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HiLIFE
BiocompleX

BIOMOLECULAR COMPLEX PURIFICATION: ASYMMETRIC FLOW FIELD FLOW FRACTIONATION

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ASYMMETRIC FLOW FIELD - FLOW FRACTIONATION (AF4)

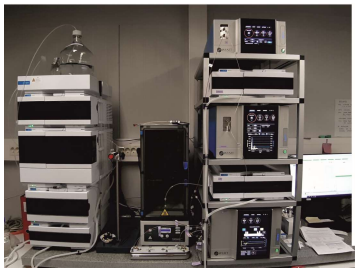
In AF4 separation is based on hydrodynamic radius, R_H. It is achieved by applying cross flow and channel flow. AF4 enables:

- Gentle separation without stationary phase;
- Versatile mobile phase compositions;
- Broad separation range: nm to µm;
- Applicability to polydisperse samples;
- Multiple online detectors (MALS, DLS, UV, dRI, FL) for biophysical characterization of sample components:
  - M_w, R_G, R_H, PDI, shape, particle concentration etc.;

Electrical AF4 (EAF4) is extension to AF4 where separation by size & charge provides data on charge and zeta potential of sample populations.

BIOCOMPLEX AF4

- Temperature controlled channels (+5°C above ambient temperature, up to +75°C)
  - Analytical and semipreparative channels
  - Dispersion inlet channel for aggregation prone samples
  - Mobility channel for EAF4
- Ultrafiltration membranes with various MWCOs and materials
  - Regenerated cellulose membranes: 2, 10 and 30 kDa
  - Polyethersulfone membranes: 10 and 30 kDa
- Spacers with various thicknesses
- Standards: BSA, polystyrene beads: R_H 10, 20, 30, 50, 60, 142 nm
- Detectors:
  - DAWN 18 angle multiangle light scattering (MALS) detector (Wyatt)
  - QELS dynamic light scattering (DLS) detector (Wyatt)
  - UV-Vis and Fluoresecence Detectors (Agilent)
  - Optilab dRI detector(Wyatt) Dilution control module (to increase signal intensity at the detectors)
- Fraction collector (temperature controlled)
- Autosampler (temperature controlled)
- ASTRA 8.2 and Vision 3.2 software



Biocomplex Neon Eclipse AF4 with detectors.

Biocomplex provides facilities for ultracentrifugation, asymmetric field flow fractionation, batch mode DLS and chromatography. Our technologies can be used to analyze and purify large macromolecular complexes such as **nanoparticles, membrane vesicles**, **protein complexes, polymers** etc. from biological, synthetic and environmental samples.

LOCATION: Biocenter 1, B-building, 6th floor, Viikki Campus, (Viikinkaari 9), University of Helsinki, Finland

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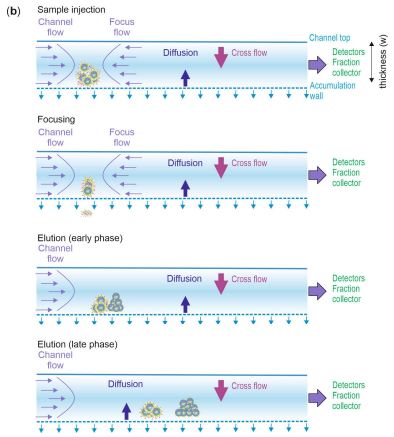
https://hilife-infra.ilab.agilent.com/account/login

WORKING PRINCIPLE



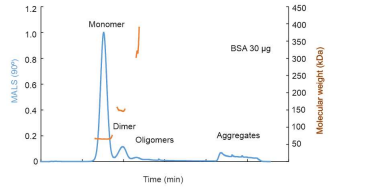
AF4 channel is thin and ribbon-like. Flow in the channel is laminar with parabolic flow profile. Separation is achieved by applying cross flow force that is perpendicular to the channel flow. Channel bottom contains a porous frit that is covered with an ultrafiltration membrane. Together they form the accumulation wall.

Experiments initiate with sample injection and focusing that concentrates the sample and promotes relaxation according to the diffusion coefficients of the sample components. During elution, the sample components migrate towards the detectors according to their diffusion coefficients. Small sample components have larger diffusivity than large sample components. This makes them to diffuse away from the accumulation wall to the higher velocity flow profiles of the channel and to elute before large sample components.

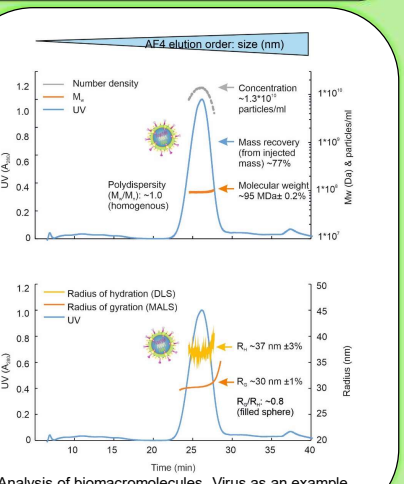
Biocomplex has also the following services:

- Ultracentrifugation: 6 ultracentrifuges and 8 rotor types (fixed angle and swing out, ThermoScientific / Sorval)
- BioComp Gradient master for gradient making
- BioComp Piston Gradient Fractionator with Triax flow cell: A260, A280, eGFP or Cy5.
- ÄktaPure25M chromatography instrument
- DynaPro NanostarII (Wyatt) batch mode dynamic light scattering instrument

APPLICATIONS



Separation of monomers from dimers and oligomers. BSA as an example.



Analysis of biomacromolecules. Virus as an example.