

# PROTEIN INTERACTION ANALYSIS ProteOn Manager™ Software

- Rapid processing and analysis of hundreds of analytes
- Detailed output of kinetic results on 36 interactions in seconds
- Novel referencing capabilities including real-time injection and double references
- Regulatory tools including 21 CFR Part 11 and IQ/OQ software tools

## A Comprehensive Tool for the Analysis of Biomolecular Interactions

### Introduction

ProteOn Manager software guides the user through complex protein interaction studies, allowing sophisticated experiments to be completed with ease. The comprehensive software offers the following benefits to ProteOn™ XPR36 protein interaction array system users:

- One software package to learn for instrument control, protocol design, data collection, processing, and analysis
- Easy to learn, workflow-oriented software
- Manual or automated processing and analysis providing robust and publication-quality results
- Intuitive software wizards to simplify kinetic, equilibrium, and concentration analysis functions
- Optional regulatory features:
  - Assistance regarding compliance with U.S. FDA 21 CFR Part 11 regulations
  - Installation qualification/operation qualification (IQ/OQ) wizard-driven software interface

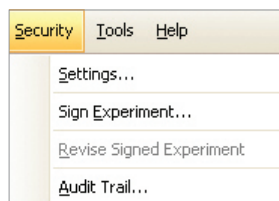


Fig. 1. ProteOn Manager software navigation with Security Edition features enabled.

### Instrument Control

ProteOn Manager software is organized to align with the typical SPR workflow. This begins with instrument control followed by protocol design, sample management, data processing, and analysis.

The software opens first to the instrument control panel, which includes all of the commonly used functions. Some of the features available in the instrument control panel are:

- Buffer selection
- Set sample and chip temperature
- Chip initialization options
- Maintenance protocols
- Log of instrument usage

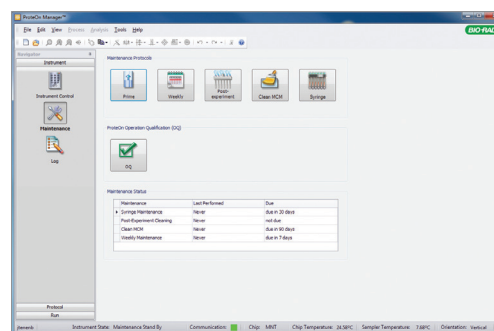


Fig. 2. Instrument controls within ProteOn Manager software.



## Protocol Design

ProteOn Manager software has flexible protocol design and allows users to choose from step or sample-driven entry. It uses orientation flow graphics to aid in the visualization of sample flow. Other features include:

- Automatic protocol generation tool
- Automated protocol looping function
- Easy-to-check protocol grid view
- Easy drag-and-drop navigation
- Ability to enter groups of steps for common functions
- Selection of data type for downstream analysis
- Ability to copy and paste steps
- Easy data management via an iterative searchable database

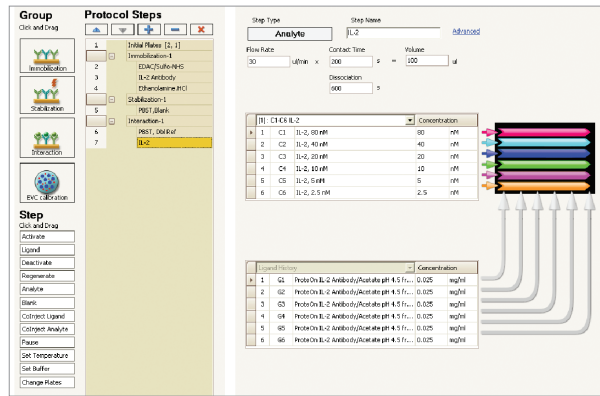


Fig. 3. Protocol design by steps.

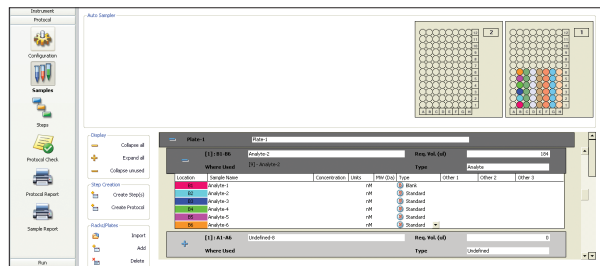


Fig. 4. Protocol design by samples.

#	Name	Group	Type	Flow Rate (µl/min)	Time (sec)	Volume (µl)	Conc. (µg/ml)	Sample Panel	Req. Vol. (µl)	Orientation	Quality	Pre Wash	Post Wash	Flow Stop. (sec)
1	Initial Plates [2, 1]		Change Rack											
2	Activate-1	Immobilization-1	Activate	30	300	150	0	[1]: H5-H6 Activator-1	218	Vertical	Minimum	Minimum	Minimum	0
3	Ligand-1	Immobilization-1	Ligand	30	300	150	0	[1]: G5-G6 Ligand-1	228	Vertical	Medium	Minimum	Minimum	0
4	Deactivate-1	Immobilization-1	Deactivate	30	300	150	0	[1]: F5-F6 Deactivator-1	218	Vertical	Medium	Minimum	Minimum	0
5	Blank-1	Stabilization-1	Blank	100	60	100	0	[2]: G5-G6 Blank-1	188	Horizontal	Maximum	Medium	Minimum	0
6	Regenerate-1	Stabilization-1	Regenerate	100	18	30	0	[1]: D5-D6 Regenerator-1	171	Horizontal	Minimum	Minimum	Minimum	0
7	Analyse-1	Interaction-1	Analyse	100	60	100	600	[1]: C5-C6 Analyte-1	184	Horizontal	Maximum	Medium	Minimum	20
8	Regenerate-1	Interaction-1	Regenerate	100	18	30	0	[1]: D5-D6 Regenerator-1	171	Horizontal	Maximum	Medium	Minimum	0
9	Analyse-2	Interaction-1	Analyse	100	60	100	600	[1]: B5-B6 Analyte-2	184	Horizontal	Maximum	Medium	Minimum	20

Fig. 5. New protocol check view.

## Sample Management

Sample management can be tedious and complex — especially if a user wants to conserve sample or reagents by performing multiple injections from a single location. ProteOn Manager software offers user-friendly features for sample management:

- User-defined sample input
- Import of sample lists
- Color-coordinated graphical display of sample locations
- Drop-down menus allowing users to change sample locations
- Creation of steps for replicate injections
- Import, export, and printing of protocols

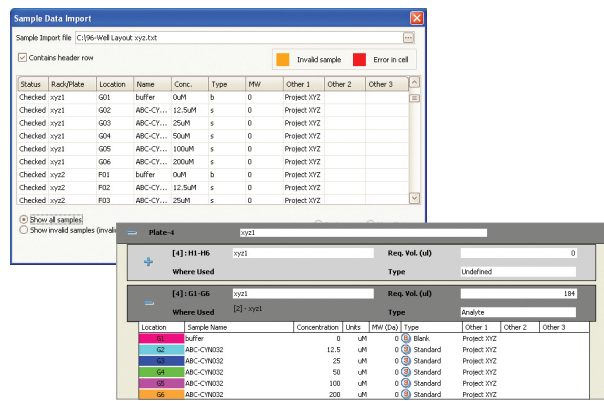


Fig. 6. Sample import dialog screen and resulting sample panel.

ProteOn Manager Sample Layout Report						
Protocol: Imported Experiment: ProteOn One Shot Kinetics Kit - 11/12/2008 2:44 PM						
Date: Friday, November 14, 2008						
Printed By: cernan						
Sample Panel: PBST:Blank						
Rack: 1	Column: H Position: 1-6					
Where Used:	PBST:Blank, PBST:Blank					
Row	Sample Name	Conc.	Units	MW	Sample Type	Vol. Req.
	ProteOn PBST, pH 7.4				Undefined	343µl
	ProteOn PBST, pH 7.4				Undefined	343µl
	ProteOn PBST, pH 7.4				Undefined	343µl
	ProteOn PBST, pH 7.4				Undefined	343µl
	ProteOn PBST, pH 7.4				Undefined	343µl
	ProteOn PBST, pH 7.4				Undefined	343µl
Sample Panel: IL-2						
Rack: 1	Column: G Position: 1-6					
Where Used:	IL-2					
Row	Sample Name	Conc.	Units	MW	Sample Type	Vol. Req.
	IL-2, 80 nM	80.00	nM		Standard	184µl
	IL-2, 40 nM	40.00	nM		Standard	184µl
	IL-2, 20 nM	20.00	nM		Standard	184µl
	IL-2, 10 nM	10.00	nM		Standard	184µl
	IL-2, 5 nM	5.00	nM		Standard	184µl
	IL-2, 2.5 nM	2.50	nM		Undefined	184µl
Sample Panel: Undefined-7						
Rack: 1	Column: F Position: 1-6					
Where Used:	IL-2					
Row	Sample Name	Conc.	Units	MW	Sample Type	Vol. Req.
	Undefined-1				Undefined	Out
	Undefined-2				Undefined	Out
	Undefined-3				Undefined	Out
	Undefined-4				Undefined	Out
	Undefined-5				Undefined	Out
	Undefined-6				Undefined	Out
	Undefined-7				Undefined	Out
	Undefined-8				Undefined	Out
	Undefined-9				Undefined	Out
	Undefined-10				Undefined	Out
	Undefined-11				Undefined	Out
	Undefined-12				Undefined	Out
	Undefined-13				Undefined	Out
	Undefined-14				Undefined	Out
	Undefined-15				Undefined	Out
	Undefined-16				Undefined	Out

Fig. 7. Sample protocol report.

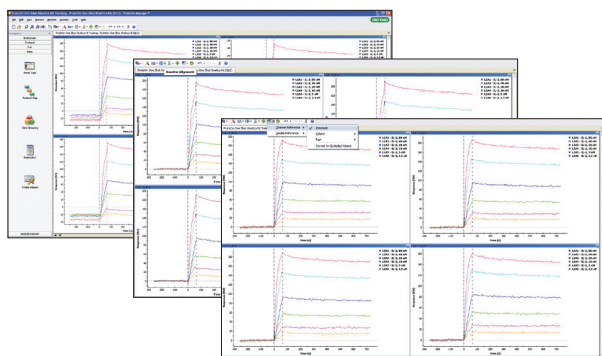
## Data Processing

Data processing in ProteOn Manager software is fast and flexible, offering single button, 3-parameter auto-processing or manual operations for in-depth replicate studies. Depending on the need, ProteOn Manager software users can process data quickly or precisely as needed. Some of the many benefits of data processing in ProteOn Manager software are:

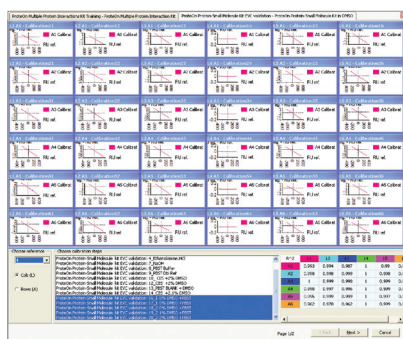
- Flexible manual or automatic processing
- Undo/redo function for rapid management of data
- Comprehensive referencing options: interspot, channel reference, real-time double reference, injection double reference, and excluded volume correction
- Double referencing independent of location with blank double reference option
- Application of processing parameters locally or globally
- Export of sensorgrams in metafile or data formats
- New data table displays report points, user-defined columns, and injection parameters
- Export of all processed data to tab-delimited files



**Fig. 8. Quick navigation to processing controls.** From left to right: 3-point auto processing; individual processing keys for manual or automated X and Y alignment, spike removal, manual sensorgram movement, and referencing; undo and redo commands; report point generation commands.



**Fig. 9. Auto-processing and referencing of the ProteOn One-Shot Kinetics™ kit.**



**Fig. 10. EVC wizard for the exclusion of bulk effects.**

## ProteOn Manager Software Offers Different Referencing Options to Suit Your Analysis Needs

Interspot and channel referencing correct for bulk effects and nonspecific binding events.

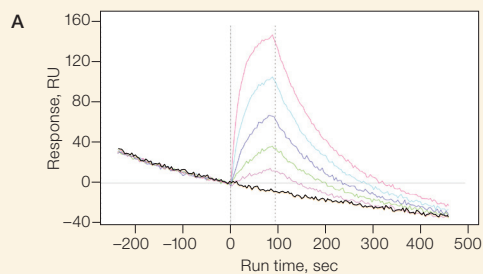
- **Interspot referencing** — unique referencing that utilizes the area in between the interaction spots for local reference subtraction. This allows the use of all six channels for data collection.
- **Channel reference** — traditional reference that uses a blank dedicated channel for global reference subtraction.

Real-time and double references can be used in conjunction with interspots or a channel reference. Injection and double references correct for baseline drift such as decay. (Figures 11A, B.)

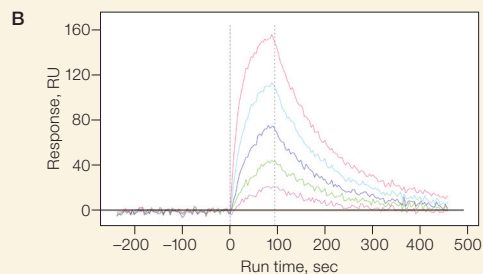
- **Real-time double reference** — unique reference that allows buffer to be run in parallel and simultaneous to an analyte injection panel.
- **Injection double reference** — traditional blank injection following an injection panel.

Small molecule applications commonly use high refractive solvents, which may need correction.

- **Excluded volume correction (EVC)** — uses a standard curve to correct for high-refractive solvents like DMSO.



**Fig. 11A. Real-time injection reference — before correction:** Five concentrations of analyte were injected (colored traces) and running buffer was injected in the sixth channel (black trace).



**Fig. 11B. Real-time injection reference — after correction:** The running buffer (black trace in top panel) was subtracted from all analyte binding curves. The drift is eliminated.

## Analysis

Analysis of protein interactions is made simple through the use of intuitive software wizard interfaces. Wizards allow step-by-step data analysis for kinetic, equilibrium, and concentration determination. Wizard options include:

- Analysis wizards to calculate results of interactions in seconds
- Default parameters for quick results or manual options for flexible analysis
- Seven kinetic fit models: Langmuir, Langmuir with mass transfer, bivalent analyte, heterogeneous analyte, heterogeneous ligand, two states, and Langmuir with drift
- Off-rate only analysis
- Sortable and filterable data results table
- User-defined columns
- Grouped, global, local, and fixed analysis parameters
- Molecular weight normalization
- Export of reports to tab-delimited files

Step Name	Sample Location	Sample Name	Concentration	Rate	RSD
L3A1	12	12	1.0E+05	1.0E+04	1.0E+04
L3A2	12	12	1.0E+05	1.0E+04	1.0E+04
L3A3	12	12	1.0E+05	1.0E+04	1.0E+04
L3A4	12	12	1.0E+05	1.0E+04	1.0E+04
L3A5	12	12	1.0E+05	1.0E+04	1.0E+04
L3A6	12	12	1.0E+05	1.0E+04	1.0E+04
L3A7	12	12	1.0E+05	1.0E+04	1.0E+04
L3A8	12	12	1.0E+05	1.0E+04	1.0E+04
L3A9	12	12	1.0E+05	1.0E+04	1.0E+04
L3A10	12	12	1.0E+05	1.0E+04	1.0E+04

Fig. 13. Single panel view of kinetic results from ProteOn One-Shot Kinetics kit. Data can be displayed grouped or ungrouped.

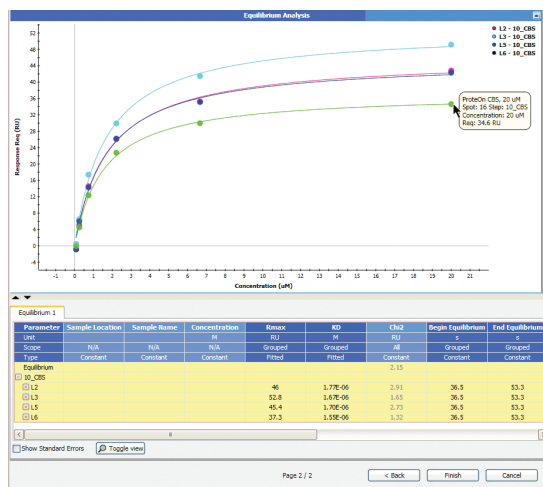


Fig. 14. Results image and table from concentration analysis.

## Regulatory Tools

Tools are available with the ProteOn Manager software to facilitate regulatory compliance in the drug discovery and development workflows. ProteOn Manager software, Security Edition and the ProteOn XPR36 IQ/OQ kit assist the user in adhering to the good practices rulings observed by the pharmaceutical industry. These rulings require procedural (notification, training, and operational), administrative, and technical (software-related) control changes.

### ProteOn Manager Software, Security Edition

New compliance features enable the user to turn on security features that comply with U.S. FDA 21 CFR Part 11 regulations. Features of the Security Edition software include audit trails, electronic signatures, data validation, user log-ins and permissions, and closed-system security. Signatures can be verified, ensuring data consistency within the instrument.

### ProteOn XPR36 Installation Qualification/Operational Qualification Kit

The ProteOn XPR36 IQ/OQ kit is designed to test critical system functions and to ensure the reliability and consistency of system performance. Key features include intuitive wizard-driven software, printable electronic reports for document control, electronic log of IQ/OQ and test results, ready-to-use reagents, and a sensor chip for testing system performance with unattended operation.

## Ordering Information

Catalog #	Description
176-0200	<b>ProteOn Manager Software</b> , for ProteOn XPR36 instrument control, experiment design, data collection and analysis
176-0210	<b>ProteOn Manager Software, Security Edition</b>
176-4200	<b>ProteOn XPR36 IQ/OQ Kit</b>
176-4220	<b>ProteOn Operation Qualification Kit</b>
176-4225	<b>Regulatory Tools Package</b> , includes ProteOn Manager Security Edition Software and ProteOn XPR36 IQ/OQ Kit



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