

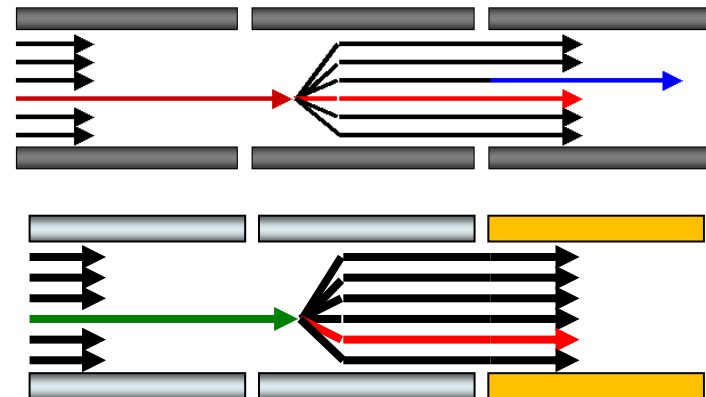


Skyline

Targeted Mass Spec Environment

What Next?

Brendan MacLean



What is New

- ▶ **What you have seen**
 - ▶ Panorama – Tom
 - ▶ Interactive tools – Sam
 - ▶ Ion mobility – Erin
 - ▶ Small molecules – Laura
 - ▶ New approaches – Sonia, Bruno
- ▶ **What else**
 - ▶ Differential statistics
 - ▶ Chorus – chromatograms from the cloud
 - ▶ Optimization libraries
 - ▶ Teaching
- ▶ **What is NEXT**



Aggregating and Publishing

- ▶ Publish fully annotated Skyline documents
- ▶ Build chromatogram libraries
- ▶ Aggregate lab QC data
- ▶ Free hosted version (<http://panoramaweb.org>)
 - ▶ >100 separate projects so far (CPTAC, LINCS & ABRF sPRG)
 - ▶ >1000 data sets uploaded
 - ▶ User controlled security
- ▶ Locally installable server application
 - ▶ Roche, Genentech, [unnamed], and Merck (2 more...)
- ▶ Free and open source (Apache 2.0)



More Panorama



Panorama

Panorama Public: A public repository for Skyline documents

TP 132 - Vagisha Sharma



Panorama

Mass spec quality control with Skyline and Panorama

TP 134 - Josh Eckels

- ▶ Panorama Partners Program – Meet to discuss
-



External Tools

Msstats (Vitek)



3946 Downloads!

grouped study statistics

QuaSAR (Carr)



1271 Downloads!

response curve statistics

SProCoP (MacCoss)



919 Downloads

system suitability

MSI Probe (Gibson)



962 Downloads!

MSI filtering statistics

Protter (Wollscheid)



363 Downloads!

transmembrane topology

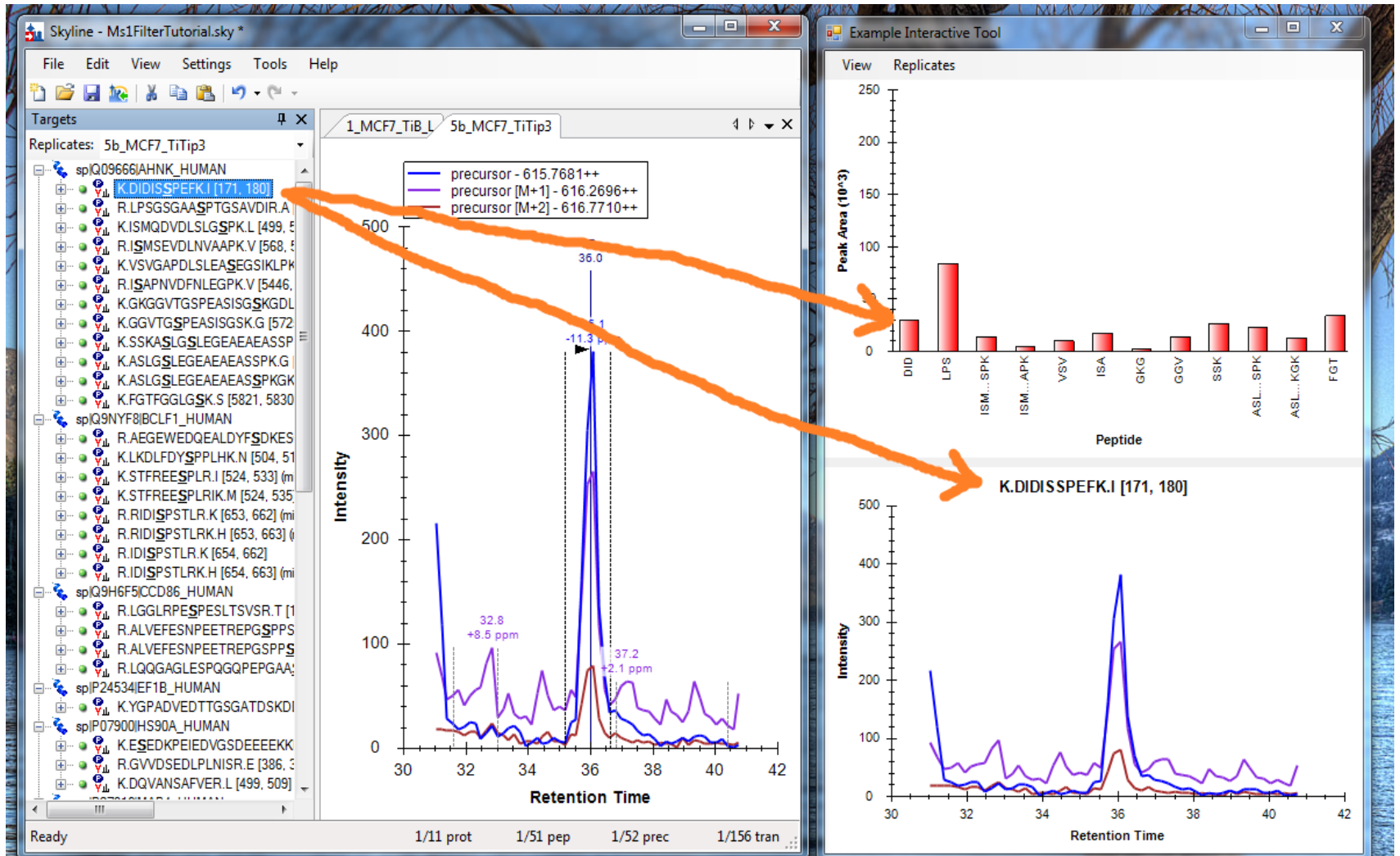
Population Variation



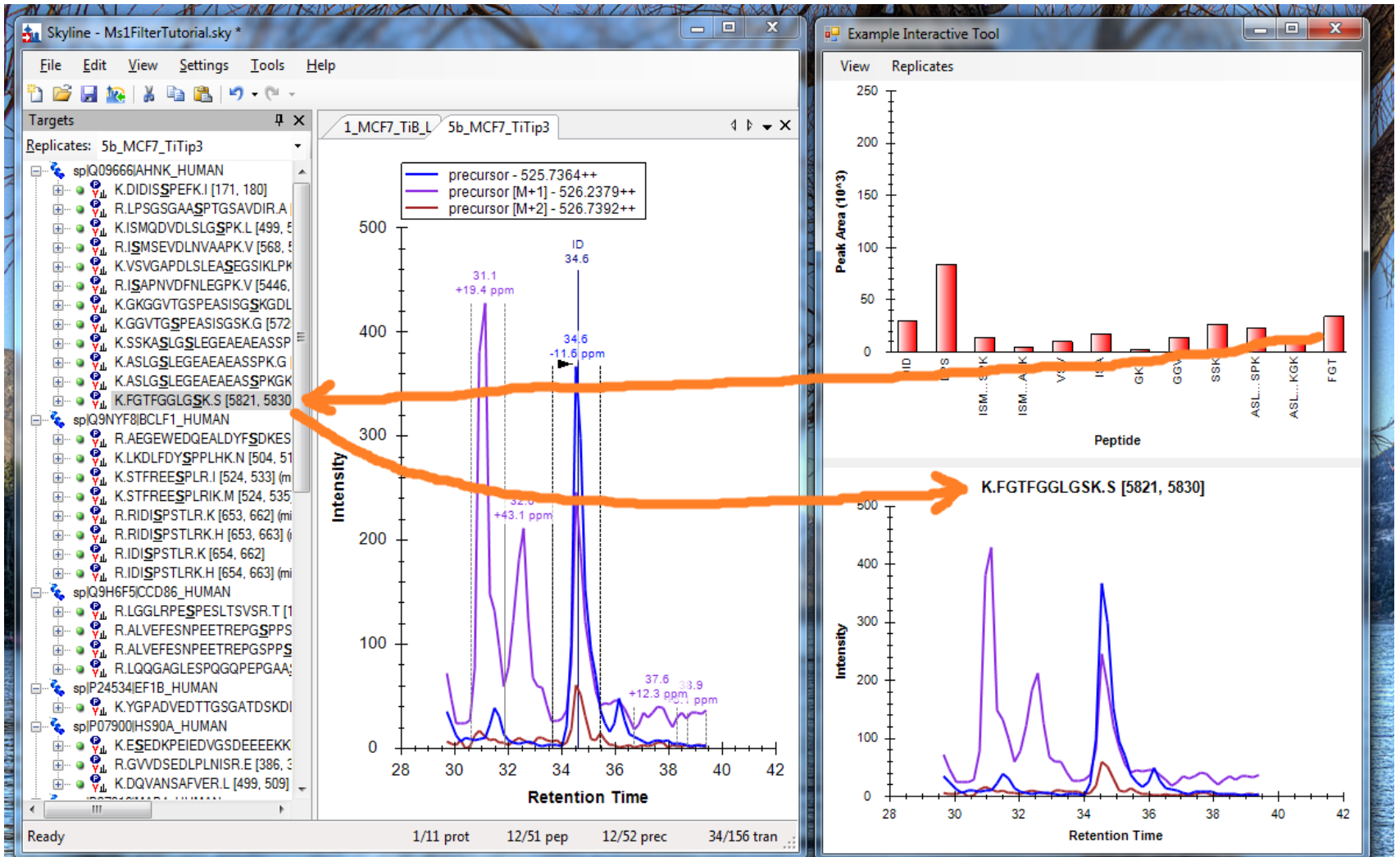
208 Downloads!

mutation frequency

Interactive Tools Interface

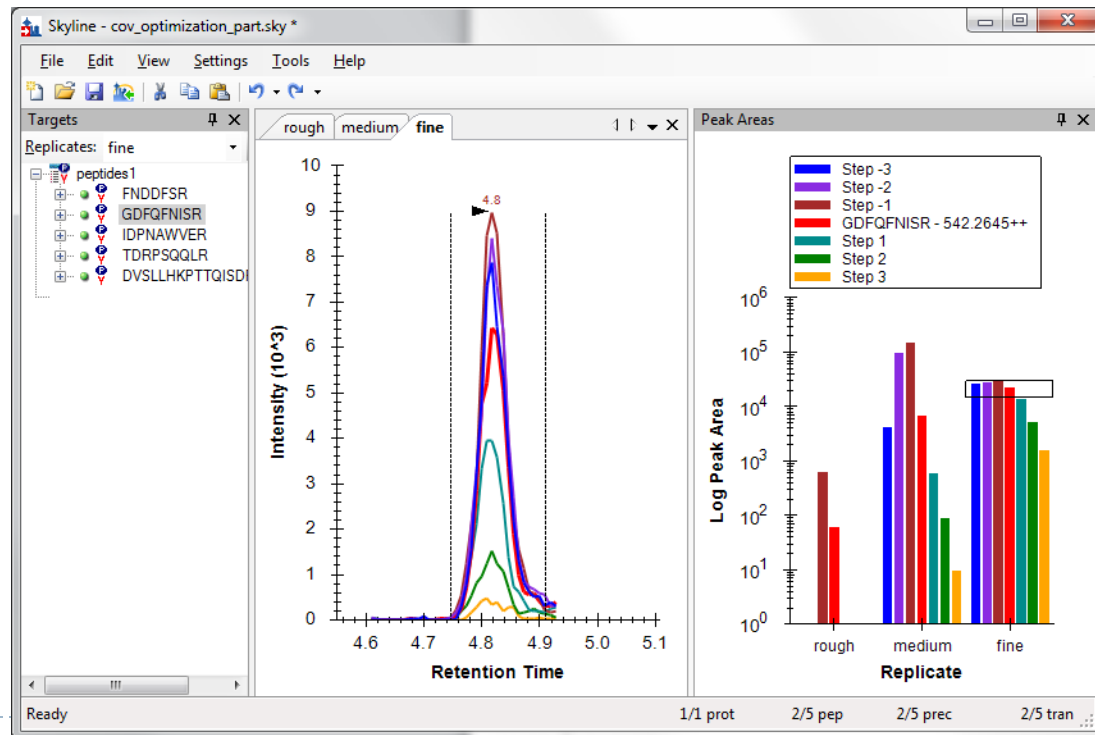


Interactive Tools Interface



Ion Mobility

- ▶ Agilent and Waters
- ▶ Waters PLGS search spectral libraries with drift times
- ▶ Sciex SelexION CoV optimization



Small Molecules

Skyline-daily - 3745_ESI+_Verif_052715_minimized.sky

File Edit View Settings Tools Help

Targets

Replicates: pool_03_UCA168_3745_060714

- Ceramide
 - Cer(d18:0/17:0)
 - Cer(d18:1/16:0)
 - Cer(d18:1/16:0) M+H-H2O
 - Cer(d18:1/22:0)
 - Cer(d18:1/22:0) M+H-H2O
 - Cer(d18:1/24:1(15Z))
 - Cer(d18:1/24:1(15Z)) M+H-H2O
- Diacylglycerol
 - DG(12:0/18:0/0:0)[iso2]
 - DG(14:1(9Z)/18:2(9Z,12Z)/0:0)[iso2]
 - DG(17:1(9Z)/22:6(4Z,7Z,10Z,13Z,16Z,19Z)/0)
 - DG(18:1(9Z)/22:6(4Z,7Z,10Z,13Z,16Z,19Z)/0)
 - DG(19:1(9Z)/22:6(4Z,7Z,10Z,13Z,16Z,19Z)/0)
 - DG(20:1(11Z)/22:6(4Z,7Z,10Z,13Z,16Z,19Z)/0)
- Monoacylglycerol
 - MG(0:0/22:6(4Z,7Z,10Z,13Z,16Z,19Z)/0:0)
- Phosphatidylcholine
 - PC(12:0/14:0)
- Phosphatidylethanolamine
 - PE(P-16:0/18:4(6Z,9Z,12Z,15Z))
- Sphingomyelin
 - SM(d18:2/24:1) M+H
 - SM(d18:2/24:1) M+NH4
- Acylcamitine
 - Stearoylcamitine
- Triacylglycerol
 - TG(12:0/12:0/13:0)[iso3]
 - TG(14:0/16:0/16:1(9Z))[iso6]

Peak Areas

Retention Times

Full Scan

Scan type: MSI

ID12990_01_UCA168_3745_060714.raw (2.10 min)

Intensity

Retention Time

Intensity

m/z

Ready

4/8 prot 15/21 pep 15/21 prec 43/63 tran

Small Molecules



MP 414 - Brian Pratt
Small Molecule Targets and Ion Mobility



WP 251 - Will Thompson
Validation of Quantitative Measurements from Lipidomics



Next for Small Molecules

- ▶ **Libraries**
 - ▶ Spectral libraries
 - ▶ Optimization libraries
 - ▶ iRT libraries?
- ▶ Integrate with discovery metabolomics tools
- ▶ Improved peak integration
- ▶ UI make-over
- ▶ More documentation



New Approaches

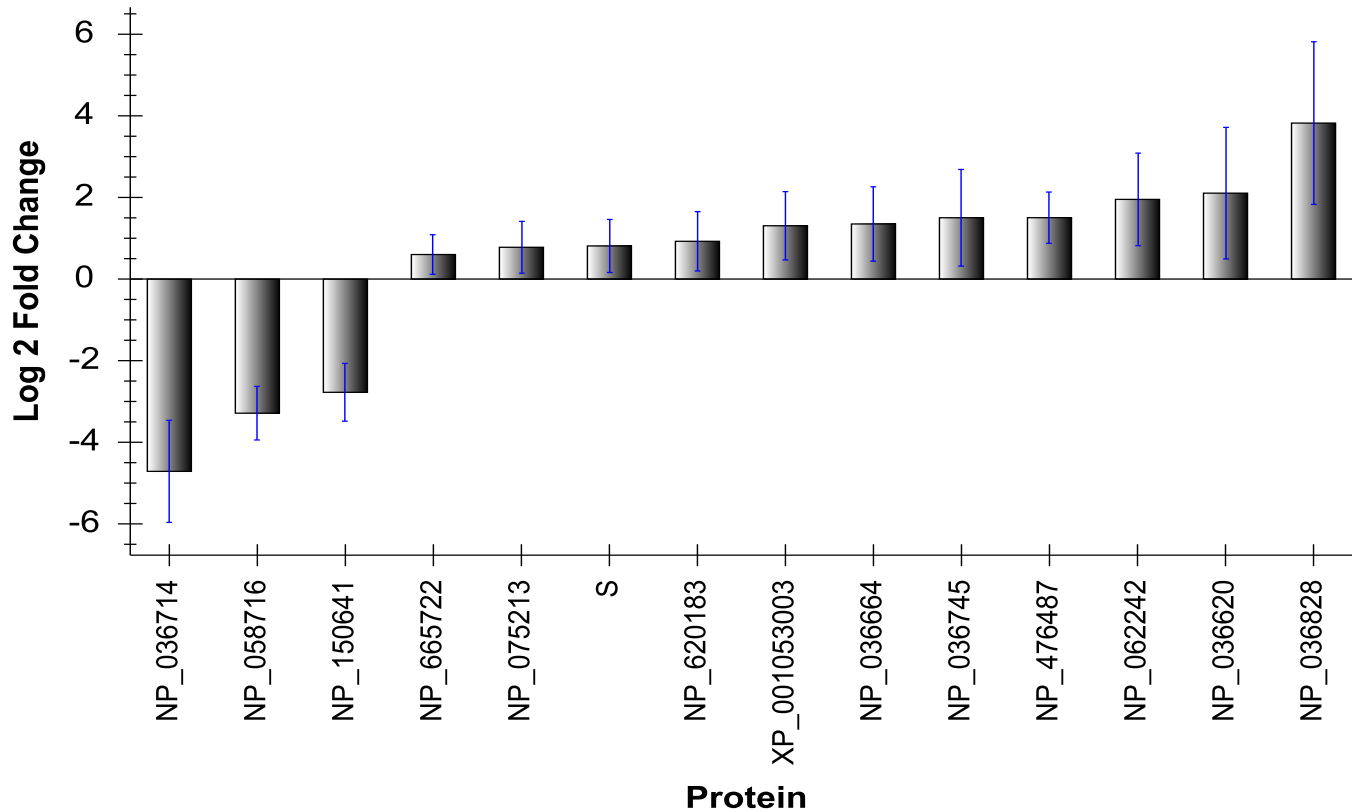
- ▶ **Library-Free DIA Peptide Search**
 - ▶ Pecan – Sonia Ting (MacCoss lab)
 - ▶ Umpire – Chih-Chiang Tsou (Nesvizhskii lab)
 - ▶ MSPLIT – Jian Wang (Bandeira lab)
- ▶ **Compressed Sensing DIA – Egertson (MOB)**
- ▶ **Triggered-PRM**
- ▶ **Assay portability from PRM to DIA**
- ▶ **Peptide Response Prediction**

Informatics – SRM & DIA

Wednesday 8:30 – 10:30 (WOB)



Differential Statistics



Webinar #8 - Brendan MacLean

DDA to Targeted: Differential Statistics

Tuesday, June 16

Chorus For Mass Spec File Storage

- ▶ Google Docs-like interface
- ▶ Lab-centered security model
- ▶ Cloud storage for raw data
 - ▶ Upload as acquired
 - ▶ Translated into distributed data structure
 - ▶ Massively parallel cloud data access
- ▶ Fast chromatogram extraction
- ▶ Fast single spectrum access
- ▶ Scalable



Usage Statistics

 **1,077** Users

 **20.52** Data volume (TB)

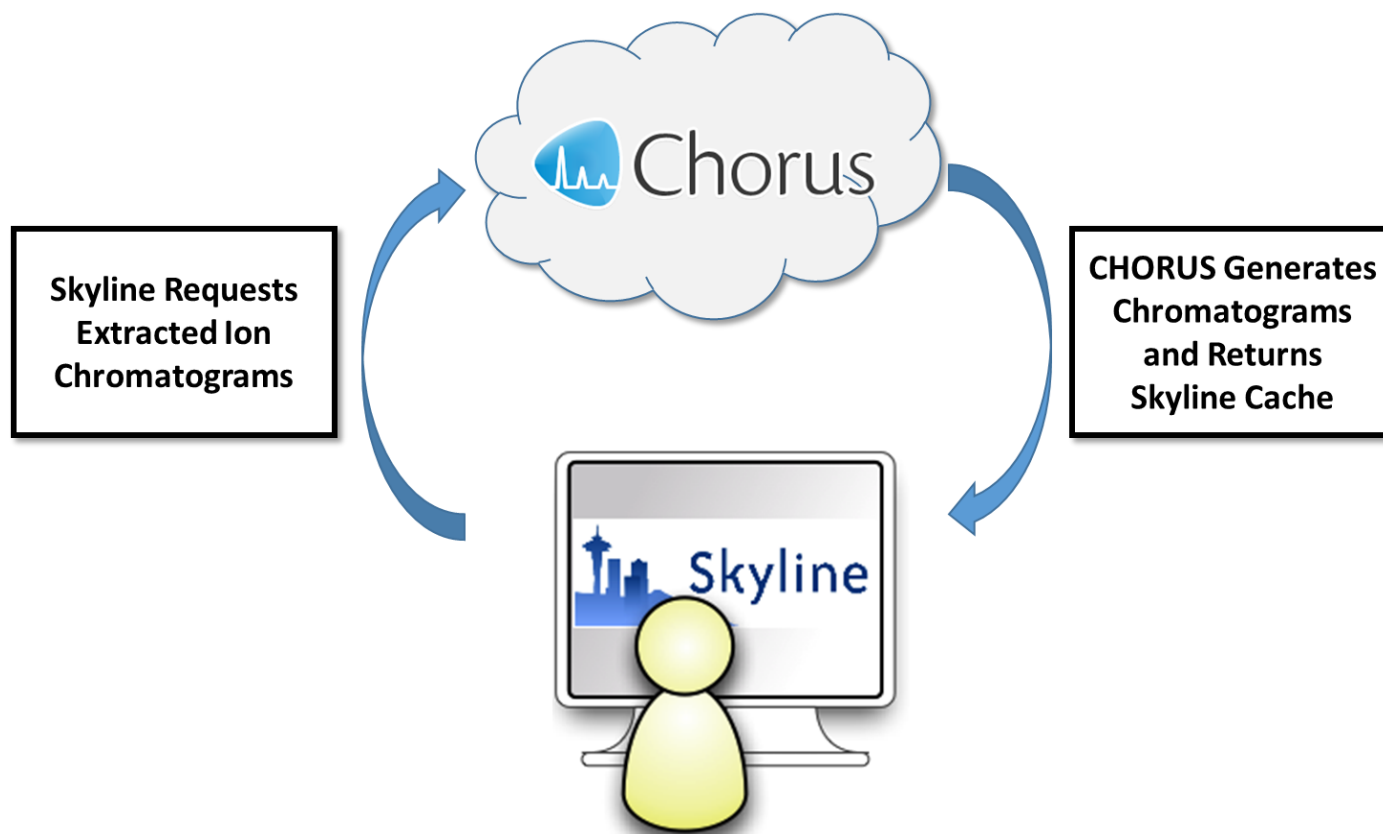
 **64,696** Data files

 **87** Public projects

 **254** Public Experiments

**>1 TB Downloaded
per Month**

Remote Extraction of Chromatograms



Wednesday Workshop
5:45 – 7:00 PM Room 260/267

Optimization Libraries

Transition Settings

Prediction Filter Library Instrument Full-Scan

Precursor mass: Monoisotopic

Product ion mass: Monoisotopic

Collision energy: ABI 5500 QTrap

Decustering potential: ABI

Optimization library: CoV library 2015

Compensation voltage: ABI

Optimize by: Precursor

OK Cancel

Edit Optimization Library

Name: CoV library 2015

OK Cancel

Optimization library: C:\proj\pwiz_clean\pwiz_tools\Skyline\TestResults\Brendan_BREND

Open... Create...

Optimization type: Compensation Voltage

Optimizations:

	Modified Sequence	Compensation Voltage
▶	GDFQFNISR++	12.34
	DVSL LHKPTTQISDFHVATR++++	23.45
	FNDDFSR++	17.00
	IDPNAWVER++	12.50
	TDRPSQQLR++	14.00

5 optimized compensation voltages

Add...

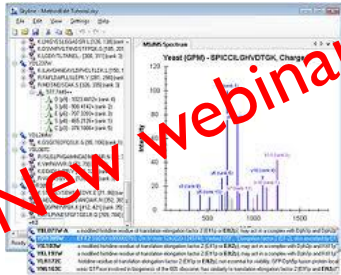
Teaching Targeted Proteomics

- ▶ **3 weeklong courses 2014**
 - ▶ Zurich, Seattle, Barcelona
- ▶ **7 workshops and short courses in 2014**
 - ▶ Albuquerque, Seattle, Baltimore, Brixen, Souel, Kyoto, Mumbai
- ▶ **5 weeklong courses 2015**
 - ▶ Seattle, Boston, Zurich, Barcelona, Mumbai
- ▶ **5 workshops and short courses in 2015**
 - ▶ Tempe, St. Louis, Rio de Janiero, Puerta Vallarta, Tokyo

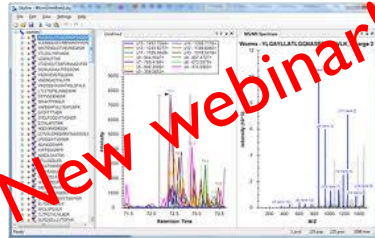


14 Tutorials (5 translated)

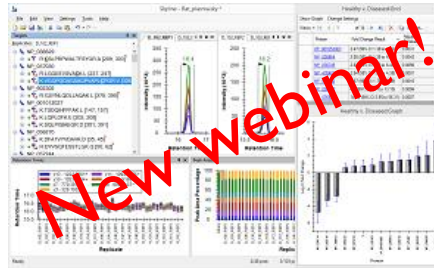
Method Editing



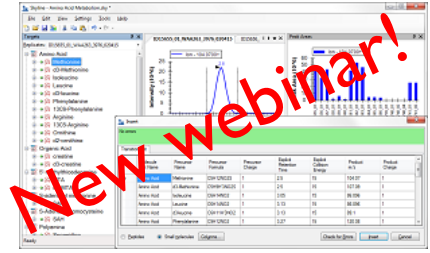
Method Refinement



Grouped Studies



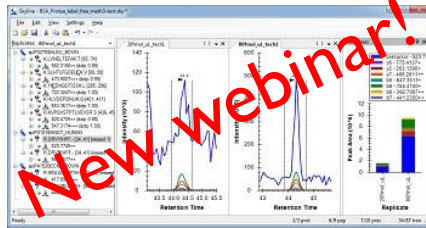
Small Molecule



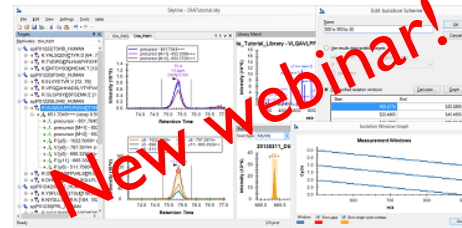
MSI Filtering



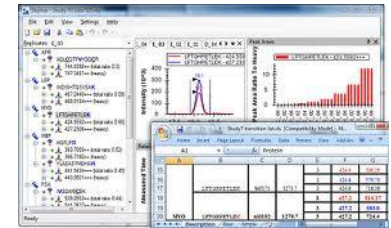
Targeted MS/MS (PRM)



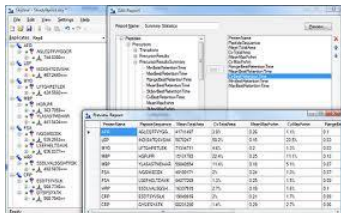
DIA with DDA



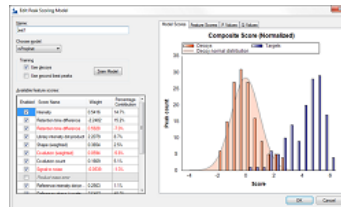
Existing Experiments



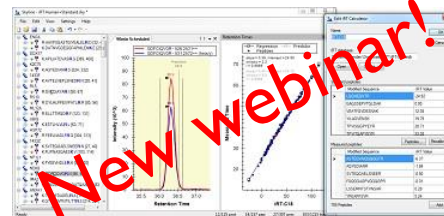
Custom Reports



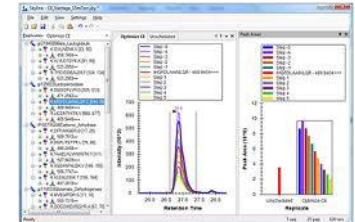
Advanced Peak Picking



iRT



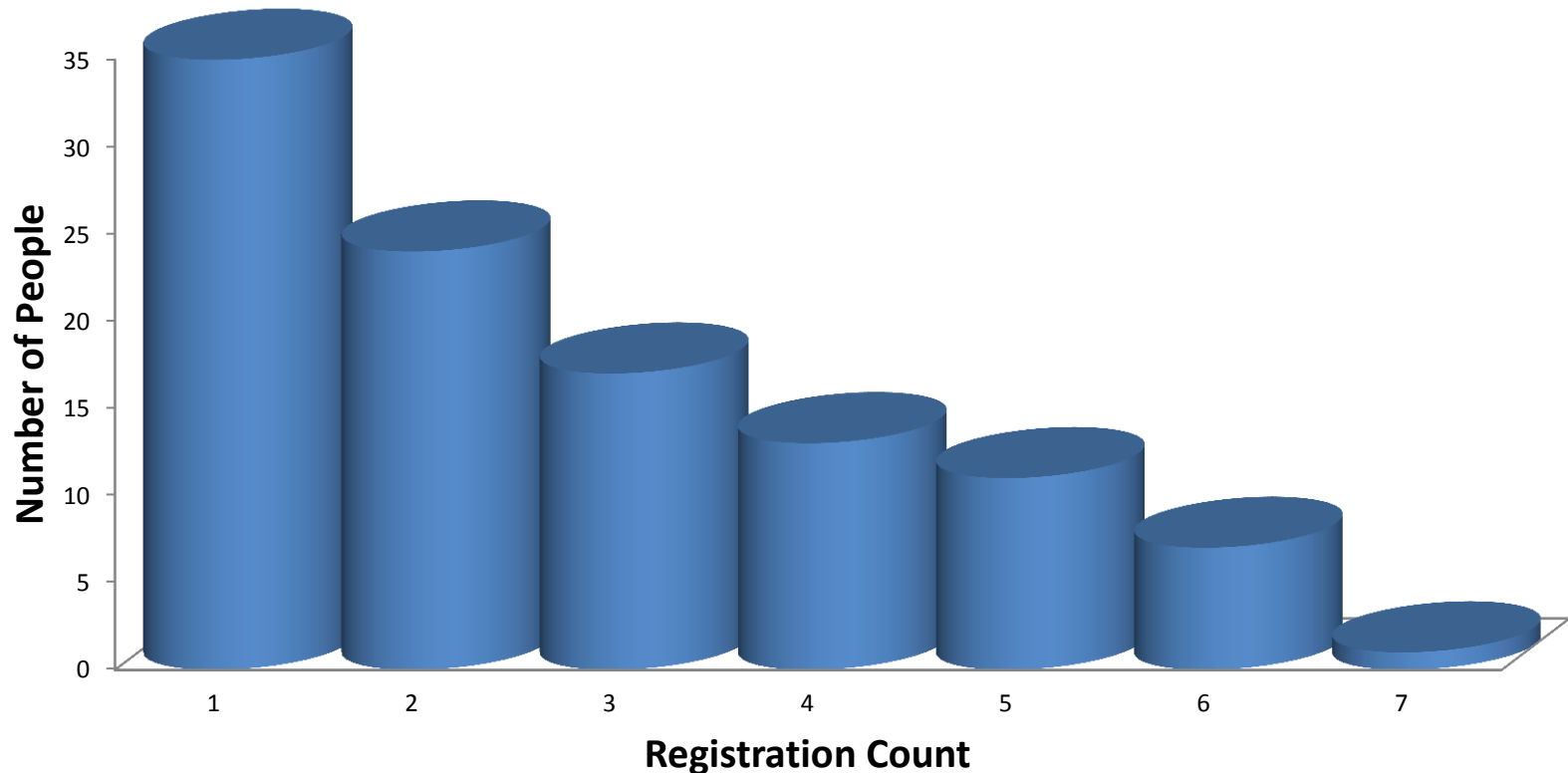
CE Optimization



Growing...

Watching the Webinars?

- ▶ 108 (43%) cross registration between UGM and webinars



Nadya Galeva



What is NEXT

- ▶ **More of ALL THAT**
- ▶ **More integrated statistics**
 - ▶ Calibrated quantification
 - ▶ Protein therapeutic characterization
- ▶ **More visualization**
 - ▶ Integrated hierarchical clustering heat maps
- ▶ **Improve support for large projects**
 - ▶ Import performance (>100,000 transitions)
 - ▶ 100s of replicates
- ▶ **Support use in validated environments**



Skyline Team

▶ Brendan MacLean



▶ Nick Shulman



▶ Don Marsh



▶ Brian Pratt



▶ Vagisha Sharma



▶ Nat Brace



▶ Kaipo Tamura



▶ Yuval Boss



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- ▶ Michael MacCoss
- ▶ Jim Bollinger
- ▶ Jarrett Egertson
- ▶ Andy Hoofnagle

▶ Broad Institute

- ▶ Sue Abbatiello
- ▶ Steve Carr
- ▶ Jake Jaffe
- ▶ D. R. Mani

▶ Buck Institute

- ▶ Birgit Schilling
- ▶ Matthew Rardin
- ▶ Brad Gibson

▶ Duke

- ▶ Will Thompson
- ▶ Arthur Moseley

▶ FHCRC

- ▶ Amanda Paulovich
- ▶ Jeffrey Whiteaker

▶ InfoClinika

- ▶ Andrey Bondarenko
- ▶ Oleksii Tymchenko

▶ IMSB

- ▶ Rudolph Aebersold
- ▶ Christina Ludwig
- ▶ Olga Schubert
- ▶ Hannes Röst
- ▶ George Rosenburger
- ▶ Lucia Espona Pernas

▶ Pitt

- ▶ Christine Wu
- ▶ Nathan Yates

▶ PNNL

- ▶ Erin Baker
- ▶ Yehia Ibrahim
- ▶ Sangtae Kim
- ▶ Tao Liu
- ▶ Sam Payne
- ▶ Chaochao Wu

▶ Purdue

- ▶ Meena Choi
- ▶ Olga Vitek

▶ Stanford

- ▶ Dario Amodei
- ▶ Parag Mallick

▶ Vanderbilt

- ▶ Matthew Chambers
 - ▶ Daniel Liebler
 - ▶ David Tabb
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Instrument Vendor Collaborators

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- ▶ Joe Roark
- ▶ Ed Darland
- ▶ Yinghang Yang



Agilent Technologies

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- ▶ Christie Hunter



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- ▶ Junko Iida
- ▶ Neil Loftus
- ▶ Kiriko Matsuo



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- ▶ Markus Kellmann
- ▶ Andreas Kuehn
- ▶ Vlad Zabrouskov



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- ▶ Kieran Neeson
- ▶ Keith Richards

