An age dependent Mathematical Model of Neurofilament Trafficking in Healthy Conditions

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Supplementary Material

$$
\text{CNS}\begin{cases}
\frac{d[\text{NfL}]_{CNS}}{dt} &= r_1^L - r_2\,[\text{NfL}]_{CNS} - r_5^L\,f(age)\,[\text{NfL}]_{CNS}\\
\frac{d[\text{NfM}]_{CNS}}{dt} &= r_1^M - r_2\,[\text{NfM}]_{CNS} - r_5^M\,f(age)\,[\text{NfM}]_{CNS}\\
\frac{d[\text{pNfH}]_{CNS}}{dt} &= r_1^H - r_2\,[\text{pNfH}]_{CNS} - r_5^H\,f(age)\,[\text{pNfH}]_{CNS}
\end{cases}
$$

$$
\text{PNS}\begin{cases}
\frac{d[\text{NfL}]_{PNS}}{dt} &= r_3^L - r_4\,[\text{NfL}]_{PNS} - r_6^L\,f(age)\,[\text{NfL}]_{PNS}\\
\frac{d[\text{NfM}]_{PNS}}{dt} &= r_3^M - r_4\,[\text{NfM}]_{PNS} - r_6^M\,f(age)\,[\text{NfM}]_{PNS}\\
\frac{d[\text{pNfH}]_{PNS}}{dt} &= r_3^H - r_4\,[\text{pNfH}]_{PNS} - r_6^H\,f(age)\,[\text{pNfH}]_{PNS}
\end{cases}
$$

$$
\text{ISF}\begin{cases}
\frac{d[\text{NfL}]_{ISF}}{dt} &= r_5^L\,f(age)\,\frac{V_{CNS}}{V_{ISF}}\,[\text{NfL}]_{CNS} - \frac{r_7}{V_{ISF}}\,[\text{NfL}]_{ISF}\\
\frac{d[\text{NfM}]_{ISF}}{dt} &= r_5^M\,f(age)\,\frac{V_{CNS}}{V_{ISF}}\,[\text{NfM}]_{CNS} - \frac{r_7}{V_{ISF}}\,[\text{NfM}]_{ISF}\\
\frac{d[\text{pNfH}]_{ISF}}{dt} &= r_5^H\,f(age)\,\frac{V_{CNS}}{V_{ISF}}\,[\text{pNfH}]_{CNS} - \frac{r_7}{V_{ISF}}\,[\text{pNfH}]_{ISF}
\end{cases}
$$

$$
\text{Endoneurial fluid}\begin{cases}
\frac{d[\text{NfL}]_{endo}}{dt} &= r_6^L\,f(age)\,\frac{V_{PNS}}{V_{endo}}\,[\text{NfL}]_{PNS} - \frac{r_8}{V_{endo}}\,[\text{NfL}]_{endo}\\
\frac{d[\text{NfM}]_{endo}}{dt} &= r_6^M\,f(age)\,\frac{V_{PNS}}{V_{endo}}\,[\text{NfM}]_{PNS} - \frac{r_8}{V_{endo}}\,[\text{NfM}]_{endo}\\
\frac{d[\text{pNfH}]_{endo}}{dt} &= r_6^H\,f(age)\,\frac{V_{PNS}}{V_{endo}}\,[\text{pNfH}]_{PNS} - \frac{r_8}{V_{endo}}\,[\text{pNfH}]_{endo}
\end{cases}
$$

$$
\text{CSF cranial}\begin{cases}
\frac{d[\text{NfL}]_{cranial}}{dt} &= \frac{r_7}{V_{cranial}}\,[\text{NfL}]_{ISF} - \frac{r_9}{V_{cranial}}\,[\text{NfL}]_{cranial} + \frac{r_{10}}{V_{cranial}}\,[\text{NfL}]_{spinal} - \frac{r_{11}}{V_{cranial}}\,[\text{NfL}]_{cranial}\\
\frac{d[\text{NfM}]_{cranial}}{dt} &= \frac{r_7}{V_{cranial}}\,[\text{NfM}]_{ISF} - \frac{r_9}{V_{cranial}}\,[\text{NfM}]_{cranial} + \frac{r_{10}}{V_{cranial}}\,[\text{NfM}]_{spinal} - \frac{r_{11}}{V_{cranial}}\,[\text{NfM}]_{cranial}\\
\frac{d[\text{pNfH}]_{cranial}}{dt} &= \frac{r_7}{V_{cranial}}\,[\text{pNfH}]_{ISF} - \frac{r_9}{V_{cranial}}\,[\text{pNfH}]_{cranial} + \frac{r_{10}}{V_{cranial}}\,[\text{pNfH}]_{spinal} - \frac{r_{11}}{V_{cranial}}\,[\text{pNfH}]_{cranial}
\end{cases}
$$

$$
\text{CSF spinal}\begin{cases}
\frac{d[\text{NfL}]_{spinal}}{dt} &= \frac{r_9}{V_{spinal}}\,[\text{NfL}]_{cranial} - \frac{r_{10}}{V_{spinal}}\,[\text{NfL}]_{spinal} - \frac{r_{12}}{V_{spinal}}\,[\text{NfL}]_{spinal} + \frac{r_8}{V_{spinal}}\,[\text{NfL}]_{endo}\\
\frac{d[\text{NfM}]_{spinal}}{dt} &= \frac{r_9}{V_{spinal}}\,[\text{NfM}]_{cranial} - \frac{r_{10}}{V_{spinal}}\,[\text{NfM}]_{spinal} - \frac{r_{12}}{V_{spinal}}\,[\text{NfM}]_{spinal} + \frac{r_8}{V_{spinal}}\,[\text{NfM}]_{endo}\\
\frac{d[\text{pNfH}]_{spinal}}{dt} &= \frac{r_9}{V_{spinal}}\,[\text{pNfH}]_{cranial} - \frac{r_{10}}{V_{spinal}}\,[\text{pNfH}]_{spinal} - \frac{r_{12}}{V_{spinal}}\,[\text{pNfH}]_{spinal} + \frac{r_8}{V_{spinal}}\,[\text{pNfH}]_{endo}
\end{cases}
$$

$$
\text{Blood}\begin{cases}
\frac{d[\text{NfL}]_{blood}}{dt} &= \frac{r_{11}}{V_{blood}}\,[\text{NfL}]_{cranial} + \frac{r_{12}}{V_{blood}}\,[\text{NfL}]_{spinal} - r_{13}^L\,[\text{NfL}]_{blood}\\
\frac{d[\text{NfM}]_{blood}}{dt} &= \frac{r_{11}}{V_{blood}}\,[\text{NfM}]_{cranial} + \frac{r_{12}}{V_{blood}}\,[\text{NfM}]_{spinal} - r_{13}^M\,[\text{NfM}]_{blood}\\
\frac{d[\text{pNfH}]_{blood}}{dt} &= \frac{r_{11}}{V_{blood}}\,[\text{pNfH}]_{cranial} + \frac{r_{12}}{V_{blood}}\,[\text{pNfH}]_{spinal} - r_{13}^H\,[\text{pNfH}]_{blood}
\end{cases}
$$

Figure S1. Ordinary Differential Equation system representing the implementation of the Nf trafficking model. The system describes the concentration levels in the seven compartments of the model for NfL, NfM, and pNfH separately. Reaction rates that have different values for the three subunits have the label "L", "M", or "H". Parameter estimates and the definition of the f(age) function are provided in Supplementary Table S1.

Parameter	Nf Subunit	Estimate	Reference
r1	NfL	1310.678 ng/(ml*day)	Assumed to be at equilibrium with degradation reaction r2 for initial concentration in CNS
	NfM	11755.78 ng(ml*day)	Assumed to be at equilibrium with degradation reaction r2 for initial concentration in CNS
	pNfH	12300.21 ng/(ml*day)	Assumed to be at equilibrium with degradation reaction r2 for initial concentration in CNS
r2	All	0.01 1/day	[1] [2]
r3	NfL	1310.678 ng/(ml*day)	Assumed to be at equilibrium with degradation reaction r4 for initial concentration in PNS
	NfM	11755.78 ng(ml*day)	Assumed to be at equilibrium with degradation reaction r4 for initial concentration in PNS
	pNfH	12300.21 ng/(ml*day)	Assumed to be at equilibrium with degradation reaction r4 for initial concentrations in PNS
r4	All	0.01 1/day	[1] [2]
r5	NfL	7.3×10^{-6} 1/day	Estimated in this work
	NfM	0.60×10^{-6} 1/day	Estimated in this work
	pNfH	0.58×10^{-6} 1/day	Estimated in this work
r6	NfL	7.3×10^{-6} 1/day	Assumed equal to r5
	NfM	0.60×10^{-6} 1/day	Assumed equal to r5
	pNfH	0.58×10^{-6} 1/day	Assumed equal to r5
r7	All	449.28 ml/day	[3] [4]
r8	All	449.28 ml/day	Assumed equal to r7
r9	All	7.4712 ml/day	[5]
r10	All	20.148 ml/day	[5]
r11	All	90 ml/day	Derived from [6] [7] [4]
r12	All	259.2 ml/day	[8]
r13	NfL	10.1 1/day	Estimated in this work
	NfM	0.0181 /day	Estimated in this work
	pNfH	7.4 1/day	Estimated in this work
f(age)	All	$0.0975 \times 1.031^{age}$	Modeled to reproduce NfL data in [9]

Table S1. Parameter estimates of the model with references.

Compartment	Volume (ml)	Reference
CNS	1265	[10]
PNS neurons	130	See the following supplementary sections
Interstitial Fluid (ISF)	225	[11] [12]
Endoneurial fluid	88	See the following supplementary sections
CSF cranial	75	[5] (half of total CSF volume)
CSF spinal	75	[5] (half of total CSF volume)
Blood	5110	[13], considering human weight of 70 kg

Table S2. Volumes of the model compartments with references. For some compartments, the derivation of the volume is detailed in the section "Estimate of PNS and endoneurial fluid volumes".

Subunit	Initial concentration in CNS and PNS	Reference
NfL	1.3×10^5 ng/ml	[14], assuming 100 mg of total proteins in 1 g of brain
NfM	1.17×10^6 ng/ml	[14], assuming 100 mg of total proteins in 1 g of brain
pNfH	1.22×10^6 ng/ml	[14], assuming 100 mg of total proteins in 1 g of brain

Table S3. Nf initial concentrations in CNS and PNS neurons used to compute the steady state condition. Nf concentration in the remaining model compartments has been deduced from the model steady state computed during parameter calibration, as indicated in the Methods section.

ESTIMATE OF PNS AND ENDONEURIAL FLUID VOLUMES

The volume of the peripheral nervous system and the endoneurial fluid were derived using a series of heterogeneous data. The starting point was an estimate of the percentage of nerve trunks in the body (about 1% of the total body weight [15]). If we consider a body weight of 70 kg, a 30-75% of connective tissue in the PNS (www.nysora.com), the percentage of fiber [16] and if we assume a 1:1 ratio between sensory and motor neurons in the nerve fascicles, we managed to derive the values reported in Supplementary Table S2.

GLOBAL SENSITIVITY ANALYSIS

r7	r6	r5	r4	r3	r2	r1	PNS neuron	ISF volume	Endoneurial	CSF spinal	CSF cranial	CNS neuron	Blood vol.	Age	
0.00000	0.00000	0.00675	0.00000	0.00000	1.00000	0.99997	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.01948	NfL CNS neurons
0.00000	0.00000	0.00044	0.00000	0.00000	1.00000	1.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00126	NfM CNS neurons
0.00000	0.00000	0.00041	0.00000	0.00000	1.00000	1.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00119	pNfH CNS neurons
0.00000	0.00818	0.00000	0.99997	0.99997	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.01953	NfL motor neurons
0.00000	0.00053	0.00000	1.00000	1.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00127	NfM motor neurons
0.00000	0.00050	0.00000	1.00000	1.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00119	pNfH motor neurons
1.00000	0.00000	0.99997	0.00000	0.00000	1.00000	0.99997	0.00000	0.00000	0.00000	0.00000	0.00000	1.00000	0.00000	2.74527	NfL ISF
1.00000	0.00000	1.00000	0.00000	0.00000	1.00000	1.00000	0.00000	0.00000	0.00000	0.00000	0.00000	1.00000	0.00000	2.74738	NfM ISF
1.00000	0.00000	1.00000	0.00000	0.00000	1.00000	1.00000	0.00000	0.00000	0.00000	0.00000	0.00000	1.00000	0.00000	2.74739	pNfH ISF
0.00000	0.99997	0.00000	0.99997	0.99997	0.00000	0.00000	1.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	2.74621	NfL endoneurial f.
0.00000	1.00000	0.00000	1.00000	1.00000	0.00000	0.00000	1.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	2.74745	NfM endoneurial f.
0.00000	1.00000	0.00000	1.00000	1.00000	0.00000	0.00000	1.00000	0.00000	0.00000	0.00000	0.00000	0.00000	0.00000	2.74746	pNfH endoneurial f.
0.00000	0.12421	0.99939	0.99997	0.99997	1.00000	0.99997	0.12947	0.00000	0.00000	0.00000	0.00000	0.99967	0.00000	2.74520	NfL CSF cranial
0.00000	0.12441	0.99964	1.00000	1.00000	1.00000	1.00000	0.12952	0.00000	0.00000	0.00000	0.00000	0.99967	0.00000	2.74738	NfM CSF cranial
0.00000	0.12442	0.99964	1.00000	1.00000	1.00000	1.00000	0.12952	0.00000	0.00000	0.00000	0.00000	0.99967	0.00000	2.74739	pNfH CSF cranial
0.00000	0.96705	0.93443	0.99996	0.99996	1.00000	0.99996	0.97369	0.00000	0.00000	0.00000	0.00000	0.94560	0.00000	2.74591	NfL CSF spinal
0.00000	0.96727	0.93497	1.00000	1.00000	1.00000	1.00000	0.97370	0.00000	0.00000	0.00000	0.00000	0.94561	0.00000	2.74744	NfM CSF spinal
0.00000	0.96727	0.93497	1.00000	1.00000	1.00000	1.00000	0.97370	0.00000	0.00000	0.00000	0.00000	0.94561	0.00000	2.74744	pNfH CSF spinal
0.00000	0.54374	0.99156	0.99997	0.99997	1.00000	0.99997	0.53731	0.00000	0.00000	0.00000	0.00000	0.99250	1.00000	2.74517	NfL blood
0.00000	0.54412	0.99196	1.00000	1.00000	1.00000	1.00000	0.53740	0.00000	0.00000	0.00000	0.00000	0.99250	1.00000	2.74739	NfM blood
0.00000	0.54412	0.99196	1.00000	1.00000	1.00000	1.00000	0.53740	0.00000	0.00000	0.00000	0.00000	0.99250	1.00000	2.74740	pNfH blood

r13	r12	r11	r10	r9	r8
0.00000	0.00000	0.00000	0.00000	0.00000	0.00000
0.00000	0.00000	0.00000	0.00000	0.00000	0.00000
0.00000	0.00000	0.00000	0.00000	0.00000	0.00000
0.00000	0.00000	0.00000	0.00000	0.00000	0.00000
0.00000	0.00000	0.00000	0.00000	0.00000	0.00000
0.00000	0.00000	0.00000	0.00000	0.00000	0.00000
0.00000	0.00000	0.00000	0.00000	0.00000	0.00000
0.00000	0.00000	0.00000	0.00000	0.00000	0.00000
0.00000	0.00000	0.00000	0.00000	0.00000	0.00000
0.00000	0.00000	0.00000	0.00000	0.00000	1.00000
0.00000	0.00000	0.00000	0.00000	0.00000	1.00000
0.00000	0.00000	0.00000	0.00000	0.00000	1.00000
0.00000	0.10867	0.98228	0.12963	0.23751	0.00000
0.00000	0.10872	0.98228	0.12965	0.23751	0.00000
0.00000	0.10872	0.98228	0.12966	0.23751	0.00000
0.00000	0.98306	0.84146	0.22324	0.83990	0.00000
0.00000	0.98306	0.84148	0.22324	0.83992	0.00000
0.00000	0.98306	0.84148	0.22324	0.83992	0.00000
1.00000	0.00000	0.00000	0.00000	0.00000	0.00000
1.00000	0.00000	0.00000	0.00000	0.00000	0.00000
1.00000	0.00000	0.00000	0.00000	0.00000	0.00000

Table S4. Global sensitivity factors of the rates and volumes of the model computed for each Nf variable. The color scale intensity, from green to white, follows the absolute values of the local sensitivity, from the greatest values to zero.

NfL blood		
Local sensitivity at 25 years	**Local sensitivity at 45 years**	**Local sensitivity at 85 years**
r13	Age	Age
Blood volume	r13	r13
r1	Blood volume	Blood volume
r2	r1	r1
CNS neuron volume	r2	r2
r5	CNS neuron volume	CNS neuron volume
Age	r5	r5
PNS neuron volume	PNS neuron volume	PNS neuron volume
r6	r6	r6
r3	r3	r3
r4	r4	r4
r7	r7	r7
r8	r8	r8
r9	r9	r9
r10	r10	r10
r11	r11	r11
r12	r12	r12
ISF volume	ISF volume	ISF volume
Endoneurial volume	Endoneurial volume	Endoneurial volume
CSF cranial volume	CSF cranial volume	CSF cranial volume
CSF spinal volume	CSF spinal volume	CSF spinal volume

Table S5. Local sensitivity factors of model rates and volumes computed for NfL in blood at different ages. The color scale intensity, from green to white, follows the absolute values of the local sensitivity, from the greatest values to zero. The values are sorted by columns in descending sensitivity order.

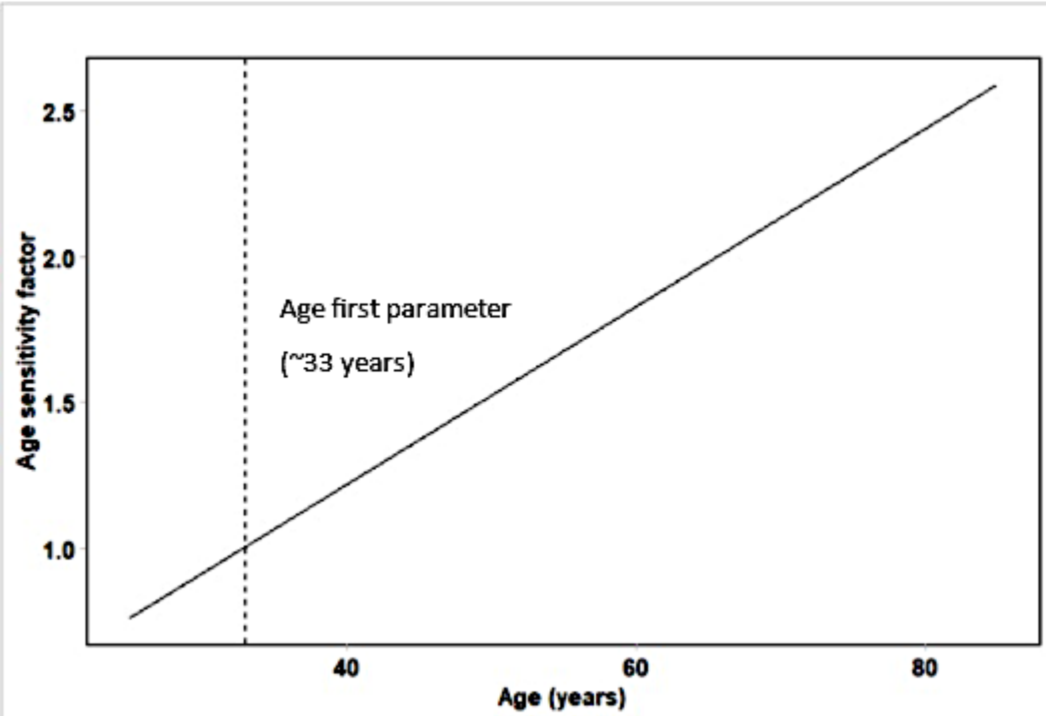

Figure S2. Local sensitivity factor of blood NfL to age as a function of the age itself. The vertical dashed line marks the age at which the age sensitivity factor becomes larger than any other factor considered in the model.

Rate change	r1	r2	r3	r4	r5	r6	r7	r8	r9	r10	r11	r12	r13
-50%	-21.46	42.88	-28.54	57.04	-21.45	-28.54	0.00	0.00	-20.81	3.46	37.46	87.45	0.00
-40%	-17.17	28.59	-22.83	38.03	-17.16	-22.83	0.00	0.00	-16.53	2.75	25.54	59.54	0.00
-30%	-12.87	18.38	-17.13	24.45	-12.87	-17.12	0.00	0.00	-12.31	2.05	16.69	38.87	0.00
-20%	-8.58	10.72	-11.42	14.26	-8.58	-11.41	0.00	0.00	-8.14	1.36	9.86	22.94	0.00
-10%	-4.29	4.77	-5.71	6.34	-4.29	-5.71	0.00	0.00	-4.04	0.67	4.42	10.29	0.00
0%	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
10%	4.29	-3.90	5.71	-5.19	4.29	5.71	0.00	0.00	3.99	-0.67	-3.67	-8.53	0.00
20%	8.58	-7.15	11.42	-9.51	8.58	11.41	0.00	0.00	7.91	-1.32	-6.77	-15.73	0.00
30%	12.87	-9.90	17.13	-13.17	12.87	17.12	0.00	0.00	11.79	-1.97	-9.42	-21.87	0.00
40%	17.17	-12.26	22.83	-16.31	17.16	22.82	0.00	0.00	15.61	-2.61	-11.71	-27.18	0.00
50%	21.46	-14.30	28.54	-19.02	21.45	28.53	0.00	0.00	19.38	-3.24	-13.71	-31.81	0.00

Table S6. Percentage variations of NfL steady state concentration in CSF spinal as a function of the percentage variations of each rate reported in the first column. The color scale intensity corresponds to the reported values, from green (positive values) to red (negative values).

Rate change	r1	r2	r3	r4	r5	r6	r7	r8	r9	r10	r11	r12	r13
-50%	-45.37	90.68	-4.63	9.25	-45.37	-4.63	0.00	0.00	0.00	0.00	0.00	0.00	100.00
-40%	-36.30	60.46	-3.70	6.16	-36.29	-3.70	0.00	0.00	0.00	0.00	0.00	0.00	66.67
-30%	-27.22	38.87	-2.78	3.96	-27.22	-2.78	0.00	0.00	0.00	0.00	0.00	0.00	42.86
-20%	-18.15	22.68	-1.85	2.31	-18.14	-1.85	0.00	0.00	0.00	0.00	0.00	0.00	25.00
-10%	-9.07	10.08	-0.93	1.03	-9.07	-0.92	0.00	0.00	0.00	0.00	0.00	0.00	11.11
0%	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
10%	9.07	-8.25	0.93	-0.84	9.07	0.92	0.00	0.00	0.00	0.00	0.00	0.00	-9.09
20%	18.15	-15.12	1.85	-1.54	18.14	1.85	0.00	0.00	0.00	0.00	0.00	0.00	-16.67
30%	27.22	-20.94	2.78	-2.13	27.21	2.77	0.00	0.00	0.00	0.00	0.00	0.00	-23.08
40%	36.30	-25.92	3.70	-2.64	36.28	3.70	0.00	0.00	0.00	0.00	0.00	0.00	-28.57
50%	45.37	-30.24	4.63	-3.08	45.35	4.62	0.00	0.00	0.00	0.00	0.00	0.00	-33.33

Table S7. Percentage variations of NfL steady state concentration in blood as a function of the percentage variations of each rate reported in the first column. The color scale intensity corresponds to the reported values, from green (positive values) to red (negative values).

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