

Name (Manually annotated)	KEGG pathway/ <i>compounds</i>	Formula	G93A/WT ratio	p-value	RT (min)	<i>m/z</i>	Adduct
Decanoic acid	<u>Fatty acid biosynthesis</u>	C <sub>10</sub> H <sub>20</sub> O <sub>2</sub>	1.703	2.85E-04	1.11	171.139	[M-H]-
Octanoic acid		C <sub>8</sub> H <sub>16</sub> O <sub>2</sub>	1.672	1.43E-07	1.112	143.1076	[M-H]-
Hexadecanoic acid		C <sub>16</sub> H <sub>32</sub> O <sub>2</sub>	1.663	1.73E-08	1.082	255.2329	[M-H]-
Dodecanoic acid		C <sub>12</sub> H <sub>24</sub> O <sub>2</sub>	1.632	2.00E-03	1.101	199.1702	[M-H]-
Hexadecenoic acid		C <sub>16</sub> H <sub>30</sub> O <sub>2</sub>	1.521	2.08E-07	1.084	253.2172	[M-H]-
Nonanoic acid	<u>Fatty acids</u>	C <sub>9</sub> H <sub>18</sub> O <sub>2</sub>	1.572	4.11E-04	1.115	157.1232	[M-H]-
Undecanoic acid	<u>Fatty acids</u>	C <sub>11</sub> H <sub>22</sub> O <sub>2</sub>	1.549	3.99E-03	1.105	185.1546	[M-H]-
Oxoglutaric acid	<u>Alanine, aspartate and glutamate metabolism</u>	C <sub>5</sub> H <sub>6</sub> O <sub>5</sub>	0.58	3.38E-08	1.41	145.0142	[M-H]-
Oxoglutaramic acid		C <sub>5</sub> H <sub>7</sub> NO <sub>4</sub>	1.5	9.66E-05	8.083	126.0196	[M-H <sub>2</sub> O-H]-
Succinic acid semialdehyde		C <sub>4</sub> H <sub>6</sub> O <sub>3</sub>	0.529	2.92E-13	1.42	101.0239	[M-H]-
Isoprene	<u>Biosynthesis of terpenoids and steroids</u>	C <sub>5</sub> H <sub>8</sub>	0.452	5.72E-04	1.352	127.0764	[M+CH <sub>3</sub> COO]-
D-Lombricine	<u>Glycine, serine and threonine metabolism</u>	C <sub>6</sub> H <sub>15</sub> N <sub>4</sub> O <sub>6</sub> P	2.081	1.09E-08	6.656	269.0669	[M-H]-
Ectoine		C <sub>6</sub> H <sub>10</sub> N <sub>2</sub> O <sub>2</sub>	1.508	1.89E-03	8.084	187.0724	[M+FA-H]-
Citraconic acid	<u>Valine, leucine and isoleucine biosynthesis</u>	C <sub>5</sub> H <sub>6</sub> O <sub>4</sub>	1.667	4.00E-06	1.401	129.0197	[M-H]-
Galactosylglycerol	<u>Glycerolipid metabolism</u>	C <sub>9</sub> H <sub>18</sub> O <sub>8</sub>	0.586	5.11E-06	1.338	275.0757	[M+Na-2H]-
Phosphatidic acid		C <sub>24</sub> H <sub>34</sub> N <sub>6</sub> O <sub>2</sub>	0.502	8.47E-08	1.269	419.257	[M-H <sub>2</sub> O-H]-
Gamma-L-glutamyl-L-tyrosine	<u>Glutathione metabolism</u>	C <sub>14</sub> H <sub>18</sub> N <sub>2</sub> O <sub>6</sub>	0.615	2.29E-04	9.16	309.1097	[M-H]-
Pantothenic acid	<u>Pantothenate and CoA biosynthesis</u>	C <sub>9</sub> H <sub>17</sub> NO <sub>5</sub>	0.663	7.48E-04	1.274	200.093	[M-H <sub>2</sub> O-H]-
Cysteine-S-sulfate	<u>Cysteine and methionine metabolism</u>	C <sub>3</sub> H <sub>7</sub> NO <sub>5</sub> S <sub>2</sub>	0.245	2.18E-20	8.079	199.9694	[M-H]-

**Supplementary Table S1 – Altered metabolites in CM of hSOD1(G93A) primary astrocytes.** Annotation and KEGG metabolic pathways of CM metabolites detected in the ESI – polarity whose expression ratio (hSOD1(G93A) vs hSOD1(WT) astrocytes) was < 0.66 and > 1.50 (p-value < 0.05, Wilcoxon rank-sum test) were reported. The measured *m/z*, the chromatographic retention time (RT), and proposed adduct were detailed.

Uniprot Accession	Gene Name	Protein Name	ratio G93A/WT	p-value
<b>P10605</b>	<b>Ctsb</b>	<b>Cathepsin B</b>	<b>1.307</b>	<b>0.006; 0.049</b>
<b>Q9WUU7</b>	<b>Ctsz</b>	<b>Cathepsin Z</b>	<b>1.484</b>	<b>0.008; 0.049</b>
O70370	Ctss	Cathepsin S	1.374	0.014; <b>0.049</b>
P99026	Psmb4	Proteasome subunit beta type-4	1.321	0.005; <b>0.049</b>
<b>Q9CPS5</b>	<b>Psmc8</b>	<b>26S proteasome non-ATPase regulatory subunit 8</b>	<b>1.356</b>	<b>0.036; 0.049</b>
A0A0R4J0I9	Lrp1	Low density lipoprotein receptor-related protein 1	1.987	0.043; <b>0.049</b>
O89053	Coro1a	Coronin-1A	1.329	0.001; <b>0.049</b>
<b>P13020</b>	<b>Gsn</b>	<b>Gelsolin</b>	<b>1.344</b>	<b>0.012; 0.049</b>
O70200	Aif1	Allograft inflammatory factor 1	1.356	0.036; <b>0.049</b>
Q5SW88	Rab1a	RAB1A, member RAS oncogene family	1.467	0.010; <b>0.049</b>
P20060	Hexb	Beta-hexaminidase subunit beta	1.429	0.005; <b>0.049</b>
A0A087WS56	Fn1	Fibronectin	1.612	0.008; <b>0.049</b>
Q9DAK9	Phpt1	14 kDa phosphohistidine phosphatase	1.388	0.020; <b>0.049</b>
Q64282	Ifit1	Interferon-induced protein with tetratricopeptide repeats 1	1.590	0.032; <b>0.049</b>
Q9D115	Znf706	Zinc finger protein 706	2.115	0.047; <b>0.049</b>
P19324	Serpinh1	Serpin H1	1.321	0.005; <b>0.049</b>
Q8C845	Efh2	EF-hand domain-containing protein D2	1.514	0.024; <b>0.049</b>
Q91VW3	Sh3bgrl3	SH3 domain-binding glutamic acid-rich-like protein 3	1.384	0.041; <b>0.049</b>

**Supplementary Table S2- Up-regulated proteins in hSOD1(G93A) primary spinal astrocytes.** Among the 640 proteins identified and quantified in hSOD1(WT) and hSOD1(G93A) astrocytes, the only proteins whose expression ratio (G93A vs WT cells) was >1.33 (p-value <0.05, two-tailed t-test) are reported. UNIPROT accessions, gene names, and protein names are reported in the first three columns. In the last column the p-values obtained with either a two-tailed Student's t-test (in black) and the Wilcoxon Rank sum test (un-paired, two-sided; confidence level 0.95) (in blue) are reported. Subsequent validation analysis by Western blot was performed only for proteins reported in bold.

Uniprot accession	Gene name	Protein Name	Ratio G93A/WT	p-value
<b>O09131</b>	<b>Gsto1</b>	<b>Glutathione S-transferase omega-1</b>	<b>0.420</b>	<b>0.033; 0.049</b>
Q9CPU0	Glo1	Lactoylglutathione lyase	0.648	0.046; <b>0.049</b>
<b>P97494</b>	<b>Gclc</b>	<b>Glutamate--cysteine ligase catalytic subunit</b>	<b>0.672</b>	<b>0.043; 0.049</b>
O88531	Ppt1	Palmitoyl-protein thioesterase 1	0.646	0.022; <b>0.049</b>
Q8VCQ8	Cald1	Caldesmon 1	0.743	0.001; <b>0.049</b>
Q3TT92	Dpysl3	Dihydropyrimidinase-related protein 3	0.751	0.031; <b>0.049</b>
P97315	Csrp1	Cysteine and glycine-rich protein 1	0.764	0.011; <b>0.049</b>
G3UVV4	Hk1	Hexokinase 1, isoform CRA_f	0.681	0.032; <b>0.049</b>
Q08091	Cnn1	Calponin-1	0.701	0.001; <b>0.049</b>
Q8BZF8	Pgm5	Posphoglucomutase-like protein 5	0.706	0.049; <b>0.049</b>
D3YYE1	Anp32a	Acidic leucine-rich nuclear phosphoprotein 32 family member A (Fragment)	0.710	0.010; <b>0.049</b>
Q9R0P9	Uchl1	Ubiquitin carboxyl-terminal hydrolase isozyme L1	0.710	0.040; <b>0.049</b>
G3X9T8	Cp	Ceruloplasmin	0.733	0.047; <b>0.049</b>
P37804	Tagln	Transgelin	0.739	0.021; <b>0.049</b>
A2AIM4	Tpm2	Tropomyosin beta chain	0.759	0.005; <b>0.049</b>
P50543	S100a11	Protein S100-A11	0.770	0.030; <b>0.049</b>
O55022	Pgrmc1	Membrane-associated progesterone receptor component 1	0.624	0.018; <b>0.049</b>
P51655	Gpc4	Glypican-4	0.667	0.004; <b>0.049</b>
E9PWK1	Ephx1	Epoxide hydrolase	0.749	0.045; <b>0.049</b>
H3BLJ9	Esd	S-formylglutathione hydrolase	0.621	0.046; <b>0.049</b>

**Supplementary Table S3 - Down-regulated proteins in hSOD1(G93A) primary astrocytes.** Among the 640 proteins identified and quantified in hSOD1(WT) and hSOD1(G93A) astrocytes, the only proteins whose expression ratio (G93A vs WT cells) was <0.77 (p-value <0.05, two-tailed t-test) are reported. Other details are as in Supplementary Table 2.