

UNIVERSITY OF TEXAS AT ARLINGTON

INSTITUTIONAL ANIMAL CARE AND USE COMMITTEE

USE OF ADJUVANTS IN ANTIBODY PRODUCTION SOP

1. Background Information

Freund's Complete Adjuvant (FCA) can cause inflammation, induration or necrosis. The following guidelines are intended to reduce animal discomfort to a minimum. Departure from these guidelines requires adequate justification in the University of Texas at Arlington (UTA) Institutional Animal Care and Use Committee (IACUC) protocol proposal.

2. Responsibilities

- A. Consider the use of Incomplete Freund's Adjuvant, or another adjuvant¹.
- B. FCA should be used only for the first antigen dose. The use of two or more doses of the complete adjuvant is rarely warranted and must be adequately justified.
- C. Injections containing FCA should be subcutaneous. Intradermal injections can cause skin necrosis and sloughing. Intramuscular injections may lead to temporary or permanent lameness. Intravenous injections have been known to produce pulmonary lipid embolism.
- D. Foot pad injections are not recommended. Injection of the hind limb foot pads in mice, or intradermal injections in rabbits, may be approved if injections at other sites are shown not to produce significant antibody titers to weak antigens. Foot pad injections must be carried out under general anesthesia.
- E. The injection should be divided into fractions so that no more than 0.25 ml is injected subcutaneously at any one site in rabbits and no more than 0.1 ml in mice and rats.
- F. The inoculum should be free of extraneous microbial contamination. Millipore ultrafiltration of the antigen prior to mixing with adjuvant is recommended. Injection site should be aseptically prepared.

¹Commercial alternatives to FCA are available, e.g., TiterMax and RiBi adjuvants, which could produce higher titers of antibodies, with less toxicity, than those usually induced by FCA. Investigators are encouraged to consider these alternatives to FCA.

Reference: [NIH Guidelines for the Research Use of Adjuvants, 4/24/19](#)