



*MS Based Proteomics:
Recent Case Studies Using
Advanced Instrumentation*

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<http://mass-spec.stanford.edu/>



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Instrumentation and Capabilities

Proteomics

Front End(s)- Eksigent Nano2D LC



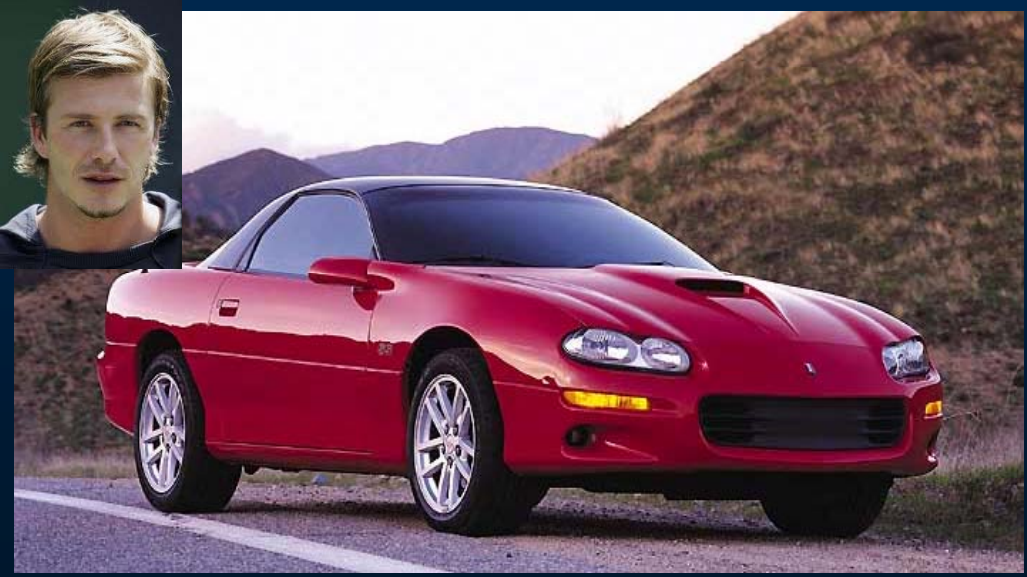
Mass Spectrometer(s)- LTQ Orbitrap Velos (ETD)
LTQ





Instrumental Upgrades:

- *More data of higher quality*





Sample Prep & Data Analysis-

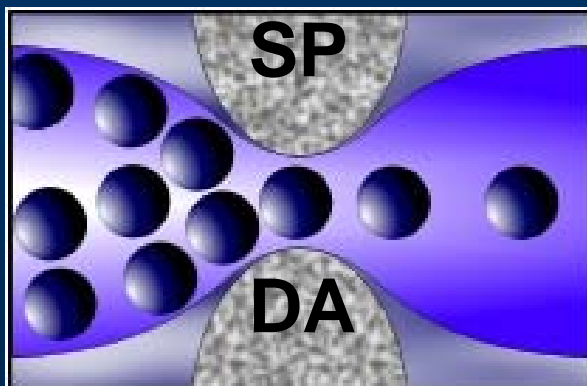
Sample Prep

-In-gel digests

-Solution

-Enrichments

**-Labels (iTRAQ,
TMT)**



Data Analysis

-Sequest

-Mascot

**-Scaffold for
visualization**

**-Statistics
(FDR)**

**-Qualitative v.
Quantitative**



Current Case Studies

- Global proteome analysis
- Quantitative Proteomics-
 - One task many alternatives.....
- PTM mapping





Project: 1 Global Proteome

Professor Virginia Walbot, Dr. Dave Skibbe
<http://www.stanford.edu/~walbot/>

Tumorigenic Fungal Peptides Causing Plant Cancer: Defining Expression Timing and Modeling Structural Similarities to Host Proteins

Ustilago maydis causes cancer within a few days of infecting maize by stimulating extra cell divisions, endopolyploidization (up to 64N), and enormous cell expansion. Although *U. maydis* grows on many hosts, only maize forms tumors where the pathogen can complete its lifecycle. Gene deletion demonstrated that secreted fungal proteins – many of which are small and encoded by gene families – are required for tumors.

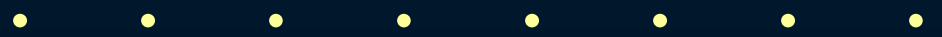
D. Skibbe, *et al.* Science 328, 89 (2010)



Project: 1 Global Proteome (cont.)

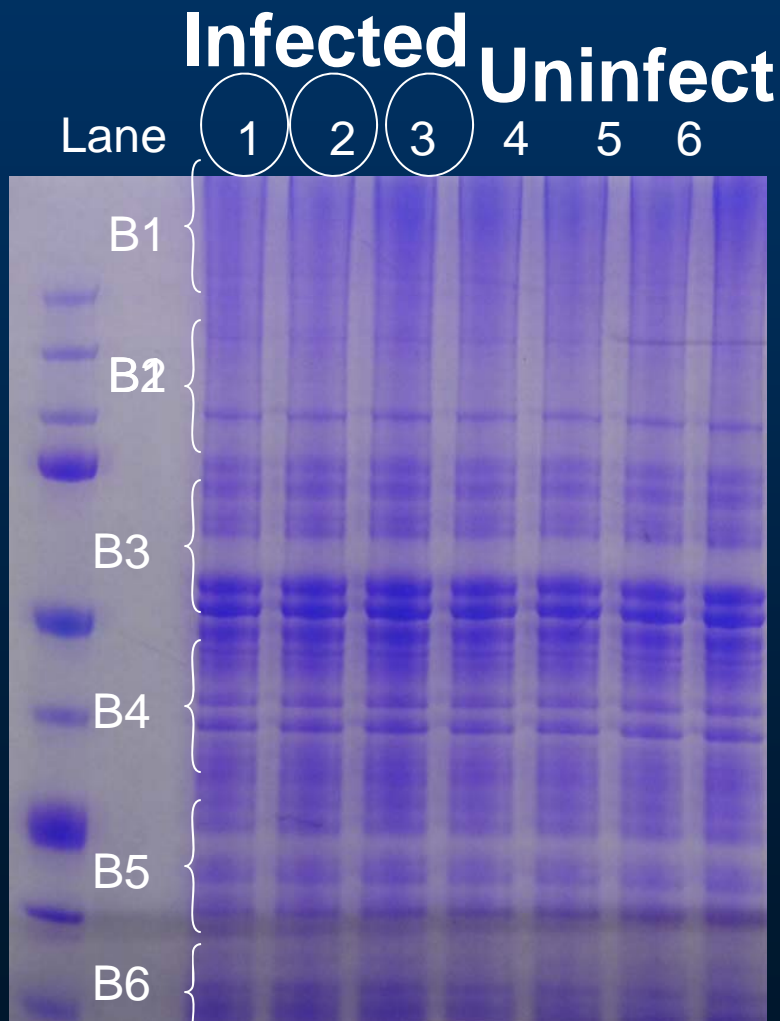


huitlacoche
WEET-LA-KO-CHEE





Global Proteome (cont.)

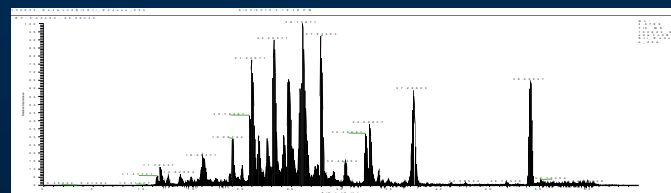


In-gel tryptic digest

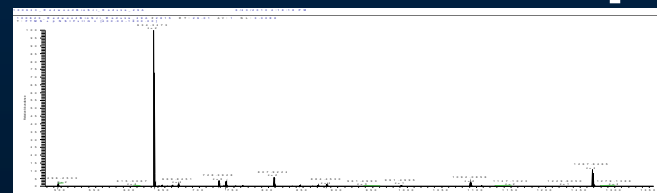


peptides

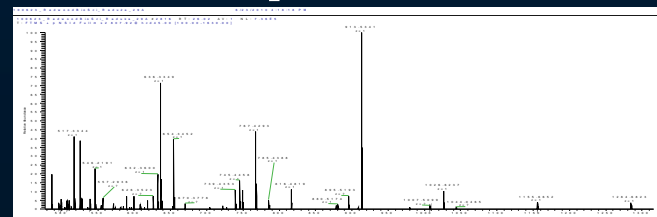
Reversed Phase nanoLC



Full Scan Orbitrap

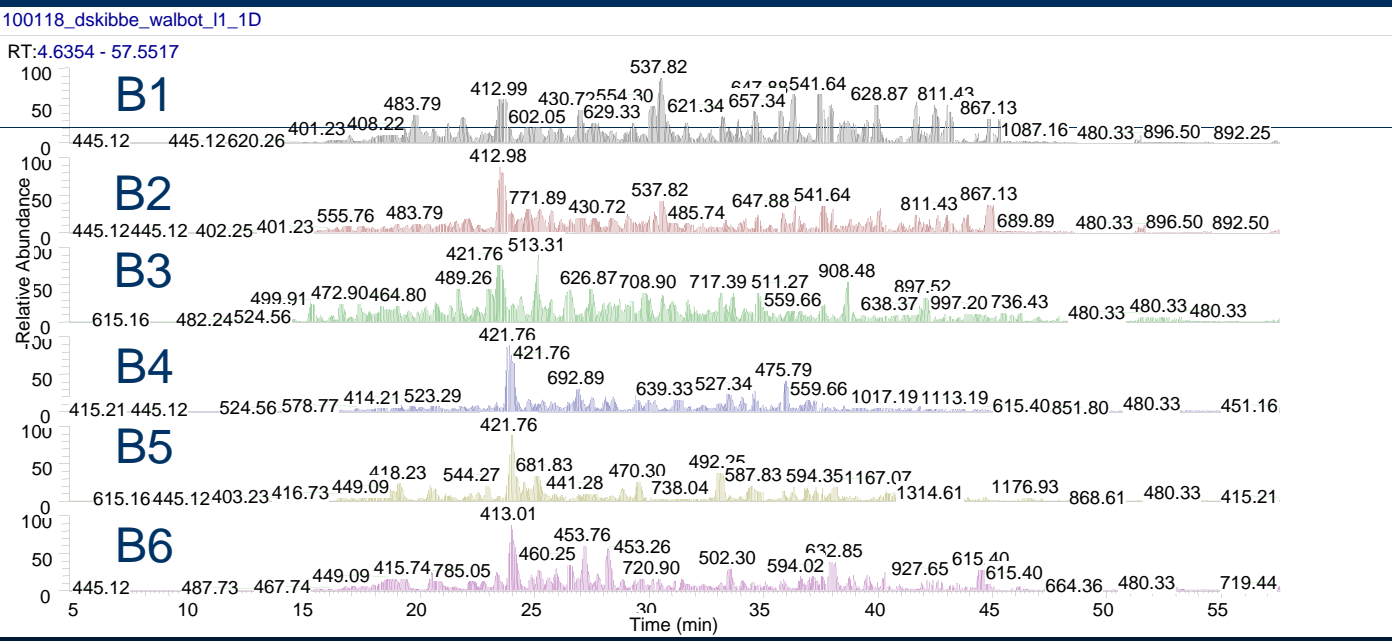


MS/MS top 15 Ion Trap





Global Proteome (cont.)



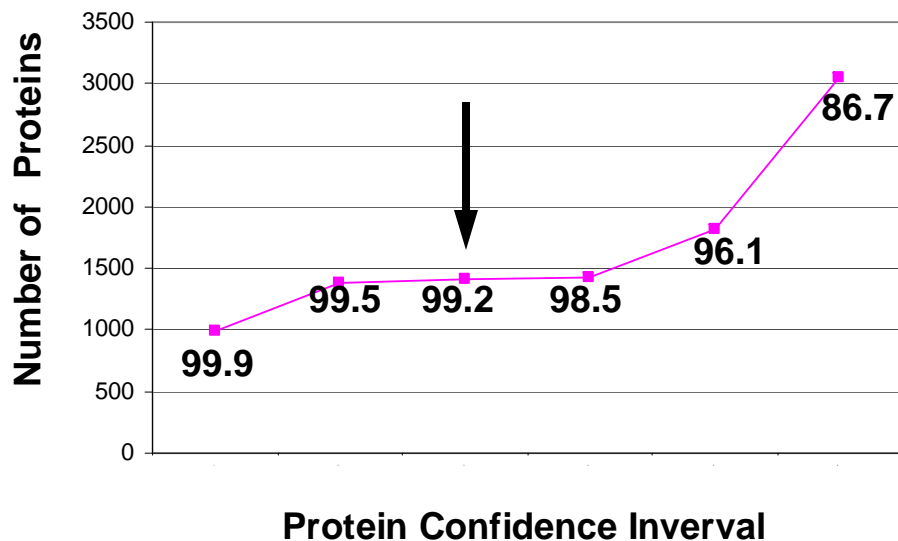
**Instrument
time:
6 bands x 6
lanes =
36 hours**



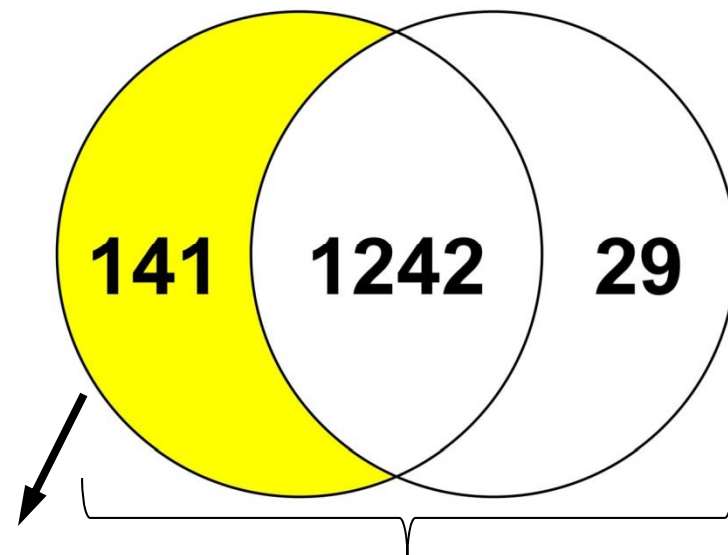
Global Proteome (cont.)

1 peptide ID
1412 proteins
0.8 % Protein FDR
5 % Peptide FDR

Protein Inference



Infected Mock



38 detected fungal proteins

Total 1412



Global Proteome (cont.)

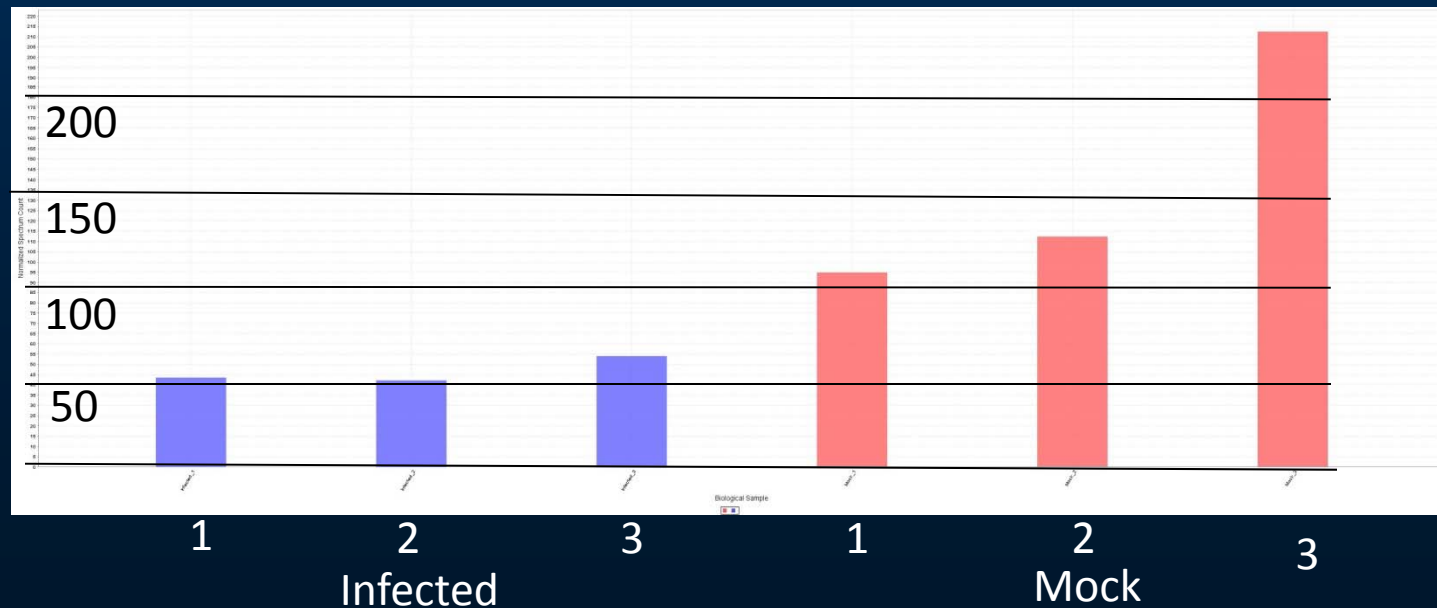
GRMZM2G021617_T01_3_3 (100%), 65,224.6 Da

XREF_ID=1

9 unique peptides, 13 unique spectra, 36 total spectra, 96/584 amino acids (16% coverage)

ERRICLPRCH	SGKRLLDRTD	ERRSAQKEKN	LGAEMAQAMA	SMTGLSQGV L
PSRRADSRT R	TAVVIVRASA	EGDAVAQAGR	RAVIGLVATG	IVGGALSQA A
RAETVKTIKI	GAPPPPSGGL	PGTLNSDQAR	DFDLPLKERF	YLQPLPPAEA
AARVK TSAQD	IINLKPLIDK	KAWPYVQNDL	RLRASYLRYD	LKTVIASPKK
EKKKSLKELT	GK LFSTIDDL	DHAAKIKSTP	EAEKYFAATK	DALGDVLA KL
GTQGLIMAMF	RTCVCSYGP G	KAMYHFLLIL	YQEALNYNA I	LSPQVPLRAY
ANIRKRYQTF	VIHYIKKVQN	KFGILNWSDE	SQITASVSHS	KLPMNSFWHF
CLSNIGMKL	KARSHTS SQF	SSESLIDSTI	QTAPQFRLLN	FQIVQNF DLV
TSVYNTLADK	PPAPKGVLLH	KPSPVCIKQQ	ASHYLPRDIQ	LPSSEFSSS
YLFVLCMRDFT	RLVITSTT CF	LPLLCQSSFC	FPTKVLRLG ST	NLVKALFNSS
LFLRREEAYF	HLTQI IHLFD	FCFPHYPHLS	IHFIVRCLQE	VF IYPTSFVN
APKRMDCNL D	SNYRYMNLAE	KTLNLNLWLR	SRSK	

Normalized Spectral Counts





Global Proteome (cont.)

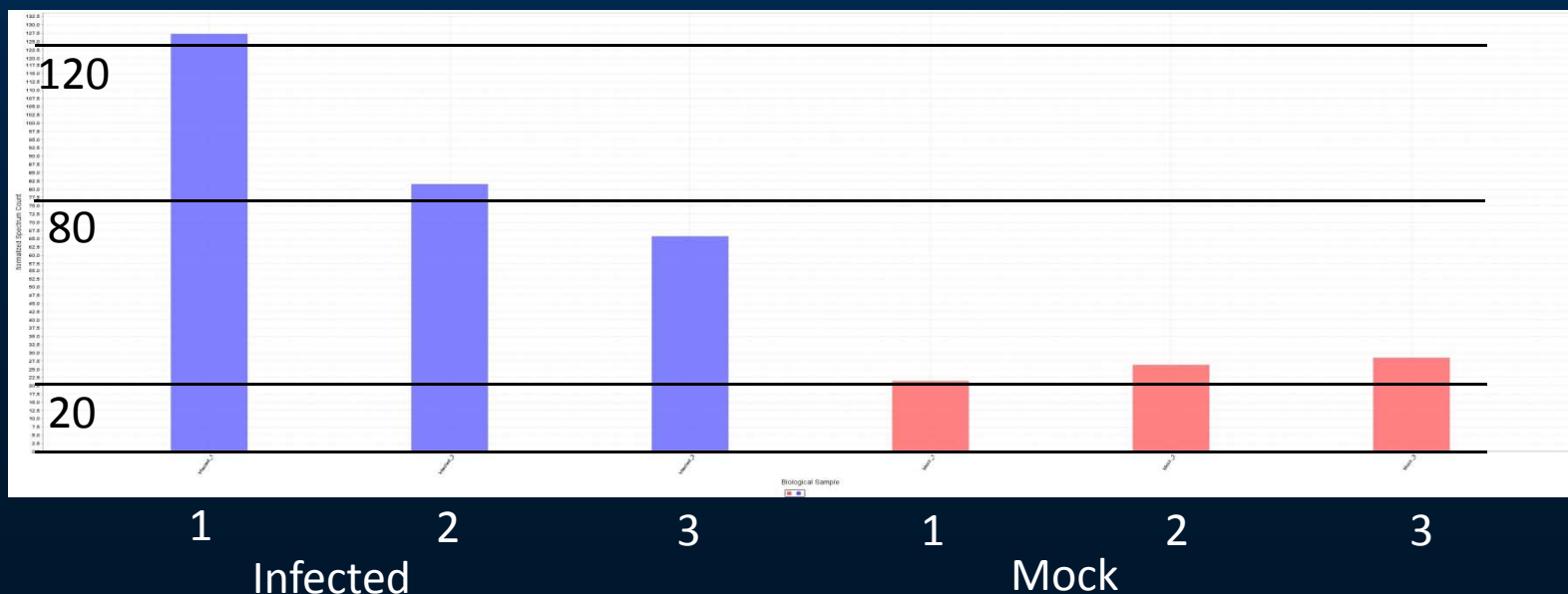
GRMZM2G020801_T02_2_2 (100%), 118,203.7 Da

XREF_ID=1

24 unique peptides, 28 unique spectra, 76 total spectra, 442/1078 amino acids (41% coverage)

A	E	A	F	L	C	S	S	S	C	C	L	T	L	C	A	A	T	V	H	A	F	K	D	I	L	T	S	L	P	K	P	G	G	G	E	Y	G	K	F	Y	S	L	P	A	L	N	D	P			
R	I	D	K	L	P	Y	S	V	I	L	L	E	S	A	I	R	N	C	D	N	F	Q	V	T	K	N	D	V	E	K	I	I	D	W	E	N	T	S	P	K	L	A	E	I	P	F	K	P			
A	R	V	L	L	Q	D	F	T	G	V	P	A	V	V	D	L	A	A	M	R	D	A	M	A	K	L	G	S	D	A	N	K	I	N	P	L	V	P	V	D	L	V	I	D	H	S	V	Q	V		
D	V	A	R	S	Q	N	A	V	Q	A	N	M	E	L	E	F	S	R	N	K	E	R	F	G	F	L	K	W	G	S	S	A	F	Q	N	M	L	V	V	S	S	A	F	Q	N	M	L	V	V		
N	L	E	Y	L	G	R	V	V	F	N	T	D	G	I	L	Y	P	D	S	V	V	G	T	D	S	H	T	T	M	I	D	G	L	G	V	A	G	W	G	V	G	I	E	A	E	A	T	M			
L	G	Q	P	M	S	M	V	L	P	G	V	V	G	F	K	L	T	G	K	L	R	S	G	V	T	A	T	D	L	V	L	T	V	T	Q	M	L	R	K	H	G	V	V	G	K	F	V	E	F		
Y	G	E	G	M	G	K	L	S	L	A	D	R	A	T	I	A	N	M	S	P	E	Y	G	A	T	M	G	F	F	P	V	D	H	V	T	L	D	Y	L	K	L	T	G	R	S	D	E	T	V		
S	M	I	E	A	Y	L	R	A	N	K	M	F	V	D	Y	N	E	P	P	T	E	R	I	Y	S	S	Y	L	E	L	N	L	D	E	V	E	P	S	M	S	G	P	K	R	H	D	R	V			
P	L	K	E	M	K	S	D	W	H	A	C	L	D	N	K	V	G	F	K	G	F	A	V	P	K	E	Q	Q	D	K	V	V	K	F	D	F	H	G	Q	P	A	E	M	K	H	G	S	V	V		
I	A	A	I	T	S	C	T	N	T	S	N	P	S	V	M	L	G	A	G	L	V	A	K	K	A	C	E	L	G	L	E	V	K	P	W	V	K	T	S	L	A	P	G	S	G	V	T	K			
Y	L	L	Q	S	G	L	Q	E	Y	L	N	Q	Q	G	F	H	I	V	G	Y	G	C	T	T	C	I	G	N	S	G	D	L	D	E	S	V	S	T	A	I	T	E	N	D	V	V	A	A	A		
V	L	S	C	N	P	N	E	F	T	C	R	V	H	P	L	T	R	A	N	Y	L	A	S	P	R	L	V	V	A	Y	A	L	A	C	T	V	D	L	D	E	F	E	K	E	R	L	C	E	C	K	D

Normalized Spectral Counts





Quantitative Proteomics Ex. 1

Spectral Counting

- Spectral count correlates well with protein abundance
- Fold change can be calculated and statistically evaluated
- Simple and straightforward implementation
- Sensitive to protein abundance changes – for abundant proteins 2 fold change easily detected with high confidence

Limitations

- The response to increasing protein amount is saturable
- Noisy data at low spectral counts – **large** difference in spectral count necessary to determine significant change



Quantitative Proteomics Ex. 1

Dr. Jonathan Rothbard, Steinman Lab

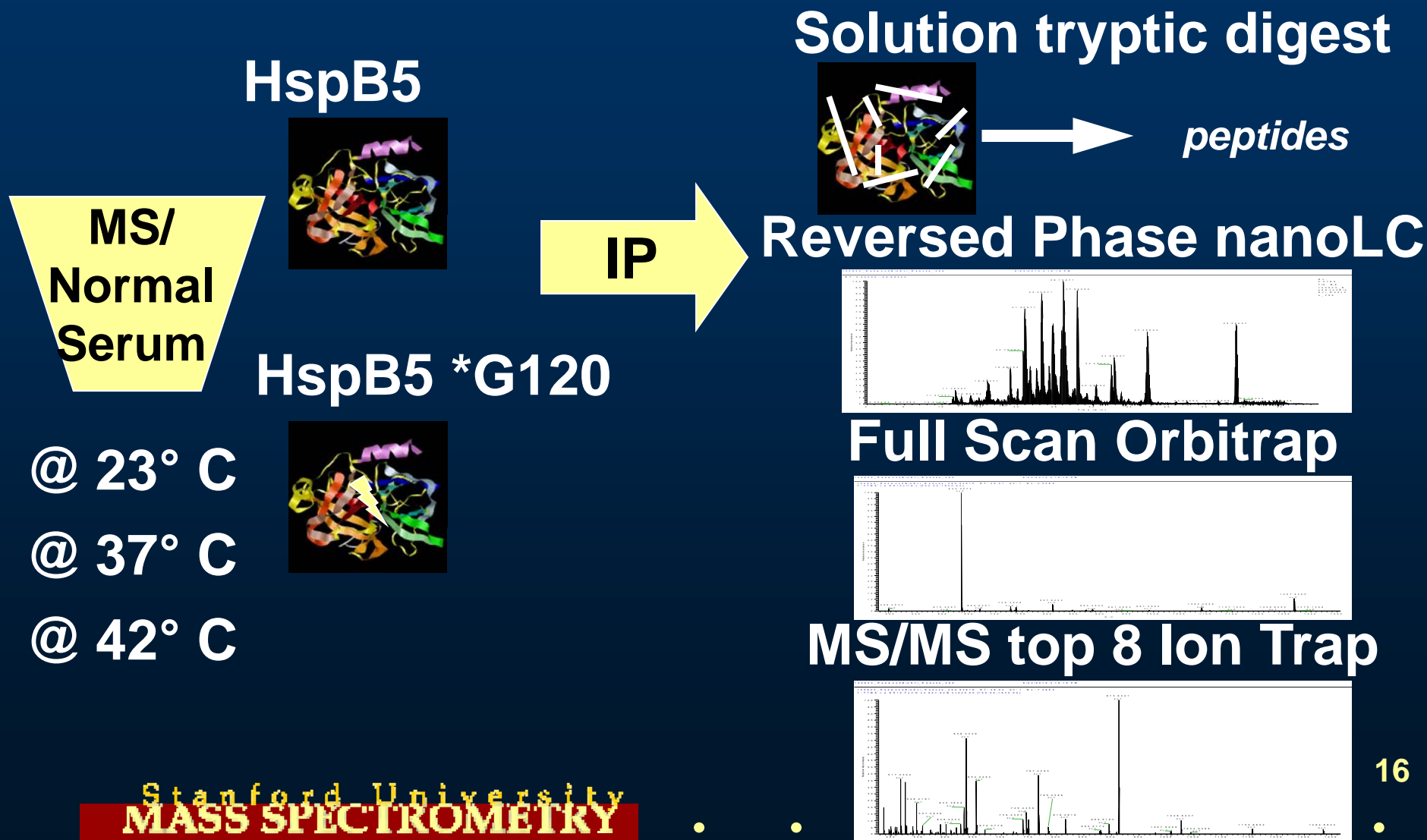
<http://steinmanlab.stanford.edu/index.html>

Define the serological clients of sHspB5 in normal and patient's plasma

Small heat shock proteins are a family of intracellular chaperones that are cytoprotective due to their ability to bind partially unfolded proteins and prevent aggregation. Recently, HspB5 (alpha B crystallin) has been shown to be therapeutically effective when administered intraperitoneally in animal models of multiple sclerosis, rheumatoid arthritis, stroke, and ischemia reperfusion injury. In all cases, there is significant immune suppression. To test the hypothesis that the mode of action is the chaperone activity of the protein in plasma, the proteomic content of clients has been defined by mass spectrometry. More specifically, proteins associated with HspB5 in normal and patient's plasma.



Quantitative Proteomics Ex. 1



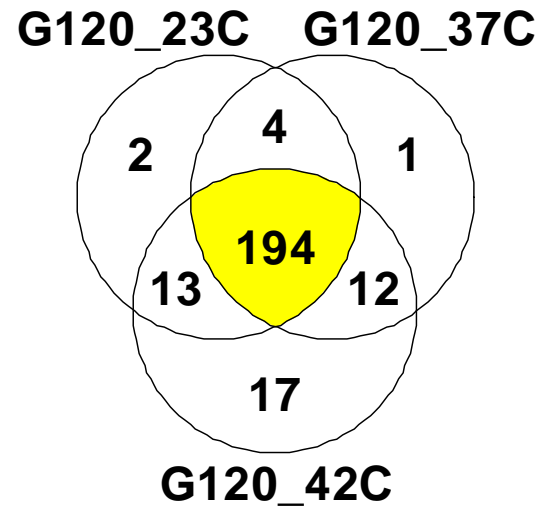
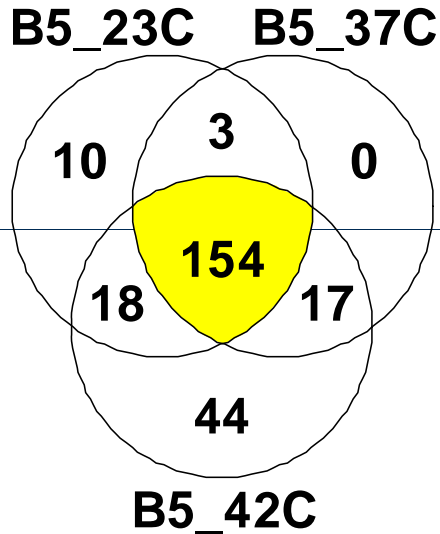


Quantitative Proteomics Ex. 1

Molecular Weight	Protein Grouping Ambiguity	B5_23C						B5_37C						B5_42C						G120_23C						G120_37C						G120_42C							
		0729_B5_23C_01	0729_B5_23C_02	0729_B5_23C_03	0729_B5_23C_04	0729_B5_23C_05	0729_B5_23C_06	0729_B5_37C_01	0729_B5_37C_02	0729_B5_37C_03	0729_B5_37C_04	0729_B5_37C_05	0729_B5_37C_06	0729_B5_42C_01	0729_B5_42C_02	0729_B5_42C_03	0729_B5_42C_04	0729_B5_42C_05	0729_B5_42C_06	0729_G120_23C_01	0729_G120_23C_02	0729_G120_23C_03	0729_G120_23C_04	0729_G120_23C_05	0729_G120_23C_06	0729_G120_37_01	0729_G120_37_02	0729_G120_37_03	0729_G120_37_04	0729_G120_37_05	0729_G120_37_06	0729_G120_42C_01	0729_G120_42C_02	0729_G120_42C_03	0729_G120_42C_04	0729_G120_42C_05	0729_G120_42C_06		
516 kDa	*	54	73	65	25	28	24	23	34	29	23	28	20	76	77	68	54	55	49	55	40	70	34	41	49	28	43	31	46	67	64	77	33	69	93	28	74		
187 kDa	*	52	66	61	58	63	51	70	75	62	60	59	49	85	84	66	70	77	53	63	58	81	51	45	54	51	67	50	51	67	67	81	45	69	84	42	64		
69 kDa	*	23	36	31	30	39	25	30	31	26	28	26	21	36	36	22	38	35	24	22	22	30	24	23	26	22	28	23	18	27	31	46	24	28	45	19	27		
280 kDa	*	19	35	31	20	28	32	1	1	1	17	34	16	7	8	5	42	49	18	10	11	24	18	20	23	12	26	13	11	23	17	7	16	40	12	29			
270 kDa	*	19	29	24	27	28	24	1	1	1	23	25	17	4	7	3	30	30	21	9	12	20	16	19	21	10	16	10	11	16	16	13	5	20	33	9	30		
56 kDa	*	16	20	20	18	20	18	16	21	20	19	22	18	33	32	31	31	31	27	22	19	26	24	23	23	22	25	18	18	23	27	35	25	32	36	24	32		
20 kDa	*	13	18	18	17	19	15	17	19	16	21	18	17	21	20	16	19	22	16	13	9	14	13	10	8	5	12	9	9	11	14	13	13	14	16	15	10	12	12
66 kDa	*	13	16	13	13	15	12	14	14	12	15	13	10	17	16	12	16	16	9	9	6	12	10	10	10	9	13	9	7	10	13	16	10	13	15	6	10		
188 kDa	*	12	16	18	17	17	17	17	23	24	35	38	29	57	59	44	57	53	38	29	29	41	19	21	23	29	45	31	20	29	28	61	25	50	54	20	43		
42 kDa	*	10	12	11	11	12	10	6	9	8	11	11	9	10	13	9	14	13	10	8	5	12	9	6	10	9	12	7	6	7	10	14	7	9	15	7	11		
16 kDa	*	10	12	10	11	11	7	10	11	8	11	9	7	11	12	6	12	13	6	7	10	10	5	5	6	6	9	7	5	10	8	13	6	10	14	10	11		
227 kDa	*	10	23	19	14	18	15	1	2	12	14	12	8	7	3	39	34	23	8	5	13	13	13	14	8	13	8	5	11	12	23	9	18	33	12	21			
501 kDa	*	10	14	11	3	3	3	4	5	5	3	3	1	12	13	13	5	4	4	8	4	9	3	4	3	2	3	2	4	7	6	5	4	3	18	5	15		
95 kDa	*	9	19	16	17	16	14	14	16	12	15	13	9	24	22	15	24	24	15	9	8	15	9	10	11	9	17	7	7	11	15	31	17	23	32	11	20		
91 kDa	*	9	13	9	8	13	7	4	10	6	13	12	10	23	25	15	25	25	19	8	3	10	12	9	11	13	18	11	5	7	9	29	18	25	26	7	21		
31 kDa	*	9	11	9	12	12	11	11	13	12	12	12	9	14	16	14	16	15	9	10	9	14	10	10	10	10	12	9	9	9	10	20	9	17	17	6	14		
52 kDa	*	8	14	14	13	13	10	12	16	11	15	12	8	20	21	15	22	22	13	10	8	15	8	10	9	11	18	7	7	14	15	27	13	19	26	13	18		
53 kDa	*	8	13	11	10	13	13	11	10	11	13	14	10	15	14	11	16	14	9	10	8	12	10	9	9	12	11	7	7	11	11	16	13	16	15	9	13		
23 kDa	*	8	9	8	7	8	7	7	10	8	8	8	9	8	9	9	12	11	10	8	7	9	7	8	7	10	10	8	8	8	8	14	7	10	11	7	8		
117 kDa	*	8	10	10	11	14	7	7	10	8	8	8	6	5	4	4	3	18	16	11	3	3	6	7	5	7	2	6	2	1	6	5	12	1	7	24	4	12	
73 kDa	*	8	7	10			0																																
47 kDa	*	7	7	7	9	9	10	8	8	8	8	10	7	13	14	13	13	14	12	10	6	12	10	9	10	11	14	11	6	8	9	12	7	16	13	4	15		
52 kDa	*	6	13	10	11	13	9	11	11	10	13	10	7	14	13	6	15	15	7	6	6	14	6	6	8	6	11	7	3	7	10	16	10	13	16	8	11		
25 kDa	*	6	7	7	8	8	8	7	7	5	8	7	7	8	9	8	8	9	5	5	5	8	5	5	6	6	6	5	4	6	7	9	5	8	8	5	8		
141 kDa	*	6	11	7	6	7	6	6	8	7	11	11	8	25	22	18	18	20	16	9	6	11	3	5	7	5	9	7	3	5	4	26	14	18	27	8	19		
15 kDa	*	6	6	6	6	6	5	6	6	7	7	7	5	8	8	6	8	8	7	7	7	8	7	4	7	6	7	6	5	7	8	8	6	7	9	6	9		
53 kDa	*	6	8	6	7	7	7	5	5	6	6	5	10	10	7	12	10	7	5	3	7	7	7	6	5	8	6	4	6	9	11	8	9	9	3	6			
51 kDa	*	6	7	5	6	6	6	5	7	6	8	7	7	10	10	11	10	10	10	7	7	8	5	6	7	8	9	6	7	8	8	14	5	9	13	6	10		
103 kDa	*	6	11	8	6	11	7	4	1	6	8	6	2	4	3	16	17	15	4	3	7	10	7	14	2	4	5	3	4	5	7	2	6	20	8	17			
47 kDa	*	5	4	4	6	5	5	4	5	5	4	3	1	10	10	4	11	9	3	3	1	6	4	4	3	2	7	3	2	3	4	13	4	6	11	1	4		
38 kDa	*	5	7	8	2	3	2	1	2	3	2	3	3	7	5	6	5	5	6	1	3	4	6	5	7	3	6	4	3	2	4	10	6	8	6	3	7		
163 kDa	*	5	12	12	24	24	22	17	21	21	27	30	22	44	44	32	51	48	32	19	20	28	20	26	22	25	39	25	14	22	22	51	24	43	49	21	38		
193 kDa	*	5	11	8	7	11	9	12	13	14	7	10	10	33	33	33	22	23	22	18	20	21	15	15	16	11	22	15	12	17	16	29	9	27	38	13	30		
77 kDa	*	5	9	9	17	19	17	6	13	9	6	10	6	19	22	15	23	22	11	8	4	11	15	9	16	11	14	8	2	4	4	26	7	23	23	6	15		

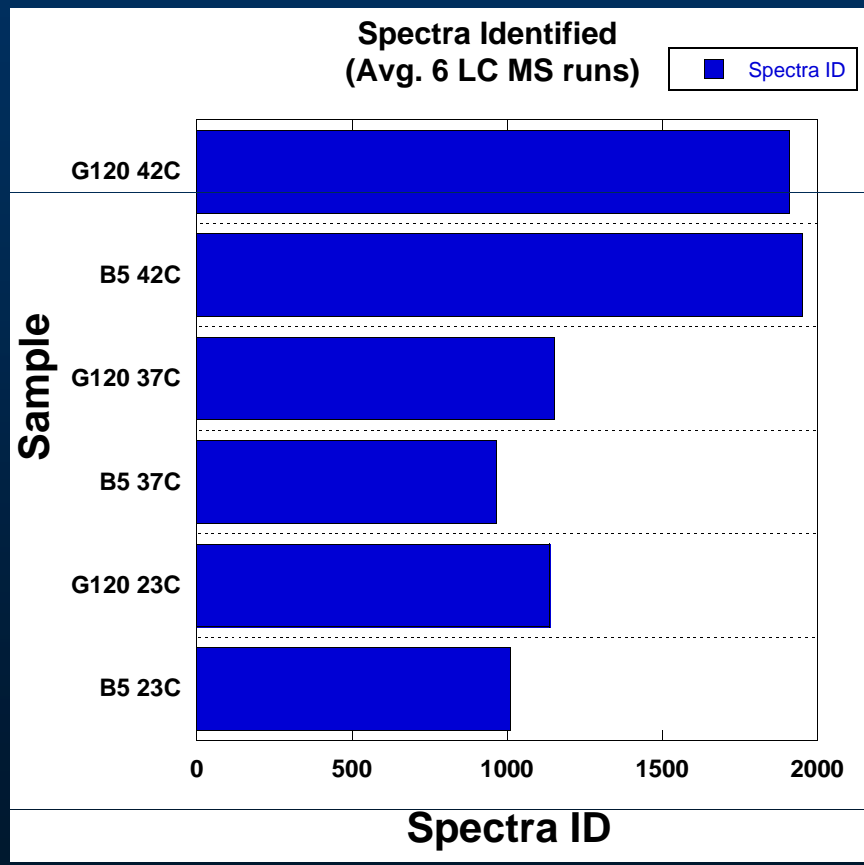
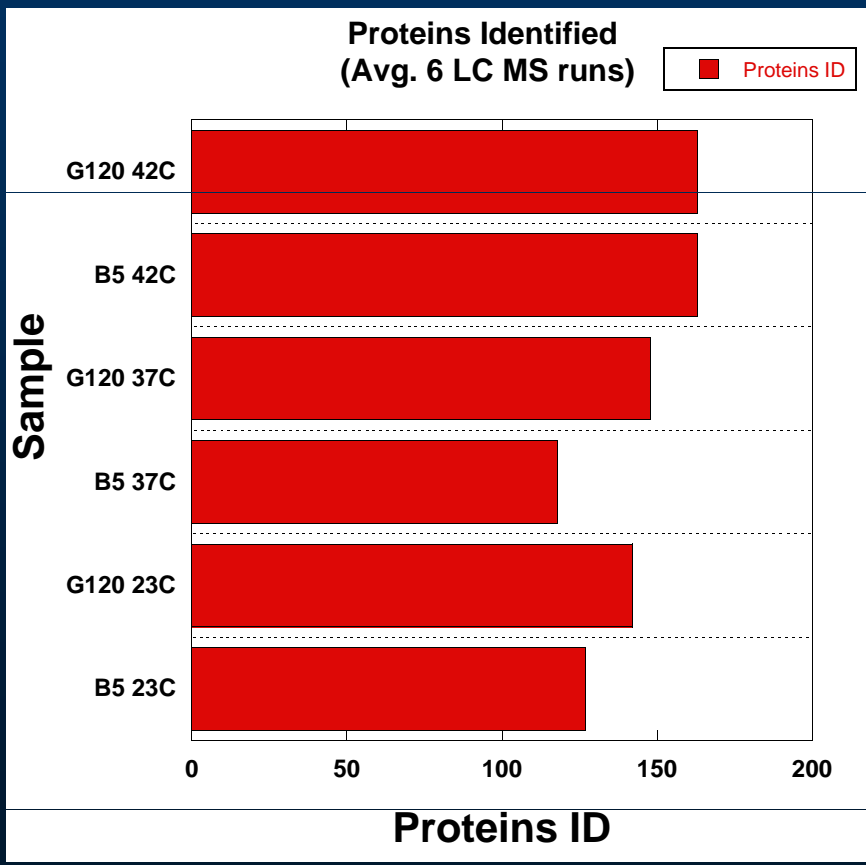


Quantitative Proteomics Ex. 1





Quantitative Proteomics Ex. 1

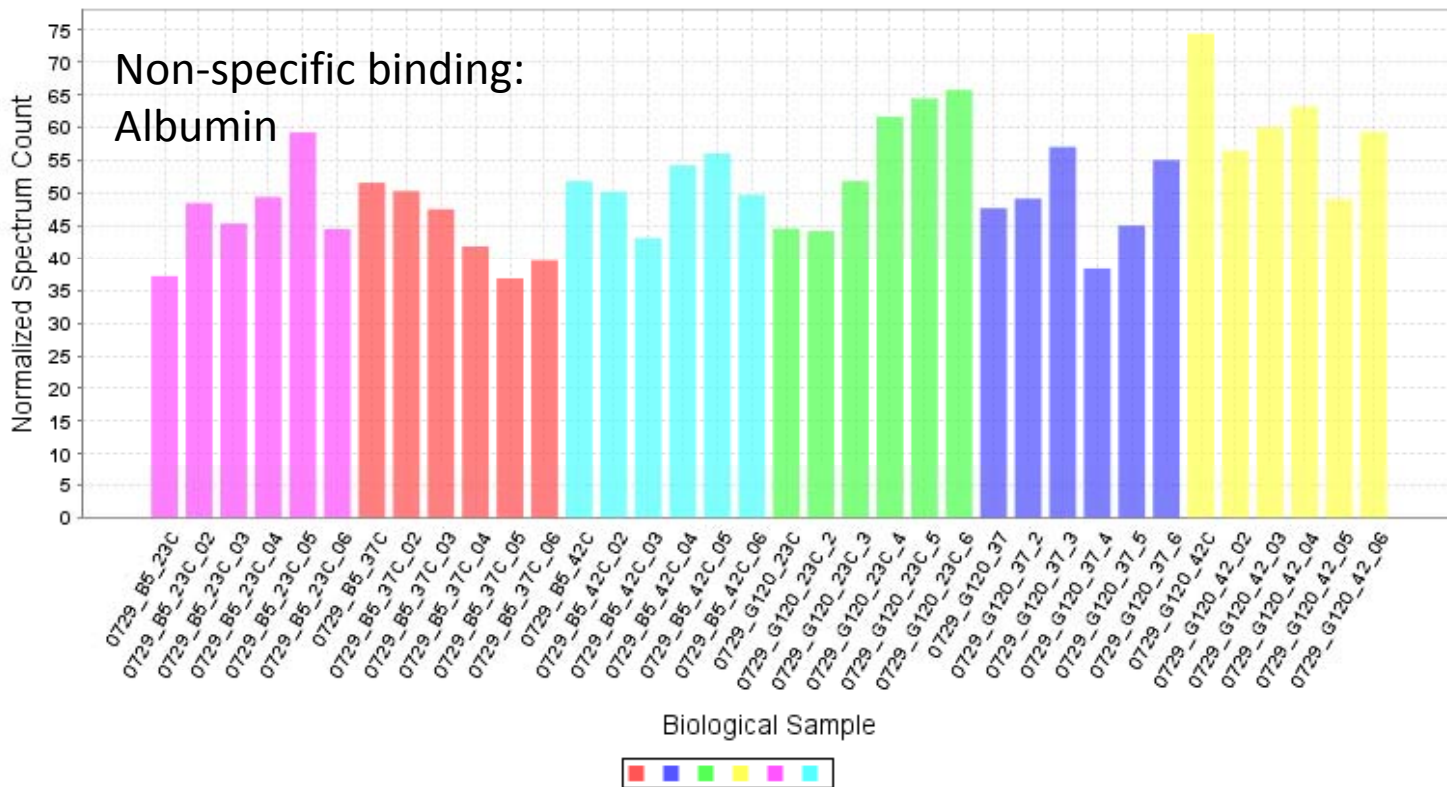




Quantitative Proteomics Ex. 1

IPI00745872 (100%), 69,366.9 Da
 IPI:IPI00745872.2 Tax_Id=9606 Gene_Symbol=ALB Isoform 1 of Serum albumin
 45 unique peptides, 64 unique spectra, 81 total spectra, 438/609 amino acids (72% coverage)

MK	WV	TF	IS	LL	FL	FSS	AYS	SR	G	V	FRR	DA	H	K	S	E	V	A	H	R	F	K	D	L	G	E	E	N	F	K	A	L	V	L	I	A	F	A	Q	Y	L	Q	C	P	F	E	D	H	V	K	L	V	N	E	V	T	E	F	A	K	T	C	V	A	D															
ES	AE	NC	DK	SL	HT	LF	GD	KL	CT	V	A	T	L	R	E	T	Y	G	E	M	A	D	C	C	A	K	Q	E	P	E	R	N	E	C	F	L	Q	H	K	D	D	N	P	N	L	P	R	L	V	R	P	E	V	D	V	M	C	T	A	F	H	D	N	E	E	T	F	L	K											
KY	LY	E	I	A	R	R	H	P	Y	F	A	P	E	L	L	F	F	A	K	R	Y	K	A	A	F	T	E	C	Q	A	A	D	K	A	A	C	L	L	P	K	L	D	E	L	R	D	E	G	K	A	S	S	A	K	Q	R	L	K	C	A	S	L	Q	K	F	G	E	R	A	F	K	A	W	A	V					
AR	LS	S	Q	R	F	P	K	A	E	F	A	E	V	S	K	L	V	T	D	L	T	K	V	H	T	E	C	C	R	A	D	L	A	K	Y	I	C	E	R	H	P	D	Y	S	V	L	L	L	R	L	A	K	T	Y	E	T	T	L	E	K	C	C	A	A	A	D	P	H	E	C	Y	A	K	V	F	D	E			
EM	P	A	D	L	P	S	L	A	A	D	F	V	E	S	K	D	V	C	K	N	Y	A	E	A	K	D	V	F	L	G	M	F	L	Y	E	Y	A	R	R	H	P	D	Y	S	V	L	L	L	L	R	L	A	K	T	Y	E	T	T	L	E	K	C	C	A	A	A	D	P	H	E	C	Y	A	K	V	F	D	E		
FK	P	L	V	E	E	P	Q	N	L	I	K	Q	N	C	E	L	F	E	Q	L	G	E	Y	K	F	Q	N	A	L	L	V	R	Y	T	K	K	V	P	Q	V	S	T	P	T	L	V	E	V	Y	V	P	K	E	F	N	A	E	T	Y	V	P	K	E	F	N	A	E	T	Y	V	P	K	E	F	N	A	E	T		
LN	Q	L	C	V	L	H	E	K	T	P	V	S	D	R	V	T	K	C	C	T	E	S	L	V	N	R	R	P	L	L	V	R	Y	T	K	K	V	P	Q	V	S	T	P	T	L	V	E	V	Y	V	P	K	E	F	N	A	E	T	Y	V	P	K	E	F	N	A	E	T												
P	K	A	T	K	E	Q	L	K	A	V	M	D	D	F	A	A	F	V	E	K	C	C	K	A	D	D	K	E	T	C	F	A	E	E	G	K	K	L	V	A	A	S	Q	A	A	L	G	L	F	T	F	H	A	D	I	C	T	L	S	E	K	E	R	Q	I	K	K	Q	T	A	L	V	E	L	V	L	V	K	H	K

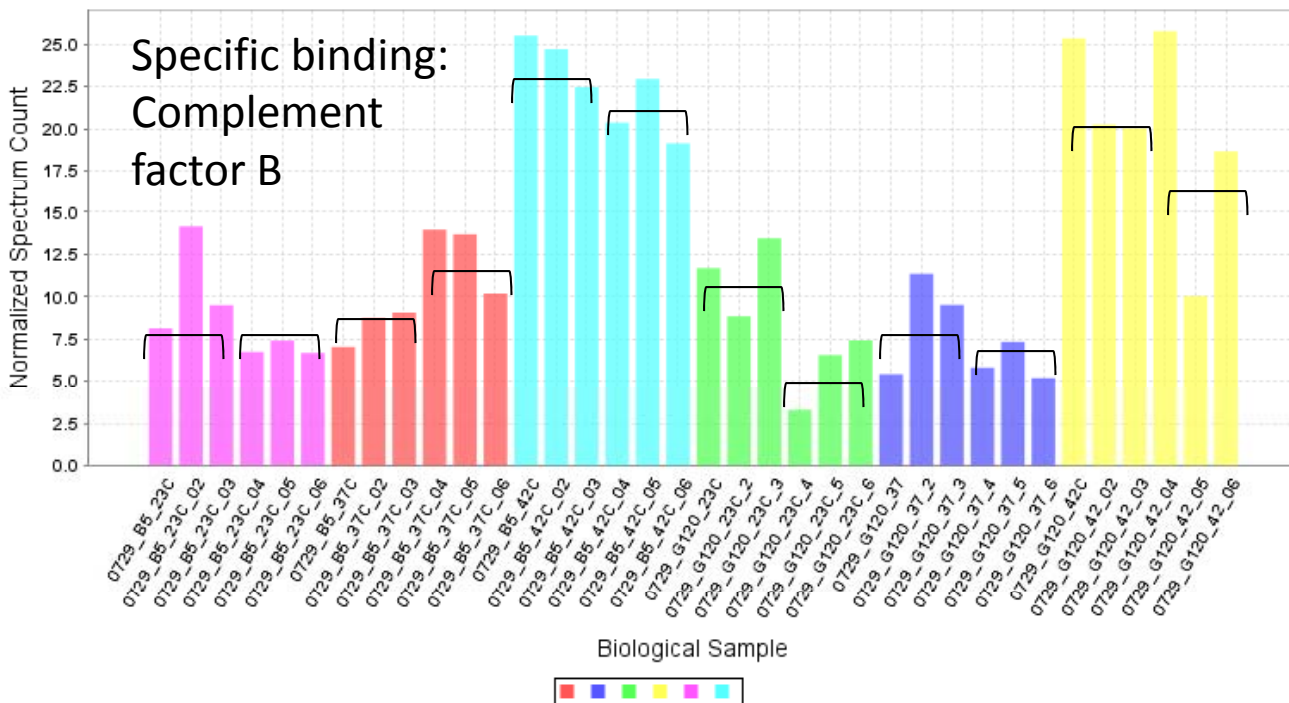




Quantitative Proteomics Ex. 1

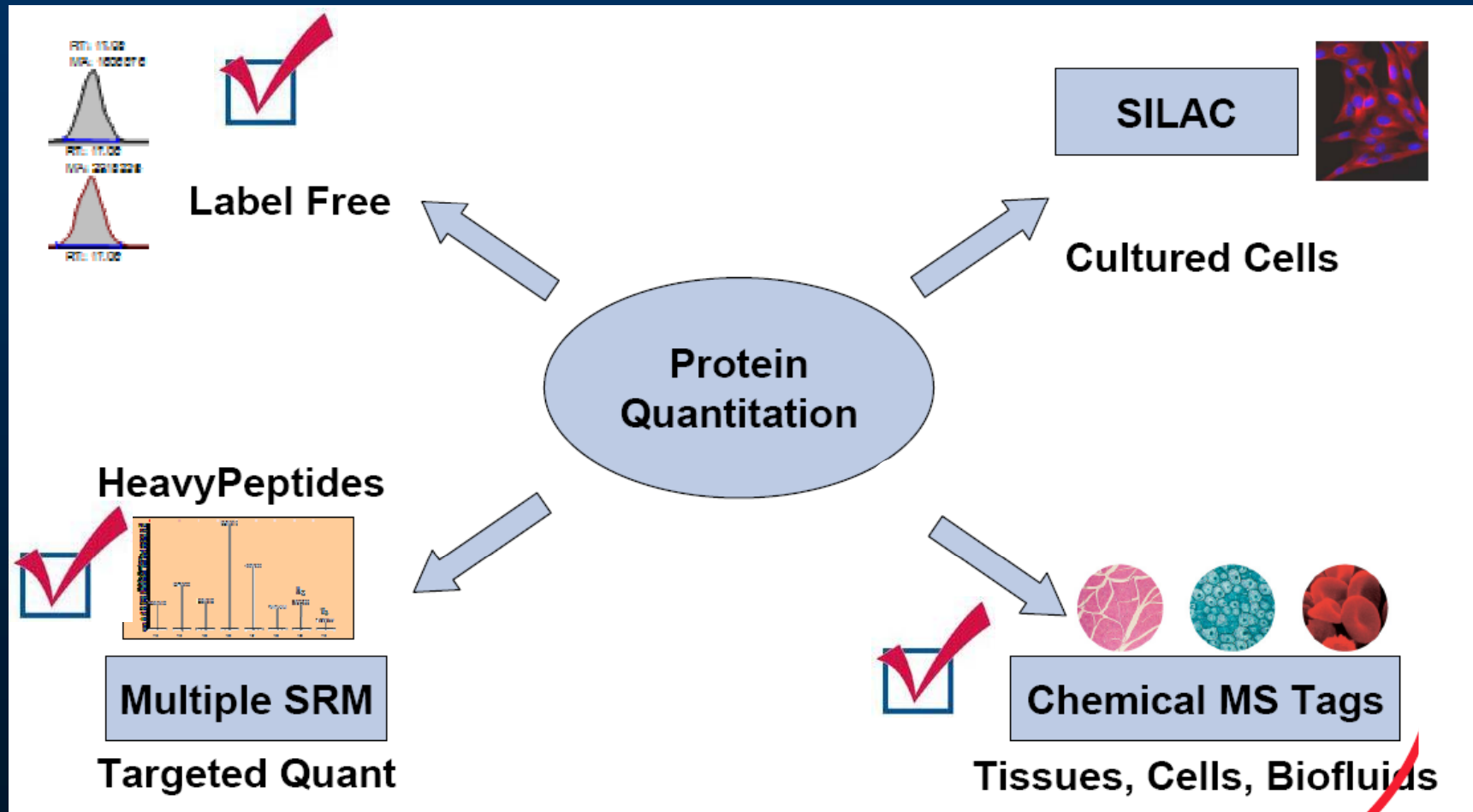
IPI00019591 (100%, 140,943.5 Da
 IPI:IPI00019591.2 Tax_Id=9606 Gene_Symbol=CFB cDNA FLJ55673, highly similar to Complement factor B
 27 unique peptides, 32 unique spectra, 33 total spectra, 333/1266 amino acids (26% coverage)

MGPLMVLVFLCL	LFLYPGLADS	APSPQPQNVNI	SGGTFETLSHG	WAPGSLLTYS	CPQGLYPSPA	SRLCK	SSGQW	QTPGATRSLS
KAVCKPGHCP	NPGISLGAVR	TGFRFGHGDK	VRYR	SSNLV	LTGSSERECO	GNGVWSGTTEP	ICRQPYSDYDF	PEDVAPALGT
SFSHMLGATN	PTQKTKDHEN	GTGNTYAAL	NSVYLMMNNO	MRLLGMETMA	WQEIRHAILL	LTDGKSNMGG	SPKTAVDHIR	GVDGNMSANAS
EILNINQKRN	DYLDIYAIGV	GKLDVDWREL	NELGSKKDG	RHAFILQDTK	ALHQVFEHML	DVSKLTDTC	LIEKAVISPG	FDVFAKKNQG
DOERTPPWHVT	IKPKSQETCR	GALISDQWVL	TAAHCFRDGN	DHSLWRVNVG	DPKSQWGKEF	ENELLNKQSV	PAHFVALNGS	KLNLINLKMVG
ILEFYGGDDIA	LLKLAQKVVM	STHARPICLP	CTMEANLALR	RPQGSSTGRDH	ENELLNKQSV	PAHFVALNGS	LEGVEIKGGS	FRLLQEGQAL
EWTSCAEVVS	QEKTMFPNLT	DVREVVTDQF	LCSGTOQEDS	PCKGVTITPW	SLARPGGSCS	PYYNVSDIIS	FHCYDGYTLR	GSWSGTEPSC
EYVLPSSGFYP	YRVTTRTCRS	TSSWSTLKTQ	DQKTVRKAEC	RAIHCRRPHD	FENGEYWFRS	GSQRRTCEG	DSIGASNFTG	AKKCLVNLIE
GSANRTCQVN	GRWSGQTAIC	DNSAGYCSNP	GIPIGTRKVG	SOYR	LEDSVT	YHCSRGLTLR	MNIYLVLDGS	LQAVYSMMSW
QDSFMYDTPQ	EVAEAFLLSSL	TETIEGVDAE	DGHGPGEQQK	RKIVLVDPSS	LKSGTNTKKA	GVGPLVNVN	YFVLTAACHF	PDDVPPEGWN
KVASYGVKPR	YGLVTTYATYP	KIIVWVSEAD	SSNADVVTKQ	LNEINYEDHK	REDELDVYVF	ESCMGAVVSE	EQHVFKVKDM	TVDDEKHSIK
RTRRHVILMT	DGLHNMGGDP	ITVIDEIRDL	LYIGKDRKNP	ISVIRPSKGH	GQTIRPICLP	CTEGTTRALR	LPPTTTCCQQQ	YADPNTCRGD
ENLEDVIFYQM	IDESQSLSLC	GMVWEHRKGT	DYHKQPWQAK	LIKLNKLY	VKDISEVVTP	RFLCTGGVSP	EDLGLFL	
VSVGGKEKRL	EIEVVLFHFN	YNINGKKEAG	IPEFYDYDVA	DAQYAPGYDK				
KEELLPAQDI	KALFVSEEEK	KLTRKEVYIK	NGDKKGCSCER	DAQYAPGYDK				
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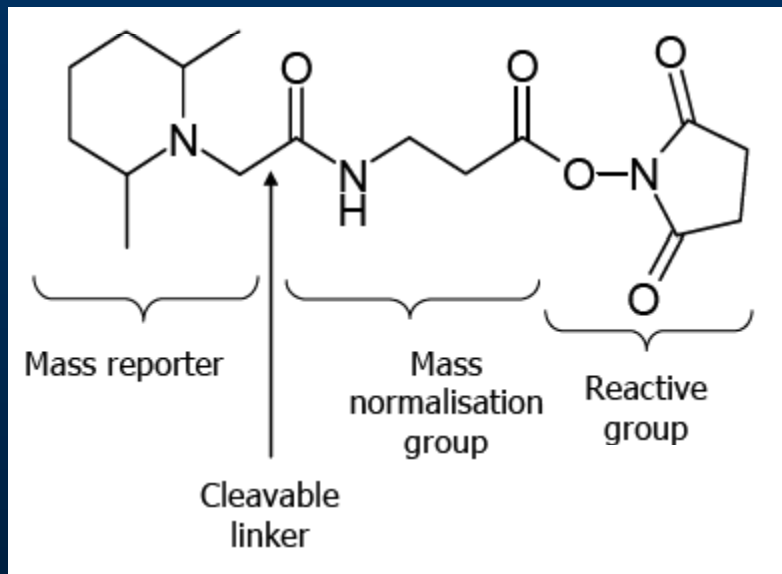


Quantitative Proteomics Continued

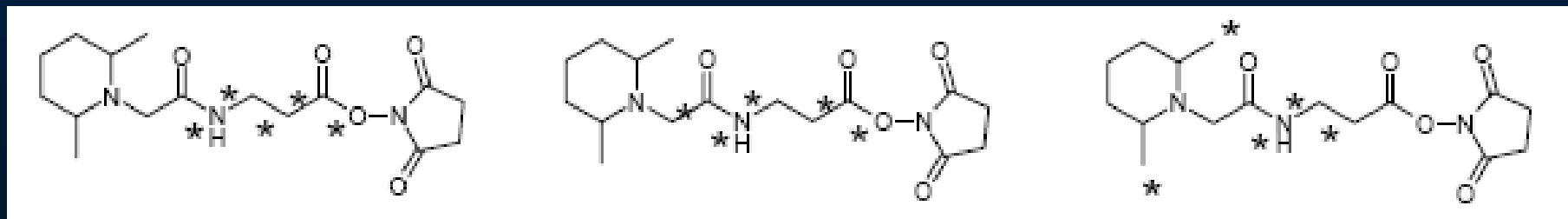




Quantitative Proteomics Ex. 2



Isobaric labels



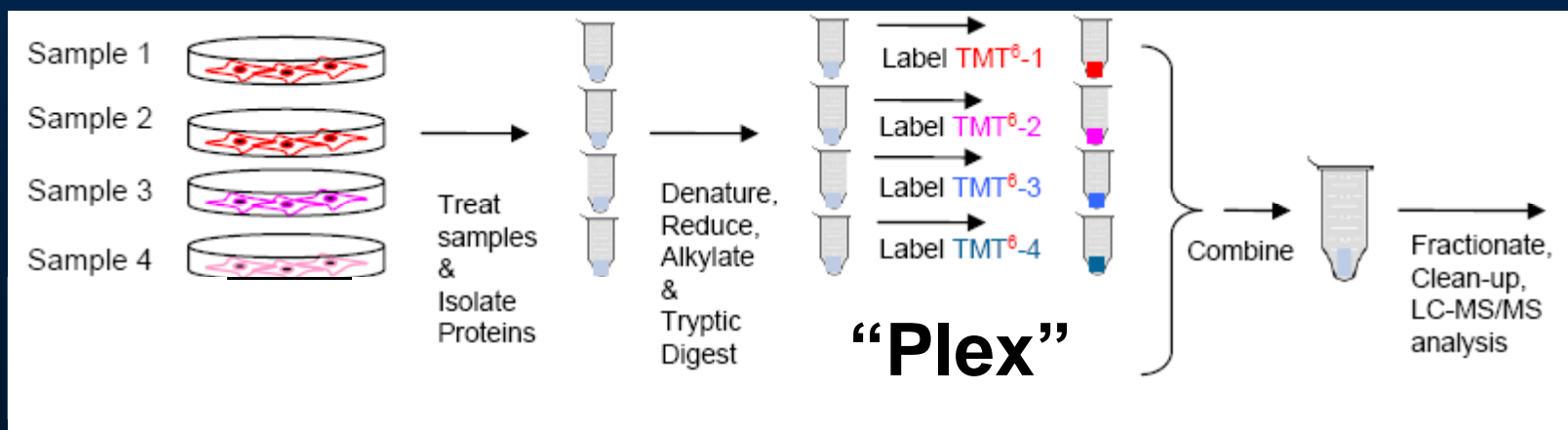


Quantitative Proteomics Ex. 2

Dr. Wen-Jun Shen, Kraemer Lab

http://med.stanford.edu/profiles/Fredric_Kraemer/

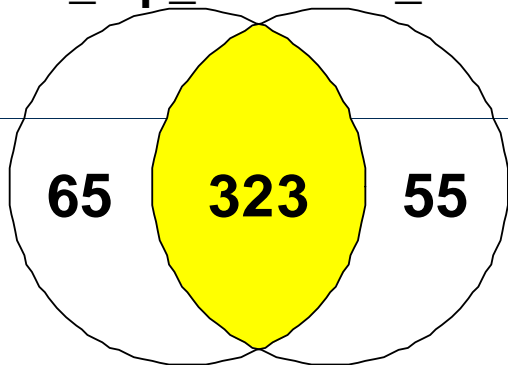
Understanding protein expression in relation to cellular response to fatty acid/HDL treatments





Quantitative Proteomics Ex. 2

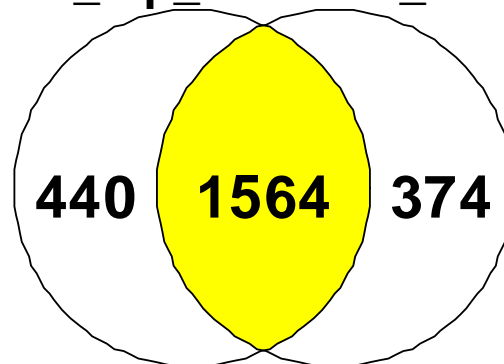
TMT_4Plex_rep_1 TMT_4Plex_rep_2



Proteins

2 peptides, >95% C.I.

TMT_4Plex_rep_1 TMT_4Plex_rep_2



Peptides



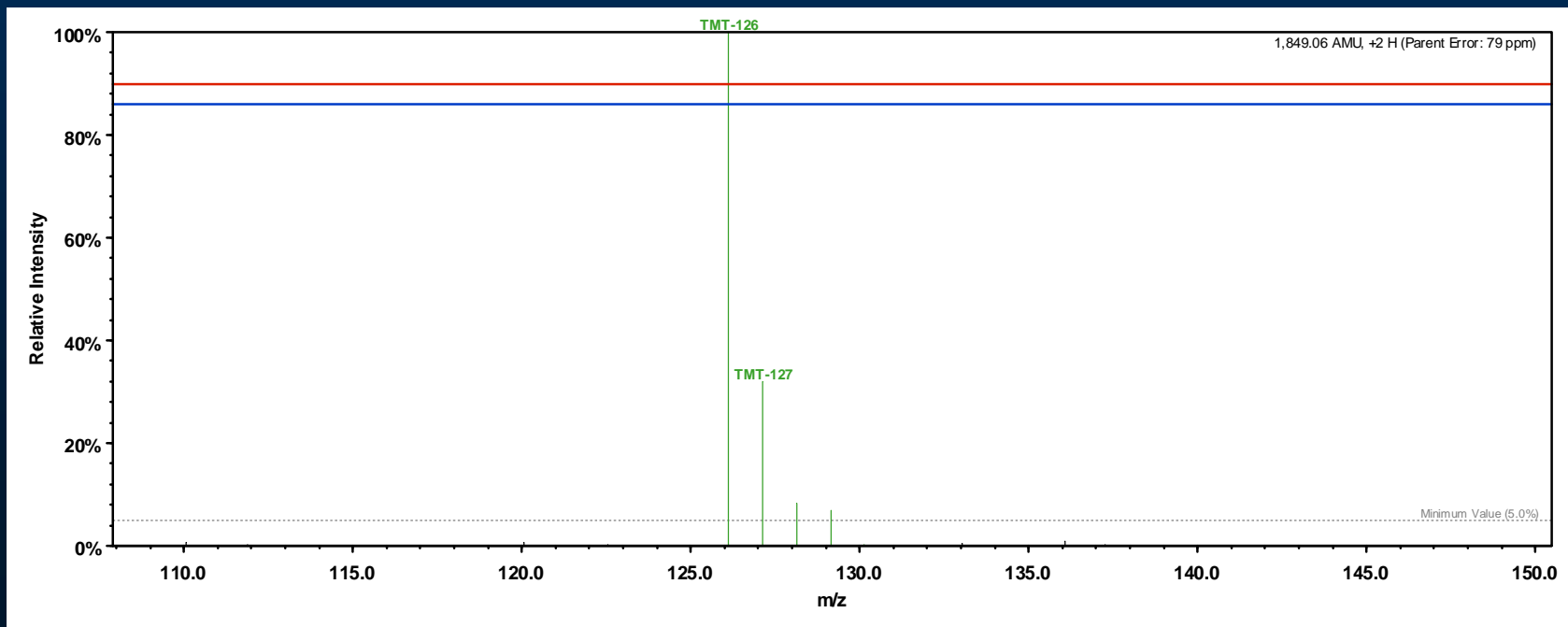
Quantitative Proteomics Ex. 2

Run in duplicate, 2 hour LC MSMS gradients

HCD for reporter ion detection

443 proteins identified, >99% Protein, >95% (2)Peptide

>70% of protein ID'd quantification values reported





Quantitative Proteomics Ex. 2

Display Options: Log₂ Fold Change Min FC: 0 (All Data) Req Mods: No Filter Search:

Relative Abundance Legend:

- Log₂ Fold Change: 2
- Log₂ Fold Change: 1
- Log₂ Fold Change: 0
- Log₂ Fold Change: -1
- Log₂ Fold Change: -2

Quant View:

Identified Proteins (469)

#	Visible?	Starred?	Accession Number	Molecular Weight	Protein Grouping Ambiguity	Reference	State_2	State_3	State_4
1	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100421874.4 Tax_Id=10116 Gene_Symbol=Vdac1 Voltage-dependent anion-selective channel protein 1	31 kDa	Ref	3.1	3.0	-0.1	
2	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100562292.2 Tax_Id=10116 Gene_Symbol=Tapbp 47 kDa protein	47 kDa	Ref	2.7	1.6	0.5	
3	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100190557.2 Tax_Id=10116 Gene_Symbol=Phb2 Prohibitin-2	33 kDa	Ref	2.5	2.4	0.4	
4	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100886470.2 Tax_Id=10116 Gene_Symbol=Hsd17b10 3-hydroxyacyl-CoA dehydrogenase type-2	28 kDa	Ref	2.4	0.9	2.1	
5	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100364616.2 Tax_Id=10116 Gene_Symbol=RGD1309676 Uncharacterized protein C10orf58 homolog	26 kDa	Ref	2.3	0.3	0.1	
6	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100480820.4 Tax_Id=10116 Gene_Symbol=Pgrmc1 Membrane-associated progesterone receptor component 1	22 kDa	Ref	2.2	0.8	-0.6	
7	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100210431.1 Tax_Id=10116 Gene_Symbol=Dhcr7 7-dehydrocholesterol reductase	54 kDa	Ref	2.1	1.2	-1.0	
8	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100209291.1 Tax_Id=10116 Gene_Symbol=Esyt1 Extended synaptotagmin-1	121 kDa	Ref	2.0	1.7	1.6	
9	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100199636.1 Tax_Id=10116 Gene_Symbol=Canx Calnexin	67 kDa	Ref	2.0	1.9	0.0	
10	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100231927.1 Tax_Id=10116 Gene_Symbol=Slc25a4 ADP/ATP translocase 1	33 kDa	Ref	1.9	3.5	0.3	
11	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100231013.2 Tax_Id=10116 Gene_Symbol=Cyb5a Isoform Short of Cytochrome b5	11 kDa	Ref	1.8	0.3	0.8	
12	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100231963.5 Tax_Id=10116 Gene_Symbol=Ddt D-dopachrome decarboxylase	13 kDa	Ref	1.8	0.9	1.4	
13	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100362470.3 Tax_Id=10116 Gene_Symbol=Hsd3b1 3 beta-hydroxysteroid dehydrogenase/Delta 5-->4-isomerase type 1	42 kDa	Ref	1.8	1.8	-0.6	
14	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100214520.1 Tax_Id=10116 Gene_Symbol=Cyp1b1 Cytochrome P450 1B1	61 kDa	Ref	1.8	-0.2	-1.0	
15	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100197344.1 Tax_Id=10116 Gene_Symbol=Mgl1 Monoglyceride lipase	33 kDa	Ref	1.7	0.1	1.4	
16	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100211756.1 Tax_Id=10116 Gene_Symbol=Phb Prohibitin	30 kDa	Ref	1.7	1.7	0.2	
17	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100561052.3 Tax_Id=10116 Gene_Symbol=Hist2h4;RGD1565073;LOC684621;LOC684887;LOC680097;Hist1h4m;Hist1h4b;LOC682518;RGD15...	11 kDa	Ref	1.7	2.8	1.5	
18	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100187747.1 Tax_Id=10116 Gene_Symbol=Rap1a Ras-related protein Rap-1A	21 kDa	Ref	1.7	1.2	0.5	
19	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100198327.2 Tax_Id=10116 Gene_Symbol=Vdac2 Voltage-dependent anion-selective channel protein 2	32 kDa	Ref	1.6	1.7	-0.3	
20	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100231200.5 Tax_Id=10116 Gene_Symbol=Por NADPH-cytochrome P450 reductase	77 kDa	Ref	1.6	0.4	-0.7	
21	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100213581.1 Tax_Id=10116 Gene_Symbol=Cyp21a1 Steroid 21-hydroxylase	56 kDa	Ref	1.6	-0.4	-0.1	
22	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100230859.5 Tax_Id=10116 Gene_Symbol=Akr1a1 Alcohol dehydrogenase [NADP+]	37 kDa	Ref	1.6	0.1	1.9	
23	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100195123.1 Tax_Id=10116 Gene_Symbol=Atp5o ATP synthase subunit O, mitochondrial	23 kDa	Ref	1.6	1.9	0.3	
24	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100365035.2 Tax_Id=10116 Gene_Symbol=Adfp Adipose differentiation related protein	46 kDa	Ref	1.5	2.4	1.4	
25	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100371036.1 Tax_Id=10116 Gene_Symbol=Fis1 Mitochondrial fission 1 protein	17 kDa	Ref	1.5	0.5	0.0	
26	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100366416.2 Tax_Id=10116 Gene_Symbol=Cyc1 cytochrome c-1	35 kDa	Ref	1.5	1.4	1.8	
27	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100371946.3 Tax_Id=10116 Gene_Symbol=LOC681252 similar to Myristoylated alanine-rich C-kinase substrate	30 kDa	Ref	1.5	4.4	1.6	
28	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100188804.1 Tax_Id=10116 Gene_Symbol=Rplp2 60S acidic ribosomal protein P2	12 kDa	Ref	1.5	1.0	1.1	
29	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100215574.5 Tax_Id=10116 Gene_Symbol=Dci 3,2-trans-enoyl-CoA isomerase, mitochondrial	32 kDa	Ref	1.4	0.0	0.2	
30	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100210635.2 Tax_Id=10116 Gene_Symbol=Nsf Vesicle-fusing ATPase	83 kDa	Ref	1.4	-0.1	-1.4	
31	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100200659.1 Tax_Id=10116 Gene_Symbol=SdhA Succinate dehydrogenase [ubiquinone] flavoprotein subunit, mitochondrial	72 kDa	Ref	1.4	1.3	-0.2	
32	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100325713.3 Tax_Id=10116 Gene_Symbol=Cyp11a1 Cholesterol side-chain cleavage enzyme, mitochondrial	61 kDa	Ref	1.4	0.7	-1.4	
33	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100190348.1 Tax_Id=10116 Gene_Symbol=LOC680312 histone H2B	14 kDa	Ref	1.4	3.1	0.8	
34	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100213659.3 Tax_Id=10116 Gene_Symbol=Decr1 2,4-dienoyl-CoA reductase, mitochondrial	36 kDa	Ref	1.3	0.5	-0.2	
35	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	IPI:IP100365944.5 Tax_Id=10116 Gene_Symbol=Myl6l Myosin light polypeptide 6	17 kDa	Ref	1.3	2.8	2.3	



Quantitative Proteomics Ex. 2

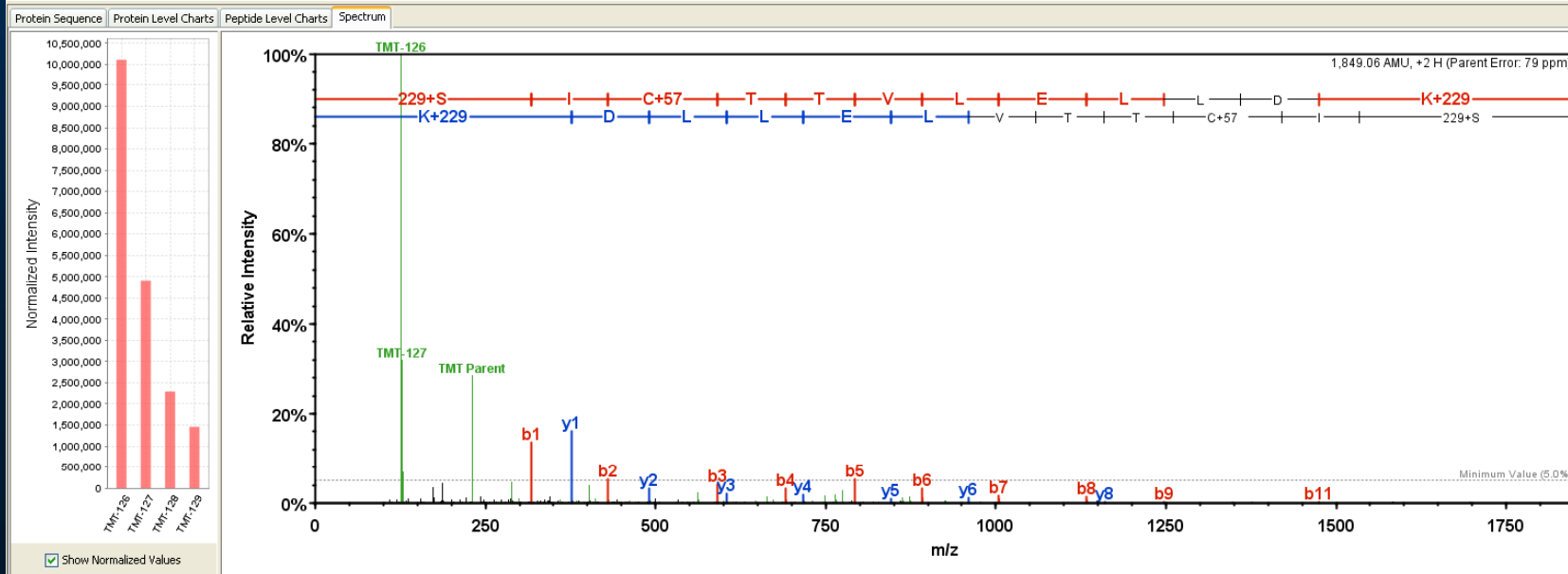
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Peptides:

Good?	Assigned	Unique	Sequence	Prob	Modifications	XCorr	deltaCn	126	Quant 2	Quant 3	Quant 4	+1H	+2H	+3H	+4H	Other Proteins
✓	✓	✓	I TTVLELLD	95%	Carbamidomethyl (+57), Pyridoxal-phos (+229), TMT-6plex (+229)	3.60	0.456	Ref	-0.81	-1.8	-2.1	0	1	1	0	
✓	✓	✓	SICTTVLELLDKYLIANATNPESK	95%	Carbamidomethyl (+57), Pyridoxal-phos (+229), Deamidation (+1), ...	3.43	0.210	Ref	No Values	No Values	No Values	0	0	0	1	
✓	✓	✓	TAFDEAIEALDTLNEDSYK	95%	Pyridoxal-phos (+229), TMT-6plex (+229)	4.37	0.424	Ref	-1.1	Value Missing	-2.5	0	0	1	0	
✓	✓	✓	YDDMATCMK	95%	Carbamidomethyl (+57), Oxidation (+16), Pyridoxal-phos (+229), T...	1.99	0.413	Ref	-0.22	-1.6	-0.68	0	1	0	0	
✓	✓	✓	YLAEVACGDDR	95%	Carbamidomethyl (+57), TMT-6plex (+229)	2.92	0.326	Ref	-0.39	-0.99	-0.71	0	1	0	0	
✓	✓	✓	YLAEVACGDDRK	95%	Carbamidomethyl (+57), Pyridoxal-phos (+229), TMT-6plex (+229)	2.54	0.277	Ref	-0.036	-0.73	-0.028	0	0	1	0	
✓	✓	✓	YLIANATNPESK	95%	Pyridoxal-phos (+229), TMT-6plex (+229)	3.49	0.375	Ref	-1.2	-2.0	-1.5	0	1	0	0	

Spectra:

Good?	Assigned	Unique	Sequence	Prob	Modifications	XCorr	deltaCn	126	Quant 2	Quant 3	Quant 4	Observed	Actual Mass	Charge	Delta...	Delta...	Spectrum ID
✓	✓	✓	I TTVLELLD	95%	Carbamidomethyl...	3.60	0.456	Ref	-1.0	-2.1	-2.8	925.54	1,849.06	2	0.15	79	100606_W3Shen...
✓	✓	✓	SICTTVLELLDK	95%	Carbamidomethyl...	3.06	0.230	Ref	-0.57	-1.5	-1.3	617.36	1,849.07	3	0.15	82	100606_W3Shen...

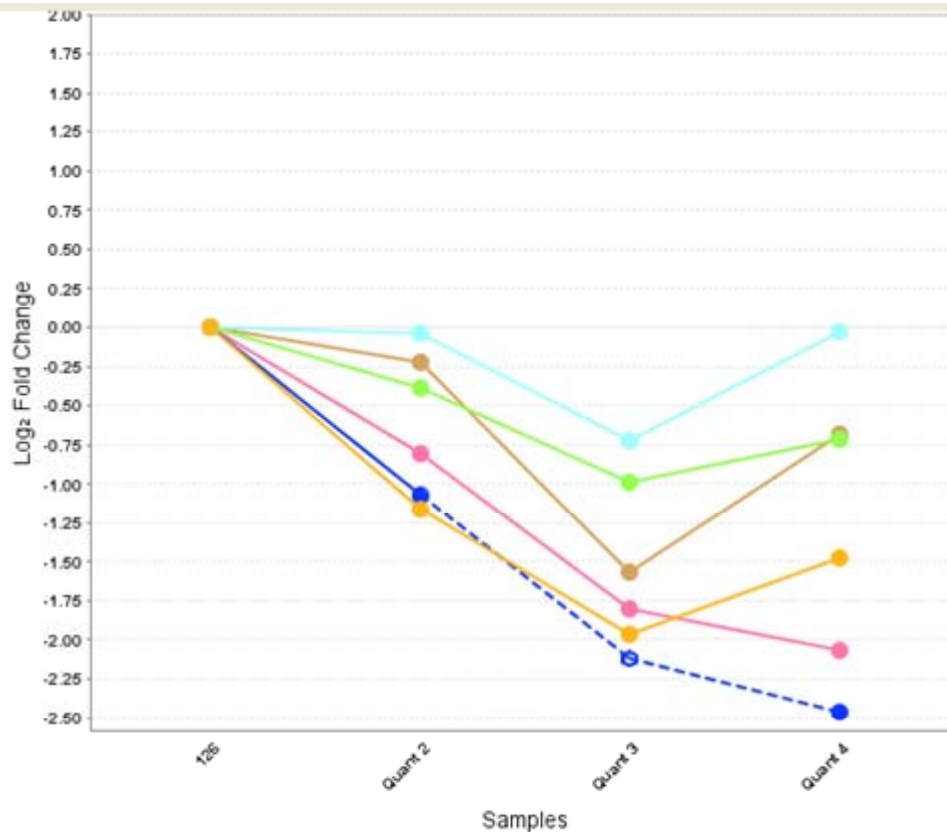




Quantitative Proteomics Ex. 2

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DYREKVESEL	R SICTTVLEL	LDKYLIAHAT	NPESHV FYLK	MKGDYFR YLA	EVACGDDRKQ	TIENSQGAYQ	EAFDISKKEM
QPTHPIRLGL	ALNFSVFYYE	TLNNPELACT	LAKTAFDEAI	AELDTLNEDS	YK DSTLIMQL	LRDNLTVFSC	VAL





Acknowledgements

SUMS

- Allis Chien
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- Pavel Aronov
- Maurizio Splendore
- Ludmila Alexandrova

- Elias Lab
- Snyder Lab

- Vincent and Stella Coates Foundation