SERVA ICPL™ Kits

SERVA
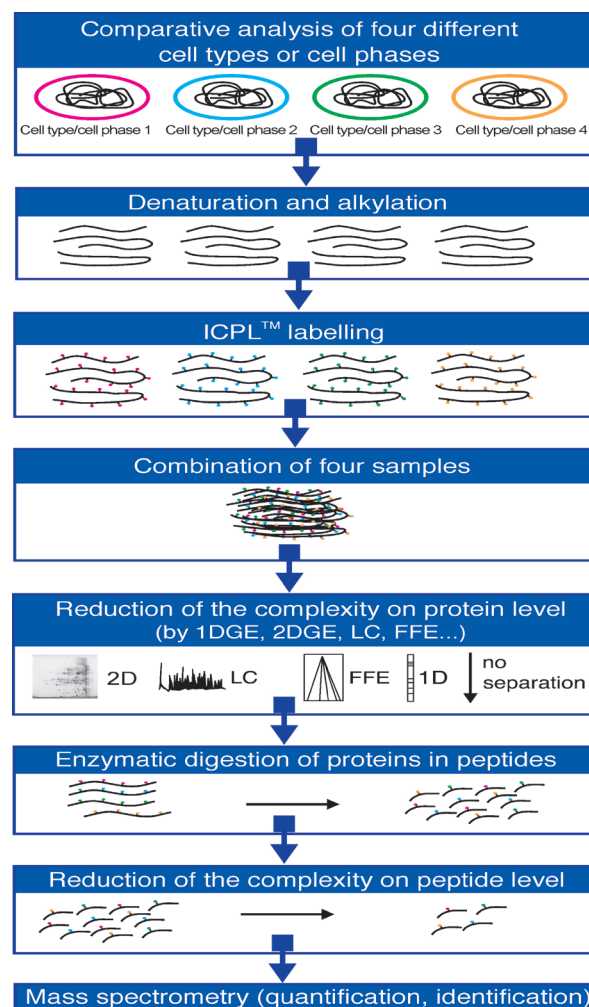*serving scientists!*

TECHNICAL NOTES

Improved proteome analysis

ICPL™ Technology

The ICPL™ technology combines the power of Isotope Coded Protein Labelling (ICPL) with an unmatched dynamic range for protein identification and quantification due to its potential combination with intact protein fractionation steps (see workflow). Applying the ICPL quadruplex method the simultaneous quantitative analysis of up to four independent proteome samples can be performed by stable protein labelling.

This powerful technology for comparative quantification of proteins is also available as ICPL Quadruplex PLUS Kit (cat. no. 39233.01) containing trypsin and endoproteinase Glu-C for double digestion of labelled proteins. The treatment of protein samples with trypsin and Glu-C will even improve the high sequence coverage of the ICPL™ Quadruplex Kit. The ICPL™-Standard PLUS included with the kit allows calibration of the analysis system. The standard contains Glu-C / Trypsin NB treated 4-plex labelled BSA peptides.

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ICPL 0	1-($^{12}C_6$$^1H_4$)-Nicotinoyloxy-succinimide (^{12}C-Nic-Reagent): M_r = 105.0215 Da	
ICPL 4	1-($^{12}C_6$$^2D_4$)-Nicotinoyloxy-succinimide (^{2}D-Nic-Reagent): M_r = 109.0715 Da	
ICPL 6	1-($^{13}C_6$$^1H_4$)-Nicotinoyloxy-succinimide (^{13}C-Nic-Reagent): M_r = 111.0419 Da	
ICPL 10	1-($^{13}C_6$$^2D_4$)-Nicotinoyloxy-succinimide (^{13}C^{2}D-Nic-Reagent): M_r = 115.0919 Da	

- Improved sequence coverage by double digestion
- Comparative analysis of up to four proteomes
- ICPL*Quant* and ICPL-ESI*Quant* for data analysis
- Analysis of PTMs and isoforms

ICPL™ Quadruplex Workflow

Comparative analysis of four different cell types or cell phases — Cell type/cell phase 1, Cell type/cell phase 2, Cell type/cell phase 3, Cell type/cell phase 4 → Denaturation and alkylation → ICPL™ labelling → Combination of four samples → Reduction of the complexity on protein level (by 1DGE, 2DGE, LC, FFE...): 2D, LC, FFE, 1D, no separation → Enzymatic digestion of proteins in peptides → Reduction of the complexity on peptide level → Mass spectrometry (quantification, identification)

Comparative analysis of proteomes

In contrast to other labelling techniques the ICPL Quadruplex Kit permits proteome analysis on 2D gel basis as well as on multidimensional LC-MS basis using ESI-MS or MALDI-MS. All approaches that work on a truly proteomics scale use pre-fractionation such as LC, PAGE, 2D-GE or magnetic beads. Protein fractionation is compatible with the ICPL-technology, as quantitative ratios remain constant throughout any fractionation step. This reduces complexity in each protein fraction which increases the dynamic range of protein detection and quantification and thus the number of identified proteins. A high sequence coverage of labelled peptides provides a sound statistical base for automatic protein quantification avoiding excessive manual validation efforts.

SERVA ICPL™ Kits

Improved proteome analysis

Quantification and analysis software ICPL*Quant* and ICPL-ESI*Quant*

ICPL*Quant* has been developed to accurately quantify ICPL-labelled peptides on the MS level during LC-MALDI and peptide mass fingerprint experiments. ICPL-ESI*Quant* allows accurate quantification of ICPL-labelled peptides on the MS level in ESI-MS runs. The software is available free of charge and includes text and video tutorials for user support (http://www.biochem.mpg.de/en/rg/lott-speich/technologies/ICPLQuant/index.html).

Main functionalities

- Pattern picking on MS data (singlets, ICPL doublets, triplets or quadruplets)
- Compound quantification to compensate for the isotope effect
- Visualization of multiplet chromatograms
- Target-orientated precurser selection for downsizing the number of MS/MS
- MASCOT result parser (data file parser) for merging protein and quantitative information
- Peptide and protein result tables in Excel and text format
- Batch process over multiple LC experiments
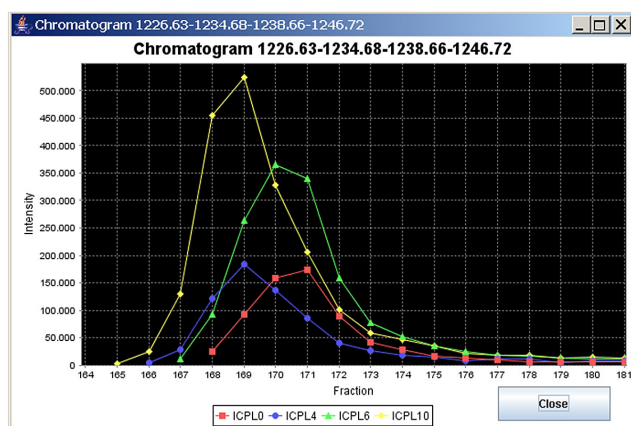
The ICPL-ESI*Quant* "Show chromatogram" option for a selected multiplet.

All SERVA ICPL Kits can also be obtained from Bruker Daltonic. For more information please contact Bruker Daltonics directly (www.bdal.de/care).

ICPL*Quant*

ICPL*Quant* is a powerful and easy-to-use "one button" software solution for ICPL-labelled proteomics experiments followed by 1D-LC and mass spectrometry. The software is able to handle deisotoped peaklist data from different vendors. Version 1.5 supports the ABI 4700/4800 Proteomics Analyser (MALDI) and the Bruker Ultraflex II mass spectrometer with WARP-LC 1.1 software included.
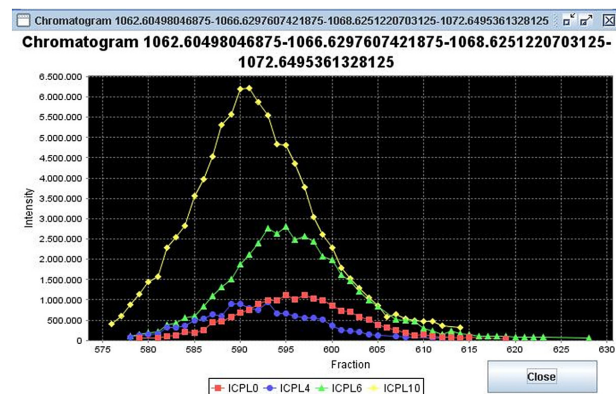
The ICPL*Quant* "Show chromatogram" option for selected quadruplet 1226-1234-1238-1246

ICPL ESI*Quant*

ICPL-ESI*Quant* is a powerful and easy-to-use "one button" software solution for ICPL-labelled proteomics experiments followed by mass spectrometry using ESI. At the moment, only Thermo ORBITRAPs (including Xcalibur Software) are supported by the program, but the support of additional vendors is planned in the future. The software is able to handle deisotoped peaklist data in mzXML format. Search results from MASCOT can be imported in the .DAT file format.

Ordering Information

Product	Quantity	Cat. no.
ICPL™ Kit	1 kit (6x2 rxn.)	39230.01
ICPL™ Triplex Kit	1 kit (6x3 rxn.)	39231.01
ICPL™ Quadruplex Kit	1 kit (6x4 rxn.)	39232.01
ICPL™ Quadruplex PLUS Kit	1 kit (6x4 rxn.)	39233.01