CONTRACT SERVICES

MOLECULAR BIOLOGY SERVICES

PCR Cloning and Sub-cloning service

Gene cloning is the act of making copies of a single gene. Amplified genes are useful in many areas of research ranging from genetic fingerprinting to large scale protein production as well as for medical applications such as gene therapy. Bhat Bio-Tech offers PCR cloning and sub-cloning services to meet the needs of the scientists. Our sub-cloning service is highly flexible to any specific project requirements. The starting materials can be bacterial culture, plasmid DNA or PCR product.

Sample requirements:

- The complete sequence of the template including the target gene and its vector
- If the starting material is DNA, 2 µg template DNA
- Sufficient amount of culture if starting material is bacterial culture

Deliverables:

- 5 µg of destination vector containing the gene
- Sequence chromatograms covering your gene
- Construct map for the plasmid
- Project report
- Quality assurance certificate

cDNA synthesis and cDNA cloning service

A cDNA is a DNA copy synthesized from mRNA. The central part of any cDNA cloning procedure is synthesis of the cDNA from an mRNA template using reverse transcriptase enzyme. Cloning and sequencing of cDNA is useful to understand various aspects of the gene as well as to express the genes in heterologous systems.

Features:

- Cloning in any commercially available cloning or expression vector
- Cloning and sequencing irrespective of the size of the gene
- Rapid turnaround time

Deliverables:

- Full length cDNA in a vector of choice
- Sequence data
- Project report
- Quality assurance certificate

Cat. No.	Service
BBCS-20101	PCR cloning and sub-cloning service
BBCS-20102	cDNA synthesis and cDNA cloning service

DNA sequencing Service

Bhat Bio-Tech offers DNA sequencing service using latest and the highest quality high throughput technologies for its sequencing.

Sample requirements:

- DNA sample: Plasmid DNA - Purified DNA or bacterial culture to isolate plasmid and to sequence
  PCR product - Purified PCR product or PCR reaction for clean up and to sequence
  Bacterial culture or template - To PCR amplify, clean up and sequence
- Primers 10 µM

Deliverables:

- Sequence data

Cat. No.	Service
BBCS-20103	DNA sequencincing up to 600 bases
BBCS-20104	Purifucation of PCR product and sequencing
BBCS-20105	PCR amplification,Clean up and sequencing
BBCS-20106	Purification of plasmid and sequencing

CONTRACT SERVICES

Large scale plasmid DNA preparation service (endotoxin free)

Plasmid DNA preparation service at Bhat Bio-Tech covers individual constructs for research labs as well as large scale manufacturing for biotech and pharmaceutical companies. The high quality plasmids will help you achieve highly efficient cell transfection, DNA vaccination, antibody production, and other preclinical studies.

Features:

- Flexible scale
- Stringent quality control
- Endotoxin level < 0.1 EU/µg DNA (Optional)
- Rapid turnaround time

Deliverables:

- Purified plasmid DNA
- Project report
- Quality assurance certificate

Cat. No.	Service
BBCS-20107	Large scale plasmid DNA preparation service
BBCS-20108	Large scale plasmid DNA preparation service (Endotoxin free)

Genetically Modified organism (GMO) detection service

A genetically modified organism (GMO) is an organism whose genetic material has been altered using genetic engineering techniques giving the organism modified or novel genes. Testing on GMOs in food and feed is routinely done by molecular techniques. We offer multiplex PCR system to screen samples for different potential GMOs. To avoid any kind of false positive or false negative testing outcome, internal controls for every step is performed.

Deliverables:

- Gel image of PCR result
- Final report of GMO detection

Cat. No.	Service
BBCS-20109	GMO detection service

PROTEIN EXPRESSION AND PURIFICATION SERVICES

At Bhat Bio-Tech, protein expression specialists work closely with clients to clone and express full-length recombinant proteins from full-length cDNAs, EST sequence information, and genomic sequences, providing service and expertise at all stages of protein expression and purification. The purified proteins can be used in number of down stream applications including monoclonal and polyclonal antibody development, immunoassay development etc. Protein expression and purification service package include sub-cloning into appropriate expression vector, sequence confirmation by gene sequencing, plasmid maxi preparation, optimization of protein expression, expression confirmation by SDS-PAGE and Western blotting, and purification of expressed protein to near homogeneity (>90% pure).

Features:

- Protein expression in-Bacteria, Yeast, Baculovirus and Mammalian cells are available
- Protein purification to near homogeneity (purity >90%)
- Expression systems with 6X His tag, GST tag, GFP tag and without any tag are available
- Large scale production of recombinant proteins upon request
- Purified proteins with endotoxin level < 0.01EU/µg of protein is available on request

Deliverables:

- All clones and sub-clones generated
- Sequence data of the final clone
- Purified recombinant protein
- Project report
- Quality assurance certificate

Cat. No.	Service
BBCS-20110	Protein expression in Bacteria
BBCS-20111	Protein expression in Yeast
BBCS-20112	Protein expression in Baculovirus
BBCS-20113	Protein expression in Mammalian cells

ANTIBODY SERVICES

Monoclonal Antibody Development Service

Bhat Bio-Tech provides customized hybridoma development service to meet the customer needs. We can develop monoclonal antibodies to a wide range of antigens including peptides. All the animal experiments are carried out in our state-of-the art facility approved by CPCSEA, Govt. of India.

The monoclonal antibody development service is divided into four phases

Phase I: Immunization and evaluation of antibody titer

Phase II: Fusion, plating and screening

Phase III: Subcloning by limiting dilution, expansion and cryopreservation of 3 selected clones

Phase IV: Amplification- production of 50 ml of culture supernatant containing monoclonal antibody.

Sample requirements:

- 5 mg of pure protein or 25-30 mg of peptide

Deliverables:

- Phase I: ELISA data for antibody titration
- Phase II: ELISA data for screening of hybridoma

  Culture supernatant from 5 of the best hybridoma containing wells
- Phase III: ELISA data for screening antibody secreting subclones

  Culture supernatant from 3 of monoclines

  Isotyping results of 3 of the monoclonal antibody
- Phase IV: 50 ml of culture supernatant containing monoclonal antibody

  Frozen hybridoma cells

  Final project report

Additional Hybridoma related services:

- Additional subcloning, expansion and screening of parental clones
- Cryopreservation of up to 20 clones per month
- Monoclonal antibody production in ascites of Balb/c mice
- Monoclonal antibody production in cell culture flask- 50 to 100 ml culture supernatant
- Isotyping of monoclonal antibodies

Cat. No.	Service
BBCS-20114	Monoclonal antibody development service
BBCS-20115	Monoclonal antibody production in ascites
BBCS-20116	Monoclonal antibody production in cell culture flask
BBCS-20117	Isotyping of monoclonal antibody

Polyclonal Antibody Production Service in Rabbit

At Bhat Bio-Tech, we develop custom polyclonal antibodies of highest quality with fast and reliable delivery times. The antibody screening will be done by direct ELISA and Western blotting

Sample requirements:

- 5 mg of immunogen

Deliverables:

- 2 ml of pre immunized rabbit serum
- 25 to 30 ml of immunized rabbit serum
- Project report

Cat. No.	Service
BBCS-20118	Polyclonal antibody production service in Rabbit

Antibody Purification Services

Bhat Bio-Tech provides a wide range of options for the purification of monoclonal and polyclonal antibodies. These include relatively common methods such as protein A and protein G affinity as well as protein L and antigen affinity chromatography.

Sample requirements:

- Serum sample
- If antigen affinity chromatography is required 5 mg of antigen to prepare purification column

Deliverables:

- Purified antibody
- Project report

Cat. No.	Service
BBCS-20119	Purification of antibody by Protein A or protein G affinity chromatography
BBCS-20120	Purification of antibody by Protein L affinity chromatography
BBCS-20121	Purification of antibody by antigen affinity chromatography

Antibody Labeling Services

Bhat Bio-Tech offers a wide range of antibody labeling services to meet the requirements of the researchers. Antibody conjugated to enzymes, biotin or FITC are widely used in a variety of immunotechniques as a detection reagent. We provide the following conjugation services

- Conjugation to HRP
- Conjugation to ALP
- Conjugation to Biotin
- Conjugation to FITC

Cat. No.	Service
BBCS-20122	Antibody- HRP conjugation
BBCS-20123	Antibody- ALP conjugation
BBCS-20124	Antibody- Biotin conjugation
BBCS-20125	Antibody- FITC conjugation

Peptide-Carrier conjugation Service

We offer conjugation of synthetic peptide to KLH, BSA or Ovalbumin depending on customer needs.

Sample requirements:

- Synthetic peptide or amino acid sequence of the peptide

Deliverables:

- Peptide carrier conjugate
- Project report

Cat. No.	Service
BBCS-20126	Peptide-KLH conjugation
BBCS-20127	Peptide-BSA conjugation
BBCS-20128	Peptide-Ova conjugation

Colloidal Gold Labeling Service

At Bhat Bio-Tech we have a strong expertise in rapid diagnostic kit development using colloidal gold conjugate. We offer conjugation of colloidal gold to a variety of proteins. We can conjugate proteins to colloidal gold of varying size such as 10nm, 15nm, 20nm, 40nm and 60nm depending on the customer requirements.

Features:

- No aggregation
- OD_{540} is 10 or above
- Uniform size
- Wide range of particle size

Cat. No.	Service
BBCS-20129	Colloidal gold labeling service

MICROBIOLOGY SERVICES

Microbial Identification Services

Microbial identification based on the gene sequencing has become the important tool for microbiologists. We offer following microbial identification services based on DNA sequencing. Identification up to species level and sometimes strain level is possible by this method.

• Bacterial identification service

It is based on 1500 bp 16S rRNA gene sequence. It involves amplification of 16S rRNA gene and sequencing of the PCR product

• Actinomycetes Identification Service

It is based on Actinomycetes species specific 750 bp 16S rRNA gene sequence.

• Fungal Identification Service

It is based on 650 bp ITS region of the rRNA gene sequence.

• Yeast Identification Service

It is based on 500 bp ITS region of the rRNA gene sequence

Features:

- Fast turnaround time
- Delivery time-3 to 4 weeks

Sample requirements:

- Microbial culture

Deliverables:

- Detailed project report
- Gel image of PCR result
- Sequencing report
- Nucleotide sequence alignment within 10 most similar neighbours
- Phylogenetic tree displaying evolutionary relationship of 10 nearest neighbours

Cat. No.	Service
BBCS-20130	Bacterial identification service
BBCS-20131	Actinomycetes identification service
BBCS-20132	Fungal identification service
BBCS-20133	Yeast identification service

Bacterial endotoxin testing service

Endotoxins are potentially toxic natural compounds of the cell wall of gram negative bacteria. The material can induce fever and acute febrile shock when present in miniscule amounts in blood. The test for bacterial endotoxin is used to detect or quantify endotoxins using Limulus Amoebocyte Lysate (LAL) which is an extract of blood cells from the horseshoe crab (Limulus polyphemus). Bhat Bio-Tech follows USP <85> Bacterial Endotoxins Test and EP 2.6.14 as the methods to detect or quantify endotoxins that may be present in or on the sample. We have highly trained personnel in both techniques and in laboratory procedures to assure that any glassware or apparatus used is endotoxin free or non-pyrogenic.

Cat. No.	Service
BBCS-20134	Bacterial endotoxin testing service

Bioburden Analysis Service

Bioburden is the population of viable microorganisms on or in a sterile product. FDA and ISO guidance recommend characterization of the microorganisms recovered from a bioburden test or by environmental monitoring. Bhat Bio-Tech offers tests to provide following information:

- Gram Stain
- Cell Morphology
- Colony Morphology
- Selective culturing for specific organisms such as *Staphylococcus aureus, Pseudomonas aeruginosa, Salmonella typhimurium, Escherichia coli, Bacillus subtilis, Candida albicans,* and *Aspergillus niger*

Features:

- Aerobic Total Count
- Aerobic Total and Spore Count
- Mold and Fungi Total Count
- Anaerobic Total Count
- Anaerobic Total and Spore Count
- Test for detection of specified organisms

Cat. No.	Service
BBCS-20135	Bioburden analysis service

CONTRACT SERVICES

CELL LINE AND CELL CULTURE SERVICES

Custom Stable Cell Line Development Service

Stable cell lines play a crucial role in cell based assay development and are the main source of membrane proteins for binding assays.

Features:

- Stable cell lines for increased reliability and reproducibility
- Reliable DHFR system, used for many commercial products
- Strict growth stability and protein expression stability evaluation
- Rapid turnaround

Deliverables:

- Stable cell line
- Project report
- Quality assurance certificate

Cat. No.	Service
BBCS-20136	Custom stable cell line development service

Cell Culture Services

At Bhat Bio-Tech we can grow either adherent or suspension cells as per customer specifications. Small or large scale cultures are carried out in cell culture flasks. The following cell culture services are available

- **Adherent cell culture service**
- **Suspension cell culture service**
- **Primary cell line derivation service**

Sample requirements:

- Small suspension culture or frozen cells

Deliverables:

- Cell pellet or cell lysate or primary cell line
- Project report

Cat. No.	Service
BBCS-20137	Adherent cell culture service
BBCS-20138	Suspension cell culture service
BBCS-20139	Primary cell line derivation service

IMMUNOASSAY DEVELOPMENT SERVICES

Our immunoassay development services are project-based and designed for diagnostics and drug discovery applications. Our scientists and project managers will discuss with each client the project design, cost estimates, and delivery specifications. We offer three basic categories of services:

- Immunoassay development service for diagnostic markers,
- Immunoassay development service for drug compound discovery and
- Immunoassay development service for therapeutic antibody detection.

The immunoassay kit development is based on following methods

- ELISA assays
- Lateral flow assays
- Flow through assays
- Dipstick assays

In general our services include the following:
Antigen or immunogen preparation
Antibody production, purification and modification
Cell line generation
Immunoassay development
Reagent preparation
Assay kit manufacturing

Cat. No.	Service
BBCS-20140	Immunoassay development services

Deliverables:

- Deliverables vary depending on the types of services requested and the requirements of the assays.
- In general, we deliver cell lines, antibodies and conjugates, assay protocols, assay reagents, assay kits, experiment records, and specific data derived from a given project.