Guidelines for the treatment of animals in behavioural research and teaching

Behavioural studies are of great importance in increasing our understanding and appreciation of animals. In addition to providing knowledge about the diversity and complexity of behaviour in nature, such studies also provide information crucial to improvements in the welfare of animals maintained in laboratories, agricultural settings and zoos, and as companion animals. The use of animals in behavioural research and teaching does, however, raise important ethical issues. While many behavioural studies are noninvasive and involve only observations of animals in their natural habitat, some research questions cannot be addressed without manipulation of animals. Studies of captive animals necessarily involve keeping animals in confinement, at times require that provision is made for trapping and subsequent release of the animals, and manipulative procedures and surgery may be necessary to achieve the aims of the research. Studies of free-living animals in their natural habitats can cause disruption to the animals' population or the wider ecosystem, particularly if feeding, capture, marking or experimental procedures are involved. While the furthering of scientific knowledge is a proper aim and may itself advance an awareness of human responsibility towards animal life, the investigator must always weigh the potential gain in knowledge against any adverse consequences for the individual animals, populations under study, and the wider ecosystem. This is equally true for the evaluation of animal use in animal behaviour teaching activities. In fact, animal behaviour courses provide an excellent opportunity to introduce students to the ethical obligations a researcher accepts when animals are studied.

To help the members make what are sometimes difficult ethical judgements about the procedures involved in the study of animals, the Association for the Study of Animal Behaviour and the Animal Behavior Society have formed Ethical and Animal Care Committees, respectively. These committees jointly produced the following guidelines for the use of all those who are engaged in behavioural research and teaching activities involving vertebrate and invertebrate animals. These guidelines are general in scope, since the diversity of species and the study techniques used in behavioural research preclude the inclusion of prescriptive standards for animal care and treatment, other than emphasizing the general principle that the best animal welfare is a prerequisite for the best science. The guidelines are used by the Editors of Animal Behaviour in assessing the acceptability or otherwise of submitted manuscripts. Submitted manuscripts may be rejected by an Editor, after consultation with the Ethical or Animal Care Committee, if the content is deemed to violate either the letter or the spirit of the guidelines.

The ethical acceptability of manuscripts considered for publication in Animal Behaviour is weighed up as a cost-benefit analysis. Costs are considered to be costs to the animals (e.g. compromises of animal welfare, reduction of survivability or reproductive success) or the environment, or reductions in the quality of science. Benefits are considered to be the value of the specific scientific insights sought to humans, other animals, or the environment (i.e. whether the science is of good quality and addresses questions of importance). Any study that allows or precipitates great costs to animals for research must have both the highest potential benefits and the highest ethical justification. Great costs can be 'offset' in the cost-benefit analysis by achieving a high quality of research and/or answering very important questions. During ethical assessment of papers submitted to Animal Behaviour, the costs and benefits are weighed up on a case-by-case basis to assess whether costs have been minimized, the benefits maximized, and whether the benefits outweighed the costs, before making a recommendation on publication.

These guidelines supplement the legal requirements in the country and/or state or province in which the work is carried out. They should not be considered an imposition upon the scientific freedom of individual researchers, but rather as an aid to provide an ethical framework that each investigator may use in making and defending decisions related to animal welfare.

1. LEGISLATION

Investigators are accountable for the care and well-being of animals used in their research and teaching activities, and must therefore abide by the spirit as well as the letter of relevant legislation. It is their responsibility to acquire knowledge about local legislation. Appendix 1 lists sources of information relating to the legislation of several countries. Investigators must familiarize themselves with legislation both on animal welfare and on threatened and endangered species, and conform with the spirit and letter of the laws. When submitting manuscripts to Animal Behaviour, all authors must complete the ASAB/ABS Confirmation Form (download from the journal Web site http://www.editorialmanager.com/anbeh/) to verify that they have identified and adhered to the legal requirements of the country in which the study was conducted, and to provide relevant permit numbers. Many nations and academic institutions require that experiments performed on captive animals or on wildlife that are manipulated in some way must first be reviewed and approved by an
animal welfare, animal care and use, or ethics committee of the sponsoring institution (Jennings 1994; Hagelin et al. 2003). It is recommended that investigators from countries without any legal requirements or guidelines voluntarily refer to one or more of the documents in Appendix 1 for guidance. A manuscript based on institutional committee-approved research may still be referred by editors or reviewers to the Ethical and Animal Care Committees of ASAB/ABS, if they feel the manuscript raises ethical concerns.

2. THE THREE R’S: REPLACEMENT, REDUCTION AND REFINEMENT

Much of the current recommendations and legislation for ensuring appropriate animal care and use are based on the three guiding principles of replacement, reduction and refinement (Russell & Burch 1959). Replacement refers to efforts to replace animal subjects and models with non-animal ones, such as tissue cultures or computer models, wherever it is possible to do so while still achieving the scientific objectives. Reduction means reducing the numbers of animals affected by the experiment to the lowest number of individuals necessary to achieve the aims of the experiment and statistical power. There will be occasions when it is possible to reduce the total number of animals used in an experiment, but only by increasing the degree or duration of discomfort for the fewer individuals that are used. In such cases, the investigator must find an ethical balance between the two principles, and decide which produces the least ethical harm. Refinement refers to efforts to design and conduct the study as carefully as possible to maximize the scientific benefit while minimizing suffering to the animals, for example by planning and implementation of humane endpoints in the event that problems arise (Richmond 1998; Rowan 1998).

3. CHOICE OF SPECIES AND NONANIMAL ALTERNATIVES

Investigators should choose species and strains for study that are appropriate and best suited for investigation of the questions posed. Choosing these requires knowledge of natural history and phylogenetic relationships. Knowledge of an individual animal’s previous experience, such as whether or not it has spent a lifetime in captivity, is also important. Recent advances in genetic characterization of many laboratory animals may also allow the investigator to control for the effects of genotype on expected behavioural traits. The specialist characteristics and needs of some genetically altered strains must also be considered (Robinson et al. 2003); these are becoming increasingly common in behavioural studies and their welfare status and responses can be considerably different to those of nonmodified animals. When research or teaching involves procedures or housing conditions that may cause pain, discomfort or stress to the animal, and when alternative species or strains can be used, the researcher should use the species or strain that is believed to be the least likely to experience pain or distress (OTA 1986). The fact that a species being studied is classified as ‘vermin’ in the country concerned does not free the researcher from normal obligations to the experimental animals. The majority of invertebrate species are usually excluded from legislation regulating scientific research on animals. This does not mean that they are all unable to experience pain, discomfort or stress (Sherwin 2001). Manuscripts involving research with invertebrates may still be reviewed by the Ethical and Animal Care Committees in light of the most current knowledge in this respect. Researchers using such species should seek expert advice and take any evidence on this matter relating to their species into account when designing experiments, and should endeavour to minimize potential harm wherever possible. Live animal subjects are generally essential in behavioural research, but nonanimal alternatives such as video records from previous work or computer simulations can sometimes be used (Smyth 1978; van Zutphen & Balls 1997). Material of this kind also exists or can be produced for teaching purposes and can sometimes be used instead of live animals to teach aspects of the behavioural sciences (van der Valk et al. 1999; Smith & Smith 2001).

4. NUMBER OF INDIVIDUALS

The researcher should use the smallest number of animals necessary and sufficient to accomplish the research goals, especially in studies that involve manipulations that are potentially detrimental to the animal or the population. The number of animals used in an experiment can often be dramatically reduced by pilot studies, good experimental design and the use of statistical tests that enable several factors to be examined simultaneously (Weigler 1995; Festing 2001). Hunt (1980), Still (1982) and McConway (1992) discuss ways of minimizing the numbers of animals used in experiments while retaining scientific validity through sound experimental design and data analysis. It is equally important not to use so few animals that the value of the research is compromised. Useful reference works are Cox (1958) and Cochran & Cox (1966).

Investigators should be aware that in studies involving the use of genetically altered animals, there may be underlying ethical and welfare problems associated with their use, including the fact that greater numbers of animals are used in the pre-experimental production phase of these animals. There are published discussions of the pros and cons of using genetically altered animals (Hubrecht 1995; Poole 1995; Wolf & Wanke 1997; Boyd Group 1999).

5. PROCEDURES

Investigators are encouraged to discuss with colleagues both the scientific value of their research proposals and possible animal welfare and ethical considerations. There are several models for evaluating animal research that
can be of use when making ethical decisions (Bateson 1986; Orlans 1987; Shapiro & Field 1988; Donnelley & Nolan 1990; Porter 1992; de Cock Buning & Theune 1994; Fraser et al. 1997; Sandee et al. 1997; Richmond 1998; Fraser 1999). If procedures used in research or teaching involve exposure to painful, stressful or noxious stimuli, whether through acts of commission or omission, the investigator must consider whether the knowledge that may be gained is justified. This will partly depend upon the goal of the research (e.g. research designed to enhance our understanding of animal welfare issues immediately may be judged differently to research designed for other purposes). Custom and practice, economic savings, convenience, or the fact that an animal might experience the same or similar stimuli in the wild are not adequate justifications. It should be borne in mind that the welfare costs of animal use may reflect not only the infliction of that which is unpleasant, but the denial of that which is pleasurable. There is a considerable amount of literature discussing the assessment of pain, suffering and well-being (e.g. Morton & Griffiths 1985; AVMA 1987; Bateson 1991; NRC 1992; Broom & Johnson 1993; Flecknell 1994; Morton 1998; Poole 1999; USDA 1999; Flecknell & Waterman-Pearson 2000; Hellbrekers 2000). Researchers are urged to consider the use of more refined procedures before using techniques that are likely to cause physical or psychological discomfort to the animal (Kreger 2000). When attempting to identify potential alternative procedures, the investigator will need to consider the pain or distress-causing potential of all aspects or stages of the procedures in an experiment. The possible procedural substitutes or refinements for each may be very different. Pain or suffering should be minimized both in duration and magnitude as far as possible under the requirements of the experimental design. However, it should be borne in mind that studies of factors such as housing or management procedures that may induce states of stress, anxiety and fear can be a necessary part of research aimed at improving animal welfare or scientific validity. Attention should be paid to the provision of proper pre- and postprocedural care to minimize preparatory stress, immediate and residual effects. When a study involves any procedure or condition likely to cause more than short-term, low-intensity pain, discomfort, or distress, then appropriate anaesthesia, analgesia, tranquillization and/or adjunctive relief measures should be used, sufficient to prevent or alleviate the discomfort, unless this would jeopardize the aims of the study. Analgesics may also be necessary after such procedures, and should be used to minimize pain and distress whenever possible (Flecknell 1985; Benson et al. 1990; Flecknell & Waterman-Pearson 2000). In certain species, appropriate procedural training of animals can reduce the stress of some experimental procedures (see Reinhardt 1997; Grandin 2000). The investigator should keep in mind that many forms of discomfort or suffering can involve experiences other than nociception, such as nausea, pruritis, thermal stress, social isolation, or fear (McMillan 2003). Many of these can be alleviated or prevented by medication, procedures or housing conditions that specifically target those experiences. Investigators should consult with experts for guidance on how to control or pre-empt suffering in its various forms.

The following more specific points may be of use.

(a) Fieldwork

Investigators studying free-living animals are expected to take precautions to minimize interference with individuals, as well as the populations and ecosystems of which they are a part. Capture, marking, radiotagging, collection of physiological data such as blood or tissue samples or field experiments may have delayed adverse consequences such as a reduced probability of survival and reproduction (Parris & McCarthy 2001; McCarthy & Parris 2004). Investigators should consider the effects of such interference, and use the least disruptive techniques (Beausoleil et al. 2004) such as individual recognition by the use of natural features rather than marking (Scott 1978), or minimally invasive techniques for monitoring physiology (Cooper 1998; Gedir 2001) where possible. Cuthill (1991) discusses the ethical issues associated with field experiments, and recommends pilot investigations to assess potential environmental disruption and follow-up studies to detect and minimize persistent effects. Pilot studies may be used to determine the minimally effective doses of chemicals required for field experiments. If favourable, results from pilot studies of other manipulations such as mark/recapture, restraint of free-living animals and disturbances of and alterations to nests and offspring, including eggs, may be used in the Methods section of submitted papers to justify procedures that might otherwise be questioned by the Animal Care or Ethical Committee. Full details of the pilot studies themselves should be available for scrutiny by the relevant committee. Investigators should weigh the potential gain in knowledge from field studies against the adverse consequences of disruption for the animals used as subjects and also for other animals and plants in the ecosystem. When an experimental protocol requires that animals be removed from the population either temporarily or on a long-term basis, investigators should ensure that suffering or discomfort is minimized not only for the removed animals but for others dependent on them (e.g. dependent offspring). Removed individuals and their dependants should be housed and cared for appropriately. Sources of further information on field techniques are the books edited by Stonehouse (1978) and Amlaner & Macdonald (1980). Researchers carrying out translocations or reintroductions of animals into wild habitats should consult the IUCN (1995) Guidelines.

(b) Aggression, Predation and Intraspecific Killing

The fact that the agent causing harm may be another nonhuman animal does not free the experimenter from the normal obligations to experimental animals. Huntingford (1984), Elwood (1991), Bekoff (1993) and Bekoff & Jamieson (1996) discuss the ethical issues involved and suggest ways to minimize suffering. Wherever possible,
field studies of natural encounters should be used in preference to staged encounters. Where staged encounters are necessary, the use of models or video/film playback should be considered, the number of animals should be kept to the minimum needed to accomplish the experimental goals, and the experiments made as short as possible. Suffering can also be reduced by continuous observation with intervention to stop aggression at predefined levels, and by providing protective barriers and escape routes for the subjects.

(c) Aversive Stimulation and Deprivation as Motivational Procedures

Aversive stimulation, deprivation or restriction of resources can cause pain or distress to animals. To minimize suffering, the investigator should investigate whether there is an alternative reward strategy that could be used to motivate the animal in the study. If an alternative reward strategy is unavailable or the effect of aversive stimulation is itself of interest (e.g. in studies of fear, stress or pain), investigators should ensure that the levels of restriction, deprivation or aversive stimulation used are no greater than necessary to achieve the goals of the experiment. Alternatives to aversive stimuli and deprivation strategies include the use of highly preferred foods and other rewards that may motivate even satiated animals (Reinhart 1997; Laule 1999; Grandin 2000). Use of minimal aversive stimuli levels requires knowledge of the technical literature in the relevant area: quantitative studies of aversive stimulation are reviewed by Church (1971) and Ruschen (1986), and the behaviour of satiated animals is considered by Morgan (1974). Before deciding in favour of aversion or deprivation, investigators should consider consulting with animal care staff, laboratory animal scientists and veterinary surgeons experienced in working with animals in research settings. Practical animal motivation and training is a highly specialized skill. The failure of positive reinforcement methods to motivate an animal may reflect only the strategy or tactic used, the skill of the research staff and level of rapport with the animal. It does not necessarily indicate that it would be impossible for a skilled trainer to motivate the animal successfully with positive reinforcement methods. Further comments on reducing distress caused by motivational procedures are to be found in Lea (1979) and Moran (1975).

(d) Social Deprivation, Isolation and Crowding

Experimental designs that require keeping animals in overcrowded conditions, or that involve social disruption, deprivation or isolation, may be extremely stressful to the animals involved, and may adversely impact the behaviours being studied. Because the degree of stress experienced by the individual animal can vary with species, age, sex, reproductive condition, developmental history and social status, the natural social behaviour of the animals concerned and their previous social experience must be considered to minimize such stress (Shepherdson et al. 1998; Poole & Dawkins 1999).
If animals are procured by capture in the wild, this must be done using methods that minimize pain, distress and suffering, and must comply with any relevant legislation. Investigators who purchase animals from local trappers should not encourage methods of trapping that cause suffering, or methods that involve killing many individuals to obtain a few live specimens. Procurement practices also should not create a local market for culling wildlife for profit. Individuals of endangered species or populations should not be taken from the wild unless they are part of an active conservation programme. The investigator should ensure that those responsible for handling purchased, donated, or wild-caught animals en route to the research facilities are well qualified and experienced in the requirements of the species being transported, and that animals are provided with adequate food, water, ventilation, space and protection from wastes (IATA 2003). The stresses associated with transport should be identified and minimized. Animals should not be subjected to unduly stressful situations (e.g. excessive climatic conditions or sensory stimulation, prolonged food or water deprivation, aggression) during transport. Generally, only healthy animals that are able to withstand the rigours of transport should be transported. Young, dependent animals are not usually suitable for transport, but if transport is necessary, they should usually be transported only with the mother or equivalent. Special care, and additional regulations, may be relevant to the transport of pregnant animals. With particularly sensitive or social species, it may be necessary for a trained attendant or veterinarian to travel with the animal(s). Preconditioning animals to transport containers prior to shipment will reduce stress during loading and shipping. Furthermore, the investigator should carefully consider modes of transport, transport schedules (so as to reduce or eliminate layovers, unless rest periods are desirable), and shipping containers to ensure that they are suitable for the species being shipped. The relevant transport regulations for the species concerned (local, national and international) must be complied with.

8. HOUSING AND ANIMAL CARE

The researcher’s responsibilities extend to the conditions under which the animals are kept when not being studied, as well as during study. Caging conditions and husbandry practices must meet, at the very least, minimal recommended requirements of the country in which the research is carried out (see Appendix 1). Although these publications provide general guidance, there is evidence that housing animals in larger or more enriched conditions than specified in these minimal requirements improves not only animal welfare (Newberry 1995; Kessler & Turner 1999; Mason et al. 2001; Olsson & Dahlborn 2002; Olsson et al. 2003; Sherwin & Olsson 2004) but also the quality of science, including behavioural studies (Poole 1997; Wurbel 2001, 2002; Sherwin 2004). Normal maintenance of captive animals should incorporate, as much as possible, aspects of the natural living conditions deemed important to maximizing welfare and survival. Consideration should be given to providing biologically relevant enrichment features such as natural material, refuges, perches, dust baths and water baths (Reinhardt & Reinhardt 2002), although it should be borne in mind that some enrichment features can sometimes create welfare problems of their own, for instance by increasing levels of competition and aggression (e.g. McGregor & Ayling 1990; Haemisch & Gartner 1994; Barnard et al. 1996). Companions should be provided for social animals where possible, providing that this does not lead to suffering or injury. Frequency of cage cleaning should represent a compromise between the level of cleanliness necessary to prevent diseases and the amount of stress imposed by frequent handling and exposure to unfamiliar surroundings, odours and bedding.

Standard housing and care regimes established for the commonly used laboratory animals are not necessarily suitable for wild animals or for individuals of wild species born in captivity. Special attention may be required to enhance the comfort and safety of these animals. Investigators may wish also to consult the most recent guidelines available from relevant taxon-oriented professional societies (Appendix 2).

The nature of human–animal interactions during routine care and experimentation should be considered by investigators. Depending upon species, rearing history and the nature of the interaction, animals may perceive humans as conspecifics, predators or symbionts (Estep & Hetts 1992). Special training of animal care personnel can help in implementing procedures that foster habituation of animals to caretakers and researchers and minimize stress. Stress can also be reduced by procedurally training animals to cooperate with handlers and experimenters during routine husbandry and experimental procedures (Biological Council 1992; Reinhardt 1997; Laule 1999).

9. FINAL DISPOSITION OF ANIMALS

When research projects or teaching exercises using captive animals are completed, it may sometimes be appropriate to distribute animals to colleagues for further study or breeding, if permitted by local legislation. However, if animals are distributed, care must be taken to ensure that the same animals are not used repeatedly in stressful or painful experiments, and that they continue to receive a high standard of care. Animals should never be subjected to major surgery more than once unless it is an unavoidable and justifiable element of a single experiment. Except as prohibited by national, federal, state, provincial or local laws, researchers may release field-trapped animals if this is practical and feasible, especially if it is critical to conservation efforts. However, the researcher should assess whether releases into the wild might be injurious or detrimental both to the released animal and to existing populations in the area. Because of potential impacts on the genetic structure of local populations in the area, animals should be released only at the site where they were trapped (unless conservation efforts dictate otherwise), and only when their ability to survive in nature has not been impaired and when they do not constitute a health or ecological hazard to existing
populations. If animals are to be killed subsequent to a study, this should be done as humanely and painlessly as possible, and death should be confirmed before their bodies are destroyed. Carcasses of wild animals killed with lethal anaesthetics or other toxic substances should be disposed of in a manner that would prevent scavengers from suffering secondary toxicity. Experts should be consulted for advice on methods of euthanasia that are appropriate for the particular species being used. Additional information on euthanasia methods can be found in Close et al. (1996) and the AVMA Report on Euthanasia (AVMA 2001).

10. OBTAINING FURTHER INFORMATION

There are a number of organizations that provide publications and detailed information about the care and use of animals. These include the Canadian Council on Animal Care (1105-151 Slater Street, Ottawa, Ontario, KIP 5H3 Canada, http://www.nal.usda.gov/awic/), the Scientists’ Center for Animal Welfare (7833 Walker Drive, Suite 410, Greenbelt, MD 20770, U.S.A. http://www.scaw.com/), and the Universities Federation for Animal Welfare (The Old School, Brewhouse Lane, Wheathampstead, Hertfordshire AL4 8AN, U.K. http://www.ufaw.org.uk/). The Animal Welfare Information Center at the National Agricultural Library (Room 205, Beltsville, MD 20705, U.S.A. http://www.nal.usda.gov/awic/), and the Universities Federation for Animal Welfare (The Old School, Brewhouse Lane, Wheathampstead, Hertfordshire AL4 8AN, U.K. http://www.ufaw.org.uk/). The Animal Welfare Information Center at the National Agricultural Library (Room 205, Beltsville, MD 20705, U.S.A., http://www.nal.usda.gov/awic/) publishes a series of bibliographies on special topics, and can also provide individualized database searches for investigators on potential alternatives, including techniques for replacement with nonanimal models or alternative species, methods for reducing the total number of animals necessary to address the research question, and experimental refinements that can reduce pain and stress.

The Internet provides a wealth of information on animal care and welfare issues. Many of these are government Web pages. A good starting place is the National Institutes of Health Office of Laboratory Animal Welfare site (http://grants.nih.gov/grants/olaw/olaw.htm), which contains considerable policy information and many links (e.g. http://grants.nih.gov/grants/olaw/links.htm), from which one can gain access to the 1996 Institute of Laboratory Animal Resources (ILAR) Guide for Care and Use of Laboratory Animals (published by the National Academies Press), as well as information on the IACUC Guidebook published by ARENA (Applied Research Ethics National Association). Another good source is the USDA/APHIS Animal Welfare Information Center site (http://www.nal.usda.gov/awic/). Additional information is available on the APHIS site (http://www.aphis.usda.gov/ac). The Animal Welfare Information Center (AWIC) at the National Agricultural Library (NAL) can be reached via the USDA Agricultural Library, 5th floor, 10301 Baltimore Avenue, Beltsville, MD 20705. AAALAC International (Association for Assessment and Accreditation of Laboratory Animal Care) also has a home page (http://www.aaalac.org/) and a toll-free phone number (1-800-926-0066). Additional sources of information are Net Vet (http://netvet.wustl.edu/), the National Academy of Sciences (http://www.nas.edu/) and the National Academies Press (http://www.nap.edu/). The American Zoo and Aquarium Association also has guidelines and advice on the husbandry of various zoo and wildlife species in captivity (http://www.aza.org/).

Acknowledgment

C. M. Sherwin was in receipt of the UFAW Hume Research Fellowship during the revising of these Guidelines.

References


United Kingdom


Laboratory Animal Breeder’s Association, Charles River (U.K.) Ltd, Manson Research Centre, Manson Road, Margate, Kent CT9 4LP (Home Office, 1995).

United States of America


Information on licensed animal dealers can be obtained from the local office of the U.S. Department of Agriculture (U.S.D.A.), or The American Association for Laboratory Animal Science, 70 Timber Creek Drive, Suite 5, Cordova, TN 38018.

Europe

http://www.homeoffice.gov.uk/docs/revieweuropeandirective86_609_ec.html.

Canada


The Canadian Association for Laboratory Animal Science, M524 Biological Sciences Building, University of Alberta, Edmonton, Alberta T6G 3E9.

Australia


New Zealand


APPENDIX 2. TAXON-ORIENTED SOCIETIES WITH ETHICAL OR ANIMAL WELFARE GUIDELINES

Ornithological Council: http://www.nmnh.si.edu/BIRDNET/GuideToUse/.
American Society of Ichthyologists and Herpetologists (ASIH), the Herpetologists’ League (HL) and the Society for the Study of Amphibians and Reptiles (SSAR): http://www.asih.org/pubs/herpcoll.html.
American Society of Ichthyologists and Herpetologists (ASIH), the American Fisheries Society (AFS) and the American Institute of Fisheries Research Biologists (AIFRB): http://www.asih.org/pubs/fishguide.html.