

Advanced BioDesign Outlines Solutions



by Advanced BioDesign

Antibody Overview

Launching an immunisation programme is an important experimental step that needs care. With Advanced BioDesign, you may develop and produce antibodies according to customer's specific needs.

- Real support and advice from a dedicated team all along your project
- Large and modern SPF animal facility
- Ethical treatment of animals

Project Start

The evaluation of your project is free-of-charge and with no obligation. Dedicated project managers will work with you from the beginning to the end of your custom antibody project to ensure the highest level of quality and service.

Immunogenicity

Immunogenicity is the ability of a molecule to solicit an immune response. A molecule must have the following characteristics to be immunogenic: non-self, high molecular weight and antigen purity. Non-self is required for the molecule to be recognized by the immunized animal. Macromolecules which have a molecular weight >6kDa are immunogenic. Smaller compounds (haptens) have to be coupled onto a larger protein to obtain a satisfying immune response.

Selecting Your Antigen

In order to obtain the best antibody, antigen has to present the most number of desirable epitopes.

Proteins

Protein antigens have the advantage of possessing a large number of possible epitopes thus enhancing the chance of generating

antibodies. However anti-protein antibodies may cross react with other isoforms due to the homology. Protein required for immunization have to be >80% pure.

Peptides

Peptide antigens are today a reliable alternative to anti-protein immunization that offer the following advantages:

- Selection of a protein region that is unique to a specific protein isoform
- Easily synthesized and highly purified material
- Antibody can be easily purified by its affinity for the antigenic peptide
- Peptide antigens can be designed in order to raise antibody against Post-Translational Modifications (PTM)

The single peptide immunization strategy is suitable for reproducible antibody production with an improved peptide. The double peptide immunization strategy involves the design of two peptides from the same protein, and coimmunized to animals. The success rate of this strategy can reach 90%.

Others

Other antigens can be used to raise antibody:

- Bacteria
- Viruses
- Non exhaustive list

Antigen Needs?

If you do not have any antigen, Advanced BioDesign can produce your protein or synthesize your peptide for immunization.

Antibody Affinity Evolution

Exposure to a specific antigen will initiate the immune response which takes place in two phases. First exposure induces a fast immune reaction resulting in IgM class antibody

secretion. IgM, mostly as a pentamer, is characterised by a lower affinity, lower specificity than other antibody classes. Further exposures lead to a faster response, induced by memory T-cells, and will generate IgG type antibodies. IgG is a monomer protein, and has a higher titer, specificity and affinity than IgM.

Polyclonal Antibody

Selecting Your Package

Advanced BioDesign develops and produces rabbit polyclonal antibodies according to customer's specific needs. We give the option of choosing either, all-inclusive service package or different modules "A la Carte" to achieve high-quality polyclonal antibodies.

All-Inclusive Package

Using the all-inclusive service package allows you to obtain antibody against the protein or peptide after a 70-day immunization. This package has been optimized to ensure the best immunoreactivity at the lowest price.

Polyclonal Anti-Post-Translational Modification (PTM) Antibody Package

Post-Translational Modifications (PTM) of proteins have been described in a large number of publications. Modifications influence protein characteristics that can modify cellular functions. To understand the particular role of any specific modification, it is crucial to detect it specifically. Using their know-how in immunization, Advanced BioDesign will assist you in the development of an antimodified peptide antibody. Modifications available for this package are: Acetylation and Phosphorylation

A La Carte Package

Advanced BioDesign can develop a tailored project in order to be close to your needs. We give you options to choose single modules covering each crucial step to achieve a high-quality antibody.

Monoclonal Antibody

Mouse Monoclonal Hybridoma Development

Project Development

The evaluation of your project is free-of-charge and with no obligation. Dedicated project managers will work with you from the beginning to the end of your custom antibody project to ensure the highest level of quality and service.

Starting Material

Protein / Peptide sequence / Haptens

Immunisation

3 Balb/c and 3 SJL are immunized following a 9-12 weeks protocol. This include five injections and two testbleeds. Additional boosts and test bleeds will be done depending on immunoreactivities.

Fusion Hybridoma Production

The splenic lymphocyte of the best responding animal will be fusionned with sp2/o-Ag14 cells.

Screening Selection of Hybridoma

Positive clones will be screened against antigen of interest. Further tests can be performed against other antigens and using different techniques in order to check antibody specificity and cross reactivities.

Cloning and Isotyping

Selected hybridoma will be recloned to select stable high producing subclones. 3 clones will be sent to customer.

Options

Additional clones can be selected during "Cloning and Isotyping" Step Selection of a the right couple of antibody for sandwich ELISA development

Additional Quality Control

Additional tests can be made for a more detailed characterisation of antibody:

- Specificity – via Western Blot Analysis
- Affinity/Sensitivity – via Surface Plasmon Resonance Technology Analysis
- Stability – possibility to constitute a cell bank by freezing cells or Advanced BioDesign proposes a complete hybridoma back-up service by sequencing hybridoma V-region.

Monoclonal Antibody Production Services

Advanced BioDesign provides cost effective cell culture services for antibody production. Quality at a reasonable price is our primary focus regardless of whether you require research or a commercial product, we strive towards customer satisfaction. Advanced BioDesign provides in vitro production methods such as roller bottles, hollow fiber, and bioreactor technologies. Optimisation of production, development of specific formulations and conditions for your cell line can be done to improve performance and quality. In vivo antibody production can be performed in our facilities.

Project Steps

Your Clone

You provide us the clone and a technical data sheet mentioning the culture medium and the antibody isotype.

Quotation

For quantities >50mg, a productivity evaluation in smaller volumes is required. A quotation for the whole production will be sent based on this evaluation.

Production Follow-Up

Viability & antibody titration will be done in order to optimize the productivity of your clone.

Purification

Supernatant is purified using affinity chromatography on Protein A or G depending on the clone's origin & isotype. Other chromatography strategies can be employed to purify your antibody using ion exchange chromatography, immuno-affinity chromatography...

Quality Control

With your final product, you will also receive quality control including SDS-PAGE control and protein concentration (Abs at 280nm). Other tests can be performed like ELISA, Western Blot.

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Additional Services

Antigen Production

Custom Peptide Synthesis

Peptide Design

The experience of the scientists at Advanced BioDesign in designing peptides will help you to select the most immunogenic sequence of your protein. Your protein sequence will be analysed following different data (antigenicity, specificity, post-translational modification,...).

With each antibody project, a complete protein analysis and peptide design report will be sent to you including:

- Selected antigens with scoring
- BLAST for homology search

Advanced BioDesign proposes custom peptide synthesis with a wide range of labels, modifications, scales... All peptides are validated by two different quality control techniques: mass spectrometry & analytical High Pressure Liquid Chromatography (HPLC).

Custom Protein Expression

Whether you are interested in crystallography, protein-protein interactions, antibody development, or activity assays, Advanced BioDesign proposes a protein expression service.

You choose your protein of interest and we will produce it according to your specification.

- Expression in E.Coli
- Expression in mammalian cells (HEK, CHO...)
- Choice of protein's tag
- From research to GMP grade

Project Description

1. Each project starts with the synthesis of a codon-optimized gene
2. We clone it into the best suitable vector following your specification (tags, producing host...)
3. Production of a pilot batch to determine protein solubility, production yields...
4. Based on previous experiment, we produce a large-scale batch
5. Purification of protein using tags or other types of purification

For starting protein expression project, please contact us, contact@a-biodesign.com.

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We may need this information for preparing a quotation:

- Your gene/protein sequence
- The host

- The tag
- Amount need
- Purity

Purification Services

Advanced BioDesign can purify your antibody using a wide range of methods, from research (mg) to scale-up (grams) quantities. We can discuss your antibody purification options in order to find the best procedure which provides the best balance between yield, purity and cost.

Available Antibody Purification services and methods are:

- Ion Exchange Chromatography (anion & cation)
- Size Exclusion Chromatography
- Hydrophobicity Chromatography
- Affinity Purification:
 - Protein A
 - Protein G
 - Antigen Immuno-affinity purification
 - Metal Chelated purification
- For any other purification technique, please contact us.

Modification Services

In order to provide you with the most efficient tool for your applications, Advanced BioDesign proposes conjugation and fragmentation services.

Conjugation Fragmentation

Biotin
 Enzyme (HRP, Phosphatase, etc.)
 Fluochrome (FITC, ALEXA, DyLight, etc.)
 Beads (Gold, Latex, etc.)
 Solids Surfaces (Polystyrene Plate, Silica, etc.)
 Fab fragment (Papain treatment)
 Fab'2 fragment (Pepsin treatment)
 Fragmentation of IgM

Characterisation Services

In addition to standardized tests, Advanced BioDesign customizes procedures to be able to live up to your goals. You can include the following technology to characterize your antibody:

- Purity assessment

- ELISA test analysis (using one or different antigens)
- Western Blot analysis
- FACS analysis
- Sequence Confirmation
- Epitope Mapping
- Size Exclusion Chromatography (SEC) analysis
- Isoelectric Focusing (IEF) analysis
- Surface Plasmon Resonance (SPR) analysis
- Non exhaustive list

Antibody Engineering

Advanced BioDesign is able to conduct projects involving recombinant antibody gene manipulation. These include V-region discovery and sequencing, isotype switching, in vitro affinity maturation as a part of a full package involving «humanisation», production of stable cell line development and delivery of quantities of purified antibody following GMP requirements.

Key areas of competence:

- V-region discovery and sequencing
- Cloning plasmid and/or expression vector assembly
- In vitro affinity maturation
- Chimeric antibodies
- Isotype switching (from IgM or IgE in IgG)
- Humanization (CDR grafting)
- Cell line development (GMP or non GMP compliancy)

Cell Banking

A cell bank is an essential element in the production of uniform biological products. Advanced BioDesign's cell banking system provides the assurance that a sufficient supply of material will be available for the life of product.

Outsourcing Services

For the successful launching of any product the following criteria are essential: speed-to-market, consistent quality and innovative design. Advanced BioDesign understands your needs and proposes solutions in antibody technology outsourcing. With over a decade of experience in R&D, Advanced BioDesign offers customer-focused, R&D services to meet and exceed your specific needs and challenges. We propose flexible, added value and

secure solutions in Biochemistry
Technology allowing you to focus on performance of your
company. Need some consultation for
your research, do not hesitate to contact us.

Assay Development

A good and robust assay to detect specific biomolecule is now essential for any process development. Advanced BioDesign can provide clients with assistance in assay design, development, enhancement and production of assays. Based on our specific know-how and experience in assay development, we can assist you in setting up a new specific assay for your targeted molecule. Our scientists will work directly with customers to define their needs and to provide the best conditions for accompanying you in your project development. Advanced BioDesign develops customized cell-based assays, immunoassays and biochemical assays to meet the biocompany's needs in different fields of applications: biomarker, quantification, immunomonitoring, drug detection, quality control methods...

Pipelines

Working with us will follow this communication path:

Phase 1: Feasibility Study

A development plan to conduct a feasibility study will be prepared based on your specifications. At the end of the feasibility study a report will be prepared and submitted along with a small number of early prototype tests.

Phase 2: Optimization and Scale-Up

A robust and reproducible test that fulfills the customer's specification will be produced. All materials needed for the assay are validated using specific protocols.

Phase 3: Validation

The validation stage involves the preparation and submission of a validation plan and the manufacture of three independent batches using different critical materials. Prior to the submission of the validation batches, draft Application Notes will be prepared and used to conduct the three batch validation.

Phase 4: Commercialisation

Advanced BioDesign proposes to evaluate the potential of your assay. If marketing your assay is an option, we are able to produce, characterise and market generated batches. Royalties

are paid then to the owner of the assay..

Microplate Coating

Advanced BioDesign provides custom microplate coatings using different functionalisation technologies. We can accommodate virtually any size of the following formats (8 to 394 wells/plate). Our quality control methods include validated microplate coating systems and the guarantee that production is feasible. These quality control measures will meet the strict demands of our biotechnology, pharmaceutical, and other life science customers.