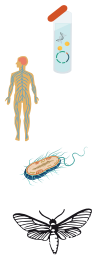
PROTEIN SERVICE

Excellency on your request

Module 1
PROTEIN EXPRESSION

- Cell-free Systems
- Hek293 cells
- *E. coli*
- Insect System



Module 2
PROTEIN SOLUBILIZATION

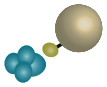
- Detergents
- Synthetic polymers

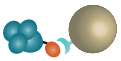
Module 3
PROTEIN STABILIZATION

- Detergent
- MSP Nanodiscs
- Synthetic Nanodiscs & Amphipols

Module 4
PROTEIN PURIFICATION

- Protein specific chromatography
- Affinity chromatography (Affinity tag)





Module 5
PROTEIN CHARACTERIZATION

- Dynamic light scattering
- Size exclusion chromatography
- Surface plasmon resonance
- ELISA

Module 6
STRUCTURE DETERMINATION

- Crystallization
- Cryo-EM

Projects tailored to your needs

Cube Biotech

Module 1 - Protein Expression

We offer four different methods of expression systems.

E.coli: The standard expression systems. Best suited for fast projects with high yield that do not require eukaryotic-exclusive post-translational modifications.

Cell free expression: A perfect match with pre-assembled nanodisc for membrane protein expression since it is an open system with no cell membrane. The membrane protein is co-translationally integrated into nanodisc. We recommend it for toxic proteins and membrane proteins that need to be stabilized in all types of nanodiscs.

Baculovirus insect system: The workhorse, for a higher output and shorter timelines than mammalian cells.

Hek293 cells: For homologue human protein expression and comprehensive post-translational modifications. We offer transient transfection in Hek293 suspension cultures.

Module 2 - Protein Solubilization

As experts for membrane proteins we offer services specialized to the challenges that membrane proteins present.
For the solubilization of membrane protein we offer two options:

Detergents: Detergents are the classic approach to solubilize membrane proteins. Using them requires a screening process to identify the detergents that fits your membrane protein the best. Of course Cube Biotech will perform this screening process.

Synthetic Polymers: Polymers like SMA and DIBMA can form complexes that are called synthetic nanodiscs. During the last decades they have increasingly started to replace detergents as the most popular tool for membrane protein solubilization. Their key advantage: They combine the essential steps of membrane proteins solubilization and subsequent stabilization. This basically fuses our modules 2 and 3.

Module 3 - Protein Stabilization

A second essential step for membrane protein sciences is the stabilization of the membrane protein after solubilization. Cube Biotech offers three options for this:

Detergents: The traditional way of membrane protein stabilization. Similar to the solubilization step, membrane proteins are constantly being kept in a detergent micelle. However this form requires constant addition of the detergent, otherwise the protein will precipitate over time.

MSP nanodiscs: Derived from apolipoprotein-1, membrane-scaffold proteins (MSPs) form disc-shapes structures composed of phospholipids. The lipid composition of these MSP nanodiscs can be 100% controlled and due to the uniformity of the disc-diameters these samples can be used for Cryo-EM.

Synthetic nanodiscs: As mentioned in module 2, synthetic nanodiscs combine solubilization and stabilization of membrane proteins. The polymers SMA and/or DIBMA cut the membrane protein out of the cell membrane similarly to a cookie cutter, while simultaneously keeping the protein stable in a disc-shaped structure.

Module 4 - Protein Purification

The purification of your protein of interest is usually the last step in a service project. As experts for protein purification, Cube Biotechcan take two approaches to this task.

Affinity tags: The easiest way to purify a protein is to add an affinity tag and purify it with matching affinity resin or MagBeads. Cube Biotech will recommend which tags suits your project the best.
The affinity chromatography is usually followed by SEC (Size Exclusion Chromatography).

Protein specific affinity chromatography: Sometimes the addition of an affinity tag is not the best way to approach protein purification. In such cases Cube Biotech can create affinity beads that are tailored to your specific protein of interest.

Example: We once added cobra venom to agarose resin beads. The venom has a natural affinity towards the Acetylcholine receptor AchR. We were then able to purify the protein with a high yield, without any modification on the protein itself.

Module 5 - Protein Characterization

Cube Biotech also offers services that go beyond the purification of your protein of interest. We can help with the characterzation of your protein. Techniques we apply in-house include:

Dynamic Light Scattering (DLS): A method to determine the diameter of your protein by measuring how light is scattered by the protein.

Surface Plasmon Resonance (SPR): We identify interaction partners of your proteins and measure the intensity of this interaction.

ELISA: Enzyme-linked Immunosorbent Assays are used to identify an antibody's reaction towards a certain protein. It can also be used to determine the amount of antibodies that react to certain protein in a blood sample.

Thermostability: We can determine how stable your protein of interest is under certain temperatures.

Protein Stability Assays: We can research which other conditions have an effect on your protein's stability.

Module 6 - Protein Structure Determination

Cube Biotech also offers service beyond the purification of your protein of interest. Beyond that step we also help with the characterzation of your protein. Techniques that we use in-house include:

Cryo-EM: We can prepare your protein sample for Cryo-EM and if ypu would like us do the Cryo-EM for you.

Cubic Phase Crystallization: The classical way to solve 3D structures of membrane proteins. Cubic phase crystallization has already solved countless tertiary structures of membrane proteins. Our company is patent owner of the CIMP (controlled in-meso phase crystallization) method which combines the lipid cubic phase with vapour diffusion. Therefore we have gathered lots of experience with this method.

Fun fact: Our Company was named after the cubic phase crystallization method.