Biacore™ X100

LABEL-FREE INTERACTION ANALYSIS

Biacore™ X100 (Fig 1) is an automated and versatile system for comprehensive, label-free analysis and characterization of biomolecular interactions in real-time. Biacore™ X100 can be used for a broad range of applications, including structure-function studies, pathway analysis, biomarker discovery and validation, drug target identification and validation, and assay development

Biacore™ X100 delivers:

- **Interaction data with added dimension:** Real-time interaction data enables kinetic characterization, highly accurate affinity determination, and detection of weak and transient binding events.
- **Versatility:** Supports many assay formats and the study of a variety of biomolecules including small molecules, proteins, DNA, and cells. These interactions can be measured in buffer solutions or in crude environments such as serum.
- **Convenience and efficiency:** Workflow oriented software provides a structured approach to assay development, minimizes hands-on time, and generates reliable data without compromising flexibility for the best assay set up. Preprogrammed workflows for supporting kits, surfaces, and reagents provide solutions for a multitude of assays.
- **Quality and sensitivity:** High quality hardware, software, and a high-sensitivity system ensure reliable detection of genuine interactions and extend the application range while minimizing sample consumption.

Biacore™ X100 Plus Package

The addition of Biacore™ X100 Plus Package extends functionality of the system, adding capabilities that enable you to:

- Study interactions involving small molecules in organic solvents
- Measure concentration based on specific binding
- Analyze interactions over a range of temperatures

Fig 1. Biacore™ X100 is an integrated system for quickly and securely generating reliable molecular interaction data.

Biacore™ X100 overview

Analysis of kinetic properties

Binding affinity can be resolved into an on-rate, which is primarily driven by molecular recognition, and off-rate, which is controlled by complex stability. This allows for a better understanding of molecular interactions and biological processes. Kinetic characterization of interactions can be performed using the traditional approach, with one sample concentration at a time followed by regeneration of the surface between each analysis cycle, or the innovative single-cycle approach. Biacore Single-Cycle Kinetics (SCK)™ is unique to Biacore™ systems and reduces time to results by eliminating one step in optimizing assay conditions (Fig 2).

cytiva

In conjunction with the use of a capture kit, Biacore™ X100 enables you to produce kinetic data in less than an hour — from assay development to final data. For capture assays, Biacore Single-Cycle Kinetics (SCK)™ typically reduces reagent consumption by a factor of four, reducing cost and time. Furthermore, biomolecular investigations that were not previously feasible due to the inability to find suitable regeneration conditions can now be performed using Biacore Single-Cycle Kinetics (SCK)™. Biacore™ X100 can measure kinetic constants over a broad range — from the fastest on-rates to the slowest off-rates:

- On-rates from 10^3 to 10^7 $M^{-1}s^{-1}$
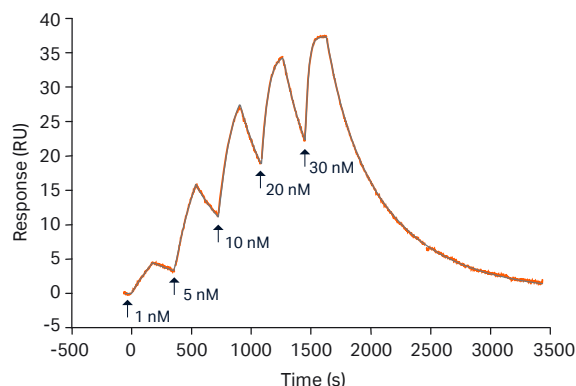- Off-rates from 10^{-5} to 10^{-1} s^{-1}

Fig 2. A sensorgram depicting a run in Biacore Single-Cycle Kinetics (SCK)™ mode. Increasing sample concentrations were injected consecutively in the same analysis cycle without a regeneration.

High sensitivity

Biacore™ X100 provides highly sensitive detection of biological interactions (Fig 3). This feature is particularly useful for the investigation of the following:

- Analysis of interactions between drug compounds and target proteins
- Biomolecules such as membrane proteins that require special conditions to remain active
- Biomolecules that are present in minute quantities, such as biomarkers in serum
- Resolution of slow off-rates, where very small decreases in signal may need to be accurately monitored during the dissociation phase
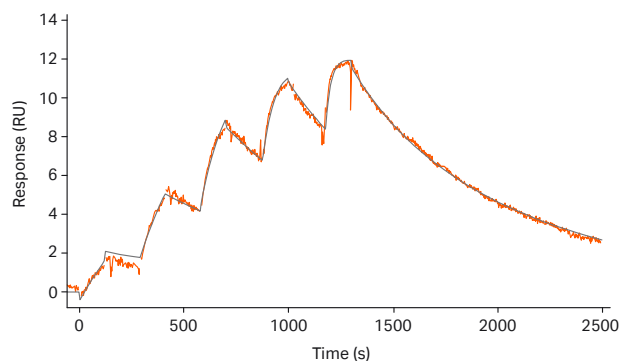

Fig 3. Low noise level enables reliable kinetic characterization at low signal levels. This helps ensure that signals are due to genuine interactions.

In addition, the high sensitivity of Biacore™ X100 facilitates improved kinetic analysis, where a low surface density of the immobilized biomolecule is generally desirable in order to avoid potential artifacts.

Display of real-time data

Biacore™ X100 provides a continuous display of the real-time data output (Fig 4), from assay development to binding analysis. This provides reassurance that the experiment is proceeding as normal, or if unexpected results are seen, you can decide whether the data is novel and worthy of further investigation, or represents a problem that requires troubleshooting.
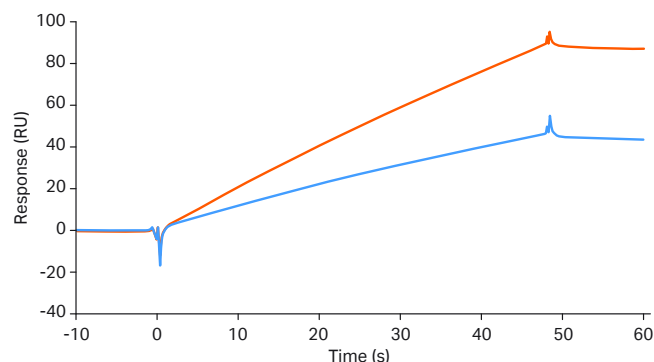

Fig 4. Overlay of two runs of the same sample 16 cycles apart. The lower slope of the curve in the later cycle indicates that the immobilized protein has lost its activity; probably due to the use of regeneration conditions that were too harsh.

A structured approach to assay development

Biacore™ X100 software incorporates the concept of guided workflows for specificity, affinity and kinetic analysis (Fig 5). This concept provides a structured approach to assay development and enables you to generate reliable data from day one without compromising assay flexibility.
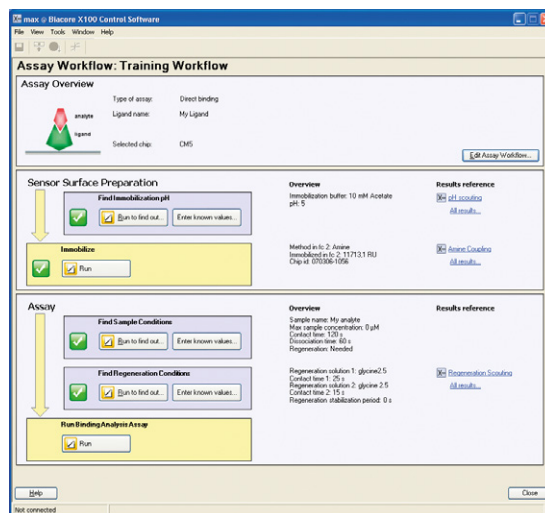

Fig 5. Workflow-oriented software simplifies assay development and minimizes hands-on time.

Based on the properties of the samples, the software suggests alternative assay configurations. After an alternative is selected, the software outlines the entire assay workflow from attaching one molecule to the sensor surface to measuring the interaction parameters for any number of interaction partners and evaluating the data. Each step of the workflow is supported by a specific software wizard, and guidance in the form of recommendations and explanations of concepts is provided to you at every step. Additionally, you have access to a built-in application library that can be reached with just one click.

Reliable data analysis

Biacore™ X100 evaluation software provides efficient data analysis in an automated and intuitive manner. Quality controls (Fig 6) are applied automatically to data fitting of kinetic parameters and if necessary, helpful suggestions or alternate models are provided for further investigation. In addition to binding profiles (i.e., sensorgram views), Biacore™ X100 provides a large number of customizable graph and chart tools, which can be used to generate visual representations of the data. The resulting graphics can be easily exported to commonly used computer applications for the production of your reports and presentations.
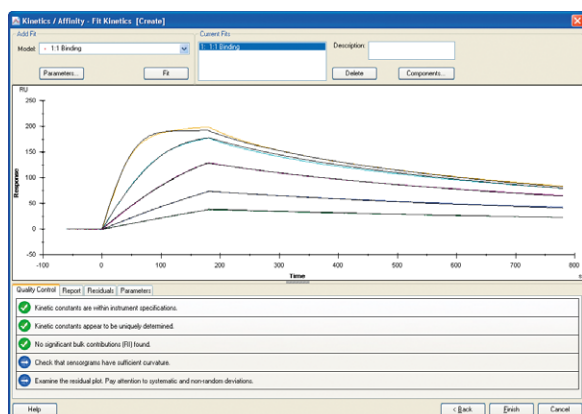

Fig 6. Easy QC tools help assess data quality and facilitate valid data interpretation.

Support for Biacore™ capture kits

Biacore™ capture kits provide optimized assay protocols that minimize the need for assay development (Fig 7). Guided workflows and wizards include preprogrammed settings for capture kits and sensor chip surfaces. This further increases the convenience and ease of use, while ensuring efficient use of reagents.

Biacore™ capture kits save cost, time, and effort by:

- Providing preoptimized capturing conditions for one of the biomolecules
- Providing preoptimized regeneration conditions
- Increasing the chances of success by using an immobilization strategy that maintains one of the biomolecules in a specific orientation on the sensor chip surface

Capture kits and surfaces are available for a variety of tags and molecules, including:

- GST Capture Kit for GST-fusion proteins
- His Capture Kit for His-tagged proteins
- Sensor Chip NTA for His-tagged proteins
- Biotin CAPture Kit for reversible capture of biotinylated molecules
- Sensor Chip SA for irreversible capture of biotinylated molecules
- Mouse Antibody Capture Kit for Mouse IgG antibodies
- Human Antibody Capture Kit for Human IgG antibodies
- Human Fab Capture Kit for human Fab fragments
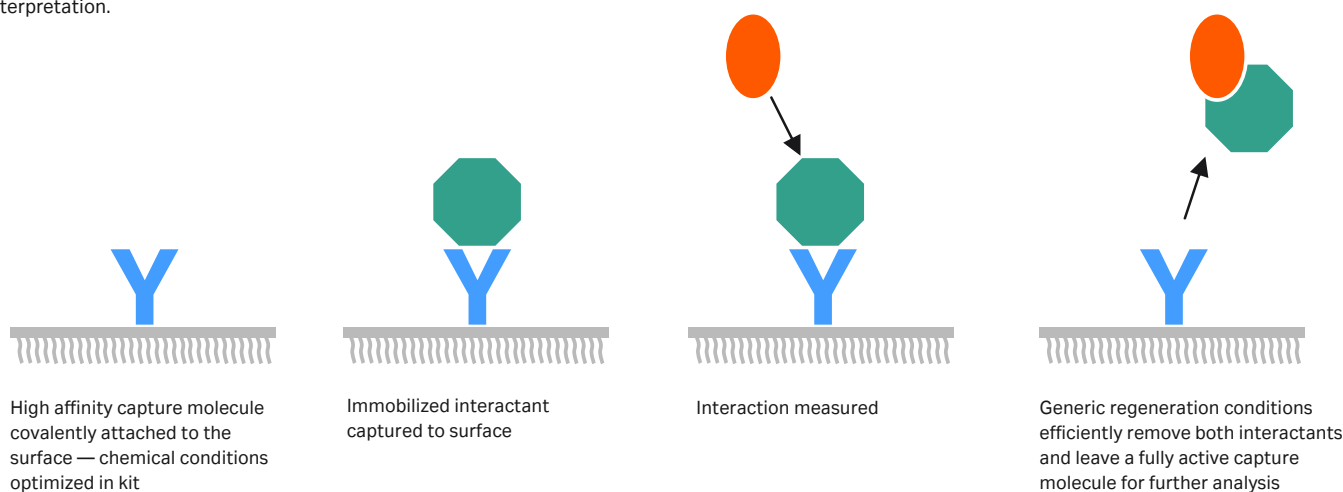- Sensor Chip L1 for liposomes

High affinity capture molecule covalently attached to the surface — chemical conditions optimized in kit

Immobilized interactant captured to surface

Interaction measured

Generic regeneration conditions efficiently remove both interactants and leave a fully active capture molecule for further analysis

Fig 7. In a capture assay, there is no need to find the conditions for attachment of the immobilized interactant to the sensor chip surface or the conditions for regenerating the surface to the original state.

Flexibility for a broad range of applications

The availability of a wide range of sensor chips allows you to choose the most suitable sensor surface for the nature of the molecule to be coupled and the requirements of the analysis. Covalent immobilization can be performed via reactive groups on the molecule, and special chips support irreversible capture of biotinylated molecules and capture of lipid monolayers and bilayers.

Equipped for multiple users

Biacore™ X100 stores all information about the immobilization step within the results files. This increases flexibility, and accessibility to the system by simplifying interrupted usage with removal of a single chip. It also reduces risk of error by recording information on sensor chip usage, which is an important issue if multiple users have access to the system.

Biacore™ X100 is accessed with regular Windows or Active Directory accounts. Each user gets a separate working area for storing methods and data.

Biacore™ X100 Plus Package

Biacore™ X100 Plus Package extends functionality of the system by adding features such as the ability to study biomolecular interactions involving small molecules in an organic solvent, to determine protein concentrations related to a specific binding event, and vary and control the temperature of different assays.

Analyze small molecules that require organic solvents for solubility

The study of how a putative drug interacts with its target protein provides crucial information for drug discovery. Putative drugs are often small organic compounds that require organic solvents like Dimethyl sulfoxide (DMSO) in order to become soluble in aqueous buffers. The analysis of an interaction in the presence of DMSO requires special care. Biacore™ X100 Plus Package provides all necessary tools for studying these types of interactions (Fig 8).
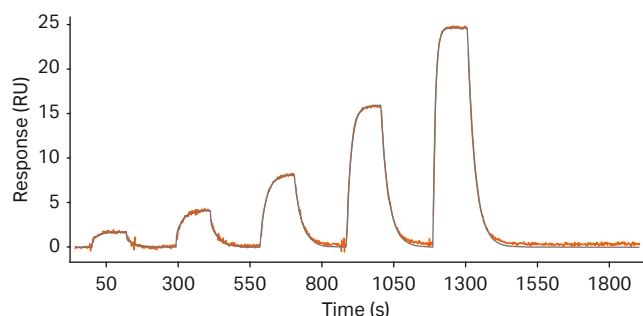
Fig 8. Biacore Single-Cycle Kinetics (SCK)™ analysis of binding of furosemide (molecular weight of 331 Da) dissolved in 3% DMSO to carbonic anhydrase.

Efficient and specific concentration analysis

Biacore™ X100 Plus Package allows you to measure the concentration of a protein via its interaction with a specific binding partner. This corresponds to the concentration of the active protein in solution (unlike total protein concentration determinations from methods such as spectrophotometry). This parameter is important to many applications because it enables you to:

- Improve accuracy of affinity and kinetic rate constants by reducing errors in concentrations that are used for fitting the binding data
- Monitor protein stability in various sample environments
- Monitor the amount of active protein throughout a process (e.g., in a series of purification steps from cell harvest to final product)

Concentration analysis without a standard

In addition to standard protein concentration analysis using a calibration curve, Biacore™ X100 Plus Package supports a method called calibration-free concentration analysis (CFCA). This method also measures protein concentrations related to specific binding activity, without the need for a protein standard. This is achieved by using the changes in binding rate that result from variations in flow rates under conditions in which the transport of molecules to the sensor surface is limited by diffusion. CFCA allows you to:

- Determine the concentration of a protein in the absence of suitable standard
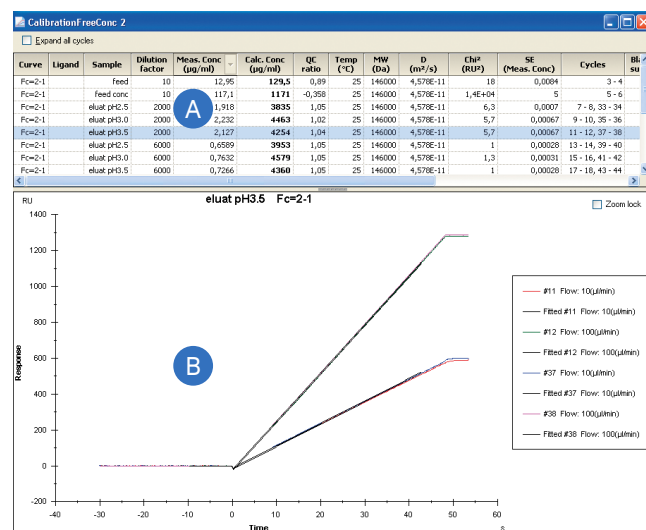- Validate the specified concentration of a protein standard

Fig 9. Calibration-free concentration analysis evaluation. Concentrations calculated for all samples included in the run (A). By clicking a sample, the underlying data and the fit of the model are displayed in (B).

Temperature control provides additional information

The ability to set the temperature of an assay enables you to perform experiments at physiological temperatures, or to study how particular biomolecular interaction properties change at different temperatures. Biacore™ X100 Plus Package can be used to:

- Study biomolecular interactions over a temperature range of 4°C to 40°C
- Better predict *in vivo* behavior by studying interactions at physiological temperatures

An in-line degasser is included in Biacore™ X100 Plus Package to preserve data quality at elevated temperatures.

Full assay and analysis flexibility

The Custom Assay Wizard provides an interface for defining assay methods with complete control over sample injection and sample flow through the system. Predefined templates are available for direct use or as a starting point to then be modified for your needs. Biacore™ X100 Plus Package also allows you to define your own evaluation models for fitting affinity and kinetic equations to collected data.

Summary

Biacore™ X100 is a robust and reliable system for comprehensive characterization of biomolecular interactions. Highly sensitive, real-time detection of molecular binding allows you to carry out studies such as kinetic characterization, highly accurate affinity determination, and detection of weak and transient binding events. Biacore™ X100 Plus Package adds additional functionality to the system, enabling you to perform experiments with molecules in organic solvents, determine specific binding concentration, and study biomolecular interactions at physiological temperatures.

Biacore™ X100 specifications

Technical specifications and characteristics

Detection technology	Surface plasmon resonance (SPR) biosensor
Data presentation	Result tables, result plots, and real-time monitoring of sensorgrams
Working principle	2–1 detection (automatic reference subtraction)
Information provided	Kinetic and affinity data (K_D, k_a, k_d) specificity, selectivity. Concentration measurements*
Sample type	LMW drug candidates to high molecular weight proteins in various sample environments (also DNA, RNA, polysaccharides, lipids, cells and viruses), e.g. in DMSO-containing buffers, plasma or serum
Kinetics:	Biacore Multi-Cycle Kinetics (MCK)™ or Biacore Single-Cycle Kinetics (SCK)™ (without the need for regeneration between injections)
Association rate constant (k_a)	10^3 to 10^7 $M^{-1}s^{-1}$ (for typical protein-protein interactions)
Dissociation rate constant (k_d)	10^{-5} to 0.1 s^{-1}
Dissociation constant (K_D)	100 µM to 1 pM
Detection limit	Typically 0.1 nM for > M_r 10 000 analytes Typically 1 nM for < M_r 10 000 analytes
Molecular weight detection	Down to M_r 100 in various sample environments
Analysis time per cycle	Typically 2 to 15 min
Sample capacity per run	Maximum 15 samples
Automation	24 h unattended operation
Injection volume	5 to 90 µL
Required sample volume	Injection volume + 20 to 30 µL (application dependent)
Immobilized interactant consumption	Typically 1 µg
Concentration analysis	With standard calibration curve or Calibration-free concentration analysis (CFCA)*
Short term noise	Typically < 0.1 RU (RMS)
Baseline drift	Typically < 0.3 RU/min
Blank subtracted drift	< ±0.03 RU/min
Sample refractive index range	1.33 to 1.40
In-line buffer degassing	Included*
Number of flow cells	2
Flow cell dimensions (W × H × L)	0.5 × 0.05 × 2.1 mm
In-line reference subtraction	Automatic
Flow rate range	1–100 µL/min
Analysis temperature range	25°C, or 4°C to 40°C* (maximum 10°C below ambient temperature)
Sample compartment temperature	Ambient
Dimensions (W × H × D)	Instrument: 596 × 563 × 593 mm
Net weight	Total 47 kg
Mains requirement	Processing unit: Autorange 100 to 240 VAC, frequency 50/60 Hz, protective earthing See the manufacturer's specification for mains requirements of the system controller
Power consumption	Processing unit: max. 400 VA See the manufacturer's specification about power consumption of the system controller

*Included in Biacore™ X100 Plus Package

Computer requirements

Processor	Minimum 2 GHz with at least two cores
Internal memory	Minimum 8 GB
Free hard disk space	Minimum 100 GB
Screen resolution	Minimum 1920×1080
Ports	Two serial communication ports for instrument connection (COM1 and COM2)

Data handling and storage

PC operating systems	Microsoft Windows 10 Professional or Enterprise, 64-bit, US English version Microsoft Windows 11 Professional or Enterprise, 64-bit, US English version
Database storage	Microsoft SQL Server Express 2022
Data export	Excel format result data export, text file raw data export
Image export	Clipboard export

Compliance

Compliant with	CE, cETLus, FCC, ICES-001, KC, RCM, EAC, UKCA

Ordering information

Product	Product code
Biacore™ X100	BR110073
Biacore™ X100 Plus Package	BR100798
Biacore™ X100 2.0 Upgrade Kit	BR100800
Biacore™ X100 Plus Package 2.0 Upgrade Kit	28956516

cytiva.com/biacore