RATIONALE FOR ANIMAL USE

Justification of Animal Numbers

Guidelines and Suggestions

A. Why a Justification for the Number of Animals is Requested

To provide IACUC with a scientific basis for decisions made by an investigator regarding the numbers of animals to be used in a research protocol.

The justification must be clear to IACUC reviewers and to informed members of the public. Therefore this justification should be written in more understandable, non-scientific terms but without jeopardizing its statistical soundness. In addition, each new completed form must be “stand alone,” so that a reviewer to understand the justification does not have to refer to other active protocols or to previously submitted Animal Subjects Approval Forms.

B. Sections of the Animal Subjects Approval Form (ASAF) Relevant to the Justification of Animal Numbers:

**ASAF III.B and III.C**

1. How many animals will be needed for this project?
2. How many experimental groups, replications, trials, etc. are required?
3. How did you determine that sample size, number of groups, replications, trials, etc. as they relate to the total number of animals requested?

C. SOME GENERAL GUIDELINES AND ASSISTANCE FOR DETERMINING THE NUMBERS OF ANIMALS TO USE IN AN EXPERIMENT

The following are intended to help researchers prepare acceptable justification statements that explain why a specific number of animals is needed. [The choice of species must be justified elsewhere (ASAF III.A).] Also included here are some suggested statement templates that, once completed, can be inserted into the Animal Subjects Approval Form.

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1 These suggestions should not replace recommendations made by a statistical consultant.
Certainly, new and novel experiments often do not enjoy the benefits of previous work that would provide the statistical basis for determining animal numbers. However, a thoughtful and demonstrable effort *must* be made to calculate and scientifically justify the numbers of animals used.

In general, there are two experimental approaches involving animals. In the first (C), animals are subjected to different treatments, and these treatment effects are statistically evaluated. In the second (D), animals indirectly aid or contribute to the study of important biological or medical phenomena, or serve in a teaching environment. The following sections discuss ways in which to apply a rationale justification to each type of experimental design.

**1. Treatment Effect**

   a) Start with reference to the experimental design discussed elsewhere in the ASAF, which itself should justify the numbers of *groups* and the treatments each group receives. The number of these treatment groups is usually driven by the scientific rationale for the study.

   b) With groups identified, then use statistical calculations to justify the minimum *numbers of animals* needed in each group to yield scientifically credible results. The number of animals in each treatment group is usually driven by statistical estimates of numbers needed (see below).
With such rationale and statistical justifications set forth, Summary Tables can be very helpful. For example:

An example—A plan for animal use (by treatment and time of sampling)

<table>
<thead>
<tr>
<th>Sample of table, which summarizes plan for animal use.</th>
<th>20 min post injection</th>
<th>40 min post injection</th>
<th>60 min post injection</th>
</tr>
</thead>
<tbody>
<tr>
<td>Treated Group</td>
<td>n=4</td>
<td>n=4</td>
<td>n=4</td>
</tr>
<tr>
<td>Control Group</td>
<td>n=4</td>
<td>n=4</td>
<td>n=4</td>
</tr>
<tr>
<td>Total</td>
<td>8</td>
<td>8</td>
<td>8</td>
</tr>
</tbody>
</table>

Total number of animals requested = 24 + 5 [for training, or other purposes (explain)] = 29.

Acceptable Statistical Approaches:

(1) Previous Research Experience

Pilot studies or previous research involving similar experimental design can form the basis for developing a statistical estimate of animal numbers needed on new ASAFs.

(2) Power Analysis

Arguments for doing a power analysis include the following:

- Assure that probability of missing an important difference is small
- Reduce cost of experiment
- Reduce wastage/suffering of animals

Resources available for doing a power analysis:

- Consult statistician: Department of Statistics (5-8645)

Requisites for doing a power analysis:
• Selection of appropriate statistical method for analyzing data.
• Specification of null and alternative hypotheses.
• Specification of minimal effect size that should be detectable (or biologically significant).
• Estimation of “nuisance” parameters (often the variance is not of interest and must be estimated by pilot data or by using crude relation).
• Significance level (\( \alpha \); chance of rejecting null hypothesis when null is correct) must be specified. Typically \( \alpha = 0.05 \) is used.
• Power level must be specified (80% is usually acceptable).

What if my power calculation is too low? How to Increase Power:

• Make significance level bigger (i.e. increase \( \alpha \)).
• Increase sample sizes.
• Specify a larger effect size.

**Suggested wording for the Animal Use Approval Form to incorporate your use of Statistical Support:**

The above power analysis [description provided] was completed by [Name and Department here]. We will use the resources of [name here] for analysis, interpretation, and presentation of our data. Additional statistical support and guidance will be provided by [Name and Department here].

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**2. Animals as Aids (a source of material)**

**a) End Products**

When animals are used to produce tissues, cells, or cellular sequences or similar biological end products (e.g., in cell culture, microarray, etc.). In such instances, the PI should work backward in their justifications. Start by justifying the number of products, and then explain how you chose the number of original animals to get there.

**b) Classroom**

Sometimes animals are used in classrooms or for clinical teaching purposes. Start with numbers of animals per section (or per student), indicate the numbers of sections/students, and then show how these add up to the totals requested.

Approved by the WSU – IACUC on 22 September 2004