A call for diagnostic and prognostic biomarkers in Amyotrophic Lateral Sclerosis: is Neurofilament Light Chain the answer?

Gaiani A, Martinelli I, Bello L, Querin G, Puthenparampil M, Ruggero S, Toffanin E, Briani C, Cagnin A, Pegoraro E, Sorarù G
Department of Neurosciences, University of Padua

Introduction: NF levels in CSF, serum and plasma of patients with neurological and neurodegenerative disorders have been extensively investigated and recognized as promising biomarkers of neuroaxonal breakdown in miscellaneous conditions. The elevation appears to be more significant in patients with ALS, with a strong discriminatory capacity. The **aim of the study** was to validate the diagnostic and prognostic role of NFL in ALS, testing the discriminatory capacity between FTD, MN and controls, among different subtypes of motor neuron disease (typical ALS, UMN-d ALS, PBP, PMA, Flail arm/leg), between genetic and sporadic ALS and in patients with ALS and cognitive impairment.

Methods: this was a single centre retrospective longitudinal study on 176 patients who underwent a lumbar puncture at the Department of Neurosciences of the University of Padua, Neurology ward, from January 2010 until February 2016 (**Table 1**). NFL levels were measured on CSF stored at -80° C by UmanDiagnostics NF-light® enzyme-linked immunosorbent assay (ELISA). NFL levels were compared between ALS, FTD, MN and CTRL groups, between sporadic and genetic ALS and between different subtypes of ALS using the Wilcoxon-Mann-Whitney U test. Correlations between log[NFL] and quantitative variables were assessed using Pearson's correlation coefficient. The ANCOVA was used to test the influence of various covariates on log[NFL]. Longitudinal changes of ALSFRSr and MITOS scores were assessed in repeated measures ANCOVA models. Effects of log[NFL] on survival of ALS patients were analyzed by Cox regression.

Group	n	M/F	Age at lumbar puncture in years (median, IR)	CSF conservation time in months (median, IR)
ALS	94	64/30	62.5 (52.3-70)	42 (25-57.5)
CTRL	44	24/20	54 (37-69.3)	40 (34-53)
FTD	20	8/12	65 (61-72.3)	37 (7.5-65.8)
MN	18	14/4	63 (57-71.5)	39.5 (14.3-48.5)
P	-	0.036*	0.009**	0.536

Table 1: Characteristics of the study groups

** p ≤ 0,01, significant; * p ≤ 0,05, significant

Results: log[NFL] was higher in ALS and FTD groups compared to MN and controls (p < 0,0001, **Figure 1**). Typical ALS, PBP, and UMND had higher levels of NFL than flail arm/leg and PMA (p < 0,0001, **Figure 2**). There was an inverse correlation between log[NFL] and overall survival (HR 2.45, 95% CI 1.66 - 3.61, p < 0,0001, **Table 2, Figure 3**). Disease progression was predicted by log[NFL] (p< 0,0001, **Figure 4**). There was no evidence of different log[NFL] and survival in genetic ALS.

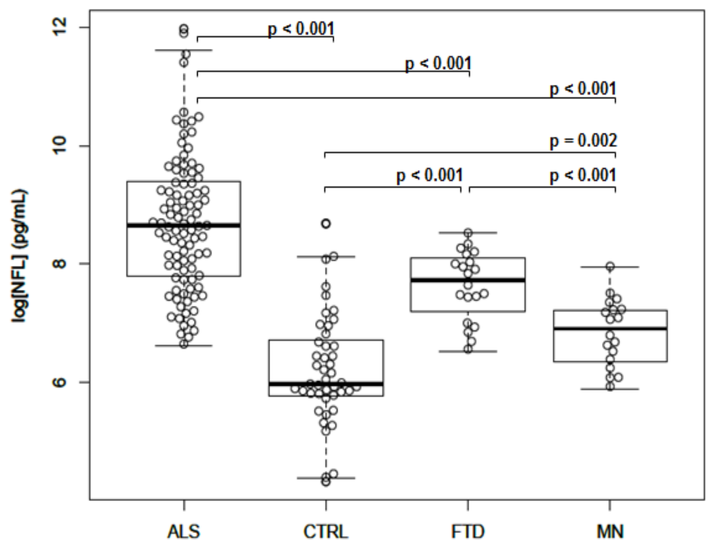

Figure 1: NFL concentration in the four study groups. Nonparametric descriptive parameters are shown (median, lower and upper quartiles) indicating the central location and scatter/dispersion of the observations.

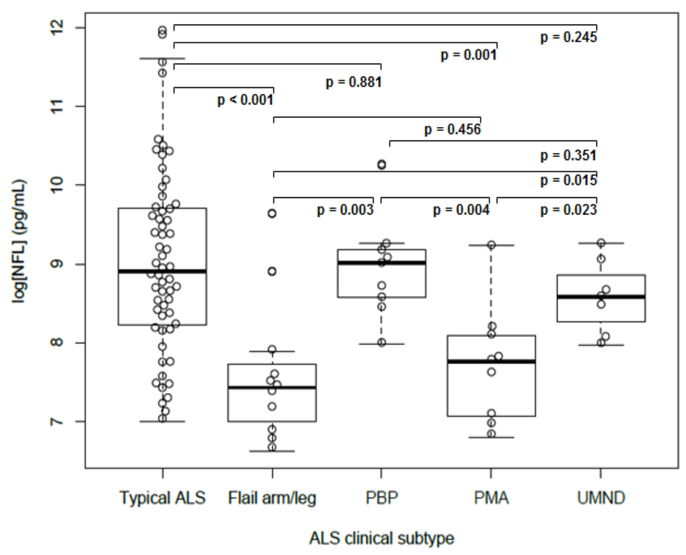

Figure 2: NFL concentration within the ALS group, divided into clinical subtypes.

Covariates		HR	SE	p Value	95% CI
Log [NFL] ALS		2.447	0.198	< 0.0001 ***	1.659 – 3.611
ALS onset age		1.08352	0.02011	< 0.0001 ***	1.042 – 1.127
ΔFS		1.16515	0.22631	0.4994	0.748 – 1.816
Subtype	Typical ALS	1°	-	-	-
	Flail Arm/Leg	0.280	0.649	0.049*	0.079 – 0.998
	PBP	1.477	0.475	0.412	0.582 – 3.750
	PMA	0.17373	1.04846	0.095	0.022 – 1.356
	UMND	0.11803	0.83463	0.0105*	0.023 – 0.606
Gender	F	1°°	-	-	-
	M	0.6158	0.36403	0.1829	0.302 – 1.257
Cognitive impairment	no	1°°°	-	-	-
	yes	0.93152	0.45231	0.8754	0.384 – 2.261

Table 2: Measures for the Cox regression analysis of effects of CSF NFL concentration, subtype, age, sex, cognitive impairment and ΔFS on overall survival.
*** p ≤ 0.0001, significant; * p ≤ 0,05, significant.

° "Typical ALS" was used as reference in the model (HR set at 1)
° ° "F" was used as reference in the model (HR set at 1)
° ° ° "no" was used as reference in the model (HR set at 1)

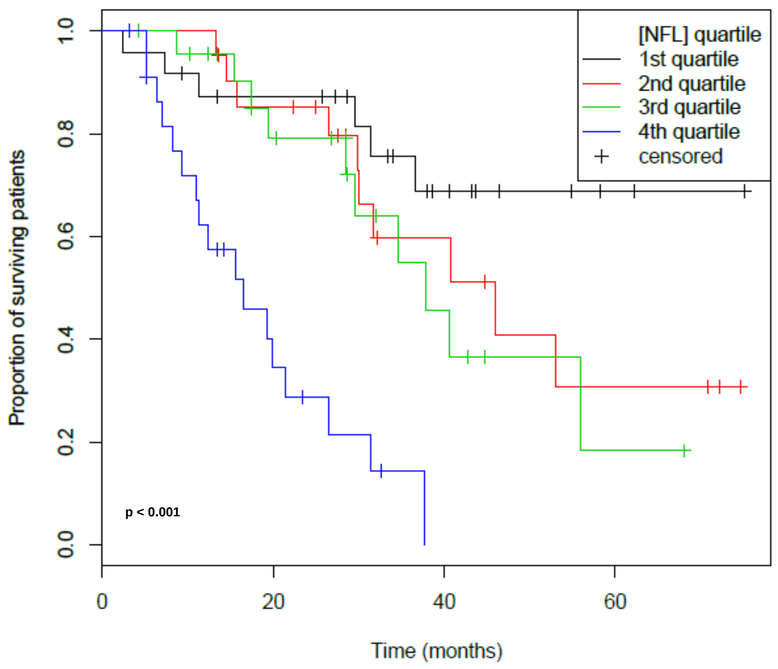

Figure 3: Kaplan-Meier plots of the proportion of surviving patients relative to time (months) from baseline, grouped by log [NFL]

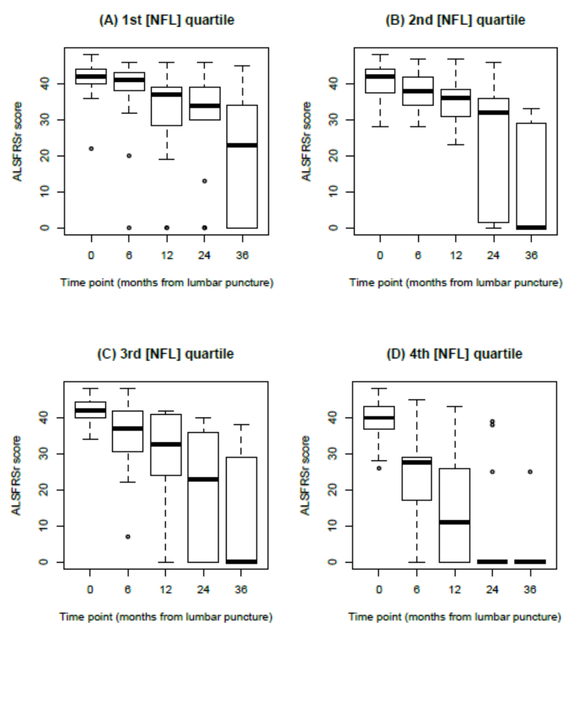

Figure 4: ALSFRSr change relative to time (months from lumbar puncture) by [NFL] quartiles.

Conclusions : our data confirm the role of NFL as biomarker in ALS. NFL elevation in patients with upper motor neuron involvement and in FTD might reflect the corticospinal tract degeneration. Low NFL levels in patients with lower motor neuron signs might predict milder phenotypes of disease.

Glossary: NF = Neurofilaments; CSF = Cerebrospinal Fluid; NFL = Neurofilament light chain; ALS = Amyotrophic Lateral Sclerosis; FTD = Fronto Temporal Dementia; MN = Motor Neuropathies; CTRL = Controls; UMN-d ALS = Upper Motor Neuron-dominant ALS; PBP = Progressive Bulbar Palsy; PMA = Progressive Muscular Atrophy; ALSFRS-r = ALS Functional Rating Scale-Revised score; MITOS = Milano Torino Staging; ANCOVA = Analysis of Covariance, CI= confidence interval; HR = Hazard Ratio; SE = standard error; ΔFS = progression rate of disease from onset to CFS;

XLVII CONGRESSO NAZIONALE
22-25 OTTOBRE 2016 – VENEZIA
Sin