

Supporting Information

Ligand-Free Strategy for Ultrafast and Highly Selective Enrichment of Glycopeptides Using Ag-Coated Magnetic Nanoarchitectures

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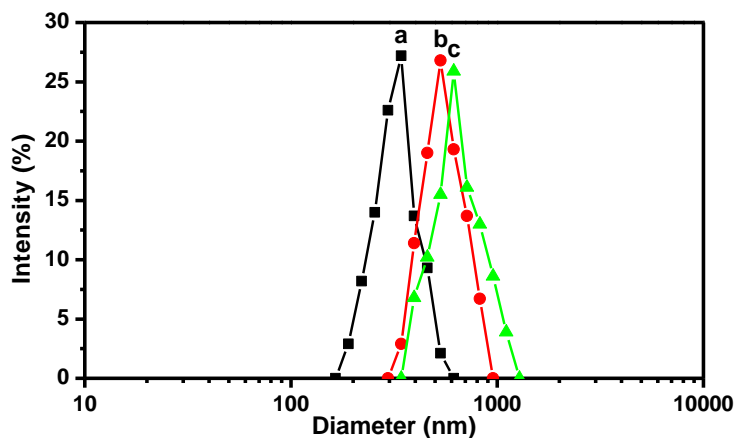


Fig. S1 Hydrodynamic diameter distribution of (a) MCNCs, (b) MCNC@PMAA, and (c) MCNC@PMAA@Ag-NPs.

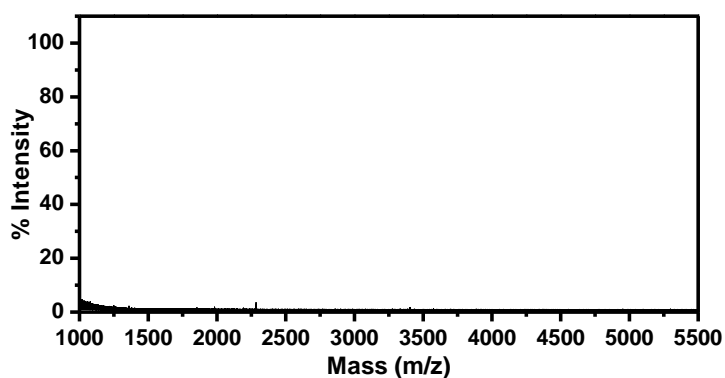


Fig. S2 MALDI mass spectrum of the tryptic digest of HRP after enrichment by MCNC@PMAA.

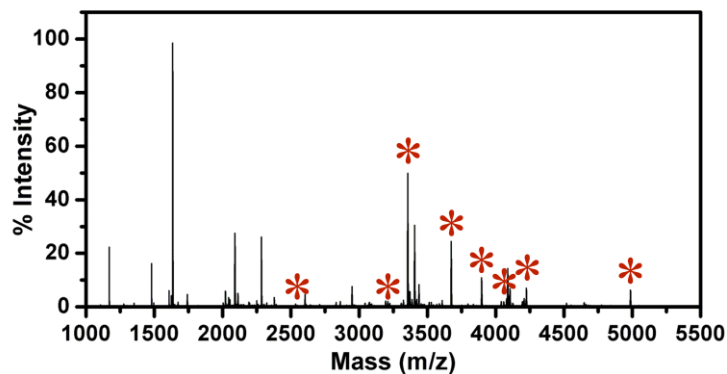


Fig. S3 MALDI mass spectrum of the tryptic digest mixture of HRP and MYO (with a molar ratio of HRP to MYO of 1:100) after enrichment by MCNC@PMAA@Ag-NPs. (Glycopeptide peaks are labeled with “*”).

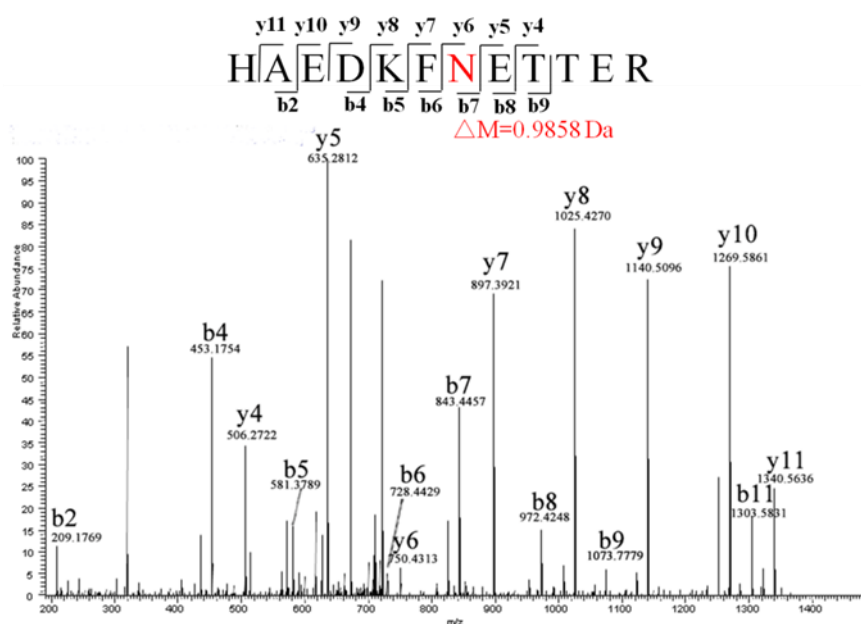


Fig. S4 Representative MS/MS spectrum of the glycopeptide HAEDKFNETTER, N represents deamidated Asn with the mass increment of 0.9858 Da.

Table S1. Overview of observed glycopeptides derived from tryptic digests of HRP after enrichment with the composite microspheres. N# denotes the N-linked glycosylation site.

m/z	glycan composition	amino acid sequence
2531.9	FucGlcNAc ₁	SFAN#STQTFNFVAFVEAMDR
2850.4	FucGlcNAc ₁	GLIQSDQELFSSPN#ATDTIPLVR
3207.4	XylMan ₃ GlcNAc ₂	SFAN#STQTFNFVAFVEAMDR
3353.4	XylMan ₃ FucGlcNAc ₂	SFAN#STQTFNFVAFVEAMDR
3526.4	XylMan ₃ GlcNAc ₂	GLIQSDQELFSSPN#ATDTIPLVR
3671.5	XylMan ₃ FucGlcNAc ₂	GLIQSDQELFSSPN#ATDTIPLVR
3893.4	XylMan ₃ FucGlcNAc ₂	LHFHDCFVNGCDASILLDN#TTSFR
4056.5	XylMan ₃ GlcNAc ₂	QLTPTFYDNSC(AAVESACPR)PN#VS NIVR-H ₂ O
4221.6	XylMan ₃ FucGlcNAc ₂	QLTPTFYDNSC(AAVESACPR)PN#VS NIVR
4838.4	XylMan ₃ FucGlcNAc ₂ ,XylMan ₃ GlcNAc ₂	LYN#FSNTGLPDPTLN#TTYLQTLR
4983.0	XylMan ₃ FucGlcNAc ₂ ,XylMan ₃ FucGlcNAc ₂	LYN#FSNTGLPDPTLN#TTYLQTLR

Table S2. The detailed information of the 127 unique glycopeptides mapped to 51 different glycoproteins.

No.	Swissprot	Description	Peptide sequence	N position	Annotation
1	P01026	Complement C3	GLSSDLWGEKPN#TSYIIGK	1617	P
2	P01048	T-kininogen 1	EGN#CSVQSGLTWQDCDFK	82	p
			IIYSIVQTN#CSK	204	p
			EGN#CSVQSGLTWQDCDFKDAEEAATGECTTTLGK	82	p
			HAVEHFNN#NTK	168	p
3	P02764	Alpha-1-acid glycoprotein	EFQTDDQCVYN#FTHLGVQR	94	p
			LIN#DTIELR	76	n
			PEPANITLGIPITN#ETLK	34	p
			HGTFMLAFN#LTDENR	134	p
			N#FTHLGVQR	94	p
4	P04937	Fibronectin	DQCIVDDITYNVN#DTFHK	528	p
			LDAPTNLQFVN#ETDR	1006	p
5	P05371	Clusterin	QELN#DSLQVAER	327	Probable
			QLEEFNL#QSSPF	144	Probable
			AFPEVCN#ETMMALWEECKPCLK	102	Probable
6	P06866	Haptoglobin	LLTTAQNLFN#HSEN#ATAK	148,152	p
			HGLTTGATLISDQWLLTTAQNLFN#HSEN#ATAK	152	p
7	P08649	Complement C4	ALN#VTLSSMGR	1323	p
			N#TTCQDLR	1386	p
8	P08932	T-kininogen 2	IIYSIVQTN#CSKEDFPFLR	204	p
9	P08934	Kininogen-1	EGN#CSVQSGFAWQDCDFK	82	p
			EALGHSIAQLNAENN#HTFYFK	294	p
10	P09006	Serine protease inhibitor A3N	LEDLTPYVRDEELN#CTVVELK	258	p
			YTGN#ASALFILPDQGK	269	p
			SMEEILEGLKFN#LTETPETEHR	104	p
11	P10959	Liver carboxylesterase 1	LDN#TSMSTVIDGVLPK	302	p
			N#TTTYPPMCSQDGVVGK	79	p
			NTQAVAQMIATLSGCN#N#TSSAAMVQCLR	274,275	p
			RFSPNLN#ISESVIPAIEK	375	p
12	P10960	Sulfated glycoprotein 1	DN#ATEEEILHYLEK	456	p
			TN#SSFVQGLVDHVK	214	p
			TVVTEAGNLLKDN#ATEEEILHYLEK	80	p
13	P12346	Serotransferrin	FDEFFSQGCAPGYKKN#STLCDLCIGPAK	512	p
	P13635	Ceruloplasmin	ANEGAIYPDND#TTDFQR	138	p
			DWEMELHHLQEQN#VSN AFLDKKEEFFIGSK	756	p
14	P14046	Alpha-1-inhibitor 3	DAGYTN#ATTTDQHGLAK	393	p
			MHQAFHVN#ATVTEEGTGSEFSGSGR	321	p
			YLN#ETQQLTEK	994	p
			ICLHLYHLN#ETVTVTASLISQR	55	p
			LDNNGCSTQEVN#ITEFQLK	301	p
			ALSCLESSWENIEQGGN#GSFVYTK	1143	p
			VPNAISVYDEIIN#VTACATYTYGK	247	p
			SLGNVN#FSVSAEAR	872	p
			VPNAISVYDEIIN#VTACATYTYGKPVPGHVK	247	p

			TEVTTNNVLLYLDQVTN#QTLFSFIIQQDIPVK	1427	p
			NYFPETWIWDLVTVN#SSGVTELEMTVPDTITIEWK	750	p
15	P14272	Plasma kallikrein	LQTPLN#YTEFQKPICLPSK	494	probable
			IYGGILN#LSEITNK	453	probable
16	P17475	Alpha-1-antiproteinase	QILEGLEFN#LTQIPEADIHK	101	p
			SSWVLMMDYLG#ATAIFLLPDDGK	265	p
		Alpha-1-antiproteinase	ELVHQS#TSNIFSPMSITTAFAMLSLGSK	64	p
17	P20059	Hemopexin	SWPAVGN#CTAALR	186	p
			GEN#GTKPDSVDIEHCSDAWSFDATMDHN#GTMLFFK	38	p
18	P20759	Ig gamma-1 chain C region	SWFVDDVEVHTAQTRPPEEQFN#STFR	176	p
19	P20762	Ig gamma-2C chain C region	VFTAQTPHEEQLN#GTFR	179	n
20	P24090	Alpha-2-HS-glycoprotein	RPFGEVYELEIDTLETTCHALDPTPLAN#CSVR	99	p
			TALAAFNAQNN#GTYFK	176	p
			FN#DTNVVHTVK	156	p
21	P26644	Beta-2-glycoprotein 1	TGN#WSALPCK	205	p
		Beta-2-glycoprotein 1	TSVGN#SSFYQDTPVFK	114	p
		Beta-2-glycoprotein 1	CLPHFAMFGNDTVTCTAHGN#WTQLPECR	145	p
		Beta-2-glycoprotein 1	YTTFEYPNTIGFACNPGYYLN#GTSSSK	169	p
22	P31211	Corticosteroid-binding globulin	EEDFYVN#ETSTVK	216	p
		Corticosteroid-binding globulin	AMLQLDEGNVLPN#STNGAPLHLR	352	p
23	P35859	Insulin-like growth factor-binding protein complex acid labile chain	N#LTHLPDDIPVSTR	64	p
24	P36953	Afamin	HAEDKFN#ETTER	402	p
			CQAYKN#NSESFLNLY	153	p
			LPTKPQDQVDHFN#ATQK	33	p
25	P51886	Lumican	AFEN#VTDLQWLILDHNLLENSK	88	p
26	P55159	Serum paraoxonase/arylesterase 1	EVPVLDLPN#CTLVK	41	n
			VTVVYAEN#GTVLQGTVAAYVK	324	p
27	P55314	Complement component C8 beta chain	YEFALTEYESYDFEHN#VTEK	242	p
28	Q03626	Murinoglobulin-1	AQEAGYTN#ATTTDQHGLAK	393	p
			ALGCLEASWETIEQGRN#GSFVYTK	1153	p
			VPNAISVYDEIIN#VTACAIY	247	p
			EEDSELDNNGCSTQEVN#ITEFQLK	301	p
			SLGNVN#FVSVEAQSPCLCGSQVATVPETGRK	882	p
			SACKEEDSELDNNGCSTQEVN#ITEFQLK	301	p
			NYFPETWIWDLVTVN#SSGVTEVEMTVPDTITIEWK	760	p
29	Q62930	Complement component C9	VGVEVSPEKN#SSKPK	261	p
30	Q62975	Protein Z-dependent protease inhibitor	LPYQGN#ATMLVVLMEK	287	p
31	Q63041	Alpha-1-macroglobulin	LTN#QTMGFSFAVEQDIPVK	1448	p
			N#LTVQVN#SVR	382	p
			SLGEVN#FTATAEALQSPCLCGNK	883	p
			PVN#ETFPVVYIENPKR	157	p
			MVLFVPNIYVLEYLN#ETQQLTEAIK	1005	p
			N#LTVQVNSVR	382	p
			VVSVDISFRPVN#ETFPVVYIENPK	157	p
			CFANTVN#LSFPSAQLSPASDTHLTVK	568	p
32	Q63416	Inter-alpha-trypsin inhibitor heavy chain H3	GDEKEN#ITAEALELSLK	580	p

			TAFITN#FTLTIDGVTPGSKV	91	p
33	Q63556	Serine protease inhibitor A3M (Fragment)	FPDGTGLGN#DTLLHK	21	n
			YTGN#SSALFILPDK	262	p
			FN#LTESYETDIHQFGHLLQR	97	p
34	Q63621	Interleukin-1 receptor accessory protein	PTLLN#DTGN#YTCMLR	117,118	p
35	Q64240	Protein AMBP	EDSCQLN#YSEGPCLMQK	233	p
36	Q6P734	Plasma protease C1 inhibitor	DTYVN#ASLSLYGSSPR	243	p
			VGQLQLSHN#LSFVIMVPQSPHQLEDMEK	356	p
37	Q7TMA5	Apolipoprotein B-100	DLEMVN#ISLAR	2662	p
			LPQQIHDYLN#ASDWER	2094	p
			LSTIDNIYIPAMGN#FTYDFSFK	3299	p
			ALN#LTMLPK	3715	p
38	Q8R2H5	Phosphatidylinositol-glycan-specific phospholipase D	LSSSPN#ITISCK	502	p
			VN#GTLTQVLLVGAPTHDDVSK	661	p
			N#LTMFISK	308	p
39	Q99J86	Attractin	VFHIHN#ESWVLLTPK	431	p
			IDSTGN#VTNELR	419	p
40	Q9EQV9	Carboxypeptidase B2	EVHFFVN#ASDVNSVK	72	p
			IPFNVLMMNVEDLIQQQTSN#DTVSPR	107	p
41	Q9JJS8	Mannan-binding lectin serine protease 2	APGN#DTFYSLGPSLK	103	Ref
			GDSGGALVFLDN#ETQR	641	p
42	Q9QX79	Fetuin-B	VLYLPAYN#CTLRPVSK	139	p
43	Q9WUW3	Complement factor I	FN#VSLIYGSTDTEGIVQVK	116	p
			IN#STECLHVR	182	p
			WGEVDLIGN#CSR	515	p
			FSNN#GTCTAEK	106	p
			DVNN#VTYVWGVISWGENCGKPEFPGVYTR	557	p
44	P05545	Serine protease inhibitor A3K	FN#LTEITEEEIHQFGHLLQR	102	p
			KFINDYVSN#QTQGK	182	p
			VPFNPN#DTFESEFYLDEK	220	p
45	P05544	Serine protease inhibitor A3L	LINDYVSN#QTQGK	182	p
46	Q6IE52	Murine globulin-2	YLN#ETQQLTENIK	968	p
47	P18292	Prothrombin	ITDNMFCAQFVN#DTK	552	p
48	Q5FVF9	Biotinidase	GAHIIVFPEDGIHGFN#FTR	97	n
49	P20761	Ig gamma-2B chain C region	EEQYN#STFR	183	n
50	Q63514	C4b-binding protein alpha chain	CGPPDLPYALPASEMN#QTDIESHTTLR	31	p
51	Q6IRK9	Plasma glutamate carboxypeptidase	EVMSLLQPLN#ITK	396	p