Supplemental Data for "Mass Spectrometric Imaging of Ceramides Tracks Therapeutic Response in Traumatic Brain Injury" by Barbacci et al.

List of Supplementary Figures and Tables

CCI Treated   CCI Vehicle   Control Treated   Control Vehicle

0   5.0×10⁵   1.0×10⁶   1.5×10⁶   2.0×10⁶
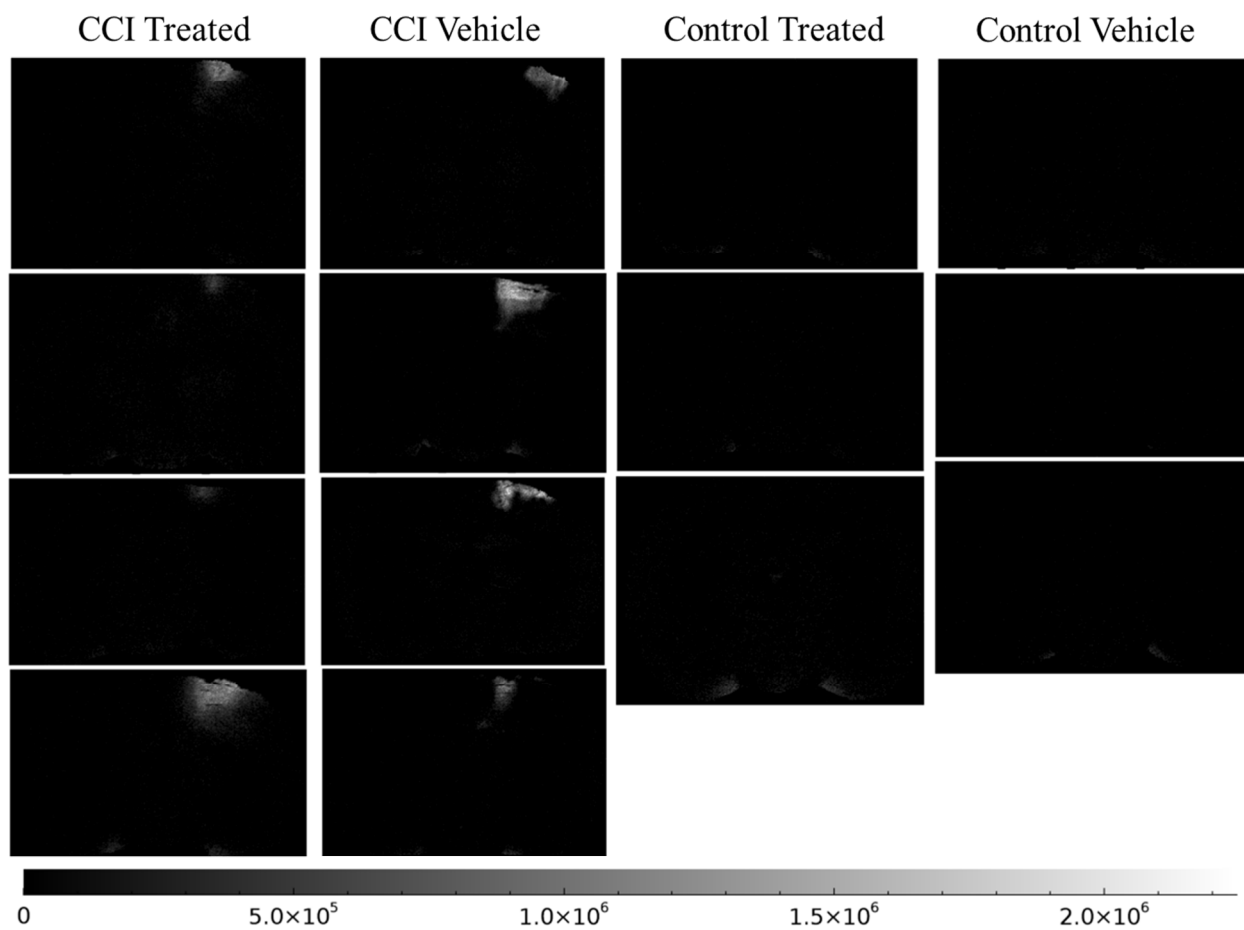
Figure S1. The distribution of CE18:1 (m/z 757.5052) displayed as 8-bit grayscale images for each study sample.

Figure S2. The distribution of CE 18:1 (m/z 757.5052) displayed as binary images of pixels exceeding peak area threshold. Black pixels represent peak area less than or equal to 953000 counts and white pixels represent peak areas greater than 953000 counts.

CE 18:1

Figure S3. Total Peak Area for pixels greater than threshold for CE 18:1. CCI Vehicle and CCI Treated Q4 are significantly different than Q4 of Control Treated (p=0.037) and Control Vehicle (p=0.037) Arms. CCI Vehicle Q4 is not significantly different from CCI Treated Q4.
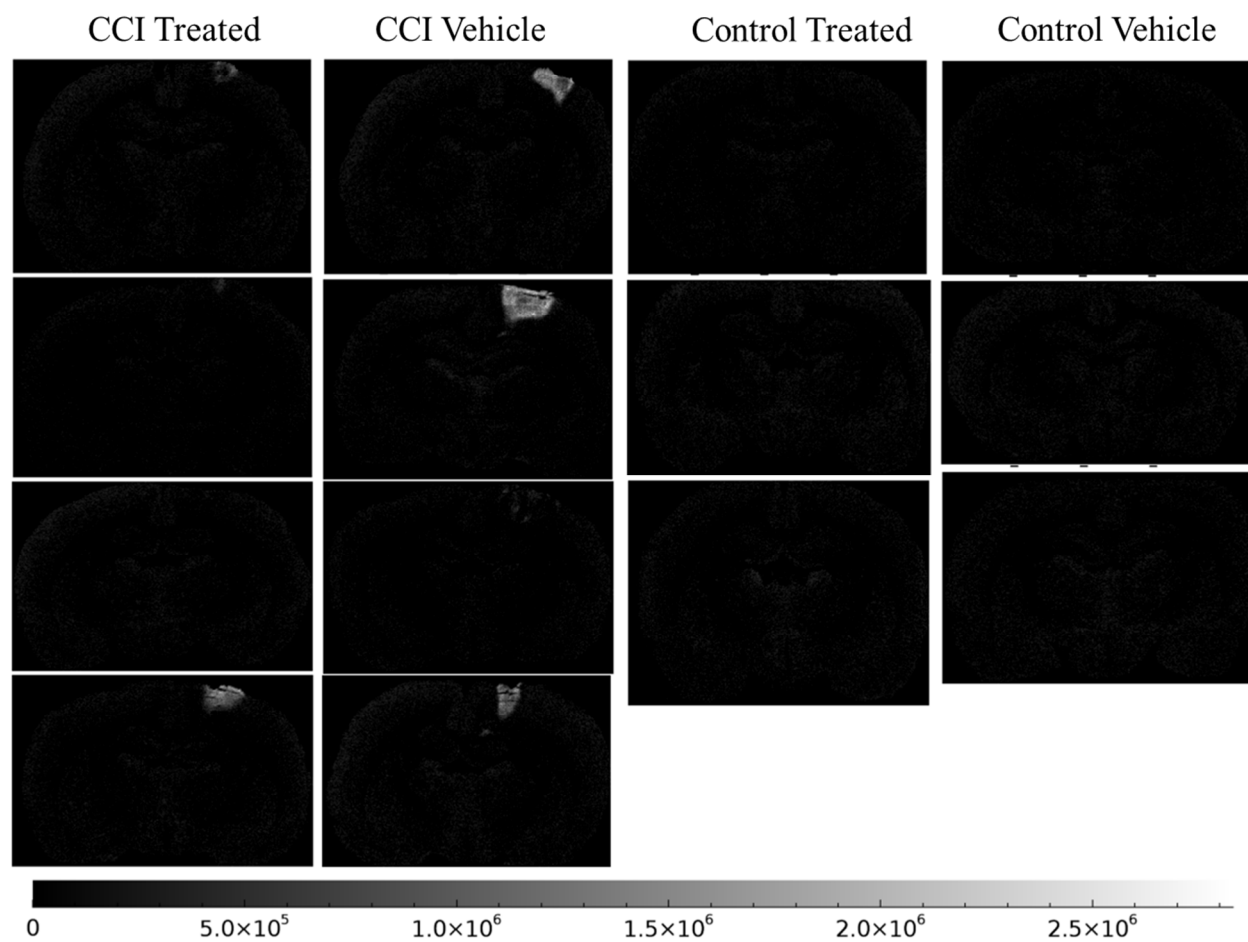
Figure S4. The distribution of DAG 40:a6 (m/z 775.4430) displayed as 8-bit grayscale images for each study sample.
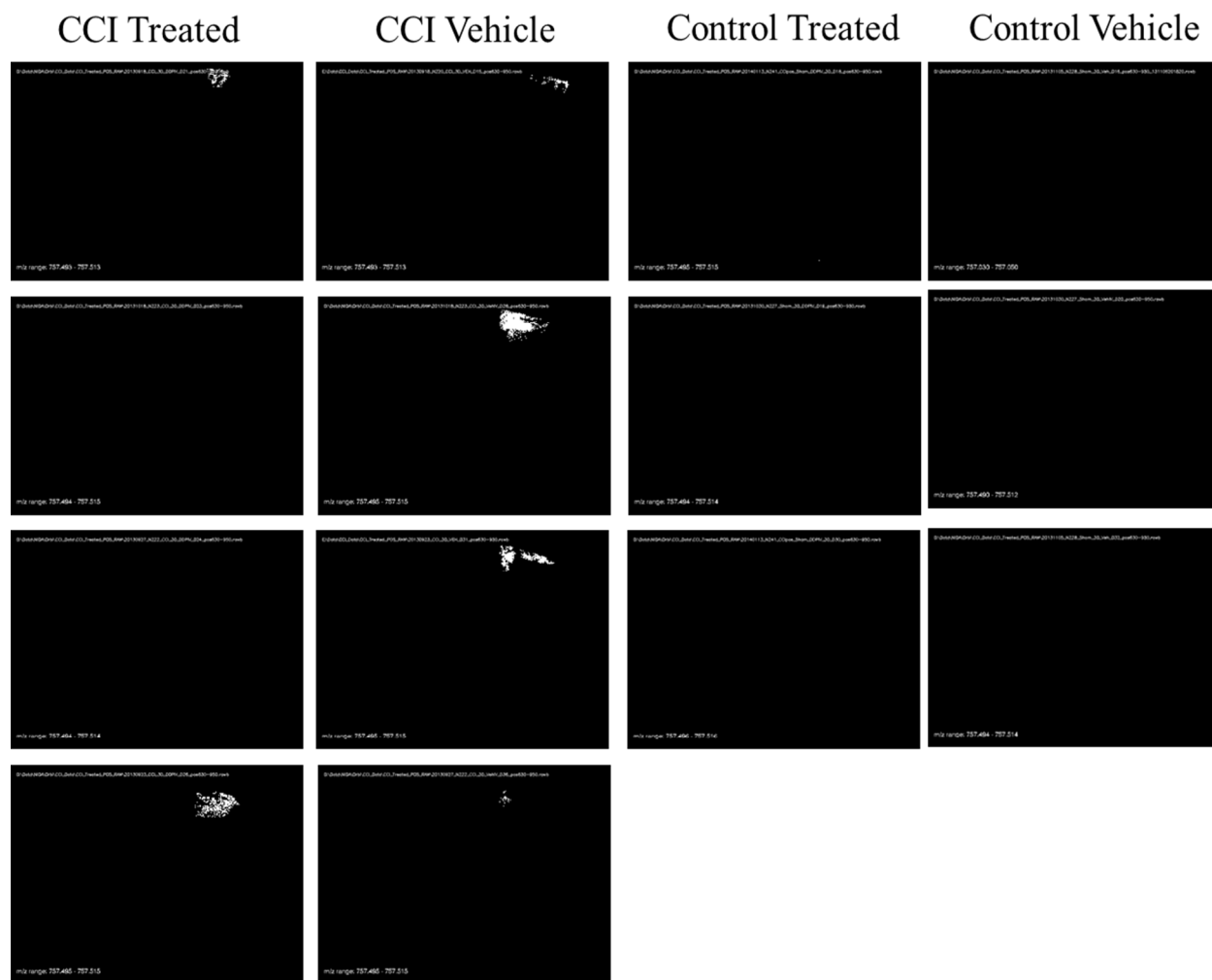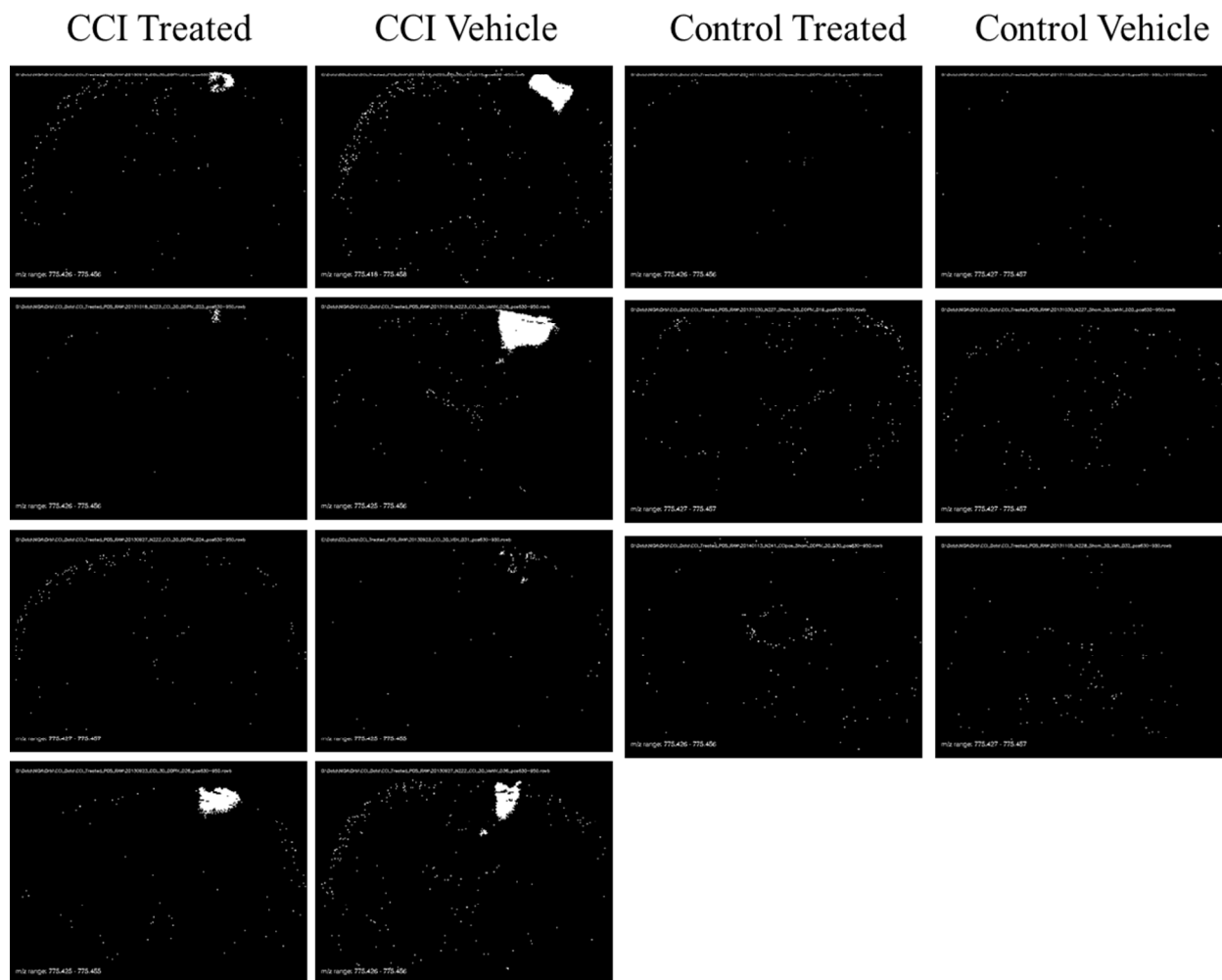
Figure S5. The distribution of DAG 40:a6 (m/z 775.4430) displayed as binary images of pixels exceeding peak area threshold. Black pixels represent peak area less than or equal to 381000 counts and white pixels represent peak areas greater than 381000 counts.
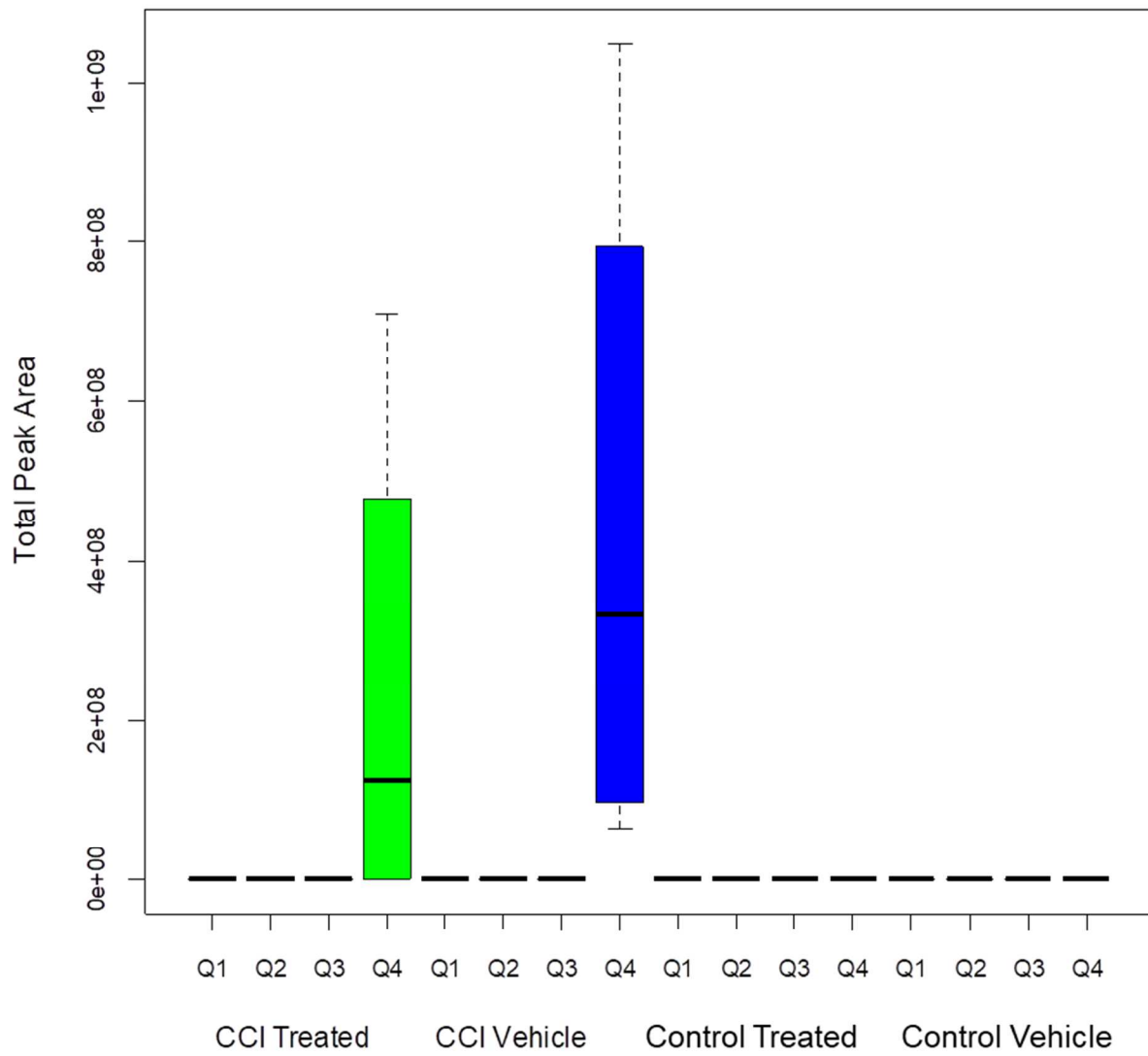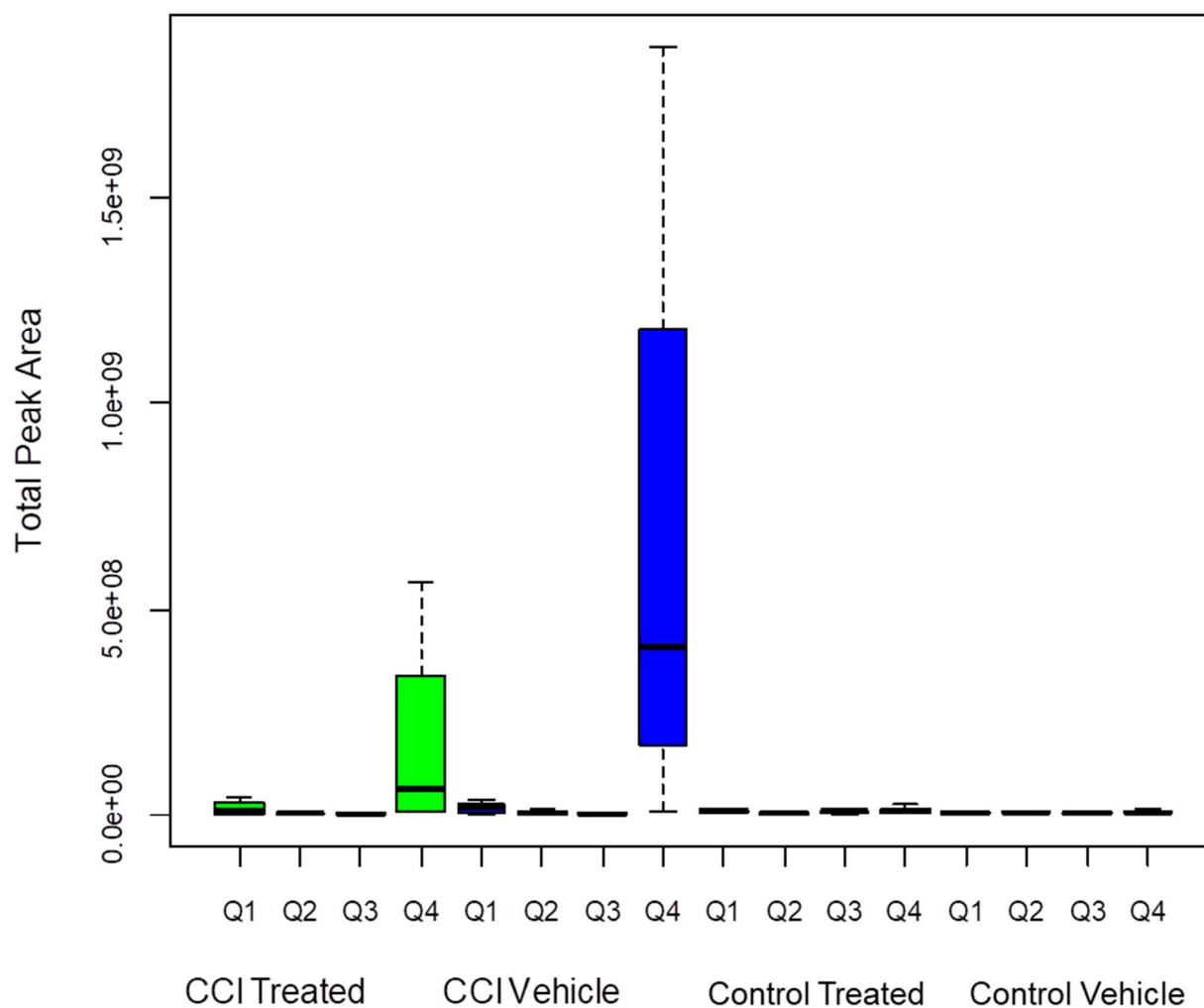
Figure S6. Total Peak Area for pixels greater than threshold for DAG 40:a6. CCI Vehicle and CCI Treated Q4 are significantly different than Q4 of Control Treated (p=0.048) and Control Vehicle (p=0.044) Arms. CCI Vehicle Q4 is not significantly different from CCI Treated Q4.
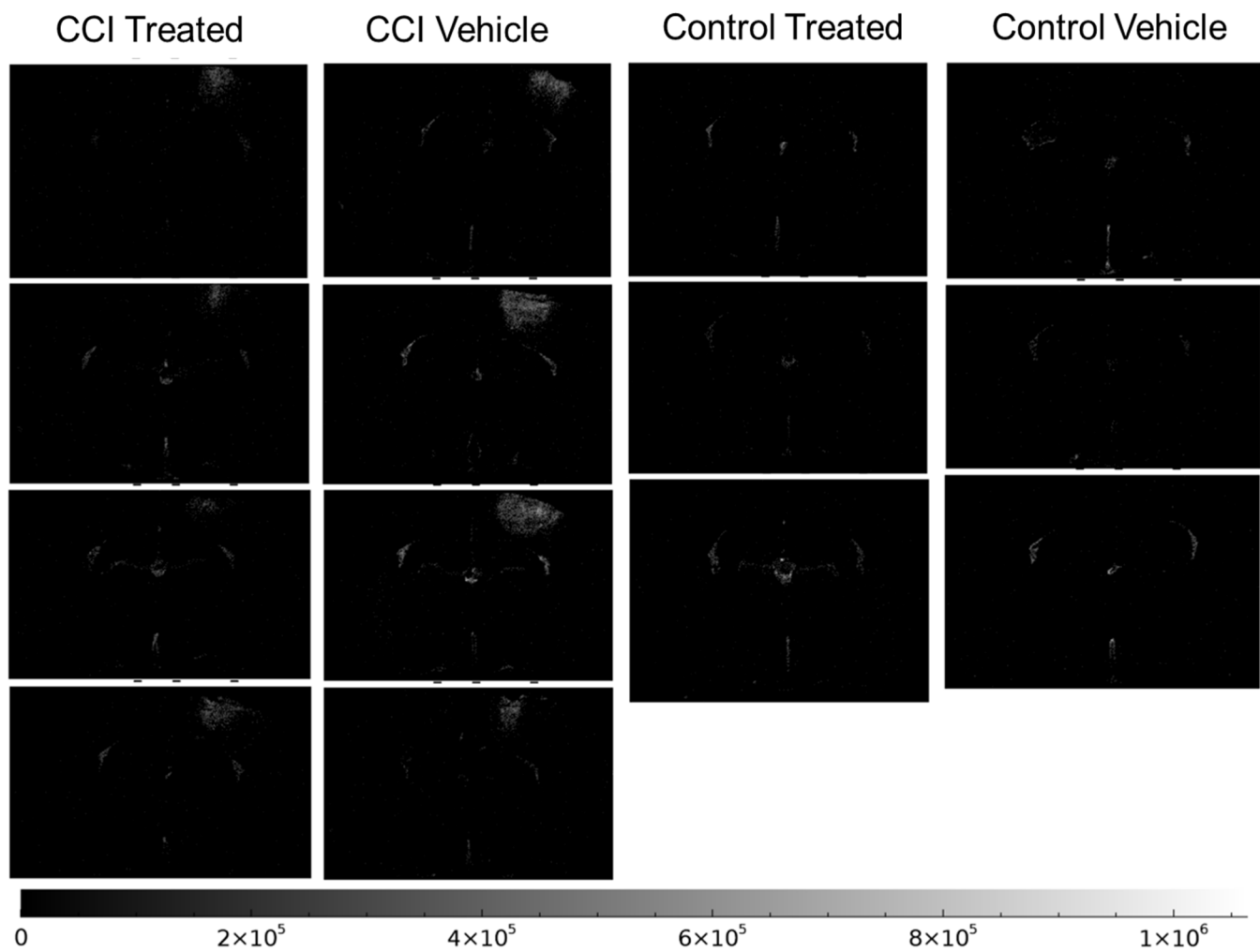
Figure S7. CER 34:1 (m/z 644.4171) 8-bit grayscale mass images of with intensity scale.
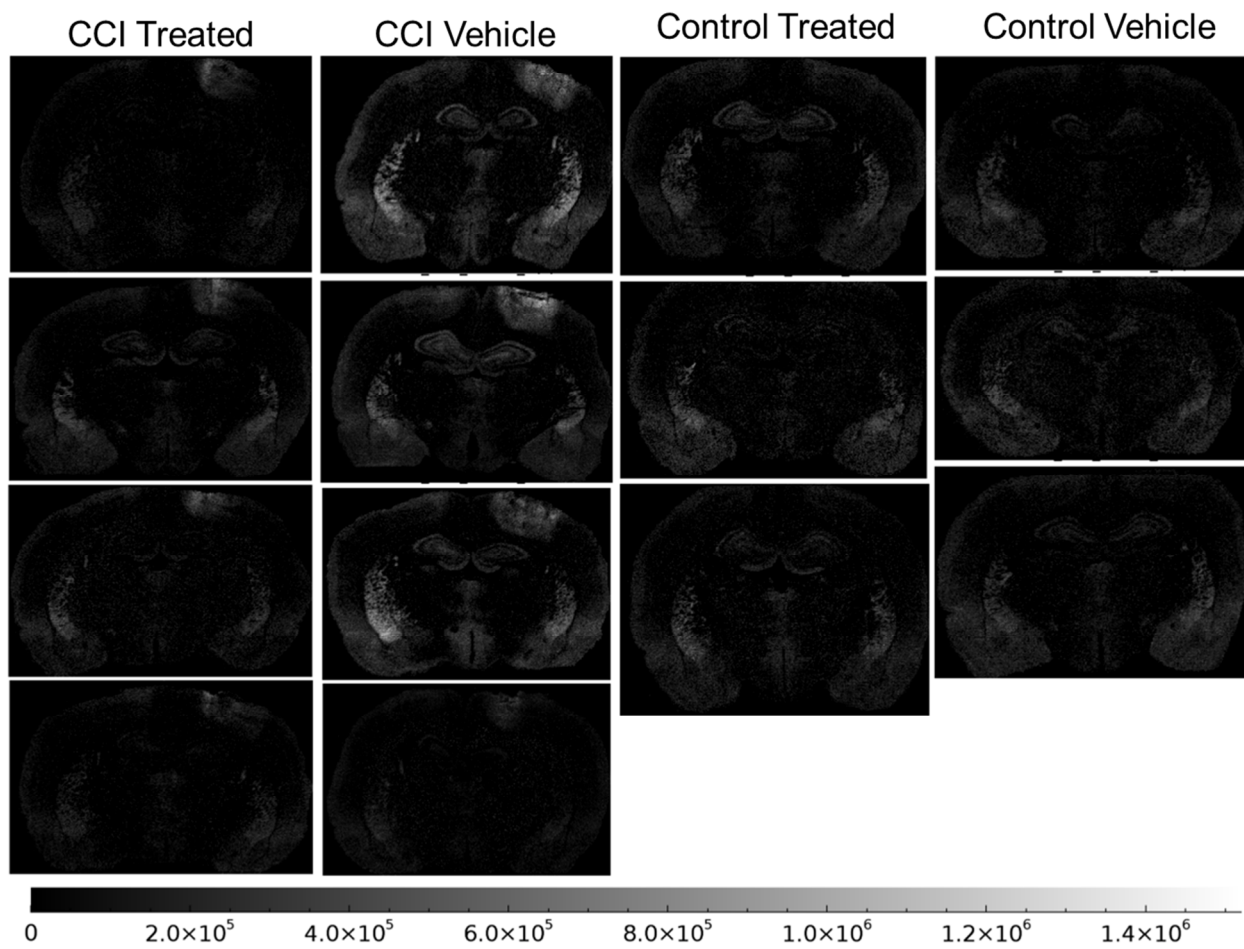
Figure S8. CER 36:2 (m/z 670.4328) 8-bit grayscale mass images of each sample with intensity scale.

Figure S9. CER 36:1 (m/z 674.4481) 8-bit grayscale mass images of each sample with intensity scale.

Figure S10. CER 38:1 (m/z 700.4797) 8-bit grayscale mass images of each sample with intensity scale.

Figure S11. Overlay of empirical cumulative distribution functions for CER 36:1 (m/z 674.4481) for each sample group.

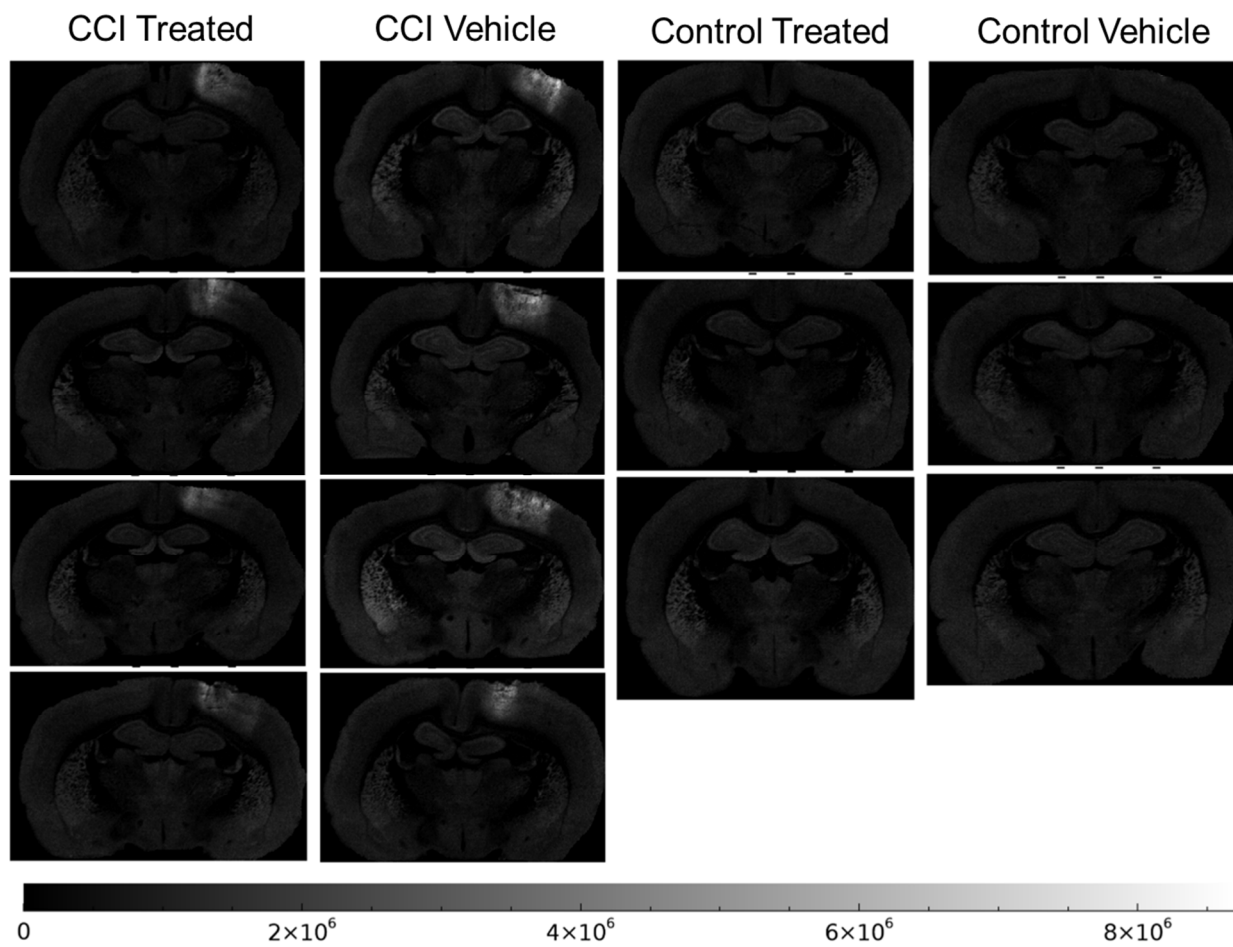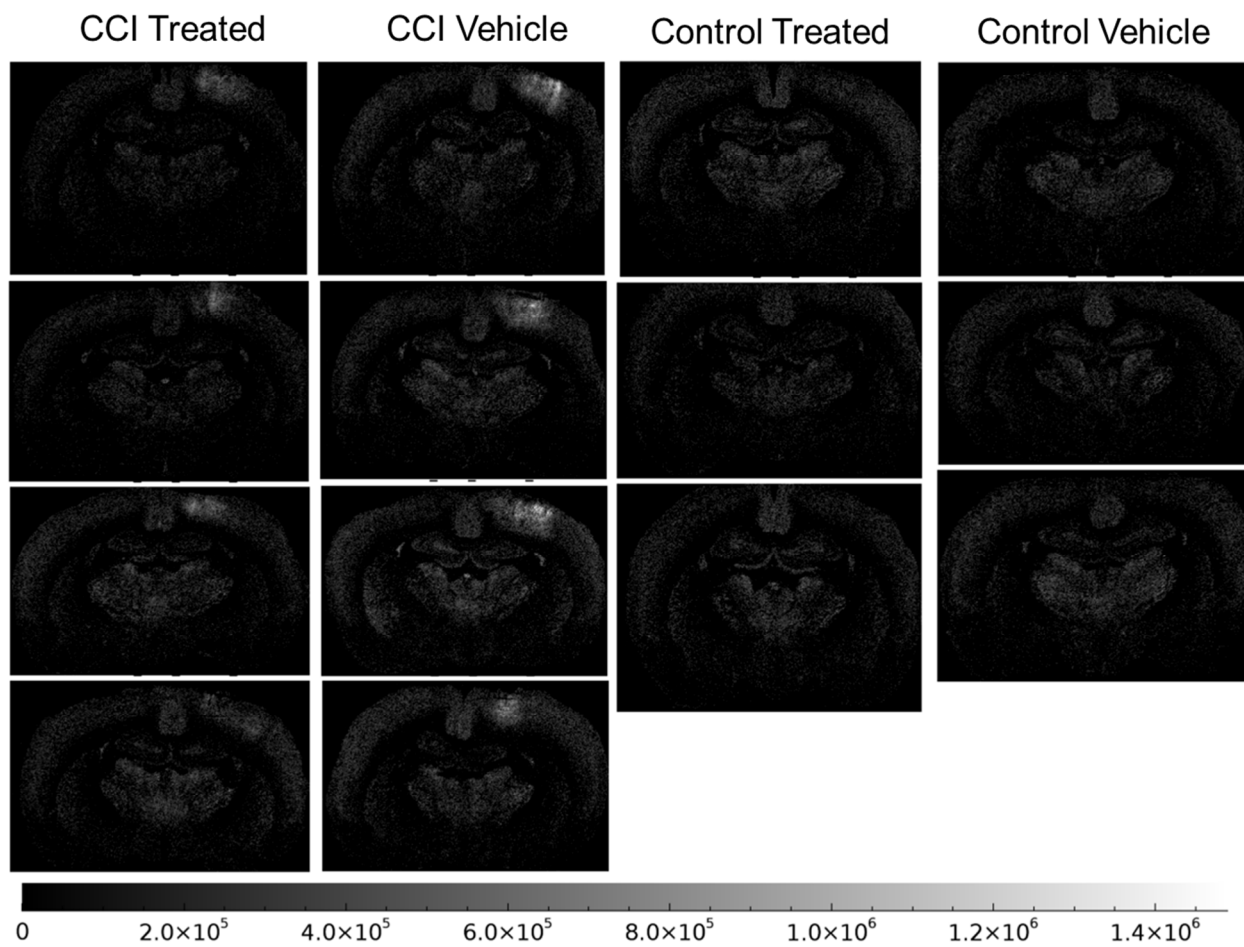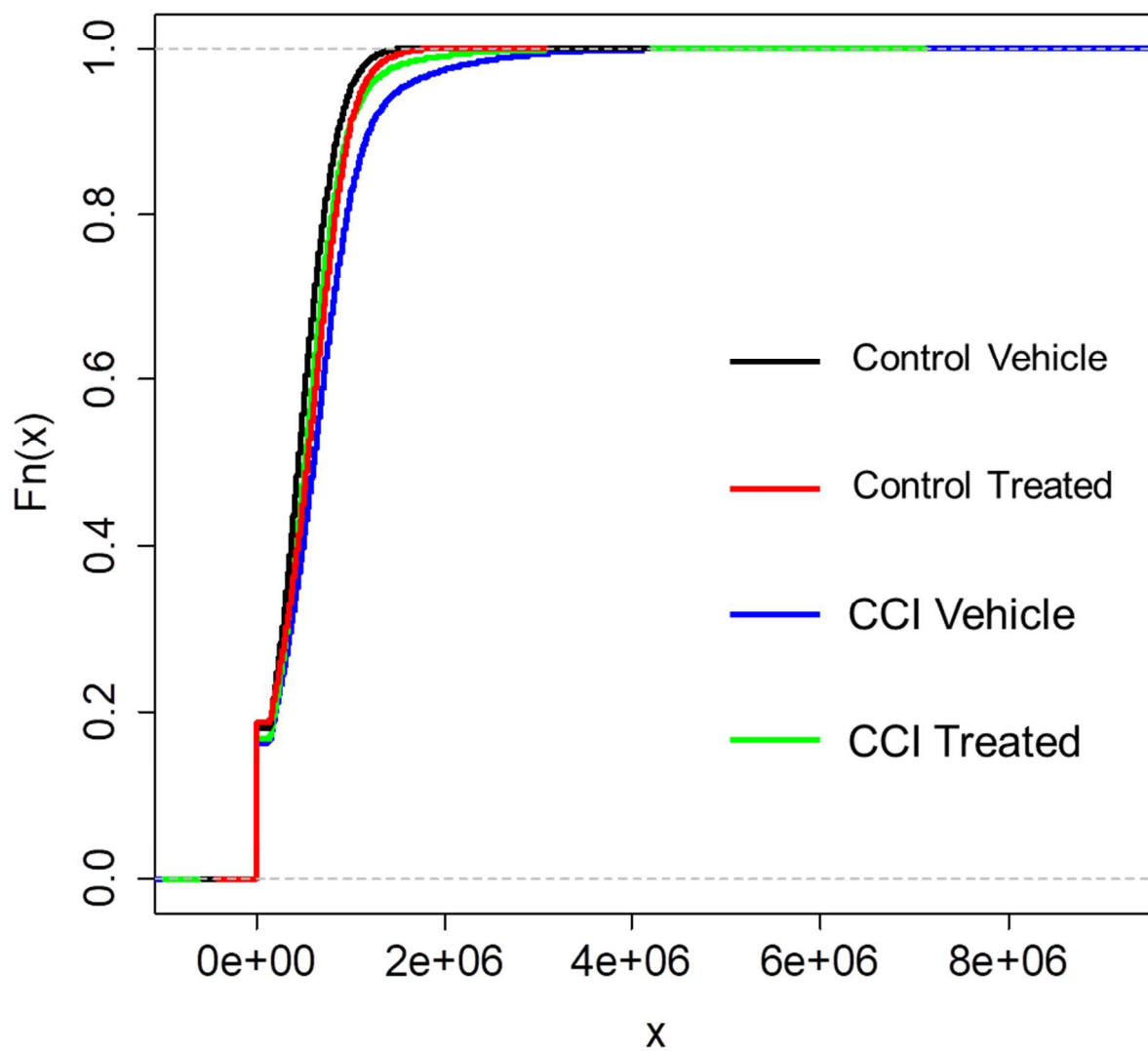| Arm	Quadrant	# Pixel				Total Area			
		CER34:1	CER36:2	CER36:1	CER38:1	CER34:1	CER36:2	CER36:1	CER38:1
CCITreated	Q1	32	7	60	199	5840000	2983143	80280000	69333000
		117	33	133	177	24801000	13820826	186390000	62003000
		205	17	200	485	42956000	7098788	280830000	176040000
		87	0	118	325	18594000	0	155760000	117515000
	Q2	23	49	190	53	3527000	20233702	257140000	18300000
		40	458	400	120	6908000	215529963	586880000	42990000
		83	183	461	365	19576000	79587013	662780000	134436000
		36	19	356	328	7037000	7732824	481210000	119617000
	Q3	15	5	37	50	2386000	2116451	48950000	17258000
		100	291	294	117	22687000	130971752	410950000	41528000
		24	19	209	246	4200000	7876140	288210000	90911000
		7	48	814	412	1075000	19882562	1116220000	153241000
	Q4	571	397	1481	613	125406000	200606999	3062610000	245029000
		471	560	1461	571	102036000	303951204	2879240000	233205000
		318	181	1091	914	64920000	81436873	2162890000	377827000
		634	55	1170	615	136137000	24928093	1988520000	228207000
CCIVehicle	Q1	84	447	900	471	19288000	218820158	1324080000	169470000
		166	320	397	472	44034000	141219513	546740000	172838000
		238	137	877	656	61517000	61566162	1339880000	242886000
		64	18	247	655	12514000	7444390	331580000	239645000
	Q2	68	992	626	103	13666000	517709160	969260000	36770000
		59	1539	796	319	10591000	783294030	1170380000	117832000
		57	1279	2475	556	10185000	699951379	4331440000	206169000
		28	154	406	316	4814000	65928498	554030000	116006000
	Q3	17	804	645	45	2859000	428361620	999370000	16017000
		53	1031	421	292	9642000	515585830	601660000	106795000
		90	669	1647	492	17325000	307858415	2424880000	184053000
		57	26	186	387	10623000	10347725	250240000	148546000
	Q4	739	1113	2083	1227	184257000	608375925	5002110000	582061000
		1542	1282	2404	1487	399213000	684099473	5384290000	656659000
		1754	471	2802	1850	449232000	214116951	5868060000	867615000
		510	344	1632	1242	113302000	161485037	3273510000	523072000
Control Treated	Q1	81	124	381	589	20293000	54303574	537510000	214258000
		64	13	78	199	12356000	6168149	113390000	70323000
		314	20	295	445	73252000	8225361	411590000	163259000
	Q2	65	99	283	218	13724000	42582691	396010000	80266000
		15	75	247	97	2443000	31340398	338050000	33799000
		21	341	924	221	3824000	155773326	1384580000	79827000
	Q3	12	138	442	353	1873000	58998013	606700000	133692000
		30	27	213	107	5935000	10823610	288690000	38469000
		79	284	896	248	16388000	125682336	1307370000	91117000
	Q4	116	37	279	750	27915000	15966925	380340000	276284000
		76	0	37	278	15244000	0	49370000	97760000
		208	20	291	439	45786000	7809978	407740000	161072000
Control Vehicle	Q1	111	10	73	297	23464000	3954762	101820000	107110000
		67	4	94	315	13540000	1622618	127330000	112990000
		120	3	149	421	34253000	1160692	196970000	152045000
	Q2	44	111	382	268	8375000	48028566	535000000	97981000
		35	87	365	102	7569000	37125466	498440000	36327000
		59	166	755	416	15231000	69813512	1028220000	158011000
	Q3	105	56	180	269	26239000	23320126	244180000	98604000
		25	28	168	110	4173000	11539144	223050000	41075000
		37	222	748	303	6924000	94432847	1038760000	115776000
	Q4	107	3	55	322	23132000	1200731	76700000	114928000
		41	5	74	335	7847000	1980391	100190000	120937000
		112	11	118	402	27657000	4756766	162370000	145999000

Table S1 Number of Pixels and Area Counts per Quadrant per Arm for each CER.