**Supplementary Information for "MALDI Mass Spectrometry Imaging in a Primary Demyelination Model of Murine Spinal Cords"** by Emily R. Sekera,<sup>1</sup> Darpan Saraswat,<sup>2</sup> Kevin J. Zemaitis,<sup>1</sup> Fraser J. Sim,<sup>2</sup> and Troy D. Wood<sup>1\*</sup>

Ranking	m/z	correlation	M1	M2
1	496.343	1	1	1
2	732.650	0.8843388	0.8366	0.8366
3	731.625	0.86776	0.8366	0.8366
4	577.534	0.8595857	0.7837	0.7837
5	248.183	0.8587007	0.8354	0.8354
6	784.595	0.8483514	0.7825	0.7825
7	733.603	0.8416425	0.756	0.756
8	758.595	0.8333636	0.7917	0.7917
9	365.830	0.8331041	0.8366	0.8366
10	759.581	0.8169438	0.7883	0.7883
11	380.303	0.8133262	0.7802	0.7802
12	380.797	0.8125086	0.7871	0.7871
13	720.588	0.7965343	0.7675	0.7675
14	735.586	0.7909163	0.7952	0.7952
15	734.557	0.7889386	0.7894	0.7894
16	367.296	0.7885805	0.7917	0.7917
17	253.552	0.7884472	0.756	0.756
18	760.570	0.7869545	0.7422	0.7422
19	761.635	0.7836034	0.7434	0.7434
20	367.810	0.7789616	0.7906	0.7906
21	244.880	0.7789535	0.7871	0.7871
22	391.806	0.7755557	0.6697	0.6697
23	253.882	0.7743311	0.7699	0.7699
24	736.617	0.7735018	0.7883	0.7883
25	1542.203	0.7706027	0.7411	0.7411

Table S1. Top 25 m/z in the Control Section Based on the Equated Pearson Correlation (n = 3).

Ranking	m/z	correlation	M1	M2
1	496.343	1	1	1
2	760.570	0.8855853	0.799337	0.799337
3	761.635	0.8795073	0.79602	0.79602
4	380.303	0.8599661	0.794362	0.794362
5	762.626	0.8410172	0.784411	0.784411
6	808.570	0.8270669	0.767828	0.767828
7	732.576	0.8262135	0.787728	0.787728
8	253.526	0.8066901	0.764511	0.764511
9	810.594	0.8045615	0.74461	0.74461
10	731.625	0.798963	0.802653	0.802653
11	577.534	0.7983043	0.74461	0.74461
12	783.576	0.78916	0.73466	0.73466
13	758.595	0.7872138	0.741294	0.741294
14	811.649	0.7866043	0.736318	0.736318
15	380.797	0.7863408	0.771144	0.771144
16	782.558	0.7862881	0.729685	0.729685
17	785.616	0.7725437	0.752902	0.752902
18	784.595	0.7601618	0.719735	0.719735
19	524.354	0.7527635	0.728027	0.728027
20	733.603	0.7521618	0.757877	0.757877
21	790.660	0.7474787	0.746269	0.746269
22	759.657	0.7447933	0.749585	0.749585
23	723.476	0.7436856	0.703151	0.703151
24	724.490	0.7372875	0.713101	0.713101
25	763.618	0.7363802	0.749585	0.749585

Table S2. Top 25 m/z in the 7 dpI Section Based on the Equated Pearson Correlation (n = 3).

Ranking	m/z	correlation	M1	M2
1	496.343	1	1	1
2	732.576	0.8711225	0.817585	0.817585
3	497.337	0.8673934	0.799213	0.799213
4	782.558	0.8598061	0.824147	0.824147
5	783.576	0.857214	0.816273	0.816273
6	733.603	0.8571726	0.784777	0.784777
7	391.297	0.8481784	0.809711	0.809711
8	391.141	0.8428891	0.809711	0.809711
9	758.595	0.8108342	0.783465	0.783465
10	720.588	0.8023305	0.783465	0.783465
11	391.806	0.7964762	0.748032	0.748032
12	260.882	0.7960478	0.75853	0.75853
13	784.595	0.7917897	0.791339	0.791339
14	759.581	0.7908774	0.778215	0.778215
15	577.534	0.7902944	0.782152	0.782152
16	731.625	0.7800646	0.784777	0.784777
17	723.476	0.7661762	0.776903	0.776903
18	380.303	0.7630641	0.787402	0.787402
19	518.307	0.7613063	0.744095	0.744095
20	380.797	0.7566262	0.783465	0.783465
21	804.537	0.7529455	0.728347	0.728347
22	808.570	0.7512867	0.740158	0.740158
23	810.594	0.7500658	0.737533	0.737533
24	745.509	0.7431823	0.728347	0.728347
25	761.635	0.7420141	0.782152	0.782152

Table S3. Top 25 m/z in the 14 dpI Section Based on the Equated Pearson Correlation (n = 3).

Ranking	m/z	correlation	M1	M2
1	496.343	1	1	1
2	732.576	0.9073122	0.842	0.842
3	782.558	0.8962942	0.822	0.822
4	783.576	0.8960431	0.826	0.826
5	733.603	0.8778388	0.802	0.802
6	731.625	0.8622493	0.856	0.856
7	758.595	0.8567485	0.824	0.824
8	577.534	0.8516148	0.824	0.824
9	723.476	0.8509324	0.8	0.8
10	784.595	0.8479682	0.828	0.828
11	760.570	0.8268713	0.82	0.82
12	391.297	0.8268091	0.786	0.786
13	761.635	0.8229695	0.826	0.826
14	808.570	0.8137183	0.814	0.814
15	804.537	0.8135809	0.792	0.792
16	734.557	0.8088015	0.788	0.788
17	735.586	0.8080369	0.794	0.794
18	724.490	0.8051679	0.744	0.744
19	736.617	0.8026913	0.798	0.798
20	380.303	0.8019672	0.818	0.818
21	736.322	0.798303	0.798	0.798
22	810.594	0.7964232	0.738	0.738
23	367.296	0.7958765	0.81	0.81
24	253.552	0.792308	0.814	0.814
25	694.552	0.7915833	0.744	0.744

Table S4. Top 25 m/z in the 28 dpI Section Based on the Equated Pearson Correlation (n = 2).

Ranking	m/z	correlation	M1	M2
1	496.343	1	1	1
2	367.296	0.8685552	0.8996	0.8996
3	734.557	0.8667451	0.8952	0.8952
4	735.586	0.866539	0.8952	0.8952
5	756.549	0.8660826	0.8908	0.8908
6	697.476	0.863227	0.8777	0.8777
7	758.595	0.8594192	0.8079	0.8079
8	244.880	0.8547116	0.9039	0.9039
9	820.544	0.8516547	0.7991	0.7991
10	736.617	0.8500314	0.8908	0.8908
11	757.533	0.8478195	0.8952	0.8952
12	367.810	0.8477932	0.9039	0.9039
13	698.523	0.841234	0.869	0.869
14	731.625	0.8390675	0.821	0.821
15	1542.203	0.8366406	0.7642	0.7642
16	784.595	0.8276637	0.8559	0.8559
17	783.576	0.8108438	0.6725	0.6725
18	1544.210	0.8076435	0.7904	0.7904
19	762.626	0.8074271	0.8384	0.8384
20	1521.219	0.8071614	0.7729	0.7729
21	577.534	0.8066179	0.7991	0.7991
22	782.558	0.8058313	0.655	0.655
23	723.476	0.8056864	0.7424	0.7424
24	551.514	0.8039598	0.8777	0.8777
25	1495.129	0.8033508	0.869	0.869

Table S5. Top 25 m/z in the Shiverer Section Based on the Equated Pearson Correlation (n = 2).

## Supplementary Figure S1.

SSC segmentation analysis of sections after lysolecithin injection (3, 7, 14, 28 dpl) and *shiverer* model.





**Supplementary Figure S2.** MALDI mass spectra over the range m/z 480-1000 for average of A) three sections for 3 dpl, B) three sections for 7 dpl, C) three sections for 14 dpl, D) two sections for 28 dpl, and E) two sections for *shiverer* model.





## Supplementary Figure S3.

Reproducibility of the MALDI MSI of spinal cord tissues using the (M+H)<sup>+</sup> and (M+Na)<sup>+</sup> ions from 16:0 and 18:0 LPCs: A) 3 dpl, B) 7 dpl, C) 14 dpl, D) 28 dpl, and E) uninjured spinal cord sections.



## Supplementary Figure S4.

PCA analysis of the full mass spectrum of the top three components in images of A) 3 dpi WT, B) 7 dpi, C) 14 dpi, D) 28 dpi, and E) control spinal cord sections. Principle component 1, 2, and 3 are shown from left to right, respectively.



**Supplementary Figure S5.** PCA analysis of the m/z range 700 – 1000 Da of the top three components in images of A) 3 dpi WT, B) 7 dpi, C) 14 dpi, D) 28 dpi, and E) control spinal cord sections. Principle component 1, 2, and 3 are shown from left to right, respectively.



**Supplementary Figure S6.** PCA analysis of the m/z range 700 – 1000 Da of the top three components in images of A) 3 dpi WT and B) *Shiverer* spinal cord sections. Principle component 1, 2, and 3 are shown from left to right, respectively.

