any institution."

Cost of Caring: Recognizing Human Emotions in the Care of Laboratory Animals. AALAS. 2001. American Association For Laboratory Animal Science, Memphis, TN.

<http://www.aalas.org/pdf/06-00006.pdf>

"The human-animal bond in the field of animal research exists in many forms. Kindness and concern for animals are desirable characteristics in animal care and research workers." Acknowledging feelings of grief or bereavement at the death of animals used for research or teaching and providing support in the workplace are important. "The bond between people and animals in the laboratory can minimize certain variables related to stress in the animals. The research community can reap the benefits of these essential relationships."

A Critical Look at Animal Experimentation. Kaufman, SR, Cohen, MJ, Cramer M, Contard PC, Hahner K and Todd B. 1995. Medical Research Modernization Committee, New York, NY.

"The value of animal experimentation has been grossly exaggerated by those with a vested economic interest in its preservation. Because animal experimentation focuses on artificially created pathology, involves confounding variables, and is undermined by differences in human and nonhuman anatomy, physiology, and pathology, it is an inherently unsound way to investigate human disease processes. Billions of dollars invested annually in animal research would be put to much more efficient, humane, and effective use if redirected to clinical and epidemiological research and public health programs."

"Editorial: Caring for animals, caring for ourselves." Spaeth GL. 1994. *Ophthalmic Surgery* 25, 426.

"When we act uncaringly toward experimental animals we become uncaring human beings. What is the worth of medical miracle achieved at the cost of inflicting trauma on others that cannot help but scar our own characters?"

"Ethical aspects of relationships between humans and research animals." Herzog H. 2002. *ILAR [Institute for Laboratory Animal Research] Journal* 43(1), 27-32.

http://dels.nas.edu/ilar_n/ilarjournal/43_1/Ethical.shtml

"Ways that research institutions can help individuals cope with the ethical consequences of relationships with research animals include the following: supporting the development of human-animal relationships in laboratories, giving animal care personnel an ethical voice through involvement in the institutional animal care and use committee decision process, publicly acknowledging the emotional and moral costs of human-laboratory animal relationships, and educating animal care staff about the purpose and possible benefits of research projects."

"Ethical decisions concerning animal biotechnology: What is the role of animal welfare science?" Olsson IAS and Sandoe P. 2004. *Science in the Service of Animal Welfare.* Kirkwood JK, Roberts EA and Vickery S, eds. Proceedings of the UFAW International Symposium, Edinburgh, 2003. *Animal Welfare* 13: S139-144.

The different ethical concerns expressed by ethicists and by the general public about animal biotechnology are summarized. Focusing on one of them, animal welfare, an introduction to the animal welfare implications of recent developments in reproductive and gene technologies is given. The importance of animal welfare aspects is discussed in relation to other ethical concerns about animal biotechnology.

"Ethical guidelines for investigations of experimental pain in conscious animals." Zimmermann M. 1983. *Pain* 16, 109-110.

Guidelines of the International Association for the Study of Pain. "It is essential that intended experiments on pain in conscious animals be reviewed beforehand by scientists and lay-persons." Investigators "should accept a general attitude in which the animal is regarded not as an object for exploitation, but as a living individual."

"The ethical socialization of animal researchers." Arluke A. 1994. *Lab Animal* 23(6), 30-32, 34-35.

"Newcomers face a closed moral universe where issues of morality are defined institutionally, and hence are rarely confronted openly by individuals. Anti-ethical training processes support ideological claims for the importance of knowledge production, the need for objectivity and professionalism, and the priority of the concerns of humans over those of animals."

"Ethics and welfare of animals used in education: an overview." King LA. 2004. *Science in the Service of Animal Welfare*. Kirkwood JK, Roberts EA and Vickery S, eds. Proceedings of the UFAW International Symposium, Edinburgh, 2-4 April 2003. *Animal Welfare* 13: S221-227.

Ethical, regulatory and scientific issues arise from the use of animals in education. The implementation of alternatives to animal use is inconsistent, and barriers to the adoption of alternatives include specific curriculum and legislative requirements, traditional educational methodology, and resource and training limitations, particularly when the alternative methods involve new technologies.

The Ethics of Animal Investigation. 1989. Canadian Council on Animal Care. Ottawa, Ontario.

http://www.ccac.ca/en/CCAC_Programs/Guidelines_Policies/POLICIES/ETHICS.HTM

"Animals should be used only if the researcher's best efforts to find an alternative have failed. A continuing sharing of knowledge, review of the literature, and adherence to the Russell-Burch "3R" tenet of "Replacement, Reduction and Refinement" are also requisites. Those using animals should employ the most humane methods on the smallest number of appropriate animals required to obtain valid information."

The Ethics of Research Involving Animals. Nuffield Council on Bioethics. 2005. London, UK.

http://www.nuffieldbioethics.org/go/ourwork/animalresearch/publication_178.html

"This Report seeks to clarify the debate and aims to help people think through the ethical issues that are raised. The ways in which animals are used in different areas of research are reviewed, including: basic or 'blue sky' research, the development of new medicines and vaccines, and toxicity testing. The Report makes practical recommendations for future policy and practice, relating, among other things, to the use of GM animals, ways of improving the quality of debate, the implementation of the Three Rs (Refinement, Reduction and Replacement), and the responsibilities of researchers, reviewers and funding bodies."

The Human Use of Animals: Case Studies in Ethical Choice. Orlans FB, Beauchamp TL, Dresser R, Morton DB and Gluck JP. 1998. Oxford University Press. New York.

This easy-to-read book includes an introductory chapter on morality providing a broad background information and describing several methods useful to resolve ethical dilemmas. Subsequent chapters are individual case studies covering most major areas of animal use, and discussing the ethical issues and welfare concerns involved. This book helps readers reflect on their own ethical outlook.

"Implications of human-animal interactions and bonds in the laboratory." 2002. *ILAR [Institute for Laboratory Animal Research] Journal* 43(1) [whole issue].

http://dels.nas.edu/ilar_n/ilarjournal/43_1/

Number of articles addressing the ethical implications of the human-animal bond in research laboratories, and its impact on caregivers and on animal well-being.

"Sacrificial symbolism in animal experimentation: Object or pet." Arluke AB. 1988. *Anthrozoos* 2, 98-117.

"Many principal investigators do not handle animals at all, although a few may do surgery after the animal has been prepared fully by a technician... Laboratories should adopt a moral attitude that sanctions the expression of emotions and condones the human side of scientific work."

"Trapped in a guilt cage." Arluke A. 1993. *Animal Welfare Information Center Newsletter* 4(2).

<http://www.nal.usda.gov/awic/newsletters/4n2.htm>

The author studied laboratories and research centers to investigate the impact of experiments on the people who carry them out. "Episodic feelings of discomfort were common and were expressed as background uneasiness and guilt... Open discussion of these feelings was taboo. Scientists, veterinarians, and administrators tended to deny that laboratory workers could be troubled by their use of animals. Uneasiness was not seen as an issue, and was not allowed to intrude on the normal course of work.... Yet within the laboratory culture were unspoken rules and resources for dealing with unwanted emotions and thoughts, despite the silence surrounding this topic." The surfacing of conflicts that prompt defensive behavior among researchers may be due to the "diffusion into the laboratory of society's heightened awareness of how animals should be viewed and treated. Coping devices will be called out when humanity's standards clash with traditional scientific practice. This is cheering to some who see this as a willingness to pay more attention to humanitarian ideals in animal experimentation."

“Understanding the emotional experiences of animal research personnel.” Halpern-Lewsi JG. 1996. *Contemporary Topics* 35(6), 58-60.

Research personnel “who demonstrate caring and compassionate behaviors add to the integrity of the animals, which ultimately results in higher quality research protocols. Individuals interacting with animals ... should be encouraged to engage in caring behaviors without fear of reprisal.” Suggestions are provided to help research participants to do their work without compromising humane relationships with experimental animals.

The Use of Animals in Higher Education : Problems, Alternatives, & Recommendations. Balcombe J. 2000. Humane Society Press. Washington, DC

“The aim of this monograph is to present a comprehensive examination of the issue of animal use in education from an ethical and humane perspective.”

ALTERNATIVES: GENERAL

“Alternatives or alternative methods *are generally regarded as those that incorporate some aspect of replacement, reduction, or refinement of animal use in pursuit of the minimization of animal pain and distress consistent with the goals of the research. These include methods that use non-animal systems or less sentient animal species to partially or fully replace animals (for example, the use of an *in vitro* or insect model to replace a mammalian model), methods that reduce the number of animals to the minimum required to obtain scientifically valid data, and methods that refine animal use by lessening or eliminating pain or distress and, thereby, enhancing animal well-being.*” USDA AWA’s policy #12.

Alternatives to Animal Testing: Refinement, Reduction, Replacement (ALTWEB Web Site). 2008*. <http://altweb.jhsph.edu/>

Up-to-date, comprehensive site on alternatives containing a number of full-text documents, abstracts of journals on alternatives (<http://altweb.jhsph.edu/publications/publications.htm>), a search engine, Frequently-Asked-Questions on alternatives, and further links.

Alternatives to Laboratory Animals (ATLA). FRAME (Fund for the Replacement of Animal in Medical Experiments.)

<http://altweb.jhsph.edu/publications/journals/atla/atla-index.htm> (abstracts available at this site)

This quarterly journal covers “all aspects of the development, validation, introduction and use of alternatives to laboratory animals in biomedical research and toxicology testing.”

Alternatives to Pain in Experiments on Animals. Pratt DP. 1980. Argus Archives. New York. http://www.awionline.org/lab_animals/pratt/prat-idx.htm

A well written account of ethically and scientifically unacceptable practices in animal experimentation. The author describes specific experiments and matches them with alternatives.

Alternatives Page of The Animal Welfare Information Center's Web Site (AWIC). 2007*.

http://awic.nal.usda.gov/nal_display/index.php?info_center=3&tax_level=1&tax_subject=183

This site contains online articles; a list of databases and organizations; help with the literature search in the form of guidelines, a thesaurus of alternatives terminology, worksheets and an AWIC alternatives workshop on the web. A number of bibliographies are available from AWIC's publications page relating to animal care, use and welfare; ethical and moral issues; and to IACUCs.

The Interagency Coordinating Committee on the Validation of Alternative Methods (ICCVAM).

<http://iccvam.niehs.nih.gov/>

This agency seeks to promote the validation and regulatory acceptance of methods that will refine, reduce, and/or replace animal use.

Fund for the Replacement of Animals in Medical Experiments: Information Resources (FRAME). 2006*.

http://www.frame.org.uk/page.php?pg_id=9

This excellent site offers a clear introduction to the three Rs, Replacement, Reduction, Refinement. Annotated links to databases, resources and organizations are provided as are very helpful guidelines in searching for alternatives.

Laboratory Animals — The Three Rs: developments in laboratory animal science. 1994. Laboratory Animals Ltd. London, England. Contains three reprints from *Laboratory Animals*, 28, 1994.

“Replacement of animal procedures: alternatives in research, education and testing.” Balls M, 193-211.

<http://www.lal.org.uk/pdf/balls2.pdf>

“Reduction of animal use: experimental design and quality of experiments.” Festing MFW, 212-221.

<http://www.lal.org.uk/pdf/festing.pdf>

“Refinement of animal use—assessment and alleviation of pain and distress.” Flecknell PA, 222-231.

<http://www.lal.org.uk/pdf/fleck.pdf>

National Centre for the Replacement, Refinement and Reduction of Animals in Research (NC3Rs) Information Portal

<http://www.nc3rs.org.uk/landing.asp?id=38>

The National Centre for the Replacement, Refinement and Reduction of Animals in Research provides a UK focus for the promotion, development and implementation of the 3Rs in animal research and testing. The Information Portal contains annotated links to online databases, websites, journal articles, legislation and other publications. These resources provide information to help apply the 3Rs and ensure the best possible standards in animal welfare.

The Principles of Humane Experimental Technique. Russell WMS and Burch RL. 1959. Methuen & Co. London, UK

http://altweb.jhsph.edu/publications/humane_exp/het-toc.htm

The authors introduce the concept of the three Rs: "Desirable as replacement is, it would be a mistake to put all our humanitarian eggs in this basket alone. The progress of replacement is gradual, nor is it ever likely to absorb the whole of experimental biology. Refinement may reach such a pitch that a given procedure employing animals is absolutely humane, but in any given field there is bound to be a latent period before such success is attained. Reduction remains of great importance, and of all modes of progress it is the one most obviously, immediately, and universally advantageous in terms of efficiency."

Progress in the Reduction, Refinement and Replacement of Animal Experimentation. Ball M, van Zeller AM and Halder ME, eds. 2000. *Developments in Animal and Veterinary Science*. Vol. 31A and 31B. Proceedings of the Third World Congress on Alternatives and Animal Use in the Life Sciences, Bologna, Italy, 29 Aug. to 2 Sept 1999. Elsevier. Amsterdam, Netherlands.

Excellent contributions reflect the present state of knowledge in many areas including adverse effects; animal models; biologicals; barrier systems *in vitro*; carcinogenicity testing; ethical committees; ethical aspects of transgenesis; humane endpoints; outreach on the three Rs to scientists and the general public; experimental design, data analysis and reduction; refinements in animal housing and husbandry; refinement in experimental design and techniques; skin and eye irritation testing; toxicogenomics; use of animals in education and training; and use of reconstituted tissues and co-cultures.

“Refinement, reduction, and replacement of animal use for regulatory testing: Current best scientific practices; future improvements and implementation within the regulatory environment; recommendations for implementation of best scientific practices.” 2002. In: Regulatory Testing and Animal Welfare. Proceedings of an International Symposium. International Council for Laboratory Animal Science & Canadian Council on Animal Care; Quebec City, Canada, June 21-23, 2001. *ILAR [Institute for Laboratory Animal Research] Journal* 43 Supplement.

http://dels.nas.edu/ilar_n/ilarjournal/43_supp/

The main objective of the symposium was to identify best practices to minimize or eliminate pain and distress for animals used in safety evaluation and testing procedures. The presentations described the current best practices for using animals in regulatory testing procedures. Widespread implementation of these best practices could improve the welfare of animals used for safety testing and contribute to reduced animal use.

The Three Rs at the Beginning of the 21st Century: Proceedings of the Fourth World Congress on Alternatives and Animal Use in the Life Sciences, 11-15 August 2002, New Orleans, LA, USA. Balls M, Firmani D and Rowan A, eds. 2004. *ATLA (Alternatives to Laboratory Animals)* 32, Suppl. 1 & 2.

<http://www.worldcongress.net/2002/proceedings/proceedings.htm>

This new volume reflects the progress that has been made in implementing the concept of the Three Rs of reduction, refinement and replacement in animal research, education and testing. Papers on the following topics were presented: Replacement and reduction alternatives; refinement and reduction alternatives; policy and ethics; education and information resources; test development, validation and implementation.

"The Three Rs: past, present and future." Russell WMS. 2005. *Animal Welfare* 14(4), 279-286.

"We originally envisaged refinement as minimising pain and distress, and by 1959, discomfort. It is now clear that we must aim positively at optimal well-being."

University of California Center for Animal Alternatives

<http://lib.ucdavis.edu/dept/animalalternatives/index.php>

Information and bibliographies on animal welfare and alternatives.

ALTERNATIVES: REPLACEMENT/IN VITRO TESTING

"We shall use the term 'replacement technique' for any scientific method employing non-sentient material which may, in the history of animal experimentation, replace methods which use conscious living vertebrates." Russell W and Burch R

Alternative Toxicological Methods. Salem H and Katz S. 2003. CRC Press. Boca Raton.

The book explores the development and validation of replacement, reduction, and refinement alternatives (the 3 Rs) to animal testing. Contributions present what has been accomplished thus far in developing acceptable alternatives to traditional animal toxicological assessment and provide potentially new initiatives.

"Evaluation of some in vitro tests to reduce and replace the sub-acute animal toxicity studies."

Chhabra RS, Ress NB, Harbell JW and Curren RD. 2004. *The Three Rs at the Beginning of the 21st Century:* Proceedings of the Fourth World Congress on Alternatives and Animal Use in the Life Sciences. Balls M, Firmani D and Rowan A, eds, 43-49. *ATLA* 32, Supplement 1 & 2.

<http://www.worldcongress.net/2002/proceedings/WA5 Chhabra 23-8-03.pdf>

"At present, in addition to refining the current testing protocols, the NTP is evaluating the potential for in vitro test methods to partially or completely avoid the need for 14-day toxicity studies, especially for chemicals where the dermal route of exposure is used. The in vitro assays used were the EpiDerm™ bioassay to estimate dermal irritation, the neutral red uptake (NRU) bioassay to estimate systemic toxicity and the primary rat hepatocyte cytotoxicity (PRHC) assay to estimate hepatotoxicity."

"In vitro and other non-animal experiments in the biomedical sciences." Blaauboer BJ. 1996. *Australian and New Zealand Council for the Care of Animals in Research and Teaching (ANZCCART) News* 9(4), insert: 1-4. An ANZCCART News Fact Sheet.
http://www.adelaide.edu.au/ANZCCART/publications/In%20Vitro_4.pdf

"Developments in cellular and molecular biology, as well as computerised modelling, provide ample opportunities for change and the incorporation of knowledge of the mechanisms of toxicity. New procedures must be validated in order to assess their reliability and relevance. Validation should have a sound scientific basis and should also be practical. In this validation process an important driving force should be the improvement of the relevance of toxicological risk assessment."

In Vitro Methods for Assessing Acute Systemic Toxicity. ICCVAM and NICEATM. 2007.
<http://iccvam.niehs.nih.gov/methods/acute/tox/acute/tox.htm>

The workshop evaluated the status of available *in vitro* methods for assessing acute toxicity. It also developed recommendations for validation efforts.

Institute for In Vitro Sciences. (IIVS website) 2008*
<http://www.iivs.org/>

The Institute (1) provides non-animal research and testing services, (2) sponsors workshops and training courses in in vitro methods, and (3) creates a forum where Industry, Government and Animal Welfare proponents can meet to determine constructive programs which effectively reduce animal use.

Selection and Use of Replacement Methods in Animal Experimentation. UFAW, FRAME. 1998.
Available from UFAW, The Old School, Brewhouse Hill, Wheathampstead, Herts AL4 8AN UK.

The booklet is "a practical guide to help ensure that those considering animal experimentation have explored all opportunities to avoid animal use and attempted to minimize the numbers involved." Detailed overview of replacement options, including a summary of the current uses, advantages and limitations for each method.

"The use of human cells in biomedical research and testing." Combes R. 2004. *ATLA* 32, Suppl. 1, 43-49.
http://www.worldcongress.net/2002/proceedings/A1_Combes_23-8-03.pdf

"It is only comparatively recently that the safe and controlled acquisition of surgical waste and non-transplantable human tissues has become feasible with the establishment of several human tissue banks. Recently, the formation of a UK and European centralised network for human tissue supply has been initiated. The problems of short longevity and loss of specialisation in culture are being approached."

Specific Topics in *In Vitro* Testing: see table 1

See also: Specialized *In Vitro* Replacement Resources p. 28

ALTERNATIVES: REFINEMENT/REDUCTION

"Techniques that improve upon the accuracy of measurements and reduce or eliminate sources of variability have the potential for reducing the number of animals required in a given protocol." Brockway B, Hassler C and Hicks N

"The standards ... shall, with respect to animals in research facilities, include requirements--(A) for animal care, treatment, and practices in experimental procedures to ensure that animal pain and distress are minimized, including adequate veterinary care with the appropriate use of anesthetic, analgesic or tranquilizing drugs, or euthanasia." AWA Section 13 (a)(3).

General

"Advancing refinement of laboratory animal use." Smaje LH. 1998. *Laboratory Animals* 32 (2), 137-142.

Various aspects of refinement are described and a series of practical recommendations for advancing refinement of laboratory animal use is given.

Altweb special section on Refinement. 2008

<http://altweb.jhsph.edu/refinement/index.htm>

"This refinement section begins with introductory text explaining the topic in non-technical language, accompanied by a set of links to relevant databases, web sites, books, articles, abstracts, and so on. The section addresses the following topics: What is pain and distress? Definitions, biology and physiology; recognition and assessment of pain and distress; alleviation and prevention of pain in animals; humane endpoints; euthanasia; enrichment ."

"Animal definition: a necessity for the validity of animal experiments?" Öbrink KJ and Rehbinder C. 1999. *Laboratory Animals* 22, 121-130.

"In most scientific journals, experimental animals are described poorly.... The animal definition should not only include species, sex and age but also ... the environmental conditions to which the animals are exposed... The prerequisites for the use of fewer animals per project, while still retaining a sufficiently high degree of accuracy are high levels of *reproducibility* and *precision* in the experimental results. Factors that may affect these will be discussed in this paper. If a researcher, through carelessness or ignorance, should use more animals for a project than is necessary, it must be considered unethical."

"Ethology recommendations for a standardized minimum description of animal treatment." Davis DE, Bennett CL, Berkson G, Lang CM, Snyder RL and Pick JR. ILAR Committee on Laboratory Animals. 1973. *ILAR [Institute for Laboratory Animal Research] News/Journal* 16(4), 3-4.

"It is clear [from this survey] that many investigators do not realize the influence of ... environmental variables [e.g., housing, handling, temperature, light] on experimental results or at least do not adequately describe the environmental history of the animals used for experimentation."

"Importance of non-statistical design in refining animal experiments." Morton D. 1998. *Australian and New Zealand Council for the Care of Animals in Research and Teaching (ANZCCART) News* 11(2), insert: 1-12. An ANZCCART News Fact Sheet.

<http://www.adelaide.edu.au/ANZCCART/publications/fs17.pdf>

Detailed discussion of approaches to refinement, including good animal care, use of anaesthetics and analgesics, humane end points; suggestions of strategies that can contribute markedly to minimising pain and suffering in an experiment and reduce animal numbers; extensive bibliography. Refinement can be defined as: "Those methods which avoid, alleviate or minimise the potential pain, distress or other adverse effects suffered by the animals involved, or which enhance animal wellbeing... If scientists wish to claim they practise humane science then they have to pay as much attention to avoiding unnecessary pain and distress to their animals as they do to their scientific objectives.... Factors leading to good animal welfare will usually also lead to reliable, accurate and economic science."

"The influences of standard laboratory cages on rodents and the validity of research data."

Sherwin CM. 2004. *Science in the Service of Animal Welfare*. Kirkwood JK, Roberts EA and Vickery S, eds. UFAW International Symposium, Edinburgh, 2-4 April 2003. *Animal Welfare* 13: S9-15.

This paper reviews various influences of standard laboratory cage design and husbandry. The studies mentioned show that the development and responses of animals from standard laboratory housing and husbandry conditions are often unrepresentative and idiosyncratic, indicating that data are likely to have reduced external validity. Standard laboratory housing may sometimes be associated both with reduced welfare and with reduced benefits gained from research.

"Minimizing stress during physiological monitoring." Brockway BP, Hassler CR and Hicks N. 1993. *Refinement and Reduction in Animal Testing*. Niemi SM and Willson JE, eds, 56-69. Proceedings of a conference held by the Scientists Center for Animal Welfare

Minimizing or eliminating extraneous stress factors from the experimental design is crucial to a true understanding of the compound, device or technique being evaluated for its effects. The elimination of sources of variability (stress, for example) may allow the use of fewer animals giving equally valid results."

"Refinement and reduction through the control of variation." Festing MFW. 2004. In: *The Three Rs at the Beginning of the 21st Century*: Proceedings of the Fourth World Congress on Alternatives and Animal Use in the Life Sciences, 11-15 August 2002, New Orleans, LA, USA. Balls M, Firmani D and Rowan A, eds. 2004. *ATLA* 32, Supplement 1, 259-263.

http://www.worldcongress.net/2002/proceedings/BW1_Festing.pdf

"The key to doing animal experiments efficiently, while using the minimum number of animals without loss of scientific information, lies in good control of random variation, and recognition and control of "fixed effect" variation, such as the sex or strain of the animals."

"Stereotypies and other abnormal repetitive behaviors: Potential impact on validity, reliability, and replicability of scientific outcomes." Garner JP. 2005. *ILAR Journal* 46(2).

http://dels.nas.edu/ilar_n/ilarjournal/46_2/html/v4602garner.shtml

"Captive environments may affect the following aspects of an experiment: validity, by introducing abnormal animals into experiments; reliability, by increasing interindividual variation through the introduction of such individuals; and replicability, by altering the number and type of such individuals between laboratories. Thus, far from increasing variability, enrichment may actually improve validity, reliability, and replicability by reducing the number of abnormal animals introduced into experiments. In this article, the specific example of abnormal repetitive behaviors (ARBs) is explored... ARBs in laboratory animals may compromise validity, reliability, and replicability, especially in behavioral experiments; and enrichments that prevent ARB may enhance validity, reliability, and replicability."

"Using Fewer Research Animals." Chamove AS. 2003. *Laboratory Primate Newsletter* 42(1), 1-2.

<http://www.brown.edu/Research/Primate/lpn42-1.html#chamove>

Sequential sampling and power analysis are more ethical alternatives to reduce animal numbers to the lowest number possible, especially when test procedures are aversive.

"Variables in animal based research: Part 2. Variability associated with experimental conditions and techniques." Reilly J 1998. Australian and New Zealand Council for the Care of Animals in Research and Teaching (ANZCCART) *News* 11(1), insert: 1-12. An ANZCCART News Fact Sheet.

http://www.adelaide.edu.au/ANZCCART/publications/fs_variables_p2.pdf

Addresses the issues of variability associated with experimental conditions and techniques and includes the effects of stress, sampling techniques, anaesthesia and euthanasia, and how these may affect research data.... Refinement of research techniques using animals will lead to less animal distress and at the same time will usually lead to higher quality and more robust data."

Variables, Refinement and Environmental Enrichment for Rodents and Rabbits kept in Research Institutions. Making Life Easier for Animals in Laboratories. Reinhardt V and Reinhardt A. 2006. Animal Welfare Institute. Washington, DC. Available from Animal Welfare Institute, PO Box 3650, Washington, DC 20007.

<http://www.awionline.org/pubs/rabrodent/rodrab.html>

This book summarizes and discusses refinement and environmental improvement techniques for rodents and rabbits kept in research facilities. The review of the literature focuses on data-supported published material and mentions descriptive and theoretical articles only if they have practical relevance. A total of 260 relevant articles published in 85 different journals have been reviewed.

Recognition and Alleviation of Pain

“AVMA Guidelines on Euthanasia” 2007.

http://www.avma.org/issues/animal_welfare/euthanasia.pdf

Useful reference to insure that appropriate methods of euthanasia are being used for different laboratory animal species.

“Adverse effects in animals and their relevance to refining scientific procedures.” Morton DB. 1990. *ATLA (Alternatives to Laboratory Animals)* 18, 29-39.

The author highlights areas in which suffering can be prevented, alleviated and avoided. The terms "pain, stress, eustress, distress and fear" are clearly defined to help determine animal suffering. "One has to recognize suffering by non-verbal means, i.e., through observing changes in behavior patterns and in physiology.... I have tried to highlight in a general way the areas in which refinement, with the specific aim of reducing laboratory animal pain, distress and anxiety, can be achieved. Important aspects are good husbandry and housing which meet the animals' behavioural needs, careful and gentle handling, competence in carrying out scientific procedures, and alleviation of unwanted side-effects."

“Assessment and alleviation of post-operative pain.” Flecknell P. 1997/98. *Animal Welfare Information Center Newsletter* 8(3-4), 8-14.

<http://www.nal.usda.gov/awic/newsletters/v8n3/8n3fleck.htm>

The author addresses a few common concerns about the use of analgesics to relieve post-operative pain, describes various methods of pain assessment such as scoring systems, gives suggestions on the clinical use of analgesics and offers recommendations on the relief of post-operative distress. Tables with recommended dosages of analgesics are given. "Providing effective post-operative pain relief can have a dramatic effect on the speed with which animals return to normality following surgical procedures. The provision of good post-operative care should be considered essential both because of a concern for the animal's welfare and also because it is good scientific practice."

Guidance Document on the Recognition, Assessment, and Use of Clinical Signs as Humane Endpoints for Experimental Animals Used in Safety Evaluation. 2000. Organisation for Economic Cooperation and Development (OECD). Paris, France.

[http://www.oalis.oecd.org/oalis/2000doc.nsf/LinkTo/env-jm-mono\(2000\)7](http://www.oalis.oecd.org/oalis/2000doc.nsf/LinkTo/env-jm-mono(2000)7)

The purpose of the document is to apply the principles of the Three Rs to the use of animals in regulatory toxicity tests. It specifically addresses refinement and describes approaches to detecting clinical signs of pain and distress and procedures that can be put in place to minimise test animal pain, distress, and suffering during testing.

“Guidelines on the recognition of pain, distress and discomfort in experimental animals and an hypothesis for assessment.” Morton DB and Griffiths PHM. 1985. *Veterinary Record* 116, 431-436.

The article helps not only newcomers inexperienced in the recognition of pain, but also experienced workers who may be called upon to evaluate the pain involved in a new model or an individual animal. Specific signs

of behavior and common clinical signs indicating pain, distress or discomfort in laboratory animals are listed and discussed.

Guidelines on Choosing an Appropriate Endpoint in Experiments Using Animals for Research, Education, and Testing. 1999. Canadian Council on Animal Care. Ottawa, Ontario.

http://www.ccac.ca/en/CCAC_Programs/Guidelines_Policies/GDLINES/ENDPTS/APOPEN.HTM

Guidelines are presented for selecting an endpoint that reduces animal pain and/or distress. “For the purposes of these guidelines, the term ‘Endpoint’ is defined as the point at which an experimental animal’s pain and/or distress is terminated, minimized or reduced, by taking actions such as killing the animal humanely, terminating a painful procedure, or giving treatment to relieve pain and/or distress.”

Humane Endpoints in Animal Experiments for Biomedical Research. Hendriksen FM, Morton DB, eds. 1998. Proceedings of the International Conference, 22-25 November 1998, Zeist, The Netherlands. Laboratory Animals Ltd. London, England.

The papers presented address issues relating to the recognition and assessment of adverse effects in animals, and the determination, validation, implementation and acceptance of humane endpoints.

“Implementing assessment techniques for pain management and humane endpoints.” Morton DB. 1998. *Pain Management and Humane Endpoints.* Proceedings of a workshop. Johns Hopkins Center for Alternatives to Animal Testing.

<http://altweb.jhsph.edu/meetings/pain/morton.htm>

Score sheets are drawn up for each scientific procedure, and for each species, listing cardinal clinical signs that are observable and measurable, developed through the experience of a team of observers. The score sheet system helps focus attention on an animal’s condition throughout the procedure. It also helps determine the effectiveness of any therapy intended to relieve adverse effects, and which experimental models cause the least pain and distress, thus helping to refine scientific procedures. This technique is especially useful with new procedures, or when users are not sure of what effects a procedure will have.

“Invasiveness scales for animal pain and distress.” Orlans FB. 1996. *Lab Animal* 25(6), 23-25.

Recommends the use of well-defined invasiveness scales to improve animal welfare standards. By using such scales “laboratory workers would acquire greater sensitivity to animal pain and distress, improving ethical decision making.”

“Pain — assessment, alleviation and avoidance in laboratory animals.” Flecknell P. 1999. An ANZCCART factsheet. *Australian and New Zealand Council for the Care of Animals in Research and Teaching (ANZCCART) News* 12 (4), insert 1-10.

http://www.adelaide.edu.au/ANZCCART/publications/Pain%20Assessment_9.pdf

Valuable suggestions are given for the recognition of pain in laboratory animals and facilitation of its assessment. Available methods of pain control and their implementation are outlined. Control of acute pain can be achieved relatively easily in most mammalian species by use of analgesic agents. It is important to consider not only measures directed towards alleviating or preventing pain, but also the overall care of the animal and the prevention of distress. Tables with analgesic dosages for ferrets, guinea pigs, mice, rats, rabbits, cats, dogs, pigs, sheep and non-human primates are included.

“Pain and distress: what really matters?” Koch VW 2006. *Lab Animal* 35(5) 27-32.

“The author argues that IACUCs and investigators should shift their focus from the word ‘pain’ to the more inclusive word ‘distress’ referring to any mentally unpleasant level of stress in an animal, including slight discomfort. She discusses criteria for defining ‘significant’ distress, and offers suggestions for the conduct of studies to determine levels of distress.”

Pain Management and Humane Endpoints. Johns Hopkins Center for Alternatives to Animal Testing. 1998. Proceedings of a workshop.

<http://altweb.jhsph.edu/meetings/pain/proceedings.htm>

Excellent papers on assessment and alleviation of pain and distress, policy, implementation and humane endpoints. Useful information for IACUCs on how to review protocols and potentially painful procedures, and on their responsibilities in the implementation of policy leading to avoidance and minimization of pain in animals.

“Physiological and ethological aspects of the assessment of pain, distress and suffering.”

Scharmann W. 1998. *Humane Endpoints in Animal Experiments for Biomedical Research*. Hendriksen FM, Morton DB, eds. Proceedings of the International Conference, 22-25 November 1998, Zeist, The Netherlands. 33-39. Laboratory Animals Ltd. London, England.

Practical approaches to the recognition of pain and grading of pain intensity are described.

“Post-operative care and analgesia of farm animals used in biomedical research.” Randolph MM. 1994. *Animal Welfare Information Center* 5(1), 11-13.

<http://www.nal.usda.gov/awic/newsletters/v5n1.htm> [scroll down]

Excellent guidance for the post-operative care and analgesia of farm animals used in research. "An astute caretaker's knowledge of the normal behavior for that particular age, sex, species, and individual is crucial in determining when animals are experiencing unacceptable levels of pain... The recovery period should be viewed as the final stage in the surgical procedure. Some investigators and their staff have underestimated the importance of this stage of the surgical endeavor. There can be no successful surgery with an unsuccessful recovery." This article includes a clear table with practical, post-operative analgesics for ruminants and pigs.

“Practical applications of animal harm scales: international perspectives.” Orlans, FB. 2000. In *Progress in the Reduction, Refinement and Replacement of Animal Experimentation*. Balls E, van Zeller AM and Halder ME, eds. 1049-1056. Elsevier. Amsterdam, Netherlands.

Harm scales have been introduced in several countries to categorise degrees of animal pain or distress resulting from experimental procedures. These scales serve three purposes: 1. They promote an understanding of fundamental concepts of humane animal experimentation, the three Rs, and sensitise investigators and reviewers to the ethical significance of animal harm. 2. Harm scales are useful in framing policies on the use of animals in education. 3. In the reporting of national statistics on laboratory animal use, increased public accountability occurs if data are reported according to level of harm.

Recognition and Alleviation of Pain and Distress in Laboratory Animals. Committee on Recognition and Alleviation of Distress in Laboratory Animals, & National Research Council. 2008. National Academy Press. Washington, DC.

http://books.nap.edu/openbook.php?record_id=11931

"The book focuses specifically on the scientific understanding of the causes and the functions of stress and distress, the transformation of stress to distress, and the identification of principles for the recognition and alleviation of distress. This book discusses the role of humane endpoints in situations of distress and principles for the minimization of distress in laboratory animals."

“Recognizing animal suffering and pain.” Mroczek NS. 1994. *Lab Animal* 23(1), 27-31.

“Recognizing pain in animals requires empathic observation, which in turn engenders identification and often sympathy. Scientists have undertaken voluminous research that is based on the assumption that animals do feel pain. Pain inherent in animal research, however, is often ignored as subjective experience of the animal's reality in a simplistic attempt to objectify animal life and hence reduce it to measurable data.”

Research Animal Anesthesia, Analgesia and Surgery. Smith AC and Swindle MM, eds. 1994. Scientists Center for Animal Welfare. Greenbelt, MD.

A conference report with excellent contributions on “**Intraoperative monitoring and equipment**” Hoyt RF, 137-146; “**Cardiopulmonary complications and emergencies in surgery**” Swearengen JR, 159-166; “**Rabbits and rodents: Anesthesia and analgesia**” Wixson SK, 59-92; “**Dogs and cats: Anesthesia and analgesia**” Daunt DA, 93-105; “**Miscellaneous species: Anesthesia and analgesia**” Schaeffer DO, 129-136.

“**Stress and distress**” Roberts SA et al. 2006. *Animal Technology and Welfare*, 5, 99-102

http://www.awionline.org/Lab_animals/biblio/atw12.html

Definitions and practical hints on how to recognize stress and distress.

Refinement of Handling and Housing Conditions (Enrichment)

“Proper housing and management of animal facilities are essential to animal well-being, to the quality of research data and teaching or testing programs in which animals are used, and to the health and safety of personnel. A good management program provides the environment, housing, and care that permit animals to grow, mature, reproduce, and maintain good health; provides for their well-being; and minimizes variations that can affect research results.”

Guide for the Care and Use of Laboratory Animals

“**Behavioral indexes of poor welfare in laboratory rats.**” Patterson-Kane EG, Hunt M and Harper DN. 1999. *Journal of Applied Animal Welfare* 2, 97-110.

Fearfulness associated with single-housing was alleviated by pair-housing, and even further alleviated by group-housing in enriched cages. Problem solving ability was improved when rats were kept in groups in enriched cages rather than in barren single cages.

“**Cage enrichment for hamsters housed in suspended wire cages.**” McClure DE and Thomson JJ. 1992. *Contemporary Topics in Laboratory Animal Science* 31(4), 33.

"When these hamsters were provided with nesting material their well-being was improved as indicated by resolution of inappetence and depression. Providing the PVC apparently resolved the aggressive behavior problem by providing a means for seclusion in addition to functioning as a burrow and as a toy."

“**Catching individual rhesus monkeys living in captive groups.**” Reinhardt V. 1990. Available from Wisconsin Regional Primate Research Center, 1220 Capitol Court, Madison, WI 53715.

This 17-minute videotape demonstrates how rhesus macaques living in a breeding troop can readily be trained to enter a chute voluntarily or on vocal command and exit one-by-one into a transport box to allow capture. The procedure avoids undue stress; it is carried out by a single person.

Comfortable Quarters for Laboratory Animals. Reinhardt V and Reinhardt A, eds. 2002. 9th edition, Animal Welfare Institute. Washington, DC. Available from Animal Welfare Institute, PO Box 3650, Washington, DC 20007.

<http://www.awionline.org/pubs/cq02/cqindex.html>

A collection of 16 articles outlining refined, i.e., species-appropriate housing conditions and handling techniques for mice, gerbils, rats, hamsters, guinea pigs, rabbits, cats, dogs, primates, pigs, sheep, cattle, horses, chickens, amphibians and reptiles. Suggestions and recommendations are made which minimize or eliminate variables such as distress, fear, anxiety, discomfort, depression and boredom "thereby maximizing the research animals' well-being and reducing the number of subjects required to achieve statistical significance of the research data."

“**The effect of feeding and housing on the behaviour of the laboratory rabbit.**” Krohn TC, Ritskes-Hoitinga J and Svendsen P. 1999. *Laboratory Animals* 33, 101-107.

"While the [individually housed] rabbits in cages spent 2-5% of the time performing abnormal behaviour like biting the bars or scratching the bottom of the cage, these activities were virtually absent in group-housed rabbits in floor pens."

“Effects of environmental enrichment on behaviour and reproductivity of growing pigs.” Beattie VW, Walker N and Sneddon IA. 1995. *Animal Welfare* 4, 207-220.

"Enriching the environment [extra area with peat and a straw hopper; four times as much floor space] reduced both the amount of time pigs spent inactive and the time involved in harmful social and aggressive behaviour. Tail biting was absent from the enriched environment but four pigs were removed from barren pens with severe tail damage."

“An enrichment object that reduces aggressiveness and mortality in caged laying hens.” Gvaryahu G, Ararat E, Asaf E, Lev M, Weller JI, Robinzon B and Snapir N. 1994. *Psychology & Behavior* 55, 313-316.

"The enrichment devices (colored key rings) significantly reduced aggressive head-pecking behavior and significantly decreased the mortality rate."

Enrichment Strategies for Laboratory Animals. (whole issue). 2005. *ILAR Journal* 46(2).

http://dels.nas.edu/ilar_n/ilarjournal/46_2/html/

"Currently, it is recognized that science also has an ethical responsibility to house animals according to their species-specific needs, and that responsibility invokes the concept of behavioral and environmental enrichment.... To develop enrichment programs and to assess the effectiveness of such programs, it is critically important for all staff members involved in the care and use of the animals to understand the behaviors of the individual species that are housed."

“Environmental enrichment of laboratory animals used in regulatory toxicology studies.” Dean SW. 1999. *Laboratory Animals* 33, 309-327.

"A creative approach to environmental enrichment is indeed compatible with regulatory toxicology. It is hoped that this will encourage those responsible for the care and welfare of animals in such a laboratory to challenge historical practices and include environmental enrichment as a fundamental necessity of study design.... The assumption that certain regulatory authorities 'prefer' single-housing should be challenged.... Group housing should become the norm whenever animals are compatible, and anything less should be justified on the basis of sound science."

Guide to the Care and Use of Experimental Animals, Vol. 2, Gilman J, ed. 1984. Canadian Council on Animal Care. Ottawa, Ontario.

http://www.ccac.ca/en/CCAC_Programs/Guidelines_Policies/GUIDES/ENGLISH/TOC_V2.HTM

Excellent guidelines for the species-appropriate keeping of fish, amphibians, reptiles, and pigeons in the research laboratory setting. Chapters on fish, amphibians, reptiles and pigeons can be accessed individually.

Making Lives Easier for Animals in Research Labs: Discussions by the Laboratory Animal Refinement & Enrichment Forum. Baumans V et al. (eds) 2007. Animal Welfare Institute, Washington, DC. Available from Animal Welfare Institute, PO Box 3650, Washington, DC 20007.

<http://www.awionline.org/pubs/LAREF/LAREF-bk.html>

This book is a collection of electronic discussions that took place on the Laboratory Animal Refinement & Enrichment Forum (LAREF) between October 2002 and May 2007. The forum serves the international animal care and animal research community to promote animal welfare and improve scientific methodology by avoiding or minimizing stress and distress resulting from husbandry and handling practices.

Of more than 5,000 comments posted, approximately 3,000 were selected for this book because they have practical animal welfare relevance and are based on first-hand experiences about ways to improve the conditions under which animals are housed and handled in research facilities.

The following main topics are discussed: Basic Issues; Maladaptive Behaviors; Environmental Enrichment; Social Housing; Working With Animals; Extraneous Variables

“Pair-housing overcomes self-biting behavior in macaques.” Reinhardt V. 1999. *Laboratory Primate Newsletter* 38(1), 4.

<http://www.brown.edu/Research/Primate/lpn38-1.html#pair>

“The transfer to a compatible social-housing arrangement [isosexual pair-housing] effectively cured the [seven] rhesus subjects from the behavioral pathology of habitual self-biting.”

“Restraint methods of laboratory nonhuman primates: A critical review.” Reinhardt V, Liss C and Stevens C. 1995. *Animal Welfare* 4, 221-238.

http://www.awionline.org/Lab_animals/biblio/aw6metho.htm

Nonhuman primates can readily be trained to co-operate rather than resist during common handling procedures, thereby avoiding data-biasing distress responses associated with traditional involuntary restraint techniques.

“The role of husbandry in promoting the welfare of laboratory animals.” Reese EP. 1991. *Animals in Biomedical Research*, Hendriksen CFM and Koëter HBWM, eds, 155-192. Elsevier. Amsterdam, Netherlands.

A very well written outline of refinement options to promote the welfare of laboratory animals. “It appears that many of our laboratory findings are based upon data from distressed animals.”

“Social housing ameliorates behavioral pathology in *Cebus apella*.” Bayne K, Dexter SL and Suomi SJ. 1991. *Laboratory Primate Newsletter* 30(2), 9-12.

<http://www.brown.edu/Research/Primate/lpn30-2.html#bayne>

Change from single- to group-housing "effectively reduced stereotypic behaviors; however, it also was associated with more passive behaviors being exhibited by the subjects. The concurrent shifts in these components of the behavioral repertoire suggests that the animals were in a calmer state when housed socially."

“Use of cornhusk nesting material to reduce aggression in caged mice.” Armstrong KR, Clark TR and Peterson MR. 1998. *Contemporary Topics in Laboratory Animal Science* 37(4), 64-66.

The provision of cornhusk reduced aggressive interactions by offering subordinate animals cover and escape routes.

See also:

➤ *Database on Refinement of Housing and Handling Conditions and Environmental Enrichment for Laboratory Animals [except primates]*

<http://www.awionline.org/SearchResultsSite/refine.aspx>

➤ *Environmental Enrichment Information Resources for Laboratory Animals.*

<http://www.nal.usda.gov/awic/pubs/enrich/intro.htm>

in the databases and bibliographies section p. 24.

Species-Specific Requirements for Enrichment

Exercise for Dogs

“... Research facilities must develop, document, and follow an appropriate plan to provide dogs with the opportunity for exercise... The plan must include written standard procedures to be followed in providing the opportunity for exercise.”

AWA Regulations. Part 3. Standards. §3.8 Exercise for dogs.

“Comfortable quarters for laboratory dogs.” Hubrecht RC. 2002. In *Comfortable Quarters for Laboratory Animals*, 9th Edition. Reinhardt V & Reinhardt A, eds, 56-64. Animal Welfare Institute. Washington, DC.

<http://www.awionline.org/pubs/cq02/Cq-dogs.html>

Well-tested refinement options for the housing and handling of laboratory dogs are outlined. The following issues are addressed in detail: Space considerations and minimum space allowances, social housing, exercise, structures and enrichment within the dog enclosure, social interaction of dogs with animal care staff, minimization of stress during interactions between people and dogs, noise, dog supply and rehoming.

“Correlates of pen size and housing conditions on the behaviour of kennelled dogs.” Hubrecht RC, Serpell JA and Poole TB.1992. *Applied Animal Behaviour Science* 34, 365-383.

Behavioral data were collected from solitary and group-housed dogs [of different breeds] from animal shelters and laboratories. "Solitary dogs were more inactive and spent more time in non-social repetitive locomotory behaviour categories." Dogs housed in pens with spacious, complex outdoor runs were more active and engaged in more species-typical locomotory behaviors than dogs kept in small and barren standard pens.

Guidelines for the Care and Housing of Dogs in Scientific Institutions. 1999. NSW Agriculture. Orange, NSW, Australia.

<http://www.animaethics.org.au/reader/animal-care>

Guidelines on exercise pen design; exercising and contact with humans apart from normal husbandry procedures are spelled out. "Regular exercise allows for dogs to increase their range of behaviours... The ideal time for social interaction [with humans] is during the exercise period...The minimum exercise period should be 30 minutes for healthy dogs...Group-housed dogs should be exercised as a group..."

“A Novel approach for addressing enrichment and exercise for dogs in a teaching institution.”

Hammer JG. 2001, *Lab Animal* 30(7), 26-29.

An IACUC approved, successfully tested [with four beagle dogs] program to foster positive human-animal interactions [a retirement home was visited on a regular basis] and exercise [via preparatory training program] is described.

Environmental Enhancement for Primates

“... Research facilities must develop, document, and follow an appropriate plan for environmental enhancement adequate to promote the psychological well-being of nonhuman primates.... The physical environment in the primary enclosures must be enriched by providing means of expressing noninjurious species-typical activities.”

AWA Regulations. Part 3. Standards. §3.81 Environmental enhancement to promote psychological well-being.

Environmental Enrichment for Caged Rhesus Macaques (Macaca mulatta): Photographic Documentation and Literature Review. Reinhardt V and Reinhardt A. 2001. Animal Welfare Institute. Washington, DC. Available free from Animal Welfare Institute, PO Box 3650, Washington, DC 20007.

http://www.awionline.org/lab_animals/rhesus/Photo.htm

A collection of 108 annotated photos dealing with all aspects of environmental enrichment – including refinement of housing and handling conditions – for caged macaques. “Sharing the same roots makes it easy for any compassionate human primate to make life easier for a nonhuman primate subjected to biomedical research.” A comprehensive bibliography is appended to this document.

The Psychological Well-Being of Nonhuman Primates. Committee on Well-Being of Nonhuman Primates. 1998. Institute for Laboratory Animal Research, Commission on Life Sciences, National Research Council. National Academy Press. Washington, DC.

<http://books.nap.edu/books/0309052335/html/index.html>

Concise guidelines for the adequate housing and handling of captive nonhuman primates.

Criteria of psychological well-being are: "Appropriate social companionship; opportunities to engage in behavior related to foraging, exploration, and other activities appropriate to the species, age, sex, and condition of the animal; housing that permits suitable postural and locomotor expression; interactions with personnel that are generally positive and not a source of unnecessary stress."

“Social enhancement for adult nonhuman primates in research laboratories: A review.” Reinhardt V and Reinhardt A. 2000. *Lab Animal* 29(1), 34-41.

Comprehensive review of safe, effective and inexpensive options (pair-housing, human interaction, training to cooperate during procedures) to address the social needs of adult nonhuman primates in accordance with the stipulations set forth in the Animal Welfare Act.

“Social-housing of previously single-caged macaques: What are the options and the risks?”

Reinhardt V, Liss C and Stevens C. 1995. *Animal Welfare* 4, 307-328.

http://www.awionline.org/Lab_animals/biblio/aw7socia.htm

“The published data show that previously single-caged macaques can be transferred to social housing adequate for the species ... without undue risk to individual animals” and without interfering with husbandry and common research protocols.

See also page 25-26:

➤ *Annotated Bibliography on Refinement and Environmental Enrichment for Primates*

http://www.awionline.org/lab_animals/biblio/

➤ *Environmental Enrichment for Primates: Annotated Database...*

<http://www.awionline.org/SearchResultsSite/enrich.aspx>

ALTERNATIVES IN EDUCATION/TEACHING

“Alternatives to the use of animals in higher education.” van der Valk J et al. 1999. *ATLA (Alternatives to Laboratory Animals)* 27(1), 39-52.

The issues reviewed and discussed include the current use of animals in higher education, the range of alternatives currently available, the advantages and disadvantages of using alternatives in education, methods of disseminating information about alternatives to those involved in education systems, and strategies for evaluating the educational effectiveness of alternatives.

AVAR (Association of Veterinarians for Animal Rights) Alternatives in Education Database. 1990-2006*. Association of Veterinarians for Animal Rights. Davis, CA.

<http://www.avar.org/alted/>

Searchable database containing audiovisual and textual materials, computer programs, simulations, models and other resources that can be used as alternatives to animals at all levels of education from primary school to the training of medical and veterinary professionals.

From Guinea Pig to Computer Mouse: Alternative Methods for a Progressive, Humane Education.

Jukes N and Chiulia M. 2nd edition, 2003. Interniche (International Network for Humane Education.) Leicester, England.

<http://www.interniche.org/news.html#book>

The book investigates state-of-the-art alternative tools and approaches to support ethical and effective knowledge and skills acquisition within biological science, medical and veterinary medical education.

“Guidelines for humane education: Alternatives to the use of animals in teaching and training.”

Smith A and Smith K. 2004. *The Three Rs at the Beginning of the 21st Century:* Proceedings of the Fourth World Congress on Alternatives and Animal Use in the Life Sciences. Balls M, Firmani D and Rowan A, eds. *ATLA (Alternatives to Laboratory Animals)* 32, Supplement 1, 29-39.

http://www.worldcongress.net/2002/proceedings/SP4_Smith.pdf

“This paper attempts to clarify the issues raised, presents an overview of the alternatives available with their strengths and weaknesses, and finally offers guidelines for humane education that take into consideration both the practical issues and the feelings of all those involved.”

Interniche: The International Website for Humane Education.

<http://www.interniche.org/>

Interniche supports progressive science teaching and the replacement of animal experiments by working with teachers to introduce alternatives and with students to support freedom of conscience. The web site is aimed at teachers and students of biological science, veterinary and human medicine, as well as university ethics committee or animal care and use committee members, legislators, and animal welfare advocates.

“Medical training using simulation: toward fewer animals and safer patients.” Balcombe J. 2004. *The Three Rs at the Beginning of the 21st Century*: Proceedings of the Fourth World Congress on Alternatives and Animal Use in the Life Sciences. Balls M, Firmani D and Rowan A, eds. *ATLA (Alternatives to Laboratory Animals)* 32, Supplement 1, 553-560.

http://www.worldcongress.net/2002/proceedings/D4_20Balcombe.pdf

”This paper presents the current status of computer-based simulation in medicine. Today, more than a dozen companies are producing virtual reality simulators and interactive manikins for training in endoscopy, laparoscopy, anaesthesia, trauma management, angiography, and needle insertion... Replacing animals with simulators in medical training is limited no longer by technical feasibility but by a willingness of the medical community to embrace it.”

The NORINA (A Norwegian Inventory of Alternatives) Database: Audiovisual Alternatives to Laboratory Animals in Teaching. Smith K and Smith A, eds. 2006*. Norwegian School of Veterinary Medicine. Oslo, Norway.

http://oslovet.veths.no/fag.aspx?fag=57&mnu=databases_1

English-language database of audiovisuals for the use of teachers and instructors in the biological sciences. Its primary purpose is to provide an overview of possible alternatives at all educational levels, from elementary school to university. The database consists of more than 3700 entries including computer programs, interactive videos, films, and traditional teaching aids such as slide sets, 3-D models, and classroom charts.

“The PVC-Rat and Other Alternatives in Microsurgical Training.” Remie R. 2001. *Lab Animal* 30(9).

"The number of animals used in educational training programs in experimental microsurgery can be reduced by using artificial devices such as the anastomoses device and the MD PVC-Rat. Such *in vitro* methods allow development of technical skills.... The use of the PVC-Rat model reduces the number of animals used during training of scientists and animal technicians by roughly 90%."

WEB RESOURCES FOR THE LITERATURE SEARCH (USDA Policy #12)

Databases, Directories, Bibliographies

Alternatives to the Use of Live Vertebrates in Biomedical Research and Testing. A Bibliography with Abstracts. Hudson VW and Nguyen Q. 2002. National Library of Medicine. Bethesda,MD.

<http://toxnet.nlm.nih.gov/altbib.html>

A quarterly publication citing techniques that replace or may replace intact vertebrates in biomedical testing to evaluate the toxicological potential of various substances. Very comprehensive. A search engine now enables users to search all issues and/or to limit the search by field, year, and category, and browse the index.

Altweb Pain Management Database. Allen T. 2007*. Altweb Web Site.

http://apps1.jhsph.edu/altweb/aadb/aadb_search.cfm

"This database includes information about anesthesia and analgesia for most commonly used laboratory animals, including: rats, mice, primates, dogs, cats, rabbits, pigs, guinea pigs, birds, sheep, fish, and exotic species. It provides information about available drugs and the side effects of commonly used drugs. Citations are from publications that have published laboratory animal studies or human clinical studies with relevance to animal research." Contains approximately 10,000 records, most of them with abstracts. It covers the period 1990 to the present, and is updated quarterly. Almost all of the records--98%--have abstracts. Records have been drawn from three major databases: MEDLINE (with records from TOXLINE as well), AGRICOLA, and AGRIS."

AnimalAlt-ZEBET Database. Spielmann H, Grune B, Dorendahl A and Skolik S. 1989-2008*. ZEBET [Center for Documentation and Evaluation of Alternative Methods to Animal Experiments]. Berlin, Germany.

<http://www.dimdi.de/static/en/db/dbinfo/zt00.htm>

Invaluable resource for the literature search. ZEBET is a database of alternative methods (replacement, refinement and reduction) to animal experiments in the English language. Each record contains a short description of a method in its most important details. An evaluation by ZEBET staff indicates whether the method results in the replacement, reduction or refinement of animal use according to the "3R's."

Annotated Bibliography on Refinement and Environmental Enrichment for Primates kept in Laboratories. Reinhardt V and Reinhardt A. 2008. 12th Edition. Animal Welfare Institute. Washington, DC.

http://www.awionline.org/lab_animals/biblio/index.html

This bibliography offers practical information on techniques that promote the expression of species-appropriate behavioral and mental activities in captive nonhuman primates. Specifically geared towards animal caregivers, animal technicians, zoo keepers, students and veterinarians. All entries are annotated.

Database on Refinement of Housing and Handling Conditions and Environmental Enrichment for Laboratory Animals: Rodents, Rabbits, Cats, Dogs, Ferrets, Farm Animals, Horses, Birds, Fishes, Amphibians and Reptiles. Reinhardt V and Reinhardt A. 2008*. Animal Welfare Institute. Washington, DC.

<http://www.awionline.org/SearchResultsSite/refine.aspx>

More than a thousand annotated entries, 13% full-text, on all aspects of environmental enrichment and refinement of housing and handling conditions of laboratory animals and farm animals used in research. Access to the database is free. It is searchable by citation and keywords and updated once every two months.

"Free Databases". FRAME Web Page. 2006*.

http://www.frame.org.uk/page.php?pg_id=146

Excellent, comprehensive annotated list of databases with links. Includes three Rs databases; alternatives in education; cell-line databases; current research; general scientific; normal, transgenic and knockout strains.

Environmental Enrichment Information Resources for Laboratory Animals: Birds, Cats, Dogs, Farm Animals, Ferrets, Rabbits, And Rodents: 1965-1995. Smith CP and Taylor V. 1995. U.S. Department of Agriculture, National Agricultural Library, Animal Welfare Information Center. Beltsville, MD.

<http://www.nal.usda.gov/awic/pubs/enrich/intro.htm>

Articles and bibliographies covering environmental enrichment techniques for birds, cats, dogs, farm animals, ferrets, rabbits, and rodents.

Environmental Enrichment for Primates: Annotated Database on Environmental Enrichment and Refinement of Husbandry for Nonhuman Primates. Reinhardt V and Reinhardt A. 2008*. New

Edition. Animal Welfare Institute. Washington, DC.

<http://www.awionline.org/SearchResultsSite/enrich.aspx>

More than 1500 annotated entries – 22% full-text – on all aspects of environmental enhancement, including social housing, feeding enrichment, inanimate enrichment and training to cooperate during procedures. Access to the database is free. It is searchable by citation and keywords – including species names – and is updated at least once every two months.

Environmental Enrichment for Nonhuman Primates Resource guide. Adams K. 2006. National Agricultural Library, Animal Welfare Information Center, Beltsville, MD.

<http://www.nal.usda.gov/awic/pubs/Primates2006/Primates.htm>

"This publication updates and expands AWIC's Environmental Enrichment for Nonhuman Primates Resource Guide, January 1992-February 1999. It covers literature published from 1999-June 2006. The bibliographic chapters are divided into subject areas that cover the concept of environmental enrichment, enrichment for nonhuman primates overall, abnormal behavior exhibited by nonhuman primates, great apes and gibbons, macaques, old world monkeys other than macaques, marmosets and tamarins, other new world monkeys, lemurs, lorises, and tarsiers, animal training as an enrichment strategy, and books and proceedings from conferences focused on environmental enrichment or nonhuman primate management."

Environmental Enrichment for Rodents and Rabbits: An Annotated Bibliography. Reinhardt A. 2007. Animal Welfare Institute. Washington, DC.

http://www.awionline.org/Lab_animals/biblio/rabrodent.html

This bibliography offers animal caregivers, animal technicians, veterinarians, and students practical information on refinement and environmental enrichment for rodents and rabbits kept in research institutions.

"Free web resources on alternatives." Donnelly TM. 2004. *Lab Animal* 33(3), 46-48.

Access to many online databases for alternatives searches requires a paid subscription and these fees may be expensive for small or not-for-profit institutions. The author provides a list of free sites that can be used to fulfill the legal requirement for an alternatives search.

Institute for In Vitro Sciences . IIVS Publications: Bibliography. 2007*

<http://www.iivs.org/pages/publications.php>

Searchable bibliography of in vitro resources with access to online documents. Articles can also be browsed under the following categories: eye, ocular; skin ; phototoxicity; percutaneous penetration; acute toxicity; validation; alternative methods.

Practical Enrichment Options for Animals Kept in Research Institutions. Reinhardt A. 2007*. Animal Welfare Institute. Washington, DC.

<http://www.awionline.org/SearchResultsSite/enriop.aspx>

This database disseminates enrichment ideas and practical tips from the "Laboratory Animals Refinement and Enrichment Forum" and from other published information.

Refinement and Environmental Enrichment for All Laboratory Animals. Reinhardt V and Reinhardt A. 2008*. Animal Welfare Institute. Washington, DC.

<http://www.awionline.org/SearchResultsSite/laball.aspx>

This information resource is a merger of the two databases on refinement and enrichment for non-primate and primate laboratory animals. It contains nearly three thousand annotated entries, 17% full-text, on all aspects of refinement of housing and handling conditions and environmental enrichment for ALL animals kept in research, testing and teaching institutions. Access to the database is free. It is searchable by citation and keywords and updated at least once every two months.

Scientific Information Service on Advanced Alternative Methods to Animal Experiments in Biomedical Sciences (SIS). ECVAM (European Centre for the Validation of Alternative Methods).
<http://ecvam-sis.jrc.it/>

SIS provides factual and evaluated information on advanced non-animal test development and validation for toxicology assessments coming from a wide range of international information sources. It includes various databases on method descriptions, validation studies, INVITTOX protocols, compounds and test results, ECVAM workshop reports.

Search Engines, Search Guidelines

“A guide to searching for alternatives to the use of laboratory animals.” Bottrill K. *FRAME*.

http://www.frame.org.uk/page.php?pg_id=139

Useful and comprehensive guide, including basic principles for searching the Internet or any online database, comparison of search engines, suggestions for how to approach a search on the Three Rs; a description, including links, of some of the databases that can be accessed free on the Internet, as well as a listing of commercial databases and database hosts of potential relevance.

“The search for refinement alternatives: When you’ve just got to use animals.” Kreger, MD. 2000. *Lab Animal* 29(4), 22-25, 28-29.

Tips for researchers and IACUCs on how to conduct a literature search for refinement. “Refinements to animal-use protocols can help reduce animal pain and distress, thus producing cleaner data. Yet, most researchers neglect a refinement search, despite legal mandates, because the information is not readily available. What’s more, IACUCs may have difficulty assessing researchers’ refinement searches. The author offers some solutions.”

“Tips for searching for alternatives to animal research and testing.” Smith CP. 1994. *Lab Animal* 23(3), 46-48.

<http://www.nal.usda.gov/awic/alternatives/tips.htm>

“Investigators can assist information specialists who perform the search by being prepared to give precise and specific information about their research or testing procedures.” The type of information the investigator may be asked to provide is listed, and the two phases of a search strategy, 1. reduction and refinement, and 2. replacement, are delineated. Sample searches are given. Worksheets with instructions and a search summary form can be found at:

http://awic.nal.usda.gov/nal_display/index.php?info_center=3&tax_level=1&tax_subject=184

EXPERT HELP WITH THE ALTERNATIVES SEARCH:

- **AWIC (Animal Welfare Information Center)** offers expertise in formulation of the search strategy and selection of key words and databases, access to unique databases, on- and off-site training of institute personnel in conducting effective alternatives searches, and is able to perform no-cost or low-cost electronic database searches. AWIC can be contacted at (301) 504-6212, via E-mail at awic@nal.usda.gov [cited from USDA Policy #12]
- **The University of California Center for Animal Alternatives:** The Center offers help with the alternatives search, including search strategy, database selection, and general guidance and performs low-cost searches. Email: (animalalternatives@ucdavis.edu). Website: http://www.vetmed.ucdavis.edu/Animal_Alternatives/service.htm

Table 1: Specific Topics in *In Vitro* Testing

<p>Eye irritation</p>	<p>“A battery of cell toxicity assays as predictors of eye irritation: a feasibility study.” Rosenkranz HS and Cunningham AR. 2000. <i>ATLA</i> 28(4).</p> <p>“Justification of the enucleated eye test with eyes of slaughterhouse animals as an alternative to the Draize eye irritation test with rabbits.” Prinsen MK and Koeter HBWM. 1993. <i>Food and Chemical Toxicology</i> 31, 69-76.</p>	<p>“Results indicate that a battery of cytotoxicity tests could provide a viable alternative to the animal-based procedure.”</p> <p>The authors examined 21 test materials and concluded that the enucleated eye test provides a very accurate means of assessing eye irritant potential without using laboratory animals.</p>
<p>Monoclonal antibodies</p>	<p>“Small-Scale monoclonal antibody production <i>in vitro</i>: Methods and resources.” Jackson LR, Trudel LJ and Lipman NS. 1999. <i>Lab Animal</i> 28(3), 20-30.</p> <p>“Proceedings of the Production of Monoclonal Antibodies Workshop August 29, 1999, Bologna, Italy.” McArdle JE and Lund CJ, eds. Alternatives Research and Development Foundation and the Third World Congress on Alternatives and Animal Use in the Life Sciences.</p> <p>“Guidance Document for IACUC Evaluation of Monoclonal Antibody Production Protocols.” Adapted from DeTolla and Smith. Available from Alternatives Research and Development Foundation, 14280 Golf View Drive, Eden Prairie, MN 55346</p>	<p>Review of <i>in vitro</i> production of monoclonal antibodies. Highlights some of the <i>in vitro</i> technologies most commonly used and points to consider when selecting an <i>in vitro</i> method for MAB production.</p> <p>Description of advantages and disadvantages of <i>in vivo</i> and <i>in vitro</i> methods; up-to-date review of laboratory-scale <i>in vitro</i> methods for producing MABs; IACUC guidance for protocol review of MABs.</p> <p>Applicable concepts and guidelines appropriate for protocol review of Mabs are presented. Each of the questions listed in the IACUC checklist are discussed.</p>
<p>Toxicology</p>	<p>“Integrating computer prediction systems with <i>in vitro</i> methods towards a better understanding of toxicology.” Barratt MD. 1998. <i>Toxicology Letters</i> Dec. 102-103:617-21.</p> <p>“Integrated <i>In Vitro</i> approaches for assessing systemic toxicity.” Forsby A and Blaauboer B. 2003. In <i>Alternative Toxicological Methods</i>. Salem H and Katz S, 43-50. CRC Press. Boca Raton.</p> <p>“The integrated use of alternative methods in toxicological risk evaluation.” Blaauboer B. 1999. <i>ATLA</i> 27(2), 229-238.</p> <p>“<i>In vitro</i> methods for predicting human toxicity.” Silber P, Ruegg CE and Myslinski N. 1994. <i>Lab Animal</i> 23(2), 33-37.</p> <p>“Predictive value of <i>in vitro</i> model systems in toxicology.” Davila JC, Rodriguez RJ, Melchert RB and Acosta D. 1998. <i>Annual Review of Pharmacology and Toxicology</i> 38, 63-96.</p>	<p>Describes ways in which computer prediction systems and <i>in vitro</i> toxicology can complement each other in the development of alternatives</p> <p>“The ECITTS prevalidation study indicated that the strategy of using cellular <i>in vitro</i> methods integrated with computer-based biokinetic modeling can be a powerful tool for risk assessment.”</p> <p>In this report, a generic scheme for local/systemic toxicity, and a specific scheme for target organ toxicity, are proposed. The scope and limitations of the approaches are discussed.</p> <p>A brief but very clear introduction to <i>in vitro</i> prediction of target organ-specific toxicity using human tissues and cells.</p> <p>Overview of the use of <i>in vitro</i> model systems to investigate target organ toxicity of drugs and chemicals; also provides selective examples of these model systems.</p>
<p>Validation</p>	<p>“Practical aspects of the validation of toxicity test procedures.” Balls M. 1995. <i>ATLA</i> 23(1), 129-147.</p> <p>“Update on the validation and regulatory acceptance of alternative tests for skin corrosion and irritation.” Fentem JH and Botham PA. 2004. <i>The Three Rs at the Beginning of the 21st Century</i>. Balls M et al. eds. <i>ATLA</i> 32, Supplement 2, , 683-688.</p>	<p>Recommendations are made concerning the practical and logistical aspects of validating alternative toxicity testing procedures.</p> <p>Discussion of validation issues. Includes tables giving an overview and summary of the validation and regulatory acceptance of <i>in vitro</i> methods for skin corrosion and irritation.</p>
<p>Cosmetics</p>	<p>“The potential use of non-invasive methods in the safety assessment of cosmetic products.” Rogiers, V et al. 1999. <i>ATLA</i> 27(4), 515-537.</p>	<p>The potential of using non-invasive techniques in safety assessment with human volunteers is reviewed.</p>

Table 2: Specialized *In Vitro* Replacement Resources

Skin irritation	<i>Alternatives to Skin Irritation Testing in Animals.</i> Huggins, J. compiler.	http://www.invitroderm.com/	Searchable database containing approx. 360 citations with abstracts.
Monoclonal antibodies	<p>“Altweb special section on monoclonal antibodies”</p> <p>“Resource Lists of <i>In Vitro</i> MABs”</p> <p>“Companies and Institutes Providing New Technologies in Mabs.” Jensen A and Smith CP, compilers.</p>	<p>http://altweb.jhsph.edu/mabs/mabs.htm</p> <p>http://altweb.jhsph.edu/mabs/where.htm</p> <p>http://www.nal.usda.gov/awic/pubs/antibody/company.htm</p>	<p>Introduction and Resources, various <i>in vitro</i> listings of facilities that produce MABs.</p> <p>Where to get <i>in vitro</i> mabs: academic core centers and commercial facilities</p> <p>AWIC Selected list of company and institute resources in MABs with annotations.</p>
Toxicity	<i>MEIC (Multicentre Evaluation of In Vitro Cytotoxicity)</i>	http://www.cctoxconsulting.a.se/sidan1.htm	The database contains <i>in vitro</i> data for the 50 MEIC reference chemicals, as well as the methodology of the used <i>in vitro</i> assays.
<i>In Vitro</i> Tests	“INVITTOX protocols” <i>In Vitro Testing Industrial Platform</i> (Europe)	http://www.ivtip.org/tests.html	Up-to-date listing of <i>in vitro</i> tests currently available.

Note: * Date when last accessed while updating the bibliography. Web sites and databases are updated regularly.

This bibliography was compiled by Annie and Viktor Reinhardt, Animal Welfare Institute, and reviewed by Barbara Orlans, Kennedy Institute of Ethics.

It is available on the web at: http://www.awionline.org/lab_animals/iacuc2.htm