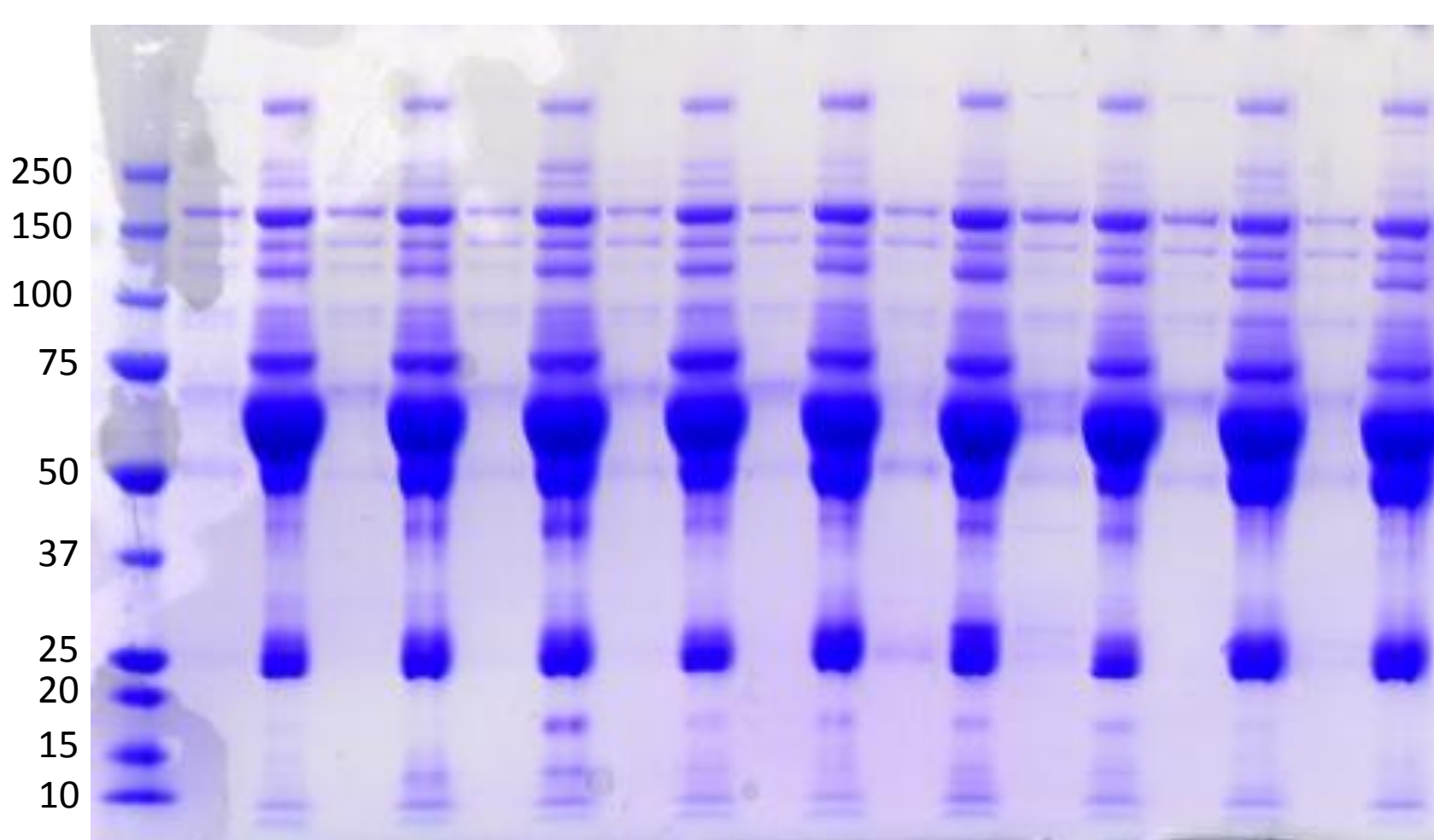
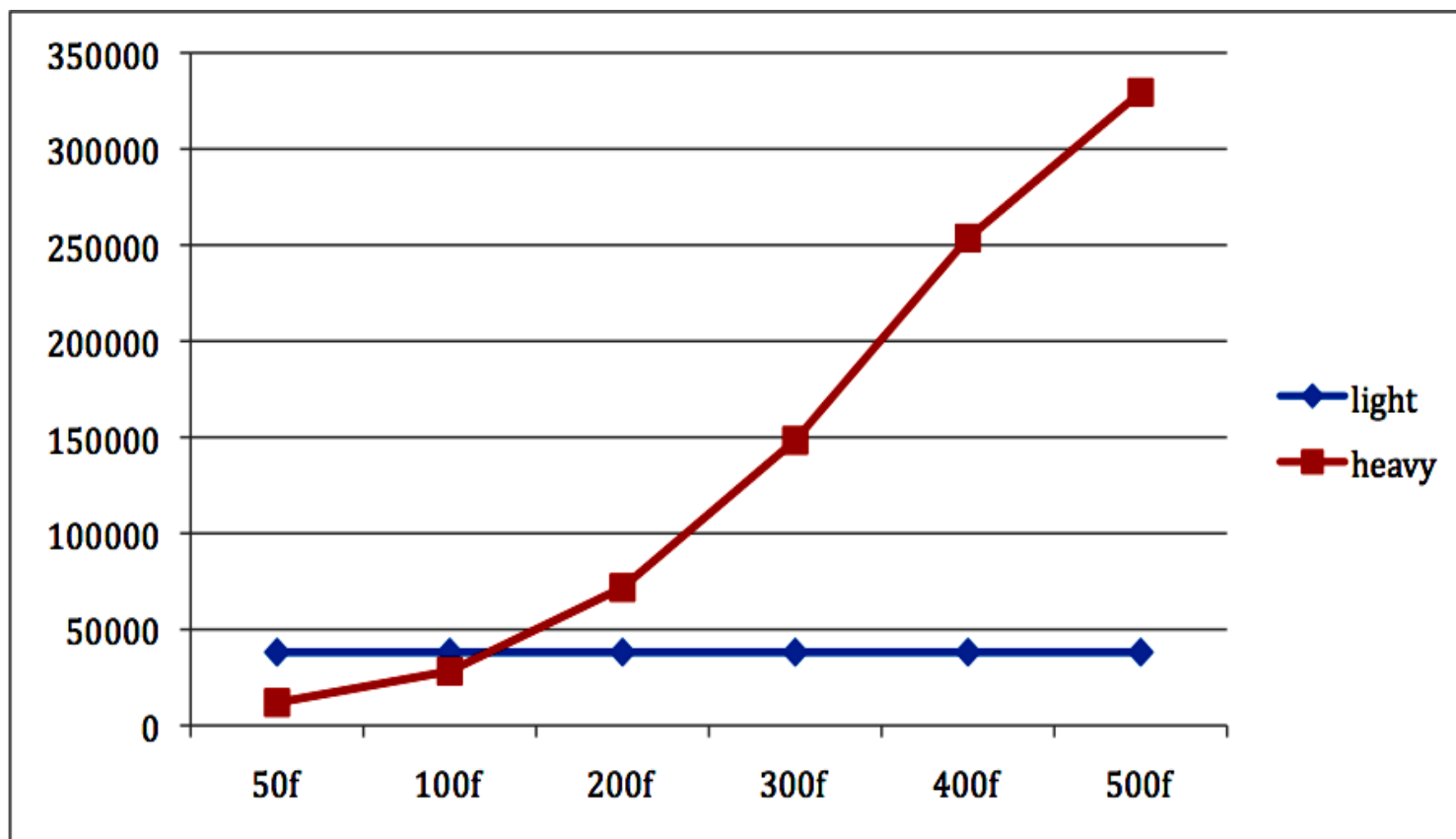


M D1 S1 D2 S2 D3 S3 D4 S4 D5 S5 D6 S6 D7 S7 D8 S8 D9 S9

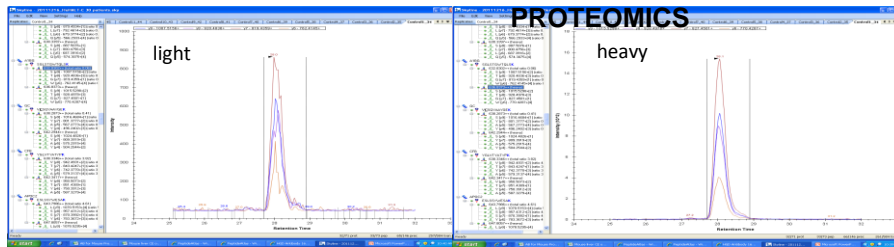


Supplemental Figure 1. A typical titration curve (peptide VANYVDWINDR) shows the correct amount of heavy peptide needed to be spiked-in to human blood samples (100 femtomole in this case). X-axis: peptide amount in femtomole, y-axis: total peak AUC (area under transition curve). Synthetic heavy-labeled peptide is in red and endogenous light peptide is in blue.

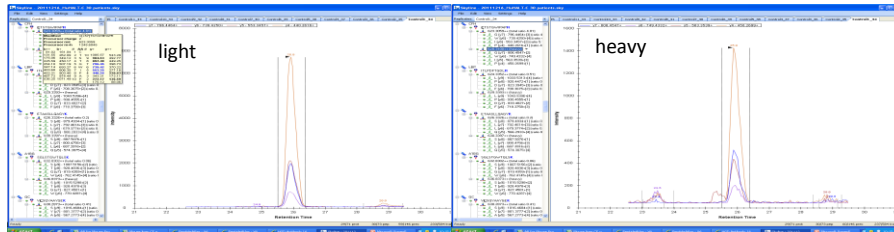


Supplemental Figure 2. A typical titration curve (peptide VANYVDWINDR) shows the correct amount of heavy peptide needed to be spiked-in to human blood samples (100 femtomole in this case). X-axis: peptide amount in femtomole, y-axis: total AUC (area under transition curve). Synthetic heavy-labeled peptide is in red and endogenous light peptide is in blue.

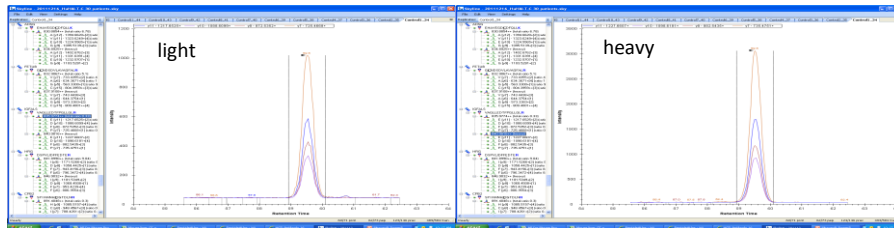
PROTEOMICS



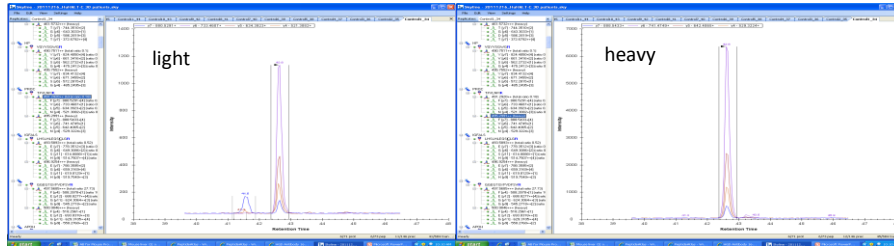
CFH/CTSTGWIPAPR



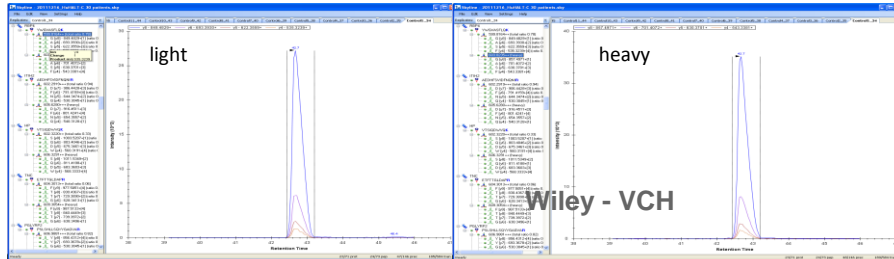
IGFALS/VAGLLEDFPGLLGLR



PROC/TFVLNFIK

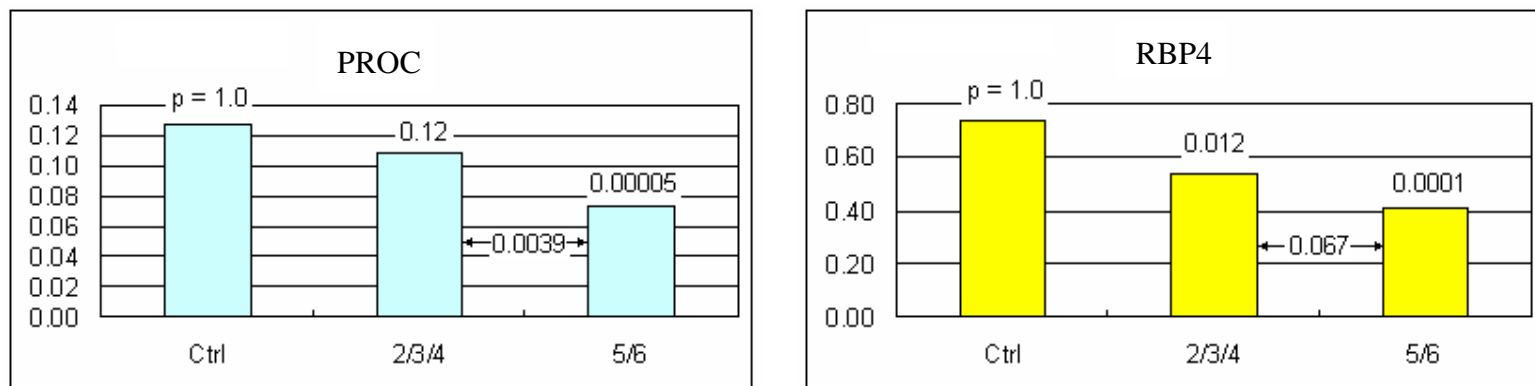


RBP4/YWGVASFLQK



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Supplementary Figure 3. Typical chromatograms of 5 targeted peptides.



Supplemental Figure 4. The mean of serum protein levels among control, Ishak 2-4 and Ishak 5-6 patient samples. There is a significant decrease of PROC levels in Ishak 5-6 patients as compared to Ishak 2-4 patients ( $P=0.004$ ). PROC levels in Ishak 5-6 patients are also significantly lower than controls ( $P=0.00005$ ). Both Ishak 2-4 and patients 5-6 patients' RBP4 levels are significantly lower than controls ( $P=0.01$  and  $P=0.0001$  with or without outliers, respectively). Comparison of RBP4 levels in Ishak 5-6 patients to Ishak 2-4 patients shows a  $P$  value of 0.07 with outliers and  $P$  value of 0.02 without outliers.

**Supp. Table 1.** Patient information on age, gender, platelet counts, AST, ALT, and alkaline phosphatase levels in blood and Ishak scores when enrolled in the HALT-C trial.

Serum sample ID	AGE	Gender	Platelets (x1000/mm3)	AST (U/L)	ALT (U/L)	Alkaline phosphatase (U/L)	Fibrosis-Ishak score
DA 000047	48	M	232	89	121	78	2
DI 000933	27	M	418	68	133	111	2
DJ 001681	58	M	208	31	27	100	2
DH 001854	48	F	222	44	67	86	2
DH 001868	44	F	270	46	56	41	2
DB 001537	47	M	132	46	73	80	3
DD 000089	54	M	188	55	46	76	3
DD 000830	53	M	243	69	113	85	3
DD 001302	49	M	218	43	68	61	3
DD 001616	45	M	216	35	60	71	3
DJ 000111	51	M	208	64	75	97	3
DI 001315	46	F	274	72	117	75	3
DJ 000004	47	F	105	113	91	69	3
DA 000043	51	M	280	58	82	46	4
DD 000019	48	M	194	62	68	80	4
DD 001632	49	M	135	89	141	82	4
DF 001110	50	M	98	46	42	117	4
DJ 001239	52	M	169	50	71	108	4
DA 001416	56	F	143	98	121	61	4
DD 000008	56	F	259	34	46	117	4
DA 000024	53	M	132	157	261	68	5
DA 000739	49	M	391	35	39	67	5
DF 000456	51	M	114	128	160	83	5
DG 001250	47	M	71	49	53	105	5
DH 001717	59	M	106	41	68	133	5
DI 000921	50	M	72	91	112	81	5
DJ 000126	46	M	93	136	107	89	5
DJ 001652	42	M	68	58	62	88	5
DB 001019	56	F	225	33	25	80	5
DH 001870	64	F	155	28	43	94	5
DD 001328	67	M	140	28	42	128	6
DD 001630	57	M	99	34	39	75	6
DF 000833	60	M	98	100	159	117	6
DF 001246	46	M	88	71	51	150	6
DI 000001	56	M	93	45	60	76	6
DJ 001705	56	M	106	89	78	142	6
DJ 002724	66	M	167	231	137	301	6
DF 000839	50	F	176	244	291	127	6
DI 000937	50	F	83	213	212	307	6
DJ 000131	46	F	95	95	77	174	6

**Supp Table 2.** 87 proteins (shown with NCBI office symbol) previously detected by tandem mass spectrometry and tested by SRM with light peptides

A1BG**	AGXT*	AHSG**	ALDOB	ALT1#	ALT2#	AMBP**	ANG	APCS**	APOA5
APOC1	APOC2**	APOC4*	APOF	APOH**	ASGR2	ASL	AST1**#	AST2#	BHMT
C8A	C8B*	C8G**	C9**	CES1*	CFB**	CFH**	CFHR2**	CFHR4	CHI3L1#
CP**	CPB2**	CPS1*	DPYS	F2**#	F9**	F10*	F12**	FAH**	FBP1*
FCN2	FETUB**	FGA*	FGB*	FGG*	GC**	GNMT	GOLM1#	GRN#	HABP2**
HAMP	HAO1	HGD	HGFAC**	HMGCS2*	HPD	HPR*	HPX	HRG**	HSPA9#
IDH1	IGFALS**	ITIH1**	ITIH2**	ITIH3**	ITIH4*	LBP**	LCAT**	LPA**	MASP2
MAT1A*	MBL2	MDH	MMP9#	PEX1	PGLYRP2**	PKLR*	PROC**	RBP4**	SAA4
SERPINF2**	SPP2	TAT	TIMPI*	TNC**#	UPB1	VTN**			

\* endogenous peptide detected in pooled control plasma

\*\* endogenous peptide detected in control sera

# proteins used in clinic practice or previously reported as liver biomarker candidates

**Supplemental Table 3,** SRM monitoring conditions of 38 proteins that detectable in control sera.

Many proteins are represented by two or more peptides. **K:**  $^{13}\text{C}_6^{15}\text{N}_2$  isotopic-labeled lysine, **R:**  $^{13}\text{C}_6^{15}\text{N}_4$  isotopic-labeled arginine

NCBI official Symbol	Description	Peptide Sequence	MH+ (mono)	Z (Q1)	Transitions				
					Q1	Q3			
A1BG	Alpha-1-B glycoprotein	HQFLLTGDTQGR	1372.7	2	686.9	734.3	847.4	960.5	1107.6
		HQFLLTGDTQGR	1382.7		691.9	744.3	857.4	970.5	1117.6
A1BG	Alpha-1-B glycoprotein	HQFLLTGDTQGR	1372.7	3	458.2	367.7	576.3	633.3	734.3
		HQFLLTGDTQGR	1382.7		461.6	372.7	586.3	643.3	744.3
A1BG	Alpha-1-B glycoprotein	SGLSTGWTQLSK	1264.7	2	632.8	762.4	819.4	920.5	1007.5
		SGLSTGWTQLSK	1272.7		636.8	770.4	827.4	928.5	1015.
AHSG	Alpha-2-HS-glycoprotein	EHAVEGDCDFLLK	1660.7	2	830.9	1095.5	1224.6	1323.6	1394.7
		EHAVEGDCDFLLK	1668.7		834.9	1103.5	1232.6	1331.6	1402.7
AHSG	Alpha-2-HS-glycoprotein	HTLNQIDEVK	1196.6	2	598.8	731.4	845.4	958.5	1059.6
		HTLNQIDEVK	1204.6		602.8	739.4	853.4	966.5	1067.6
AMBP	Alpha-1-microglobulin/bikunin	AFIQLWAFDAVK	1408.8	2	704.9	579.3	650.4	836.4	949.5
		AFIQLWAFDAVK	1416.8		708.9	587.3	658.4	844.4	957.5
AMBP	Alpha-1-microglobulin/bikunin	GVCEETSGAYEK	1329.5	2	665.3	755.4	884.4	1013.4	1173.5
		GVCEETSGAYEK	1337.5		669.3	763.4	892.4	1021.4	1181.5
APCS	Amyloid P component, serum	AYSLFSYNTQGR	1406.7	2	703.84	575.3	738.4	825.4	972.5
		AYSLFSYNTQGR	1416.7		708.84	585.3	748.4	835.4	982.5
APCS	Amyloid P component, serum	VGEYSLYIGR	1156.6	2	578.8	508.3	621.4	708.4	871.5
		VGEYSLYIGR	1166.6		583.8	518.3	631.4	718.4	881.5
APOC2	Apolipoprotein C-II	ESLSSYWESAK	1286.6	2	643.8	783.4	870.4	957.4	1070.5
		ESLSSYWESAK	1294.6		647.8	791.4	878.4	965.4	1078.5
APOC2	Apolipoprotein C-II	TAAQNLYEK	1037.5	2	519.3	552.3	666.4	794.4	865.4
		TAAQNLYEK	1045.5		523.3	560.3	674.4	802.4	873.4
APOH	Apolipoprotein H	ATVVYQGER	1037.5	2	511.8	489.2	652.3	751.4	850.4
		ATVVYQGER	1045.5		516.8	499.2	662.3	761.4	860.4
APOH	Apolipoprotein H	EHSSLAFWK	1104.6	2	552.8	551.3	664.4	751.4	838.5
		EHSSLAFWK	1112.6		556.8	559.3	672.4	759.4	846.5
AST1	glutamic-oxaloacetic transaminase 1	LALGDDSPALK	1099.6	2	550.3	745.4	802.4	915.5	986.5
		LALGDDSPALK	1107.6		554.3	753.4	810.4	923.5	994.5
AST1	glutamic-oxaloacetic transaminase 1	TDDCHPWVLPVVK	1565.8	3	522.6	617.8	654.5	675.4	732.9
		TDDCHPWVLPVVK	1573.8		525.3	621.8	662.5	679.4	736.9

C8G	Complement component 8, gamma polypeptide	SLPVSDSVLSGFQQR SLPVSDSVLSGFQQR	1620.8 1630.8	3	541.0 544.3	579.3 589.3	636.3 646.3	662. 667.3	710.9 715.9
C9	Complement component 9	AIEDYINEFSVR AIEDYINEFSVR	1455.7 1465.7	2	728.4 733.4	637.3 647.3	751.4 761.4	864.5 874.5	1271.6 1281.6
CFB	Complement factor B	DFHINLFQVLPWLK DFHINLFQVLPWLK	1770.0 1778.0	3	590.7 593.3	543.3 551.3	656.4 664.4	755.5 763.5	883.5 891.5
CFB	Complement factor B	YGLVTYATYPK YGLVTYATYPK	1275.7 1283.7	2	638.3 642.3	579.3 587.3	742.9 750.9	843.4 851.4	942.5 950.5
CFH	Complement factor H	CTSTGWIPAPR CTSTGWIPAPR	1245.6 1255.6	2	623.3 628.3	440.3 450.3	553.4 563.4	739.4 749.4	796.5 806.5
CFHR2	Complement factor H-related 2	ITCAEEGWSPTPK ITCAEEGWSPTPK	1475.7 1483.7	2	738.4 742.4	529.3 537.3	772.4 780.4	901.4 909.4	1030.5 1038.5
CFHR2	Complement factor H-related 2	TGDIVEFVCK TGDIVEFVCK	1167.6 1175.6	2	584.3 588.3	553.3 561.3	682.3 690.3	781.4 789.4	894.5 902.5
CP	Ceruloplasmin (ferroxidase)	ALYLQYTDETFR ALYLQYTDETFR	1519.7 1529.7	2	760.4 765.4	667.3 677.3	931.4 941.4	1059.5 1069.5	1172.6 1182.6
CP	Ceruloplasmin (ferroxidase)	IYHSHIDAPK IYHSHIDAPK	1180.6 1188.6	3	394.2 396.9	430.2 438.2	452.7 456.7	534.3 538.3	767.4 775.4
CPB2	Carboxypeptidase B2 (plasma)	SFYANNHCIGTDLNR SFYANNHCIGTDLNR	1781.8 1791.8	2	891.4 896.4	675.3 685.3	788.4 798.4	948.5 958.5	1085.5 1095.5
CPB2	Carboxypeptidase B2 (plasma)	SFYANNHCIGTDLNR SFYANNHCIGTDLNR	1781.8 1791.8	3	594.6 597.9	657. 662.3	692.8 697.8	774.4 779.4	948.5 958.5
F12	Coagulation factor XII	CFEPQLLR CFEPQLLR	1062.5 1072.5	2	531.8 536.8	401.3 411.3	626.4 636.4	755.4 765.4	902.5 912.5
F2	Coagulation factor II (thrombin)	ELLESYIDGR ELLESYIDGR	1194.6 1204.6	2	597.8 602.8	460.3 470.3	623.3 633.3	710.4 720.4	839.4 849.4
F2	Coagulation factor II (thrombin)	TATSEYQTFNPR TATSEYQTFNPR	1561.7 1571.7	2	781.4 786.4	533.3 543.3	680.6 690.6	909.5 919.5	1072.5 1082.
F9	Coagulation factor IX	WIVTAAHCVETGVK WIVTAAHCVETGVK	1570.8 1578.8	3	524.3 526.9	536.3 540.3	586.8 590.8	636.3 640.3	692.9 696.9
FAH	Fumarylacetoacetate hydrolase	VFLQNLLSVSQAR VFLQNLLSVSQAR	1474.8 1484.8	2	737.9 742.9	560.3 570.3	647.4 657.4	760.4 770.4	987.6 997.6
FETUB	Fetuin B	IFFESVYGQCK IFFESVYGQCK	1377.63 1385.63	2	689.3 693.3	492.2 500.2	655.3 663.3	841.4 849.4	1117.5 1125.5
GC	Group-specific component (vitamin D binding protein)	HLSLLTTLNLR HLSLLTTLNLR	1254.7 1264.7	3	418.9 422.3	376.2 386.2	489.3 499.3	590.3 600.3	691.4 701.4
GC	Group-specific component (vitamin D binding protein)	VCSQYAAAYGEK VCSQYAAAYGEK	1275.6 1283.6	2	638.3 642.3	496.2 504.2	567.3 575.3	801.4 809.4	1016.5 1024.5
HABP2	Hyaluronan binding protein 2	DEIPHNDIALLK DEIPHNDIALLK	1377.7 1385.7	3	459.9 462.6	444.3 452.3	462.3 466.3	510.8 514.8	567.3 571.3
HABP2	Hyaluronan binding protein 2	GQCLITQSPYYR GQCLITQSPYYR	1582.7 1592.7	2	791.9 796.9	695.4 705.4	782.4 792.4	910.4 920.4	1011.5 1021.5
HGFAC	HGF activator	TTDVTQTFGIEK TTDVTQTFGIEK	1339.7 1347.7	2	670.3 674.3	694.4 702.4	822.4 830.4	923.5 931.5	1137.6 1145.6
HGFAC	HGF activator	VANYVDWINDR VANYVDWINDR	1364.7 1374.7	2	682.8 687.8	703.4 713.4	818.4 828.4	917.5 927.5	1080.5 1090.5
HP	Haptoglobin	VGYSVSGWGR VGYSVSGWGR	980.5 990.5	3	490.8 495.8	475.2 485.2	562.3 572.3	661.3 671.3	824.4 834.4
HP	Haptoglobin	VTSIQDWVQK VTSIQDWVQK	1203.6 1211.6	2	602.3 606.3	560.3 568.3	675.4 683.4	803.4 811.4	1003.5 1011.5
HRG	Histidine-rich glycoprotein	DSPVLIDFFEDTER DSPVLIDFFEDTER	1682.8 1692.8	2	841.9 846.9	796.4 806.4	943.4 953.4	1058.4 1068.4	1171.5 1181.5
HRG	Histidine-rich glycoprotein	GGEGTGYFVDFSVR GGEGTGYFVDFSVR	1490.7 1500.7	2	745.9 750.9	869.6 879.6	1032.5 1042.5	1089.5 1099.5	1190.6 1200.6
IGFALS	Insulin-like growth factor binding protein, acid labile subunit	LHSLHLEGSCLGR LHSLHLEGSCLGR	1478.7 1488.7	3	493.6 496.9	514.8 519.8	614.8 619.8	649.3 659.3	778.4 788.4
IGFALS	Insulin-like growth factor binding protein, acid labile subunit	VAGLLEDTFPGLLGLR VAGLLEDTFPGLLGLR	1671.0 1681.0	2	836.0 841.0	725.5 735.5	872.5 882.5	1088.6 1098.6	1217.7 1227.7

ITIH1	Inter-alpha (globulin) inhibitor H1	AAISGENAGLVR AAISGENAGLVR	1157.6 1167.6	2	579.3 584.	629.4 639.4	758.4 768.4	815.4 825.4	902.5 912.5
ITIH1	Inter-alpha (globulin) inhibitor H1	LDAQASFLPK LDAQASFLPK	1089.6 1097.6	2	545.3 549.3	591.4 599.4	662.4 670.4	790.5 798.5	861.5 869.5
ITIH2	Inter-alpha (globulin) inhibitor H2	TEVNVLPGAK TEVNVLPGAK	1027.6 1035.6	2	514.3 518.3	372.2 380.2	485.3 493.3	698.4 706.4	797.5 805.5
ITIH3	Inter-alpha (globulin) inhibitor H3	DYIFGNFYIER DYIFGNFYIER	1289.6 1299.6	2	645.3 650.3	694.4 704.4	751.4 761.4	898.4 908.4	1011.5 1021.5
LBP	Lipopolysaccharide binding protein	ITLPDFTGDLR ITLPDFTGDLR	1247.7 1257.7	2	624.3 629.3	708.4 718.4	823.4 833.4	920.5 930.5	1033.5 1043.5
LCAT	Lecithin-cholesterol acyltransferase	SSGLVSNAPGVQIR SSGLVSNAPGVQIR	1384.8 1394.8	2	692.9 697.9	740. 750.45	854.5 864.5	941.5 951.5	1040.6 1050.6
LCAT	Lecithin-cholesterol acyltransferase	TYSVEYLDSSK TYSVEYLDSSK	1291.6 1299.6	2	646.3 650.3	712.4 720.4	841.39 849.41	940.5 948.5	1027.5 1035.5
LPA	Lipoprotein, Lp(a)	NPDAVAAPCYTR NPDAVAAPCYTR	1497.7 1507.7	2	749.3 754.3	859.4 869.4	930.41 940.42	1001.5 1011.5	1100.5 1110.5
PGLYRP2	Peptidoglycan recognition protein 2	PSLSHLLSQYYGAGVAR PSLSHLLSQYYGAGVAR	1819.0 1829.0	3	607.0 610.3	473.3 483.3	530.30 540.31	693.4 703.4	856.4 866.4
PGLYRP2	Peptidoglycan recognition protein 2	TDCPGDALFDLLR TDCPGDALFDLLR	1492.7 1502.7	2	746.9 751.	663.4 673.4	776.47 786.47	847.5 857.5	1116.6 1126.6
PON1	Paraoxonase 1	EVQPVELPNCNLVK EVQPVELPNCNLVK	1638.8 1646.8	2	819.9 823.9	844.4 852.4	957.5 965.5	1086.6 1094.6	1282.7 1290.7
PON1	Paraoxonase 1	IFFYDSENPPASEVLR IFFYDSENPPASEVLR	1883.9 1893.9	2	942.5 947.5	603.4 613.4	868.5 878.5	982.5 992.5	1198.6 1208.6
PON1	Paraoxonase 1	IQNILTEEPK IQNILTEEPK	1184.7 1192.7	2	592.8 596.8	603.3 611.3	716.4 724.4	829.5 837.5	943.5 951.5
PROC	Protein C	EVFVHPNYSK EVFVHPNYSK	1219.6 1227.6	2	610.3 614.3	608.3 616.3	745.4 753.4	844.4 852.4	991.5 999.5
PROC	Protein C	TFVLNFIK TFVLNFIK	981.6 989.6	2	491.3 495.3	521.3 529.3	634.4 642.4	733.5 741.5	880.5 888.5
RBP4	Retinol binding protein 4, plasma	YWGVASFLQK YWGVASFLQK	1198.6 1206.6	2	599.8 603.8	535.3 543.3	622.4 630.4	693.4 701.4	849.5 857.5
SERPINF2	Serpin peptidase inhibitor, clade F, member 2	LCQDLGPGAFR LCQDLGPGAFR	1233.6 1243.6	2	617.3 622.3	547.3 557.3	604.3 614.3	717.4 727.4	960.5 970.5
TNC	Tenascin C (hexabrachion)	EEFWLGLDNLNK EEFWLGLDNLNK	1477.7 1485.7	2	739.4 743.4	773.4 781.4	886.5 894.5	1072.6 1080.6	1219.7 1227.7
TNC	Tenascin C (hexabrachion)	ETFTTGLDAPR ETFTTGLDAPR	1207.6 1217.6	2	604.3 609.3	628.3 638.3	729.4 739.4	830.4 840.4	977.5 987.5
VTN	Vitronectin	DVWGIEGPIDAAFTR DVWGIEGPIDAAFTR	1646.8 1656.8	2	823.9 828.9	890.5 900.5	947.5 957.5	1076.5 1086.5	1246.6 1256.6
VTN	Vitronectin	GQYCYELDEK GQYCYELDEK	1304.5 1312.5	2	652.8 656.8	504.3 512.3	633.3 641.3	796.4 804.4	956.4 964.4