GE Healthcare
Life Sciences

Biacore™ X100

Successful assays from day one


power
temperature
sensor chip
run

Boost your protein interaction research

Biacore X100 is a complete solution for biochemistry, molecular biology, or other research laboratories involved in the study of molecular interactions. The system contains all the key functionalities needed for day-to-day molecular interaction research with the purpose of understanding protein function and biological mechanisms.

Real-time monitoring of binding events gives a deep understanding of the dynamics of molecular interactions, including valuable kinetic data. Biacore analysis allows for straightforward and rapid data generation, leading to dependable conclusions and enabling the design of new innovative studies.

Biacore X100 can be used for a broad range of applications, including structure-function studies, pathway analysis, drug target identification and validation, and assay development.

Versatility and flexibility, in combination with unparalleled support, make it easy to generate top-quality data for your publication. With Biacore X100 on your bench, take your experiments to the next level and boost your protein interaction research.

Biacore systems, used extensively in academic research worldwide, are cited in almost 10 000 peer-reviewed scientific publications.


Biacore X100

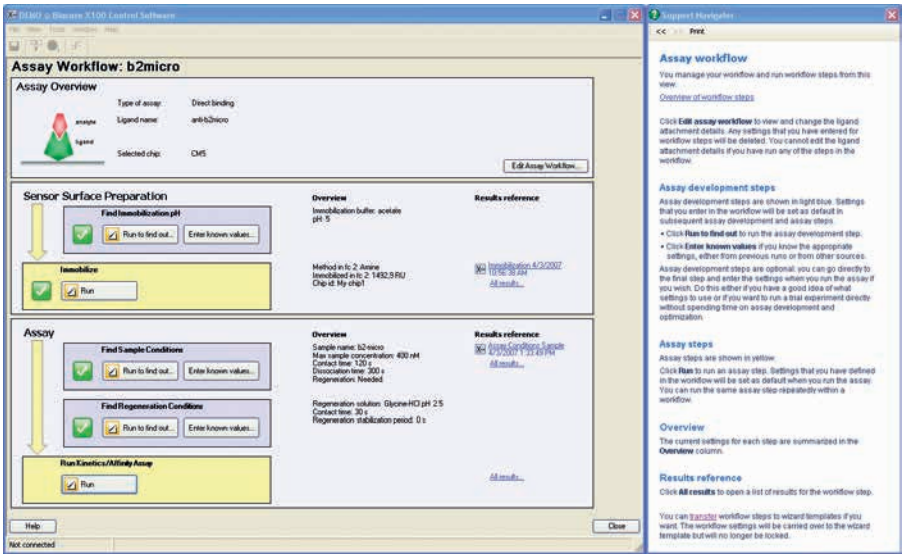
Run successful assays from day one

Biacore X100 uses workflow-oriented software that provides a supportive framework for assay development and data interpretation, building your expertise as you work.

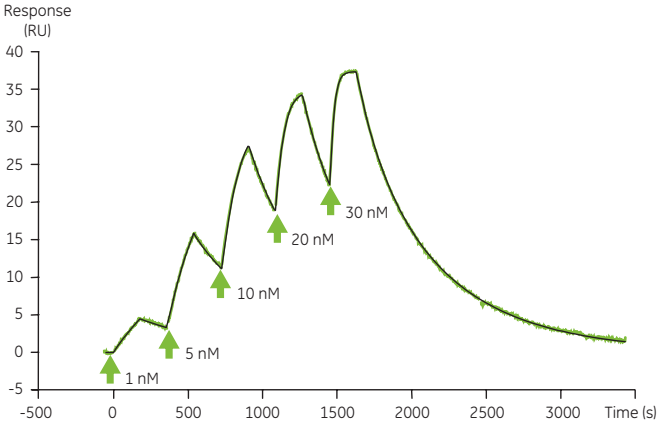
Researchers at Uppsala University used Biacore X100 to determine the binding characteristics of various Affibody™ molecules to HER2 with regards to specificity, affinity, and kinetic association- and dissociation-rates.

Specificity was quickly established by comparing binding levels of the different Affibody molecules to immobilized HER2 and a reference protein. As a next step, the kinetics of high-affinity binders was studied.

Affibody molecules turned out to be very difficult to regenerate. This problem was overcome with single-cycle kinetics which eliminates the need for finding regeneration conditions.



Guided workflows and software wizards enable generation of reliable data and rapid development of your expertise.



No regeneration required – Affibody molecules allowed to partially dissociate from immobilized HER2 between injections (arrows)

Sequential injection of increasing concentrations of sample in one continuous analysis cycle

Single-cycle kinetics sensorgram.

"The software wizards were easy to use, and the help function was extremely useful to have. I got good at this very quickly!"

Thuy Tran, PhD student, Uppsala University, Sweden

Acknowledgement: Thuy Tran and Professor Jörgen Carlsson, Uppsala University, Sweden

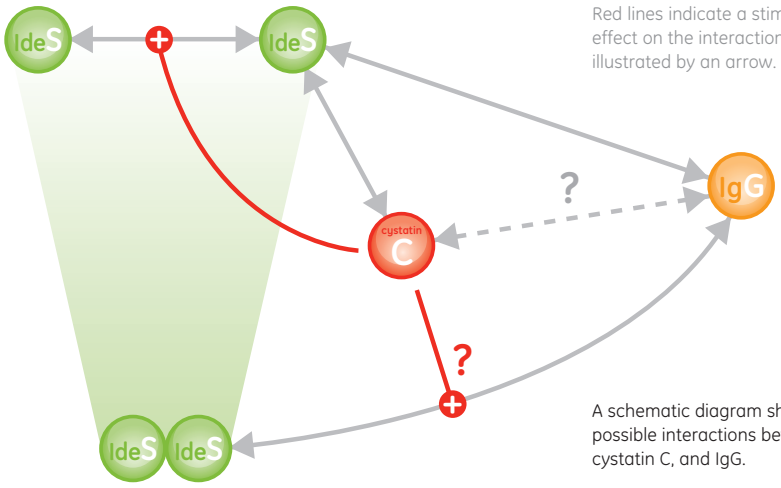
Elucidate complex molecular mechanisms

IdeS is a papain-like cysteine protease from the human pathogen *Streptococcus pyogenes* that specifically degrades IgG. The enzyme has been studied by Dr. Ulrich von Pawel-Rammingen's group at Umeå University in Sweden. They discovered that human cystatin C has an unexpected stimulatory effect on the IgG-endopeptidase activity of IdeS.

Evidence of an interaction between the two proteins was first found using a combination of gel filtration and Western blotting. The mechanisms of the interactions between IdeS, cystatin C, and IgG were further investigated using Biacore X100.

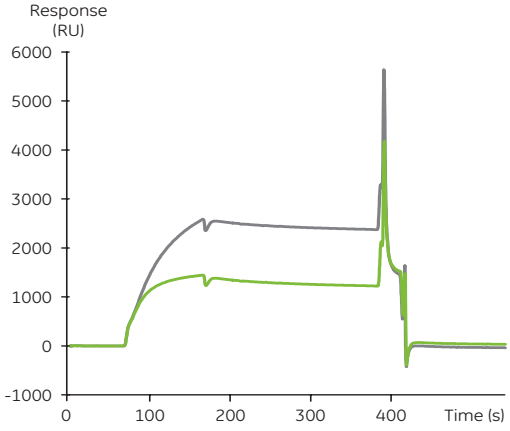
Their studies showed that cystatin C binds to IdeS, that IdeS can form homodimers, and that IdeS binding to IgG was not affected by cystatin C. The first research paper including data from Biacore X100 was published only five months after the system was installed.

The precise mechanisms of this complex network of interactions are now being further investigated. To aid in this study, a standardized enzyme activity assay for IdeS cleavage of IgG is employed, which is run on Biacore X100.



Red lines indicate a stimulatory effect on the interaction, illustrated by an arrow.

A schematic diagram showing the possible interactions between IdeS, cystatin C, and IgG.



Activity assay for IdeS cleavage of IgG. IgG_1 binding to immobilized Protein A is measured. Upper curve shows a sensogram of uncleaved IgG_1, while the lower curve shows a sensorgram for IgG_1 cleaved by addition of IdeS.

Reference
Vincents, B. *et al.* The human protease inhibitor cystatin C is an activating cofactor for the streptococcal cysteine protease IdeS. *Chemistry & Biology* **15**, 960-968 (2008).

"Having a Biacore X100 instrument in our own lab is of great value for our research and allows us to employ many new types of studies. Especially the possibility to run SPR experiments according to our own research agenda is a valuable tool in increasing the understanding of this sophisticated molecular interaction network."

Dr. Ulrich von Pawel-Rammingen, Umeå University, Sweden

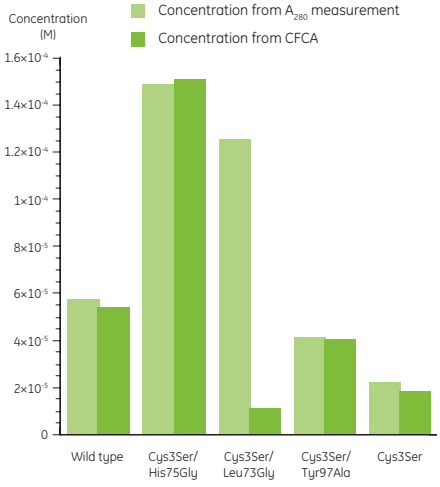
Acknowledgement: Dr. Ulrich von Pawel-Rammingen, Umeå University, Sweden

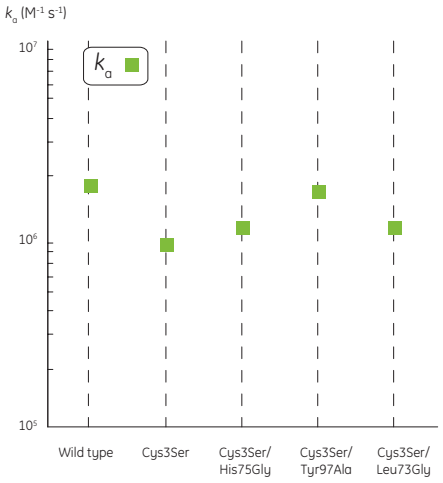
Increase the reliability of kinetic analysis

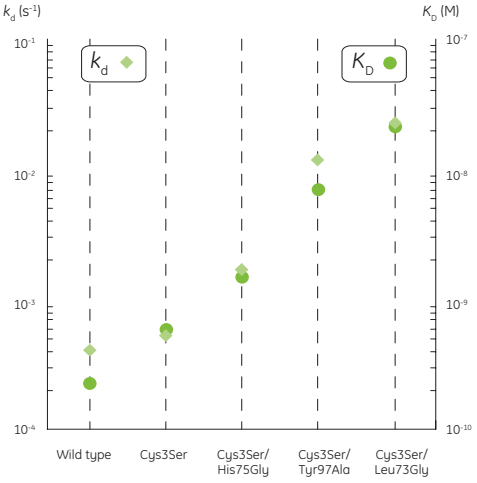
Cystatin B is an inhibitor of papain-like cysteine proteases. Four mutants were produced in order to study the importance of three different amino acids at the C-terminal and second binding loop for its binding to papain. Since no standard was available, Calibration-free concentration analysis (CFCA) was used to assess protein concentrations. CFCA is an innovative tool to measure protein concentrations, based on specific binding activity, without using a standard curve.

Mutants Cys3Ser/His75Gly, Cys3Ser/Tyr97Ala, and Cys3Ser showed a good agreement between concentration values obtained with A_{280} and CFCA, while mutant Cys3Ser/Leu73Gly had a very low concentration as measured by CFCA. Introduction of mutations can destabilize protein folding and prolonged storage or freeze/thaw procedures may also decrease the amount of protein capable of binding.

Kinetic analysis based on the A_{280} concentration measurement would lead to the conclusion that leucine 73 is important for the association rate, giving about ten times slower binding than the other mutants. In contrast, CFCA allowed for correct assessment of association rate (k_a) and affinity (K_D), allowing an appropriate interpretation of the interaction mechanism. The decreased affinities for all mutants were due to an increased dissociation rate (k_d), while the association rate (k_a) remained relatively constant.





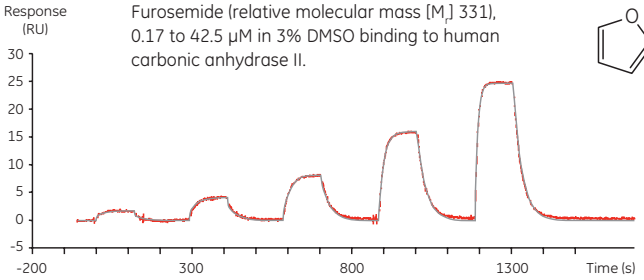


The combination of concentration, kinetics, and affinity analysis in a single study enabled the right interpretation of the interaction mechanism.

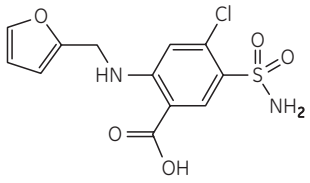
Develop and run assays involving small molecule interactions

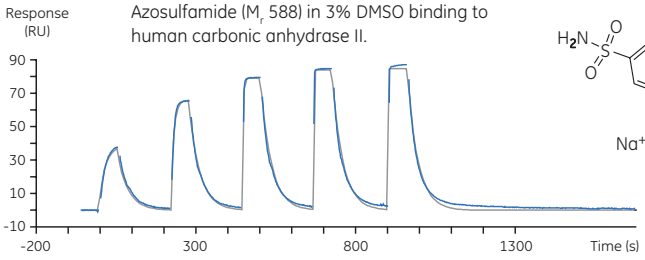
Biacore X100 provides the sensitivity required for challenging applications. Low molecular weight (LMW) compounds represent one challenging application, because a small molecular size reduces the signal levels. In addition, LMW compounds often require organic solvents for solubility reasons. The optional Biacore X100 Plus Package supports correction of bulk effects from high refractive index solvents such as DMSO.

Human carbonic anhydrases are known drug targets and inhibitors are used for treatment of various diagnoses. The figures show solvent-corrected sensorgrams of single-cycle kinetic profiles for furosemide, azosulfamide, and sulfanilamide binding to human carbonic anhydrase II in buffer containing 3% DMSO. Azosulfamide has the highest affinity, due to a slow k_d as compared to the other compounds.

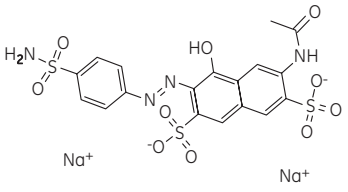


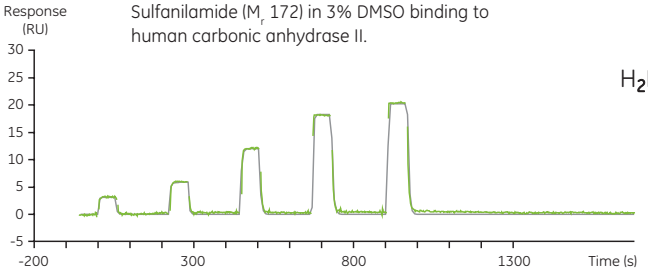
Furosemide (relative molecular mass [M_r] 331), 0.17 to 42.5 µM in 3% DMSO binding to human carbonic anhydrase II.



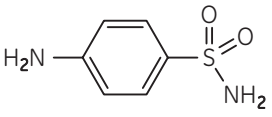


Azosulfamide (M_r 588) in 3% DMSO binding to human carbonic anhydrase II.





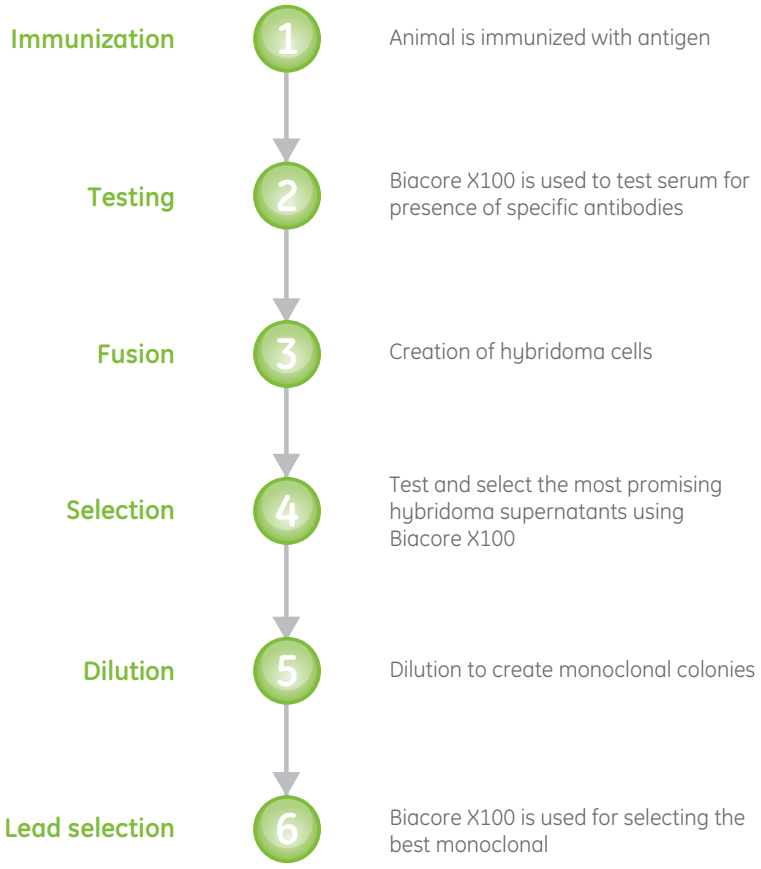
Sulfanilamide (M_r 172) in 3% DMSO binding to human carbonic anhydrase II.



Add Biacore X100 Plus Package if you are using DMSO for aqueous solubility.

Make critical decisions with confidence

Antibody selection workflow



Mercodia AB develops, manufactures, and markets *in vitro* diagnostic kits and assays for clinical and research applications.

Antibody selection is based on characteristics such as specificity and affinity. Biacore X100 has replaced ELISA in Mercodia's evaluation of monoclonal antibodies in early product development. This has resulted in reduced reagent costs and higher data quality, leading to significantly reduced project risk.

As Biacore X100 is a critical component of Mercodia's entire workflow, the reliability of the system and extensive service support available from GE Healthcare were important aspects in the decision to purchase a Biacore system.

"Using Biacore systems allows us to approach our product development and production questions from a new angle, providing a better basis for key decision making than we had previously."

Robert Gunnarsson, Product Development Manager, Mercodia AB

Acknowledgement: Robert Gunnarsson, Product Development Manager, Mercodia AB, Uppsala

Biacore X100 – a complete system solution

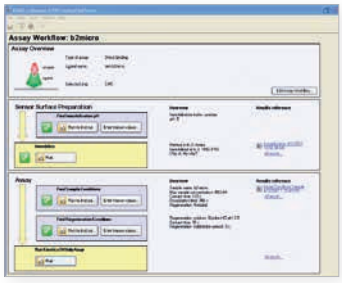
Biacore X100 integrates all the components needed to give quick and reliable molecular interaction data from day one. Automated instrumentation collects high quality data from small amounts of sample while minimizing hands-on time.

The software is workflow-oriented and provides a structured, yet flexible framework for assay development and data interpretation. A broad range of consumables are available for various assay alternatives. Biacore capture kits and surfaces are supported by preprogrammed workflows in the Biacore X100 software and flexibility is provided through a number of software wizards.

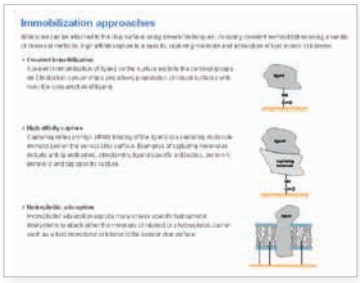
The system software helps you build expertise as you work, with an integrated support functionality that provides assay tips and guidelines. In addition, all Biacore X100 users have access to e-learning tools and an extensive methodology knowledge database on our Web site. Biacore X100 provides unparalleled support from assay development to data interpretation.

Robust and accessible instrumentation for acquisition of top-quality data

Supporting assay chemistry in capture kits and sensor surfaces allows a broad range of applications and saves time



Workflow-oriented software with built-in guidance ranging from assay development to data interpretation



Online tools speed up the learning process

Get the most out of label-free interaction analysis

A comprehensive range of service programs, support tools, and information services is available for Biacore X100.

Instrument service

During installation, dedicated service engineers provide you with quality service to ensure long-term, trouble-free performance of your system. Service contracts and extended warranty options are available for cost control and priority service.

Training

A range of courses and self-training tools are available to ensure that you get the most out of your protein interaction analyses.

Application support

Our experienced application specialists offer customized support to answer your specific questions.

Support tools on our Web site

- Technical tips and protocols
- BIA simulation software to perform dry-run experiments
- Interactive tutorials to learn how to setup, run, and evaluate Biacore analyses
- Extensive methodology knowledge database
- Material Safety Data Sheets relating to consumables
- Download section provides you with the latest software version, handbooks, news, and product-specific material about your system
- Biacore X100 basics e-learning course

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imagination at work

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