



## Antigen Requirements

We accept proteins, peptides (conjugated or unconjugated), enzymes, heat-killed bacteria and viruses, cells ... and more. Please contact us if you need antibodies against a different antigen. If providing a fusion protein made using a large fusion partner such as GST or MBP, we recommend cleaving the fusion partner prior to submitting the protein for immunization. Large fusion partners tend to be highly antigenic and host species often produce antibodies against them as well. This dilutes polyclonal antiserum and requires additional purification to yield specific antibodies against the target. It also makes monoclonal antibody development screening more difficult. Antigens with molecular weights less than 10 kDa should be conjugated to a carrier protein such as KLH (Keyhole Limpet Hemocyanin) or BSA (Bovine Serum Albumin) to make them large enough for recognition by the host animals' immune system. KLH is preferred as it is large and is also not common in laboratory samples, so antibodies generated against this protein will not cause background.

### Antigen Quantities:

- We can start a custom polyclonal antibody production project with 50% of the total amount of antigen needed. Additional quantities can be supplied during the project's progress.
- *Immunization:* Please send 10% more than listed below to account for residual loss when mixing the emulsions and ensure that the quantities below are immunized into the animals:
  - 1 mg per pair of rabbits
  - 1 mg per pair of chickens
  - 0.625 mg per five mice
  - 1.25 mg per goat
  - 0.5 mg per llama
- *ELISA:*
  - 100 µg per bleed per animal tested
- *Immuno-Affinity Purification:*
  - 5 mg for a standard column (Purified Package) if the protein is less than 50 kDa in size, 8 mg for a standard column if it is more than 50 kDa in size.
  - Avoid using free amines in the buffers, such as Tris, etc.
  - Protein should be concentrated, with 1 mg/ml minimum.
- *Mouse Monoclonal Antibody Development:*
  - 5 mg for immunization and screening
- *Single Domain Antibody Development:*
  - 0.5 mg for immunization plus 0.5 mg for basic screening
- *Rabbit Monoclonal Antibody Development:*
  - 2.5 mg for immunization and ELISAs in Phase I
  - 1 mg for Phase II and ELISA screening of clones

Please complete the Antigen Submission Form and email to [services@prosci-services.com](mailto:services@prosci-services.com) for approval prior to shipping the antigen.



Where Scientists Meet Antibody Success

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**Antigens in Solution:** Antigens in solution should be provided at a concentration of 1 mg/ml or higher. Preferred buffers include PBS and Tris for immunization, but samples used for immuno-affinity purification should not contain Tris or other buffers with primary amines as they interfere with the coupling chemistry. Soluble proteins are preferred and up to 4 M urea is acceptable, but precipitated proteins can be used for immunization as well. For services such as ELISA and immuno-affinity purification, however, the antigen must be soluble.

**Lyophilized Antigens:** Lyophilized proteins or lyophilized peptides already conjugated to a carrier protein should be supplied in the quantities listed above. 5 mg of unconjugated lyophilized peptides should be supplied for conjugation to a carrier protein. Include the reconstitution method on the antigen submission form.

**Gel Strips as Antigens:** Proteins can be submitted as a gel strip cut from an SDS PAGE gel. Please note that we cannot perform an immuno-affinity purification or ELISA when the protein is supplied in this form. However, you may send antigen in a gel strip for immunization and another batch in solution for the add-on services. The protein needs to be reasonably concentrated (2-5 mg/ml) and then run on the gel. If the protein precipitates, use a lower concentration. 100 µg to 200 µg of protein in 500 µL of gel is the maximum we can inject into rabbits. Stain the gel with coomassie blue, de-stain it, and rinse with water at least twice to remove traces of methanol. Cut the desired strip from the gel and place it in a 15 ml or 50 ml conical vial with deionized water and ship with an icepack – do not freeze. The strip will still have some color.

**Shipping Guidelines:**

If the antigen is stored frozen, ship on dry ice; if refrigerated, ship on gel ice packs; ship antigens in a Styrofoam shipping container by an overnight delivery company such as FedEx, DHL, or UPS.

**Ship Antigens To:**

ProSci Incorporated  
Attn: Antigen Receiving  
12170 Flint Place  
Poway, CA 92064 USA