

Ab^{Max} Peptide Antibody Package (Cat# 60001RB)

The package includes the following items:

- Consultation of epitope selection and confirmation.
- ❖ 20 mg of peptide (80% checked by HPLC, up to 15 mer); BSA/KLH conjugation,
- ❖ 2 rabbits with AbboMax immunization protocol,
- ❖ Pre-immune serum (2-5 ml/rabbit); 4 s.c. immunizations; 2 bleeds with total 100 ml of serum
- ❖ ELISA titration for all bleeds. Western blot (Optional); Dot blot (Optional)

The data will be provided with the project:

- Peptide: HPLC and Mass Spec
- KLH/BSA Conjugation datasheet
- ELISA titration for all antisera

Timetable

- ❖ 1st test bleed will be shipped within 10 weeks upon the order is taken, please see the details:
- Epitope selection and consultation: 2-3 days
- Peptide synthesis/purification/conjugation: 2-3 weeks
- 1st test bleed shipment: 7 weeks
- ❖ 2nd production sera: 4 weeks

Standard Operation Protocol:

Outline:

Peptide synthesis -Fmoc solid phase peptide synthesis, and purified by reversed-phase HPLC **Peptide-carrier protein conjugation-** According to the protein/peptide chemistry.

Immunization--7-8 weeks old of New Zealand White Rabbits will be immunized with 1st dosage of immunogen with Complete Freund's Adjuvant (CFA) followed by subsequent boosting of 2nd, 3rd and 4th immunogen with incomplete Freund's Adjuvant (IFA). The dosages will be variable according to the nature of immunogen and resources. Typically, it will be 50-500 μg/injection.

Procedure:

Prepare the rabbits 3 days prior to the immunization. The equal amount of protein (at appropriate concentration) and adjuvant are homogenized to form an identical emulsion, and immunize the rabbits subcutaneously. Label the rabbit with antigen and ID. The following immunizations will be performed according to the immunization schedule. Test bleed will be taken 10 days later after 3rd immunization. The whole blood is collected and incubated at 37°C to maximize the yields, and followed by refrigerating and centrifugation. (The labels will be matched by the rabbits and tubes, including the project name, rabbit ID, bleeding date, and volume).

3) Schedule:

- ❖ Day –3: Prepare 2-5 ml of pre-immune serum.
- ❖ Day 0: 1st immunization with 1st dosage of antigen with CFA.
- ❖ Day 20th: 2nd boosting with 2nd dosage of antigen with IFA
- ❖ Day 40th: 3rd boosting with 3rd dosage of antigen with IFA
- ❖ Day 50th: 1st test bleeding will be performed to check the titer, the antisera (20-25 ml/rabbit) will be shipped to customers, allow them to have additional 3 weeks to evaluate the antisera.
- ❖ Day 70th, 4th boosting with 4th dosage of antigen with IFA
- ❖ Day 80th: Terminal bleed will be performed and evaluated by ELISA. The antisera (~30-40 ml/rabbit) will be shipped to customers.

Note: If customer would like to get more antisera production, please inform us before day 80th, we will extend the immunization schedule and continue the bleeding process, in this case, each bleeding will yield 20-25 ml/rabbit to maintain the healthy rabbits until the customer would like to get terminal sera before exsanguinations. Please e-mail: info@abbomax.com.