CUSTOM SERVICES
Screening & Profiling | Expression | Cell Line Development

Find the perfect solutions to advance your drug discovery programs.
Primary Manufacturer, Custom Capabilities

Table of Contents

BPS Bioscience Advantages 1
Expression & Purification 2
Protein Binding Studies - BLI 4
Screening & Profiling 5
Biochemical Screening & Profiling 6
Cell-Based Screening & Profiling 7
Cell Line Development 8
CRISPR 9
Lentiviruses 10
CAR-T Cell Development 11
Conducted In-house
- All services are conducted in the USA at our San Diego, California laboratory
- Get customized, personal support directly from the source

Break the Bottlenecks
- Services aligned with:
  - Pre-clinical drug development - Discovery Biology - Medicinal Chemistry
  - Small and large project capabilities
  - Process development & execution
  - Enzymatic, structural, stability, & binding studies
  - Cellular toxicity measurements
  - Scale-up & validation
  - Data reporting & management

Customized For Your Research Needs
- Screening & Profiling: >400 biochemical or cell-based assays manufactured by BPS Bioscience to screen your compounds of interest
- Cell Line Development: Choose from >70 cell types and >20 reporter genes
- Protein Expression: Choice of tags, labeling, host species, and QC

Committed to Excellence
- ISO 9001:2015-certified Quality Management System
Expression and Purification
High Purity, High Yield

- Expertise in expressing highly active enzymes
- Scale-up and bulk production available
- Flexible deliverables: supernatants, cell pellets, plasmids
- FPLC methods include: SEC, IEX, HIC
- Glutamine synthetase expression capabilities
- Lyophilization option
- Protein Immobilization

Expression Systems

<table>
<thead>
<tr>
<th>Baculovirus/ Sf9 Insect Cells</th>
<th>E. coli</th>
<th>Mammalian (HEK293 or CHO-K1)</th>
</tr>
</thead>
</table>

Purification Options

- Additional rounds of column purification
- Endotoxin testing
- Inclusion body purification
- Biotinylation (Avi-Tag or Side Chain) and pull down QC testing
- Protein refolding
- Phosphorylation and dephosphorylation

Protein Labeling

- Biotinylation
- Fluorescence labeling
- Antibody labeling
- Enzyme conjugation
### Customize Your Project Milestones

<table>
<thead>
<tr>
<th></th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Cloning</strong></td>
<td><strong>Expression &amp; Purification</strong></td>
<td><strong>Activity Testing</strong></td>
<td><strong>Bulk Protein Production</strong></td>
<td><strong>Additional Options</strong></td>
<td></td>
</tr>
<tr>
<td>BPS Bioscience can supply the image clone or synthetic DNA. Mutations will be introduced if needed. Confirmation of successful cloning will be completed through gene sequencing.</td>
<td>The plasmids will be transfected using lipofectamine, electroporation, or lentiviral transduction in the desired expression system.</td>
<td>Choose to test your custom protein using BPS Bioscience's collection of &gt;400 biochemical and cell-based assays, or we can develop a new custom activity assay.</td>
<td>BPS Bioscience has the capabilities to scale up protein production and produce the product in bulk. Our team of scientists will re-express and re-purify the protein.</td>
<td>Mutagenesis studies</td>
<td>Protein conjugation with probes and dyes</td>
</tr>
</tbody>
</table>
Protein Binding Studies
Analytics with BLI Services

BPS Bioscience offers Bio-Layer Interferometry (BLI) services to evaluate and analyze protein interactions. Label free analysis is a critical research method to determine the binding kinetic parameters of compounds. These studies are of key importance for pharmaceutical and biotechnological preclinical drug development.

Utilize BPS Bioscience’s BLI services for deeper insight into your protein studies through measuring binding kinetics, steady state affinity, and through target validation. BPS Bioscience will provide accurate and sensitive kinetic studies in a timely manner to further progress your research.

BLI Binding Analysis

Figure 1: BLI binding analysis of polyclonal human anti-ACE2 antibody to immobilized ACE2-His (BPS Bioscience #11003) via anti-His probes. Kd = 144 nM.

Figure 2: BLI binding analysis of SARS-CoV-2 Spike (RBD) to immobilized ACE2-His (BPS Bioscience #11003) via anti-His probes. Kd = 1.2 nM.

Available Services

- Protein immobilization
- Binding affinity measurements
- Data fitting and reporting
- Access to BPS Bioscience’s extensive protein portfolio of target proteins
- Detailed final report includes all data, methods, and results
Screening & Profiling
Biochemical & Cell-Based Assays

Evaluate Lead Compounds
Use our extensive panel of assays for biochemical and cell-based screening and profiling services.

Save Time
Avoid troubleshooting assays in-house by using our portfolio of >400 validated assays to determine potency and selectivity.

Get Detailed Results
- Extensive report with raw and analyzed data, graphs, and detailed protocols.
- Proteins and enzymes synthesized in-house to ensure the highest level of inter and intra-assay consistency.

BPS Bioscience Advantages
- Receive results within 2-3 weeks
- Fully customizable
- Numerous unique screening & profiling services
- Orthogonal screening platforms
- Standardized screening protocols
Our team of experts along with our broad services portfolio make it easy to:
- Screen for inhibitors/targets
- Select from IC50 determination or single concentration assays
- Receive data within days of compound submission
- Perform follow-up studies using the same protein lots manufactured in-house
- Get questions answered and project guidance in a time-efficient manner

**BPS Bioscience Advantages**

- Extensive immunotherapy panel, including many unique immunotherapy targets
- 1st commercially available and largest HDM panel
- Over 20 unique histone methyltransferases
- Complete PARP isozyme panel
- Largest PDE isozyme panel
- 1st complete suite of HDAC and SIRT enzymes
- Extensive bromodomain & HSP90 panels

**Biochemical Assay Target Classes**

- Acetyltransferase
- Apoptosis
- Bromodomain
- Coronavirus
- Cell Surface Receptor
- DNA Methyltransferase
- HDAC/Sirtuin
- Histone Demethylase
- Histone Methyltransferases
- HSP90
- Immune Checkpoints
- Kinase
- Metabolic Enzymes
- Methyl-lysine Reader
- PARP
- PCSK9
- PDE
- Protein Phosphatase
- Protease

**Inhibition of PD-1[B]: PD-L1 Interaction by PD-1 Antibody**

<table>
<thead>
<tr>
<th>Luminescence</th>
<th>Log([anti-PD-1 Ab]/nM)</th>
<th>IC50 = 1.2 nM</th>
</tr>
</thead>
<tbody>
<tr>
<td>3500</td>
<td>0</td>
<td>2000</td>
</tr>
<tr>
<td>2000</td>
<td>3</td>
<td>1500</td>
</tr>
<tr>
<td>1000</td>
<td>-3</td>
<td>500</td>
</tr>
</tbody>
</table>

**CD73 Activity**

<table>
<thead>
<tr>
<th>% Activity</th>
<th>AMPCP Log [μM]</th>
<th>IC50 = 6 μM</th>
</tr>
</thead>
<tbody>
<tr>
<td>100</td>
<td>-1.5</td>
<td>50%</td>
</tr>
<tr>
<td>50</td>
<td>-0.5</td>
<td>30%</td>
</tr>
<tr>
<td>10</td>
<td>1.5</td>
<td>10%</td>
</tr>
</tbody>
</table>

**BCL-XL Activity**

<table>
<thead>
<tr>
<th>% Activity</th>
<th>A-1155643 Log(μg/ml)</th>
<th>IC50 = 0.01 μg/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>100</td>
<td>-4.3</td>
<td>40%</td>
</tr>
<tr>
<td>50</td>
<td>-3</td>
<td>20%</td>
</tr>
<tr>
<td>10</td>
<td>-2</td>
<td>10%</td>
</tr>
</tbody>
</table>
BPS Bioscience has developed a number of recombinant reporter cell lines to screen inhibitors or activators of various cell signaling pathways. If you are developing inhibitors specific to one of our pathway reporters, these cell-based systems offer a more complex and physiologically relevant setting than cell-free systems. This screening service is also a great tool that can be used to tease out the specific pathway that a particular compound is targeting. Screen against our entire portfolio or select a few pathways.

### Cell-Based Assays

- CAR-T Cell Screening
- Cytokine Assays
- Ion Channel Assays

- Tumor Proliferation Assays
- Reporter Gene Assays

### Cell-Based Assay Target Classes

- Cell Signaling Pathways
- Hedgehog Pathway
- Histone Deacetylases
- Immune Checkpoints
- Nrf2 Antioxidant Pathway

- NF-κB Pathway
- Phosphodiesterases
- T-Cell Activation
- Wnt/β-catenin Pathway
Cell Line Development
Customized for your Research Needs

- Delivery of multiple stable clones for internal validation
- >70 available cell types and >20 possible reporters
- Use for antibody and compound screening

Cell Line Formats

Expression Cell Lines

Reporter Cell Lines

Lentivirus Generated

Project Milestones

1. **Molecular Biology**
   Expression vectors will be generated using available image clones, or through the use of synthetic DNA to stably transfect the gene of interest.

2. **Selection and Pool Generation**
   Parental cells will be transfected with the expression vector the desired targets. The cell pool will be selected for using antibiotics.

3. **Limiting Dilution and Clonal Selection**
   Based on the results of the initial pool testing, the cell pool will be diluted and a single cell-derived clone will be selected.

4. **Confirmation of Expression**
   The expression level of the target protein will be analyzed via Western Blot or FACS.

5. **Functional Validation**
   Cells will be treated with a reference control compound to obtain dose-response titration data.

6. **Stability Testing**
   The desired number of clones will be selected for passage stability testing. Mycoplasma testing and cell banking services are also available.
CRISPR
Knock-out or Knock-in

- Introduce a specific point mutation or add a tag to your endogenous gene of interest
- Knock-out your gene(s) of interest for mechanistic or screening studies
- Customized lentivirus generated cell lines can be used for knock-down or knock-out cell pools

Knock-out or Knock-in

![CRISPR mechanism diagram]

Project Milestones

1. Molecular Biology
   We will synthesize three short guide RNA sequences for knock-out cell lines. We can also design the HDR template for knock-in cell lines.

2. CRISPR Transfection
   Depending on the cell-type, cells can be transfected via electroporation, liposome-based transfection, or viral infection.

3. Limiting Dilution
   Based upon the results of the initial pool testing, the cell pool will be clonally diluted and the clones will be expanded.

4. Confirmation of Expression
   The expression level of the gene of interest will be analyzed via Western Blot or FACS.

5. Confirmation
   Genes showing loss of expression of the gene will be analyzed through genomic sequencing. For knock-in mutations, functional validation is available.
**Custom Lentivirus Services**

- Utilize our services for a custom lentivirus generated for your research needs
- Customize your lentivirus and cell line with reporters, selection markers, variants, and mutations
- Generate custom overexpression and reporter cell lines using your lentivirus
- Choose knock-out/knock-in cell lines or integrating/non-integrating lentiviruses

**Project Milestones**

<table>
<thead>
<tr>
<th>Milestone</th>
<th>Description</th>
</tr>
</thead>
</table>
| 1 | Molecular Biology  
BPS Bioscience will generate lentiviral vectors using available image clones, or through the use of synthetic DNA |
| 2 | Lentivirus  
The custom lentivirus is manufactured for the development of the stable cell line |
| 3 | Selection and Pool Generation  
Parental cells will be transduced with lentivirus. The cell pool will be selected for using antibiotics. |
| 4 | Clonal Selection  
Based on the results of the initial pool testing, the cell pool will be diluted and the single cell derived clone will be selected. |
| 5 | Confirmation of Expression  
The expression level of the target protein will be analyzed via Western Blot or FACS. |
| 6 | Functional Validation  
Cells will be treated with a reference control compound to obtain dose-response titration data. |
| 7 | Stability Testing  
The desired number of clones will be selected for passage stability testing. Mycoplasma testing and cell banking services are also available. |
CAR-T Cell Development
Comprehensive CAR-T Services

- Primary screening and validation of CAR activity using reporter cell lines
- Cytokine detection from CAR-T cells
- CAR-T cell killing assays

Researcher provides Ab sequence against cancer antigen

Engineering & validation of ScFv for specificity and affinity

CAR Lentivirus production and initial validation

T-cell preparation & transduction

Functional validation of CAR-T cells

Anti-CD19 CAR/Jurkat NFAT Stable Pool in Co-culture Reporter Assay

IFNγ Production of Anti-CD19 CAR-T (CD4:CD8=1:1) Cells Induced by CD19/CHO Cells (E:T=10:1)

CD19 Luc/CHO Target Cell Induced Killing by CAR-T

Stimulations

Fold Induction

Fold Change in IFNγ Production

Target Cells

Effector cells:Target cell = 10:1