Guidelines For Polyclonal Antibody Production In Rabbits

Purpose
To provide basic guidelines for immunization that will minimize complications and assure humane care and use of rabbits.

Background
Polyclonal antibody production is commonly performed on rabbits and typically involves the use of Freund’s Complete and/or Incomplete Adjuvant (FCA, FIA). Immunization sites occasionally become excessively inflamed, resulting in large granulomas, sterile abscesses and/or ulcerated areas that cause distress and pain in the animals and require treatment.

Procedure

Antigen preparation and injection
- Add a concentrated antigen solution to adjuvant to reduce quantity of emulsion injected.
- Use multiple injection sites (up to 10) with limitation of volume at each site (up to 100ul per site).
- Maintain sterility of antigen solution.
- Separate injection sites to avoid fusion of inflammatory lesions.
- Inject into the dorsum. Avoid injections on extremities.
- Use FCA initially and boost with FIA. 10 days intervals required between injections.
- Prior to boost examine the rabbit for granulomas. Rabbits cannot be boosted and veterinary staff should be consulted if:
  - An ulcerated lesion is detected
  - Five or more non-ulcerated reactions (granulomas) are present
  - A reaction granuloma exceeds 3cm in diameter

Blood collection
- Sedate rabbits with 1-2mg/kg acepromazine intramuscular (quadriceps muscle, hind leg, 23” needle)
- Disinfect skin on the dorsal surface of the ear with alcohol.
- Use 23”-21” butterfly cannula to draw blood from the central artery of the ear.
- Allow blood to fill the tubing and attach a 10ml syringe to the cannula hub.
- Once needle is removed assure hemostasis by applying digital pressure.
- Use up to 10ml bleeds for testing the quality and quantity of antigen and larger quantities for production bleeds.
- Interval for production bleeds 2 weeks or longer to allow appropriate regeneration.

Safe amount of blood withdrawn in millimeters every 14 days:

Animal weight in kilograms x 0.06 x 0.20 x 1000
It is recommended to monitor the rabbit’s packed cell volume by performing monthly hematocrites (normal 36-48%). If hematocrite falls below 30%, delay blood collection. If hematocrite drops below 20%, contact veterinary staff.