The IACUC Proposal Form – Key Features

Although efforts to completely revise the “Proposal to Use Laboratory Animals in Research and Teaching,” which will likely be re-named as well, are well underway, this Information Sheet is meant to serve as a resource for Principal Investigators’ completing and Committee members’ reviewing the existing form.

**Hint #1: Read the instructions.** They contain important information not only about the responsibilities of the PI, but also specific instructions for several of the key questions. No, really, go ahead and read the instructions (then delete them from the submission)!

**Section V.A. Description of Experimental Groups.**
This section requests a brief description of the experimental groups. So that the committee can understand what will happen to each animal requested, please provide an outline (a table or flowchart is recommended) of each experimental group. For each group, list the species used (one species per group), a few-word description of each procedure performed (e.g., phlebotomy, test article A injection, echocardiogram, survival surgery-subcutaneous implantation, non-survival surgery blood pressure measurement, euthanasia, tissue collection), the time line of each procedures, and the number of animals for each group in each experiment.

**Hint #2:** Don’t just cut-and-paste your Specific Aims from a grant application. Although that might be a good start, it probably doesn’t adequately share with the reviewers the sequence of events that will occur with each experimental group.

**Hint #3:** Describe breeding colonies as they relate to production of animals needed for experiments listed above. Don’t forget donor animals and small number of animals needed for dose determination, new staff training, and pilot studies, if needed.

**Hint #4:** If this is a 3-year renewal, the Proposal will benefit from a brief description of findings-to-date.

**Hint #5:** If animal use is limited to tissue donation for in vitro studies, a brief description of the in vitro studies will assist the reviewer in understanding the number of animals needed. Note that a number of studies per time period is rarely appropriate; rather, your description should suggest how many different in vitro experiments are planned in the time frame of the Proposal (maximum 3 years) and the number of animals needed per experiment.

**Section V.B. Assurance of Non-Necessary Duplication.**
This is somewhat intuitive, but regulations require that you provide a written description of how you determined that the work has not been performed before or, if duplicating others’ work, why repeating studies are necessary. When using literature databases to support your assurance (which is strongly recommended), USDA Policy 12 requires that you list: a) the database (more than one is recommended), b) the date the search was performed, c) the keywords used, and d) the years/range included in the search.

**Section V.C. Use of Animals.**
Responses to this question are also usually intuitive. However, the Guide (2011) also states that “the IACUC is obliged to weigh the objectives of the study against potential animal welfare concerns.” In this section, you should address a “harm-benefit analysis” that compares potential animal welfare concerns with anticipated gains in knowledge leading to benefits to human and/or animal health and well-being.

**Section VI. Project Participants.**
See the following two IACUC Policies:
- **Required Training**
**Individual Risk Assessment and Medical Surveillance**

**Hint #6:** Describe each individual’s role and experience in this study, i.e. who will be performing each procedure and what is their specific experience in performing such procedures – such as drug administration, observations during behavioral experiments, post-operative monitoring, and euthanasia. This is especially important for personnel who have little or no experience with laboratory animals -- how, and by whom, will they receive training relative to his activities in this project?

**Section VII. Hazardous Agents.**
IACUC approval is contingent upon the approval by the appropriate safety unit. A written Special Animal Safety Protocol (SASP) may be required. This will be determined by the DEHS (chemicals), IBC (biological agents), and/or RSO (radioactive materials, radiation, and lasers). Use care in listing all potentially hazardous substances in this section. Otherwise, the concurrent review by the appropriate safety unit will be delayed. See the following two IACUC Policies:

- **Hazardous Chemical Review and Approval**
- **Biological Hazard Review and Approval**

**Subsection C. Personnel Risks.** Don’t go into great detail. If a Special Animal Safety Protocol (SASP) will be required, simply state that fact with an assurance that it will be followed.

**Section VIII. Study Site.**
See the following two IACUC Policies:

- **Transporting Animals to Research Laboratories**
- **Animal Security in Investigator Laboratories**

**Hint #7:** For animal and occupational health reasons, the IACUC is becoming more and more concerned with the transport and use of animals in laboratories. Please describe which components of the study MUST be performed in your laboratory and what components may be completed within the procedural areas of the centralized holding facilities.

**Hint #8:** As instructed, copy and complete the entire **Section IX and X for each species employed.** Failure to do so will likely result in the Proposal returned to you without review.

**Section IX.C. Justification of Animal Numbers.**
A description of the experimental groups should have been included in Section V.A. Therefore, in this section you only need to provide a specific rationale for your selection of the number of animals per group (“N”). Statistical calculation given a known or expected error rate and difference between groups is preferred, but experience with the model and/or literature may be deemed acceptable.

**Hint #9:** Subdivide your description into categories such as “Animals Used in Experiments,” “Breeding Colony,” “Animals for *In Vitro* Studies,” “Pilot Studies,” “Animals Used for Teaching/Training,” etc.

**Hint #10:** For a more thorough description of justifying animal numbers, including for breeding colonies, see the Information Sheet entitled, “Animal Number Justification.”

**Section IX.D. Special Housing and Care.**
This section should describe any housing or husbandry practices that are different from RRF standard procedures, e.g., use of special diets, a need to avoid environmental enrichment devices, altered light cycles, and increased cage density. Especially important are descriptions of the rationale and monitoring when food or fluids are restricted or regulated, movement is restricted (e.g., use of tethers), or enclosures smaller than those recommended by the Guide or USDA Animal Welfare Regulations are employed.

See also the following two IACUC Policies:

- **Social Housing of Animals.** If the conduct of the study requires individual housing, you should provide a justification here. Note that there are a few exceptions to social housing for which you do not need specific IACUC approval (and therefore an explanation here), such as post-surgery (unless for more than 14 days), rodents in breeding colonies, attrition of established research groups, or acquisition of single animals.

- **Laboratory and Satellite Rodent Housing.** If you plan to take animals to a non-RRF location and keep them there for more than 12 hours, a detailed justification must be included in this section. Please note that leaving animals in a laboratory overnight for monitoring after surgery is often not justifiable unless someone plans to stay all night with them. Biosecurity, physical security, and environmental control is much better within the RRF.
Section X.C. Alternatives to Procedures Potentially Causing Pain and/or Distress.
This one also warrants its own Information Sheet, aptly entitled, “Consideration of Alternatives.” Check it out.

Section X.D.1. Non-Surgical Procedures.
In this section you should provide a step-by-step description of procedures that do not involve surgery, e.g., blood or other non-surgical tissue collection, test article administration, behavioral assays, etc. Procedures should be documented in records available to the IACUC (see: Individual Animal Records).

Hint #11: For breeding colonies, describe:
1) Breeding techniques employed. Note that the RRF and IACUC strongly encourage pair-mating. If harem (2 females with one male) is required, pregnant females should be relocated as soon as possible. Similarly, in cages where a post-partum mating is allowed or encouraged, litters should be weaned as soon as possible after 21 days; at no time should two litters be allowed to remain in a single cage.
2) Disposition of those animals not deemed genetically useful (presumably euthanatized).
3) Adjunct procedures such as tissue collection for genotyping and animal identification methods.

See also the following IACUC Policies:
- Rodent Breeding Colonies
- Tissue Harvesting for Rodent Genotyping
- Rodent Identification

Hint #12: For tumor studies:
1) All transplantable tumors that may have been exposed to mice, rats, or murine tissues (e.g., serum) must be tested for potential contamination with adventitious murine viruses (see: Testing of Cell Lines and Other Biological Materials for Rodent Pathogens).
2) Tumor implantation sites must be chosen to minimize damage to normal structures. Sites involving special senses and intramuscular implantation should be avoided. Subcutaneous or intradermal implantation in the flank is considered least painful and is recommended. Tumors should not exceed 10% of the animal's total body weight. Tumors should not be allowed to grow so large that they interfere with the animal's normal physiologic functions. Animals should be euthanatized before tumors ulcerate. Experimental end points should be clearly identified in the animal care and use protocol, such as tumors size, time post-implantation, or other well-defined parameters. Simply observing animals for clinical evidence of pain or distress is discouraged and requires justification. Investigative staff should observe the animals frequently (at least daily) after tumor implantation.

Hint #13: When describing the use of test substances, drugs, or other agents to be administered to animals, provide the dose in mg/kg, total volume (mL), route (e.g., IV, PO, IM, SC), site (location on the animal), and frequency (number of times per day, number of total doses, time interval between doses, etc.). Recall that pharmaceutical-grade agents, including research test articles, should be employed if available. Use of non-pharmaceutical-grade drugs must be specifically described and justified. For each such product, consideration should be given to the quality of the material (grade, purity, etc.), methods of ensuring sterility and physiological compatibility (pH, pyrogenicity, osmolality, etc.), expected shelf-life (stability, expiration/discard timeframe that will be used), site and route of administration, and effectiveness (pharmacokinetics, etc.) of the chemical or substance to be administered, as well as any potential animal welfare and scientific issues relating to its use.

Hint #14: When describing blood collection, provide site (e.g., tail, facial, auricular, cephalic, jugular vein), volume collected (mL), frequency (number of times per day, time interval between collections, etc.), and total volume withdrawn.

Section X.D.2. Prolonged Physical Restraint.
Brief physical restraint for administering drugs or tissue collection is not considered “prolonged,” which has been defined by the IACUC as greater than 15 minutes for rodents or 30 minutes for other mammals (see: Prolonged Physical Restraint).

Section X.D.3. Methods of Avoiding Distress.
Describe all potential adverse effects of the procedures above. Outline any objective signs or criteria that will be used to determine humane endpoints (that is, signs that should they occur indicate pain/distress levels warranting...
euthanasia regardless of experimental timelines). Often, basic “sick animal” signs such as inappetance or lethargy lasting over 24-48 hours or weight loss exceeding 10% are used. Other signs/criteria, such as Body Condition Indices, may be more appropriate. Note that death is not an acceptable experimental end point unless clear and detailed justification is provided to the IACUC.

**Hint #15**: When using genetically-engineered rodents, also include a description of any adverse effects of the desired phenotype and means by which the laboratory will respond to such effects (e.g., if the transgene produces diabetes mellitus, a description of how hyperglycemia will be monitored and treated; if the transgene produces partial blindness, a description of how the laboratory will support the animals). This may include alterations in standard husbandry (e.g., providing food on the floor, supplements to drinking water, extra bedding or enrichment) if the ability to reach food/water or ambulate is impacted.

**Section X.E. Surgery.**
See: Performing Rodent Survival Surgery

For projects involving multiple survival surgery, describe why the two procedures could not be performed during the same anesthetic period. Also specifically state the time interval between surgeries, and any additional health monitoring necessary.

**Section X.E.2. Methods of Avoiding Pain and Distress.**
Describe all potential adverse effects of the surgical procedures, including a description of humane endpoint criteria similar to Section X.D.3.

**Hint #16**: While use of appropriate anesthetics and analgesics should be included in this section, avoid including the dose, route, and duration. This precludes conflicts with that information that must be provided in Section X.F. Don’t forget the “at least 48-hour rule” for providing post-operative analgesics following major survival surgery (see: Use of Postoperative Analgesia Following Major Survival Surgery).

**Section X.E.3. Aseptic Technique.**
See the following two IACUC Policies:

- Performing Rodent Survival Surgery
- Use of Pharmaceutical-Grade Medications and Outdated Drug Supplies

**Hint #17**: The IACUC recently revised the policy “Performing Rodent Survival Surgery,” which now includes tables or recommended disinfectants, sterilants, and suture materials, and a check-list for preparation of the surgical area, instruments, patient, and surgeon.

**Section X.E.3. Personnel Involved and Recordkeeping.**
Records must be kept that document anesthetic use (including dose administered), procedures performed, anesthetic monitoring, complications and emergency procedures, analgesic use, post-operative monitoring, and suture/wound clip removal. Note that records should document absence of reflexes prior to surgery and periodically throughout the procedure. Special means of assuring anesthetic depth is required when using neuromuscular-blocking agents. “Performing Rodent Survival Surgery” also contains a template form for recordkeeping. Date(s) of surgical procedures must also be indicated on rodent cage cards. For non-rodent mammals, all animal observations and procedures must be documented within Individual Animal Records, as outlined in the aptly-named Policy, Individual Animal Records.

**Section X.F. Anesthetics and Analgesics.**
See the following two IACUC Policies:

- Use of Pharmaceutical-Grade Medications and Outdated Drug Supplies. Include a scientific justification of why pharmaceutical-grade medications are not appropriate for the study. If not available, you should at least consider a drug for the same purpose that is available in a pharmaceutical-grade. Note the other considerations for using non-pharmaceutical-grade drugs in **Hint #12** above. This is also applicable to Section X.G. Euthanasia.
- Use of Postoperative Analgesia Following Major Survival Surgery.pdf

**Remarks**: Ironic that the “helpful hints” approaches the length of the form itself! But it’s hoped that this document will assist you in collating (and reviewing) the Proposal, with the ultimate aim of providing sufficient information for the IACUC to readily understand, and thereby more efficiently authorize requests for animal use.