

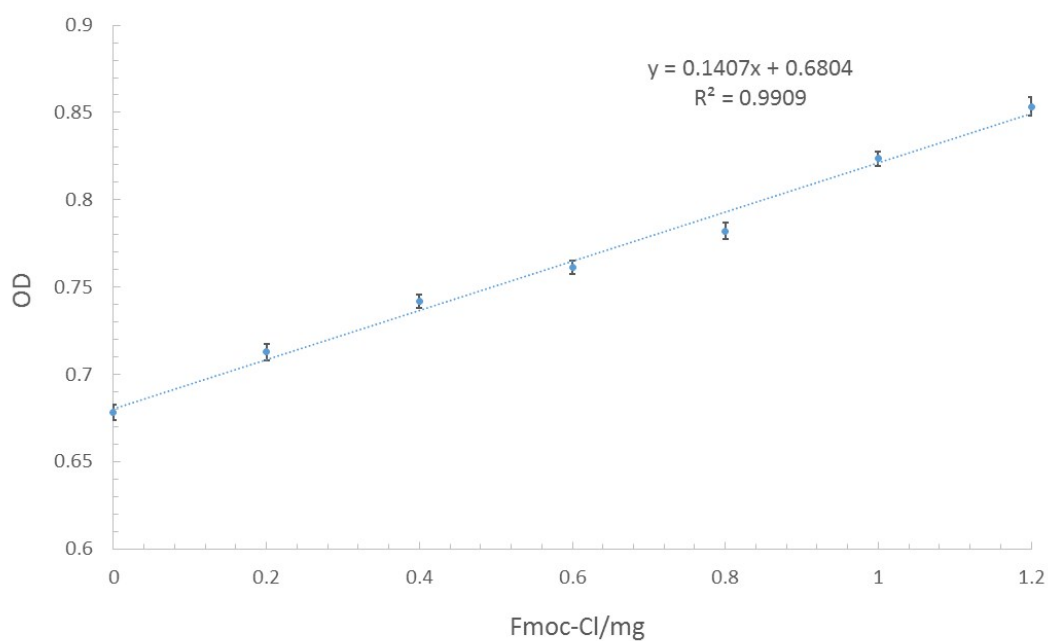
## Supporting Information

### **Optimized Conditions for Enrichment of N-Linked Glycopeptides with Amine-Functionalized Mesoporous Silica Nanoparticles**

The peptides were first dissolved in a sodium acetate buffer (100 mM sodium acetate and 150 mM NaCl, pH = 5.5). Then the peptides were oxidized at R. T. in the dark with 10 mM sodium periodate. Sodium sulfite was added to the solution until a final concentration of 20 mM after 1 hour of incubation. The mixture was incubated for another 10 min with shaking at R. T. in the dark. The oxidized samples were lyophilized and redissolved in the coupling solution of 70% methanol and 30% acetic acid (v/v) with a final concentration of 0.5 mg/mL. Amine-functionalized nanoparticles were prewashed by the coupling solution and then added to the peptide sample with a final concentration of 2 mg/mL. After shaking for 0.5 hour at room temperature, sodium cyanoborohydride was added to the reaction system with a final concentration of 1 M. The suspension was under shaking for another 0.5 hour at room temperature. After incubation, nanoparticles were washed by the following solutions sequentially twice: coupling solution, water, 80% ACN/20% H<sub>2</sub>O (v/v), 50 mM NH<sub>4</sub>HCO<sub>3</sub>. The as-cleaned nanoparticles were re-dispersed in 50mM NH<sub>4</sub>HCO<sub>3</sub> aqueous solution with a concentration of 4 mg/mL. At last, PNGase F (500 units per μL) were added until a concentration of 1 U/mL and the suspension was kept under shaking at 37 °C for 3 hours. The supernatant was collected through centrifugation for MALDI-MS analysis.

**Table S1.** Result of Fmoc UV assay

	Sample 1	Sample 2	Sample 3
SBA-15-APTES-Fmoc/ $\times 10^{-3}$ g	6.52	8.64	12.56
Average OD	0.7749	0.8023	0.8632
$-\text{NH}_2/\times 10^{-8}$ mol	259.5	334.8	502.3
Content of $-\text{NH}_2/\times 10^{-5}$ mol $\cdot$ g $^{-1}$	39.80	38.75	39.99



**Fig S1.** The standard curve of Fmoc UV assay

**Table S2.** List of identified glycoproteins from 5  $\mu$ L human serum after solid phase extraction with amino-functionalized SBA-15 after three parallel runs, N# denotes the N-linked glycosylation site.

No.	Protein	Description	N-linked deglycosylated Peptide sequence
1	P00450	Ceruloplasmin	EN#LTAPGSDSAVFFEQGTTR EHEGAIYPDN#TTDFQR
2	A2VEC9	SCO-spondin	LDSCNN#CTCVSGK
3	O43520	Probable phospholipid-transporting ATPase IC	N#TSFPLDADKILLR
4	O75882	Attractin	IDSTGN#VTNELR
5	P00738	Haptoglobin	MVSHHN#LTTGATLINEQWLLTTAK VVLHPN#YSQVDIGLIK
6	P00450	Ceruloplasmin	EN#LTAPGSDSAVFFEQGTTR
7	P01008	Antithrombin-III	LGACN#DTLQQLMEVFK
8	P01011	Alpha-1-antichymotrypsin	YTGN#ASALFILPDQDK TLN#QSSDELQLSMGNAMFVK FN#LTETSEAEIHQSFQHLLR
9	P02750	Leucine-rich alpha-2-glycoprotein	MFSQN#DTR
10	P00739	Haptoglobin-related protein	MVSHHN#LTTGATLINEQWLLTTAK NLFLN#HSEN#ATAK
11	P02763	Alpha-1-acid glycoprotein 1	QDQCIYN#TTYLNVQREN#GTISR
12	P20851	C4b-binding protein beta chain	LGHCPDPVLVNGEFSSSGPVN#VSDK
13	P01009	Alpha-1-antitrypsin	YLG N#ATAIFFLPDEGK ADTHDEILEGLNFN#LTEIPEAQIHEGFQEL LR
14	P01024	Complement	TVLTPATNHMGN#VTFTIPANR

15	P01042	Kininogen-1	ITYSIVQTN#CSK LNAENN#ATFYFK
16	P01616	Ig kappa chain V-II region MIL	FSGSGSGTN#FTLK
17	P01833	Polymeric immunoglobulin receptor	VPGN#VTAVLGETLK
18	P01861	Ig gamma-4 chain C region	EEQFN#STYR
19	P01871	Ig mu chain C region	YKN#NSDISSTR GLTFQQN#ASSMCVPDQDTAIR
20	P01876	Ig alpha-1 chain C region	LAGKPTHVN#VSVVMAEVDGTCY LSLHRPALEDLLLGSEAN#LTCTLTGLR
21	P01877	Ig alpha-2 chain C region	TPLTAN#ITK
22	P02745	Complement C1q subcomponent subunit	RNPPMGGNVVIFDTVITNQEEPYQN#HSG R
23	P02749	Beta-2-glycoprotein 1	LGN#WSAMPSCK DTAVFECLPQHAMEFGN#DTITCTTHGN#W TK
24	P02751	Fibronectin	LDAPTNLQFVN#ETDSTVLVR DQCIVDDITYNVN#DTFHK
25	P02765	Alpha-2-HS-glycoprotein	KVCQDCPLLAPLN#DTR AALAAFNAQNN#GSNFQLEEISR VCQDCPLLAPLN#DTR
26	P02790	Hemopexin	N#GTGHGN#STHHGPEYMR ALPQPQN#VTSLLGCTH
27	P03952	Plasma kallikrein	IYPGVDFGGEELN#VTFVK
28	P04004	Vitronectin	NN#ATVHEQVGGPSLTSDLQAQSK
29	P04114	Apolipoprotein	FN#SSYLQGTNQITGR
30	P04196	Histidine-rich glycoprotein	VIDFN#CTTSSVSSALANTK
31	P04745	Alpha-amylase	INGN#CTGIK

32	P05090	Apolipoprotein D		ADGTVNQIEGEATPVN#LTEPAKLEVK
33	P05155	Plasma protease inhibitor	C1	DTFVN#ASR VGQLQLSHN#LSLVILVPQNLK
34	P05546	Heparin cofactor 2		DFVN#ASSKYEITTIHNLFR
35	P06681	Complement C2		LTDTICGVGN#MSAN#ASDQER
36	P08185	Corticosteroid-binding globulin		AQLLQGLGFN#LTER
37	P08603	Complement factor H		ISEEN#ETTCYMGK IPCSQPPQIEHGTIN#SSR MDGASN#VTCINSR
38	P0C0L4	Complement C4-A		FSDGLESN#SSTQFEVK GLN#VTLSSSTGR
39	P10909	Clusterin		HN#STGCLR MLN#TSSLLEQLNEQFNWVSR LAN#LTQGEDQYYLR
40	P17936	Insulin-like growth factor-binding protein 3		GLCVN#ASAVSR
41	P19652	Alpha-1-acid glycoprotein 2		EN#GTVSR
42	P19827	Inter-alpha-trypsin inhibitor heavy chain H1		AN#LSSQALQMSLDYGFVTPLTMSIR
43	P22792	Carboxypeptidase subunit 2	N	LYLGSNN#LTALHPALFQN#LSK
44	P25311	Zinc-alpha-2-glycoprotein		DIVEYYN#DSNGSHVLQGR DIVEYYN#DSN#GSHVLQGR
45	P27169	Serum paraoxonase/arylesterase 1		VTQVYAEN#GTVLQGSTVASVYK
46	P36955	Pigment epithelium-derived factor		VTQN#LTLIEESLTSEFIHDIDR

47	P36980	Complement factor H-related protein 2	H-	LQNNENN#ISCVER
48	Q03591	Complement factor H-related protein 1	H-	LQNNENN#ISCVER
49	Q08380	Galectin-3-binding protein		ALGFEN#ATQALGR
50	Q14624	Inter-alpha-trypsin inhibitor heavy chain H4		LPTQN#ITFQTESSVAEQEAEFQSPK
51	Q6T4R5	Nance-Horan syndrome protein		N#STFDVKNR
52	Q86U70	LIM domain-binding protein 1		RKMSGGSTMSSGGGNTN#NSNSK
53	Q8NE71	ATP-binding cassette sub-family F member 1		QAMLEN#ASDIK
54	Q9NY74	Ewing's tumor-associated antigen 1		FLGATN#LTMYSK
55	Q9QSQ7	Envelope glycoprotein gp160		DGGN#ESNIETFRPEGGMK
56	Q96PD5	N-acetylmuramoyl-L-alanine amidase		LEPVHLQLQCMSQEQLAQAAN#ATK

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