PLASMA GFAP LEVELS ARE INCREASED IN SYMPTOMATIC FTD MUTATION CARRIERS WITHIN THE GENFI COHORT

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Introduction

Why are biomarkers so important for FTD?

Frontotemporal dementia (FTD) is a progressive, neurodegenerative condition with 30% of cases caused by an autosomal dominant gene mutation, including in progranulin (GRN), chromosome 9 open reading frame 72 (C9orf72) or microtubule-associated protein tau (MAPT). As clinical trials are fast approaching, identification of robust and easily accessible biomarkers is paramount to monitor treatment response.

Neurofilament light (NfL), a constituent of the neural cytoskeleton, is a marker of neuronal death and axonal degeneration. Glial fibrillary acidic protein (GFAP) is a filamentous structure expressed by mature astrocytes. It is a marker of astrogliosis, which is the abnormal proliferation of astrocytes in response to neuronal damage. Both biomarkers can be measured in CSF and blood-based samples including plasma and serum.

Aim

Investigating GFAP and NfL within GENFI

To test plasma levels of the markers GFAP and NfL in patients from the GENFI cohort. Establish biomarker differences in FTD mutation carriers versus controls and investigate correlations with psychological and imaging measures.

Methods

Participants from the GENFI cohort

Table 1. Participant characteristics

	Controls	*C9orf72*	*GRN*	*MAPT*
	n = 185	n = 111	n = 120	n = 53
Female gender (%)	102 (55.14)	57 (51.35)	70 (58.33)	29 (54.72)
Age at collection, years (IQR)	43.78 (36.38 - 55.06)	52.90 (40.44 - 64.58)	50.28 (38.42 - 59.79)	44.86 (34.60 - 57.56)
MMSE (IQR)	30 (29 - 30)	29 (27 - 30)	30 (28 - 30)	30 (28 - 30)
FTLD-CDR-SOB (IQR)	0 (0 - 0)	0 (0 - 7)	0 (0 - 2.25)	0 (0- 2.5)
GFAP median (IQR)	105.97 (80.40 - 147.23)	129.78 (95.77 - 207.96)	140.38 (89.60 - 226.00)	98.75 (73.13 - 157.99)
NfL median (IQR)	9.38 (6.79 - 13.46)	16.35 (9.53 - 37.29)	11.64 (7.56 - 32.58)	10.74 (7.30 - 19.96)
Gene-specific information		*C9orf72 PS*	*C9orf72 S*	*GRN PS*	*GRN S*	*MAPT PS*	*MAPT S*
	n = 185	n = 71	n = 40	n = 90	n = 30	n = 34	n = 19
Age at onset, years (range)			59 (34 -71)		60 (49 - 76)		54 (37 - 66)
Disease duration, years (IQR)			3.63 (0.87 - 5.58)		1.59 (0.84 - 2.67)		2.52 (0.88 - 4.26)

Table 1. Participant information divided into presymptomatic and symptomatic genetic subgroups, C9orf72, GRN and MAPT. Demographic information portrayed as median and interquartile ranges (IQR).
Abbreviations: MMSE = Mini-mental state examination; FTLD-CDR-SOB = Frontotemporal Lobar Degeneration – Clinical Dementia Rating – Sum of Box Scores

Measurement of plasma biomarkers

Markers were multiplexed using the Neurology 4-plex assay on the Single Molecule Array (SIMOA) platform. 469 plasma samples were included in total and measured in duplicates using one batch of reagents.

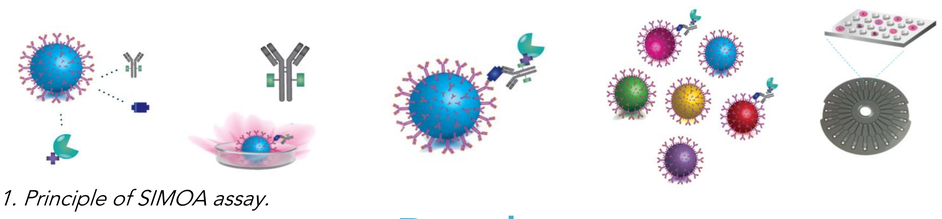

Figure 1. Principle of SIMOA assay.

Results

GFAP and NfL within the GENFI cohort

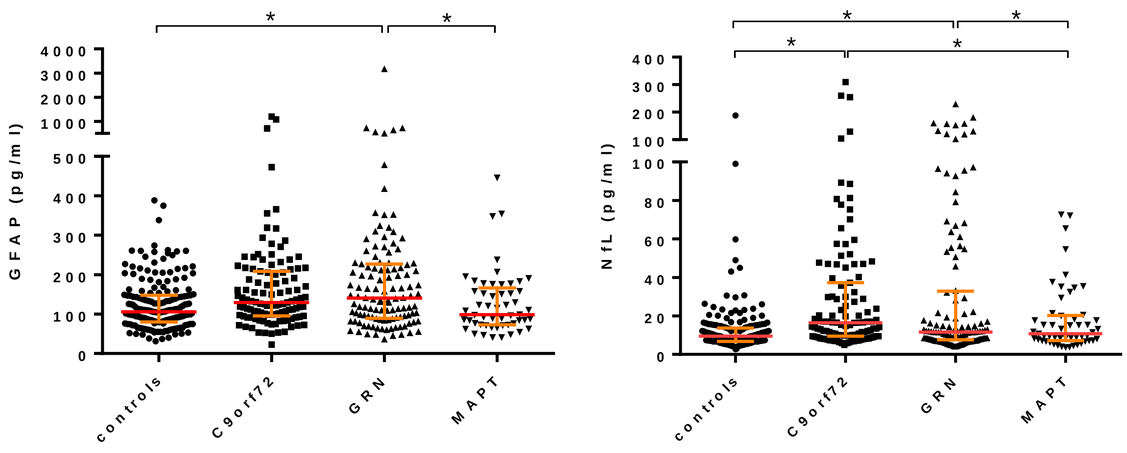

Figure 2. GFAP (left) and NfL concentrations (right) in control and genetic FTD groups C9orf72, GRN and MAPT. Median designated by red line, interquartile ranges indicated by orange error bars.

Results

Statistical analysis of biomarker differences

Group differences			Plasma GFAP bootstrap regression model				
Group	vs	Group	Observed Coef.	Bias	Bootstrap Std. Err.	95% CI	
Controls		C9	18.51	-0.08	16.48	-6.90	63.58
Controls		GRN	59.89	-0.34	22.98	**31.33**	**150.89**
Controls		MAPT	1.67	0.15	11.56	-19.53	25.57
C9		GRN	41.37	-0.26	31.16	-1.49	143.62
C9		MAPT	-16.84	0.24	19.61	-61.52	15.43
GRN		MAPT	-58.22	0.49	25.26	**-140.25**	**-23.66**

Group differences			Plasma NfL bootstrap regression model				
Group	vs	Group	Observed Coef.	Bias	Bootstrap Std. Err.	95% CI	
Controls		C9	12.65	-0.09	4.55	**5.78**	**24.77**
Controls		GRN	15.78	0.00	3.51	**9.46**	**23.19**
Controls		MAPT	4.56	0.00	2.52	-0.62	9.32
C9		GRN	3.13	0.10	5.57	-9.26	13.42
C9		MAPT	-8.09	0.10	4.84	**-20.79**	**-0.62**
GRN		MAPT	-11.22	0.00	3.86	**-19.25**	**-4.26**

Table 2 & 3. Plasma GFAP and NfL data was not normally distributed (D'Agostino-Pearson omnibus normality test); therefore group mean differences were compared using a linear regression model with 95% bias-corrected bootstrap confidence intervals (CIs) with 2000 replications, adjusting for age in Stata v.14. Gender had no statistically significant effect on biomarker levels. Significant levels in bold.

Correlations of GFAP and NfL with age

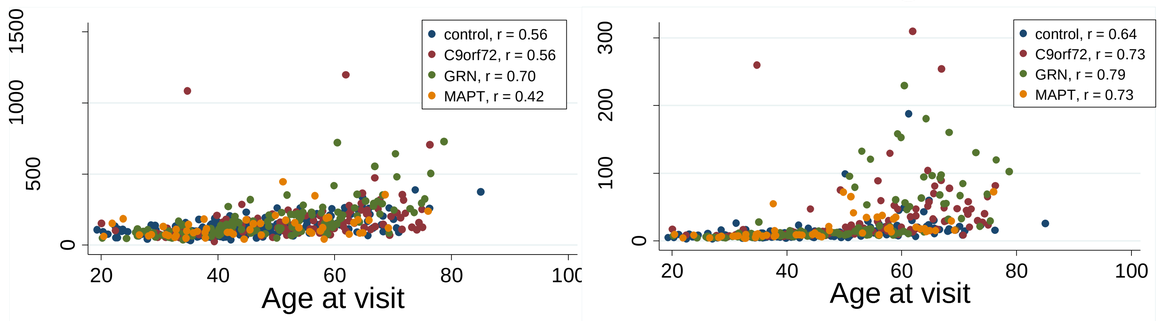

Figure 3. Spearman's correlations were run to assess the relationship between age at sample collection and levels of GFAP in FTD mutation carriers and controls. There was a positive correlation between GFAP and age, which was statistically significant in all groups. ($p < 0.05$).

Correlations of GFAP with psychological measures

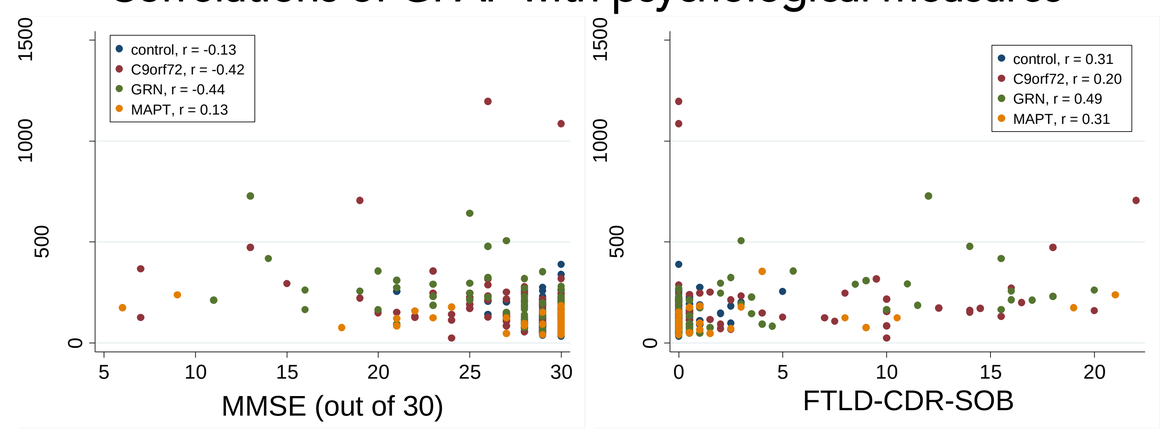

Figure 3. Spearman's correlations were run to assess the relationship between psychological measures, MMSE and FTLD-CDR-SOB, with levels of GFAP in FTD mutation carriers and controls. There was a negative correlation between GFAP and MMSE in C9orf72 and GRN ($p < 0.05$). There was a positive correlation with FTLD-CDR-SOB and GFAP in GRN and controls ($p < 0.05$).

Correlations of GFAP with imaging data

Cross-sectional imaging correlation

Brain region		*Controls*	*C9orf72*	*GRN*	*MAPT*
Total brain	r	-0.32	-0.49	-0.46	-0.17
	p	**0.000**	**0.0000**	**0.0000**	0.2354
Frontal lobe	r	-0.22	-0.43	-0.47	-0.31
	p	**0.004**	**0.0000**	**0.0000**	**0.0309**
Temporal lobe	r	-0.34	-0.35	-0.56	-0.31
	p	**0.0000**	**0.0001**	**0.0000**	**0.0333**
Parietal lobe	r	-0.35	-0.41	-0.35	-0.25
	p	**0.0000**	**0.0000**	**0.0003**	0.0874
Occipital lobe	r	-0.17	-0.24	-0.14	-0.14
	p	**0.0252**	**0.0128**	0.1586	0.3312
Cingulate	r	-0.27	-0.45	-0.50	-0.12
	p	**0.0003**	**0.0000**	**0.0000**	0.4138
Insula	r	-0.32	-0.42	-0.52	-0.22
	p	**0.0000**	**0.0000**	**0.0000**	0.129

Longitudinal imaging correlation

Brain region		*Controls*	*C9orf72*	*GRN*	*MAPT*
Total brain	r	0.05	0.10	0.32	-0.06
	p	0.5818	0.4676	**0.0129**	0.7672
Frontal lobe	r	-0.17	0.11	0.20	0.03
	p	0.0805	0.4278	0.1313	0.8639
Temporal lobe	r	-0.06	-0.14	0.32	0.05
	p	0.5395	0.3055	**0.0117**	0.7906
Parietal lobe	r	-0.19	-0.01	0.32	-0.03
	p	0.0595	0.9187	**0.0119**	0.8748
Occipital lobe	r	-0.02	0.17	0.21	-0.33
	p	0.8296	0.2312	0.1001	0.082
Cingulate	r	-0.06	0.17	0.17	-0.09
	p	0.5252	0.2312	0.1886	0.6496
Insula	r	-0.05	0.00	0.37	-0.09
	p	0.6119	0.9804	**0.004**	0.6537

Table 3 & 4. Spearman's correlations were run to assess the relationship between cross-sectional and longitudinal Imaging data and levels of GFAP in FTD mutation carriers and controls. There was a negative correlation between GFAP and most brain regions (cross-sectional imaging data) primarily in the C9orf72 and GRN mutation carriers and controls. Negative correlations of GFAP with brain regions known to be affected in FTD was only significant in the GRN mutation carriers.

Conclusions

GFAP and NfL were measured in 469 plasma samples from the GENFI cohort. Plasma GFAP levels were significantly higher in the *GRN* mutation carriers compared to controls and *MAPT* carriers. GFAP correlated with age and psychological measures in all groups. Cross-sectional imaging data correlated with GFAP in most brain regions across various groups. Longitudinal atrophy of brain regions known to be affected in FTD were only seen in the *GRN* mutation carriers. These findings further strengthen the hypothesis that inflammation plays a key role in FTD, in particular in the *GRN* mutation carriers. Further investigation of longitudinal changes in plasma GFAP is required, to determine its use as a biomarker.

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