

MACROMOLECULAR AFFINITY CHARACTERIZATION FACILITY

DIVISION OF BIOLOGICAL SCIENCES

INDIAN INSTITUTE OF SCIENCE

S
P
R

ABOUT

This facility was established in 1997 with support from DBT, Govt. of India. There are 5 instruments in the facility. They are Biacore2000, Octet Red96, Biacore T200,Monolith NT.115 & Prometheus NT.48.

PEOPLE

Faculty

Prof. Raghavan Varadarajan , Molecular Biophysics Unit

Phone: +91- 080- 22932612 E-Mail: varadar@iisc.ac.in

Facility Incharge

Mrs. Jisna V, Technical Assistant.

Phone: +91-080-22932299 E-Mail: jisna@iisc.ac.in

We have several different instruments to measure macromolecular interaction kinetics and thermodynamics. These comprise of two instruments that use Surface Plasmon Resonance (SPR), one that uses Biolayer Interferometry, one that uses Microscale Thermophoresis and the one that uses Differential fluorometry . The facilities are used by multiple faculties from across the division as well as some external users.

SURFACE PLASMON RESONANCE (SPR) is a non-invasive optical measuring technique which measures the mass concentration of biomolecules in close proximity to a specially prepared surface. The technique does not require any labelling of the interacting components. The response is largely independent of the nature of the biomolecule, so that all steps in an interaction analysis may be followed with the same detection technique. The process involves flow of one molecule (analyte) over a surface with an immobilized binding partner (ligand). Binding results in a change in refractive index at the surface which is transduced to an optical signal. Dissociation of bound analyte is monitored by flowing buffer over the surface.



They are two very robust SPR instruments. These instruments use a single chip with four surfaces to which different molecules can be attached, making it possible to simultaneously determine 3 different affinities.



Octet is multi-functional instrument platform based on Bio-Layer

Interferometry (BLI), a label-free technology that measures molecular interactions in real time for the purpose of detection, quantitation and kinetic analysis. Octet has the advantage of negligible analyte consumption and low consumable costs and was procured with funding from MHRD. The Octet system is ideally suited for 96-well characterization of protein-protein and protein-small molecule binding kinetics, and for the determination of protein concentrations and titer. This uses tips which are coated with wide selection of biosensor chemistries available for highly specific binding of target molecules in a sample.

Monolith NT.115(Nano temper technologies)



Monolith NT.115 is a laboratory instrument from NanoTemper Technologies that uses MicroScale Thermophoresis (MST) to precisely measure biomolecular interactions and binding affinities in solution. MST works by creating a tiny temperature gradient with an

infrared laser in a capillary, and detecting how molecules move in response—changes that occur when two molecules bind. The system is highly sensitive, can work in close-to-native conditions without immobilizing the molecules, and can quantify interactions from very small ligands (including ions) to large complexes like ribosomes across a broad affinity range. It requires only small sample volumes ($\approx 4 \mu\text{L}$ per capillary) and supports a variety of fluorophores for flexible experimental design.

Prometheus NT.48(Nano temper technologies)



Prometheus NT.48 is a protein stability analysis instrument from NanoTemper Technologies that uses nanoDSF (nano Differential Scanning Fluorimetry) to measure thermal and chemical stability of proteins in a label-free way. It monitors intrinsic tryptophan and tyrosine fluorescence as proteins unfold when heated or exposed to

denaturants, providing high-resolution unfolding curves and precise stability data such as melting temperatures (Tm) and aggregation onset. The system can analyze up to 48 samples at once in small capillaries ($\approx 10 \mu\text{L}$ each) and works over a broad concentration range, making it suitable for protein engineering, formulation screening, membrane proteins, antibodies and other biopharmaceutical research without needing external dyes or modifiers.

Experiments

For Biacore2000 and BiacoreT200 the chips have to be procured by the individual investigators from Wipro GE Healthcare Pvt. Ltd. The tips for Octet Red96 and Capillaries for Monolith as well as prometheus instruments will be provided by us and will be charged.

Requests

The request can be made via email or call. The slots would be given according to the availability of the instruments.

Payments

The charges are subsidised for academic and Govt. users. The request should be made from the investigators.

For IISc users the charges will be Rs.500/- for half a day and

Rs.1000/day. For users from external institute the charges will be Rs.3000/day + 18% GST. For Companies and other users it will be charged Rs.10,000/day + 18% GST. The invoice will be provided and the payment should be made online to Institute's bank account. The Bank account details will be provided along with the invoice.