FIDA Technology
Rapid protein analysis using nanoliter samples of native composition

FIDA - enabling development and characterisation of biopharmaceuticals

Flow Induced Dispersion Analysis (FIDA) is a new proprietary technology developed by FIDA-Tech Aps for quantification and characterisation of proteins (including biologics), early detection of immune responses (including immunogenicity testing) and affinity assessment. FIDA is characterised by being fast (minutes), requiring very small sample amounts (nL-µL), and being exceptionally tolerant to the sample matrix. Contrary to most other procedures, the FIDA methodology is based on binding in homogenous solution; complications related to non-specific surface adsorption and challenging assay development is therefore avoided. The unique features of FIDA enable characterisation and quantification in native (biorelevant) environments, in-built assay quality control and automation.

FIDA principle
FIDA is based on dispersion analysis of a parabolic hydrodynamic flow profile. The dispersion of an injected sample zone of an indicator depends on flow channel dimensions, flow rate and apparent diffusivity of the indicator. The FIDA Analyser allows accurate quantification of the apparent diffusivity of the indicator in different environments. Particularly, the interaction with an analyte such as an antibody based drug may be quantified. The principle is illustrated in figure 1.

The apparent diffusivity is fitted to a binding isotherm to obtain the non-covalent binding constant. Additional information includes size determination of the indicator and the formed complex. The binding curve is also used to quantify proteins in solution such as biologics and autoantibodies developed in relation to immune responses.

FIDA principle diagram:
- Unbound indicator
- Bound indicator
- Flow
- Indicator + Analyte = Complex
- Indicator + Analyte = Complex
- Flow

Figure 1 The dispersion of the indicator in a uniform flow channel (top). When the indicator interacts with an analyte its apparent size increases causing an increased dispersion (bottom). The change in apparent dispersion is used to obtain non-covalent binding constants and protein concentration.

Figure 2 The apparent diffusivities are fitted to a binding isotherm to obtain diffusivities and non-covalent binding constants.
Key Features
- Detection in native conditions (sensitivity pM - mM)
- Sizes of affinity complexes from 1 - 300nm (hydrodynamic radius)
- Larger aggregates can be detected qualitatively
- Fast assays (minutes)
- Swift assays development (hours - days)
- Low sample volumes (nL - µL)
- Built-in quality control - high level robustness
- Walk away automation as standard

Applications in BioPharma
- Detection of auto-antibodies (protein - DNA interactions) in up to 85 % plasma
- Protein - protein interactions
- Drug - DNA interactions
- Assessment of binding kinetics
- Protein binding and quantification (small molecule - protein interactions)

![Figure 3 Taylorgram of Fluorescein (dotted line) and the fluorescein HSA complex (full line, left). Binding isotherm allowing quantification of affinity constant (K) and diffusivity of the indicator (fluorescein) D_I and the HAS - fluorescein complex, D_{IA} (right).](image)

$$D_I = 4.3 \times 10^{-10} m^2 s^{-1}$$

$$D_{IA} = 6.3 \times 10^{-11} m^2 s^{-1}$$

$$K = 2.8 \times 10^4 M^{-1}$$

Literature
Nicklas N. Poulsen, Nina Z. Andersen, Jesper Østergaard, Guisheng Zhuang, Nickolaj J. Petersen and Henrik Jensen; Flow Induced Dispersion Analysis Rapidly Quantifies Proteins in Human Plasma Samples Analyst, 2015, 140, 4365-4369.


Henrik Jensen and Jesper Østergaard; Flow Induced Dispersion Analysis Quantifies Noncovalent Interactions in Nanoliter Samples J. Am. Chem. Soc., 2010, 132, 4070-4071