

A Workflow for Quality Assessment, Quantitation and Statistical Inference of Targeted Proteomics Data using Skyline and Panorama

Genentech
A Member of the Roche Group

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Skyline User Meeting
June 4, 2017

Data analysis is a challenge for development of clinical biomarkers using targeted mass spectrometry

Drug development



Targeted MS for development of clinical biomarkers



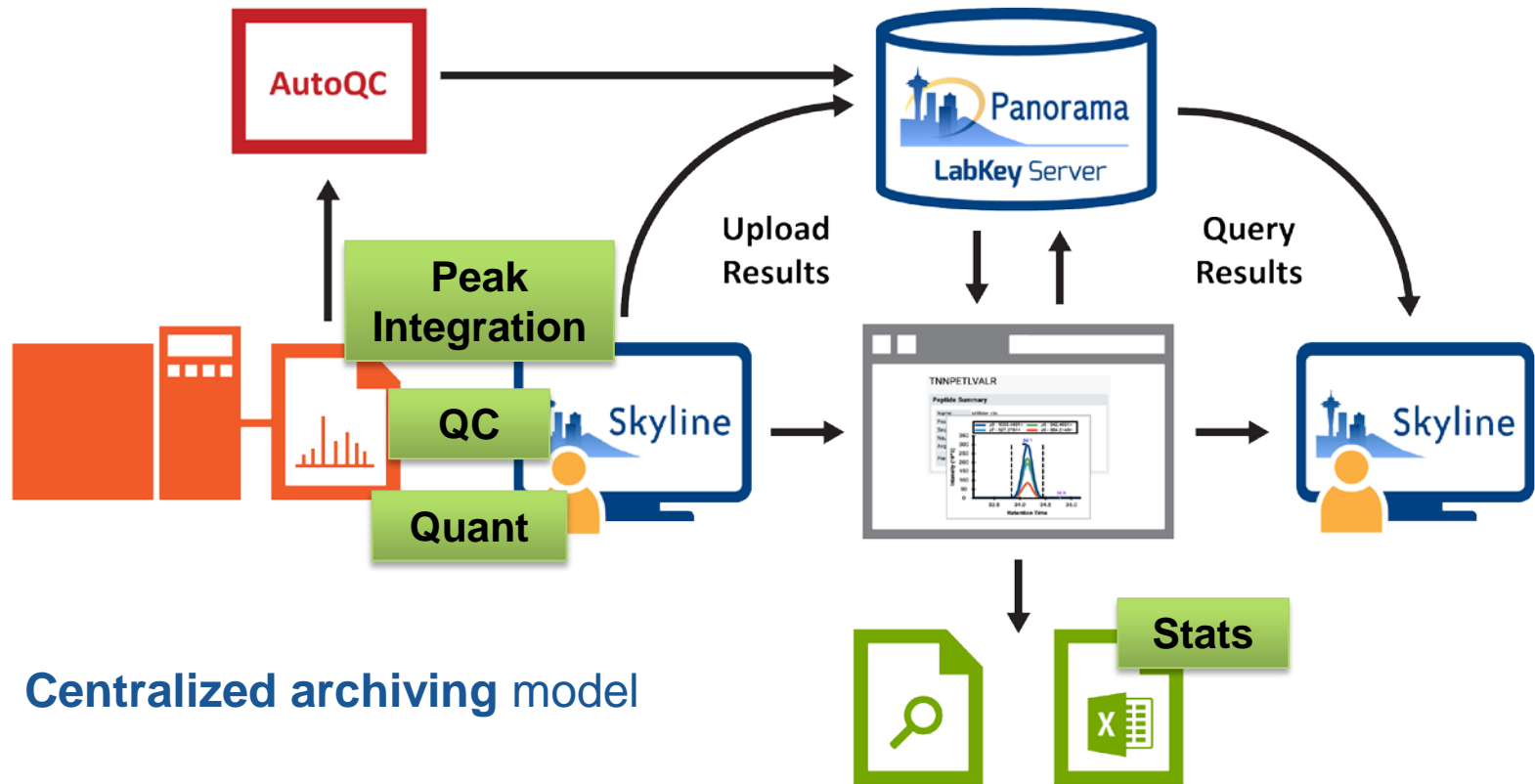
Triple Quad
(High sensitivity)

- 100 patients
- 2 timepoints
- 40 target analytes
- 5 transitions per analyte
- **8,000 peak groups**
- **40,000 transitions**

An ideal data analysis pipeline would be:

- **High throughput**
- **Reproducible**
- **Objective**
- **Automated**

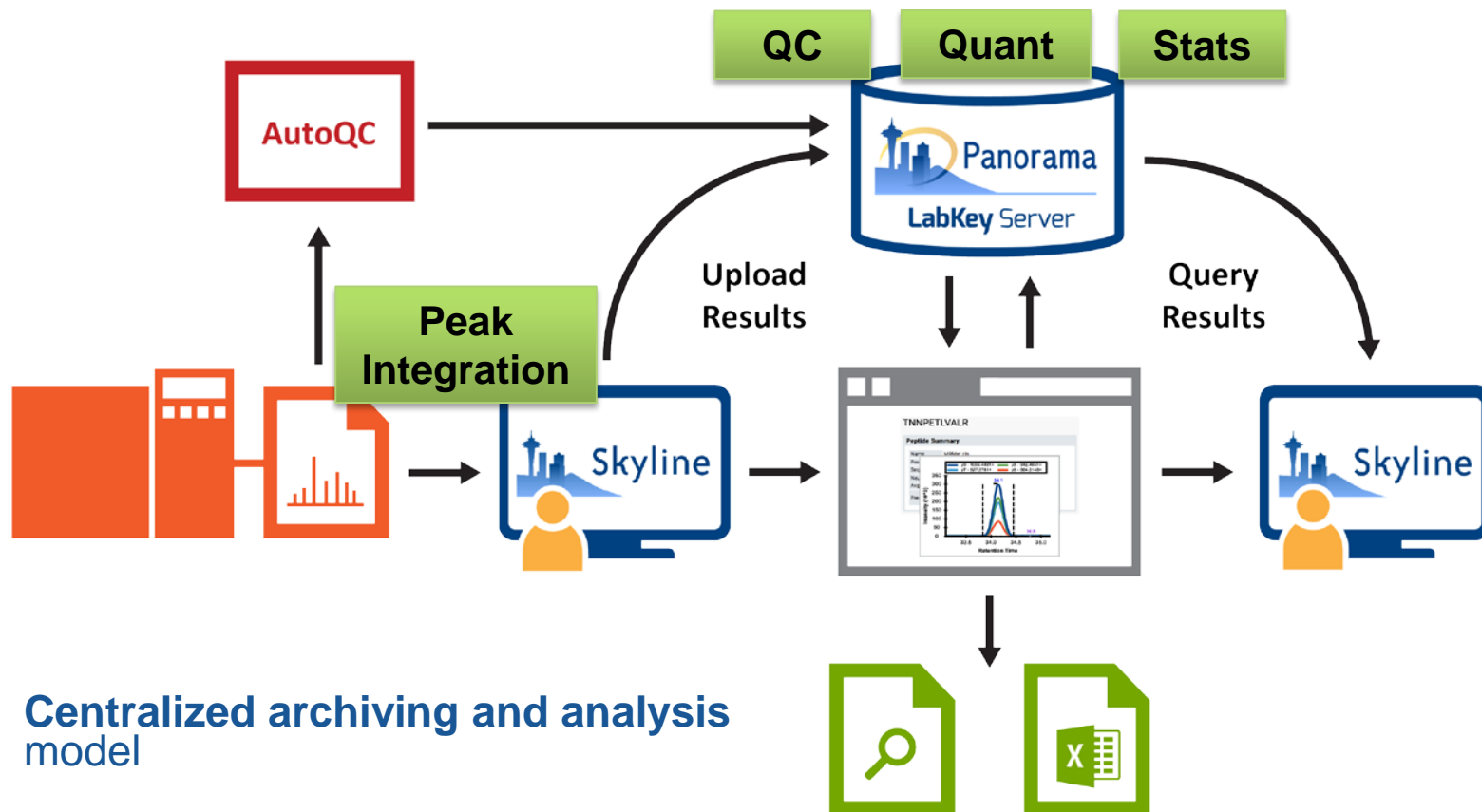
Panorama and skyline simplify data management and analysis for targeted MS assays



- Centralized archiving model
- Distributed analysis model

JASMS, November 2016

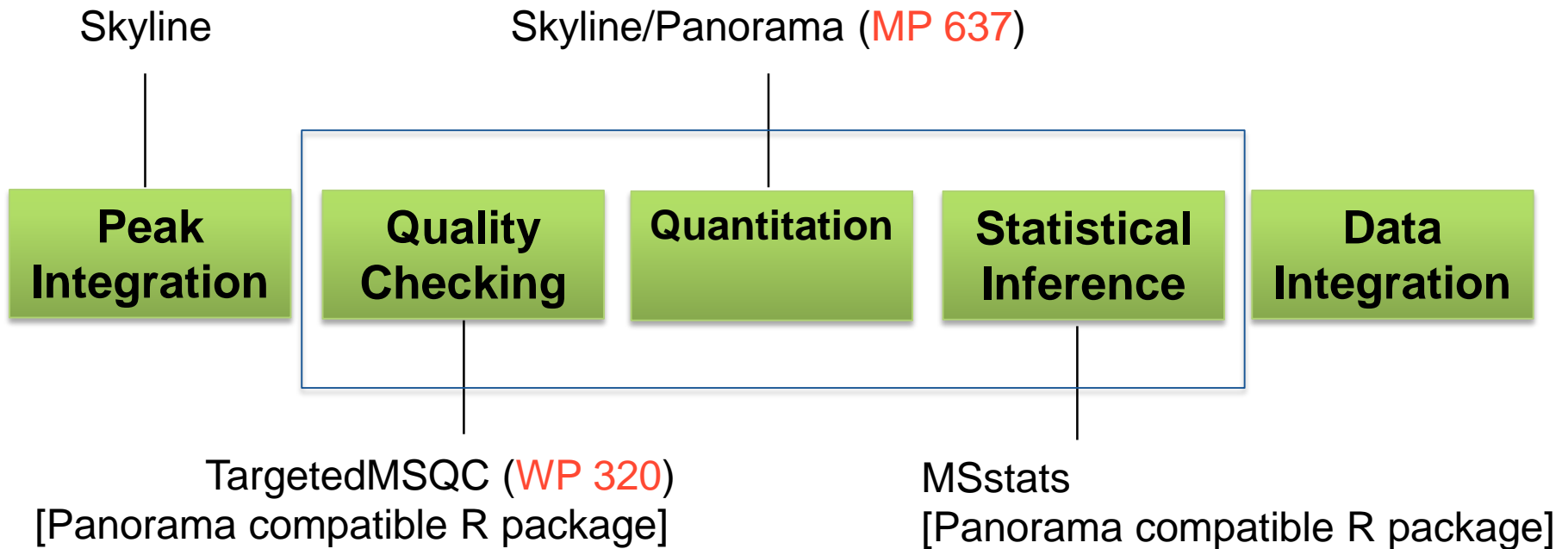
Enabling targeted MS post-processing on Panorama centralizes data archiving and analysis



- **Centralized archiving and analysis model**
- **Automated and objective QC**
- **Generation of results within minutes post data acquisition**

JASMS, November 2016

Targeted MS data analysis pipeline



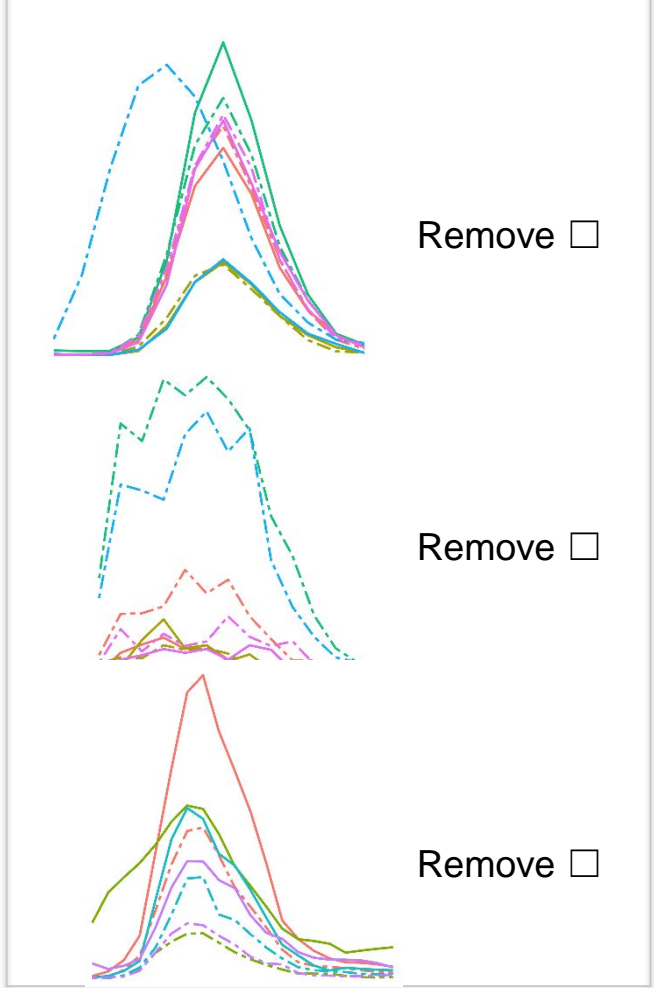
Quality Checking Report

QC Summary

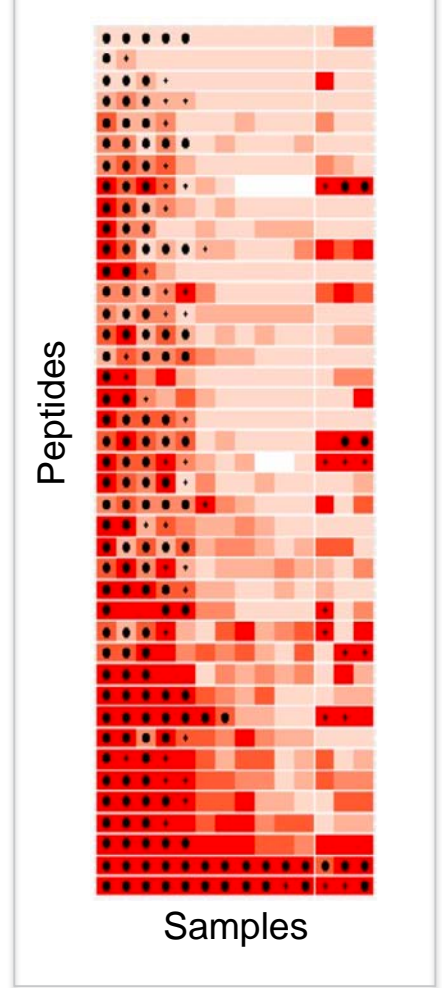
Samples	100
Peptides	30
Transitions	12000
High Quality	11500
Poor Quality	500

Remove all flagged transitions from downstream analysis

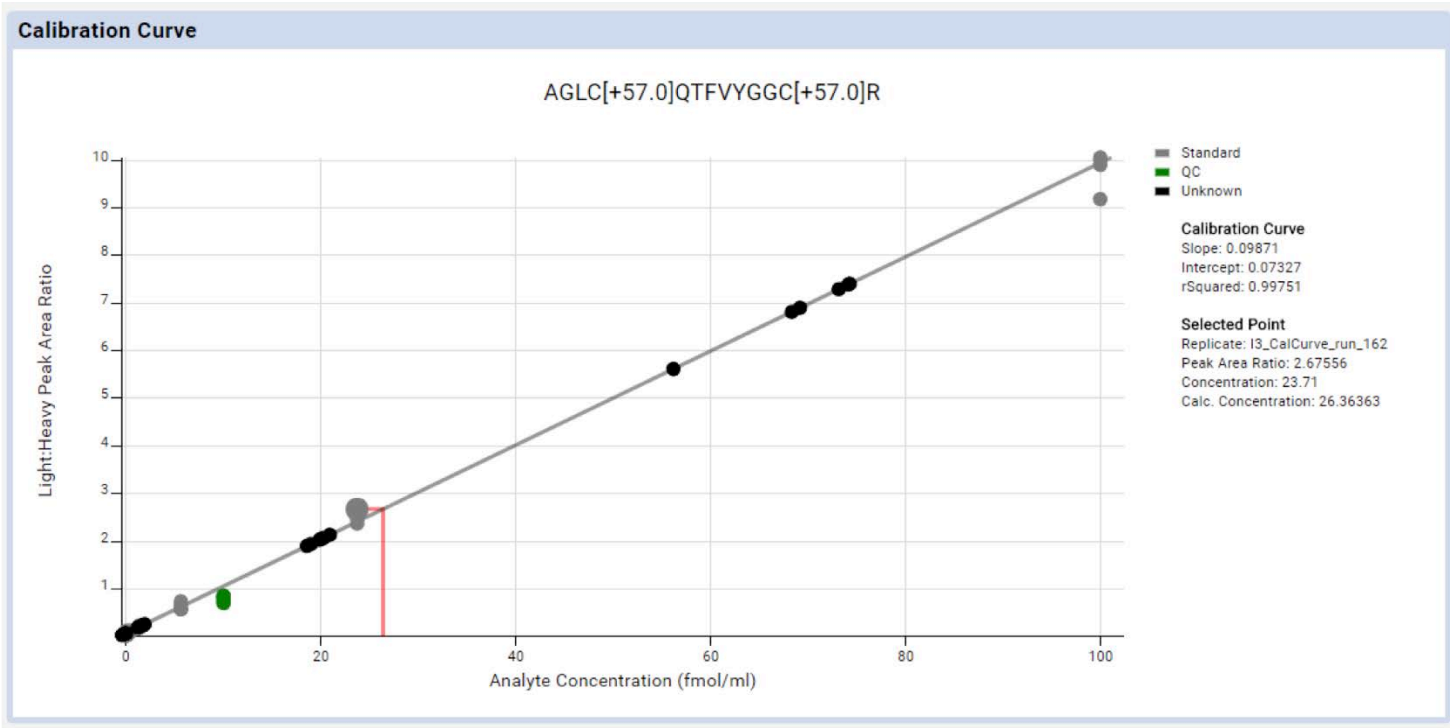
Chromatographic Peaks



Peptide QC Summary



Quantitation Report



Peptide Calibration Curves

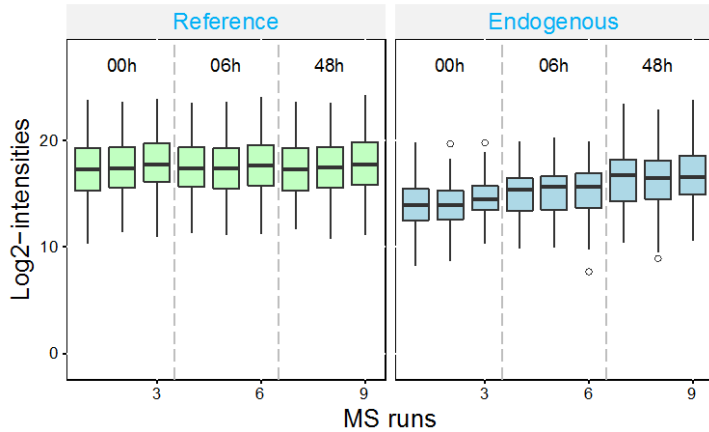
GRID VIEWS ▾ REPORTS ▾ CHARTS ▾ PAGING ▾ 1 - 5 of 5

<input type="checkbox"/>	Peptide	Slope	Intercept	Point Count	Quadratic Coefficient	RSquared	Error Message
<input type="checkbox"/> DETAILS ▾	FIC[+57.0]VTP TTC[+57.0]SNTIDLPMSPR	3.2642E-2	-2.7814E-2	27	0.0000E0	0.9994	
<input type="checkbox"/> DETAILS ▾	YC[+57.0]RPESQEHPEADPGSAAPYLK	5.5256E-2	2.1355E-2	27	0.0000E0	0.9995	
<input type="checkbox"/> DETAILS ▾	TSMGGTQQQFVEGVR	3.1720E-2	3.0357E-2	27	0.0000E0	0.9997	
<input type="checkbox"/> DETAILS ▾	RTSMGGTQQQFVEGVR	2.9297E-2	2.2693E-2	27	0.0000E0	0.9998	
<input type="checkbox"/> DETAILS ▾	LNTSDFQK	5.6347E-2	7.7697E-2	27	0.0000E0	0.9995	



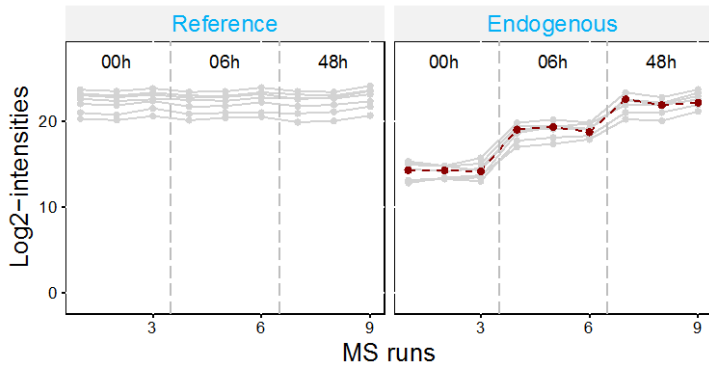
Profile Plots

All proteins



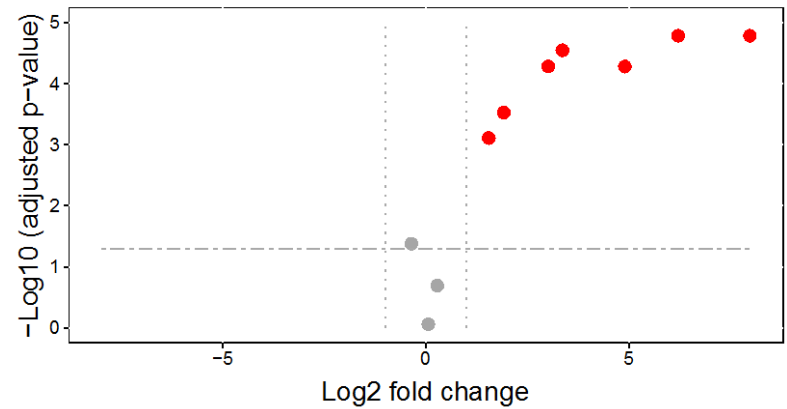
Rv2031c_hspX

— Processed feature-level data ● Run summary • Detected data



Group Comparison

48h-00h



Peptide	Sample	Condition	Concentration
Pep 1	A	0 h	0.13
Pep 2	A	0 h	0.24
Pep 1	B	48 h	0.48
Pep 2	B	48 h	0.19

Acknowledgements

Genentech

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Panorama Partners



Josh Eckels (Labkey)

Brendan MacLean (UW)

Vagisha Sharma (UW)

Nick Shulman (UW)

Michel Petrovic (Roche)

Daniel Spellman (Merck)

Related ASMS Posters:

TargetedMSQC: WP 320

Absolute Quant on Panorama: MP 637