Supplementary Table 1. Example of the dataset structure.

Patient _ID	Time from baseline	sNfL_change	Outcome _measure1	Outcome _measure2-5
1	0_12	-2.1% (change between M1 and M12)	-0.56% (change between M0 and M12)	…
1	0_24	-26.3% (change between M1 and M24)	-0.95% (change between M0 and M24)	…
1	0_36	15.3% (change between M1 and M36)	-1.26% (change between M0 and M36)	…
2	0_12	-59.6% (change between M1 and M12)	-0.44% (change between M0 and M12)	…
2	0_24	-12.5% (change between M1 and M24)	-0.84% (change between M0 and M24)	…
2	0_36	-0.3% (change between M1 and M36)	-0.99% (change between M0 and M36)	…
…	…	…	…	…
172	0_12	10.6% (change between M1 and M12)	-0.90% (change between M0 and M12)	…
172	0_24	-64.3% (change between M1 and M24)	-1.80% (change between M0 and M24)	…
172	0_36	12.3% (change between M1 and M36)	-2.50% (change between M0 and M36)	…

Example of the structure of univariate mixed models (results shown in Table 2):

lme (sNfL_change ~ Outcome_measure + Time_from_baseline, random = ~1|Patient_ID, data = Name_of_dataset)

Example of the structure of the multivariate mixed models (results shown in Table 3):

lme (sNfL_change ~ Outcome_measure1 + Outcome_measure2 + Outcome_measure3 + Outcome_measure4

+ Outcome_measure5 + Time_from_baseline, random=~1|Patient_ID, data = Name_of_dataset)

Legend: lme: liner mixed effect model analysis; **sNfL_change:** logarithmically transformed relative change in serum neurofilament light chain level between month 1 and specific timepoint; **Outcome_measure1-5:** change in clinical or imaging measure (one of the following: absolute change of EDSS, absolute change of T1 and T2 lesion volume from baseline, cumulative number of T2 lesions from baseline, number of gadolinium-enhancing lesions at particular timepoints and whole brain, grey matter and corpus callosum volume percentage volume changes from baseline) between baseline and specific timepoint; **Time_from_baseline:** specification of the timepoint; **Patient_ID:** identification of certain patient (1|Patient_ID = random intercept)

Supplementary Table 2. Spearman cross-sectional correlations between serum neurofilament light chain (sNfL) level at M1 and clinical and MRI parameters at baseline.

Variable	Rho	p-value
Age at onset	-0.09	0.281
Time between onset and baseline	-0.09	0.27
EDSS	0.21	0.01
T2 lesion volume (cm^3)	0.46	<0.001
T1 lesion volume (cm^3)	0.36	<0.001
T2 lesion number	0.23	0.006
GAD lesion number	0.35	<0.001
Grey matter fraction (%)	0.02	0.793
Corpus callosum fraction (%)	-0.14	0.083
Brain parenchymal fraction (%)	0.08	0.338

Legend: Rho: Spearman rho; **EDSS:** Expanded Disability Status Scale; **GAD lesion number:** number of gadolinium-enhancing lesions

Supplementary Table 3. Serum neurofilament light chain (sNfL) levels at different timepoints.

	N	Mean	Median	Minimum	Maximum	IQR
sNfL at screening	156	38.66	20.71	1.39	416. 21	13.84 - 42.30
sNfL at month 0	64	43.99	22.68	4.97	475.78	12.62 - 39.89
sNfL at month 1	157	32.23	17.70	0.44	268.94	10.99 - 31.05
sNfL at month 12	155	19.00	13.86	1.69	147.57	9.51 - 21.29
sNfL at month 24	135	17.46	12.48	0.54	171.73	8.61 - 18.00
sNfL at month 36	126	15.17	12.24	3.11	137.26	8.96 - 16.49

Legend: N: number of available measures; **IQR:** interquartile range; **sNfL:** serum neurofilaments light chain

sNfL levels are in pg/ml.

Supplementary Table 4. Spearman correlations among serum neurofilament light chain (sNfL) levels at different timepoints.

	sNfL at screening	sNfL at month 0	sNfL at month 1	sNfL at month 12	sNfL at month 24	sNfL at month 36
sNfL at screening	1.00	**0.71*****	**0.79*****	**0.47*****	**0.41*****	**0.23***
sNfL at month 0	**0.71*****	1.00	**0.93*****	**0.39****	0.21	0.10
sNfL at month 1	**0.79*****	**0.93*****	1.00	**0.41*****	**0.31*****	0.18
sNfL at month 12	**0.47*****	**0.39****	**0.41*****	1.00	**0.58*****	**0.58*****
sNfL at month 24	**0.41*****	0.21	**0.31*****	**0.58*****	1.00	**0.68*****
sNfL at month 36	**0.23***	0.10	0.18	**0.58*****	**0.68*****	1.00

*** are Spearman correlations with $p<0.001$, ** are $p<0.01$, * are $p<0.05$.

Supplementary Table 5. The longitudinal relationship between percentage changes of serum neurofilament light chain (sNfL) levels (change between screening and 12; screening and 24; screening and 36) and change of clinical and imaging explanatory variables (change between: month 0 and 12; month 0 and 24; month 0 and 36) analysed by multivariate mixed-effects models.

Variable	Regression coefficient	p-value
Intercept	1.825	<0.001
Time from baseline	-0.086	0.005
EDSS absolute change	-0. 055	0.252
Cumulative relapse number	0.059	0.124
T1 lesion volume absolute change	0.191	<0.001
Cumulative number or T2 lesions	0.049	0.001
Whole brain volume % change	5.797	0.097

Legend: EDSS: Expanded Disability Status Scale; **AIC (**Akaike information criterion) = 494.0

Supplementary Table 6. Spearman cross-sectional correlations between serum neurofilament light chain (sNfL) levels at different timepoints and MRI parameters at 48 months.

Timepoint	BPF	Log T2 lesion volume	Whole brain volume % change
sNfL at screening	-0.27***	0.45***	-0.36***
sNfL at 1 month	-0.24**	0.40***	-0.33***
sNfL at 12 months	-0.16	0.42***	-0.26**
sNfL at 24 months	-0.29**	0.49***	-0.15
sNfL at 36 months	-0.27**	0.29**	-0.16

Legend: BPF = brain parenchymal fraction; reported Spearman rho

*** p<0.001; ** p<0.01; * p<0.05

Supplementary Table 7. Multivariate linear regression models (adjusted for sex and age) showing the best predictors of global or regional brain volume loss over 48 months.

Variable	Predictors at baseline	Regression coefficient	p-value
WB %	Intercept	0.007	0.210
	sNfL at month 1	0.005	0.001
	T2 lesion volume	0.001	0.106
	Gender	0.001	0.701
	Age	<0.001	0.031
CC %	Intercept	-0.064	0.012
	sNfL at month 1	0.031	<0.001
	T2 lesion volume	0.004	0.016
	Gender	0.012	0.208
	Age	<0.001	0.46
GM %	Intercept	0.004	0.828
	sNfL at month 1	0.014	0.001
	T2 lesion volume	<0.001	0.816
	Gender	0.005	0.418
	Age	-0.001	0.021
WB %	Intercept	0.007	0.259
	sNfL at month 1	0.005	<0.001
	T1 lesion volume	0.002	0.163
	Gender	0.001	0.726
	Age	<0.001	0.033

	Intercept	-0.068	0.008
	sNfL at month 1	0.033	<0.001
CC %	T1 lesion volume	0.010	0.038
	Gender	0.012	0.219
	Age	<0.001	0.464
	Intercept	0.004	0.794
	sNfL at month 1	0.013	0.001
GM %	T1 lesion volume	0.002	0.597
	Gender	0.005	0.399
	Age	-0.001	0.019
	Intercept	0.008	0.158
	sNfL at month 1	0.004	0.003
	GAD lesion number	0.002	0.023
WB %	Gender	-0.001	0.760
	Age	<0.001	0.104
	Intercept	-0.047	0.032
	sNfL at month 1	0.023	<0.001
CC %	GAD lesion number	0.020	<0.001
	Gender	-0.002	0.836
	Age	<0.001	0.895
	Intercept	0.009	0.611
	sNfL at month 1	0.010	0.008
GM%	GAD lesion number	0.004	0.061
	Gender	0.002	0.798

	Age	-0.001	0.073
WB %	Intercept	0.009	0.114
	sNfL at month 1	0.004	0.002
	T2 lesion number	<0.001	0.004
	Gender	<0.001	0.948
	Age	<0.001	0.011
CC %	Intercept	-0.062	0.009
	sNfL at month 1	0.030	<0.001
	T2 lesion number	0.001	0.002
	Gender	0.008	0.355
	Age	<0.001	0.388
GM %	Intercept	0.009	0.606
	sNfL at month 1	0.011	0.004
	T2 lesion number	<0.001	0.069
	Gender	0.002	0.714
	Age	-0.001	0.028

Legend: WB%: percentage change of whole brain volume between baseline and month 48 (%); **CC%:** percentage change of corpus callosum volume between baseline and month 48 (%); **GM%:** percentage change of grey matter volume between baseline and month 48 (%); **sNfL:** logarithmically transformed serum neurofilament light chain level; T1 and T2 lesion volume (cm^3) and T2 lesion number were measured at baseline; **GAD:** gadolinium enhancing