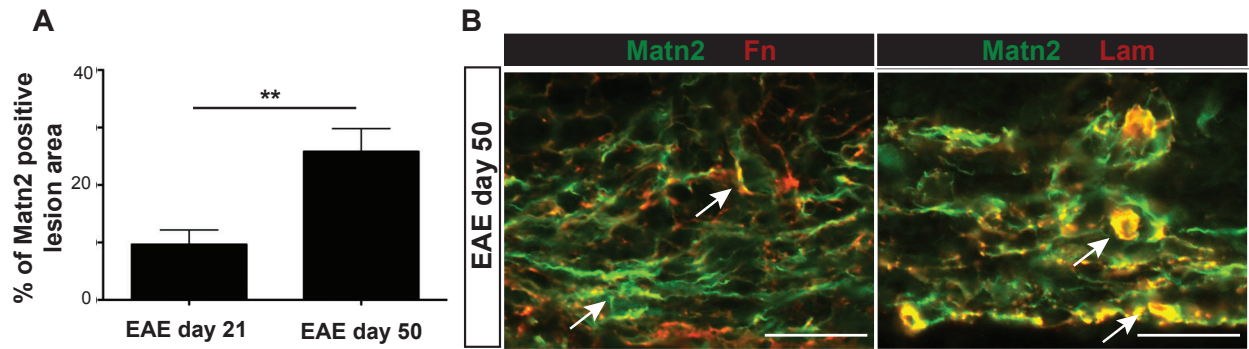
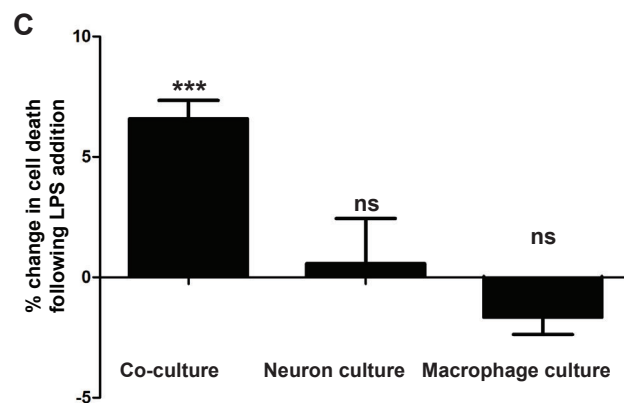
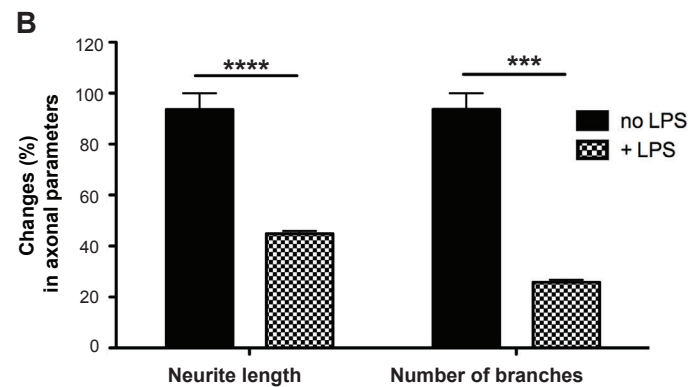
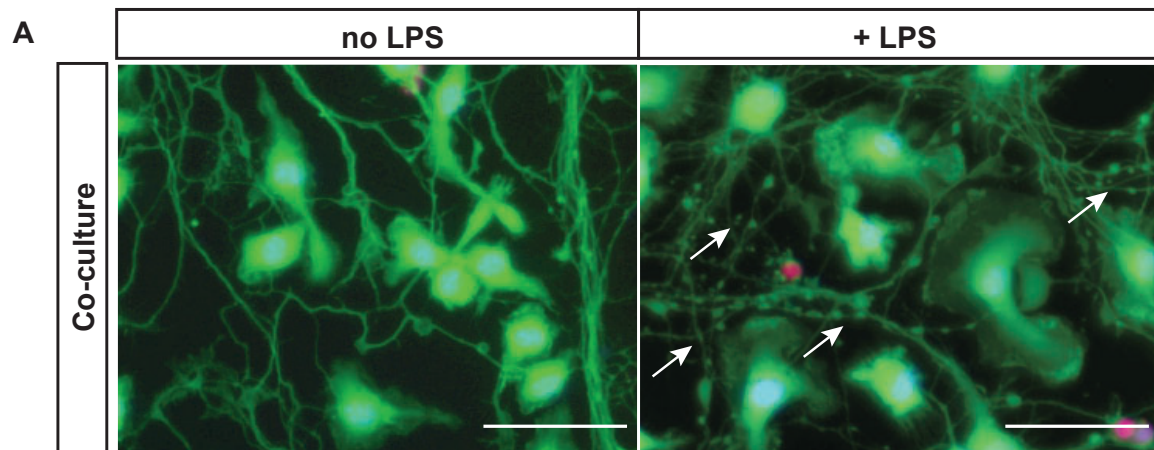


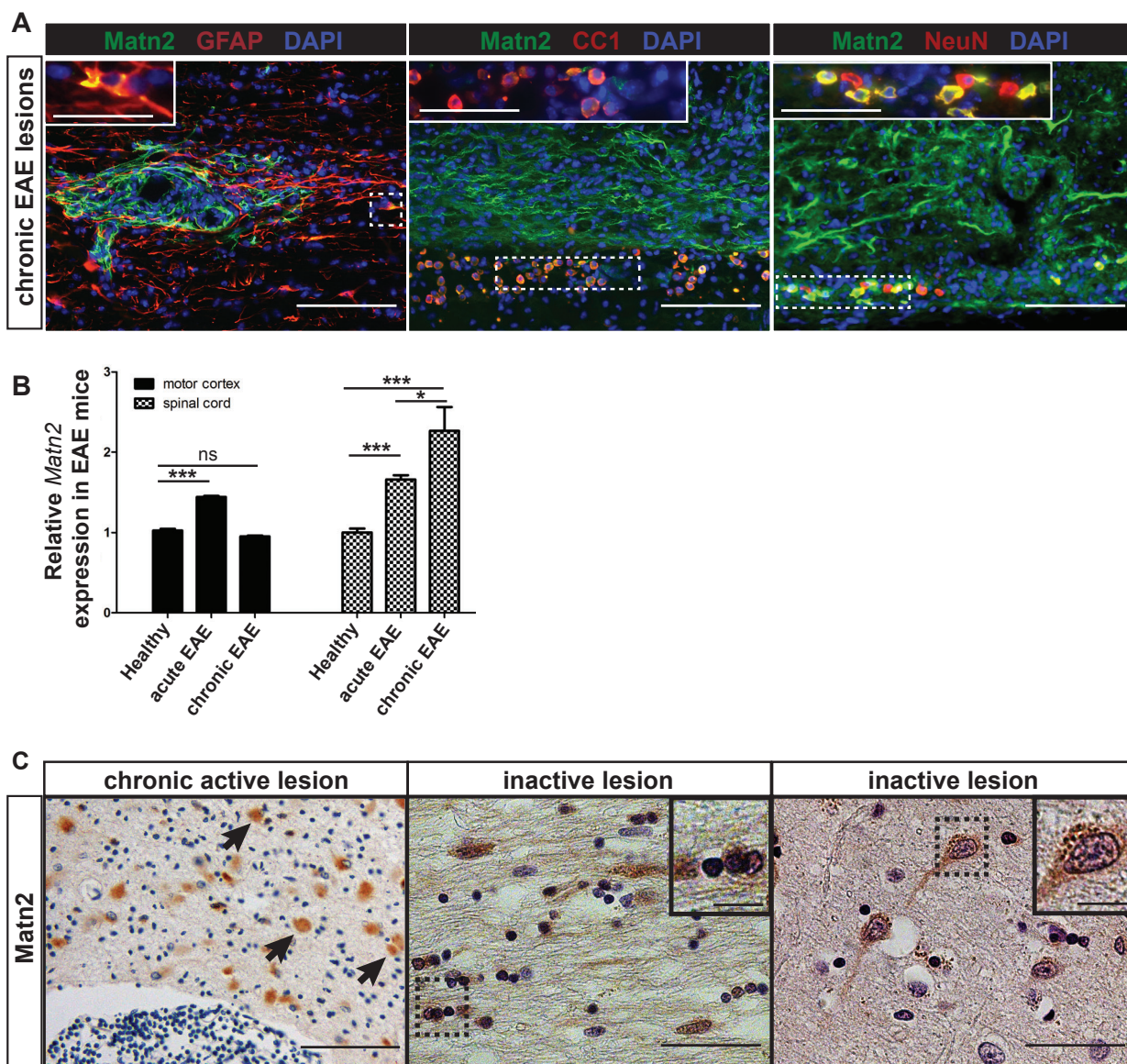
**Figure S1. *Matn2* expression in specific neurons of the brain.** (A) Co-labeling for *Matn2* and *Ctip2* (cortical layer 5 marker) or *Cux1* (cortical layer 2/3 marker) in the frontal cortex of healthy mice shows specific expression by neurons of CSMN layers. Scale bars, 50µm. (B) Co-labeling for *Matn2* and NF (pan Neurofilament marker) in the mouse hippocampus and *Matn2* IHC labeling in the hippocampus of human brain. Scale bars, 50µm. (C) *Matn2* expression (mean±SEM, qRT-PCR) in primary neuronal, astrocytic, oligodendroglial and microglial monocultures shows highest expression of *Matn2* in neurons (n=4-6/group). \*\*\*\*p<0.0001; One-way ANOVA relative to expression in microglia.



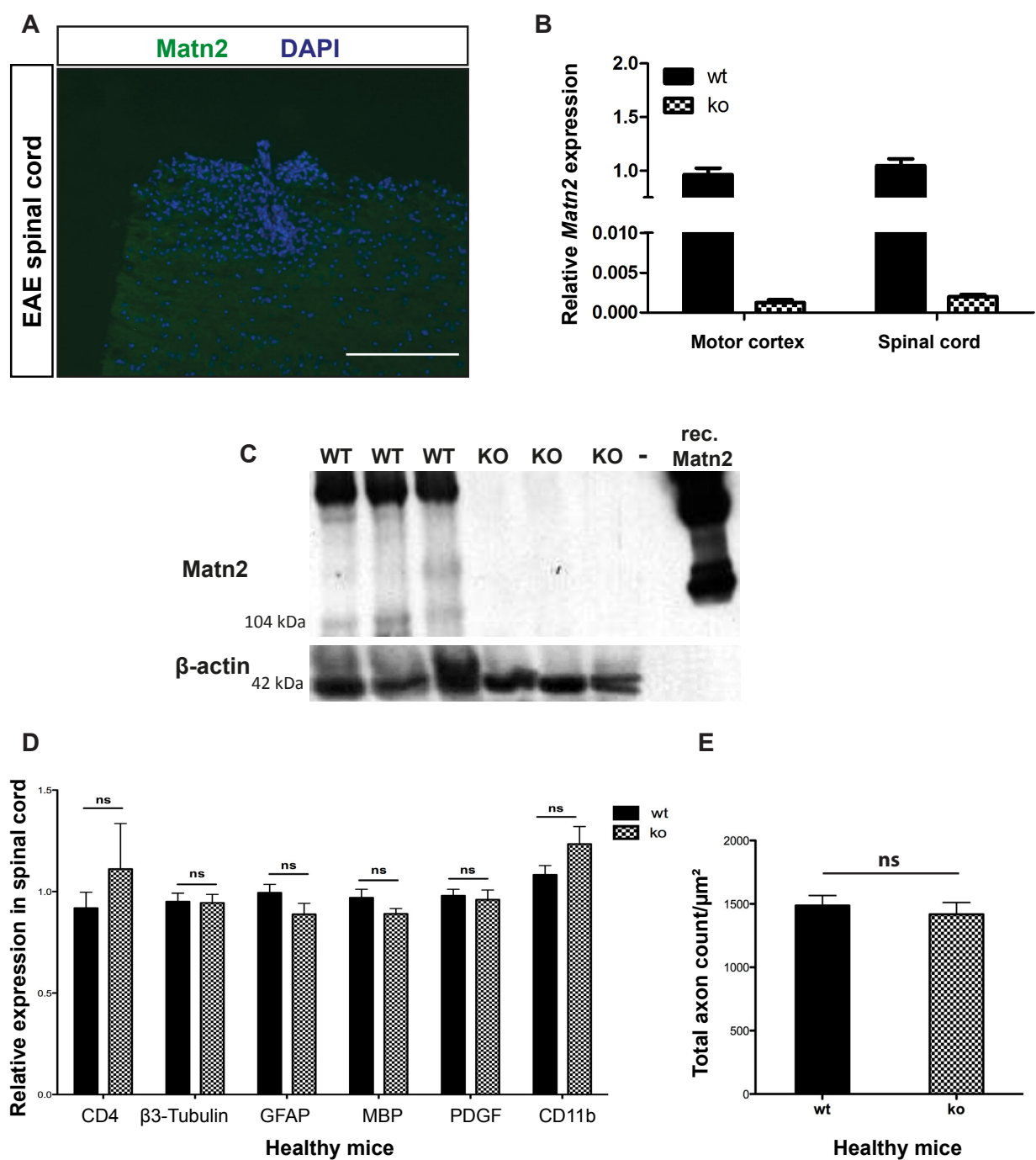
**Figure S2. Expression of Matn2 in post-acute EAE lesions.** (A) Quantitation of Matn2-positive lesional areas as a proportion of total lesion area (mean±SEM; %) at EAE day 21 and 50 (n=10/group, 2 independent cohorts). \*\* $p < 0.01$ ; Student's Ttest. (B) Co-labeling of Matn2 and fibronectin (Fn) or laminin (Lam) in lumbar spinal cord at EAE day 50 showing an association of Matn2 with the altered lesional ECM. Scale bars, 25 $\mu$ m.



**Figure S3. Macrophage-induced axonal injury increases *Matn2* gene expression in neurons *in vitro*.** Neuron/macrophage co-cultures and respective monocultures were stained with calceinAM (green), ethidium homo-dimer (Ethd1, red) and DAPI (blue). (A) Co-cultures develop axon injury (beading and transection, arrows) 24h following LPS addition compared to untreated cultures. Scale bars, 25 $\mu$ m. (B) Assessment of neurite length and number of branches (mean $\pm$ SEM; %) of 100 neurons in co-culture after 24h LPS addition (n=3 cultures/condition, 4 independent experiments). \*\*\*\*p<0.0001; \*\*\*p<0.001; Student's Ttest. (C) Change in cell death (mean $\pm$ SEM, %) detected by Ethd1 staining in co- and monocultures after 24h LPS addition compared to untreated controls (n=3 cultures/condition, 4 independent experiments).

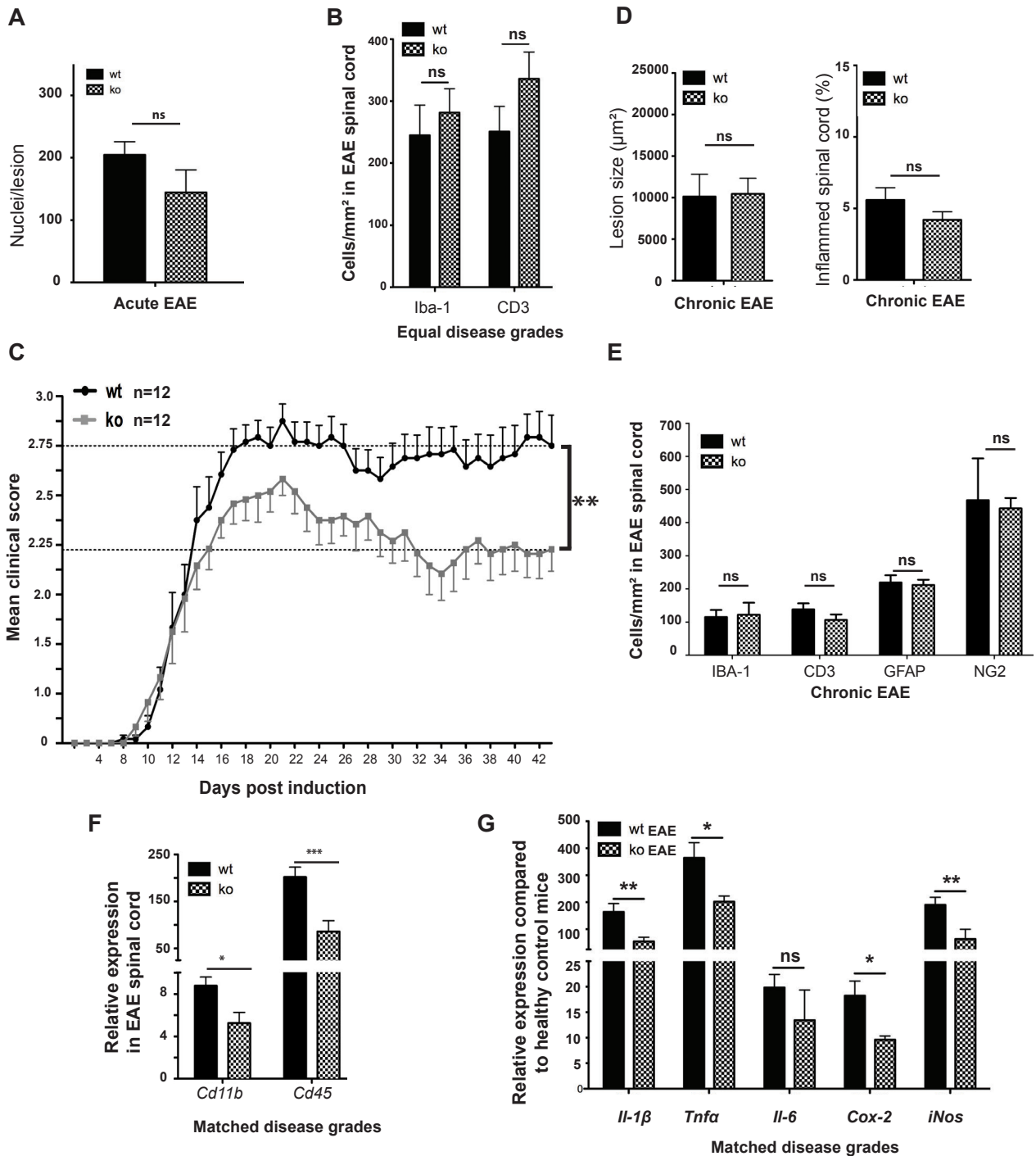


**Figure S4. Cell-specific Matn2 expression in chronic EAE and MS lesions.** (A) Co-labeling of Matn2 and GFAP, CC1 or NeuN in longitudinal lumbar cord sections at EAE day50 shows localization in astrocytes, oligodendrocytes and neurons, respectively. Nuclei were visualized with DAPI. Scale bars, 25µm and 10µm, respectively. (B) Matn2 expression (mean±SEM, qRT-PCR) in healthy and EAE mice (day21 and 50) in motor cortex and whole spinal cord (n=4-6/cohort). \*\*\*p<0.001, \*p<0.05; p≥0.5 = not significant (ns), One-way ANOVAs. (C) IHC labeling of a chronic active and inactive human MS lesions for Matn2 show widespread lesional astrocytic reactivity with Matn2 expression also found in local lesional oligodendrocytes and neurons. Scale bars, 50µm and 25µm.

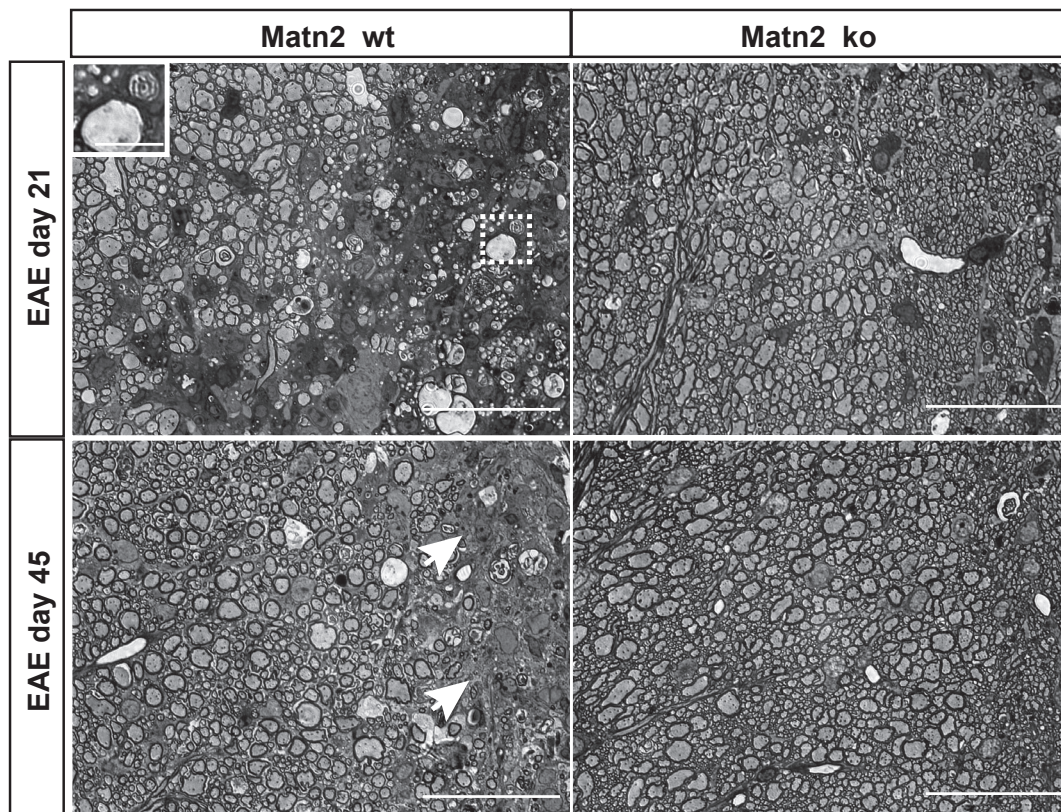


**Figure S5. Confirmation of Matn2 deletion and basic expression analysis in Matn2 ko mice.**

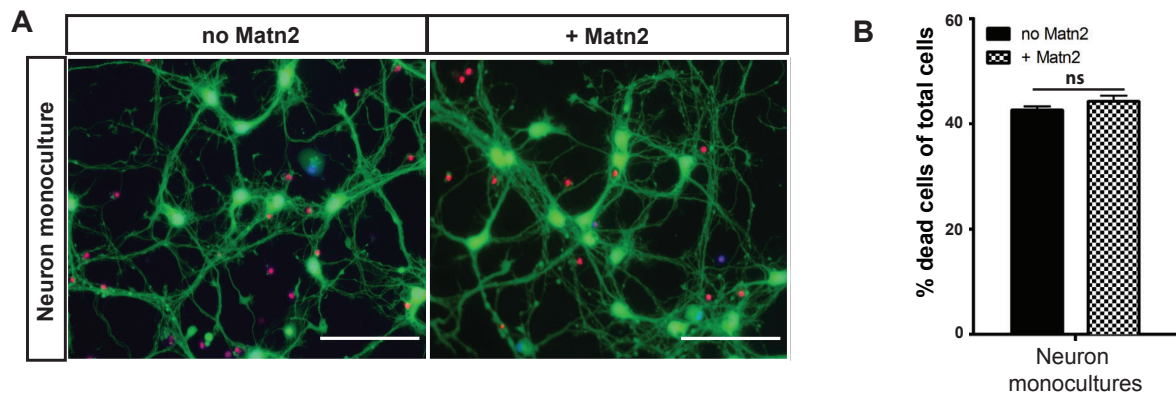
(A) Immunofluorescence analysis of Matn2 in longitudinal sections of lumbar spinal cord from EAE day 21 Matn2 ko mice shows absence of Matn2 staining in lesions. Nuclei were visualized with DAPI. Scale bar, 50 $\mu\text{m}$ . (B) Matn2 gene expression (mean $\pm$ SEM, qRT-PCR) in the motor cortex and whole spinal cord of Matn2 ko mice is reduced to undetectable levels compared to health mice (n=4/group). (C) Immunoblot analysis (native) for Matn2 in the frontal cortex of healthy wt and ko mice. Recombinant Matn2 was used as positive control, bands above 104kDa represent oligomeric Matn2. (D) Gene expression analysis (mean $\pm$ SEM, qRT-PCR) for CNS cell markers in spinal cord of healthy wt and Matn2 ko mice. Note that no difference in baseline expression levels was detected (n=5-7/group). p $\geq$ 0.5 = not significant (ns), Student's Ttest. (E) Axon counts (mean $\pm$ SEM) in the dorsal spinal cord column of healthy wt and Matn2 ko mice are similar (n=4-5/group). p $\geq$ 0.5 = not significant (ns); Student's Ttest.



**Figure S6. Matn2 deletion attenuates disease severity in acute EAE.** (A) Comparison of EAE scores (mean±SEM) of mice surviving to day 43 in wt and Matn2 ko mice shows that EAE scores stabilise beyond EAE day 21 (n=12/group). \*\*p<0.01; Mann-Whitney rank sum test. (B) Average cell density (mean±SEM) in lumbar spinal cord lesions of wt and Matn2 ko mice is similar at EAE day21 (n=10/genotype). p≥0.5 = not significant (ns); Student's Ttest. (C) Quantification of Iba-1-positive macrophages and CD3-positive T-cell density (mean±SEM) in the lumbar spinal cord of wt and Matn2 ko mice with equal disease severities in acute EAE (n=4/cohort). \*p<0.05; Student's Ttest. (D) Comparison of lesion size and cross-sectional cord area (%) with inflammatory lesions in the lumbar spinal cord (mean±SEM) in Matn2 ko mice and wt littermates in chronic EAE (n=10/genotype). \*\*p<0.01; Student's Ttest. (E) Quantification of Iba-1-positive macrophages CD3-positive T-cells, NG2-positive oligodendroglia and GFAP-positive astrocyte density (mean±SEM) in the lumbar spinal cord of wt and Matn2 ko mice in chronic EAE (n=10/cohort). \*p<0.05; Student's Ttest. (F-G) Expression of inflammation-associated genes (mean±SEM, qRT-PCR) in whole spinal cord from acute EAE wt and Matn2 ko mice. (n=4-6/group). \*\*\*\*p<0.0001, \*\*\*p<0.001, \*\*p<0.01, p≥0.5 = not significant (ns), Student's Ttest. Note that EAE mice with matched disease grades for each genotype were compared to healthy control mice (n=4-6/group). \*\*\*p<0.001, \*\*p<0.01, \*p<0.05, p≥0.5 = not significant (ns), Student's Ttest.

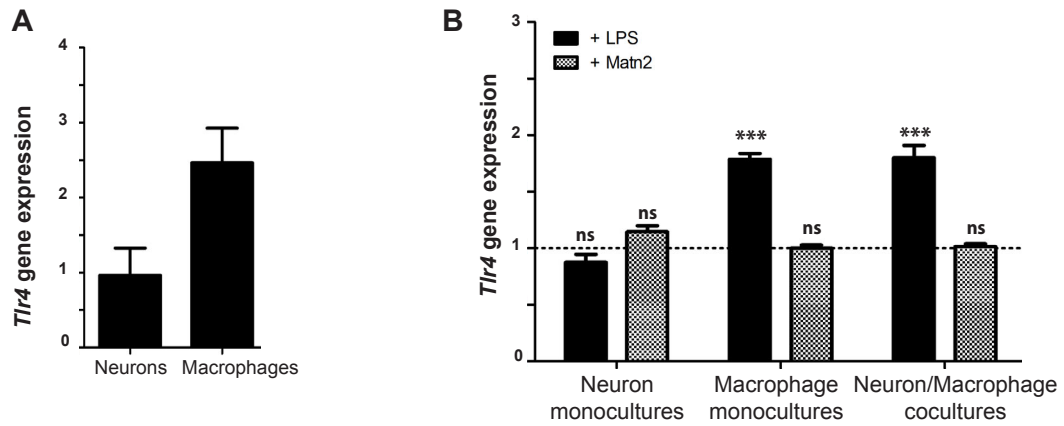


**Figure S7. Matn2 deletion reduces inflammatory gene expression and improves axonal integrity in EAE mice.** (A) Methylene-blue stained semi-thin cross-sections of the dorsal columns from EAE day 21 and 45 (converted to grey scale). Wt mice show marked inflammatory cell infiltration and abnormally shaped axons (inset) at EAE day 21 and noticeably more ECM tissue at day 45 (arrowheads) compared to their ko littermates. Scale bars, 25um.



**Figure S8. Exogenous Matn2 has no effect on pure neuronal cultures.** (A) Representative images of neuronal monocultures stained with CalceinAM (green), Ethidium homodimer (Ethd1, red) and DAPI (blue) show no morphological differences following 24h incubation with Matn2 (1 $\mu$ g/ml). Scale bars, 50 $\mu$ m. (B) Proportion of dead cells (mean $\pm$ SEM, %) detected by Ethd1 staining in neuronal monocultures at 24h post Matn2 addition (1 $\mu$ g/ml) compared to untreated control cultures (n=5 cultures/condition, 3 independent experiments).  $p \geq 0.5$  = not significant (ns), Student's Ttest.





**Figure S9. Tlr4 gene expression in primary cultures.** (A) Tlr4 gene expression (mean±SEM, qRT-PCR) difference in neuron and macrophage monocultures. (B) Tlr4 gene expression changes (mean±SEM, qRT-PCR) in neuron- and macrophage monocultures and cocultures following LPS (1µg/ml) or Matn2 (1µg/ml) addition after 24h incubation. (n=4 cultures/condition, 3 independent experiments). Results are shown relative to untreated controls (dotted line). \*\*\*p<0.001; p≥0.5 = not significant (ns); Student's Ttest.

## Genes differentially expressed in the motor cortex of EAE mice (fc $\geq$ 1.3, p<0.05).

Probe set	Gene name	Gene symbol	p-value	Fold change
1418595_at	plasma membrane associated protein, S3-12	S3-12	2,76E-08	3,90
1419394_s_at	S100 calcium binding protein A8 (calgranulin A)	S100a8	2,94E-04	3,22
1452975_at	alanine-glyoxylate aminotransferase 2-like 1	Agxt2l1	9,53E-08	3,11
1427345_a_at	sulfotransferase family 1A, phenol-preferring, member 1	Sult1a1	1,39E-06	2,93
1417290_at	leucine-rich alpha-2-glycoprotein 1	Lrg1	1,19E-07	2,64
1418536_at	histocompatibility 2, Q region locus 7	H2-Q7	3,66E-04	2,58
1416125_at	FK506 binding protein 5	Fkbp5	2,42E-06	2,34
1444681_at	ELKS/RAB6-interacting/CAST family member 2	Erc2	1,82E-05	2,25
1435999_at	serine peptidase inhibitor, Kazal type 8	Spink8	1,25E-04	2,21
1451006_at	xanthine dehydrogenase	Xdh	8,33E-06	2,18
1426260_a_at	similar to UDP glycosyltransferase 1 family, polypeptide A8	LOC632297 /// Ugt1a	1,86E-05	2,03
1455301_at	WAS/WASL interacting protein family, member 3	Wipf3	2,47E-05	1,80
1416612_at	cytochrome P450, family 1, subfamily b, polypeptide 1	Cyp1b1	4,28E-05	1,79
1448231_at	FK506 binding protein 5	Fkbp5	1,05E-05	1,76
1419442_at	matrilin 2	Matn2	2,12E-05	1,71
1451784_x_at	histocompatibility 2, D region locus 1	H2-D1	1,42E-04	1,71
1425545_x_at	histocompatibility 2, D region locus 1	H2-D1	1,88E-04	1,70
1429183_at	plakophilin 2	Pkp2	6,58E-05	1,70
1426261_s_at	similar to UDP glycosyltransferase 1 family, polypeptide A8	LOC632297 /// Ugt1a	2,71E-05	1,68
1444564_at	apolipoprotein D /// similar to apolipoprotein D	Apod /// LOC100047583	2,69E-04	1,63
1417495_x_at	ceruloplasmin	Cp	3,38E-04	1,61
1435386_at	Von Willebrand factor homolog	Vwf	5,85E-05	1,59
1455978_a_at	matrilin 2	Matn2	1,90E-05	1,57
1417494_a_at	ceruloplasmin	Cp	2,10E-04	1,55
1424783_a_at	similar to UDP glycosyltransferase 1 family, polypeptide A8	LOC632297 /// Ugt1a	5,72E-06	1,55
1448397_at	gap junction protein, beta 6	Gjb6	3,19E-05	1,50
1423619_at	RAS, dexamethasone-induced 1	Rasd1	2,29E-04	1,49
1425428_at	hypoxia inducible factor 3, alpha subunit	Hif3a	1,80E-05	1,48
1431043_at	kelch repeat and BTB (POZ) domain containing 5	Kbtbd5	1,44E-04	1,47
1457373_at	cadherin 19, type 2	---	1,41E-04	1,46
1449316_at	cytochrome P450, family 4, subfamily f, polypeptide 15	Cyp4f15	2,86E-05	1,44
1421384_at	lysosomal trafficking regulator	Lyst	1,31E-04	1,43
1451601_a_at	spinster homolog 2 (Drosophila)	Spns2	2,30E-04	1,43
1418306_at	crystallin, beta B1	Crybb1	2,13E-05	1,43
1435941_at	rhomboid, veinlet-like 3 (Drosophila)	Rhbdl3	1,85E-04	1,42
1442025_a_at	zinc finger and BTB domain containing 16	---	6,99E-05	1,42
1424714_at	aldolase C, fructose-bisphosphate	Aldoc	8,03E-06	1,41
1417629_at	proline dehydrogenase	Prodh	1,57E-04	1,40
1453187_at	OCIA domain containing 2	Ociad2	1,92E-07	1,39
1434674_at	lysosomal trafficking regulator	Lyst	5,24E-05	1,38
1428223_at	major facilitator superfamily domain containing 2	Mfsd2	9,83E-05	1,38
1448392_at	secreted acidic cysteine rich glycoprotein	Sparc	4,77E-05	1,38
1435917_at	OCIA domain containing 2	Ociad2	8,04E-05	1,37
1448188_at	uncoupling protein 2 (mitochondrial, proton carrier)	Ucp2	1,29E-05	1,35
1426915_at	death associated protein kinase 1	Dapk1	1,87E-04	1,34
1446769_at	tetratricopeptide repeat domain 39C	Ttc39c	2,05E-04	1,34
1422465_a_at	nucleoredoxin	Nxn	4,37E-06	1,34
1424715_at	retinol saturase (all trans retinol 13,14 reductase)	Retsat	1,75E-04	1,34
1416266_at	prodynorphin	Pdyn	7,52E-06	1,33
1420764_at	scrapie responsive gene 1	Scrg1	4,26E-05	1,33
1417785_at	phospholipase A1 member A	Pla1a	1,27E-04	1,32
1434203_at	cDNA sequence BC055107	BC055107	8,01E-05	1,32
1436978_at	wingless-type MMTV integration site 9A	Wnt9a	2,31E-04	-1,31
1422977_at	glycoprotein lb, beta polypeptide	Gp1bb	4,03E-05	-1,31
1420877_at	septin 6	Sep06	3,43E-06	-1,31
1454995_at	dimethylarginine dimethylaminohydrolase 1	---	2,40E-04	-1,32
1448812_at	hippocalcin-like 1	Hpcal1	1,67E-04	-1,32
1434414_at	FAD-dependent oxidoreductase domain containing 2	Foxred2	2,65E-04	-1,33
1424475_at	calcium/calmodulin-dependent protein kinase kinase 2, beta	Camkk2	2,62E-05	-1,33
1416474_at	neighbor of Punc E11	Nope	3,96E-04	-1,33
1424659_at	slit homolog 2 (Drosophila)	Slit2	2,58E-04	-1,33
1455008_at	guanine nucleotide binding protein, alpha 12	---	1,20E-04	-1,33
1451555_at	neurolysin (metallopeptidase M3 family)	Nln	1,34E-04	-1,34
1422889_at	protocadherin 18	Pcdh18	4,46E-05	-1,34
1453746_at	formin binding protein 1	Fnbp1	3,68E-04	-1,35
1448502_at	solute carrier family 16 (monocarboxylic acid transporters), member 7	Slc16a7	9,37E-05	-1,35
1418788_at	endothelial-specific receptor tyrosine kinase	Tek	1,25E-04	-1,35
1426460_a_at	UDP-glucose pyrophosphorylase 2	Ugp2	4,52E-05	-1,37
1418139_at	doublecortin	Dcx	3,23E-04	-1,37
1435947_at	TSC22 domain family, member 1	---	9,82E-05	-1,37
1433489_s_at	fibroblast growth factor receptor 2	Fgfr2	1,43E-05	-1,39
1418706_at	solute carrier family 38, member 3	Slc38a3	1,54E-06	-1,40
1448609_at	thiosulfate sulfurtransferase, mitochondrial	Tst	8,89E-05	-1,40
1415802_at	solute carrier family 16 (monocarboxylic acid transporters), member 1	Slc16a1	2,83E-04	-1,40
1417963_at	phospholipid transfer protein	Pltp	3,33E-04	-1,40
1433787_at	NEL-like 1 (chicken)	Nell1	2,57E-04	-1,40
1436568_at	junction adhesion molecule 2	Jam2	2,43E-05	-1,40

1436672_at	G protein-coupled receptor kinase 5	Grk5	1,26E-04	-1,42
1418937_at	deiodinase, iodothyronine, type II	Dio2	7,81E-05	-1,42
1428922_at	RIKEN cDNA 1200009O22 gene	1200009O22Rik	6,26E-05	-1,42
1424701_at	protocadherin 20	Pcdh20	5,56E-06	-1,42
1435171_at	hypothetical protein LOC100046169; RIKEN cDNA 2810416G20 gene	2810416G20Rik	3,71E-04	-1,43
1416673_at	beta-site APP-cleaving enzyme 2	Bace2	1,73E-05	-1,43
1451122_at	isopentenyl-diphosphate delta isomerase	Idi1	7,08E-08	-1,43
1420905_at	interleukin 17 receptor A	Il17ra	1,31E-04	-1,43
1418979_at	aldo-keto reductase family 1, member C14	Akr1c14	6,90E-05	-1,43
1422706_at	prostate transmembrane protein, androgen induced 1	Pmepa1	1,56E-04	-1,44
1440981_at	p21 (CDKN1A)-activated kinase 7	Pak7	1,45E-05	-1,45
1436642_x_at	expressed sequence AW047730	AW047730	1,42E-05	-1,45
1459679_s_at	myosin IB	Myo1b	2,26E-04	-1,45
1418976_s_at	cell death-inducing DNA fragmentation factor, alpha subunit-like effector B	Cideb	3,71E-04	-1,45
1458954_at	predicted gene 13308; similar to chemokine (C-C motif) ligand 27	621580	2,48E-04	-1,46
1460219_at	myelin-associated glycoprotein	Mag	2,48E-04	-1,46
1420895_at	transforming growth factor, beta receptor I	Tgfb1	7,40E-06	-1,47
1416444_at	elongation of very long chain fatty acids (FEN1/Elo2, SUR4/Elo3, yeast)-like 2	Elov2	3,05E-04	-1,47
1428792_at	breast carcinoma amplified sequence 1	Bcas1	8,82E-05	-1,47
1448752_at	carbonic anhydrase 2	Car2	2,67E-05	-1,47
1449514_at	G protein-coupled receptor kinase 5	Grk5	4,31E-05	-1,47
1416832_at	solute carrier family 39 (metal ion transporter), member 8	Slc39a8	1,01E-05	-1,48
1435581_at	beta-site APP-cleaving enzyme 2	Bace2	1,08E-05	-1,48
1456722_at	chordin-like 1	Chrd1	5,19E-05	-1,49
1443888_at	expressed sequence AU023762	AU023762	3,33E-07	-1,50
1451047_at	integral membrane protein 2A	Itm2a	3,22E-05	-1,50
1423343_at	solute carrier organic anion transporter family, member 1c1	Slco1c1	5,80E-08	-1,50
1420904_at	interleukin 17 receptor A	Il17ra	9,20E-05	-1,51
1423608_at	integral membrane protein 2A	Itm2a	1,38E-04	-1,51
1436195_at	cDNA sequence BC046404	BC046404	7,78E-06	-1,52
1450728_at	four jointed box 1 (Drosophila)	Fjx1	2,18E-05	-1,52
1438169_a_at	FERM domain containing 4B	Frm4b	2,04E-04	-1,52
1428303_at	BTB (POZ) domain containing 17	1500005I02Rik	3,92E-04	-1,53
1435418_at	solute carrier family 22 (organic anion transporter), member 8	Slc22a8	2,36E-04	-1,53
1426314_at	endothelin receptor type B	Ednrb	6,44E-06	-1,53
1426294_at	hyaluronan and proteoglycan link protein 1	Hapln1	2,31E-04	-1,53
1426973_at	G protein-coupled receptor 153	Gpr153	1,13E-04	-1,54
1439015_at	glial cell line derived neurotrophic factor family receptor alpha 1	Gfra1	2,37E-04	-1,55
1417430_at	cerebellar degeneration-related 2	Cdr2	3,99E-05	-1,56
1434984_at	polo-like kinase 5 (Drosophila)	6330514A18Rik	4,62E-05	-1,57
1428700_at	purinergic receptor P2Y, G-protein coupled 13	P2ry13	2,07E-04	-1,59
1456603_at	family with sequence similarity 101, member B	1500005K14Rik	2,88E-05	-1,59
1448945_at	plasma membrane proteolipid	Plip	1,05E-04	-1,59
1437347_at	endothelin receptor type B	Ednrb	4,97E-07	-1,60
1416966_at	solute carrier family 22 (organic anion transporter), member 8	Slc22a8	2,88E-04	-1,60
1417319_at	poliovirus receptor-related 3	Pvr3	3,59E-05	-1,60
1417143_at	lysophosphatidic acid receptor 1	Lpar1	3,72E-04	-1,61
1438030_at	RAS, guanyl releasing protein 3	Rasgrp3	2,94E-04	-1,61
1435642_at	proline rich region 18	Prr18	3,80E-05	-1,63
1418310_a_at	retinaldehyde binding protein 1	Rlbp1	3,96E-05	-1,63
1425582_a_at	endomucin	Emcn	4,74E-05	-1,64
1438540_at	collagen, type XXV, alpha 1	Col25a1	1,89E-05	-1,66
1438648_x_at	RIKEN cDNA 1190003M12 gene	1190003M12Rik	4,26E-05	-1,66
1437918_at	RIKEN cDNA 4930539E08 gene	4930539E08Rik	6,96E-05	-1,67
1436593_at	RIKEN cDNA 1700016K19 gene	1700016K19Rik	1,47E-04	-1,70
1444633_at	adenylate cyclase 2	---	9,74E-07	-1,72
1439933_at	RIKEN cDNA B430316J06 gene	B430316J06Rik	9,99E-05	-1,72
1451038_at	apelin	Apln	5,37E-05	-1,75
1444089_at	spectrin beta 2	Spnb2	3,22E-04	-1,76
1438751_at	solute carrier family 30, member 10	Slc30a10	4,02E-04	-1,78
1420352_at	protease, serine, 22	Prss22	2,39E-04	-1,79
1419063_at	UDP galactosyltransferase 8A	Ugt8a	4,13E-05	-1,79
1419905_s_at	hydroxyprostaglandin dehydrogenase 15 (NAD)	Hpgd	3,81E-04	-1,83
1434201_at	chordin-like 1	Chrd1	2,21E-05	-1,85
1449519_at	growth arrest and DNA-damage-inducible 45 alpha	Gadd45a	3,85E-04	-1,85
1429286_at	RIKEN cDNA 1190003M12 gene	1190003M12Rik	1,82E-04	-1,93
1431724_a_at	purinergic receptor P2Y, G-protein coupled 12	P2ry12	1,88E-04	-1,99
1450779_at	fatty acid binding protein 7, brain	Fabp7	2,57E-06	-2,34
1416318_at	serine (or cysteine) peptidase inhibitor, clade B, member 1a	Serpinb1a	9,20E-05	-2,58
1422542_at	G protein-coupled receptor 34	Gpr34	9,59E-05	-2,61
1427053_at	ABI gene family, member 3 (NESH) binding protein	Abi3bp	7,86E-06	-2,92
1427054_s_at	ABI gene family, member 3 (NESH) binding protein	Abi3bp	7,38E-05	-3,24

**Table S1 continued. Genes differentially expressed in the motor cortex (mc) of EAE mice (fc $\geq$ 1.3, p $<$ 0.05).** List of differentially expressed genes in the motor cortex of healthy and EAE mice determined by microarray analysis (Affymetrix Moe430 2.0). The table shows the probe set number, gene name, gene symbol, p-value and fold change between healthy and EAE mice. Genes with positive values are up-regulated and genes with negative values are down-regulated in the motor cortex of EAE mice.