Title: Immunization of Rodents and Rabbits in Antibody Production

Purpose:
To provide guidelines for minimizing pain and distress to rodents and rabbits used in antibody production. The following are considered the recommended practices by the IACUC; deviations from these guidelines require scientific justification in the Animal Subject Approval Form (ASAF) and be approved by the IACUC before work can begin.

Procedure:

1) Principal Investigators must justify the use of animals for the production of antibodies in their ASAF. They should also cite specific references to explain why in vitro methods are inappropriate.

2) Alternatives to Complete Freund’s Adjuvant (CFA), such as RIBI, Titer Max. etc., must be considered. If used, CFA can be injected only once per animal. Incomplete Freund’s Adjuvant (IFA) will be used for subsequent booster immunizations. Immunogens should be separated as completely as possible from polyacrylamide gels or other separation media. The amount of adjuvant in the injection should be minimized.

3) Hair should be clipped from intradermal and subcutaneous injection sites, and the site should be aseptically prepared with betadine, nolvasan scrub, or other appropriate antiseptic followed by isopropyl alcohol.

4) The following are the recommended injection sites and volumes:
   a) Subcutaneous: 0.10 ml in mice and rats (2-4 sites), 0.2 ml in rabbits (3-5 sites)
   b) Intradermal: 0.05 ml in rabbits (10-20 sites)
   c) Intraperitoneal: 0.10 ml in mice and rabbits
   d) Intravenous: Cannot use adjuvant
   e) Intramuscular: Should provide strong scientific justification.
   f) Foot pad: Not allowed without strong scientific justification and explicit approval of the IACUC

5) Subcutaneous and intradermal injection should be given in the dorsal thoracic and lumbar regions, avoiding the cervical and intracaspular regions, which may interfere with handling.

Approved by WSU IACUC on: August 28, 2015