INTRODUCTION

This handout contains basic information concerning IACUC protocol review and approval criteria contained in the Federal policies and regulations which govern the use of animals in research, training, experimentation, biological testing, or for related purposes (hereinafter referred to as activities). For more detailed information, the reader is referred to the Federal policies, regulations and guidelines listed below as well as the many existent lab animal welfare web sites and other available resources.

A. FEDERAL POLICIES, REGULATIONS AND GUIDELINES

1. What Federal policies, regulations and guidelines should the IACUC use during protocol review?

The criteria the IACUC should use when reviewing protocols are contained in the following:

- PHS Policy on Humane Care and Use of Laboratory Animals (Promulgated 1986, revised 1996, reprinted 2000) which includes the U.S. Government Principles for the Utilization and Care of Vertebrate Animals Used in Testing, Research and Training (USGP)
- Animal Welfare Act (USDA) Regulations (9 CFR 1,2,3, Promulgated 1989, 1991)
- USDA Animal Care Policies
- The institution’s Animal Welfare Assurance.
- The Guide for the Care and Use of Animals in Agricultural research and Teaching, 1999 (AgGuide)

NOTE: The PHS Policy at IV.1. requires assured institutions to base their animal care and use program on The Guide and comply with USDA regulations which apply to USDA covered species. Since Part 2 of the USDA regulations is reasonably consistent with the PHS Policy, it makes “ethical sense” and sound public policy for institutions to use one uniform standard of animal care and use.
2. **What species of animals are covered by the PHS Policy?**

   Any live (warm blooded or cold blooded) vertebrate animal to be used in research, research training and biological testing “activities”, which are PHS conducted, PHS supported or subject to the requirements of the PHS Policy and the institution’s Animal Welfare Assurance.

3. **What species of animals are covered by USDA**

   Any warm blooded animal to be used by a “research facility” in “activities,” as defined previously, except birds, rats (Rattus), mice (Mus) bred for use in research, and horses or other farm animals used or intended for use as food or fiber. “Research facility” means any school (except an elementary or secondary school), institution, organization or person who conducts “activities” involving animals covered by USDA regulations.

B. **SUBMISSION AND REVIEW PROCEDURES**

1. **Must all protocols be reviewed initially by the full IACUC at a convened meeting?**

   “Prior to the review, each IACUC member shall be provided with a list of proposed activities to be reviewed. Written descriptions of all proposed activities that involve the care and use of animals shall be available to all IACUC members, and any member of the IACUC may obtain, upon request, full committee review of those activities. If full committee review is not requested, at least one member of the IACUC, designated by the chairperson and qualified to conduct the review, shall review those research projects and have the authority to approve, require modifications in (to secure approval) or request full committee review of any of those activities. If full committee review is requested, approval of that activity may be granted only after review at a convened meeting of a quorum of the IACUC and with the approval vote of a majority of the quorum present. No member may participate in the IACUC review or approval of an activity in which the member has a conflicting interest...” PHS Policy at IV.C.2.; USDA 9 CFR 2.31(d)(2)

   *Note: The PHS Policy and USDA regulations require only that a list of proposed activities be provided to each IACUC member and that written descriptions be made available. However, in order for each IACUC member to effectively determine whether or not full committee review is warranted, sufficient information about the proposed activity must be provided. Some IACUCs provide each member with a copy of an abbreviated protocol whereas other IACUCs distribute the complete protocol.*
2. **Do some IACUCs use a classification system to determine which types of protocols qualify for review by the designated reviewer method versus those requiring full committee review?**

Some IACUCs have found it useful to categorize research protocols on the basis of invasiveness and the potential to cause animal pain, discomfort and distress. For example, protocols which may cause more than momentary pain, discomfort or distress are subject to full committee review at some institutions. There is, however, no regulatory requirement for this type of review system. And, even if an IACUC categorizes protocols in this way, written descriptions must still be made available to all IACUC members and any member may still request full committee review of any animal related activity.

3. **Is a protocol application form useful?**

Most IACUCs utilize protocol application forms which instruct the investigator on what information must be provided. A well-constructed form helps facilitate IACUC review.

4. **How should the IACUC review proposed changes related to the care and use of animals in on-going activities?**

The IACUC is required by the PHS Policy and USDA regulations to review proposed significant changes regarding the use of animals in on-going activities. PHS Policy at IV.C.1.; USDA 9 CFR 2.31 (B)(7).

*Note: Proposed “significant changes” should be reviewed using the same criteria used during initial review. While the term “significant” is not defined in the PHS Policy or USDA regulations, the Office for the Protection from Research Risks (OPRR), now known as the Office of Laboratory Animal Welfare (OLAW), has provided the following examples as guidance for IACUCs: changes in the objectives of a study; changes in the degree of invasiveness; changes in species; changes in anesthetic agent(s) or methods of euthanasia ... These examples, however, are not meant to be inclusive and each institution should establish a written policy concerning what constitutes a “significant change.” (Lab Animal 24(9):24-26, 1995.)*

5. **How often must the IACUC conduct continuing review of previously approved protocols?**

   a. “The IACUC shall conduct continuing review of each previously approved, ongoing activity covered by this Policy at appropriate intervals as determined by the IACUC, including a complete review in accordance with IV.C.1-4 at least once every three years.” PHS Policy At IV.C.5.
b. “The IACUC shall conduct continuing review of activities covered by this subchapter at appropriate intervals as determined by the IACUC but not less than annually.” USDA 9 CFR 2.31(d)(5)

NOTE: The NIH/OLAW interprets triennial continuing review to be a de novo process which must meet all new proposal review criteria. It is also considered a monitoring process to determine that the study remains in compliance. APHIS also interprets continuing review to be a monitoring process to determine that the study remains in compliance. USDA continuing review requirements are not, however, as comprehensive as the PHS requirements for triennial continuing review. However, the PI must reconsider alternatives at least once every three years and the IACUC should review this reconsideration (i.e. the written narrative) as part of triennial review. [Contemporary Topics, 35(5) 1996; pgs. 53-56; ILAR News 33(4):68-70, 1991; USDA Policy 12]

6. Is the IACUC required to notify the investigator and the institution in writing of its decisions?

“The IACUC shall notify investigators and the institution in writing of its decision to approve or withhold approval of those activities related to the care and use of animals, or of modifications required to secure IACUC approval. If the IACUC decides to withhold approval of an activity, it shall include in its written notification a statement of the reasons for its decision and give the investigator an opportunity to respond in person or in writing.” PHS Policy at IV.C.4.; USDA 9 CFR 2.31(d)(4)

NOTE: USDA uses the term “research facility” instead of “institution.”

NOTE: The written notification to investigators, i.e., the IACUC review letter, should contain a clear rationale which supports the IACUC’s decision regarding clarification(s) and/or change(s) which the investigator must address before final approval can be granted. The IACUC review letter can serve as an excellent educational tool and foster a positive relationship between the committee and the investigator.

7. Under what circumstances may an IACUC suspend an activity it previously approved?

a. “The IACUC may suspend an activity that is previously approved if it determines that the activity is not being conducted in accordance with applicable provisions of the Animal Welfare Act, the Guide, the institution’s Assurance, or IV.C.1.a.-g. of this Policy. The IACUC may suspend an activity only after review of the matter at a convened meeting of a quorum of the IACUC and with the suspension vote of a majority of the quorum present.” PHS Policy at IV.I.C.6.
b. “If the IACUC suspends an activity involving animals, the Institutional Official in consultation with the IACUC shall review the reasons for suspension, take appropriate corrective action, and report that action with a full explanation to OLAW.” PHS Policy at IV.C.7.

c. “The IACUC may suspend an activity that it previously approved if it determines that the activity is not being conducted in accordance with the description of that activity provided by the principal investigator and approved by the Committee. The IACUC may suspend an activity only after review of the matter at a convened meeting of a quorum of the IACUC and with the suspension vote of a majority of the quorum present.” USDA 9 CFR 2.31 (d)(6).

d. “If the IACUC suspends an activity involving animals, the Institutional Official, in consultation with the IACUC, shall review the reasons for suspension, take appropriate corrective action, and report that action with a full explanation to APHIS and any Federal agency funding that activity.” USDA 9 CFR 2.31(d)(7).

C. PROTOCOL REVIEW QUESTIONS AND IACUC APPROVAL CRITERIA

1. What is the potential relevance and value of the activities involving animals to human or animal health, the advancement of knowledge or the good of society?

IACUC Approval Criteria

a. “Procedures involving animals should be designed and performed with due consideration of their relevance to human or animal health, the advancement of knowledge, or the good of society.” USGP II.

b. “A proposal ... must contain a rationale for involving animals ... “ USDA 9 CFR 2.31 (e)(2)

c. “A proposal ... must contain a description of procedures designed ... for the conduct of scientifically valuable research ...” USDA 9 CFR 2.31 (e)(4)

NOTE: The PHS Policy requires the IACUC to consider criteria which reflect “scientific merit review”, e.g., “relevance” (USGP II.); “consistent with sound research design” (Section IV.C.1.a.); “conduct of scientifically valuable research” (Section IV.D.1.d); and “valid results” (USGP III.) While these criteria presume the IACUC will consider “general scientific relevance” this should not be construed as the equivalent of peer review by a PHS funding component. USDA regulations are far less specific than the PHS Policy with regard to scientific merit review. The comment section of the regulations implies the IACUC should not be involved in reviewing scientific merit (FR 54, No. 168, p. 36114). However, the USDA Regulations, like the PHS Policy, use the term...
2. Is the research entirely new or is there duplication of previous experiments? If there is duplication, why?

**IACUC Approval Criteria**

“The principal investigator must provide written assurance that the activities do not unnecessarily duplicate previous experiments.” USDA 9 CFR 2.31(d)(iii)

**NOTE:** *PHS Policy does not directly address the issue of “unnecessary duplication.” However, section IV.D.1.d. and USGP II. clearly imply that activities involving animals must not be unnecessarily duplicative.*

3. What species will be used in the activity and what is the rationale for selecting the species?

**IACUC Approval Criteria**

a. “The animals selected for a procedure should be of an appropriate species and quality ... to obtain valid results.” USGP III.

b. “A proposal ... must contain ... identification of the species ... and a rationale for the appropriateness of the species ...” 9 CFR 2.31(e)(l)(2)

**NOTE:** *Selection of the species should be based upon anatomical, physiological or other biological characteristics of the species in consideration of the scientific objectives and the need to relate study data to already existent relevant data derived from previous research using the same species.*

4. What is the justification for the required number of animals based upon scientific and statistical considerations? Is the required number of animals minimized to the greatest extent possible?

**IACUC Approval Criteria**

a. “The number of animals selected ... should be the minimum number required to obtain valid results.” USGP III.

b. “A proposal ... must contain ... the approximate number of animals to be used ... and a rationale for ... numbers of animals to be used.” USDA 9 CFR 2.31 (e)(1)(2)
NOTE: USDA regulations do not directly address the issue of “minimization” of animal numbers, but clarification is contained in USDA Animal Care Policy #12 which requires the investigator to consider the principle of “reduction.” Many IACUCs require the investigator to justify animal numbers using scientific and statistical considerations.

5. What are the animals’ living conditions? Are the living conditions appropriate for the species? Will the living conditions contribute to the health and comfort of the animals?

**IACUC Approval Criteria**

a. “The living conditions of animals will be appropriate for their species and contribute to their health and comfort. The housing, feeding, and nonmedical care of the animals will be directed by a veterinarian or other scientist trained and experienced in the proper care, handling, and use of the species being maintained or studied.” PHS Policy at IV.C.d.; USGP VII.

b. “The animals’ living conditions will be appropriate for their species in accordance with Part 3 of this subchapter and contribute to their health and comfort.” USDA 9 CFR 2.31(d)(vi)

6. What procedures will be performed on animals? Which, if any, procedures will cause pain, discomfort or distress? Are procedures designed to avoid or minimize pain, discomfort or distress to the animals? Are there any suitable non-animal alternatives or other alternatives to procedures which may cause pain, discomfort or distress which will have less of an adverse effect on the animal?

**IACUC Approval Criteria**

a. “A proposal ... must contain a complete description of the proposed use of the animals.” USDA 9 CFR 2.31(e)(3)

b. “Procedures with animals will avoid or minimize discomfort, distress, and pain to the animals, consistent with sound research design.” PHS Policy at IV.C.I.a.

c. “Proper use of animals, including the avoidance or minimization of discomfort, distress, and pain when consistent with sound scientific practices, is imperative. 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.” USGP IV.
d. “A painful procedure is defined as any procedure that would reasonably be expected to cause more than momentary pain and/or distress in a human to which that procedure is applied ...” “Examples of procedures that can be expected to cause more than momentary or slight pain include, but are not limited to the following: terminal surgery ...; Freund’s complete adjuvant used for antibody production ...; ocular and skin irritancy testing ... .” USDA Animal Care Policy #11, April 14, 1997.

e. “A proposal ... must contain a description of procedures designed to assure that discomfort and pain to animals will be limited to that which is unavoidable for the conduct of scientifically valuable research ...” USDA 9 CFR 2.31(e)(4)

f. “The Principal Investigator has considered alternatives to procedures that may cause more than momentary or slight pain or distress to the animals and has provided a written narrative description of the methods and sources ... used to determine that alternatives were not available.” USDA 9 CFR 2.31(d)(ii)

g. “The minimal written narrative should include: the databases searched and/or other sources consulted (e.g. named experts, conferences, etc.), the date of the search, the years covered by the search and the key words and/or search strategy. Reduction, replacement, and refinement (the “3 R’s”) must be addressed, not just animal replacement.” USDA Animal Care Policy #12, June 21, 2000

h. “… Methods such as mathematical models, computer simulation, and in vitro biological systems should be considered.” USGP III.

i. “Procedures that may cause more than momentary or slight pain or distress to the animals will be performed with appropriate sedation, analgesic, or anesthesia, unless the procedure is justified for scientific reasons. ...” PHS Policy at IV.C.I.b.

j. “Procedures that may cause more than momentary or slight pain or distress ... will be performed with appropriate sedatives, analgesics or anesthetics unless withholding such agents is justified for scientific reasons ...” USDA 9 CFR 2.31(d)(iv)(A)

k. “Procedures that may cause more than momentary or slight pain or distress to the animals will involve, in their planning, consultation with an attending veterinarian or his or her designee.” USDA 9 CFR 2.31 (d)(iv)(B)

l. “Procedures with animals that may cause more than momentary or slight pain or distress should be performed with appropriate sedation, analgesia, or anesthesia. Surgical or other painful procedures should not be performed on unanesthetized animals paralyzed by chemical agents.” USGP V.
m. “Procedures that may cause more than momentary or slight pain or distress to the animals will not include the use of paralytics without anesthesia.”
USDA 9 CFR 2.31 (d)(iv)(c)

NOTE: Both the PHS Policy and USDA regulations require “minimization” of pain, discomfort and distress to the animals within justifiable scientific constraints. This requires a diligent application of the principles of replacement, reduction and refinement (the 3 Rs). USDA, however, also requires the investigator to provide a written narrative as described in Section 2.31 (d) (ii) of the regulations and USDA Animal Care Policy #12 which addresses the methods and sources used to determine that alternatives to the use of live animals and/or painful procedures are not available or feasible.

The following is an example of a template for the required written narrative:

“I have performed the following database searches: (Insert database titles and key words used). These searches were performed on (insert date) and cover the period of (insert years). Based on (insert number) years of experience in this field, in conjunction with periodic consultation of bibliographic sources (insert source titles) and a number of references, including (insert major journal titles), I believe there is no alternative to performing this potentially painful/distressful procedure in order to achieve the scientific objectives of this research. Therefore, based on the aforementioned references and experience, this protocol utilizing (insert species) is the most appropriate for conducting my research.”

8. Will major and/or minor survival and/or non-survival surgery be performed? What is the pre-operative regimen? What is the anesthetic regimen? What is the frequency and duration of post-operative care and monitoring? What is the post-operative analgesic regimen?

IACUC Approval Criteria

a. “All survival surgery will be performed using aseptic procedures, including surgical gloves, masks, sterile instruments and aseptic techniques.”
USDA 9 CFR 2.31 (d)(ix)

b. “Major operative procedures on non-rodents will be conducted only in facilities intended for that purpose ... non-major operative procedures and all surgery on rodents do not require a dedicated facility but must be performed using aseptic procedures.” USDA 9 CFR 2.31(d)(ix)

“Major survival surgery penetrates and exposes a body cavity or produces substantial impairment of physical or psychological functions (such as laparotomy, thoracotomy, craniotomy, joint replacement, and limb amputation. Minor survival surgery does not expose a body cavity and
causes little or no physical impairment (such as would suturing; peripheral-
vessel cannulation; ... castration ... .” The Guide, Pgs. 61-62.

c. “No animal will be used in more than one major operative procedure from
which it is allowed to recover unless justified for scientific reasons ... or is
required to protect the health and well being of the animal as required by the
attending veterinarian ...” USDA 9 CFR 2.31(d)(x)

NOTE: For further guidance regarding multiple major survival operative
procedures, refer to USDA Animal Care Policy #14, April 14, 1997.

d. “It is important ... to ensure that good surgical technique is practiced, that is,
asepsis, gentle tissue handling, minimal dissection of tissue, appropriate
use of instruments, effective hemostasis, and correct use of suture
materials ... “ The Guide, pgs. 60-64.

e. “Procedures with animals that may cause more than momentary or slight
pain or distress should be performed with appropriate sedation, analgesia,
or anesthesia. Surgical or other painful procedures should not be
performed on unanesthetized animals paralyzed by chemical agents.”
USGP V.

f. “Activities that involve surgery include appropriate provision for pre-
operative and post-operative care ... in accordance with established
veterinary ... practice.” USDA 9 CFR 2.31(d)(ix)

NOTE: For further guidance regarding pre- and post-procedure care refer to
USDA Animal Care Policy #3, April 14, 1997.

device(s) will be used? How are animals conditioned to the restraint
device? What is the duration of the restraint? Is the restraint time
minimized as much as possible? How often are animals observed?

IACUC Approval Criteria

Physical restraint procedures should be used on awake animals only after
alternative procedures have been considered and found to be inadequate. The
restraint device must provide the animal with the opportunity to assume its
normal postural adjustments. The animal should be conditioned to the device,
the duration of restraint must be minimized, and the animal observed at
appropriate intervals. The Guide, Pg. 11
10. What method(s) of euthanasia will be used? What is the rationale for selecting the method(s) of euthanasia? Is the method(s) of euthanasia the most humane possible given the scientific constraints? How will death be confirmed? If the animal will not be euthanized, what will be its disposition?

a. “Methods of euthanasia used will be consistent with the recommendations of the American Veterinary Medical Association (AVMA) Panel on Euthanasia, unless a deviation is justified for scientific reasons in writing by the investigator.” PHS Policy at IV.C.I.g.

b. “Methods of euthanasia used must be in accordance with the definition of the term set forth in 9 CFR Part 1 ... unless a deviation is justified for scientific reasons ...” Euthanasia means, “the humane destruction of an animal accomplished by a method that produces rapid unconsciousness and subsequent death without evidence of pain or distress, or a method that utilizes anesthesia produced by an agent that causes painless loss of consciousness and subsequent death.” USDA 9 CFR 2.31(d)(xi)

c. “The methods of euthanasia must be consistent with the current report of the AMVA Panel on Euthanasia ...” USDA Animal Care Policy #3, April 14, 1997.

d. “Euthanasia is the act of killing animals by methods that induce rapid unconsciousness and death without pain or distress. Unless a deviation is justified for scientific or medical reasons, methods should be consistent with the 1993 Report of the AVMA Panel on Euthanasia (AVMA 1993 or later editions). In evaluating the appropriateness of methods, some of the criteria that should be considered are ability to induce loss of consciousness and death with no or only momentary pain, distress, or anxiety; reliability; nonreversibility; time required to induce unconsciousness; species and age limitations; compatibility with research objectives; and safety of and emotional effect on personnel.” The Guide, Pg. 65.

e. “Death should be confirmed by ... cessation of vital signs in the species being euthanized.” The Guide, Pg. 66.

11. What is the criteria for premature euthanasia?

**IACUC Approval Criteria**

a. “Animals that would otherwise suffer severe or chronic pain or distress that cannot be relieved should be painlessly killed at the end of the procedure or, if appropriate, during the procedure.” USGP VI.
b. “Animals that would otherwise experience severe or chronic pain or distress that cannot be relieved will be painlessly euthanized at the end of the procedure or, if appropriate, during the procedure.” USDA 9 CFR 2.31(d)(v)

c. “Protocols should include criteria for initiating euthanasia, such as a physical or behavioral defect or tumor size, that will enable a prompt decision to be made by the veterinarian and the investigator to ensure that the end point is humane and the objective of the protocol is achieved.” The Guide, Pgs. 65-66.  

NOTE: The investigator should choose criteria for premature euthanasia which reflect the earliest possible signs of pain, discomfort, distress and morbidity which cannot be treated. The principle of “refinement” is particularly applicable when deciding when the condition of an animal warrants euthanization.

12. Who are the scientists, research technicians and other personnel involved in the care and use of animals? What are their assigned responsibilities? Are they qualified by training and experience?

a. “Personnel conducting procedures on the species being maintained or studied will be appropriately qualified and trained in those procedures.” PHS Policy at IV.C.f.; USDA 9 CFR 2.31(d)(viii)

b. “Investigators and other personnel shall be appropriately qualified and experienced for conducting procedures on living animals. Adequate arrangements shall be made for their in-service training, including the proper and humane care and use of laboratory animals.” USGP VIII.

c. “Investigators, technical personnel, trainees, and visiting investigators who perform animal anesthesia, surgery, or other experimental manipulations must be qualified through training and experience to accomplish these tasks in a humane and scientifically acceptable manner.” The Guide, Pg. 14.

13. Will any personnel be exposed to occupational health hazards as a consequence of conducting activities involving animals? If so, what are the hazards? How are risks minimized and personnel protected?

a. “The IACUC must be satisfied there is an effective occupational health and safety program which ensures that the risks associated with the experimental use of animals are reduced to acceptable levels and that personnel are appropriately trained.” The Guide, Pgs. 14-15.

NOTE: Many institutions have safety committees which review protocols involving animals for considerations related to occupational health and convey the results of their reviews to the IACUC.