

Supplementary Materials: Neuron-Specific Enolase (NSE) Predicts Long-Term Mortality in Adult Patients after Cardiac Arrest: Results from a Prospective Trial

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Table S1: Association between NSE serum levels at day 5 & 7 and long-term outcomes.

Primary endpoint: Long-term mortality									
	N	All	Survivors	Non-Survivors	p-value	Univariable HR (95%CI), p-value	Multivariable * adjusted HR (95%CI), p-value	Multivariable ** adjusted HR (95%CI), p-value	ROC AUC (95% CI)
NSE day 5, median (IQR)	122	22.9 (15.4, 47.5)	17.7 (13.8, 25.5)	40.1 (18.8, 84.3)	<0.001	1.42 (1.27, 1.59), p<0.001	1.42 (1.26, 1.6), p<0.001	1.44 (1.26, 1.65), p<0.001	0.76 (0.68, 0.85)
NSE day 7, median (IQR)	78	21.4 (14.4, 33.3)	20 (14.1, 26.3)	24.9 (15.4, 45)	0.040	1.14 (1.01, 1.29), p=0.028	1.17 (0.98, 1.39), p=0.076	1.18 (1.05, 1.33), p=0.007	0.64 (0.51, 0.76)
Secondary endpoint: Neurological outcome (CPC) after 2 years									
	N	All	Good neurological outcome (CPC 1-2)	Poor neurological outcome (CPC 3-5)	p-value	Univariable (OR (95%CI), p-value)	Multivariable * adjusted OR (95%CI), p-value	Multivariable ** adjusted OR (95%CI), p-value	ROC AUC (95%CI)
NSE day 5, median (IQR)	93	25.3 (17.3, 65.9)	17.8 (14.1, 23.1)	46 (22.4, 91.7)	<0.001	1.55 (1.27, 1.88), p<0.001	1.7 (1.3, 2.22), p<0.001	1.52 (1.19, 1.93), p=0.001	0.81 (0.72, 0.89)
NSE day 7, median (IQR)	57	22.6 (15.6, 34.5)	19.3 (14.3, 27.4)	26.2 (17.4, 42.7)	0.068	1.19 (0.97, 1.46), p=0.095	1.37 (0.96, 1.96), P=0.086	1.21 (0.95, 1.54), p=0.121	0.65 (0.50, 0.80)

Data presented as median (interquartile range) or mean (95% confidence interval). NSE, neuron specific enolase; CPC, cerebral performance category; HR, hazard ratio; OR, odds ratio; ROC, receiver operating characteristics curve; AUC, area under the curve; IQR interquartile range. * adjusted for gender, age and comorbidities (coronary artery disease, congestive heart failure, COPD, hypertension, diabetes mellitus, chronic kidney disease, malignant disease, neurological disease). ** adjusted for gender, age and resuscitation circumstances (ROSC, setting of cardiac arrest, bystander CPR, initial rhythm, initial lactate).

Table S2: Performance of NSE serum levels at day 3 to predict mortality at different time points.

Mortality	Survivors below cutoff (n)	Survivors above cutoff (n)	Non-survivors below cutoff (n)	Non-survivors above cutoff (n)	Sensitivity Pr(+A), % (95%CI)	Specificity Pr(-N), % (95%CI)	ROC area, (95%CI)	Likelihood ratio (+), (95%CI)	Likelihood ratio (-), (95%CI)	PPV, % (95%CI)	NPV, % (95%CI)
at 6 month	100	26	23	76	76.8% (67.2%-84.7%)	79.4% (71.2%-86.1%)	0.78 (0.73-0.84)	3.72 (2.60-5.33)	0.29 (0.20-0.42)	74.5% (64.9-82.6%)	81.3% (73.3-87.8%)
at 12 month	99	26	24	76	76.0% (66.4%-84.0%)	79.2% (71.0%-85.9%)	0.78 (0.72-0.83)	3.65 (2.55-5.23)	0.30 (0.21-0.43)	74.5% (64.9-82.6%)	80.5% (72.4-87.1%)
at 18 month	99	26	24	76	76.0% (66.4%-84.0%)	79.2% (71.0%-85.9%)	0.78 (0.72-0.83)	3.65 (2.55-5.23)	0.30 (0.21-0.43)	74.5% (64.9-82.6%)	80.5% (72.4-87.1%)
at 24 month	98	26	25	76	75.2% (65.7%-83.3%)	79.0% (70.8%-85.8%)	0.77 (0.72-0.83)	3.59 (2.50-5.14)	0.31 (0.22-0.45)	74.5% (64.9-82.6%)	79.7% (71.5-86.4%)

Table S3: Performance of NSE serum levels at cut-off 60ng/mL at day 3 to predict long-term outcomes.

Overall long-term mortality												
Survivors below cutoff (n)	Survivors above cutoff (n)	Non-survivors below cutoff (n)	Non-survivors above cutoff (n)	Sensitivity Pr(+A), % (95%CI)	Specificity Pr(-N), % (95%CI)	ROC area, (95%CI)	Likelihood ratio (+), (95%CI)	Likelihood ratio (-), (95%CI)	Hazard ratio (95%CI)	PPV, % (95%CI)	NPV, % (95%CI)	
NSE 60 ng/mL	105	6	53	61	53.5% (43.9%-62.9%)	94.6% (88.6%-98.0%)	0.74 (0.69-0.79)	9.90 (4.46-21.96)	0.49 (0.40-0.60)	20.14 (8.35-48.39)	91.0% (81.5%-96.6%)	66.5% (58.5%-73.8%)
CPC after 2 years												
Good neurological outcome (CPC 1-2) below cutoff (n)	Good neurological outcome (CPC 1-2) above cutoff (n)	Poor neurological outcome (CPC 3-5) below cutoff (n)	Poor neurological outcome (CPC 3-5) above cutoff (n)	Sensitivity Pr(+A), % (95%CI)	Specificity Pr(-N), % (95%CI)	ROC area, (95%CI)	Likelihood ratio (+), (95%CI)	Likelihood ratio (-), (95%CI)	Odds ratio (95%CI)	PPV, % (95%CI)	NPV, % (95%CI)	
NSE 60 ng/mL	59	3	44	61	58.1% (48.1%-67.7%)	95.2% (86.5%-99.0%)	0.77 (0.71-0.82)	12.01 (3.93-36.64)	0.44 (0.35-0.56)	27.27 (8.47-86.94)	95.3% (86.9%-99.0%)	57.3% (47.2%-67.0%)

Neuron specific enolase at a cut-off 60ng/mL day 3; ROC, receiver operating curve; PPV, positive predictive value; NPV, negative predictive value.

Table S4: Performance of NSE serum levels on day 5 & day 7 at different cutoff points to predict overall long-term mortality.

	Survivors below cutoff (n)	Survivors above cutoff (n)	Non-survivors below cutoff (n)	Non-survivors above cutoff (n)	Sensitivity Pr(+A), % (95%CI)	Specificity Pr(-N), % (95%CI)	ROC area, (95%CI)	Likelihood ratio (+), (95%CI)	Likelihood ratio (-) (95%CI)	Hazard ratio (95%CI)	PPV, % (95%CI)	NPV, % (95%CI)
Day 5 NSE												
33 ng/mL	54	9	26	33	55.9 (42.4-68.8)	85.7 (74.6-93.3)	0.71 (0.63-0.79)	3.29 (2.05-7.47)	0.51 (0.38-0.70)	7.62 (3.21-17.98)	78.6 (63.2-89.7)	67.5 (56.1-77.6)
27.4 ng/mL	51	12	23	36	61.0 (47.4-73.5)	81.0 (69.1-89.8)	0.71 (0.63-0.79)	3.20 (1.85-5.54)	0.48 (0.34-0.68)	6.65 (2.96-14.95)	75.0 (60.4-86.4)	68.9 (57.1-79.2)
Day 7 NSE												
33 ng/mL	33	5	25	15	37.5 (22.7-54.2)	86.8 (71.9-95.6)	0.62 (0.53-0.72)	2.85 (1.15-7.08)	0.72 (0.55-0.94)	3.96 (1.31-11.89)	75.0 (50.9-91.3)	56.9 (43.2-69.8)
26.2 ng/mL	28	10	21	19	47.5 (31.5-63.9)	73.7 (56.9-86.6)	0.61 (0.50-0.71)	1.80 (0.97-3.37)	0.71 (0.50-1.01)	2.53 (0.99-6.48)	65.5 (45.7-82.1)	57.1 (42.2-71.2)

At each day cutoff at 33 ng/mL as recommended and cutoff based on Youden index is shown. NSE Neuron specific enolase; ROC, receiver operating curve; PPV, positive predictive value; NPV, negative predictive value

Table S5: Performance of NSE serum levels at different cutoff points to predict neurological outcome after two years.

	Good neurological outcome below cutoff (n)	Good neurological outcome above cutoff (n)	Poor neurological outcome below cutoff (n)	Poor neurological outcome above cutoff (n)	Sensitivity Pr(+A), % (95%CI)	Specificity Pr(-N), % (95%CI)	ROC area, (95%CI)	Likelihood ratio (+), (95%CI)	Likelihood ratio (-), (95%CI)	Odds ratio (95%CI)	PPV, % (95%CI)	NPV, % (95%CI)
Max. NSE												
33 ng/mL	49	39	45	170	79.1% (73.0-84.3%)	55.7% (44.4-66.3%)	0.67 (0.61-0.73)	1.78 (1.4-2.28)	0.38 (0.27-0.52)	4.75 (2.79-8.08)	81.3% (75.4-86.4%)	52.1% (41.6-62.5%)
42.1 ng/mL	64	24	63	152	70.7% (64.1-76.7%)	72.7% (62.2-81.7%)	0.72 (0.66-0.77)	2.59 (1.82-3.69)	0.40 (0.32-0.51)	6.43 (3.71-11.16)	86.4% (80.4-91.1%)	50.4% (41.4-59.4%)
Day 0 NSE												
33 ng/mL	40	28	72	107	59.8% (52.2-67.0%)	58.8% (46.2-70.6%)	0.59 (0.52-0.66)	1.45 (1.07-1.98)	0.68 (0.52-0.89)	2.12 (1.21-3.73)	79.3% (71.4-85.8%)	35.7% (26.9-45.3%)
34.7 ng/mL	44	24	76	103	57.5% (49.9-64.9%)	64.7% (52.2-75.9%)	0.61 (0.54-0.68)	1.63 (1.15-2.30)	0.66 (0.51-0.84)	2.48 (1.40-4.42)	81.1% (73.2-87.5%)	36.7% (28.1-45.9%)
Day 1 NSE												
33 ng/mL	54	25	56	121	68.4% (61.0-75.1%)	68.4% (56.9-78.4%)	0.68 (0.62-0.75)	2.16 (1.54-3.03)	0.46 (0.36-0.60)	4.67 (2.65-8.23)	82.9% (75.8-88.6%)	49.1% (39.4-58.8%)
38 ng/mL	62	17	66	111	62.7% (55.1-69.9%)	78.5% (67.8-86.9%)	0.71 (0.65-0.76)	2.91 (1.88-4.51)	0.48 (0.38-0.59)	6.13 (3.32-11.31)	86.7% (79.6-92.1%)	48.4% (39.5-57.4%)
Day 2 NSE												
33 ng/mL	59	13	40	96	70.6% (62.2-78.1%)	81.9% (71.1-90.0%)	0.76 (0.70-0.82)	3.91 (2.37-6.47)	0.36 (0.27-0.48)	10.89 (5.42-21.87)	88.1% (80.5-93.5%)	59.6% (49.3-69.3%)
31.9 ng/mL	57	15	37	99	72.8% (64.5-80.1%)	79.2% (68.0-87.8%)	0.76 (0.70-0.82)	3.49 (2.20-5.55)	0.34 (0.25-0.46)	10.17 (5.16-20.01)	86.8% (79.2-92.4%)	60.6% (50.0-70.6%)
Day 3 NSE												
33 ng/mL	52	10	26	79	75.2% (65.9-83.1%)	83.9% (72.3-92.0%)	0.80 (0.73-0.86)	4.66 (2.62-8.32)	0.30 (0.21-0.42)	15.80 (7.09-35.1)	88.8% (80.3-94.5%)	66.7% (55.1-76.9%)
37.1 ng/mL	54	8	28	77	73.3% (63.8-81.5%)	87.1% (76.1-94.3%)	0.80 (0.74-0.86)	5.68 (2.95-10.96)	0.31 (0.22-0.43)	18.56 (7.95-43.15)	90.6% (82.3-95.8%)	65.9% (54.6-76.0%)

Day 5 NSE												
33 ng/mL	34	3	22	34	60.7% (46.8- 73.5%)	91.9% (78.1- 98.3%)	0.76 (0.68- 0.84)	7.49 (2.48- 22.61)	0.43 (0.30-0.60)	17.52 (5.04- 59.81)	91.9% (78.1- 98.3%)	60.7% (46.8- 73.5%)
25.7 ng/mL	32	5	18	38	67.9% (54.0- 79.7%)	86.5% (71.2- 95.5%)	0.77 (0.69- 0.85)	5.02 (2.18- 11.57)	0.37 (0.25-0.56)	13.51 (4.62- 39.15)	88.4% (74.9- 96.1%)	64.0% (49.2- 77.1%)
Day 7 NSE												
33 ng/mL	17	3	23	14	37.8% (22.5- 55.2%)	85.0% (62.1- 96.8%)	0.61 (0.50- 0.73)	2.52 (0.82- 7.75)	0.73 (0.54-1.00)	3.45 (0.9- 12.88)	82.4% (56.6- 96.2%)	42.5% (27.0- 59.1%)
21.7 ng/mL	13	7	15	22	59.5% (42.1- 75.2%)	65.0% (40.8- 84.6%)	0.62 (0.49- 0.76)	1.70 (0.88- 3.27)	0.62 (0.38-1.03)	2.72 (0.90- 8.24)	75.9% (56.5- 89.7%)	44.1% (27.2- 62.1%)

At each day cutoff at 33 ng/mL as recommended and cutoff based on Youden index is shown. CPC 1-2 was considered good neurological outcome and CPC 3-5 as poor neurological outcome. NSE, neuron specific enolase; ROC, receiver operating curve; PPV, positive predictive value; NPV, negative predictive value.

Table S6: Performance of NSE serum levels to predict overall long-term mortality in patients aged 16–53.9 years.

Overall Mortality					
Max. NSE	Sensitivity Pr(+A), % (95%CI)	Specificity Pr(-N), % (95%CI)	ROC area, (95%CI)	PPV, % (95%CI)	NPV, % (95%CI)
33 ng/mL	100.0% (88.8–100.0%)	44.6% (31.3–58.5%)	0.72 (0.66–0.79)	50.0% (37.0–63.0%)	100.0% (86.3–100.0%)
Day 0 NSE					
33 ng/mL	71.4% (51.3–86.8%)	57.8% (42.2–72.3%)	0.65 (0.53–0.76)	51.3% (34.8–67.6%)	76.5% (58.8–89.3%)
Day 1 NSE					
33 ng/mL	88.9% (70.8–97.6%)	60.0% (45.9–73.0%)	0.74 (0.66–0.83)	52.2% (36.9–67.1%)	91.7% (77.5–98.2%)
Day 2 NSE					
33 ng/mL	92.0% (74.0–99.0%)	80.0% (66.3–90.0%)	0.86 (0.78–0.94)	69.7% (51.3–84.4%)	95.2% (83.8–99.4%)
Day 3 NSE					
33 ng/mL	100.0% (83.9–100.0%)	80.0% (65.4–90.4%)	0.90 (0.84–0.96)	70.0% (50.6–85.3%)	100.0% (90.3–100.0%)
Day 5 NSE					
33 ng/mL	90.0% (55.5–99.7%)	83.3% (62.6–95.3%)	0.87 (0.74–0.99)	69.2% (38.6–90.9%)	95.2% (76.2–99.9%)
Day 7 NSE					
33 ng/mL	100.0% (29.2–100.0%)	83.3% (58.6–96.4%)	0.92 (0.83–1.00)	50.0% (11.8–88.2%)	100.0% (78.2–100.0%)

Cutoff at 33 ng/mL as recommended is shown; NSE, neuron specific enolase; ROC, receiver operating curve; PPV, positive predictive value; NPV, negative predictive value.