

## Monoclonal Antibody Development Services

### Introduction:

Bangalore Genei offers a very convenient, reliable and timely custom hybridoma development service to suit the customer needs. We have experience in generating hybridomas against a variety of antigens ranging from peptides, large proteins - both pure as well as fusion proteins, viruses, bacterial proteins and mammalian cell surface proteins.

All experiments are carried out in our facility. Our technical staff has extensive training and experience with bleeding, injecting, caring for and handling animals. We have an independent Institutional Animal Ethics Committee (IAEC) governed by the Committee for the Purpose of Control of Scientific Experiments in Animals (CPCSEA). We have a pre-drawn protocol for antibody production, but if you suggest a unique protocol we can usually follow it if it is within our guidelines. We have an environmentally controlled and monitored animal facility in keeping with the CPCSEA's Guide to the Care and Use of Experimental Animals. This ensures results are not altered by environmental stress.

### Features:

Custom Monoclonal Antibody Development service at Bangalore Genei normally takes 5-6 months and is divided into 4 phases. This allows the researchers to evaluate the progress prior to the beginning of each phase.

**Cat # 105518: Phase I:** The purity and concentration of the antigen provided will be determined by SDS-PAGE.

(> 95% pure by SDS-PAGE for immunization and > 99% pure for sensitive assays).

Peptide antigens will be conjugated to carrier proteins using suitable methods.

Immunization of three Balb/c mice on a predetermined schedule with one primary immunization and at least 3 boosters with the immunogen provided. This is followed by monitoring the antibody response in serum by an antibody capture ELISA. This regime will continue until sufficient antiserum titre is achieved. The success of a custom hybridoma project is dependent on the quality of the immunogen used.

Phase I report in the form of antibody titration results will be provided to the customer.

For the entire project, about 5 mg of the protein or 20-30 mg peptide is required.

The duration of Phase I is about 12-16 weeks.

**Cat # 105519: Phase II:** Best antibody producing mouse is used to get splenocytes for fusion with myeloma cells using the PEG method. The fused products are cultured in HAT selection medium. The cultures are monitored for growth and fed weekly. Clones are screened by ELISA. Positive parental clones are amplified and frozen.

The other two mice are kept in reserve until a positive fusion clone is obtained.

**5 ml of culture supernatant from 3-10 positive clones is provided to the customer for evaluation in specific application.**

The duration of Phase II is about 3-4 weeks.

**Cat # 105520: Phase III:** Selected 3 clones will be subcloned by limiting dilution. Subclones are screened by antibody capture ELISA, positives expanded and screened again. Up to 3 positive subclones from each parental clone will be expanded and frozen.

**3 x 20 ml of culture supernatant from each monoclonal is provided to the customer for evaluation in specific application along with frozen cells from these clones.**

The duration of Phase III is about 6-8 weeks.

**Cat # 105521: Phase IV:** Isotype determination of all the monoclonal antibodies generated will be done.

**Phase IV report in the form of isotyping results and final service report will be provided to the customer.**

**Cat # 105525: Additional colony expansions:** Additional subcloning and expansion of parental clones from Phase II can be performed at an extra cost.

The time frame for this service is about 6-8 weeks

### TERMS AND CONDITIONS:

- Mab production service does not include the cost of making or purifying antigens. User is required to supply these to us or arrange for their production.
- User must specify, if the Monoclonal antibodies are to be generated against the native antigen or denatured antigen and supply the antigen appropriately.
- Mab production service is divided into phases with the option of halting the hybridoma production prior to the beginning of the next phase.
- Any additional phase II service to be done after billing will be charged 75% extra of Cat # 105519 service.**
- We cannot guarantee that a Mab will be produced that will work in the users own application. Antibody secreting clones are selected by using antigen coated plates (ELISA). The user is required to do further testing.
- The isotype of the monoclonal generated will be known only at phase IV of the service. They could be either of the IgGs or IgM. Hence, Phase wise payment is mandatory irrespective of the result of the following phase.
- We will make every effort to provide the customer with the service outlined above in the shortest possible time. Minimum of 5-6 months will normally be required for the completion of the project. Delays may be incurred if the antigen is poorly immunogenic or produces unstable clones.
- All clones, cell lines, antibodies and other materials which are the products of this project shall become the exclusive property of the customer at the conclusion of the contract.
- Antibody clones/cell lines can be preserved in liquid nitrogen for a period of 6 months after the completion of the service. If the clones have to be preserved at our facility beyond this period, kindly place order for our service Cat # 105531.
- Complete confidentiality is guaranteed.