



**AGRO-BIO**  
The Proteomic &  
Immune Response

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# Protocol for the production of mouse polyclonal antibodies

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Agro-Bio has developed a time-optimized immunization protocol, yielding quality antibodies, needing low amounts of antigen. We recommend the use of 5 animals minimum to follow the protocol of immunization in order to overcome the differences obtained due to the genetic variability between each animal.

The mouse (*Mus musculus*) represents species of choice when **small quantities of antigens** are **available** and when the production of **weak volumes of antibodies** are enough for the foreseen use.

Our standard protocol **35 days** with 5 mice allows you to obtain after 3 injections of antigen, **100 - 200 µL of antiserum** (possibility of obtaining until 500 µL from antiserum after exsanguination of the animals).

<b>Species:</b>	Balb / C mouse. Other mice are available on inquiry.
<b>Antigens:</b>	Peptides, proteins, bacteria, viruses or any other inactivated or nontoxic organism whose antigenicity and immunogenicity have been demonstrated. Concentration advised $\geq 1$ mg / mL.
<b>Antigen quantities:</b>	To obtain good titers of antibodies, AGRO-BIO uses for a protocol 35 days: - 200 µg by mouse (that is 1 mg for 5 mice) - $3 \cdot 10^8$ CFU or PFU by mouse (that is $15 \cdot 10^8$ CFU or PFU for 5 mice).
<b>Injections:</b>	The first injection is carried out by subcutaneous and/or intraperitoneal injection in FCA whereas the booster injections are made in incomplete Freund's adjuvant.



## Immunization protocol

Day	0	14	28	35
<b>Injection</b>	1 <sup>st</sup>	2 <sup>nd</sup>	3 <sup>rd</sup>	
<b>Volume of serum</b>	Pre-immune <sup>(1)</sup> 50-100 µL			50-100 µL

<sup>(1)</sup> Pre-immune serum enables to determine whether the animals have naturally occurring antibodies that can recognize non-specifically the antigen of interest.

Within two weeks following the dispatch of your order, you will receive a provisional timetable listing the various stages of your service.

### Included in our protocol:

- The purchase of animals, the quarantine period and transfer to experimentation,
- The guard of animals during the protocol,
- The injections and withdrawals of animals,
- The 10-day guard while awaiting your decision on the fate of animals,
- The sacrifice of animals without recovery of serum.

### Not included in our protocol:

- The charges,
- The guard of animals for a period exceeding 10 days (you have 10 days starting from last collection reception date to inform us of your decision to continue or not your protocol. After this deadline and in the absence of response from you, we are forced to sacrifice your animals),
- Withdrawals of end protocol (3 withdrawals at 1 day interval corresponding to « exsanguination »).

*Cf. General terms of sale.*

Agro-Bio is approved by the prefecture of Loir-et-Cher for experimentation on vertebrate animals.  
**Accreditation number: B 41-285-4**

