

NU – IACUC

POLICY

Northeastern University Institutional Animal Care and Use Committee

Polyclonal Antibody Production

General:

The in vivo production of antibodies using laboratory animals is considered to be an essential part of research. The intent is to obtain high titer, high affinity antisera generated by animals while causing only momentary pain or discomfort. Polyclonal antibody production is not considered painful or distressful to animals under normal conditions. Proper technique and monitoring must be incorporated into this procedure to assure the animal's welfare.

A cocktail composed of an adjuvant and an antigen stimulates the production of polyclonal antibodies. Below is a short discussion of these components and their properties, as well as recommended methodologies.

Definition:

Adjuvant is described as any substance that improves the immune response to an antigen.

Antigen is the substance that you want an antibody produced against.

Freund's Adjuvants: These are the most commonly used adjuvants for polyclonal antibody production. Freund's Complete Adjuvant (FCA) is composed of heat-killed mycobacterium, mineral oil and an antigen. Freund's Incomplete Adjuvant (FIA) is the same as FCA except that killed mycobacterial cells or cellular components, are absent. FIA is a water-in oil emulsion.

FCA is used only for the initial immunization, while FIA is used for subsequent "booster" immunizations (usually 2-8 weeks apart). Re-injection of FCA can cause painful hypersensitivity reactions. Additional immunizations with FCA must be specifically justified by the investigator and approved by the NU-IACUC.

RIBI Adjuvant System: An oil-in-water emulsion in which the antigen is first blended with a minimal amount of metabolizable oil (squalene) and then emulsified in a saline solution and surfactant. The immunostimulant used in this system include mycobacterial product (TDM) and/or endotoxin product. There are different species oriented formulations.

- Total volume per injection: 50-200 ul

- Rat:
 - Divide into multiple subcutaneous sites
 - SC: 0.1-0.25 ml/site
 - IM or IP: 0.5-0.75 ml/injection
 - Total volume per injection: 1-2 ml

Approved: