Services Offered by the Centre of Molecular Structure at IBT

Our Centre offers techniques of production, characterisation, and measurements of samples of biological macromolecules. Contact us with questions or requests at

magdalena.schneiderova@ibt.cas.cz.

All prices are in CZK and do not include VAT. A detailed price list is available upon request.

Pure proteins and their intermediates

Preparation of plasmid DNA constructs

Input: Target plasmid, DNA encoding the target protein, or purchased gene from

common suppliers.

Output: Finished plasmid DNA, sequencing results confirming cloning success.

Time: Approx. 2 weeks

Approximate price: From 21 000 (cloning of 1 construct in one step) to 40 000 (multistep cloning

of 1 construct)

Expression of protein constructs in prokaryotic expression systems

Description: Various *E.coli* strains, expression optimization, bulk expression.

Input: Expression plasmid.

Output: Bacterial pellets containing the desired protein. Optimized expression

conditions.

Time: Several days to a week

Approximate price: From 10 000 (1 liter of culture)

Purification of expressed proteins for further use

Description: Affinity chromatography, ion-exchange chromatography, gel permeation

chromatography, etc.

Input: Protein, in the form of a soluble sample, cell lysate or cell pellet.

Output: Purified protein sample.

Time: Several days to several weeks (in case of optimizations).

Approximate price: From 8 000 (affinity chromatography), depending on the method

<u>Properties of biomolecules: size distribution, structure, stability and conformation</u>

Dynamic light scattering

Description: Offers measurement of the particle size distribution in solution, particle

oligomerization, aggregation. Particle size range 1-1000 nm.

Intut: Min. 1 μl of solution, min. concentration 0.5 mg/ml.

Output: Particle size distribution

Time: Next day

Approximate price: 1 000 (1 hour, ~ 6 samples per hour)





Small Angle X-ray Scattering (SAXS)

Description: In solution, it is possible to determine size of the particle, molecular weight,

structural information at the level of particle shape (low resolution 3D structure), sample quality (aggregation). Analysis of mixture of components

with known structure. The measurement can be coupled to liquid chromatography (SEC). Screening of various conditions is possible.

Input: Protein/DNA/RNA in monodisperse solution, concentration series 1-10 mg/ml,

min. 3x 20 µl.

Output: Sample quality (aggregation), particle size and molecular weight estimate,

ratio of components in the case of mixtures.

Time: Basic measurement and analysis - next day; advanced analysis - several days

Approximate price: From 2 000 to 5 000 per sample (depending on analysis level)

Differential scanning fluorimetry

Description: Characterization of protein thermostability using intrinsic tryptophan and

tyrosine fluorescence to measure melting transitions.

Input: Min. 10 µl of protein solution, concentration from 0.1 mg/ml.

Output: Plot of melting transition, melting temperature

Time: Next day

Approximate price: From 300 per sample

Differential scanning calorimetry

Description: Characterization of thermostability of biopolymers.

Input: Min. 500 µl of biomolecule solution, concentration from 1 mg/ml.

Output: Thermodynamic parameters of melting transition.

Time: Next day

Approximate price: From 5 000 per sample

Characterization of interactions of biomolecules

Microscale thermophoresis

Description: Characterization of affinities of biomolecular interaction

Input: Min. 40 μl of protein at concentration 100*Kd. Min 200 μl of labeled molecule

in the order of the Kd.

Output: Binding curves, Kd.

Time: Next day

Approximate price: From 4 000 to 20 000 per sample (depending on technique and device)

Surface plasmon resonance

Description: Characterization of affinities and rates of interaction of biomolecules

immobilized on the sensor chip with molecules in solution. Real-time

interaction measurements.

Input: Min. 200 µl of protein solution at concentration 0.2 mg/ml. Concentrated ligand

solution.

Output: Binding curves and calculated Kd, kon a koff.





Time: 7 days

Approximate price: From 17 000 (1 chip, measurement of interaction of two interaction partners)

Bio-layer interferometry

Description: Characterization of affinities of interaction of molecules (immobilized on the

sensor) with molecules in solution. Real-time interaction measurements.

Input: Min. 200 μl of protein solution at concentration 0.1 mg/ml. Concentrated ligand

solution.

Output: Binding curves and calculated Kd, kon a koff.

Time: Next day

Approximate price: From 2 000 per measurement

Isothermal titration calorimetry

Popis: Determination of thermodynamic parameters of an interaction. No labeling or

immobilization is needed.

Input: 300 μl of molecule and 60 ul of ligand, μM concentration.

Output: Thermodynamic parameters of an interaction: Kd, stoichiometry, enthalpy and

entropy.

Time: 3-5 days

Approximate price: From 5 000 per 1 interaction measurement

Spectrophotometric and luminescence study of biomolecules

Spectrophotometry and luminescence

Description: Sample characterization by the measurement of their absorbance and

fluorescence. Allows studying enzyme activity, interaction with ligands (by the measurement of fluorescence polarization), FRET, ELISA, to determine protein and DNA concentrations. Can be measured in cuvettes and plates.

Input: Min.100 μl of biomolecule solution.

Output: Spectra, kinetics and its analysis.

Time: Up to one week

Approximate price: From 1 500 (measurement only, no sample preparation)

Fourier Transform Infrared Spectroscopy

Description: Characterization of samples in the infrared spectral region by measuring

absorption frequencies of some specific chemical groups (amide bond in proteins). It allows to determine protein secondary structure, interaction with

ligand and structural changes.

Input: Min. 30 µl biomolecule solution at concentration 3-5 mg/ml.

Output: FTIR spectrum and its characterization. Information on the secondary

structure and the interaction of the molecule with the ligand.

Time: One week

Approximate price: From 1 500 (1 hour of measurement without analysis)





Secondary structure of proteins and conformation of biomolecules

Circular dichroism

Description: Determining the secondary structure types in protein: approximate fractions -

% of helix, sheet, turns, and random structures. Testing of the impact of point mutations on structural integrity and comparing the secondary structure of protein in different conditions (temperature, pH, salt, concentration, ligands,

etc.).

Input: Min. 100 μl molecule solution, concentration around 0.2 mg/ml.

Output: Spectra of circular dichroism (ellipticity at different wavelengths) with the

analysis of secondary structure of proteins.

Time: Next day

Approximate price: 1 500 per sample

Mass spectrometry of biomolecules

Identification and quantification of proteins in mixtures

Description: Identification of proteins in complex mixtures and their relative quantification.

In-gel or in-solution analysis.

Input: SDS PAGE gel or solution, min. amount of protein: several μg.

Output: List of identified proteins and their relative abundance.

Time: 2-3 days

Approximate price: from 5 000 per sample

Characterization of proteins

Description: Determination of recombinantly expressed or isolated proteins.

Characterization of protein modifications.

Input: Protein solution, min. amount: several µg.

Output: Exact molecular weight of proteins and their forms.

Time: 1-2 days

Approximate price: From 5 000 per sample

Characterization of protein-protein interactions

Description: Determination of protein-protein interaction areas and structural changes by

hydrogen-deuterium exchange and chemical cross-linking in combination with

mass spectrometry.

Input: Protein in solution, min. amount: units of mg.

Output: Description of interaction area and protein structural changes.

Time: 3-7 days

Approximate price: From 10 000 per 1 complex analysis





High-resolution 3D structure of (bio)molecules

Crystallization of proteins, nucleic acids, and complexes

Description: For the subsequent experimental determination of the 3D structure at atomic

level by diffraction analysis, (using the home source or synchrotron).

Input: At least 200 µl of sample, with a minimal macromolecule concentration of 5

mg/ml, optimally 10-20 mg/ml. Purity at least 95 %.

Output: Crystals suitable for diffraction analysis.

Time: Weeks to months (depending on the complexity of the project and the

methods chosen).

Approximate price: From 2 000 for 1 screening (1-3 proteins), a typical campaign (1 protein)

requires e.g. 5 screenings + manual work

Determination of 3D structure of biological (macro)molecules at high resolution

Description: Single crystal X-ray diffraction. Measurement of diffraction data of molecular

crystals, at cryo-conditions or at room temperature. Measurement in

crystallization plates. Upon agreement, it is possible to measure diffraction of

small molecule crystals.

Input: Single crystal of the molecule (in the crystallization plate or vitrified in liquid

nitrogen) of size at least 50 µm.

Output: High resolution structure of the molecule or the complex, typically 1.5-3.5 Å

(atomic level). Diffraction data, symmetry space group, 3D maps of electron

density, atomic coordinates of the molecule (PDB or mmCIF format).

Time: Weeks or months, depending on the project complexity

Approximate price: From 25 000 for the least demanding projects

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