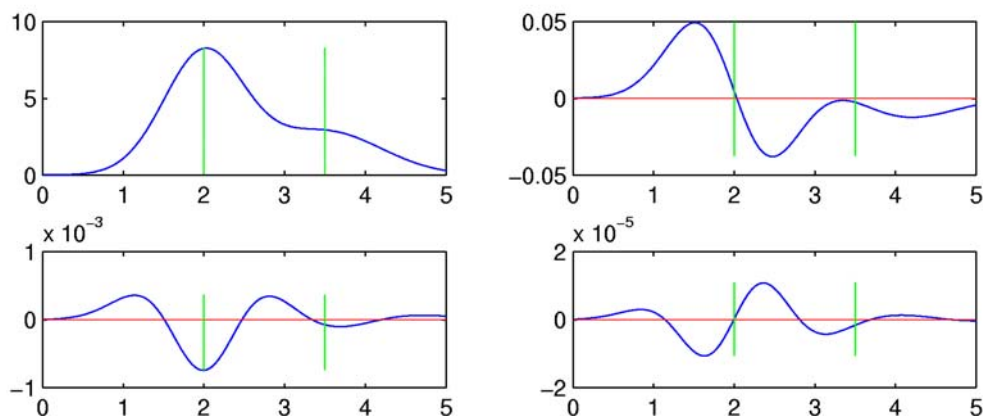
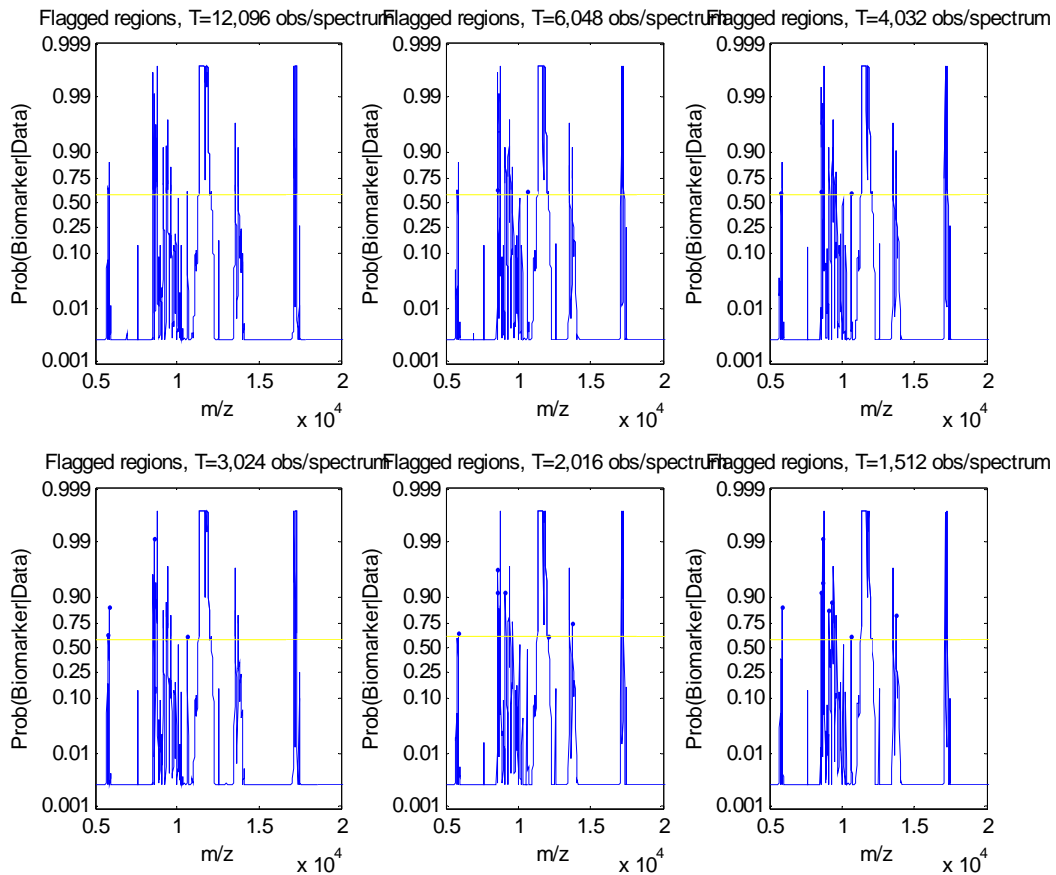


Supplementary Materials for “Bayesian Analysis of Mass Spectrometry Proteomic Data using Wavelet Based Functional Mixed Models”

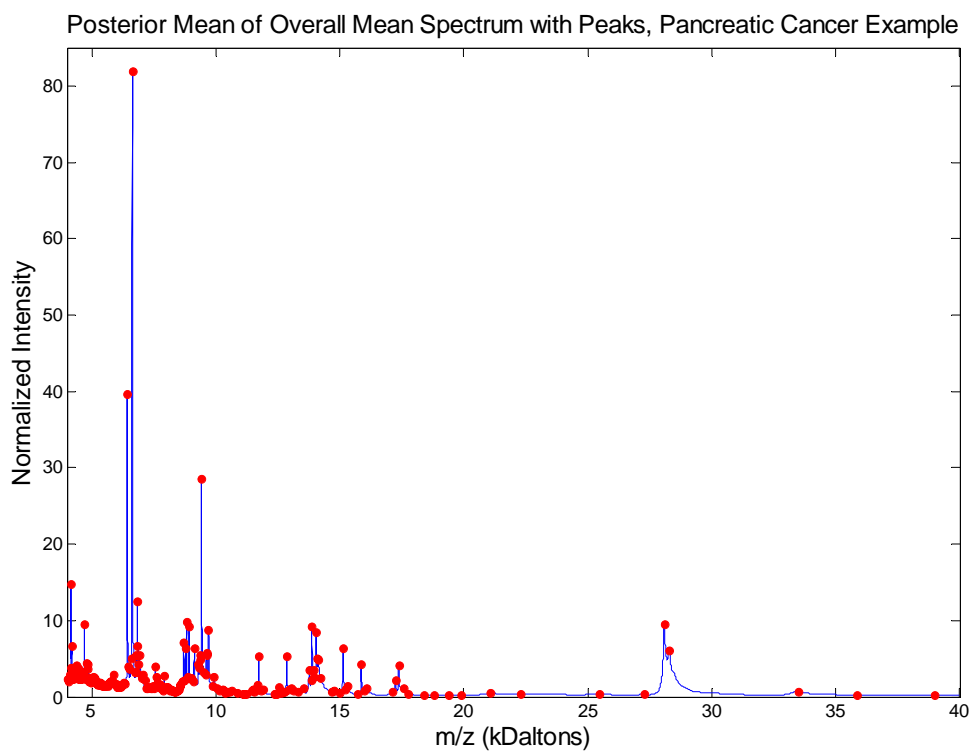
by Jeffrey S. Morris, Philip J. Brown, Richard C. Herrick, Keith A. Baggerly, and Kevin R. Coombes



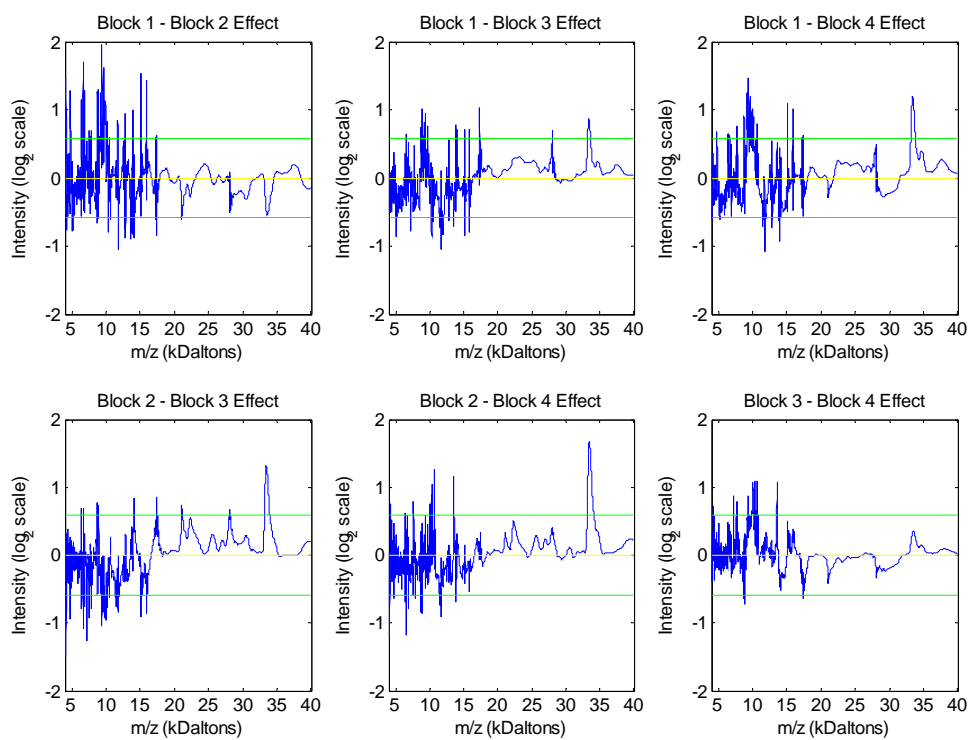
Supplementary Figure 1: Two peaks. To illustrate the fact that not all “peaks” will show up as local maxima in a spectrum, we constructed a sum of two Gaussian peaks, one centered at 2.0 with standard deviation 0.5 and height 8, and the other centered at 3.5 with standard deviation 0.7 and height 3.5. The upper left plot is of this curve, and the upper right, lower left, and lower right are the first, second, and third difference curves, respectively. Note that the “peak” at 3.5 does not appear as a local maximum, as indicated by the lack of a local maximum in the upper left figure, and the absence of a zero in the upper right figure of 1st differences. The failure of the peak to manifest as a local maximum is related to the fact that it is near the shoulder of a larger peak – if the neighboring peak were slightly smaller, or slightly further away, then the smaller peak would show up better. This illustrates the benefit of functional modeling methods for mass spectrometry data – they are not limited by the sensitivity of the peak detection methods used, but may find significant differences in spectral regions that are related to proteins but are not manifest as a local maximum.



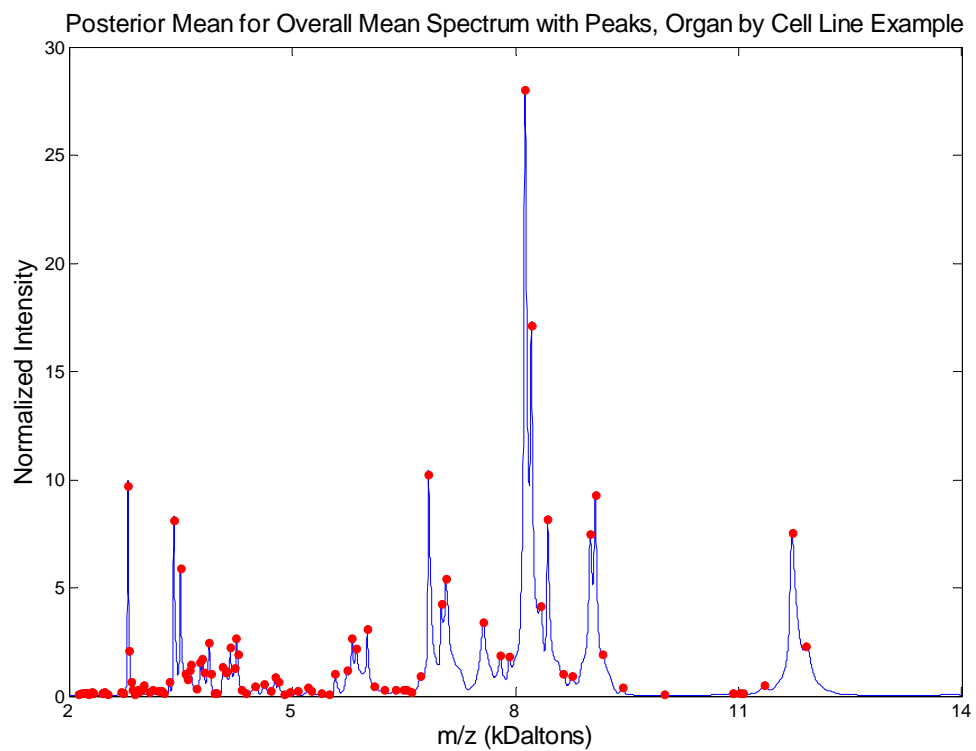
Supplementary Figure 2: Robustness to Sampling Frequency. For the pancreatic cancer example, we repeated the Bayesian FDR procedure described in Section 4 of the paper while downsampling the spectra's posterior probabilities by factors of 2, 3, 4, 6, and 8, respectively. The plots demonstrate the posterior probabilities of at least 1.5-fold expression differences between cancer and normal at each location on the grid, as a function of m/z , and the yellow line indicates the threshold corresponding to an FDR of 0.10, computed for each sampling frequency. Note that the threshold and flagged regions are robust to changes in the sampling frequency.



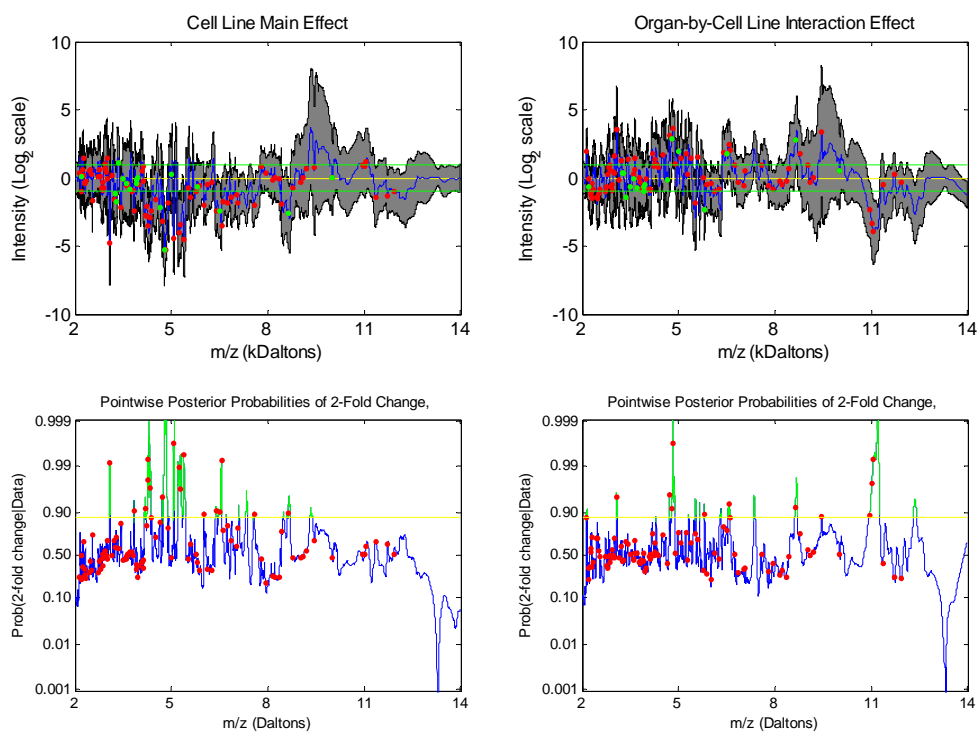
Supplementary Figure 3: *Mean Spectrum with Peaks, Pancreatic Cancer Example.* Plot of the posterior mean of the overall mean spectrum for the pancreatic cancer example, with detected peaks indicated by the red dots.



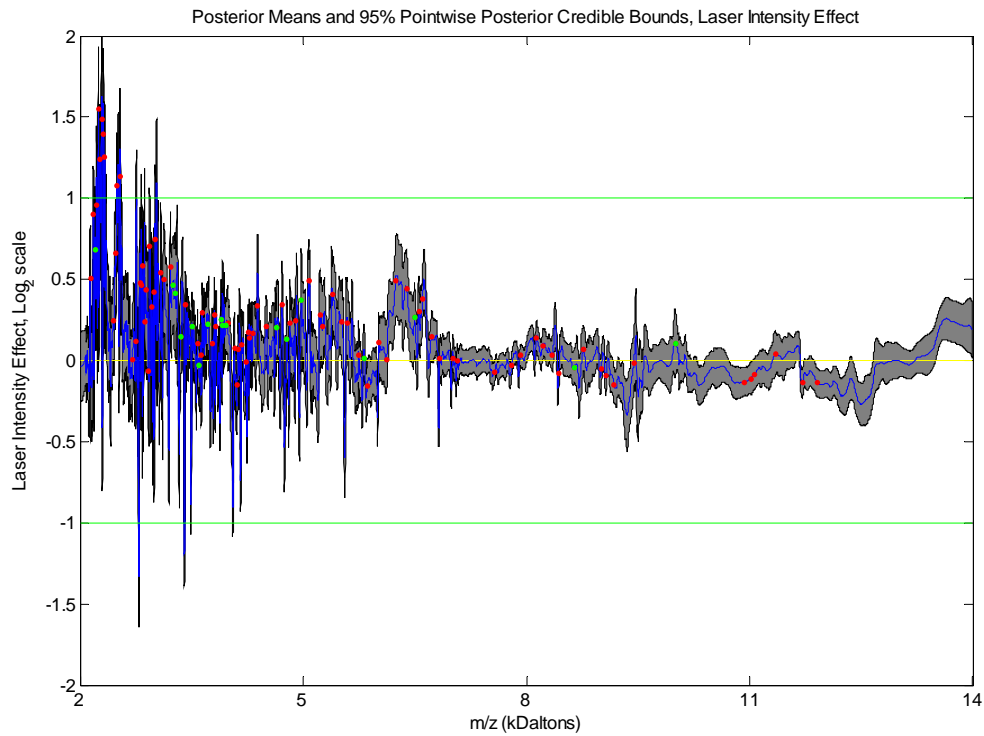
Supplementary Figure 4: Block Effects, Pancreatic Cancer Example. Posterior means for all possible pairwise block effects for the pancreatic cancer example. The green lines indicate a threshold on the \log_2 scale corresponding to a 1.5-fold difference.



Supplementary Figure 5: *Mean Spectrum with Peaks, Organ-by-Cell-Line Example.* Plot of the posterior mean of the overall mean spectrum for the organ-by-cell-line example, with detected peaks indicated by the red dots.



Supplementary Figure 6: Cell-Line and Interaction Effect Functions, Organ-by-Cell-Line example. Top plots: Posterior mean and 95% pointwise posterior credible bands for cell line main effect function and organ-by-cell-line interaction effect function, respectively. The green lines indicate 2-fold differences, and dots indicate peaks detected using the average spectrum. Bottom Plots: Posterior probabilities of at least 2-fold expression differences. The red dots indicate detected peaks, and the green lines mark the flagged regions. The yellow dotted lines indicate the threshold for flagging a location as significant, controlling the expected Bayesian FDR to be less than 0.10.



Supplementary Figure 7: *Laser Intensity Effect, Organ-by-Cell-Line Example.* Plot of the posterior mean of the laser intensity effect function (blue line), along with 95% pointwise posterior bounds (gray shaded regions). The green lines mark 2-fold expression differences. The green/red dots indicate detected peaks that were/were not flagged as significant for either the organ, cell-line, or interaction main effects.

Supplementary Table 1: Pancreatic Cancer Results, Flagged Regions. Regions of spectra flagged with significant 1.5-fold differences between cancers and normals for Pancreatic Cancer example, holding Bayesian FDR<0.10. Columns, from left to right, include the start and endpoint of the region in the m/z scale and the clock tick scale, the mean probability of at least 1.5-fold expression within the region, and the maximum height of the mean normalized spectrum (\log_2) scale within that region.

**wfmm results MYO Fraction 16 significant regions
with Bayesian FDR<0.10 (1.5-fold)**

x_start	x_end	index_s	index_e	mean(prob)	max height mean spectrum
5819.1	5824.2	1156	1159	0.626	0.98
5836.3	5846.7	1166	1172	0.751	0.99
8555.6	8566	2592	2597	0.917	0.66
8576.5	8578.6	2602	2603	0.624	0.54
8618.3	8628.8	2622	2627	0.960	1.04
8670.8	8683.5	2647	2653	0.879	2.62
8729.8	8786.9	2675	2702	0.885	2.66
9126.8	9141.9	2861	2868	0.820	2.68
9337.3	9354.8	2958	2966	0.804	2.45
9389.8	9453.4	2982	3011	0.840	4.83
9621	9645.4	3087	3098	0.745	2.52
10644	10646	3537	3538	0.617	-0.25
11314	12037	3820	4116	0.944	2.40
12071	12099	4130	4141	0.608	-1.15
13528	13573	4700	4717	0.829	0.13
13750	13763	4784	4789	0.820	1.79
17103	17171	5980	6003	0.954	-0.45
17230	17311	6023	6050	0.899	1.17

Supplementary Table 2: Pancreatic Cancer Results, Flagged Peaks. Spectral peaks flagged with significant 1.5-fold differences between cancers and normals for Pancreatic Cancer example, holding Bayesian FDR<0.10. Columns, from left to right, include the m/z value and clock tick location of the peak, the posterior probability of 1.5-fold expression difference at the peak, the log₂ fold change estimate at the peak, and the fold-change estimate at the peak.

**wfmm results MYO Fraction 26 significant peaks
with Bayesian FDR<0.10 (1.5-fold)**

peak	index_peak	Prob(1.5x)	log ₂ (fc)	fc
5844.9	1171	0.656	0.667	1.59
8561.8	2595	0.997	-0.704	0.61
8578.6	2603	0.634	-0.561	0.68
8624.6	2625	0.992	-0.746	0.60
8734.1	2677	0.862	-0.603	0.66
8763.6	2691	0.998	-1.028	0.49
9131.1	2863	0.919	-0.632	0.65
9354.8	2966	0.598	-0.688	0.62
9420.5	2996	0.953	-0.710	0.61
9627.7	3090	0.821	-0.656	0.63
9641	3096	0.806	-0.668	0.63
10644	3537	0.621	-0.576	0.67
11439	3872	0.998	1.366	2.58
11471	3885	0.998	1.404	2.65
11524	3907	0.998	1.411	2.66
11568	3925	0.998	1.065	2.09
11631	3951	0.998	1.350	2.55
11680	3971	0.998	1.299	2.46
11726	3990	0.789	0.811	1.75
11822	4029	0.997	0.864	1.82
11925	4071	0.966	0.769	1.70
13536	4703	0.958	-0.719	0.61
13758	4787	0.919	-0.682	0.62
17124	5987	0.997	-0.912	0.53
17144	5994	0.996	-0.854	0.55
17245	6028	0.998	-1.160	0.45

Supplementary Table 3: Pancreatic Cancer Results, Koomen Results. Spectral peaks flagged with significant differences between cancers and normals for Pancreatic Cancer example, using t-tests, and the BUM method to control FDR<0.01. Columns, from left to right, include the m/z value and clock tick location of the peak, the t-statistic and p-value at the peak location, the log₂ fold change estimate at the peak, and the fold-change estimate at the peak.

**Koomen et al. MYO Fraction 28 sig
with FDR<0.01 (t-tests)**

peak	Index_peak	t	pv	log ₂ (fc)	fc
4135.6	95	1.42	0.078723	0.211	1.16
4403.9	277	-2.89	0.002066	-0.445	0.73
4711	478	-6.89	0.000000	-0.980	0.51
4788.7	528	1.41	0.079762	0.210	1.16
5747.7	1114	2.10	0.018407	0.430	1.35
5840.5	1168	4.92	0.000001	1.140	2.20
6939.3	1777	-5.47	0.000000	-0.558	0.68
8622.9	2624	-7.19	0.000000	-1.118	0.46
8685.9	2654	-5.61	0.000000	-0.730	0.60
8761.8	2690	-9.65	0.000000	-1.622	0.32
9129.2	2862	-6.02	0.000000	-0.922	0.53
9348.5	2963	-7.10	0.000000	-0.966	0.51
9418.5	2995	-6.80	0.000000	-0.939	0.52
9506.4	3035	-4.25	0.000015	-0.501	0.71
9632.3	3092	-5.09	0.000000	-0.823	0.57
11381	3847	5.09	0.000000	0.994	1.99
11465	3882	6.72	0.000000	1.477	2.78
11523	3906	6.33	0.000000	1.419	2.67
11619	3945	6.10	0.000000	1.261	2.40
11677	3969	5.54	0.000000	1.291	2.45
11724	3988	3.52	0.000254	0.539	1.45
11849	4039	6.21	0.000000	1.162	2.24
13534	4702	-3.22	0.000731	-0.848	0.56
13755	4785	-5.24	0.000000	-0.668	0.63
13874	4830	-5.68	0.000000	-0.728	0.60
13905	4842	-5.56	0.000000	-0.549	0.68
17114	5983	-6.30	0.000000	-1.004	0.50
17241	6026	-6.53	0.000000	-1.095	0.47

Supplementary Table 4: Organ-by-Cell-Line Results, Flagged Regions for Organ Main Effect. Regions of spectra flagged with significant 2.0-fold differences for organ effect in organ-by-cell-line experiment, holding Bayesian FDR<0.05. Columns, from left to right, include the start and endpoint of the region in the m/z scale and the clock tick scale, the mean probability of at least 2.0-fold expression within the region, the maximum height of the mean normalized spectrum (log₂) scale within that region, the maximum probability of at least 2-fold expression, the maximum absolute fold change on the log₂ scale, and the maximum fold change.

index_s	index_e	x_start	x_end	Mean (prob)	mean spectrum	Max (prob)	Max (log ₂ effect)	max(FC)
1894	1985	3866.3	3971.3	0.991	1.35	1.000	-6.55	93.92
1710	1782	3658.3	3739	0.990	-0.68	1.000	-6.89	118.45
1542	1626	3473.4	3565.3	0.981	2.59	1.000	-5.13	35.02
2004	2062	3993.4	4061.3	0.980	0.36	1.000	4.39	21.03
2442	2584	4519.9	4697.5	0.976	-0.80	1.000	4.78	27.52
2783	2826	4952.3	5008.2	0.973	-2.54	1.000	-5.04	32.79
5945	6022	9902.6	10044	0.954	-3.81	1.000	5.53	46.05
2603	2673	4721.6	4810.6	0.959	-0.22	0.996	4.14	17.59
1309	1353	3225	3271.2	0.949	-2.13	0.995	-4.04	16.47
3410	3433	5798.9	5831.2	0.964	1.42	0.994	-3.55	11.73
3739	3809	6269.9	6372.4	0.946	-1.79	0.993	3.38	10.39
207	225	2174.7	2190.1	0.944	-3.16	0.991	-3.10	8.58
3820	3910	6388.6	6521.9	0.940	-1.68	0.979	3.08	8.46
5638	5683	9347.6	9427.9	0.936	-1.60	0.969	5.42	42.91
6570	6634	11082	11207	0.939	-2.89	0.967	-2.92	7.59
2336	2370	4389.5	4431.1	0.938	-3.12	0.958	3.03	8.16
1632	1656	3571.9	3598.4	0.918	-0.06	0.944	-2.95	7.71
165	171	2138.7	2143.8	0.910	-4.19	0.944	-2.28	4.85
5218	5276	8614.1	8713.6	0.917	0.14	0.942	2.97	7.82
5710	5768	9476.3	9580.6	0.905	-2.88	0.936	4.43	21.52
5078	5094	8376.3	8403.3	0.904	1.30	0.926	2.44	5.44
4618	4638	7618.3	7650.5	0.904	1.00	0.924	-2.30	4.94
1420	1432	3342.2	3355	0.899	-0.63	0.923	-2.68	6.43
5329	5347	8805	8836.2	0.904	-0.74	0.920	2.67	6.36
3308	3313	5656.6	5663.6	0.891	-2.90	0.910	2.36	5.12
798	810	2712.3	2723.8	0.893	-2.77	0.910	-2.26	4.78
228	236	2192.7	2199.6	0.896	-3.10	0.909	-2.15	4.44
2692	2707	4834.9	4854.2	0.894	-1.58	0.907	2.35	5.10
2828	2842	5010.8	5029.1	0.889	-4.10	0.905	-2.75	6.71
344	347	2294	2296.6	0.877	-3.30	0.898	2.14	4.41
1244	1253	3157.3	3166.6	0.887	-3.81	0.898	-2.56	5.92
1024	1027	2933.5	2936.5	0.880	-2.26	0.891	-2.53	5.78
258	261	2218.7	2221.3	0.878	-3.34	0.888	-1.97	3.93
366	369	2313.5	2316.1	0.871	-3.67	0.883	2.11	4.32
5773	5786	9589.7	9613.1	0.878	-4.20	0.881	3.32	9.96
6397	6414	10749	10782	0.877	-3.76	0.880	-2.44	5.44
1017	1020	2926.6	2929.6	0.875	-1.90	0.880	-2.41	5.31
542	544	2472.1	2473.9	0.871	-2.92	0.880	-2.08	4.24
1528	1529	3458.2	3459.3	0.874	0.41	0.877	-2.28	4.86
5770	5772	9584.3	9587.9	0.875	-4.15	0.875	3.28	9.74
2887	2889	5088.1	5090.7	0.874	-3.14	0.875	-2.64	6.24

Supplementary Table 5: Organ-by-Cell-Line Results, Flagged Regions for Cell Line Main Effect. Regions of spectra flagged with significant 2.0-fold differences for cell line effect in organ-by-cell-line experiment, holding Bayesian FDR<0.05. Columns, from left to right, include the start and endpoint of the region in the m/z scale and the clock tick scale, the mean probability of at least 2.0-fold expression within the region, the maximum height of the mean normalized spectrum (log₂) scale within that region, the maximum probability of at least 2-fold expression, the maximum absolute fold change on the log₂ scale, and the maximum fold change.

index_s	index_e	x_start	x_end	Mean (prob)	mean spectrum	Max (prob)	Max (log ₂ effect)	Max (FC)
2624	2718	4748.2	4868.3	0.987	-0.22	1.000	-5.31	39.68
2859	2891	5051.3	5093.3	0.972	-2.01	1.000	-4.56	23.53
2205	2303	4231	4349.3	0.970	1.44	0.999	-4.11	17.21
2969	3066	5196.5	5326.1	0.968	-1.29	0.995	-4.03	16.32
3087	3142	5354.4	5428.9	0.965	-2.91	0.995	-4.48	22.37
3862	3944	6450.7	6572.6	0.928	-1.74	0.993	-3.46	11.02
1150	1185	3060.7	3096.5	0.958	-2.57	0.992	-4.75	26.84
4421	4454	7304.7	7356.8	0.931	-0.51	0.967	-2.25	4.75
2588	2599	4702.6	4716.5	0.930	-1.92	0.956	-3.25	9.51
5226	5268	8627.8	8699.9	0.923	0.07	0.955	-3.01	8.03
2161	2179	4178.4	4199.8	0.920	0.47	0.953	-2.68	6.39
1851	1863	3817.2	3830.9	0.912	0.12	0.944	-2.63	6.20
4278	4294	7081.2	7106	0.897	2.22	0.930	-2.20	4.58
3807	3832	6369.5	6406.3	0.906	-1.68	0.928	-2.40	5.28
2146	2157	4160.5	4173.6	0.898	1.19	0.923	-2.73	6.64
5616	5643	9308.4	9356.5	0.900	-0.85	0.921	3.74	13.36
3559	3563	6009.9	6015.6	0.894	1.65	0.907	-2.00	4.00
4577	4595	7552.5	7581.3	0.887	1.77	0.904	-2.04	4.12
5131	5151	8465.9	8499.9	0.886	1.90	0.903	-2.54	5.83
5646	5662	9361.8	9390.4	0.884	-2.67	0.897	3.47	11.05
4031	4037	6703.3	6712.3	0.888	-0.17	0.894	-1.97	3.90
5211	5220	8602.1	8617.5	0.885	0.29	0.892	-2.44	5.41
2744	2748	4901.8	4907	0.879	-3.78	0.885	-2.40	5.29
2322	2328	4372.4	4379.7	0.876	-2.98	0.879	-2.42	5.34
2329	2331	4380.9	4383.4	0.875	-3.06	0.877	-2.35	5.11

Supplementary Table 6: Organ-by-Cell-Line Results, Flagged Regions for Interaction Effect. Regions of spectra flagged with significant 2.0-fold differences for interaction effect in organ-by-cell-line experiment, holding Bayesian FDR<0.05. Columns, from left to right, include the start and endpoint of the region in the m/z scale and the clock tick scale, the mean probability of at least 2.0-fold expression within the region, the maximum height of the mean normalized spectrum (log₂) scale within that region, the maximum probability of at least 2-fold expression, the maximum absolute fold change on the log₂ scale, and the maximum fold change.

index_s	index_e	x_start	x_end	Mean (prob)	mean spectrum	Max (prob)	Max (log ₂ effect)	max(FC)
6482	6666	10912	11269	0.969	-2.32	1.000	-4.03	16.36
2635	2723	4762.2	4874.8	0.952	-0.22	1.000	3.78	13.70
5216	5269	8610.7	8701.6	0.940	0.17	0.982	3.49	11.21
1154	1175	3064.8	3086.2	0.930	-2.57	0.962	3.69	12.91
2585	2600	4698.8	4717.8	0.925	-1.92	0.961	3.23	9.40
4437	4471	7329.9	7383.7	0.920	-1.21	0.956	2.11	4.33
7186	7236	12310	12412	0.925	-2.75	0.948	-2.68	6.40
3204	3224	5513.4	5540.8	0.920	-3.35	0.948	-2.94	7.65
3302	3312	5648.3	5662.2	0.913	-2.72	0.939	-2.55	5.87
3922	3938	6539.8	6563.6	0.913	-1.74	0.938	2.47	5.54
3405	3416	5791.9	5807.3	0.913	1.42	0.936	-2.54	5.80
3756	3773	6294.7	6319.6	0.899	-2.89	0.920	-2.37	5.18
3939	3961	6565.1	6598	0.898	-2.14	0.919	2.29	4.90
154	161	2129.3	2135.3	0.894	-3.76	0.916	2.10	4.29
3051	3060	5306	5318.1	0.886	-3.19	0.894	2.46	5.49
5689	5698	9438.7	9454.8	0.882	-1.36	0.886	3.42	10.68
2260	2263	4297.2	4300.8	0.872	-1.13	0.881	2.21	4.61
3740	3747	6271.3	6281.5	0.877	-2.50	0.880	-2.16	4.46

Supplementary Table 7: Flagged Peaks, Organ-by-Cell Line Example. Flagged peaks for organ-by-cell-line example, for 2.0-fold change with FDR<0.05. Columns from left to right include, m/z and clock tick location of peak, probability of 2-fold expression for organ, cell-line, and interaction effects, estimate of fold-change for organ, cell-line, and interaction effects, and indication of which effect(s) is significant (1=organ, 2=cell line, 3=both organ and cell line, 4=interaction).

peak	Index peak	Prob(2.0x)			Fold-change			Effect
		Organ	Cell line	Interaction	Organ	Cell Line	Interaction	
2130.2	155	0.226	0.242	0.875	1.157	1.264	3.856	4
2197.9	234	0.899	0.223	0.349	0.244	1.109	0.628	1
3077	1166	0.746	0.992	0.953	3.405	0.038	11.960	4
3243.8	1327	0.981	0.596	0.424	0.077	0.452	1.269	1
3269.1	1351	0.912	0.726	0.444	0.166	0.312	1.281	1
3346.4	1424	0.912	0.584	0.667	0.161	2.188	0.377	1
3497.4	1564	0.998	0.441	0.511	0.036	0.969	0.631	1
3591.7	1650	0.921	0.449	0.439	0.151	0.773	0.963	1
3707.5	1754	0.999	0.538	0.529	0.022	0.576	0.612	1
3819.5	1853	0.682	0.911	0.439	0.374	0.191	0.613	2
3885.8	1911	0.998	0.324	0.531	0.012	0.901	0.497	1
3905.4	1928	0.998	0.382	0.434	0.048	0.758	0.628	1
3951.6	1968	0.998	0.246	0.309	0.025	1.010	0.737	1
4167.7	2152	0.424	0.919	0.757	0.981	0.151	3.572	2
4250.2	2221	0.411	0.993	0.652	0.657	0.088	2.591	2
4268.2	2236	0.272	0.980	0.549	0.928	0.141	2.103	2
4321.3	2280	0.421	0.970	0.519	1.629	0.134	1.946	2
4372.4	2322	0.636	0.874	0.502	2.519	0.189	1.743	2
4624.5	2526	0.998	0.846	0.402	25.277	0.220	0.918	1
4711.4	2595	0.753	0.954	0.958	3.536	0.111	8.993	4
4769.8	2641	0.968	0.998	0.918	11.322	0.026	7.417	4
4814.5	2676	0.812	0.998	0.997	3.468	0.026	12.901	4
4971.7	2798	0.993	0.473	0.762	0.031	1.233	3.884	1
5068.4	2872	0.857	0.997	0.657	0.197	0.047	2.735	2
5216.4	2984	0.556	0.990	0.761	1.924	0.063	3.523	2
5259.1	3016	0.542	0.969	0.434	1.955	0.096	1.195	2
5385.5	3110	0.536	0.995	0.664	1.760	0.045	2.716	2
5805.9	3415	0.985	0.416	0.896	0.110	0.645	0.203	4
6011.3	3560	0.501	0.894	0.225	0.508	0.262	0.770	2
6387.2	3819	0.868	0.910	0.815	4.154	0.193	3.605	2
6496.6	3893	0.975	0.907	0.786	8.089	0.194	3.493	3
6548.7	3928	0.532	0.993	0.935	2.098	0.092	5.496	4
6598	3961	0.325	0.785	0.876	1.388	0.305	4.124	4
7571.7	4589	0.257	0.892	0.509	0.805	0.254	1.985	2
8636.4	5231	0.917	0.899	0.924	6.472	0.171	6.856	4
9444	5692	0.808	0.688	0.883	5.187	1.747	10.418	4
10007	6002	0.999	0.471	0.509	46.045	1.059	1.482	1
10927	6490	0.736	0.511	0.890	0.322	1.917	0.206	4
11016	6536	0.757	0.515	0.977	0.289	1.829	0.098	4
11063	6560	0.841	0.609	0.993	0.218	2.315	0.065	4