

Electronic Supplementary Information

Protein Profiling by Protein Imprinted Polymer Array

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Protein extraction (%)

	Cyt-MIP	Rib-MIP	Lac-MIP
AA based-MIP	80.5	99.3	92.5
DMA based MIP	88.4	99.1	92.0

Total conversion data

Total conversions of monomers in the obtained polymers were about 90 %. Reproducibility of the total conversion of Cyt-MIP_{AA} was examined by five independent preparation of Cyt-MIP_{AA}: average 91 % (91%, 89%, 93%, 93%, 88%), C.V.=2.5%.

Protein-MIP_{AA}

	Cyt-MIP _{AA}	Rib-MIP _{AA}	Lac-MIP _{AA}	NIP _{AA}
protein	Cyt 7.5 μ mol	Rib 7.5 μ mol	Lac 7.5 μ mol	-
AA	10 μ l (0.15mmol)	10 μ l (0.15mmol)	10 μ l (0.15mmol)	10 μ l (0.15mmol)
GEMA	395mg (1.35mmol)	395mg (1.35mmol)	395mg (1.35mmol)	395mg (1.35mmol)
MBAA	231mg (1.5mmol)	231mg (1.5mmol)	231mg (1.5mmol)	231mg (1.5mmol)
Polymer obtained	579mg	553mg	550mg	583mg
conversion	91%	87%	86%	92%

Protein-MIP_{DMA}

	Cyt-MIP _{DMA}	Rib-MIP _{DMA}	Lac-MIP _{DMA}	NIP _{DMA}
protein	Cyt 7.5 μ mol	Rib 7.5 μ mol	Lac 7.5 μ mol	-
DMA	25 μ l (0.15mmol)	25 μ l (0.15mmol)	25 μ l (0.15mmol)	25 μ l (0.15mmol)
GEMA	395mg (1.35mmol)	395mg (1.35mmol)	395mg (1.35mmol)	395mg (1.35mmol)
MBAA	231mg (1.5mmol)	231mg (1.5mmol)	231mg (1.5mmol)	231mg (1.5mmol)
Polymer obtained	542mg	582mg	535mg	547mg
conversion	85%	91%	84%	86%

Specific surface area and pore size

The specific surface area and pore size of the AA-based polymer were estimated to be 2.36 m²/g (BET method) and <5 nm pores (DFT method) by nitrogen gas adsorption/desorption.

The specific surface area and pore size of the DMA-based polymer were estimated to be 2.71 m²/g (BET method) and <5 nm pores (DFT method) by nitrogen gas adsorption/desorption.

IR data of Cyt-MIP

AA-based polymers; (KBr pellet, cm⁻¹): 3740-3130 (ν_{O-H}), 3030-2830 (ν_{C-H}), 1730 and 1680 (ν_{C=O}), 1050 (ν_{C-O}). Rib-MIP; (KBr pellet, cm⁻¹): 3720-3140 (ν_{O-H}), 3030-2850 (ν_{C-H}), 1725 and 1670 (ν_{C=O}), 1050 (ν_{C-O}). Lac-MIP; (KBr pellet, cm⁻¹): 3730-3120 (ν_{O-H}), 3020-2840 (ν_{C-H}), 1720 and 1660 (ν_{C=O}), 1040 (ν_{C-O}). NIP; (KBr pellet, cm⁻¹): 3720-3130 (ν_{O-H}), 3030-2840 (ν_{C-H}), 1730 and 1670 (ν_{C=O}), 1050 (ν_{C-O}).

DMA-based polymers; (KBr pellet, cm⁻¹): 3720-3130 (ν_{O-H}), 3030-2830 (ν_{C-H}), 1730 and 1670 (ν_{C=O}), 1050 (ν_{C-O}). Rib-MIP; (KBr pellet, cm⁻¹): 3720-3120 (ν_{O-H}), 3030-2820 (ν_{C-H}), 1730 and 1670 (ν_{C=O}), 1050 (ν_{C-O}). Lac-MIP; (KBr pellet, cm⁻¹): 3730-3130 (ν_{O-H}), 3030-2820 (ν_{C-H}), 1730 and 1670 (ν_{C=O}), 1050 (ν_{C-O}). NIP; (KBr pellet, cm⁻¹): 3720-3120 (ν_{O-H}), 3030-2820 (ν_{C-H}), 1730 and 1670 (ν_{C=O}), 1050 (ν_{C-O}).

Multivariate analysis (Principal Component Analysis)

PCA was performed by Multivariate Analysis Add-in for Excel v1.2 (Bristol Centre for Chemometrics). The results are as follows.

AA					
Analyte		Bound (mg/g-polymer)			
		Cyt-MIP	Rib-MIP	Lac-MIP	NIP
Cyt	Run2	15.64	11.86	9.42	7.79
Cyt	Run3	16.52	11.27	9.17	7.94
Cyt	Run4	16.08	10.88	8.58	7.50
Cyt	Run5	17.01	11.57	9.12	7.21
Rib	Run1	7.45	17.00	7.71	15.89
Rib	Run3	7.60	15.80	6.64	18.73
Rib	Run4	7.40	16.24	7.78	18.09
Rib	Run5	7.77	16.90	8.78	18.48
Lac	Run1	8.06	9.32	11.33	4.42
Lac	Run2	8.88	9.21	11.26	4.05
Lac	Run4	8.17	8.89	10.15	4.58
Lac	Run5	8.22	9.27	11.26	3.97
Alb	Run1	0.56	1.20	2.29	8.56
Alb	Run2	0.63	0.97	1.98	9.24
Alb	Run4	0.56	1.09	1.87	9.76
Alb	Run5	0.67	1.30	2.13	8.97
Myo	Run1	5.57	5.40	6.40	14.87
Myo	Run2	5.70	5.25	6.43	15.50
Myo	Run3	5.89	3.32	6.70	15.34
Myo	Run4	5.40	3.11	7.00	15.80

PCA - Principal Components Analysis (Standardised)						
N. PCs	Eigenvalues		Scores			Loadings
	E-value	%				
Total SS	80	100	1.743912	-0.4806	0.636902	0.588117 -0.09233 0.787461
#1	46.95765	58.69706	1.734354	-0.50824	0.846627	0.530712 0.450158 -0.20396
#2	23.85602	29.82003	1.540014	-0.58607	0.894821	0.59693 -0.12203 -0.57158
#3	5.584652	6.980815	1.828692	-0.61884	0.904921	-0.12703 0.879739 0.107724
			0.760497	1.577294	-0.32692	
			0.377683	2.012198	0.004102	
			0.638655	1.894961	-0.27233	
			0.934105	1.971366	-0.42103	
			1.07948	-1.2173	-0.87114	
			1.160102	-1.303	-0.73495	
			0.814384	-1.18127	-0.61342	
			1.091595	-1.30042	-0.84081	
			-2.47014	-0.67223	0.069916	
			-2.56239	-0.56172	0.162008	
			-2.59348	-0.45538	0.178522	
			-2.48901	-0.58808	0.121665	
			-0.82711	0.520651	0.04468	
			-0.83752	0.614418	0.078927	
			-0.94688	0.411572	0.126023	
			-0.97693	0.470716	0.011482	

DMA					
Analyte		Bound (mg/g-polymer)			
		IP-Cyt	IP-Rib	IP-Lac	BP
Cyt	Run2	7.77	3.79	3.48	3.66
Cyt	Run3	6.80	3.79	2.65	2.17
Cyt	Run4	7.65	3.97	2.89	3.61
Cyt	Run5	8.01	3.67	3.61	2.89
Rib	Run1	5.34	15.38	10.72	12.43
Rib	Run3	5.15	15.66	11.10	12.60
Rib	Run4	5.23	13.68	10.88	12.86
Rib	Run5	4.90	15.64	10.65	12.00
Lac	Run1	7.69	6.26	12.60	9.96
Lac	Run2	8.25	6.78	12.75	9.63
Lac	Run3	7.53	6.73	14.60	9.69
Lac	Run5	7.58	7.08	13.20	10.09
Alb	Run1	7.47	8.24	17.86	10.45
Alb	Run2	6.34	8.45	17.02	10.56
Alb	Run3	5.98	8.52	17.56	11.20
Alb	Run4	6.57	7.65	18.00	10.67
Myo	Run2	14.67	21.98	18.68	21.98
Myo	Run3	15.56	22.67	18.88	23.00
Myo	Run4	14.67	21.68	18.23	22.65
Myo	Run5	14.75	22.03	19.21	22.87

PCA - Principal Components Analysis (Standardised)						
N. PCs	Eigenvalues		Scores			Loadings
	E-value	%				
Total SS	80	100	-2.14023	1.026708	0.137292	0.453338 0.695566 0.540859
#1	62.39326	77.99157	-2.46927	0.951416	-0.04039	0.518873 0.091363 -0.67955
#2	10.80479	13.50599	-2.19486	1.080262	0.052292	0.458935 -0.70955 0.459922
#3	6.656153	8.320191	-2.17538	1.065789	0.221092	0.560923 -0.06613 -0.18481
			-0.18078	-0.31776	-1.10135	
			-0.13797	-0.40265	-1.13383	
			-0.2782	-0.38804	-0.94337	
			-0.26302	-0.39097	-1.19148	
			-0.64991	-0.17765	0.435348	
			-0.55207	-0.07159	0.49364	
			-0.49434	-0.45431	0.534347	
			-0.53897	-0.26564	0.380383	
			-0.04934	-0.86638	0.614805	
			-0.24152	-0.98862	0.342471	
			-0.18254	-1.13624	0.303485	
			-0.1833	-1.07808	0.538118	
			3.079117	0.568188	0.075284	
			3.358354	0.72283	0.131774	
			3.078028	0.614045	0.049746	
			3.216202	0.508692	0.10034	

Reproducibility of polymer preparation (Cyt-MIP)

Reproducibility of the polymer preparation was evaluated by the binding amounts of Cyt to 5 independently prepared Cyt-MIP. Five independently prepared Cyt-imprinted polymers were prepared with the same recipe in the text and the amounts of Cyt bound to the Cyt-MIP were determined as follows: Cyt (0.25 mg/mL) was dissolved in 50 mM phosphate buffer pH 7.4 (5 mL) containing a polymer (5 mg).

After 16 h incubation with rotation

at 25 °C, the mixtures were filtered to remove the polymer and the proteins in the filtrate were analyzed by the same HPLC described in the text. Bound protein amounts were calculated by subtracting the amount in the filtrate from the initial amount.

A coefficient of variation was calculated to be 2.3 % (n=5), suggesting that the MIPs can be prepared reproducibly and the MIP array could give us reliable data sets for the protein identification.

