

Choroid plexus LAT2 and SNAT3 as partners in CSF amino acid homeostasis maintenance

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Additional file 2 Supplementary tables

Table S1: Antibody dilutions used

Target protein	Target sequence	Species of target protein	Host	Dilution	Manufacturer
SNAT3 (SLC38A3)	MEIPRQTEMVELVPNGKC (N-terminus)	Mouse	Rabbit	1:100	In house via Pineda
SNAT1 (SLC38A1)	MMHFKSGLELTELQNMTVC (N-terminus)	Mouse	Rabbit	1:100	In house via Abent
LAT2 (SLC7A8)	CPIFKPTPVKDPDSEEQP (C-terminus)	Mouse	Rabbit	1:100, or 1:1000	In house via Eurogentech
AE2 (SLC4A2)	100-255 AA (N-terminus)	Human	Mouse	1:100	Santa Cruz Biotechnology SC-376632
Na ⁺ -K ⁺ -ATPase α (H-3)	551-850 AA	Human	Mouse	1:100	Santa Cruz Biotechnology SC-48345

Table S2: Primers and probes used for qRT-PCR

	Gene	Full name	Accession number	Sequence	
Markers					
1	<i>Cd31/Pecam1</i>	platelet/endothelial cell adhesion molecule 1	NM_008816	forward reverse probe	TCCCCGAAGCAGCACTCT CCGCAATGAGCCCTTTCTT TCAGAGTCTTCCTTGCCCCA
2	<i>Gfap</i>	glial fibrillary acidic protein	NM_010277	forward reverse probe	CACAGCGGCCCTGAGAGA GTCTGCAAACCTTAGACCGATACCA ACTCAATACGAGGCAGTGGCCACCAGTAA
3	<i>Syp</i>	Synaptophysin	NM_009305	forward reverse probe	GGGCCAATGATGGACTTCCT GGATGAGCTAACTAGCCACATGAA CACAGCAGTGTTTCGC
4	<i>Slc2a1</i>	Glut1	NM_011400	forward reverse probe	TCGTCGTTGGCATCCTTATTG TGCATTGCCCATGATGGA CCAGGTGTTTGGCTTAG
5	<i>Ttr</i>	Transthyretin	NM_013697.5	forward reverse probe	CATGAATTCGCGGATGTG GATGGTGTAGTGGCGATGG Roche probe#76

Transporters					
1	<i>Slc1a1</i>	Eaat3	NM_009199.2	forward reverse probe	GGTCTGCGCGCTGTAGTAT TGAGTGACACCAGGCTTGATA Roche probe #77
2	<i>Slc1a2</i>	Eaat2	NM_001077514	forward reverse probe	TGCTCATCCTCCCCTTATCATC GGCCGCTGGCTTTAGCAT AGTTTAATCACAGGGTTGTC
3	<i>Slc1a3</i>	Eaat1	NM_148938	forward reverse probe	GAAGCCATCATGCGATTGGT CCCTGCGATCAAGAAGAGGAT CGGTGATAATGTGGTATGC
4	<i>Slc1a4</i>	Asct1	NM_018861.3	forward reverse probe	TGGACTCTTTCCTCGATTACTCA AATCGGTTGCAGACGTAGTAA CCTGTTCCCTTCCAATCTTGTGGTTGC
5	<i>Slc1a5</i>	Asct2	D85044	forward reverse probe	CATCCTGGAAGCAGTCAGCC CACCTTCCACGTTGAGGACA TGATCTTGGCCGTGGACTGGCTAGTG
6	<i>Slc1a6</i>	Eaat4	NM_009200	forward reverse probe	CCCCAGGCAGGACTGGTTA TTGTACGAAGTCGGTCAAGGAA TGATTGTGCTCACATCCGTCGGC
7	<i>Slc6a5</i>	Glyt2	NM_148931.3	forward reverse probe	ATGTTTGGAGGTTTCCCTACC TCAAGTAAGGGATGAGGAACG Roche probe #97
8	<i>Slc6a6</i>	Taut	NM_009320	forward reverse probe	GCAACGTCTGGCGTTTCC AGGCCGCTCCCAAACAG TGC GTT CCTCATACCGT
9	<i>Slc6a7</i>	Prot	NM_201353.1	forward reverse probe	GGAAGGGCATCCAGTTCTATC AGCAGCTTCAATCCACACCT Roche probe #77
10	<i>Slc6a9</i>	Glyt1	NM_008135.4	forward reverse probe	AATGTCTGGCGTTTCCATA TGATGAAGTAGGGGAACATGAA Roche probe #52
11	<i>Slc6a20b</i>	SIT1	NM_011731.3	forward reverse probe	GGAAGCATGCTTGGAAACACAG AACACCATGCCAACAGCACA AGGAGGCCATTTAGGTTTGGTATGCCTC
12	<i>Slc7a1</i>	Cat1	NM_007513.4	forward reverse probe	GGCAGCTCACGGAGAAAAATT TCCCTCACCGTATTTACGTT TCCTGTAACAACAACGACAC
13	<i>Slc7a2</i>	Cat2	NM_007514.3	forward reverse probe	CATCACTGGCTGGAATCTCATC ACTCCATGCTCTTGC GACT TGTCATATGCATAGGTACGTC
14	<i>Slc7a3</i>	Cat3	NM_007515.2	forward reverse probe	ACTCCCTGGTGTCCATTTGTG TCTTCAACTCTTCATTTCTGATC TCTCATCCTCAGATATCA
15	<i>Slc7a4</i>	Cat4	NM_144852.3	forward reverse probe	GGCTCTATCTGTGCCATGAACA GCGCGGCAGGGAGAA TCCTGCTCAGCAACC
16	<i>Slc7a5</i>	Lat1	NM_011404	forward reverse probe	GGAGGATGGA ACTATCTGAATTTG GGTTGGTAGAGGGGTAGTGAATAGG CTACAGGAACCTGCCCTGGCCA

17	<i>Slc7a6</i>	y+Lat2	BC038404	forward reverse probe	TACATCCTGACCAACGTGGC ATGCCGAATGTCTGGTCAGC TCCATAAGAGTGACGCTGTGCCTGTGA
18	<i>Slc7a7</i>	y+Lat1	NM_011405	forward reverse probe	AATTCCAGTAGCGGTTGCATT GGAGGTGGCCTTCTCTCGAG TTGCTTTGGTGGGCTCAACGCC
19	<i>Slc7a8</i>	Lat2	NM_016972	forward reverse probe	TCCACGTTTGGTGGAGTCAA TCACACAACCGGTACTAGGT CTCCCTCTTCACCTCCTCCCGGCT
20	<i>Slc7a10</i>	Asc1	NM_017394.4	forward reverse probe	TGGCTGGAACCTCCTCAACT GATGGCACGAGGTAGGTTCT Roche probe #17
21	<i>Slc7a11</i>	xCT	NM_011990.2	forward reverse probe	ACCATCAGTGC GGAGGAG AAGATCGGGACTGCTAATGAGA Roche probe #3
22	<i>Slc7a12</i>	Asc2	NM_080852	forward reverse probe	CAGAACCCTGATATACCAAGACCT GGAAAAGTGATAAAGCTATAGATCCAA Roche probe #5
23	<i>Slc16a10</i>	TAT1	NM_028247.4	forward reverse probe	CGCCTACGGGGTGCTCTTC ACTCACGATGGGGCAGCAG CGAGCCCACCCACGCTGTCTTG
24	<i>Slc38a1</i>	Snat1	AF184240	forward reverse probe	CGCGTGCACACCAAAGTATG AGATTGGCAGGACGGACG TACCAACCATCGCCTTCGCGTTTG
25	<i>Slc38a2</i>	Snat2	BC041108	forward reverse probe	CCAATGAGATCCGTGCAAAA TGGACCCAATCCAGCACAAAT TCTGTGTTTTCTCCTGAGTGGCATAGTGGTG
26	<i>Slc38a3</i>	Snat3	AF159856	forward reverse probe	CGAATCATGCCACTGACAA AACCGCAGCGAAACAAAGG AGCCTGCAAGATCCACCCCTAAATCCT
27	<i>Slc38a4</i>	Snat4	AK051023	forward reverse probe	AGGGTGCTGAGA AATGCCAA GACCTCAGGGTGGCAGAC AA AATACTTTGTGTTCAATTCCCGGACGGC
28	<i>Slc38a5</i>	Snat5	AK054485	forward reverse probe	CCCTCACTGTGCCTGTTGTG CTTGCTTGAAAGAGCAGCTG CCCTATCCGCCGAGCCCTCCA
29	<i>Slc38a6</i>	Snat6	BC043932	forward reverse probe	CGCCGTGCTTCTGACAGCC GGGAATGTCGGATCCAGGA CTTTAATGATGATCCTGTTCTCCAATTACCCGTT
30	<i>Slc38a7</i>	Snat7	NM_172758.4	forward reverse probe	GGCTCTGACCCCATACCTTT GGGGAACCTTTGTCAACCTG Roche probe #6
31	<i>Slc43a1</i>	Lat3	NM_001081349	forward reverse probe	CCCTGAATGAGAATGCTTCCTT ATGGCATTGGT GAGCTTTTGT AGCACCAAGTTCCTAGACCACGCTACCG
32	<i>Slc43a2</i>	Lat4	NM_173388	forward reverse probe	GCTGATTGCATATGGAGCAAGTAAC CGAAGTGAACGT CATGCACAT CTCTCTGTGCTCATCTTTATCGCCTTGGC

Table S3: AA concentrations measured in CSF and plasma samples of wt and Lat2^{-/-} animals

Amino acids	Plasma concentrations, $\mu\text{mol/L} \pm\text{SD}$		CSF concentrations, $\mu\text{mol/L} \pm\text{SD}$	
	LAT2 ^{+/+}	LAT2 ^{-/-}	LAT2 ^{+/+}	LAT2 ^{-/-}
Ala	551.84 \pm 276.83	685.12 \pm 272.34	65.13 \pm 17.40	116.60 \pm 22.84*
Arg	105.74 \pm 22.15	118.54 \pm 25.76	24.60 \pm 5.35	37.18 \pm 6.95*
Asn	45.13 \pm 6.20	78.42 \pm 13.81**	5.28 \pm 0.84	9.09 \pm 1.39**
Asp	12.43 \pm 5.27	15.25 \pm 7.59	5.08 \pm 3.39	42.86 \pm 28.70*
Gln	738.08 \pm 143.83	1103.16 \pm 235.80*	553.43 \pm 141.16	1191.18 \pm 653.97
Glu	24.05 \pm 11.86	42.92 \pm 37.36	13.29 \pm 9.47	97.44 \pm 36.85**
Gly	280.23 \pm 48.56	344.90 \pm 67.12	27.14 \pm 9.30	101.53 \pm 21.88***
His	76.28 \pm 7.56	86.01 \pm 17.19	5.76 \pm 1.63	11.73 \pm 5.77
Leu	134.80 \pm 30.83	141.02 \pm 26.64	5.52 \pm 1.22	12.04 \pm 2.28**
Lys	286.73 \pm 54.65	318.88 \pm 74.51	48.28 \pm 13.46	62.87 \pm 16.61
Met	60.18 \pm 8.20	63.29 \pm 10.63	7.97 \pm 1.91	10.63 \pm 3.46
Phe	70.87 \pm 15.26	75.29 \pm 19.53	4.30 \pm 1.03	8.63 \pm 5.79
Pro	83.49 \pm 24.56	129.50 \pm 31.84	2.57 \pm 1.63	14.62 \pm 3.26***
Ser	143.03 \pm 16.32	181.59 \pm 19.50*	41.29 \pm 8.70	57.37 \pm 14.83
Thr	159.46 \pm 12.67	181.22 \pm 35.38	26.42 \pm 5.75	33.58 \pm 9.04
Trp	65.22 \pm 9.41	65.21 \pm 10.92	5.02 \pm 1.36	15.10 \pm 10.24
Tyr	73.34 \pm 15.35	84.14 \pm 27.84	7.35 \pm 1.18	13.55 \pm 6.31
Val	214.17 \pm 39.40	244.16 \pm 31.09	8.34 \pm 1.32	20.86 \pm 2.38***
Ile	88.20 \pm 20.92	94.97 \pm 16.00	not reliably detected in CSF	
Tau	486.92 \pm 212.65	586.43 \pm 220.02	207.24 \pm 128.24	713.53 \pm 243.52*

¹Data compared with unpaired t-test and statistically significant changes are indicated as ***p<0.001, ** p<0.01, *p<0.05.