Supporting Information for

Array-Based Sensing of Proteins Using Conjugated Polymers

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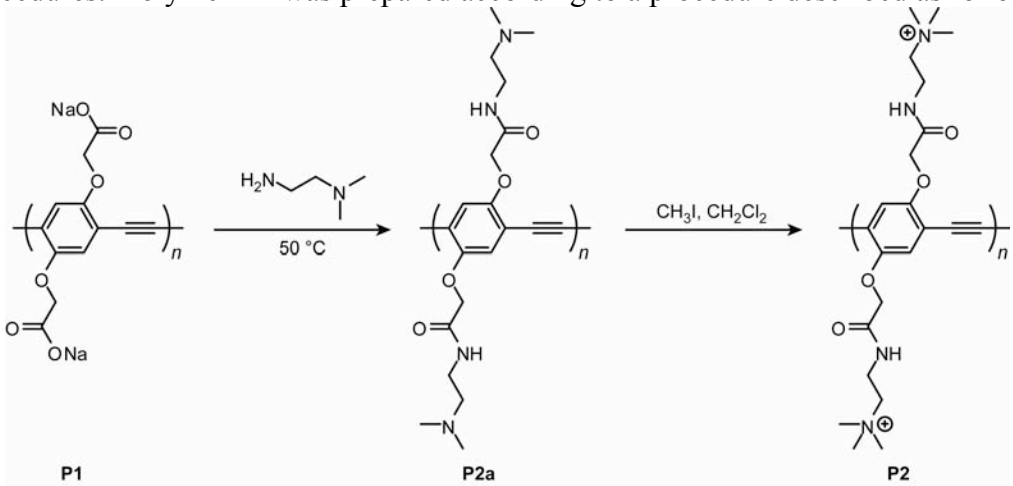
Experimental Section

Materials. α-Amylase (α-Am, from *Bacillus licheniformis*), bovine serum albumin (BSA), α-chymotrypsin (ChT, from bovine pancreas, type I-S), cytochrome *c* (CytC, from equine heart), ferritin (Fer, from equine spleen), β-galactosidase (β-Gal, from *Escherichia coli*), hemoglobin (Hem, from human), histone (His, from calf thymus, type III-S), human serum albumin (HSA), lipase (Lip, from *candida rugosa*, type VII), lysozyme (Lys, from chicken egg white), myoglobin (Myo, from equine heart), papain (Pap, from papaya latex), acid phosphatase (PhosA, from potato), alkaline phosphatase (PhosB, from bovine intestinal mucosa), ribonuclease A (RibA, from bovine pancreas, type I-A), and subtilisin A (SubA, from *Bacillus licheniformis*) were purchased from Sigma and used as received. Phosphate buffered saline (PBS, pH 7.4, 1×) was purchased from Invitrogen and used as the solvent throughout the fluorescence assays. Polymers **P1**,[1] **P3**,[2] and **P4**-**P6**[3] were synthesized according to the reported procedures. Polymer **P2** was prepared according to a procedure described as follows.



Scheme S1. Synthesis of cationic polymer **P2**.

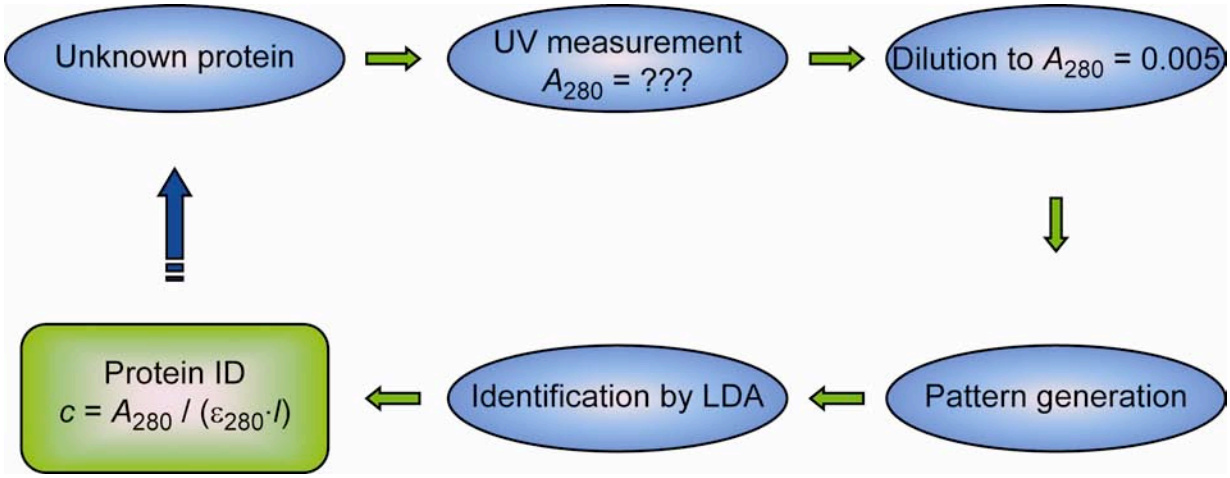
Polymer P2a: The mixture of polymer **P1** (25 mg, 0.082 mmol) and *N*,*N*-dimethylethylenediamine (25 mL) was stirred at 50 ºC for 24 h. After removal of the solvent, the residue was washed thoroughly with hexane. After being dried under vacuum, polymer **P2a** was obtained as a dark orange solid (52 mg, 0.077 mmol, 94 %). ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.14 (s, 2H), 4.52 (s, 4H), 3.31 (s, 4H), 2.81

(s, 4H), 2.17 (s, 12H). ^{13}C NMR (300 MHz, $CDCl_3$): δ 168.97, 157.57, 118.45, 113.49, 92.94, 90.67, 68.57, 58.10, 44.97, 36.44.

Polymer P2: Polymer **2a** (40 mg, 0.1 mmol) was dissolved in dichloromethane (25 mL) and iodomethane (15 mL) was added for methylation. The reaction mixture was stirred at room temperature overnight. After removal of the solvent, the polymer was washed with hexane and dried under vacuum. The product is a dark orange powder (63 mg, 0.094 mmol, 94%). ^{1}H NMR (300 MHz, D_2O): δ 7.09 (s, 2H), 4.50 (s, 4H), 3.74 (s, 4H), 3.52 (s, 4H), 3.27 (s, 18H). ^{13}C NMR (300 MHz, D_2O): δ 167.51, 156.13, 118.10, 113.54, 93.03, 90.86, 66.21, 65.33, 54.16, 36.38.

Methods. UV-vis spectra were measured in a rectangular quartz cuvette (light path = 10 mm) on a Hewlett Packard 8452A diode array spectrophotometer. Fluorescence spectra were recorded in a conventional quartz cuvette (10 × 10 × 40 mm) on a Jasco FP-6500 fluorimeter. The polymers were dissolved in phosphate buffered saline (1 ×) to make 100 nM of stock solutions on the basis of their molecular weights. In the protein sensing study, each polymer solution (100 nM, 200 μL) in PBS was respectively loaded into a well on a 96-well plate (300 μL Whatman® Glass Bottom microplate) and the fluorescence intensity values at 465 nm was recorded on a Molecular Devices SpectraMax M5 microplate reader with excitation at 430 nm. Subsequently, 10 μL of protein solution was added to each well and the final protein concentrations in the wells were A = 0.005 at 280 nm, which was initially calibrated using UV/vis spectroscopy. The fluorescence intensity values at 465 nm were recorded again. The difference between two reads before and after addition of proteins was treated as the fluorescence response. This process was repeated for 17 protein targets to generate six replicates of each. Thus, the 17 proteins were tested against the six-polymer array (**P1**-**P6**) six times, to afford a training data matrix of 6 polymers × 17 proteins × 6 replicates (ref. Table S1). The raw data matrix was processed using classical linear discriminant analysis (LDA) in SYSTAT (version 11.0). In LDA, all variables were used in the model (complete mode) and the tolerance was set as 0.001. The raw fluorescence response patterns were transformed to canonical patterns where the ratio of between-class variance to the within-class variance was maximized according to the preassigned grouping. The Mahalanobis distances of each individual pattern to the centroid of each group in a multidimensional space were calculated and the assignment of the case was based on the shortest Mahalonobis distance.

In the studies featuring unknown analyte protein concentrations, sixty-eight unknown protein solutions were randomly selected from the 17 protein species and subjected to an analysis procedure as illustrated in Scheme S2. The protocol included UV absorption measurement of protein samples at 280 nm, dilution to A_{280} = 0.005, fluorescence response pattern recording against the sensor array (**P1**~**P6**, 100 nM), and LDA. The new cases were classified to the groups generated through the training matrix (6 polymers × 17 proteins × 6 replicates) according to their shortest Mahalanobis distances. After the protein identity was recognized by LDA, the initial protein concentration (c) was deduced from the A_{280} value and corresponding molar extinction coefficient (ε_{280}) according to the Beer-Lambert law: $c = A_{280}/(\varepsilon_{280} \cdot l)$ (Table S3 for original data). In the experimental setup, the protein sample preparation, data collection and LDA analysis were performed by different persons.



Scheme S2. Schematic representation for the detection procedure of unknown proteins using array-based sensors.

(1) Kim, I.-B.; Dunkhorst, A.; Gilbert, J.; Bunz, U. H. F. *Macromolecules* **2005**, *38*, 4560-4562.
(2) Tan, C.-Y.; Pinto, M. R.; Schanze, K. S. *Chem. Commun.* **2002**, 446-447.
(3) Kim, I.-B.; Phillips, R. L.; Bunz, U. H. F. *Macromolecules* **2007**, DOI: 10.1021/ma070795v.

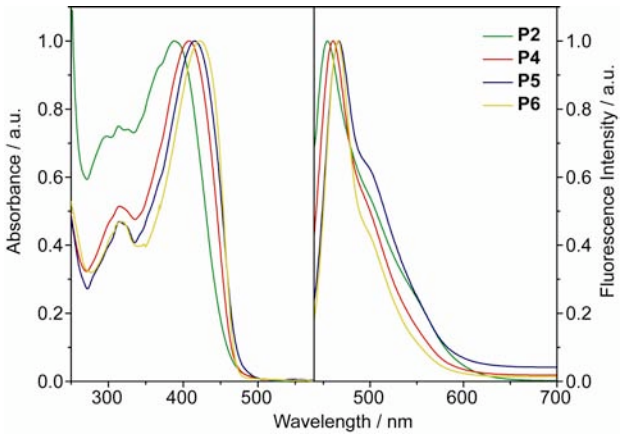


Figure S1. UV/vis and fluorescence emission spectra of conjugated polymers **P2**, **P4**, **P5**, and **P6** in PBS (pH 7.4). The excitation wavelength was 430 nm.

Table S1. Training Matrix of Fluorescence Response Patterns of PPE Sensor Array (**P1** – **P6**) Against Various Proteins with Identical Absorption Values of A = 0.005 at 280 nm.

Protein	**P1**	**P2**	**P3**	**P4**	**P5**	**P6**
α-Am	-141.689	-193.194	-15.466	-123.073	-135.605	-601.813
α-Am	-158.462	-181.726	-20.109	-115.962	-102.329	-520.650
α-Am	-154.865	-219.207	-15.642	-124.749	-131.664	-470.636
α-Am	-192.150	-214.235	-13.995	-128.604	-127.777	-543.929
α-Am	-199.672	-220.482	-18.554	-140.331	-128.792	-513.562
α-Am	-199.082	-228.261	-18.236	-131.178	-128.892	-446.606
BSA	213.277	-153.690	11.660	-88.343	-207.538	-750.476
BSA	284.778	-296.412	10.579	-88.882	-202.425	-773.870
BSA	299.881	-203.497	14.776	-79.468	-205.392	-694.852
BSA	298.518	-210.909	7.806	-68.319	-208.266	-784.255
BSA	310.192	-277.552	7.535	-87.684	-201.372	-810.050
BSA	303.641	-219.795	8.130	-58.529	-201.517	-808.743
ChT	-199.860	-192.310	-34.458	-160.788	-159.249	-641.747
ChT	-231.035	-186.575	-27.128	-177.833	-168.873	-589.265
ChT	-246.360	-227.314	-44.399	-180.100	-149.027	-543.333
ChT	-270.663	-254.628	-43.239	-178.590	-149.978	-592.137
ChT	-278.277	-226.102	-29.326	-178.060	-164.042	-652.320
ChT	-241.126	-224.725	-19.703	-184.200	-146.494	-543.265
CytC	-620.575	-170.756	-101.931	-280.652	-244.480	-858.686
CytC	-642.671	-193.748	-79.658	-274.883	-238.558	-858.861
CytC	-595.649	-204.236	-106.364	-265.394	-236.064	-794.694
CytC	-638.408	-219.283	-107.057	-261.689	-238.536	-878.212
CytC	-646.416	-189.995	-101.884	-263.673	-246.894	-863.988
CytC	-634.266	-202.221	-88.695	-287.188	-249.150	-905.274
Fer	-204.790	73.559	26.414	-86.839	-97.322	-367.614
Fer	-186.660	-86.968	21.038	-111.210	-95.382	-504.612
Fer	-187.125	22.110	-7.693	-96.493	-131.301	-548.468
Fer	-193.481	-111.773	2.669	-101.956	-90.267	-454.272
Fer	-211.882	94.118	2.809	-113.195	-103.113	-442.128
Fer	-187.427	-90.777	13.812	-101.199	-110.762	-600.072
β-Gal	348.929	-39.373	30.169	-72.885	-134.097	-404.059
β-Gal	346.984	-37.710	18.790	-74.803	-94.019	-361.810
β-Gal	336.811	-29.672	28.485	-44.826	-138.642	-327.500
β-Gal	342.446	-33.510	17.190	-54.269	-104.904	-370.782
β-Gal	344.958	-44.636	33.528	-69.320	-116.494	-385.750
β-Gal	302.395	-47.893	43.243	-52.503	-115.031	-395.621
Hem	-356.009	-246.625	-90.568	-238.971	-249.266	-979.830

Hem	-380.870	-329.243	-102.824	-250.558	-255.146	-920.807
Hem	-263.760	-266.048	-103.688	-233.923	-247.659	-903.936
Hem	-244.617	-258.521	-98.737	-240.795	-257.765	-877.392
Hem	-272.388	-271.081	-101.856	-249.450	-280.669	-905.413
Hem	-431.450	-255.928	-90.208	-265.975	-273.969	-876.136
His	-120.885	-189.531	2.176	309.030	-179.825	-669.412
His	-115.866	-174.040	2.247	359.820	-164.455	-613.784
His	-184.227	-220.205	3.370	335.786	-166.842	-689.833
His	-137.870	-233.411	-1.020	392.092	-173.855	-615.942
His	-137.851	-223.287	-4.126	308.271	-153.846	-683.530
His	-136.696	-217.466	5.425	355.298	-155.573	-666.808
HSA	-166.956	-313.811	-10.835	-206.007	-187.693	-865.609
HSA	-179.802	-260.686	-16.466	-170.667	-196.444	-735.332
HSA	-189.343	-235.524	-15.308	-206.404	-199.141	-731.155
HSA	-185.840	-283.917	-32.640	-215.115	-203.337	-810.859
HSA	-186.606	-302.867	-14.860	-219.830	-205.050	-705.139
HSA	-180.831	-269.950	-22.953	-224.517	-165.390	-715.520
Lip	-163.133	-88.981	38.564	-31.485	-201.059	-727.808
Lip	-191.148	-96.009	43.994	28.392	-190.689	-738.343
Lip	-202.713	-113.530	22.945	28.921	-197.574	-693.060
Lip	-161.390	-120.217	3.947	-7.151	-192.658	-778.290
Lip	-240.012	-122.978	33.178	-26.963	-221.690	-783.278
Lip	-187.111	-112.533	50.253	-36.303	-217.065	-769.396
Lys	-34.810	-96.478	-9.629	-110.054	-191.803	-830.871
Lys	-123.219	-156.686	0.539	-151.766	-191.354	-844.617
Lys	-62.738	-132.002	-1.148	-136.427	-195.743	-727.195
Lys	-292.354	-124.226	-3.932	-139.318	-202.180	-779.685
Lys	-260.238	-138.461	-11.833	-126.255	-191.801	-794.622
Lys	-268.321	-123.347	-3.691	-129.838	-173.123	-794.885
Myo	-223.919	-340.502	-64.173	-157.238	-194.821	-706.549
Myo	-255.898	-318.343	-64.709	-153.595	-200.330	-712.824
Myo	-311.824	-324.417	-65.231	-166.470	-210.297	-557.516
Myo	-282.382	-311.498	-73.533	-166.367	-218.299	-749.187
Myo	-325.775	-322.881	-63.397	-167.768	-223.494	-713.666
Myo	-311.053	-302.761	-64.301	-143.249	-207.223	-700.533
Pap	-418.560	-220.978	-17.847	-252.893	-212.157	-822.198
Pap	-425.792	-252.336	-2.145	-238.774	-246.838	-1019.127
Pap	-434.589	-218.509	-15.055	-231.009	-240.862	-996.012
Pap	-453.547	-195.680	-14.887	-240.099	-188.394	-977.860
Pap	-485.930	-65.817	-25.253	-234.677	-197.496	-982.076
Pap	-438.417	-229.094	-21.823	-249.522	-260.781	-984.072
PhosA	-28.124	-195.891	-38.787	-140.815	-152.478	-448.033
PhosA	-49.139	-201.727	-51.303	-140.870	-131.251	-477.898
PhosA	-53.841	-204.691	-52.975	-126.789	-140.601	-451.528
PhosA	-66.956	-212.469	-46.901	-114.366	-140.121	-527.754
PhosA	-72.710	-226.651	-44.792	-135.707	-145.671	-518.576
PhosA	-35.822	-214.351	-40.849	-113.215	-146.749	-512.660
PhosB	-336.756	-160.921	-61.755	-172.724	-153.730	-515.216
PhosB	-354.731	-182.285	-43.269	-153.826	-209.907	-674.810
PhosB	-348.775	-189.189	-53.967	-145.625	-120.116	-608.777
PhosB	-335.291	-200.894	-58.080	-135.150	-156.292	-627.410
PhosB	-336.736	-206.211	-59.749	-160.909	-162.815	-628.732
PhosB	-334.932	-195.475	-54.239	-150.343	-149.882	-624.484
RibA	-37.842	-190.959	16.116	37.353	-131.310	-656.554
RibA	-82.202	-198.734	15.883	72.840	-124.595	-540.348
RibA	-137.361	-220.208	21.538	77.099	-126.286	-588.219
RibA	-174.601	-188.341	15.254	72.615	-149.609	-566.822
RibA	-168.459	-207.485	15.103	57.919	-143.764	-554.431
RibA	-178.655	-168.200	28.272	67.146	-138.649	-587.515

SubA	-376.386	-33.952	-28.039	-129.602	-155.696	-477.609
SubA	-374.395	-102.481	-2.284	-155.446	-156.393	-419.252
SubA	-296.507	-8.686	-30.221	-138.276	-124.144	-380.636
SubA	-362.440	-81.274	-7.021	-126.481	-139.298	-533.678
SubA	-288.540	0.702	-61.465	-133.844	-140.186	-546.621
SubA	-355.101	35.528	-38.348	-111.863	-122.310	-579.666

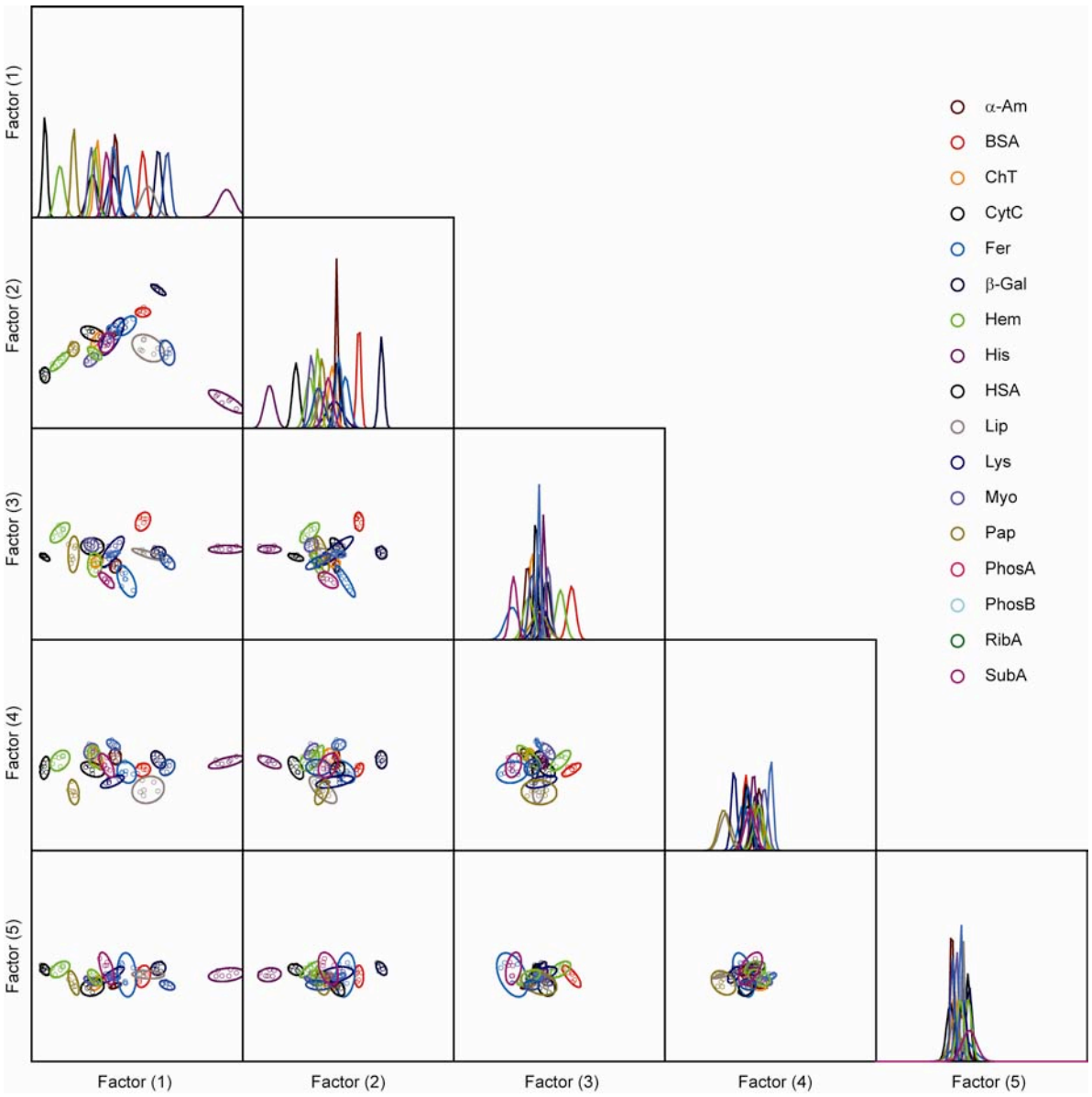


Figure S2. Two-dimensional correlations of canonical fluorescence response patterns. The 95% confidence ellipses for the individual proteins are also shown.

Table S2. LDA Classification Accuracy of Protein Analytes (A_{280} = 0.005) by Using Individual Fluorescent Polymers and the Array of the Fluorescent Polymers as Sensors. The Values are Taken from the Jackknifed Classification Matrix Based on LDA Analysis of the Raw Data (6 Replicates) Listed in Table S1.

Protein	**P1**	**P2**	**P3**	**P4**	**P5**	**P6**	**P1-P6**
α-Am	0%	0%	0%	33%	50%	50%	100%
BSA	83%	0%	17%	67%	17%	17%	100%
ChT	50%	0%	17%	83%	0%	33%	100%
CytC	100%	0%	0%	100%	100%	67%	100%
Fer	50%	17%	0%	83%	67%	0%	100%
β-Gal	83%	67%	33%	50%	33%	100%	100%
Hem	0	67%	17%	33%	67%	50%	100%
His	50%	0%	67%	100%	50%	33%	100%
HSA	33%	17%	17%	67%	17%	0%	100%
Lip	33%	67%	50%	50%	17%	33%	100%
Lys	0%	67%	50%	0%	50%	33%	100%
Myo	17%	100%	100%	50%	33%	67%	100%
Pap	100%	0%	33%	67%	0%	83%	100%
PhosA	100%	0%	50%	0%	33%	0%	100%
PhosB	33%	33%	67%	50%	0%	67%	100%
RibA	0%	0%	83%	100%	17%	33%	100%
SubA	0%	17%	33%	17%	33%	0%	100%
Average	43%	26%	37%	56%	34%	39%	100%

Table S3. Detection and Identification of Unknown Proteins Using LDA Combined with UV Measurements.[a]

#	Fluorescence response pattern						Identification			Verification	
	P1	**P2**	**P3**	**P4**	**P5**	**P6**	Protein	A_{280}	Conc.	Protein/Conc.	Deviation
1	205.868	-288.573	2.877	-93.543	-221.483	-908.288	BSA	1.07930	23.03 μM	BSA/22.57 μM	+2.0%
2	-36.525	-314.851	-71.272	-104.615	-155.643	-481.945	PhosA	0.64561	2.50 μM	PhosA/2.55 μM	-2.0%
3	-336.923	-184.637	-46.871	-168.878	-209.000	-627.381	PhosB	0.94714	15.09 μM	PhosB/14.92 μM	+1.1%
4	206.340	-236.713	11.880	-120.416	-225.059	-863.051	BSA	0.81725	17.44 μM	BSA/16.93 μM	+3.0%
5	-625.736	-257.496	-113.244	-333.300	-227.026	-993.710	CytC	0.90680	39.09 μM	CytC/38.74 μM	+0.9%
6	-284.205	-334.328	-62.777	-157.899	-217.322	-679.430	Myo	0.48907	35.08 μM	Myo/35.77 μM	-1.9%
7	-49.604	-313.123	-50.599	-137.788	-158.459	-478.620	PhosA	0.51670	2.00 μM	PhosA/1.91 μM	+4.7%
8	-154.183	-286.984	-24.395	-138.970	-263.185	-775.770	HSA	0.50154	13.27 μM	HSA/13.56 μM	-2.1%
9	-658.843	-279.765	-96.019	-346.453	-251.424	-1000.404	CytC	0.68408	29.49 μM	CytC/29.06 μM	+1.5%
10	-214.473	-117.325	-22.328	-80.036	-117.566	-445.444	α-Am	1.01160	7.78 μM	Fer/1.08 μM	Fail
11	-43.228	-297.157	-51.463	-140.573	-149.820	-496.900	PhosA	0.38010	1.47 μM	PhosA/1.31 μM	+12.2%
12	-344.979	-181.457	-42.907	-156.618	-205.868	-601.347	PhosB	0.72899	11.61μM	PhosB/11.32μM	+2.6%
13	-168.590	-270.777	-31.311	-165.386	-294.068	-734.892	Myo	0.38649	27.72 μM	HSA/10.17 μM	Fail
14	-119.696	-104.469	-8.754	-149.563	-175.239	-837.361	Lys	0.78323	20.61 μM	Lys/20.54 μM	+3.4%
15	-423.310	-248.712	-24.704	-214.249	-238.414	-920.972	Pap	0.41248	7.17 μM	Pap/7.52 μM	-4.6%
16	-316.202	-97.323	-8.979	-115.029	-137.247	-480.756	SubA	0.59009	22.67 μM	SubA/22.23 μM	+2.0%
17	-264.767	-384.261	-71.518	-164.819	-222.815	-727.058	Myo	0.36684	26.32 μM	Myo/26.83 μM	-1.9%
18	-195.353	-210.723	-16.912	-120.438	-134.817	-460.488	α-Am	0.53223	4.09 μM	α-Am/3.97 μM	+3.0%
19	254.058	-285.751	8.347	-125.631	-214.080	-892.752	BSA	0.53607	11.44 μM	BSA/11.28 μM	+1.4%
20	-172.790	-241.992	-33.462	-168.568	-275.764	-768.605	HSA	0.25797	6.82 μM	HSA/6.78 μM	+0.6%
21	317.077	-36.383	20.041	-60.316	-123.071	-402.854	β-Gal	0.25548	226.37 nM	β-Gal/217.65 nM	+4.0%
22	-289.911	-343.192	-63.100	-158.628	-194.320	-563.914	Myo	0.24780	17.78 μM	Myo/17.89 μM	-0.6%
23	-441.928	-241.634	-25.557	-247.700	-232.498	-880.193	Pap	0.30777	5.35 μM	Pap/5.38 μM	-0.6%
24	328.639	-44.038	35.605	-54.494	-110.318	-393.907	β-Gal	0.23970	212.39 nM	β-Gal/217.65 nM	-2.4%
25	-53.716	-168.207	16.848	42.080	-135.905	-560.393	RibA	0.53206	53.21 μM	RibA/52.72 μM	+0.9%
26	-331.410	-64.966	-22.203	-122.880	-145.980	-484.169	SubA	0.45741	17.57 μM	SubA/16.67 μM	+5.4%
27	-150.597	-234.214	-26.464	-177.453	-260.834	-762.498	HSA	0.13061	3.45 μM	HSA/3.39 μM	+1.8%
28	250.597	-239.809	18.168	-116.409	-209.376	-904.084	BSA	0.26260	5.60 μM	BSA/5.64 μM	-0.7%
29	-664.228	-275.116	-80.047	-299.953	-242.608	-975.049	CytC	0.45815	19.75 μM	CytC/19.37 μM	+2.0%
30	285.219	-33.982	19.723	-70.765	-115.287	-371.571	β-Gal	0.12735	112.84 nM	β-Gal/108.82 nM	+3.7%
31	-310.828	-58.049	-17.462	-143.332	-132.607	-481.866	SubA	0.29666	11.40 μM	SubA/11.11 μM	+2.6%
32	-249.518	-212.190	-21.015	-174.885	-165.739	-621.645	ChT	0.47876	9.39 μM	ChT/9.41 μM	-0.2%
33	-51.895	-294.680	-47.899	-120.775	-143.166	-468.379	PhosA	0.19010	736.88 nM	PhosA/715.95 nM	+2.9%
34	-150.139	-213.785	2.513	389.243	-156.292	-672.272	His	0.12856	33.48 μM	His/30.41 μM	+10.1%
35	-190.097	-204.420	-15.266	-128.871	-135.684	-496.873	α-Am	0.41243	3.17 μM	α-Am/3.07 μM	+3.3%
36	-349.466	-177.124	-57.398	-159.064	-206.045	-594.142	PhosB	0.49704	7.92 μM	PhosB/7.54 μM	+5.0%
37	-181.375	-56.973	19.250	-74.390	-143.897	-469.844	Fer	0.76428	804.5 nM	Fer/808.2 nM	-0.5%
38	-224.347	-111.656	25.100	-97.686	-135.439	-401.962	Fer	0.51793	545.2 nM	Fer/538.8 nM	+1.2%
39	-338.490	-188.395	-57.511	-172.196	-195.702	-586.439	PhosB	0.24387	3.88 μM	PhosB/3.77 μM	+2.9%
40	-88.915	-140.570	-9.232	-137.743	-193.456	-815.462	Lys	0.59082	15.55 μM	Lys/15.41 μM	+0.9%
41	-414.860	-215.897	-5.307	-255.164	-204.247	-825.423	Pap	0.21159	3.68 μM	Pap/3.57 μM	+3.1%
42	-182.102	-97.822	27.183	25.424	-190.947	-737.328	Lip	0.62106	11.43 μM	Lip/11.89 μM	-3.9%
43	-173.336	-209.254	1.005	375.435	-153.752	-646.665	His	0.12541	32.66 μM	His/28.85 μM	+13.2%
44	-39.052	-185.779	15.634	57.754	-136.644	-552.595	RibA	0.39310	39.31 μM	RibA/39.54 μM	-0.6%
45	-89.850	-156.538	-2.117	-152.476	-202.934	-768.488	Lys	0.40562	10.67 μM	Lys/10.27 μM	+3.9%
46	-158.068	-211.578	-0.204	371.246	-170.745	-670.916	His	0.11400	29.69 μM	His/28.85 μM	+2.9%
47	-81.326	-165.988	27.036	74.332	-129.806	-558.697	RibA	0.27234	27.23 μM	RibA/26.36 μM	+3.3%
48	-191.250	-215.892	-17.602	-124.124	-133.234	-456.393	α-Am	0.27539	2.12 μM	α-Am/2.05 μM	+3.4%
49	-299.945	-46.623	-14.677	-138.380	-132.360	-438.623	SubA	0.16156	6.21 μM	SubA/5.56 μM	+11.7%
50	-167.399	-107.760	25.454	-1.012	-196.484	-740.253	Lip	0.47704	8.78 μM	Lip/8.92 μM	-1.6%
51	-103.958	-188.526	24.108	68.478	-145.776	-627.343	RibA	0.13203	13.20 μM	RibA/13.18 μM	+0.2%
52	-396.593	-257.742	-95.951	-206.813	-322.691	-809.428	Hem	0.72591	5.81 μM	Hem/5.67 μM	+2.5%
53	311.498	-42.459	21.425	-56.801	-113.723	-382.695	β-Gal	0.12434	110.17 nM	β-Gal/108.82 nM	+1.2%
54	-194.450	-207.251	-18.628	-129.723	-135.082	-508.999	α-Am	0.13434	1.03 μM	α-Am/1.02 μM	+1.0%
55	-75.437	-148.366	-10.066	-114.924	-172.711	-734.844	Lys	0.19581	5.15 μM	Lys/5.13 μM	+0.4%
56	-187.048	-85.633	-2.829	-92.817	-113.697	-497.479	Fer	0.25693	270.5 nM	Fer/269.4 nM	+0.4%
57	-394.271	-240.323	-92.243	-205.339	-319.933	-832.554	Hem	0.48975	3.91 μM	Hem/4.25 μM	-8.0%
58	-175.346	-122.719	30.094	29.634	-192.020	-737.559	Lip	0.29781	5.48 μM	Lip/5.94 μM	-7.7%

59	-418.975	-67.661	-19.792	-247.076	-237.987	-861.259	Pap	0.11971	2.08 μM	Pap/1.95 μM	+6.7%
60	-185.271	-203.892	3.336	374.216	-172.600	-680.270	His	0.11243	29.28 μM	His/28.85 μM	+1.5%
61	-379.188	-241.094	-100.835	-195.308	-325.642	-814.008	Hem	0.28161	2.25 μM	Hem/2.83 μM	-20.5%
62	-648.420	-260.398	-89.514	-312.376	-244.209	-991.173	CytC	0.22690	9.78 μM	CytC/9.68 μM	+1.0%
63	-227.662	-213.101	-26.985	-163.228	-167.147	-619.821	ChT	0.36388	7.13 μM	ChT/7.05 μM	+1.1%
64	-229.732	-340.367	-68.626	-154.962	-211.994	-754.397	Myo	0.12285	8.81 μM	Myo/8.94 μM	-1.5%
65	-243.902	-205.961	-29.839	-168.568	-153.006	-606.213	ChT	0.26436	5.18 μM	ChT/4.70 μM	+10.2%
66	-182.125	-106.110	45.288	-21.243	-214.394	-783.599	Lip	0.13147	2.42 μM	Lip/2.64 μM	-8.3%
67	-399.131	-246.467	-94.688	-219.078	-307.087	-789.120	Hem	0.14966	1.20 μM	Hem/1.42 μM	-15.5%
68	-247.815	-200.642	-24.480	-176.112	-156.580	-642.720	ChT	0.11827	2.32 μM	ChT/2.35 μM	-1.3%

[a] α-Am: α-amylase, ε (280 nm) = 130000 M^{-1} cm^{-1}; BSA: bovine serum albumin, ε (280 nm) = 46860 M^{-1} cm^{-1}; ChT: α-chymotrypsin, ε (280 nm) = 51000 M^{-1} cm^{-1}; CytC: cytochrome *c*, ε (280 nm) = 23200 M^{-1} cm^{-1}; Fer: ferritin: ε (280 nm) = 950000 M^{-1} cm^{-1}; β-Gal: β-galactosidase, ε (280 nm) = 1128600 M^{-1} cm^{-1}; Hem: hemoglobin, ε (280 nm) = 125000 M^{-1} cm^{-1}; His: histone: ε (280 nm) = 3840 M^{-1} cm^{-1}; HSA: human serum albumin, ε (280 nm) = 37800 M^{-1} cm^{-1}; Lip: lipase: ε (280 nm) = 54350 M^{-1} cm^{-1}; Lys: lysozyme: ε (280 nm) = 38000 M^{-1} cm^{-1}; Myo: myoglobin, ε (280 nm) = 13940 M^{-1} cm^{-1}; Pap: papain: ε (280 nm) = 57500 M^{-1} cm^{-1}; PhosA: acid phosphatase, ε (280 nm) = 257980 M^{-1} cm^{-1}; PhosB: alkaline phosphatase, ε (280 nm) = 62780 M^{-1} cm^{-1}; RibA: ribonuclease A, ε (280 nm) = 10000 M^{-1} cm^{-1}; SubA: subtilisin A, ε (280 nm) = 26030 M^{-1} cm^{-1}.