

An exploratory study of the association between thyroid hormone and survival of amyotrophic lateral sclerosis

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Abstract The objective of the study was to determine the correlation of thyroid hormones (THs) and survival in patients with amyotrophic lateral sclerosis (ALS). A total of 278 patients with ALS were enrolled and followed up prospectively every 3 months by face-to-face interviews or phone calls. Level of fasting serum TH was determined at the time of enrollment. Clinical data including age, onset age, onset body region, ALS Functional Rating Scale-Revised (ALS-FRS-R), and survival were collected. Patients were separated into four subgroups according to the quartile distribution of TH, and survival was compared among them. There was no significant difference in baseline characteristics, including onset age, gender, onset form as well as treatment with riluzole among different levels of TH subgroups. Chi-square test indicated that the lowest quartile of FT3 (<4.26 mg/L) might correlate with higher incidences of death (OR 0.374, 95 % CI 0.173–0.813), while second and third quartiles of FT4 (range 14.25–18.0 mg/L) with lower incidences of death in ALS. However, after correction for onset age and onset form using logistic regression analysis, no statistical difference was revealed in survival among patients with different TH levels. TH does not correlate with survival and is not a prognostic factor for ALS.

Keywords Amyotrophic lateral sclerosis · Motor neuron disease · Thyroid hormone · Survival · Prognosis · Quartile-specific analysis

Introduction

Amyotrophic lateral sclerosis (ALS) is a chronic, typically adult-onset neurodegenerative disease, involving both the upper and lower motor neurons in the motor areas of cerebral cortex, brainstem and spinal cord, leading to death within 3–5 years through failure of respiration mostly [1].

Despite the traditional view considering that ALS was a pure motor neuron disorder affecting motor functions simply, increasing evidence has been suggesting that ALS is a multi-system disease with additional extra-motor neurologic and extra-neurological manifestations [2–5]. Beyond the nervous system, intriguing metabolic abnormalities have been reported in ALS patients, such as elevated systematic metabolism [6, 7], dyslipidemia [8], uric acid abnormality [9], disturbed glucose tolerance [10] and insulin resistance [11].

It has been well known that thyroid hormone (TH) plays a major role in normal development, metabolism and growth of many organ systems including central nervous system [12]. Several basic researches reported that thyroid hormone was involved in the up-regulation of growth of neuronal cells including motor neurons [13–15]. There was evidence that hypermetabolism could exacerbate the rate of motor neuron degeneration by increasing the production of reactive oxygen species in mitochondria [16], whose dysfunction closely correlated with the pathogenesis of ALS [17]. Therefore, reducing metabolism systemically may in theory prolong the survival in ALS. Nevertheless, the life span of SOD1G93A ALS mice did not get prolonged using non-pharmacologic calorie restriction [18], nor by a pharmacologic approach such as methimazole which can lead to hypothyroidism by inhibiting TH synthesis [19].

Therefore, there has been increasing interest in investigating the role of TH in the progression of ALS. We

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Table 1 Baseline characteristics of 341 patients with ALS in stratified thyroid hormones

	Baseline thyroid hormone concentration quartiles							
	TSH		FT3		FT4			
Thyroid hormones	1 (<1.51)	2 (1.51–2.09)	3 (2.10–3.14)	4 (>3.14)	1 (<14.25)	2 (14.25–15.89)	3 (15.90–18.00)	4 (>18.00)
Number	69	71	69	69	69	67	70	69
Onset age	53.0 (11.3)	52.4 (12.4)	52.0 (12.0)	54.7 (13.8)	55.8 (12.5)	51.4 (14.4)	52.1 (14.3)	53.8 (11.0)
Age	54.0 (11.3)	53.2 (12.4)	53.0 (12.1)	54.6 (12.1)	56.7 (12.7)	52.0 (12.0)	52.1 (12.4)	54.9 (11.0)
Male no. (%)	43 (62.3)	42 (59.2)	39 (56.5)	35 (50.7)	29 (42.0)	40 (60.6)	36 (51.4)	44 (62.9)
Area of onset								
Spinal no. (%)	54	54	51	55	45	61	55	54
Bulbar no. (%)	15	17	18	14	24	5	15	21
Patients on riluzole no. (%)	16 (23.2)	16 (22.5)	18 (26.1)	15 (21.7)	14 (20.3)	13 (19.7)	15 (21.4)	19 (27.5)

FT3 free triiodothyronine, FT4 free thyroxine, TSH thyroid stimulating hormone, ALS amyotrophic lateral sclerosis

determined the TH status in a large sample of patients with ALS, and analyzed the correlation of different levels of TH with the survival of ALS patients.

Methods

Patients

Patients with clinical diagnosis of definite or probable ALS (El Escorial revised criteria) from a large, regional referral ALS clinic of Southwest China, from May 2006 to March 2013 were enrolled in the study. Level of fasting serum TH including free triiodothyronine (FT3), free thyroxine (FT4) and thyroid stimulating hormone (TSH) was determined at enrollment. The demographic features and clinical data such as age, onset age, onset body region, disease duration, and survival time were collected. Forced vital capacity (FVC) and ALS Functional Rating Scale-Revised (ALS-FRS-R) assessed the severity of ALS patients. Patients were followed up every 3 months by face-to-face interviews or by phone calls. Survival was determined when patients were alive without tracheotomy. Patients with pre-ALS thyroid diseases diagnosed by endocrine specialists were excluded. The study was approved by the local ethics committee of West China Hospital, Sichuan University. All the patients were informed about the purpose of the study.

Statistical analysis

Analysis was performed using SPSS version 17.0 software. All continuous data including mean age, onset age, disease duration, and ALS-FRS-R were presented in the form of mean \pm standard deviation, and compared between subgroups with Kruskal–Wallis *H* test. The levels of TH were separated into quartiles based on their distribution of the ALS patients, and the highest quartile of each variable was applied as a reference for analysis of investigating its possible role in ALS survival (Table 1). Odds ratios (OR) and 95 % confidence intervals (CI) were estimated by Chi-square test, and by Logistic Regression analysis. Kaplan–Meier curves using log-rank test were performed in survival analysis of ALS patients.

Results

A total of 278 patients with probable or definite diagnosed ALS were enrolled, with the baseline demographic data collected at the date of the study enrollment displayed in Table 1. Among them, there were 119 females and 159 males, i.e., male-to-female ratio was 1.34:1. The mean age at ALS onset was 53.0 ± 12.4 years (range

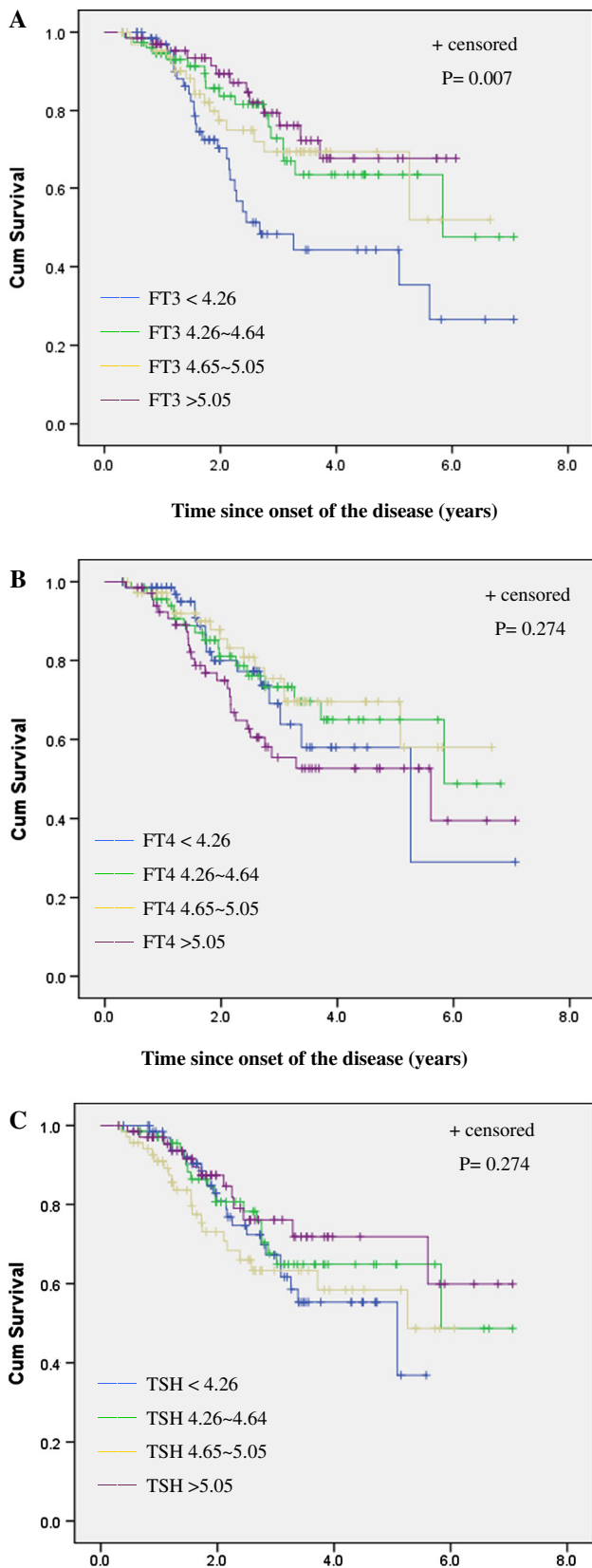


Fig. 1 Correlation between TH levels and survival in ALS patients. Kaplan–Meier survival curves of 278 patients with ALS stratified according to their thyroid hormones’ levels. **a** FT3 level. *Blue line* <math><4.26\text{ mg/dL}</math>, *green line* orange line purple line b FT4 level. *Blue line* <math><14.25\text{ mg/dL}</math>, *green line* orange line purple line c TSH level. *Blue line* <math><1.51\text{ mg/dL}</math>, *green line* orange line purple line

Table 2 TSH, FT3 and FT4 compared between different gender, onset form and age group

	TSH	P_{TSH}	FT3	P_{FT3}	FT4	P_{FT4}
Gender						
Male	2.37 (1.51)	0.086	4.75 (0.69)	0.002	16.90 (5.43)	0.118
Female	2.88 (3.25)		4.50 (0.68)		16.62 (2.45)	
Onset form						
Limb	2.47 (1.61)	0.311	4.71 (0.69)	0.108	16.64 (4.94)	0.166
Bulbar	2.82 (4.08)		4.45 (0.69)		15.75 (2.36)	
Onset age (years)						
Early (<math><45</math>)	2.47 (1.44)	0.589	4.64 (0.66)	0.922	15.95 (2.25)	0.335
Late (>math>>45</math>)	2.65 (2.72)		4.65 (0.72)		16.53 (5.07)	

FT3 free triiodothyronine, FT4 free thyroxine, TSH thyroid stimulating hormone, ALS amyotrophic lateral sclerosis

22.6–78.5 years), and 64 patients (23.0 %) had a bulbar form at onset. The mean TSH value was $2.59 \pm 2.42\text{ mg/dL}$ (range 0.21–33.48 mg/dL), and FT3 $4.64 \pm 0.70\text{ mg/dL}$ (2.58–7.89 mg/dL), FT4 $16.35 \pm 4.45\text{ mg/dL}$ (ranged 9.30–76.7 mg/dL). A total of 20 (7.2 %) patients had abnormal TH levels, of whom 14 (5.1 %) ALS had lower FT3, 10 (3.6 %) lower FT4, and 6 (2.2 %) ALS patients increased FT4. None of the ALS patients had abnormal TSH and increased FT3. None of these patients needed to receive medication treatment according to endocrine specialists. Three patients with higher serum TSH level had hypertension managed with antihypertensive agents. One patient with normal TH levels had type 2 diabetes mellitus, and was diet controlled. None of these patients reported any significant hypertensive or diabetic complications. Two patients with lower FT3 levels and six patients with normal TH levels had a history of taking creatine or enzyme Q10.

Table 3 Association analysis of thyroid hormone levels with survival of amyotrophic lateral sclerosis

	Number of ALS	Number departed	OR (95 % CI)	OR after correction for onset age and onset symptom (95 % CI)
TSH				
1	69	22	0.496 (0.226, 1.090)	2.083 (0.928, 4.674)
2	71	18	0.684 (0.305, 1.531)	1.500 (0.656, 3.430)
3	69	21	0.531 (0.240, 1.171)	2.096 (0.922, 4.767)
4	69	13	1 (reference value)	1 (reference value)
FT3				
1	69	27	0.374 (0.173, 0.813)	2.376 (0.973, 5.261)
2	76	18	0.776 (0.347, 1.733)	1.216 (0.537, 2.756)
3	66	16	0.752 (0.329, 1.720)	1.366 (0.589, 3.168)
4	67	13	1 (reference value)	1 (reference value)
FT4				
1	69	16	2.003 (0.921, 4.204)	0.503 (0.233, 1.054)
2	70	17	1.885 (1.012, 3.919)	0.547 (0.258, 2.281)
3	70	15	2.217 (1.047, 4.695)	0.427 (0.599, 1.013)
4	69	26	1 (reference value)	1 (reference value)

FT3 free triiodothyronine, FT4 free thyroxine, TSH thyroid stimulating hormone, ALS amyotrophic lateral sclerosis

FT3 was significantly higher in males than in females (4.75 ± 0.69 vs. 4.50 ± 0.68 , $P = 0.002$), but not between limb- and bulbar-onset patients ($P = 0.108$), nor between early and late onset groups ($P = 0.922$). No statistical differences were revealed while analyzing TSH and FT4 levels in different gender, onset form as well as early or late onset groups ($P > 0.05$) (Table 2). The patients were separated into four subgroups according to the quartile distribution of TH. There were no significant differences in baseline characteristics including onset age, gender, onset form, treatment with riluzole as well as survival among the four subgroups (Table 1).

At the end of our study, 74 ALS patients had died, with 70 (94 %) of respiratory dysfunction. With the highest quartile of each variable being applied as a reference for analysis, we investigated the possible role in ALS survival by Chi-square test, which showed that lowest quartile of FT3 (<4.26 mg/L) correlated with higher incidences of death in ALS, second and third quartile of FT4 (range

14.25–18.0 mg/L) correlated with less incidences of death, while no correlation was found between different levels of TSH and survival in ALS. Kaplan–Meier curve with log-rank test indicated that the lowest quartile of FT3 was significantly associated with shorter survival time of ALS than the highest quartile of FT3 ($P = 0.007$; Fig. 1a). There were no differences in survival time between the first, second, third, and highest quartile of FT4 and TSH ($P > 0.05$; Fig. 1b, c). However, considering the relation between FT3 and FT4, and the close correlations of onset age, onset form with survival in ALS [20], we, therefore, performed the logistic regression analysis with adjusted for onset age, onset form to further analyze the effects on the survival of different levels of TH. As illustrated in Table 3, after correction for onset age and onset form, there was no correlation between different levels of TH and survival of ALS.

Discussion

To the best of our knowledge, this is the first study to analyze the correlation of TH and survival in a relatively large cohort of ALS patients. The demographic data of included patients were in accordance with previous reports, showing that the patients included in the present study are good representatives of the ALS population.

In the current study, ALS patients who had abnormal level of TH were not common (7.2 %). Although the association of FT3 level with survival of ALS in mono-variate analysis was found in the current study, the significance of which was not maintained in multivariate analysis after adjusting for gender, onset age and onset form. Moreover, it has been indicated that thyroid hormone could be influenced by age, gender and even tested season in previous studies [21, 22]. In a word, the data we presented here indicated that TH does not have impact on survival of patients with ALS. Our findings were in accordance with these human studies and animal studies. Many researchers found that TH status was not relevant to the pathogenesis of ALS [23–25], although Malin et al.'s study found elevation of TH (r-T3) in the cerebrospinal fluid in 12 definite ALS patients [26]. Furthermore, no prolonged lifespan was observed in hypometabolism in SOD1-G93A mice treated with methimazole and calorie restriction [19].

TH has been demonstrated to play direct roles in the proliferations and differentiations of neuronal cell and glial cells [14], but also indirect actions through the presence of TH receptors, which have been demonstrated in cultured astrocytes [27]. On the other hand, there was evidence of TH action on the neuronal–glial interactions. In vitro, TH has been demonstrated to regulate the

expression of extracellular matrix and adhesion molecules that are important for neuronal migration and development, such as tenascin- C [28], neural cell adhesion molecule [29], laminin and fibronectin [30–32]. And in vivo, TH was proved to be important in the *Xenopus* spinal cord neurogenesis by comparing between controls and those with injections of different dosage of T3, which indicated that TH administrations could result in increased numbers of neurons and up-regulate the neurogenesis of *Xenopus* spinal cord.

Taken together, despite the restrictions of study designs and sample sizes, the inconsistent views drawn by in vitro, in vivo and human studies indicate that the regulation of neuronal cells in human subjects may involve more mechanism, but not simply through TH regulation.

Our study and together with those basic studies give the evidence that TH does not correlate with survival time in ALS and is not a prognostic factor for ALS patients.

Conflict of interest The authors report no conflict of interest.

References

- Brooks BR, Sanjak M, Belden D, Juhasz-Poscine K, Waclawik A (eds) (2000) Natural history of amyotrophic lateral sclerosis. Dunitz, London
- Dupuis L, Corcia P, Fergani A, Gonzalez De Aguilar J-L, Bonnefont-Rousselot D, Bittar R, Seilhean D, Hauw J-J, Lacomblez L, Loeffler J-P, Meininger V (2008) Dyslipidemia is a protective factor in amyotrophic lateral sclerosis. *Neurology* 70(13):1004–1009
- Hiroo Ichikawa MK (2010) Language impairment in amyotrophic lateral sclerosis. *Brain Nerve* 62:435–440
- Donaghy C, Thurtell MJ, Pioro EP, Gibson JM, Leigh RJ (2011) Eye movements in amyotrophic lateral sclerosis and its mimics: a review with illustrative cases. *J Neurol Neurosurg Psychiatry* 82:110–116
- Hammad M, Silva A, Glass J, Sladky JT, Benatar M (2007) Clinical, electrophysiologic, and pathologic evidence for sensory abnormalities in ALS. *Neurology* 69:2236–2242
- Bouteloup C, Desport JC, Clavelou P, Guy N, Derumeaux-Burel H, Ferrier A, Couratier P (2009) Hypermetabolism in ALS patients: an early and persistent phenomenon. *J Neurol* 256:1236–1242. doi:10.1007/s00415-009-5100-z
- Desport JC, Torny F, Lacoste M, Preux PM, Couratier P (2005) Hypermetabolism in ALS: correlations with clinical and para-clinical parameters. *Neurodegener Dis* 2:202–207. doi:10.1159/000089626
- Dupuis L, Corcia P, Fergani A, Gonzalez De Aguilar JL, Bonnefont-Rousselot D, Bittar R, Seilhean D, Hauw JJ, Lacomblez L, Loeffler JP, Meininger V (2008) Dyslipidemia is a protective factor in amyotrophic lateral sclerosis. *Neurology* 70:1004–1009. doi:10.1212/01.wnl.0000285080.70324.27
- Paganoni S, Zhang M, Quiroz Zarate A, Jaffa M, Yu H, Cudkowicz ME, Wills AM (2012) Uric acid levels predict survival in men with amyotrophic lateral sclerosis. *J Neurol* 259:1923–1928. doi:10.1007/s00415-012-6440-7
- Harno K, Rissanen A, Palo J (1984) Glucose tolerance in amyotrophic lateral sclerosis. *Acta Neurol Scand* 70:451–455
- Reyes ET, Perurena OH, Festoff BW, Jorgensen R, Moore WV (1984) Insulin resistance in amyotrophic lateral sclerosis. *J Neurol Sci* 63:317–324
- Shulga A, Rivera C (2012) Interplay between Thyroxin, BDNF and GABA in injured neurons. *Neuroscience* 239:241–252. doi:10.1016/j.neuroscience.2012.12.007
- Sampson D, Pickard M, Evans I, Leonard A, Sinha A, Ekins R (2002) Thyroid hormone regulates the expression of alpha-internexin in neurons in culture. *NeuroReport* 13:273–276
- Konig S, Moura Neto V (2002) Thyroid hormone actions on neural cells. *Cell Mol Neurobiol* 22:517–544
- Schlosser G, Koyano-Nakagawa N, Kintner C (2002) Thyroid hormone promotes neurogenesis in the *Xenopus* spinal cord. *Dev Dyn* 225:485–498. doi:10.1002/dvdy.10179
- Funalot B, Desport JC, Sturtz F, Camu W, Couratier P (2009) High metabolic level in patients with familial amyotrophic lateral sclerosis. *Amyotroph Lateral Scler* 10:113–117. doi:10.1080/17482960802295192
- Cozzolino M, Ferri A, Valle C, Carri MT (2012) Mitochondria and ALS: implications from novel genes and pathways. *Mol Cell Neurosci*. doi:10.1016/j.mcn.2012.06.001
- Hamadeh MJ, Rodriguez MC, Kaczor JJ, Tarnopolsky MA (2005) Caloric restriction transiently improves motor performance but hastens clinical onset of disease in the Cu/Zn-superoxide dismutase mutant G93A mouse. *Muscle Nerve* 31:214–220. doi:10.1002/mus.20255
- Li J, Paulson JM, Ye FD, Sung M, Hollenberg AN, Rutkove SB (2012) Reducing systemic hypermetabolism by inducing hypothyroidism does not prolong survival in the SOD1-G93A mouse. *Amyotroph Lateral Scler* 13:372–377. doi:10.3109/17482968.2012.662988
- Zoccollella S, Beghi E, Palagano G, Fraddosio A, Guerra V, Samarelli V, Lepore V, Simone IL, Lamberti P, Serlenga L, Logroscino G (2008) Predictors of long survival in amyotrophic lateral sclerosis: a population-based study. *J Neurol Sci* 268:28–32. doi:10.1016/j.jns.2007.10.023
- Pallavi C, Bhautik M, Sarita M, Pranay J, Rita S (2011) Variation in thyroid hormones level among people of different age, gender and seasons, Piparia, Gujarat. *Natl J Med Res* 2:57–59
- Jang YY, Kim CY, Hwang TY, Kim KD, Lee CH (2008) Reference interval of serum thyroid hormones in healthy Korean adults. *J Prev Med Public Health* 41:128–134 [pii] 200803128
- Iwasaki Y, Kinoshita M, Ikeda K, Takamiya K, Shiojima T (1989) Amyotrophic lateral sclerosis and thyroid function. *J Neurol* 236:373–374
- Ilzecka J, Stelmasiak Z (2003) Thyroid function in patients with amyotrophic lateral sclerosis. *Ann Univ Mariae Curie Sklodowska Med* 58:343–347
- Kiessling WR (1982) Thyroid function in 44 patients with amyotrophic lateral sclerosis. *Arch Neurol* 39:241–242
- Malin JP, Koddig R, Fuhrmann H, von zur Muhlen A (1989) T4, T3 and rT3 levels in serum and cerebrospinal fluid of patients with amyotrophic lateral sclerosis. *J Neurol* 236:57–59
- Trentin AG (2006) Thyroid hormone and astrocyte morphogenesis. *J Endocrinol* 189:189–197. doi:10.1677/joe.1.06680
- Alvarez-Dolado M, Gonzalez-Sancho JM, Bernal J, Munoz A (1998) Developmental expression of the tenascin-C is altered by hypothyroidism in the rat brain. *Neuroscience* 84:309–322 [pii] S0306452297005113
- Iglesias T, Caubin J, Stunnenberg HG, Zaballos A, Bernal J, Munoz A (1996) Thyroid hormone-dependent transcriptional repression of neural cell adhesion molecule during brain maturation. *EMBO J* 15:4307–4316
- Trentin AG, Moura Neto V (1995) T3 affects cerebellar astrocyte proliferation, GFAP and fibronectin organization. *NeuroReport* 6:293–296

31. Martinez R, Gomes FC (2005) Proliferation of cerebellar neurons induced by astrocytes treated with thyroid hormone is mediated by a cooperation between cell contact and soluble factors and involves the epidermal growth factor-protein kinase a pathway. *J Neurosci Res* 80:341–349. doi:[10.1002/jnr.20463](https://doi.org/10.1002/jnr.20463)
32. Trentin AG, De Aguiar CB, Garcez RC, Alvarez-Silva M (2003) Thyroid hormone modulates the extracellular matrix organization and expression in cerebellar astrocyte: effects on astrocyte adhesion. *Glia* 42:359–369. doi:[10.1002/glia.10228](https://doi.org/10.1002/glia.10228)