AGRO-BIO
The Proteomic &
Immune Response

AGRO-BIO

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Protocol for the development of mouse monoclonal antibodies

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EN

Agro-Bio offers a development program that combines skills and know-how in developing hybridomas. Before starting your project, a discussion with our Cell Culture department is required to identify your needs and constraints and thereby define precise specifications.

Standard hybridoma development is as follows (minimum: **5 months**):

Initialization phase of the project:	- **Fill in the specifications sheet for the development of your monoclonal antibodies** and return it, - Agro-Bio will assess the feasibility of your program, - Upon request, a bibliography on your antigen or developed antibody can be achieved. Additional costs will be charged.
Phase I: Immunization **(9 weeks)**	Our standard protocol on 5 mice includes : - Immunization of animals according to a 63-day protocol including 4 injections and 2 control samples, - Development of **ELISA method specifically developed** to follow the immune response of animals in your protocol, - Monitoring the immune response of immunized mice in order to determine mice with the best serum titer, - Choice of mice for fusion.
Phase II: Fusion **(2 weeks)**	- Splenocytes from the best mice are merged with myeloid cells, - The fusion cells are divided into 96-well plates, - Wells in which a single clone has grown are identified 6 days after the fusion.
Phase III: Primary screening **(4-5 weeks)**	- The culture supernatants from the identified wells are tested via the ELISA method previously developed, - The positive hybridomas are then divided into 24-well plates so as to undergo a second round of screening on their **specificity towards the antigen,** - A bank of 5 cryotubes is made.
Phase IV: Cloning **(2 weeks)**	In collaboration with the customer, the chosen hybridoma is cloned by successive dilutions.
Phase V: Secondary screening **(3-4 weeks)**	- The clones are divided into 96-well plates and a secondary screening is done by ELISA, - The best positive clone is selected and **a bank of 10 cryotubes** is made.

The characterization of antibodies produced for a particular application can be achieved even if it is not included in the standard service. Additional costs will be charged.

Agro-Bio can send upon request the supernatant so you can test the hybridomas according to your own methods.
Within two weeks following 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 the transfer to experiment,
- The guard of animals during the protocol,
- The injections and withdrawals of animals.

Not included in our protocol:

- The charges.

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