

# Polyclonal Production Protocol

## Sheep Protein Schedule of Events, 118 Day

Day:	Procedure:
0	Sheep Pre-bleed (Avg. 5 ml serum) 1° SC: P&G: 1.0 mg with FCA
21	Boost SC/IM: 0.5 mg with FIA
31	Test Bleed (Avg. 5 ml serum)
42	Boost SC/IM: 0.25 mg with FIA
52	Test Bleed (Avg. 5 ml serum)
53-59	ELISA Titer Assay of Bleed *
63	Boost SC/IM: 0.25 mg with FIA
73	Production Bleed (Avg. 200 ml serum)**
84	Boost SC/IM: 0.25 mg with FIA
94	Production Bleed (Avg. 200 ml serum)**
95-101	ELISA Titer Assay of Bleed *
105	Boost SC/IM: 0.25 mg with FIA
115	Production Bleed (Avg. 200 ml serum)**
118	Terminal bleed (Avg. 500 ml serum) Carcass removal (disposal)

**Total material required for injection for the above program is 2.5 mg/animal.**

**If we are conjugating protein for you we assume a 20% loss of protein during conjugation.**

**Total amount of serum expected from a sheep on the above schedule of events is approximately 1.0 L.**

### Please Note:

- It is recommended that a minimum of two sheep be used due to biological variations.
- Immunogen is emulsified in Freund's Complete Adjuvant (FCA) for initial injections. Freund's Incomplete Adjuvant (FIA) is used for all subsequent injections (boosts). Projects follow a three-week cycle of boosts. Test bleeds are taken approximately 10 days after the boosts.

**\*Optional ELISA:** We suggest testing the first bleed of the project to evaluate the effect of antigen on the animal. Information gained from an ELISA at this point will provide an opportunity for you to make adjustments to your immunization schedule. Additional ELISA's performed later in the project will give you valuable information that can be used to determine which sera samples to include in your research or which bleeds you would like to have purified.

\*\*Plasmapheresis can be performed in lieu of production bleeds. Each plasmapheresis would yield approximately 800-1,000 ml of plasma per bleed.