

Supporting information

Facile synthesis of magnetic covalent organic frameworks for hydrophilic enrichment of N-glycopeptides

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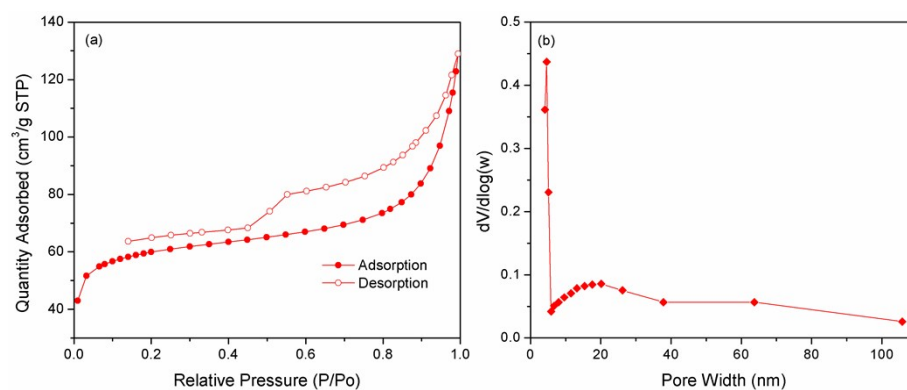


Fig. S1 N₂ adsorption isotherms (77 K) (a) and pore size distribution (b) of Fe₃O₄@TpPa-1.

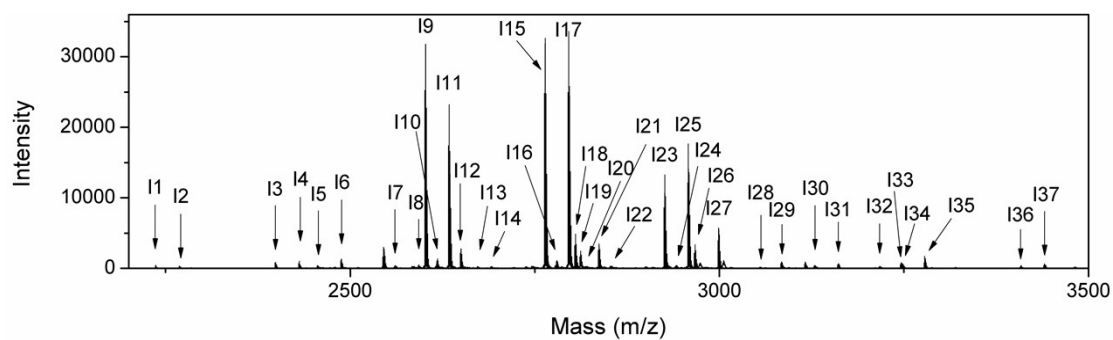


Fig. S2 MALDI-TOF MS spectra of human IgG digests (70 fmol μL^{-1}) after enrichment by Fe₃O₄@TpPa-1.

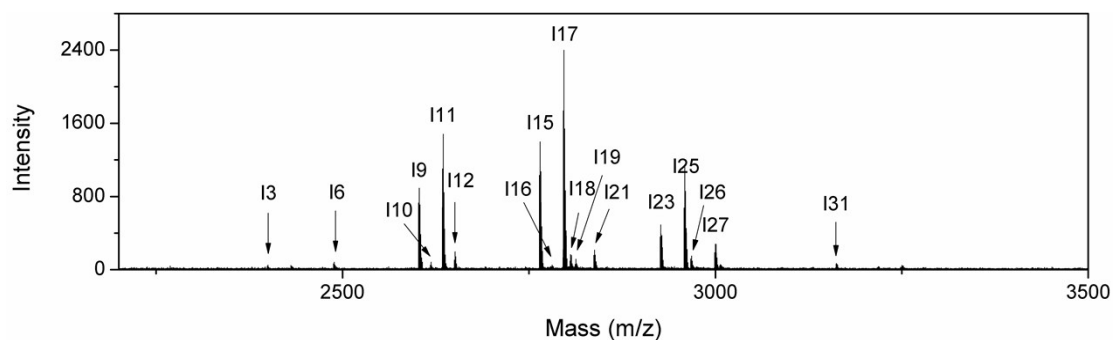


Fig. S3 MALDI-TOF MS spectra of human IgG digests (70 fmol μL^{-1}) after enrichment by commercial HILIC materials.

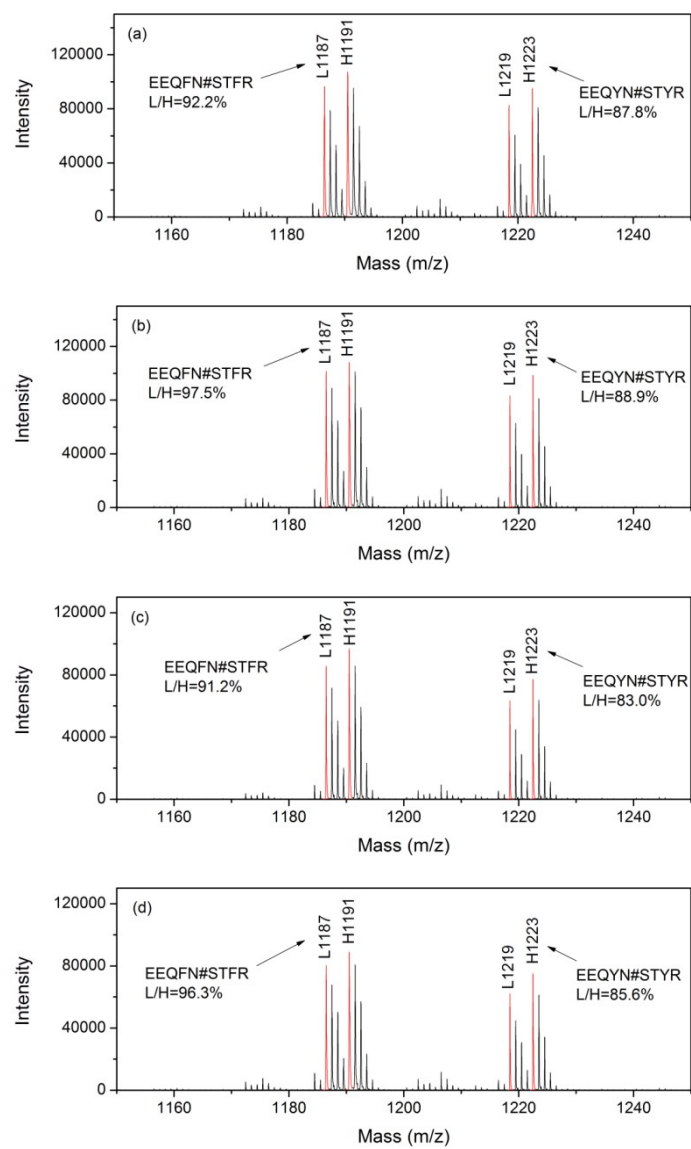


Fig. S4 Recovery of N-glycopeptides from human IgG digests using $\text{Fe}_3\text{O}_4@\text{TpPa-1}$.

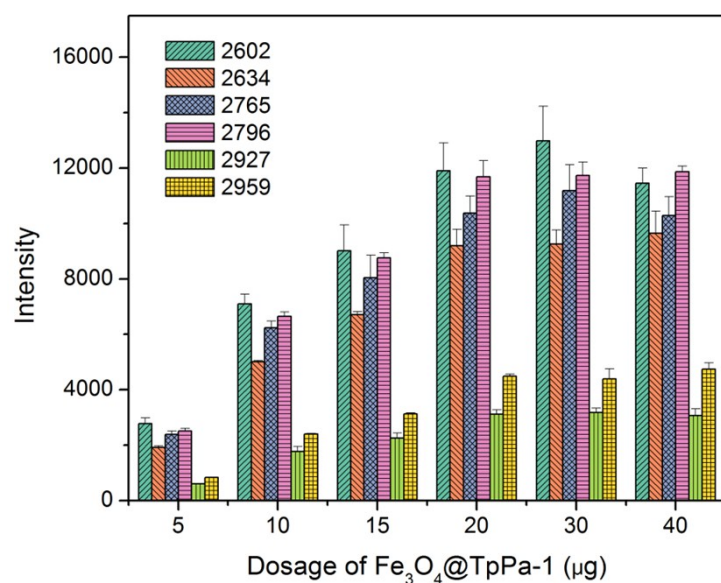


Fig. S5 The signal intensities of six selected glycopeptides from 3 µg of IgG digests after enrichment by Fe₃O₄@TpPa-1.

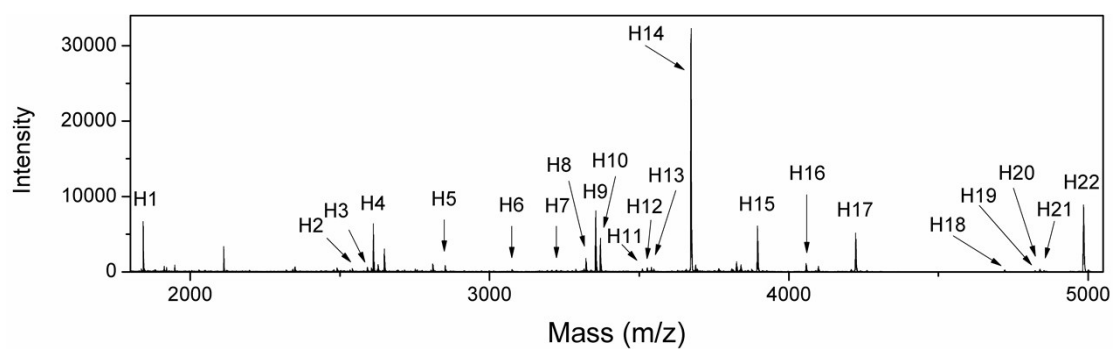


Fig. S6 MALDI-TOF MS spectra of HRP digests (125 fmol µL⁻¹) after enrichment by Fe₃O₄@TpPa-1.

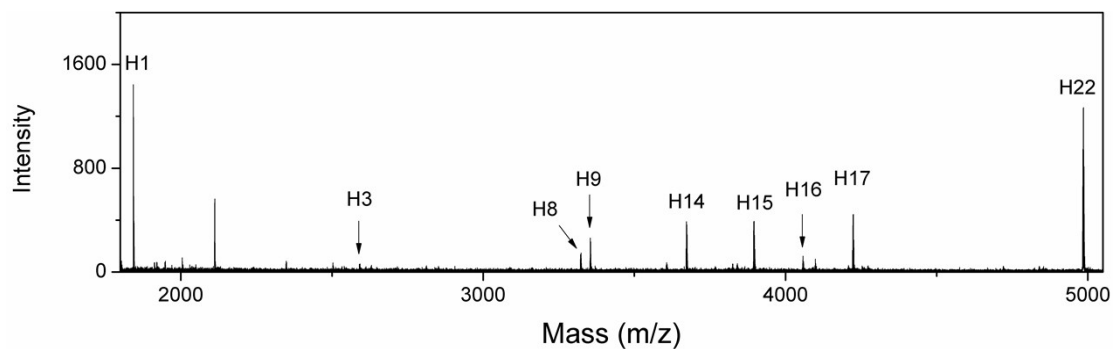


Fig. S7 MALDI-TOF MS spectra of HRP digests (125 fmol µL⁻¹) after enrichment by commercial HILIC materials.

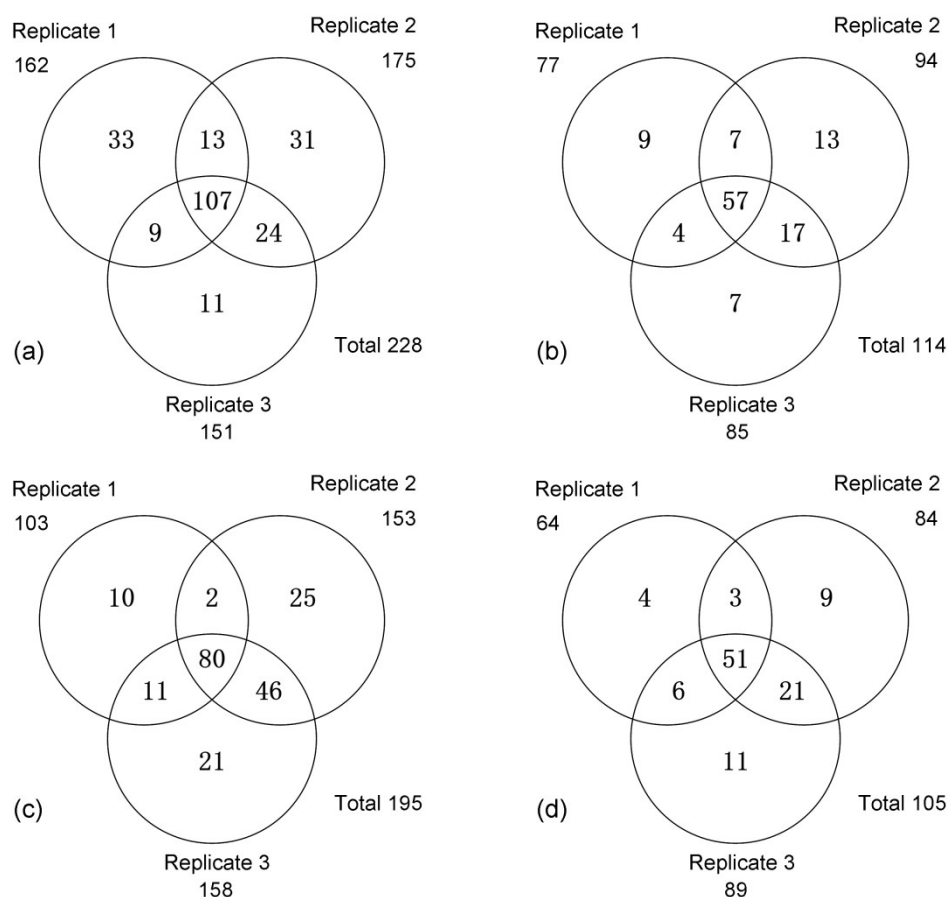


Fig. S8 The overlapping of identified glycopeptides (a, c) and glycoproteins (b, d) by triplicate analysis from tryptic digests of human serum after enrichment by $\text{Fe}_3\text{O}_4@\text{TpPa-1}$ (a, b) or commercial HILIC materials (c, d). All of Venn's diagrams were drawn with online Venny 2.1.0 (<http://bioinfogp.cnb.csic.es/tools/venny/>)

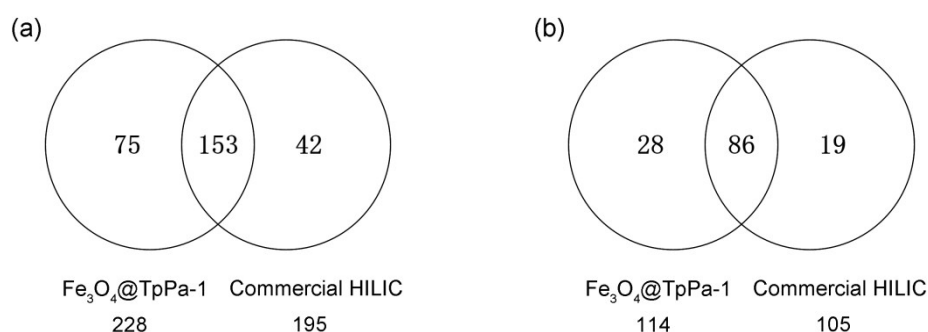


Fig. S9 The overlapping of identified glycopeptides (a) and glycoproteins (b) from digests of human serum after enrichment by $\text{Fe}_3\text{O}_4@\text{TpPa-1}$ and commercial HILIC materials.

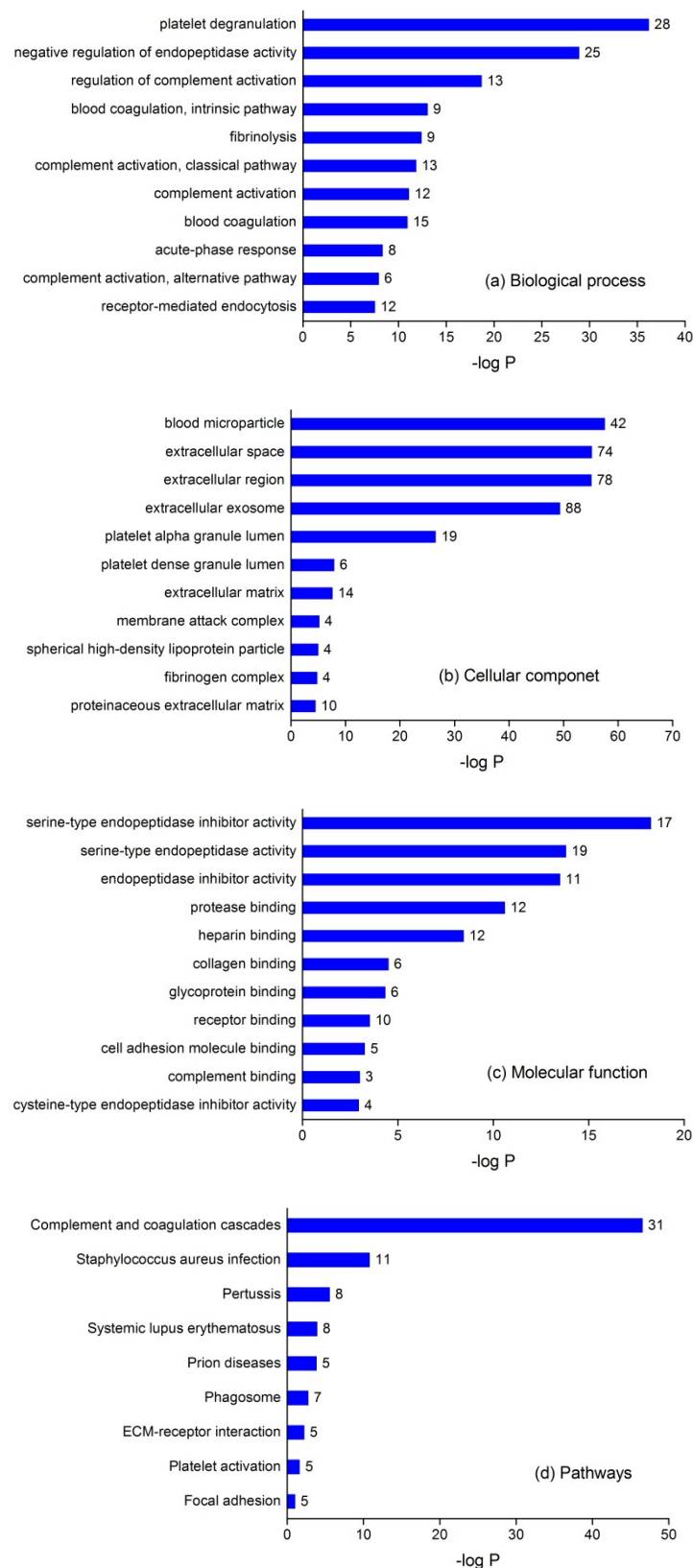


Fig. S10 The gene ontology annotations (a-biological process, b-cellular component, c-molecular function) and (d) pathway analysis of the identified glycoproteins from digests of human serum by $\text{Fe}_3\text{O}_4@\text{TpPa-1}$.

Table S1 Detailed information of the glycopeptides enriched by Fe₃O₄@TpPa-1 from human IgG tryptic digests.

NO.	m/z	Glycan composition	Peptide sequence
I1	2237.4	[Hex]3[HexNAc]2[Fuc]1	EEQFN#STFR
I2	2268.8	[Hex]3[HexNAc]2[Fuc]1	EEQYN#STYR
I3	2400.1	[Hex]3[HexNAc]3[Fuc]1	EEQFN#STFR
I4	2431.5	[Hex]3[HexNAc]3[Fuc]1	EEQYN#STYR
I5	2456.4	[Hex]3[HexNAc]4	EEQFN#STFR
I6	2488.4	[Hex]3[HexNAc]4	EEQYN#STYR
I7	2562.3	[Hex]4[HexNAc]3[Fuc]1	EEQFN#STFR
I8	2593.2	[Hex]4[HexNAc]3[Fuc]1	EEQYN#STYR
I9	2602.4	[Hex]3[HexNAc]4[Fuc]1	EEQFN#STFR
I10	2618.2	[Hex]4[HexNAc]4	EEQFN#STFR
I11	2634.3	[Hex]3[HexNAc]4[Fuc]1	EEQYN#STYR
I12	2650.3	[HexNAc]4[HexNAc]4	EEQYN#STYR
I13	2674.8	[Hex]3[HexNAc]5	EEQFN#STYR
I14	2691.7	[Hex]3[HexNAc]5	EEQYN#STYR
I15	2764.6	[Hex]4[HexNAc]4[Fuc]1	EEQFN#STFR
I16	2780.6	[Hex]5[HexNAc]4	EEQFN#STFR
I17	2796.4	[Hex]4[HexNAc]4[Fuc]1	EEQYN#STYR
I18	2805.9	[Hex]3[HexNAc]5[Fuc]1	EEQFN#STFR
I19	2812.7	[Hex]5[HexNAc]4	EEQYN#STYR
I20	2822.6	[Hex]4[HexNAc]5	EEQFN#STFR
I21	2838.3	[Hex]3[HexNAc]5[Fuc]1	EEQYN#STYR
I22	2853.9	[Hex]4[HexNAc]5	EEQYN#STYR
I23	2926.8	[Hex]5[HexNAc]4[Fuc]1	EEQFN#STFR
I24	2943.6	[Hex]5[HexNAc]4[Fuc]1	EEQFN#STYR
I25	2959.3	[Hex]5[HexNAc]4[Fuc]1	EEQYN#STYR
I26	2967.7	[Hex]4[HexNAc]5[Fuc]1	EEQFN#STFR
I27	2999.8	[Hex]4[HexNAc]5[Fuc]1	EEQYN#STYR
I28	3057.3	[Hex]4[HexNAc]4[Fuc]1[NeuAc]1	EEQFN#STFR
I29	3085.3	[Hex]4[HexNAc]4[Fuc]1[NeuAc]1	EEQYN#STYR
I30	3130.9	[Hex]5[HexNAc]5[Fuc]1	EEQFN#STFR
I31	3162.6	[Hex]5[HexNAc]5[Fuc]1	EEQYN#STYR
I32	3219.5	[Hex]5[HexNAc]4[Fuc]1[NeuAc]1	EEQFN#STFR
I33	3247.5	[Hex]4[HexNAc]4[Fuc]1	TKPREEQFN#STFR
I34	3251.7	[Hex]5[HexNAc]4[Fuc]1[NeuAc]1	EEQYN#STYR
I35	3279.6	[Hex]4[HexNAc]4[Fuc]1	TKPREEQYN#STYR
I36	3409.6	[Hex]5[HexNAc]4[Fuc]1	TKPREEQFN#STFR
I37	3441.8	[Hex]5[HexNAc]4[Fuc]1	TKPREEQYN#STYR

The N-glycosylation sites are marked with N#. HexNAc=N-acetylglucosamine, Hex=mannose, Fuc=fucose, NeuAc=sialic.

Table S2 Detailed information of the glycopeptides enriched by Fe₃O₄@TpPa-1 from HRP tryptic digests.

NO.	m/z	Glycan composition	Peptide sequence
H1	1842.3	XylMan ₃ FucGlcNAc ₂	NVGLN#R
H2	2541.1	XylMan ₃ FucGlcNAc ₂	SSPN#ATDTIPLVR
H3	2590.8	XylMan ₃ FucGlcNAc ₂	PTLN#TTYLQTLR
H4	2611.0	XylMan ₃ GlcNAc ₂	MGN#ITPLTGTQGQIR
H5	2850.4	FucGlcNAc	GLIQSDQELFSSPN#ATDTIPLVR
H6	3074.5	FucGlcNAc	LHFHDCFVNGCDASILLDN#TTSFR
H7	3222.0	Man ₃ FucGlcNAc ₂	SFAN#STQTFNFAFVEAMDR
H8	3320.9	XylMan ₃ FucGlcNAc ₂	QLTPTFYDNSCPN#VSNIVR
H9	3353.0	XylMan ₃ FucGlcNAc ₂	SFAN#STQTFNFAFVEAMDR
H10	3369.6	XylMan ₃ FucGlcNAc ₂	SFAN#STQTFNFAFVEAM*DR
H11	3508.7	XylMan ₂ FucGlcNAc ₂	GLIQSDQELFSSPN#ATDTIPLVR
H12	3525.6	XylMan ₃ GlcNAc ₂	GLIQSDQELFSSPN#ATDTIPLVR
H13	3539.6	Man ₃ FucGlcNAc ₂	GLIQSDQELFSSPN#ATDTIPLVR
H14	3671.6	XylMan ₃ FucGlcNAc ₂	GLIQSDQELFSSPN#ATDTIPLVR
H15	3894.6	XylMan ₃ FucGlcNAc ₂	LHFHDCFVNGCDASILLDN#TTSFR
H16	4056.9	XylMan ₃ GlcNAc ₂	QLTPTFYDNSC(AAVESACPR)PN#VSNIVR-H ₂ O
H17	4222.5	XylMan ₃ FucGlcNAc ₂	QLTPTFYDNSC(AAVESACPR)PN#VSNIVR
H18	4720.1	Man ₃ FucGlcNAc ₂ , Man ₃ FucGlcNAc ₂	LYN#FSNTGLPDPTLN#TTYLQTLR
H19	4821.7	XylMan ₂ FucGlcNAc ₂ , XylMan ₂ GlcNAc ₂	LYN#FSNTGLPDPTLN#TTYLQTLR
H20	4838.2	XylMan ₃ FucGlcNAc ₂ , XylMan ₃ GlcNAc ₂	LYN#FSNTGLPDPTLN#TTYLQTLR
H21	4852.7	Man ₃ FucGlcNAc ₂ , XylMan ₃ FucGlcNAc ₂	LYN#FSNTGLPDPTLN#TTYLQTLR
H22	4984.3	XylMan ₃ FucGlcNAc ₂ , XylMan ₃ FucGlcNAc ₂	LYN#FSNTGLPDPTLN#TTYLQTLR

The N-glycosylation sites are marked with N#. GlcNAc = N-acetylglucosamine, Fuc = fucose, Man = mannose, Xyl = xylose.

Chemical Communications, 2016, 52(15): 3251-3253.

Nanoscale, 2014, 6(6): 3150-3156.

ACS applied materials & interfaces, 2015, 7(29): 16011-16017.

Table S3 Matching situation of the glycopeptides enriched by Fe₃O₄@TpPa-1 from IgG tryptic digests in each figure.

NO.	m/z	Fig. 3a	Fig. 3b	Fig. 3c	Fig. 4a	Fig. 4b	Fig. 4c	Fig. 4d	Fig. 5b	Fig. 5c
I1	2237.4			√						
I2	2268.8			√						
I3	2400.1			√	√					
I4	2431.5			√	√					
I5	2456.4			√						
I6	2488.4			√	√					
I7	2562.3			√						
I8	2593.2			√						
I9	2602.4	√	√	√	√	√	√	√	√	√
I10	2618.2			√	√					
I11	2634.3	√		√	√	√	√	√	√	√
I12	2650.3			√	√	√			√	
I13	2674.8			√						
I14	2691.7			√						
I15	2764.6	√	√	√	√	√	√	√	√	√
I16	2780.6			√	√				√	√
I17	2796.4	√	√	√	√	√	√	√	√	√
I18	2805.9		√	√	√					
I19	2812.7			√	√					
I20	2822.6			√						
I21	2838.3			√	√	√	√		√	
I22	2853.9			√						
I23	2926.8	√	√	√	√	√	√		√	√
I24	2943.6			√						
I25	2959.3	√		√	√	√	√	√	√	√
I26	2967.7		√	√	√					
I27	2999.8			√	√	√	√		√	√
I28	3057.3			√						
I29	3085.3			√	√					
I30	3130.9			√						
I31	3162.6			√	√					
I32	3219.5			√						
I33	3247.5			√	√					
I34	3251.7			√						
I35	3279.6			√	√					
I36	3409.6			√						
I37	3441.8			√						
Total		6	6	37	21	9	8	5	10	8

Table S4 List of identified glycoproteins and glycopeptides from digests of human serum after enrichment by Fe₃O₄@TpPa-1.

Master Protein Accessions	Definition	Sequence
4501987	afamin precursor [Homo sapiens]	YAEDKFN#ETTEK HN#FSHCCSK DIENFN#STQK ICAMEGLPQKHN#FSHCCSK FN#ETTEK
4502027	serum albumin preproprotein [Homo sapiens]	QEPERN#ECFLQHKDDNPNLPR
4502163	apolipoprotein D precursor [Homo sapiens]	CIQAN#YSLMENGK ADGTVNQIEGEATPVN#LTEPAK ADGTVNQIEGEATPVN#LTEPAKLEVK
4502261	antithrombin-III precursor [Homo sapiens]	LGACN#DTLQQLMEVFK WVSN#KTEGR AAINKWVSN#KTEGR
4502337	zinc-alpha-2-glycoprotein precursor [Homo sapiens]	DIVEYYN#DSN#GSHVLQGR FGCEIENN#RSSGAFWK
4502503	C4b-binding protein alpha chain precursor [Homo sapiens]	SRPAN#HCVYFYGDEISFSCHETSR
4502511	complement component C9 preproprotein [Homo sapiens]	CNGDN#DCGDFSDEDDCESEPRPPCR
4503625	coagulation factor X isoform 1 preproprotein [Homo sapiens]	GDNN#LTR
4503635	prothrombin isoform 1 preproprotein [Homo sapiens]	SRYPHKPEIN#STTHPGADLQEN#FCR YPHKPEIN#STTHPGADLQEN#FCR SEGSSVN#LSPPLEQCVPDRGQQYQGR
4503689	fibrinogen alpha chain isoform alpha-E preproprotein [Homo sapiens]	GGSTSYGTGSETESPRN#PSSAGSWNSGSSGPGST GNR N#PSSAGSWNSGSSGPGSTGNR
4504383	hepatocyte growth factor activator isoform 2 preproprotein [Homo sapiens]	CFLGN#GTGYR
4504489	histidine-rich glycoprotein precursor [Homo sapiens]	VIDFN#CTTSSVSSALANTK KYWN#DCEPPDSR
4504893	kininogen-1 isoform 2 precursor [Homo sapiens]	HGIQYFNN#NTQHSSLFMLNEVKR

	sapiens]	YNSQN#QSNNQFVLYR HGIQYFNN#NTQHSSLFMLNEVK LNAENN#ATFYFK ITYSIVQTN#CSK KYNSQN#QSNNQFVLYR
4505047	lumican precursor [Homo sapiens]	LHINHNN#LTESVGPLPK KLHINHNN#LTESVGPLPK
4505529	alpha-1-acid glycoprotein 2 precursor [Homo sapiens]	QN#QCFYN#SSYLVNQR
4505881	plasminogen isoform 1 precursor [Homo sapiens]	FSPATHPSEGLEEN#YCRNPDNDPQGPWCYTDP EKR TPENYPNAGLTMN#YCR
4506115	vitamin K-dependent protein C preproprotein [Homo sapiens]	EVFVHPN#YSK
4508007	zinc finger protein 174 isoform a [Homo sapiens]	GNGLQNPEPRGAN#MSEPR
4557287	angiotensinogen preproprotein [Homo sapiens]	VYIHPFHLVIHN#ESTCEQLAK
4557351	cholinesterase precursor [Homo sapiens]	WSDIWN#ATK DN#NSIITR EN#ETEIK DN#YTKAEEILSR DN#NSIITRK
4557373	biotinidase isoform 3 [Homo sapiens]	FN#DTEVLQR
4557389	complement component C8 alpha chain preproprotein [Homo sapiens]	RAATPAAVTCQLSN#WSEWTDPCQDKK
4557485	ceruloplasmin precursor [Homo sapiens]	ELHHLQEQN#VSNAFLDK EN#LTAPGSDSAVFFEQTTR ELHHLQEQN#VSNAFLDKGEFYIGSK EHEGAIYPD#TTDFQR
4557871	serotransferrin precursor [Homo sapiens]	CGLVPVLAENYN#KSDNCEDTPEAGYFAVAVVK K ILRQQHFLFGSN#VTDCSGN#FCLFR QQHFLFGSN#VTDCSGN#FCLFR LCMGSGLN#LCEPNNKEGYGYTGAFR IN#HCRFDEFFSEGCAPGSKK DSSLCKLCMGSGLN#LCEPNNKEGYGYTGAFR CGLVPVLAENYN#KSDNCEDTPEAGYFAVAVVK
4826762	haptoglobin isoform 1 preproprotein [Homo sapiens]	VVLHPN#YSQVDIGLIK
5031863	galectin-3-binding protein precursor	ALGFEN#ATQALGR

	[Homo sapiens]	TVIRPFYLTN#SSGVD GLN#LTEDTYKPR AAIPSALDTN#SSK DAGVVCTN#ETR YKGLN#LTEDTYKPR
5453914	phospholipid transfer protein isoform a precursor [Homo sapiens]	VSN#VSCQASVSR
11321561	hemopexin precursor [Homo sapiens]	GKEGHFYYN#ISEVK ERSWPAVGN#CSSALR SWPAVGN#CSSALR SLGPN#SCSANGPGLYLIHGPNLYCYSDVEKLNA AK SLGPN#SCSANGPGLYLIHGPNLYCYSDVEK ALPQPQN#VTSLLGCTH NGTGHGN#STHHGPEYMR
14550407	complement C2 isoform 1 preproprotein [Homo sapiens]	QSVPAHFVALN#GSK
16418467	leucine-rich alpha-2-glycoprotein precursor [Homo sapiens]	TMFPN#LTDVR KLPPGLLAN#FTLLR
16933540	prolyl endopeptidase FAP isoform 1 [Homo sapiens]	MFSQN#DTR LPPGLLAN#FTLLR DKMFSQN#DTR
19115964	1-phosphatidylinositol 4,5-bisphosphate phosphodiesterase delta-3 [Homo sapiens]	SVN#ASNYGLSPDR
19923106	serum paraoxonase/arylesterase 1 precursor [Homo sapiens]	MNSAN#YSPQEMWNSGCQLVALNFQTPGYEMD LNAGR
21071030	alpha-1B-glycoprotein precursor [Homo sapiens]	HAN#WTLTPLK VTQVYAEN#GTVLQGSTVASVYK EGDHEFLEVPEAQEDVEATFPVHQPGN#YSCSYR
21264359	mannan-binding lectin serine protease 1 isoform 2 precursor [Homo sapiens]	NN#LTTYK
21361302	kallistatin isoform 2 precursor [Homo sapiens]	FLN#DTMAVYEAK
21361306	BDNF/NT-3 growth factors receptor isoform a precursor [Homo sapiens]	NSNLQHIN#FTR
21450861	attractin isoform 1 preproprotein [Homo sapiens]	N#HSCSEGQISIFR VFHIHN#ESWVLLTPK AATCINPLN#GSVCERPAN#HSAK

		IDSTGN#VTNELR
		CIN#QSICEK
		GICN#SSDVR
		CEN#LTTGK
21489959	immunoglobulin J chain precursor [Homo sapiens]	IIVPLNNREN#ISDPTSPLR
		EN#ISDPTSPLR
22091452	apolipoprotein M isoform 1 [Homo sapiens]	TELFSSSCP GGIMLN#ETGQGYQR
24308201	adipocyte plasma membrane-associated protein [Homo sapiens]	AGPN#GTLFVADAYK
27894376	neural cell adhesion molecule L1-like protein isoform 1 precursor [Homo sapiens]	IIPSN#NSGTFR
28373117	contactin-1 isoform 1 precursor [Homo sapiens]	GTEWLVN#SSR
29171717	phosphatidylinositol-glycan-specific phospholipase D precursor [Homo sapiens]	LNVEAAN#WTVR
		N#LTTSLTESVDR
31377566	E3 ubiquitin-protein ligase RNF168 [Homo sapiens]	DAIPSLSECQCGICMEILVEPVTLP CN#HTLCKPCF QSTVEK
31377806	polymeric immunoglobulin receptor precursor [Homo sapiens]	VPGN#VTAVLGETLK
32171249	prostaglandin-H2 D-isomerase precursor [Homo sapiens]	SVVAPATDGGLN#LTSTFLR
32307128	nuclear receptor coactivator 6 isoform 1 [Homo sapiens]	MNNPATVMIPP GGN#VSSMMAPGPNPELQPR
33519481	neural cell adhesion molecule 2 precursor [Homo sapiens]	YN#CTATNHIGTR
38016904	ADAMTS-like protein 4 isoform 1 precursor [Homo sapiens]	LVSGN#LTDR
38016947	complement C5 isoform 1 preproprotein [Homo sapiens]	AN#ISHKDMQLGR
40217843	cartilage oligomeric matrix protein precursor [Homo sapiens]	CN#DTIPEDYETHQLR
40549451	lymphatic vessel endothelial hyaluronic acid receptor 1 precursor [Homo sapiens]	ANQQLN#FTEAK
45592949	transmembrane and immunoglobulin domain-containing protein 1 isoform 1 precursor [Homo sapiens]	SGNKIN#SSSVCVSSISENDNGISFTCR
47078292	integrin beta-3 precursor [Homo sapiens]	LRPDDSKN#FSIQVR

47132557	fibronectin isoform 1 precursor [Homo sapiens]	LDAPTNLQFVN#ETDSTVLVR DQCIVDDITYNVN#DTFHK RHEEGHMLN#CTCFGQGR HEEGHMLN#CTCFGQGR GGNSNGALCHFPFLYNNHN#YTDCTSEGR DQCIVDDITYNVN#DTFHKR
50659080	alpha-1-antichymotrypsin precursor [Homo sapiens]	YTGN#ASALFILPDQDK FN#LTETSEAEIHQSFQHLLR TLN#QSSDELQLSMGNAMFVK LINDYVKN#GTR KLINDYVKN#GTR
58331240	fetuin-B isoform 1 precursor [Homo sapiens]	VLYLAAYN#CTLRPVSK
58530840	desmoplakin isoform I [Homo sapiens]	HQNQNTIQELLQN#CSDCLMR
62243248	insulin-like growth factor-binding protein 3 isoform a precursor [Homo sapiens]	GLCVN#ASAVSR
62739186	complement factor H isoform a precursor [Homo sapiens]	MDGASN#VTCINSR SPDVIN#GSPISQK IPCSQPPQIEHGTIN#SSR ISEEN#ETTCYMGK
62912468	C4b-binding protein beta chain isoform 1 precursor [Homo sapiens]	EWDN#TTTECR TLFCN#ASK
66347875	complement C1r subcomponent precursor [Homo sapiens]	CN#YSIRVER
66932947	alpha-2-macroglobulin isoform a precursor [Homo sapiens]	VSN#QTLSLFFTVLQDVPVR SLGNVN#FTVSAEALSEQELCGTEVPSVPEHGRK
67190163	proteoglycan 4 isoform A precursor [Homo sapiens]	N#GTLVAFR
67782358	complement factor B preproprotein [Homo sapiens]	SPYYN#VSDEISFHCYDGYTLR
70778918	inter-alpha-trypsin inhibitor heavy chain H2 precursor [Homo sapiens]	GAFISN#FSMTVDGK
70906435	fibrinogen beta chain isoform 1 preproprotein [Homo sapiens]	N#YCGLPGEYWLGNDKISQLTR N#YCGLPGEYWLGNDK QGFGNVATNTDGKN#YCGLPGEYWLGNDK
70906439	fibrinogen gamma chain isoform gamma-B precursor [Homo sapiens]	VDKDLQSLEDILHQVEN#KTSEVK

		DN#CCILDER
73858564	corticosteroid-binding globulin precursor [Homo sapiens]	AQLLQGLGFN#LTER
73858566	heparin cofactor 2 precursor [Homo sapiens]	DFVN#ASSK
		N#LSMPLLPADFHK
73858570	plasma protease C1 inhibitor precursor [Homo sapiens]	GVTSVSQIFHSPDLAIRDTFVN#ASR
		VLSN#NSDANLELINTWVAK
		DTFVN#ASR
78191798	plasma kallikrein isoform 1 preproprotein [Homo sapiens]	IYSGILN#LSDITK
		LQAPLN#YTEFQKPICLPSK
		GVN#VCQETCTK
		GVNFN#VSK
86788132	EGF-containing fibulin-like extracellular matrix protein 1 precursor [Homo sapiens]	IQCAAGYEQSEHN#VCQDIDECTAGTHNCR
88702793	vasorin precursor [Homo sapiens]	LHEITN#ETFR
88853069	vitronectin precursor [Homo sapiens]	NN#ATVHEQVGGPSLTSDLQAQSK
		N#GSLFAFR
89191868	von Willebrand factor preproprotein [Homo sapiens]	MEACMLN#GTVIGPGK
105990532	apolipoprotein B-100 precursor [Homo sapiens]	FN#SSYLQGTNQITGR
		YDFN#SSMLYSTAK
		FVEGSHN#STVSLTTK
		QVLFLDTVYGN#CSTHFTVK
105990535	coagulation factor V preproprotein [Homo sapiens]	TNIN#SSRDPDNIAAWYLR
		TNIN#SSR
110624774	C-type mannose receptor 2 precursor [Homo sapiens]	VTPACN#TSLPAQR
112380628	lysosome-associated membrane glycoprotein 1 precursor [Homo sapiens]	GHTLTLN#FTR
115298678	complement C3 preproprotein [Homo sapiens]	TVLTPATNHMGN#VTFTIPANREFK
		TVLTPATNHMGN#VTFTIPANR
		MN#KTVAVR
		TVLTPATNHMGN#VTFTIPANREFKSEK
115529484	CD109 antigen isoform 1 preproprotein [Homo sapiens]	HLN#GTITAK
116292750	apolipoprotein(a) precursor [Homo sapiens]	TPENYPNAGLTEN#YCR

	sapiens]	TTENYPNAGLIMN#YCR WEYCN#LTR
119392081	complement factor I isoform 2 preproprotein [Homo sapiens]	LSDLSIN#STECLHVHCR
		LISN#CSK N#GTAVCATNR
119964726	cation-independent mannose-6- phosphate receptor precursor [Homo sapiens]	SLLEFN#TTVSCDQQGTNHR
		YHN#QTLR
126012562	prolow-density lipoprotein receptor- related protein 1 preproprotein [Homo sapiens]	WTGHN#VTVVQR
		LYWISSGN#HTINR FN#STEYQVVTR
133925809	inter-alpha-trypsin inhibitor heavy chain H3 preproprotein [Homo sapiens]	NAHGEEKEN#LTAR
		KNAHGEEKEN#LTAR
145275213	coagulation factor XII preproprotein [Homo sapiens]	RN#HSCEPCQTLAVR
148277022	selenoprotein P isoform 2 [Homo sapiens]	EGYSN#ISYIVVNHQGISSR
153266841	beta-2-glycoprotein 1 precursor [Homo sapiens]	LGN#WSAMPSCK
		VYKPSAGN#NSLYR
154759291	protein Z-dependent protease inhibitor precursor [Homo sapiens]	ETFFN#LSK
156119625	inter-alpha-trypsin inhibitor heavy chain H1 isoform a preproprotein [Homo sapiens]	ICDLLVANNHFAHFFAPQN#LTNMNK
156523970	alpha-2-HS-glycoprotein preproprotein [Homo sapiens]	AALAAFNAQNN#GSNFAQLEEISR
		KVCQDCPLLAPLN#DTR VCQDCPLLAPLN#DTR VCQDCPLLAPLN#DTRVVHAAK
156616294	N-acetylmuramoyl-L-alanine amidase precursor [Homo sapiens]	LEPVHLQLQCMSQEQLAQVAAN#ATK
157266300	aminopeptidase N precursor [Homo sapiens]	KLN#YTLSQGHR
162809334	pregnancy zone protein precursor [Homo sapiens]	VLNCDKQEVCEEFSQQLNSN#GCITQQVHTK
166795301	prenylcysteine oxidase 1 precursor [Homo sapiens]	GELN#TSIFSSRPIDK

167857790	alpha-1-acid glycoprotein 1 precursor [Homo sapiens]	QDQCIYN#TTYLVNQVR
		QDQCIYN#TTYLVNQREN#GTISR EN#GTISR
169790833	lysosome-associated membrane glycoprotein 2 isoform C precursor [Homo sapiens]	VQPFN#VTQ GK
		LN#SSTIK
189163542	alpha-1-antitrypsin precursor [Homo sapiens]	YLGN#ATAIFFLPDEGK
189242612	complement component C6 precursor [Homo sapiens]	VLN#FTTK
		KLECNGEN#DCGDNNDERDCGR
205277441	thyroxine-binding globulin precursor [Homo sapiens]	VTACHSSQPN#ATLYK
256217721	carboxypeptidase N subunit 2 precursor [Homo sapiens]	AFGSNPN#LTK
310923208	microfibril-associated glycoprotein 4 isoform 1 precursor [Homo sapiens]	FN#GSVSFFR
322302700	extracellular matrix protein 1 isoform 3 precursor [Homo sapiens]	HIPGLIHN#MTAR
333944015	cadherin-13 isoform 2 [Homo sapiens]	IN#NTHALVSLQLNLK
355594753	clusterin preproprotein [Homo sapiens]	LAN#LTQGEDQYYLR EDALN#ETR EIRHN#STGCLR KKEDALN#ETR HN#STGCLR LKELPGVCN#ETMMALWEECKPCLK
429836859	multiple epidermal growth factor-like domains protein 8 isoform 1 precursor [Homo sapiens]	VN#STELFHVDR
		ALLTN#VSSVALGSR
410170896; 178557739	complement C4-A isoform 1 preproprotein [Homo sapiens]/ complement C4-B preproprotein [Homo sapiens]	FSDGLESN#SSTQFEVK
		N#TTCQDLQIEVTVK FSDGLESN#SSTQFEVKK GLN#VTLSSTGR
45580723; 4826762	haptoglobin-related protein precursor [Homo sapiens]/ haptoglobin isoform 1 preproprotein [Homo sapiens]	MVSHHN#LTTGATLINEQWLLTTAK
		NLFLN#HSEN#ATAKDIAPTLTLYVGKK NLFLN#HSEN#ATAK

		NLFLN#HSEN#ATAKDIAPTLTLYVGK
5031695;	complement factor H-related protein 2	LQNNENN#ISCVER
118442839	isoform 1 precursor [Homo sapiens]/ complement factor H-related protein 1 precursor [Homo sapiens]	

The N-glycosylation sites are marked with N#.

Table S5 List of identified glycoproteins and glycopeptides from digests of human serum after enrichment by commercial HILIC materials.

Master Protein Accessions	Definition	Sequence
4501987	afamin precursor [Homo sapiens]	ICAMEGLPQKHNF#FSHCCSK DIENFN#STQK HN#FSHCCSK YAEDKFN#ETTEK DLLRN#CCNTENPPGCYR YAEDKFN#ETTEKSLK FN#ETTEK
4502027	serum albumin preproprotein [Homo sapiens]	QEPERN#ECFLQHKDDNPNLPR
4502119	membrane primary amine oxidase isoform 1 [Homo sapiens]	YLYLASN#HSNK
4502163	apolipoprotein D precursor [Homo sapiens]	ADGTVNQIEGEATPVN#LTEPAKLEVK CIQAN#YSLMENGK ADGTVNQIEGEATPVN#LTEPAK
4502261	antithrombin-III precursor [Homo sapiens]	AAINKWVSN#KTEGR WVSN#KTEGR
4502337	zinc-alpha-2-glycoprotein precursor [Homo sapiens]	FGCEIENN#RSSGAFWK DIVEYYN#DSN#GSHVLQGR
4502951	collagen alpha-1(III) chain preproprotein [Homo sapiens]	ASQN#ITYHCK
4503143	cathepsin D preproprotein [Homo sapiens]	GSLSYLN#VTR
4503635	prothrombin isoform 1 preproprotein [Homo sapiens]	YPHKPEIN#STTHPGADLQEN#FCR SRYPHKPEIN#STTHPGADLQEN#FCR
4503689	fibrinogen alpha chain isoform alpha-E preproprotein [Homo sapiens]	GGSTSYGTGSETESPRN#PSSAGSWNSGSSGPGST GNR
4504383	hepatocyte growth factor activator isoform 2 preproprotein [Homo sapiens]	CFLGN#GTGYR
4504489	histidine-rich glycoprotein precursor [Homo sapiens]	VIDFN#CTTSSVSSALANTK
4505047	lumican precursor [Homo sapiens]	KLHINHNN#LTESVGPLPK LHINHNN#LTESVGPLPK
4505529	alpha-1-acid glycoprotein 2 precursor	QN#QCFYN#SSYLVNQR

	[Homo sapiens]	
4505881	plasminogen isoform 1 precursor	FSPATHPSEGLEEN#YCRNPDNDPQGPWCYTDP
	[Homo sapiens]	EKR
4506115	vitamin K-dependent protein C preproprotein [Homo sapiens]	EVFVHPN#YSK
4557287	angiotensinogen preproprotein [Homo sapiens]	VYIHPFHLVIHN#ESTCEQLAK
4557351	cholinesterase precursor [Homo sapiens]	WSDIWN#ATK
		EN#ETEIIK
		DN#NSIITR
		DN#YTKAEEILSR
		LTGCSREN#ETEIIK
4557373	biotinidase isoform 3 [Homo sapiens]	FN#DTEVLQR
4557485	ceruloplasmin precursor [Homo sapiens]	EHEGAIYPDN#TTDFQR
		ELHHLQEQN#VSNAFLDK
		EN#LTAPGSDSAVFFEQTTR
4557871	serotransferrin precursor [Homo sapiens]	QQQHLFGSN#VTDCSGN#FCLFR
		IN#HCRFDEFFSEGCAPGSK
4557892	phosphatidylcholine-sterol acyltransferase precursor [Homo sapiens]	AELSN#HTRPVILVPGCLGNQLEAK
4826762	haptoglobin isoform 1 preproprotein [Homo sapiens]	NLFLN#HSENATAK
		VVLHPN#YSQVDIGLIK
		MVSHHN#LTTGATLINEQWLLTTAK
5031863	galectin-3-binding protein precursor [Homo sapiens]	AAIPSALDTN#SSK
		GLN#LTEDTYKPR
		ALGFEN#ATQALGR
		DAGVVCTN#ETR
		TVIRPFYLTN#SSGVD
		YKGLN#LTEDTYKPR
5453914	phospholipid transfer protein isoform a precursor [Homo sapiens]	VSN#VSCQASVSR
11321561	hemopexin precursor [Homo sapiens]	ALPQPQN#VTSLLGCTH
		ERSWPAVGN#CSSALR
		SWPAVGN#CSSALR
		N#GTGHGN#STHHGPEYMR
14550407	complement C2 isoform 1 preproprotein [Homo sapiens]	QSVPAHFVALN#GSK
		TMFPN#LTDVR

16418467	leucine-rich alpha-2-glycoprotein precursor [Homo sapiens]	MFSQN#DTR
		KLPPGLLAN#FTLLR
19923106	serum paraoxonase/arylesterase 1 precursor [Homo sapiens]	HAN#WTLTPLK
		VTQVYAEN#GTVLQGSTVASVYK
21264359	mannan-binding lectin serine protease 1 isoform 2 precursor [Homo sapiens]	NN#LTTYK
21361302	kallistatin isoform 2 precursor [Homo sapiens]	FLN#DTMAVYEAK
21361306	BDNF/NT-3 growth factors receptor isoform a precursor [Homo sapiens]	N#LTIVDSGLK
		NSNLQHIN#FTR
21450861	attractin isoform 1 preproprotein [Homo sapiens]	IDSTGN#VTNELR
		N#HSCSEGQISIFR
		GICN#SSDVR
		AATCINPLN#GSVCERPAN#HSAK
		VFHIHN#ESWVLLTPK
		CEN#LTTGK
21489959	immunoglobulin J chain precursor [Homo sapiens]	EN#ISDPTSPLR
		IIVPLNNREN#ISDPTSPLR
22091452	apolipoprotein M isoform 1 [Homo sapiens]	TELFSSSCPGGIMLN#ETGQGYQR
24308201	adipocyte plasma membrane-associated protein [Homo sapiens]	AGPN#GTLFVADAYK
27262659	macrophage colony-stimulating factor 1 receptor precursor [Homo sapiens]	HTN#YSFSPWHGFTIHR
27894376	neural cell adhesion molecule L1-like protein isoform 1 precursor [Homo sapiens]	IIPSN#NSGTFR
28373117	contactin-1 isoform 1 precursor [Homo sapiens]	GTEWLVN#SSR
29171717	phosphatidylinositol-glycan-specific phospholipase D precursor [Homo sapiens]	LNVEAAN#WTVR
		LGTSLSGGHVLNMN#GTLK
		N#LTTSLSLVDR
31377806	polymeric immunoglobulin receptor precursor [Homo sapiens]	VPGN#VTAVLGETLK
33519481	neural cell adhesion molecule 2 precursor [Homo sapiens]	YN#CTATNHIGTR
34734066	fibulin-1 isoform D precursor [Homo sapiens]	CATPHGDN#ASLEATFVK

	sapiens]	CATPHGDN#ASLEATFVKR
38016947	complement C5 isoform 1 preproprotein [Homo sapiens]	AN#ISHKDMQLGR
40217843	cartilage oligomeric matrix protein precursor [Homo sapiens]	CN#DTIPEDYETHQLR
40317626	thrombospondin-1 precursor [Homo sapiens]	VVN#STTGPGEHLR
40549451	lymphatic vessel endothelial hyaluronic acid receptor 1 precursor [Homo sapiens]	KANQQLN#FTEAK
47078292	integrin beta-3 precursor [Homo sapiens]	LRPDDSKN#FSIQVR
47132557	fibronectin isoform 1 precursor [Homo sapiens]	HEEGHMLN#CTCFGQGR
		RHEEGHMLN#CTCFGQGR
		GGNSNGALCHFPFLYNNHN#YTDCTSEGR
50659080	alpha-1-antichymotrypsin precursor [Homo sapiens]	LINDYVKN#GTR
		KLINDYVKN#GTR
		YTGN#ASALFILPDQDK
54112390	voltage-dependent calcium channel subunit alpha-2/delta-1 preproprotein [Homo sapiens]	DAVNN#ITAK
58331240	fetuin-B isoform 1 precursor [Homo sapiens]	VLYLAAYN#CTLRPVSK
		GCN#DSDVLAVAGFALR
62243248	insulin-like growth factor-binding protein 3 isoform a precursor [Homo sapiens]	GLCVN#ASAVSR
62739186	complement factor H isoform a precursor [Homo sapiens]	IPCSQPPQIEHGTIN#SSR
		MDGASN#VTCINSR
		ISEEN#ETTCYMGK
62912468	C4b-binding protein beta chain isoform 1 precursor [Homo sapiens]	TLFCN#ASK
		KTLCN#ASK
66347875	complement C1r subcomponent precursor [Homo sapiens]	CN#YSIRVER
67190163	proteoglycan 4 isoform A precursor [Homo sapiens]	N#GTLVAFR
70778918	inter-alpha-trypsin inhibitor heavy chain H2 precursor [Homo sapiens]	GAFISN#FSMTVDGK
73858564	corticosteroid-binding globulin	AQLLQGLGFN#LTER

	precursor [Homo sapiens]	
73858566	heparin cofactor 2 precursor [Homo sapiens]	N#LSMPLLPADFHK
		DFVN#ASSK
73858570	plasma protease C1 inhibitor precursor [Homo sapiens]	DTFVN#ASR
78191798	plasma kallikrein isoform 1 preproprotein [Homo sapiens]	GVN#VCQETCTK
		IYSGILN#LSDITK
		LQAPLN#YTEFQKPICLPSK
81158226	neuronal cell adhesion molecule isoform A precursor [Homo sapiens]	FN#HTQTIQKQ
88702793	vasorin precursor [Homo sapiens]	LHEITN#ETFR
88853069	vitronectin precursor [Homo sapiens]	N#GSLFAFR
		NN#ATVHEQVGGPSLTSDLQAQSK
89191868	von Willebrand factor preproprotein [Homo sapiens]	MEACMLN#GTVIGPGK
105990532	apolipoprotein B-100 precursor [Homo sapiens]	FVEGSHN#STVSLTTK
		YDFN#SSMLYSTAK
		FN#SSYLQGTNQTGR
		SYN#ETKIKFDK
105990535	coagulation factor V preproprotein [Homo sapiens]	TNIN#SSRDPDNIAAWYLR
		TWN#QSIALR
110618248	cadherin-related family member 5 isoform 1 precursor [Homo sapiens]	ITN#HSHFR
112380628	lysosome-associated membrane glycoprotein 1 precursor [Homo sapiens]	GHTLTLN#FTR
115298678	complement C3 preproprotein [Homo sapiens]	TVLTPATNHMGN#VTFTIPANR
		MN#KTVAVR
		TVLTPATNHMGN#VTFTIPANREFK
		TVLTPATNHMGN#VTFTIPANREFKSEK
115529484	CD109 antigen isoform 1 preproprotein [Homo sapiens]	HLN#GTITAK
		TQDEILFSN#STR
116292750	apolipoprotein(a) precursor [Homo sapiens]	WEYCN#LTR
118442839	complement factor H-related protein 1 precursor [Homo sapiens]	LQNNENN#ISCOVER
119392081	complement factor I isoform 2 preproprotein [Homo sapiens]	LISN#CSK

		N#GTAVCATNR
119964726	cation-independent mannose-6-phosphate receptor precursor [Homo sapiens]	TN#ITLVCKPGDLESAPVLR
		MN#FTGGDTCHK
126012562	prolow-density lipoprotein receptor-related protein 1 preproprotein [Homo sapiens]	N#STTLVMHMK
		LYWISSGN#HTINR
		WTGHN#VTVVQR
133925809	inter-alpha-trypsin inhibitor heavy chain H3 preproprotein [Homo sapiens]	NAHGEEKEN#LTAR
145275213	coagulation factor XII preproprotein [Homo sapiens]	RN#HSCEPCQTLAVR
		N#HSCEPCQTLAVR
148277022	selenoprotein P isoform 2 [Homo sapiens]	EGYSN#ISYIVVNHQGISSR
153266841	beta-2-glycoprotein 1 precursor [Homo sapiens]	LGN#WSAMPSCK
		VYKPSAGN#NSLYR
154759291	protein Z-dependent protease inhibitor precursor [Homo sapiens]	ETFFN#LSK
156231037	kininogen-1 isoform 1 precursor [Homo sapiens]	ITYSIVQTN#CSK
		LNAENN#ATFYFK
		YNSQN#QSNNQFVLYR
		KYNSQN#QSNNQFVLYR
		HGIQYFNN#NTQHSSLFMLNEVKR
		HGIQYFNN#NTQHSSLFMLNEVK
156523970	alpha-2-HS-glycoprotein preproprotein [Homo sapiens]	KVCQDCPLLAPLN#DTR
		VCQDCPLLAPLN#DTR
		VCQDCPLLAPLN#DTRVVHAAK
		AALAAFNAQNN#GSNFQLEEISR
157266300	aminopeptidase N precursor [Homo sapiens]	KLN#YTLSQGHR
		AEFN#ITLIHPK
		LN#YTLSQGHR
166795301	prenylcysteine oxidase 1 precursor [Homo sapiens]	GELN#TSIFSSRPIDK
167466198	intercellular adhesion molecule 1 precursor [Homo sapiens]	LNPTVITYGN#DSFSAK
167857790	alpha-1-acid glycoprotein 1 precursor [Homo sapiens]	QDQCIYN#TTYLNVQR

		EN#GTISR
168229159	ADP-ribosyl cyclase/cyclic ADP-ribose hydrolase 2 precursor [Homo sapiens]	NKN#CTAIWEAFK
169790833	lysosome-associated membrane glycoprotein 2 isoform C precursor [Homo sapiens]	VQPFN#VTQGK
		VASVININPN#TTHSTGSCR
178557739	complement C4-B preproprotein [Homo sapiens]	GLN#VTLSSSTGR
		FSDGLESN#SSTQFEVKK
		FSDGLESN#SSTQFEVK
182508169	neuropilin-1 isoform a precursor [Homo sapiens]	IGYSNN#GSDWK
189163542	alpha-1-antitrypsin precursor [Homo sapiens]	YLGN#ATAIFFLPDEGK
189242612	complement component C6 precursor [Homo sapiens]	VLN#FTTK
		KLECNGEN#DCGDNSDERDCGR
		KLECNGEN#DCGDNSDER
190341024	SPARC-like protein 1 isoform 1 precursor [Homo sapiens]	N#YSHHQLNR
205277441	thyroxine-binding globulin precursor [Homo sapiens]	VTACHSSQPN#ATLYK
223633991	pantetheinase precursor [Homo sapiens]	MTGSGIYAPN#SSR
225579152	insulin-like growth factor-binding protein complex acid labile subunit isoform 1 precursor [Homo sapiens]	YLSLRN#NSLR
256217721	carboxypeptidase N subunit 2 precursor [Homo sapiens]	AFGSNPN#LTK
310923208	microfibril-associated glycoprotein 4 isoform 1 precursor [Homo sapiens]	FN#GSVSFFR
322302700	extracellular matrix protein 1 isoform 3 precursor [Homo sapiens]	HIPGLIHN#MTAR
324021745	vitamin D-binding protein isoform 3 precursor [Homo sapiens]	LCDN#LSTK
333944015	cadherin-13 isoform 2 [Homo sapiens]	IN#NTHALVSLLQNLNK
355594753	clusterin preproprotein [Homo sapiens]	LAN#LTQGEDQYYLR
		KKEDALN#ETR
		KEDALN#ETR
		EIRHN#STGCLR
		EDALN#ETR
		HN#STGCLR
		EDALN#ETRESETK
514239916	complement component C8 beta chain	EYESYSDFERN#VTEK

isoform 1 preproprotein [Homo
sapiens]

The N-glycosylation sites are marked with N#.