

Prenatal exposure to lipopolysaccharide or valproate leads to abnormal accumulation of the NMDA receptor agonist D-aspartate in the adolescent rat brain

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Supplementary Table 1. Statistical analysis of amino acid levels in the plasma of LPS-treated rats.

Amino acid	Age	Saline (μM)			LPS (μM)			Two-way ANOVA		
		Median	IQR		Median	IQR		Factor	F (DFn, DFd)	p value
D-Aspartate	P35	0.1718	0.1139	0.2007	0.1139	0.06661	0.1971	Treatment Age Treatment x Age	- - -	- - -
	P75	-	-	-	-	-	-			
L-Aspartate	P35	2.497	1.884	4.006	2.160	1.464	4.741	Treatment Age Treatment x Age	0.199 (1, 18) 0.0006 (1, 18) 0.128 (1, 18)	0.6605 0.9793 0.7239
	P75	2.604	2.084	3.051	3.219	2.175	3.664			
D-Asp/total Asp (%)	P35	5.702	5.095	6.516	4.922	2.834	5.216	Treatment Age Treatment x Age	- - -	- - -
	P75	-	-	-	-	-	-			
L-Asparagine	P35	15.64	10.66	27.93	13.98	9.916	27.33	Treatment Age Treatment x Age	0.061 (1, 18) 0.371 (1, 18) 0.014 (1, 18)	0.8073 0.5500 0.9048
	P75	20.05	15.14	23.98	21.44	17.19	24.52			
D-Serine	P35	1.382	1.311	1.466	1.484	1.270	1.729	Treatment Age Treatment x Age	0.966 (1, 18) 8.412 (1, 18) 0.048 (1, 18)	0.3385 0.0095 0.8277
	P75	1.148	0.9573	1.172	1.275	0.9978	1.371			
L-Serine	P35	100.4	62.53	141.1	92.54	61.81	173.7	Treatment Age Treatment x Age	0.584 (1, 18) 0.002 (1, 18) 0.015 (1, 18)	0.4544 0.9629 0.9026
	P75	106.7	81.11	120.3	124.6	94.84	141.4			
D-Ser/total Ser (%)	P35	1.353	0.9548	2.179	1.392	1.011	2.017	Treatment Age Treatment x Age	0.076 (1, 18) 6.194 (1, 18) 0.003 (1, 18)	0.7849 0.0228 0.9553
	P75	1.097	0.9522	1.179	1.005	0.9238	1.104			
Glycine	P35	245.8	166.2	349.6	204.6	154.8	270.5	Treatment Age Treatment x Age	0.155 (1, 18) 3.133 (1, 18) 2.052 (1, 18)	0.6980 0.0937 0.1692
	P75	161.5	150.4	175.7	192.5	167.8	227.5			
L-Glutamate	P35	26.94	19.83	33.56	21.04	15.54	34.00	Treatment Age Treatment x Age	0.003 (1, 18) 3.782 (1, 18) 0.262 (1, 18)	0.9512 0.0676 0.6145
	P75	34.91	26.22	38.29	33.54	26.17	43.63			
L-Glutamine	P35	200.5	124.0	290.4	158.0	113.6	297.0	Treatment Age Treatment x Age	0.132 (1, 18) 2.498 (1, 18) 0.255 (1, 18)	0.7200 0.1314 0.6195
	P75	238.7	193.9	293.5	275.0	230.3	315.5			
L-Glutamine/ L-Glutamate	P35	7.618	6.982	8.231	7.576	7.132	8.707	Treatment Age Treatment x Age	1.346 (1, 18) 0.068 (1, 18) 0.286 (1, 18)	0.2612 0.7961 0.5990
	P75	7.401	6.893	7.888	7.926	6.746	9.832			

Supplementary Table 2. Statistical analysis of amino acid levels in the plasma of VPA-treated rats.

Amino acid	Age	Saline (μM)			VPA (μM)			Two-way ANOVA		
		Median	IQR		Median	IQR		Factor	F (DFn, DFd)	p value
D-Aspartate	P40	-	-	-	-	-	-	Treatment	-	-
	P90	-	-	-	-	-	-	Age	-	-
								Treatment x Age	-	-
L-Aspartate	P40	6.652	5.463	7.743	5.726	5.315	9.035	Treatment	0.756 (1, 16)	0.3972
	P90	7.275	5.709	7.976	5.173	4.665	6.444	Age	0.648 (1, 16)	0.4324
								Treatment x Age	1.618 (1, 16)	0.2215
D-Asp/total Asp (%)	P40	-	-	-	-	-	-	Treatment	-	-
	P90	-	-	-	-	-	-	Age	-	-
								Treatment x Age	-	-
L-Asparagine	P40	13.30	10.53	21.84	19.07	13.15	29.61	Treatment	0.099 (1, 16)	0.7569
	P90	16.79	11.99	22.88	11.40	8.674	20.33	Age	0.794 (1, 16)	0.3861
								Treatment x Age	2.129 (1, 16)	0.1639
D-Serine	P40	2.585	2.278	3.167	2.892	2.299	3.296	Treatment	1.595 (1, 16)	0.2248
	P90	1.821	1.702	2.265	2.506	1.956	2.603	Age	9.928 (1, 16)	0.0062
								Treatment x Age	0.414 (1, 16)	0.5287
L-Serine	P40	103.2	77.31	151.8	106.1	78.77	161.6	Treatment	0.812 (1, 16)	0.3808
	P90	128.1	84.79	144.4	72.86	56.62	112.3	Age	0.820 (1, 16)	0.3786
								Treatment x Age	1.457 (1, 16)	0.2450
D-Ser/total Ser (%)	P40	2.904	1.503	3.641	2.735	1.433	3.963	Treatment	2.282 (1, 16)	0.1504
	P90	1.718	1.399	1.968	2.748	2.107	4.044	Age	0.491 (1, 16)	0.4933
								Treatment x Age	1.860 (1, 16)	0.1915
Glycine	P40	227.6	186.1	260.5	225.7	180.6	261.2	Treatment	1.504 (1, 16)	0.2378
	P90	197.8	140.8	222.0	140.1	102.6	177.9	Age	10.09 (1, 16)	0.0059
								Treatment x Age	1.223 (1, 16)	0.2851
L-Glutamate	P40	3.873	3.336	3.959	3.545	3.385	4.159	Treatment	0.645 (1, 16)	0.4334
	P90	4.083	3.490	4.216	3.629	3.162	3.974	Age	0.028 (1, 16)	0.8673
								Treatment x Age	0.988 (1, 16)	0.3349
L-Glutamine	P40	186.0	125.7	216.8	160.0	121.8	238.2	Treatment	0.871 (1, 16)	0.3645
	P90	210.6	164.1	262.0	149.7	117.1	221.9	Age	0.331 (1, 16)	0.5728
								Treatment x Age	1.013 (1, 16)	0.3291
L-Glutamine/ L-Glutamate	P40	48.03	37.54	54.73	46.19	35.55	57.17	Treatment	0.954 (1, 16)	0.3432
	P90	51.44	47.07	62.48	42.08	36.74	55.87	Age	0.552 (1, 16)	0.4681
								Treatment x Age	0.873 (1, 16)	0.3638

Supplementary Table 3. Statistical analysis of amino acid levels in the feces of LPS-treated rats.

Amino acid	Age	Saline (nmol/g feces)			LPS (nmol/g feces)			Two-way ANOVA		
		Median	IQR		Median	IQR		Factor	F (DFn, DFd)	p value
D-Aspartate	P40	25.99	17.73	31.87	22.56	14.67	37.63	Treatment Age Treatment x Age	0.278 (1, 36) 7.516 (1, 36) 0.073 (1, 36)	0.6007 0.0095 0.7877
	P90	11.86	8.769	18.71	12.46	7.013	19.07			
L-Aspartate	P40	59.13	46.90	95.57	72.03	58.34	97.78	Treatment Age Treatment x Age	0.182 (1, 36) 5.561 (1, 36) 0.912 (1, 36)	0.6715 0.0239 0.3459
	P90	43.29	32.41	84.41	47.71	34.64	64.32			
D-Asp/total Asp (%)	P40	25.47	22.07	31.56	27.25	14.72	28.59	Treatment Age Treatment x Age	1.685 (1, 36) 3.413 (1, 36) 0.017 (1, 36)	0.2025 0.0729 0.8962
	P90	19.74	17.40	27.80	21.19	15.32	23.60			
L-Asparagine	P40	2.477	1.876	5.301	3.240	1.444	5.997	Treatment Age Treatment x Age	0.116 (1, 35) 3.837 (1, 35) 0.016 (1, 35)	0.7348 0.0581 0.8995
	P90	1.855	1.233	3.149	1.611	0.7036	3.375			
D-Serine	P40	2.252	1.550	3.535	1.720	0.9791	3.194	Treatment Age Treatment x Age	0.008 (1, 35) 0.077 (1, 35) 0.988 (1, 35)	0.9281 0.7818 0.3270
	P90	2.028	0.9140	2.667	2.178	1.315	3.644			
L-Serine	P40	30.88	22.94	80.04	26.33	25.58	44.25	Treatment Age Treatment x Age	2.120 (1, 35) 2.001 (1, 35) 0.436 (1, 35)	0.1543 0.1660 0.5130
	P90	21.51	19.50	55.01	24.78	15.37	37.15			
D-Ser/total Ser (%)	P40	6.250	2.679	8.559	4.418	2.938	8.768	Treatment Age Treatment x Age	0.359 (1, 34) 1.805 (1, 34) 0.949 (1, 34)	0.5528 0.1880 0.3368
	P90	5.094	3.765	8.873	8.306	5.331	9.344			
Glycine	P40	41.96	13.92	148.2	73.01	30.73	121.7	Treatment Age Treatment x Age	1.804 (1, 34) 0.085 (1, 34) 0.110 (1, 34)	0.1881 0.7718 0.7416
	P90	69.13	27.17	133.5	48.81	5.872	84.69			
L-Glutamate	P40	308.7	194.5	421.6	244.0	211.4	387.0	Treatment Age Treatment x Age	0.006 (1, 36) 0.554 (1, 36) 3.1e-005 (1, 36)	0.9385 0.4614 0.9956
	P90	264.5	141.6	420.0	225.1	177.1	445.0			
L-Glutamine	P40	29.01	12.10	38.67	23.35	18.98	43.02	Treatment Age Treatment x Age	0.020 (1, 36) 3.017 (1, 36) 0.029 (1, 36)	0.8875 0.0910 0.8645
	P90	18.84	10.52	31.86	19.23	16.69	31.94			
L-Glutamine/ L-Glutamate	P40	0.08979	0.05702	0.1229	0.08913	0.07298	0.1142	Treatment Age Treatment x Age	0.107 (1, 36) 2.181 (1, 36) 0.185 (1, 36)	0.7449 0.1484 0.6695
	P90	0.06673	0.05405	0.08768	0.08946	0.05688	0.1125			

Supplementary Table 4. Statistical analysis of amino acid levels in the feces of VPA-treated rats.

Amino acid	Age	Saline (nmol/g feces)			VPA (nmol/g feces)			Two-way ANOVA		
		Median	IQR		Median	IQR		Factor	F (DFn, DFd)	p value
D-Aspartate	P30	13.73	8.786	17.19	11.49	8.504	28.53	Treatment Age Treatment x Age	3.235 (1, 36) 0.022 (1, 36) 0.0001 (1, 36)	0.0805 0.8827 0.9906
	P80	12.03	4.779	16.88	14.85	8.253	46.86			
L-Aspartate	P30	39.89	29.47	78.19	48.51	25.78	76.10	Treatment Age Treatment x Age	0.928 (1, 36) 0.329 (1, 36) 0.004 (1, 36)	0.3417 0.5694 0.9448
	P80	39.32	21.46	58.33	44.32	26.03	98.28			
D-Asp/total Asp (%)	P30	21.77	17.69	27.44	27.49	16.14	31.25	Treatment Age Treatment x Age	2.629 (1, 36) 0.638 (1, 36) 0.619 (1, 36)	0.1137 0.4296 0.4363
	P80	21.93	16.19	29.05	30.98	20.77	34.92			
L-Asparagine	P30	4.655	3.166	7.786	4.787	2.611	9.676	Treatment Age Treatment x Age	0.893 (1, 36) 1.030 (1, 36) 0.052 (1, 36)	0.3508 0.3170 0.8197
	P80	2.979	2.212	5.578	4.494	2.313	7.610			
D-Serine	P30	1.229	0.6118	2.200	1.269	0.7242	3.061	Treatment Age Treatment x Age	0.807 (1, 36) 0.344 (1, 36) 0.106 (1, 36)	0.3748 0.5607 0.7460
	P80	1.520	0.8087	2.786	2.155	1.115	4.149			
L-Serine	P30	22.86	16.62	61.44	26.84	15.46	46.14	Treatment Age Treatment x Age	0.001 (1, 36) 2.039 (1, 36) 1.292 (1, 36)	0.9719 0.1619 0.2632
	P80	21.01	9.348	27.91	27.21	11.87	45.15			
D-Ser/total Ser (%)	P30	7.127	1.011	10.92	5.148	3.183	8.061	Treatment Age Treatment x Age	0.345 (1, 36) 3.086 (1, 36) 0.060 (1, 36)	0.5602 0.0875 0.8070
	P80	7.456	5.836	11.67	8.751	7.149	10.53			
Glycine	P30	129.0	11.16	412.6	161.9	21.77	358.6	Treatment Age Treatment x Age	0.016 (1, 36) 4.250 (1, 36) 0.134 (1, 36)	0.8999 0.0465 0.7161
	P80	54.86	26.70	127.4	76.82	29.30	193.2			
L-Glutamate	P30	291.7	160.4	420.1	359.9	105.2	710.0	Treatment Age Treatment x Age	0.327 (1, 36) 0.469 (1, 36) 0.426 (1, 36)	0.5708 0.4976 0.5178
	P80	256.6	199.4	460.0	280.3	200.3	651.7			
L-Glutamine	P30	17.41	13.25	36.58	20.93	12.97	41.12	Treatment Age Treatment x Age	1.124 (1, 36) 6e-005 (1, 36) 0.953 (1, 36)	0.2961 0.9938 0.3354
	P80	14.90	12.84	29.17	25.28	12.94	53.27			
L-Glutamine/ L-Glutamate	P30	0.08487	0.06994	0.1026	0.07781	0.05587	0.1271	Treatment Age Treatment x Age	0.719 (1, 36) 2.010 (1, 36) 0.773 (1, 36)	0.4019 0.1649 0.3850
	P80	0.05776	0.03090	0.08157	0.07228	0.05163	0.09342			

Supplementary Table 5. Statistical analysis of DASPO, DAAO and SR activity and predicted enzyme concentrations in the prefrontal cortex and dorsal striatum of LPS-treated rats.

Enzyme	Tissue	Age	Saline ($\mu\text{U}/\mu\text{g protein}$) [ng enzyme/ $\mu\text{g protein}$]		LPS ($\mu\text{U}/\mu\text{g protein}$) [ng enzyme/ $\mu\text{g protein}$]		Two-way ANOVA		
			Mean	SEM	Mean	SEM	Factor	F (DFn, DFd)	P value
DASPO	PFC	P35	0.0984 [0.00190]	0.0078 [0.00015]	0.1089 [0.00211]	0.0067 [0.00013]	Treatment Age Treatment x Age	2.19 (1, 20) 5.65 (1, 20) 7.16 (1, 20)	0.1540 0.0276 0.0144
		P75	0.1011 [0.00196]	0.0125 [0.00024]	0.0649 [0.00125]	0.0063 [0.00012]			
	STR	P35	0.0510 [0.000986]	0.0053 [0.00010]	0.0468 [0.000906]	0.0045 [0.00009]	Treatment Age Treatment x Age	2.699 (1, 19) 7.130 (1, 19) 0.713 (1, 19)	0.1169 0.0149 0.4089
		P75	0.0692 [0.00134]	0.0064 [0.00012]	0.0563 [0.00109]	0.0047 [0.00009]			
DAAO	PFC	P35	0.0076 [0.00019]	0.0015 [0.00004]	0.0091 [0.00023]	0.0008 [0.00002]	Treatment Age Treatment x Age	0.14 (1, 20) 15.9 (1, 20) 1.77 (1, 20)	0.7079 0.0007 0.1981
		P75	0.0162 [0.00040]	0.0025 [0.00006]	0.0134 [0.00033]	0.0013 [0.00003]			
	STR	P35	0.0027 [0.000067]	0.0015 [0.000038]	0.00275 [0.000068]	0.0016 [0.000039]	Treatment Age Treatment x Age	0.166 (1, 19) 22.44 (1, 19) 0.171 (1, 19)	0.6886 0.0001 0.6835
		P75	0.0154 [0.000380]	0.0026 [0.000064]	0.0134 [0.00033]	0.0035 [0.000087]			
SR	PFC	P35	1.26 [0.574]	0.09 [0.043]	1.20 [0.543]	0.08 [0.039]	Treatment Age Treatment x Age	5.259 (1, 20) 0.636 (1, 20) 2.309 (1, 20)	0.0328 0.4345 0.1443
		P75	1.46 [0.672]	0.05 [0.026]	1.13 [0.515]	0.10 [0.045]			
	STR	P35	1.11 [0.502]	0.23 [0.107]	1.44 [0.652]	0.32 [0.145]	Treatment Age Treatment x Age	3.083 (1, 19) 1.526 (1, 19) 0.511 (1, 19)	0.0952 0.2318 0.4833
		P75	1.27 [0.578]	0.16 [0.071]	2.05 [0.934]	0.44 [0.198]			

Supplementary Table 6. Statistical analysis of DASPO, DAAO and SR activity and predicted enzyme concentrations in the prefrontal cortex and dorsal striatum of VPA-treated rats.

Enzyme	Tissue	Age	Saline ($\mu\text{U}/\mu\text{g protein}$) [ng enzyme/ $\mu\text{g protein}$]		VPA ($\mu\text{U}/\mu\text{g protein}$)[ng enzyme/ $\mu\text{g protein}$]		Two-way ANOVA		
			Mean	SEM	Mean	SEM	Factor	F (DFn, DFd)	P value
DASPO	PFC	P40	0.0998 [0.00193]	0.0090 [0.00017]	0.0903 [0.00174]	0.0090 [0.00018]	Treatment Age Treatment x Age	0.057 (1, 20) 0.599 (1, 20) 0.878 (1, 20)	0.8132 0.4480 0.3600
		P90	0.0860 [0.00166]	0.0072 [0.00014]	0.0917 [0.00177]	0.0067 [0.00013]			
	STR	P40	0.0693 [0.00134]	0.0056 [0.00011]	0.0782 [0.00151]	0.0047 [0.00009]	Treatment Age Treatment x Age	2.087 (1, 19) 0.0001 (1, 19) 0.164 (1, 19)	0.1649 0.9907 0.6905
		P90	0.0658 [0.00127]	0.0091 [0.00018]	0.0815 [0.00158]	0.0124 [0.00024]			
DAAO	PFC	P40	0.0145 [0.00036]	0.0002 [0.00001]	0.0131 [0.00032]	0.0007 [0.00002]	Treatment Age Treatment x Age	3.928 (1, 20) 32.93 (1, 20) 0.128 (1, 20)	0.0614 <0.0001 0.7240
		P90	0.0100 [0.00025]	0.0011 [0.00003]	0.0081 [0.00020]	0.0010 [0.00002]			
	STR	P40	0.0142 [0.00036]	0.0008 [0.00002]	0.0158 [0.00039]	0.0030 [0.00007]	Treatment Age Treatment x Age	0.1405 (1, 19) 7.866 (1, 19) 0.1020 (1, 19)	0.7119 0.0113 0.7530
		P90	0.0082 [0.00020]	0.0027 [0.00007]	0.0083 [0.00021]	0.0025 [0.00006]			
SR	PFC	P40	1.58 [0.718]	0.12 [0.056]	1.07 [0.487]	0.12 [0.056]	Treatment Age Treatment x Age	7.22 (1, 20) 13.1 (1, 20) 4.55 (1, 20)	0.0142 0.0017 0.0454
		P90	0.97 [0.442]	0.08 [0.035]	0.92 [0.415]	0.09 [0.041]			
	STR	P40	1.66 [0.755]	0.14 [0.066]	2.11 [0.956]	0.26 [0.118]	Treatment Age Treatment x Age	2.67 (1, 19) 22.1 (1, 19) 1.00 (1, 19)	0.1185 0.0002 0.3295
		P90	1.04 [0.473]	0.11 [0.049]	1.15 [0.5212]	0.08 [0.037]			

Supplementary Table 7. Statistical analysis of DASPO, DAAO and SR activity and predicted enzyme concentrations in the prefrontal cortex and dorsal striatum of *Fmr1-Δexon 8* rats.

Enzyme	Tissue	Wild-type (μU/μg protein) [ng enzyme/μg protein]		<i>Fmr1-Δexon 8</i> (μU/μg protein) [ng enzyme/μg protein]		Unpaired t-test	
		Mean	SEM	Mean	SEM	t, df	P value
DASPO	PFC	0.0380 [0.000736]	0.0040 [0.000077]	0.0280 [0.000541]	0.0072 [0.000139]	1.224, 10	0.1246
	HIPP	0.0401 [0.000776]	0.0082 [0.000160]	0.0383 [0.000741]	0.0032 [0.000063]	0.209, 10	0.4194
	NAc	0.0146 [0.000283]	0.0031 [0.000060]	0.0141 [0.000273]	0.0033 [0.000063]	0.117, 10	0.4547
	STR	0.0252 [0.000488]	0.0104 [0.000200]	0.0173 [0.000335]	0.0027 [0.000053]	0.736, 10	0.2392
DAAO	PFC	0.0025 [0.000061]	0.0005 [0.000011]	0.0027 [0.000066]	0.0008 [0.000019]	0.238, 10	0.4084
	HIPP	0.0017 [0.000043]	0.0005 [0.000011]	0.0037 [0.000091]	0.0004 [0.000010]	3.151, 10	0.0052
	NAc	0.0012 [0.000029]	0.0007 [0.000017]	0.0023 [0.000056]	0.0006 [0.000015]	1.196, 10	0.1296
	STR	0.0031 [0.000077]	0.0013 [0.000032]	0.0025 [0.000060]	0.0008 [0.000019]	0.438, 10	0.3355
SR	PFC	0.464 [0.211]	0.098 [0.044]	0.518 [0.235]	0.115 [0.052]	0.358, 10	0.3638
	HIPP	0.619 [0.282]	0.109 [0.050]	0.479 [0.218]	0.086 [0.039]	1.011, 10	0.1680
	NAc	0.754 [0.343]	0.140 [0.063]	0.737 [0.335]	0.169 [0.077]	0.076, 10	0.4703
	STR	0.646 [0.294]	0.114 [0.052]	0.486 [0.221]	0.117 [0.053]	0.981, 10	0.1749