Neurofilament light chain evaluated with a new automated commercially available method for outcome prediction in out-of-hospital patients with cardiac arrest

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Highlights

- Atellica NfL accurately predicts OHCA outcomes, similar to Simoa NfL.
- NfL levels measured early post-OHCA help guide prognosis and treatment.
- Cutoff values identified for good and poor outcomes at 24, 48, and 72 h.

Abstract

Aim of the study: Neurofilament light (NfL) has high prognostic accuracy as a biomarker for predicting poor outcome when its plasma concentration is measured 12–72 h after out-of-hospital cardiac arrest (OHCA). However, methods suitable for 24/7 clinical use are limited. We performed a post hoc analysis of NfL using samples from 112 patients in the COMACARE trial, applying the Siemens Atellica NfL method, which meets these criteria.

Methods: We analyzed samples collected between 2016 and 2017 using the Siemens Atellica NfL and compared the results with those from the Quanterix Simoa NF-Light method. We assessed the ability to predict poor outcomes, defined as the Cerebral Performance Category (CPC) 3–5, with high specificity at six months post-OHCA by calculating the areas under the receiver operating characteristic curves (AUROC) for NfL concentrations at 24–72 h and compared the results obtained from the two methods.

Results: The Atellica NfL showed high prognostic performance, with AUROCs (0.97–0.98) comparable to those of the Simoa NfL method, having minimal difference (-0.008–0.001). Cut-off values for predicting poor outcome at six months after OHCA with 99% specificity were 112, 229, and 331 ng/L at 24, 48 and 72 h, respectively, whereas they were below 29 ng/mL at 24, 48, and 72 h for predicting good outcome (CPC 1–2) with 100% sensitivity.

Conclusion: The Atellica NfL assay accurately predicts neurological outcomes after OHCA, comparable to that obtained using the Simoa method. It is readily available and can be used in clinical practice.

Keywords: Cardiac arrest, Prognostication, Biomarker, Neurofilament light (NfL)

Introduction

Accurate prognosis of brain injury severity is crucial for patients who remain comatose after resuscitation from cardiac arrest. This information supports clinical decisions regarding the continuation or withdrawal of life-sustaining treatments, assessing the potential for neurological recovery, guiding family discussions, and preparing for long-term rehabilitation or palliative care. Current prognostication guidelines recommend a multimodal approach incorporating clinical evaluations, neurophysiological assessments (somatosensory-evoked potentials and electroencephalogram assessment), blood biomarkers, and brain imaging (computed tomography [CT] and Magnetic Resonance Imaging [MRI]).[1,2] Prognostication is not advised 72 h after cardiac arrest; however, many prognostication tests can be done earlier.[1] A substantial proportion of patients remain indeterminate after prognosis using the current approach[1,3].

Neuron-specific enolase (NSE) remains the only blood biomarker endorsed by the European Resuscitation Council / European Society of Intensive Care Medicine guidelines for detecting neuronal damage.[1,4] Elevated serum/plasma NSE levels at 48 h and/or 72 h after cardiac arrest predict a poor neurological outcome with high specificity but low sensitivity.[1] NSE levels measured early (before 12–24 h) have limited prognostic value, restricting its usefulness for early identification in patients with poor outcomes.[5] Additionally, other conditions, such as hemolysis, ischemic or hemorrhagic stroke, and neuroendocrine tumors, may increase NSE concentration in the blood.[5,6] Furthermore, results may vary considerably, up to 30–40%, depending on the different NSE methods used by different clinical laboratories.[7]

The neurofilament light chain, also called neurofilament light (NfL), is a biomarker of neuroaxonal injury. Elevated blood NfL levels are associated with various neurological diseases, including multiple sclerosis, Alzheimer's disease, frontotemporal dementia, amyotrophic lateral sclerosis, stroke, cerebrovascular disease, traumatic brain injury, and Parkinson's disease.[8] In patients with poor clinical outcomes, NfL levels begin to increase 12 h after cardiac arrest.[9] Several studies have shown that blood NfL has prognostic value for predicting neurological outcomes after cardiac arrest compared with NSE, S100B, clinical examination, neuroimaging, and neurophysiological investigations.[5,10–16] A previous meta-analysis identified NfL as the most accurate

prognostic biomarker after cardiac arrest.[5] It has been suggested that NfL should be incorporated into algorithms and multivariable models alongside other diagnostic tests and clinical scores to enhance the sensitivity and specificity of prognostic models.[8,17,18]

Most previous studies on NfL in patients with cardiac arrest have used the Quanterix Simoa NfL method. However, the Simoa instrument has limited availability in 24/7 clinical laboratories and is less suitable for this type of service. It is a plate-based method that analyses samples in batches, with a relatively long turnaround time (approximately 2.5 h per plate). The International Liaison Committee on Resuscitation (ILCOR) has not recommended the use of NfL.[2] Siemens Healthineers introduced an NfL method using an Atellica immunoanalyzer, which is readily available in clinical laboratories worldwide. Alternatives for fully automated instruments include the Fujirebio Lumipulse, and Roche Cobas Elecsys NFL assays.[19]

In this study, we used samples from the carbon dioxide, oxygen, and mean arterial pressure after cardiac arrest and resuscitation (COMACARE) trial, which involved 112 patients treated in the intensive care unit (ICU) after resuscitation from out-of-hospital cardiac arrest (OHCA).[20–22] We conducted post-hoc analyses using the Siemens Healthineers Atellica NfL method and compared the results with those previously obtained in 2020 with Quanterix Simoa NF-Light.[23] We evaluated the agreement between the two methods and the ability of the Siemens method to predict outcomes after OHCA. Cutoff values for good and poor outcomes were assessed at 24, 48, and 72 h after OHCA.

Material and Methods

Study population and blood samples

The COMACARE study results have been published previously.[20–22] In brief, COMACARE was a randomized clinical trial that compared the effects of low-normal vs. high-normal arterial carbon dioxide targets, normoxia vs. moderate hyperoxia targets, and low-normal vs. high-normal mean arterial pressure targets in comatose patients resuscitated from OHCA. In this study, we obtained frozen samples from 112 patients involved in the COMACARE study between March 2016 and November 2017. We collected samples at the time of ICU admission (0 h) and at 24, 48 and 72 h after OHCA.

Baseline characteristics, resuscitation-associated factors, and the number of blood samples available at the sample collection time points are presented in Table 1 and Figure 1.

Of the samples from 112 patients, one 24-hour, three 48-hour, and six 72-hour blood samples were not available for analysis with either of the method. In addition, one sample with a result available from the Simoa lacked from the sample set analyzed with the Atellica. Conversely, among the samples analyzed with the Simoa, four 24-hour and three 72-hour samples were missing, although these were available for Atellica NfL measurement (Figure 1).

We stored centrifuged plasma/serum samples (à 500 µL) at −70 ^{0}C. These samples had been refrozen at least once and had previously undergone analysis with the Quanterix Single molecule array (Simoa®) NF-Light™ immunoassay (Billerica, Massachusetts, USA) in the Clinical Neurochemistry Laboratory of the University of Gothenburg, Mölndal, Sweden 2019.[23]

Siemens Atellica NfL method

The Atellica IM Neurofilament Light Chain (NfL) is a fully automated method designed for *in vitro* diagnostic use for the quantitative measurement of NfL in human serum and plasma (EDTA) using the Atellica® IM Analyzer. The analyzer operates on a random-access principle, enabling individual sample loading without requiring batch runs, with the first result available in 51 min. The method is CE-IVD-marked but not yet FDA-approved and is intended by Siemens Healthineers to assist in identifying adult patients aged between 18–55 years with Relapsing Multiple Sclerosis. [24] We tested precision and reproducibility using the Atellica IM NfL QC three-level quality control material (Siemens Healthineers, REF 11643634).

Specimen equivalency and stability

Serum and EDTA plasma are recommended specimen types for the Atellica IM NfL method by Siemens [24]. We compared NfL concentrations in corresponding serum and plasma samples by analyzing the samples with an Atellica IM (n=14). We also used surplus samples from the laboratory (n=6) to compare NfL concentrations in unfrozen samples using both the Siemens Atellica IM NfL and Quanterix NfL methods at Labor Berlin-Charité Vivantes GmbH, Berlin, Germany. According to the Siemens NfL reagent

instructions for use, NfL remains stable in serum and plasma for up to four freeze-thaw cycles.[24]

Atellica NfL method comparison to Simoa NF-Light method

We compared the Atellica NfL results from EDTA plasma samples with those obtained using Simoa. In 2024, we thawed, mixed, recentrifuged and analyzed the samples with Siemens Atellica IM NfL reagents (REF 11553991) using the Siemens Atellica® Solution IM 1600 Analyzer (Siemens Healthineers, Erlangen, Germany) at the HUS Diagnostic Center Automation Laboratory, Helsinki, Finland. The Atellica NfL method has a direct measurement range of 3–300 ng/L. Samples with higher concentrations were automatically diluted 1:10 with Atellica IM NfL diluent (Siemens Healthineers, REF 11562429). For samples with NfL concentrations > 3000 ng/L, we manually pre-diluted them 1:10 with 0.9% NaCl to obtain quantitative results, although Siemens does not recommend manual dilution/predilution for this method. For analysis using the Simoa method, we diluted the samples four-fold. Clinical laboratories currently report NfL results of up to 1800 ng/L for undiluted samples, with higher concentrations reported as >1800 ng/L. We used single-reagent lots for both the Simoa and Atellica NfL measurements. According to the manufacturer, the Siemens Atellica NfL method does not exhibit interference exceeding a 10% change in NfL results with hemoglobin at 10 g/L, conjugated bilirubin at 783 umol/L, unconjugated bilirubin at 1129 umol/L or with lipemia at 20 g/L (tested with Intralipid).[24]

Outcomes

A neurologist, blinded to patient data and laboratory results, evaluated neurological outcomes six months after OHCA via telephone interviews. We used the Cerebral Performance Category (CPC) scale [25] to assess outcomes, considering CPC 1–2 as a good outcome and CPC 3–5 as a poor outcome.

Statistical methods

We performed statistical analysis using Analyze-it ® version 4.81.1 for Microsoft Excel. We employed Passing-Bablok regression analysis, Bland–Altman constant, relative bias plots, and Pearson's correlation estimation for method comparison. We tested the normality of distribution using the Shapiro–Wilk test. We compared continuous

variables using the median and interquartile range (IQR), applying the Wilcoxon–Mann–Whitney test to compare central locations (medians).

We determined the ability of the Siemens Atellica IM NfL test to predict poor neurological outcomes at 6 months was assessed by calculating the area under the receiver operating characteristic curve (AUROCs). We compared the AUROCs of the results obtained with the Siemens Atellica IM Atellica NfL to those obtained with the Simoa NfL at ICU admission and at 24, 48, and 72 h after OHCA.

We determined the optimal cutoff value for predicting poor neurological outcomes with Siemens Atellica NfL at 24, 48, and 72 h after cardiac arrest from the ROC curves. We assessed the cutoff value with a specificity higher than 99%. Additionally, we used the Youden method to determine cutoff values for specificities 95–100% and determined corresponding sensitivities, positive predictive values (PPV), negative predictive values (NPV), and positive likelihood ratios (LR+) for NfL in predicting poor outcomes at 6 months. We also determined cutoff values for predicting good outcomes with high sensitivity (100%).

Ethics and approval

The Ethics Committee of the Northern Savo Hospital District, Finland, approved the original COMACARE study protocol (decision no. 295/2015). The Helsinki University Hospital initially approved an extended research permit until 31.12.2024 (decision no.: HUS/334/2023), and further until 31.12.2026 (decision no.: HUS/1110/2025).

The COMACARE study protocol and main results have been published previously, [20–22] and the prognostic accuracy of NfL after OHCA using the Quanterix Simoa NF-Light method has also been published. [23]

Results

Prognostic accuracy of Siemens Atellica IM NfL method

High accuracy in discriminating between patients with poor and good outcome at 24, 48, and 72 h after cardiac arrest was demonstrated using the Siemens Atellica IM NfL method, with AUROC values of 0.975 [95% confidence interval (CI) 0.949 –1.000], 0.983 (0.963–1.000) and 0.978 (0.953–1.000), respectively. In contrast, the Nfl concentration

at ICU admission showed poor prognostic ability, with an AUROC of 0.692 (0.590–0.795) (Figure 2).

No statistically significant difference was observed between the Siemens Atellica IM NfL and Quanterix Simoa NF-Light methods in their ability to discriminate patients with poor and good outcomes at ICU admission and at 24, 48, and 72 h after cardiac arrest (Figure 2). NfL concentrations measured with the Siemens Atellica IM NfL method were significantly higher in patients with poor outcome compared with those with good outcome at all studied time points (Table 2 and Figure 3). The greatest difference in NfL concentrations was observed in samples drawn 48 h after cardiac arrest; the median concentration was 1704 ng/L (interquartile range 583–3408) ng/L in patients with poor outcome and 21 (15-32) ng/L in patients with good outcomes ($p<0.001$) (Table 2).

Cutoff values for poor and good outcome

The cutoff values for predicting poor outcome with 99% specificity were 112 ng/L at 24 h, 229 ng/L at 48 h, and 331 ng/L at 72 h after OHCA, with corresponding sensitivities of 76%, 86%, and 85%, respectively (Table 2). According to the Youden method, the cutoff values were 48 ng/L at 24 h, 80 ng/mL at 48 h, and 236 ng/mL at 72 h, yielding specificities of 90%, 96%, and 99% and sensitivities of 95%, 92%, and 88%, respectively. The cutoff values for predicting poor outcome with specificities of 95–100%, along with corresponding sensitivities, PPV, NPV, and LR+ values, are presented in Table S1.

For predicting good outcome with low NfL concentrations and 100% sensitivity, the cut-off values were 19 ng/mL at 24 h, 29 ng/mL at 48 h, and 27 ng/mL at 72 h after OHCA (Table S2).

Siemens Atellica NfL method compared to Quanterix Simoa NF-Light method

The correlation between the Atellica and Simoa NfL methods was strong (Pearson's coefficient $r >0.95$) at concentrations within the measurement range stated by the manufacturer (Table S2). On average, Atellica NfL results were 12.6% (95% CI 9.3–15.9%) higher compared with the mean of the two methods for NfL concentrations <3000 ng/L. The constant difference was −83 ng/L (95% CI, −121 to −46), indicating an increasing negative bias at higher concentrations. The method comparison showed a positive bias at lower NfL concentrations (<50 ng/L) and a negative bias at higher

concentrations (Figure 4, Table S3). Therefore, the analysis was conducted separately for different NfL concentration ranges (Figures S1 and S2). The Atellica NfL method exhibited good precision and repeatability (Table S4). Serum and EDTA plasma levels were found to be equivalent (Table S5, Figure S3). Additionally, results from the comparison of non-frozen samples were consistent with those obtained from frozen COMACARE samples (Table S6, Figure S4).

Discussion

In this study, we demonstrated that the NfL results obtained using the commercially available Siemens Atellica NfL method were equivalent to those from the more labor-intensive Quanterix Simoa NF-Light method. A key difference for clinical use in this indication is that the Siemens Atellica NfL, being a fully automated instrument, has a shorter turn-around time, allowing single-sample analysis and results to be reported to clinicians more quickly. The Siemens Atellica NfL method also demonstrated high accuracy in predicting neurological outcome after OHCA. Previous studies have demonstrated the efficacy of the Simoa NfL method in these patients. [9,10,23] The prognostic performance of Atellica NfL in predicting neurological outcome at six months after OHCA was consistent, with very high AUROCs (0.97–0.98), comparable to that of the Simoa NfL method. Plasma NfL levels, measured as early as 12 h after cardiac arrest, appear to predict long-term neurological outcomes. [9] A readily available method for 24/7 use has the potential to transform prognostication for these patients.

The cutoff values with 99% specificity for poor neurological outcome were 112 ng/L at 24 h, 229 ng/L at 48 h, and 331 ng/L at 72 h. The cutoff values for Atellica NfL were somewhat lower than those obtained with Simoa NfL in the same samples, [23] which aligns with the observed bias between these methods. Specifically, a positive bias was noted at concentrations below 50 ng/L, while an increasing negative bias occurred at concentrations above 50 ng/L when comparing Siemens Atellica NfL with Quanterix Simoa NF-Light. All NfL immunoassays on the market show a strong correlation; however, bias is observed between certain methods. [19] This highlights the need to validate the cutoff values for each method separately in each laboratory, even though the differences in concentrations were small.

Certified reference materials for NfL are not available; hence, NfL methods still lack standardization. Efforts to produce certified reference materials are ongoing. [26] Similarly, certified reference material for the widely used biomarker NSE are also lacking, and NSE levels vary across laboratories using different NSE methods. [7]

This study has some limitations. The samples were not analyzed simultaneously with the two different methods. The other results were from samples that were originally analyzed several years ago. Nevertheless, the effect of storage was tested with small comparison of unfreezed samples suggesting that storage has no significant effect on the results. Clinical laboratories typically do not report quantitative NfL results above 1800 ng/L using the Simoa method. In our study, we obtained quantitative results for concentrations higher than 1800 ng/L. The systematic negative bias observed at concentrations above 1800 ng/L might be related to the uncertainty of the Simoa NfL method at very high concentrations. Furthermore, the Atellica NfL results >3000 ng/L in this study should be interpreted with caution, as the manufacturer does not recommend manual dilution for these high NfL concentrations. However, the NfL cut-off concentrations associated with poor outcomes in patients with OHCA are considerably lower than these values. [9,23] To the best of our knowledge, this is the first comparison between these two methods that contain samples with such high (>1000 ng/L) NfL concentrations.

NfL levels increase during neuronal injury; however, this increment is not disease-specific. In addition to primary neurological disease, other factors such as age, pregnancy, obesity, diabetes, chronic kidney disease, cardiovascular risk factors, and unrecognized head trauma are also known to affect blood NfL levels. [8,27] Reference values for apparently healthy adults, as established by Siemens, show a median NfL concentration of 7.0 ng/L and a 95% upper percentile limit of 13.9 ng/L. [24] The participants, who were from a US blood donor population (n=684), did not present with known clinically symptomatic diseases; however, underlying disorders, such as neurodegenerative disorders or metabolic diseases, were not studied. [24]

Low NfL levels 24 h after OHCA predict good neurological outcomes. [13,23] In previous studies using the Simoa NfL method, NfL levels lower than 55 ng/L [13] and lower than 30 ng/L (with 100% sensitivity) [23] at 24, 48, and 72 h were predictive of good neurological outcome after OHCA. In our study, the cut-offs for Atellica NfL were 19, 29, and 27 ng/L,

respectively, for the same time points. Confounding factors affecting NfL levels need to be considered; however, increases in NfL caused by other conditions are typically lower than those caused by hypoxic brain injury. [28–30] As a highly sensitive biomarker, NfL has the potential to identify individuals likely to have good outcome. Predicting good outcome using sensitive methods can help reduce the proportion of individuals with indeterminate prognoses. [13,31] Integrating a rapid and accurate biomarker with a low false positive rate is essential for the prognostication of patients at risk of poor neurological outcomes. Early and reliable prediction supports informed decision-making about the continuation or withdrawal of life-sustaining therapies and facilitates timely communication with families regarding prognosis.

Conclusion

Plasma NfL concentration measured with the Atellica NfL predicts outcome after OHCA with high accuracy, comparable with the Simoa method. A 24/7 available method that delivers results quickly can provide a more valuable tool for physicians in accurately predicting outcomes following OHCA.

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Conflict of interest statement

KB has served as a consultant and at advisory boards for Abbvie, AC Immune, ALZPath, AriBio, Beckman-Coulter, BioArctic, Biogen, Eisai, Lilly, Moleac Pte. Ltd, Neurimmune,

Novartis, Ono Pharma, Prothena, Quanterix, Roche Diagnostics, Sanofi and Siemens Healthineers; has served at data monitoring committees for Julius Clinical and Novartis; has given lectures, produced educational materials and participated in educational programs for AC Immune, Biogen, Celdara Medical, Eisai and Roche Diagnostics; and is a co-founder of Brain Biomarker Solutions in Gothenburg AB (BBS), which is a part of the GU Ventures Incubator Program, outside the work presented in this paper. HZ has served at scientific advisory boards and/or as a consultant for Abbvie, Acumen, Alector, Alzinova, ALZpath, Amylyx, Annexon, Apellis, Artery Therapeutics, AZTherapies, Cognito Therapeutics, CogRx, Denali, Eisai, Enigma, LabCorp, Merck Sharp & Dohme, Merry Life, Nervgen, Novo Nordisk, Optoceutics, Passage Bio, Pinteon Therapeutics, Prothena, Quanterix, Red Abbey Labs, reMYND, Roche, Samumed, ScandiBio Therapeutics AB, Siemens Healthineers, Triplet Therapeutics, and Wave, has given lectures sponsored by Alzecure, BioArctic, Biogen, Cellectricon, Fujirebio, LabCorp, Lilly, Novo Nordisk, Oy Medix Biochemica AB, Roche, and WebMD, is a co-founder of Brain Biomarker Solutions in Gothenburg AB (BBS), which is a part of the GU Ventures Incubator Program, and is a shareholder of MicThera (outside submitted work). The other authors declare that they have no competing financial or non-financial interests that could have influenced the work reported in this paper.

CRediT authorship contribution statement:

Christel Pussinen: Conceptualization, Resources, Methodology, Investigation, Data curation, Validation, Formal analysis, Visualization, Writing –original draft, writing – review, and editing.

Mikko Anttonen: Conceptualization, Investigation, Visualization, Writing – review & editing

Lauri Wihersaari: Conceptualization, Resources, Methodology, Investigation, Data curation, Validation, Formal analysis, and visualization

Marjaana Tiainen: review and editing

Matti Reinikainen: Conceptualization, Writing – review & editing

Johanna Hästbacka: Review and editing

Henrik Zetterberg: Conceptualization, Investigation (sample analysis), and writing (review and editing)

Kaj Blennow Conceptualization, Investigation (sample analysis), writing (review and editing).

Kari Pulkki: Conceptualization, Writing – review & editing

Markus B. Skrifvars: Conceptualization, Financial resources, Methodology, Visualization, Writing, review, and editing.

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Tables

Table 1. Characteristics of the patients (n=112) in the study as published by Wihersaari, et al. 2021 [23].

	All patients (n=112)
Age, median (IQR) (years)	62 (53–68)
Male sex [*n* (%)]	92 (82.1)
Weight [median (IQR), kg]	85 (72.3–93)
Neurological function before cardiac arrest	
Normal, CPC 1 [*n* (%)]	103 (92)
Some disability, CPC 2 [*n* (%)]	9 (8)
Medical history	
Hypertension [*n* (%)]	56 (50)
Chronic heart failure (NYHA 3 or 4) [*n* (%)][a]	9 (8)
Smoker [*n* (%)]b	35 (31.3)
Resuscitation factors	
Bystander life support [*n* (%)]	93 (83)
Time to ROSC [median (IQR), min]	21 (16–26)
Clinical status on ICU admission	
GCS [median, (IQR)]c	3 (3–3)
APACHE II score, median (IQR)	28 (24–31)
TTM	
33 °C [*n* (%)]	75 (67)
36 °C [*n* (%)]	36 °C [*n* (%)] 37 (33)

IQR, interquartile range; *NYHA,* New York Heart Association; *ROSC,* return of spontaneous circulation; *ICU,* intensive care unit; *GCS,* Glasgow Coma Scale, *APACHE II,* Acute Physiology and Chronic Health Evaluation; TTM, Targeted temperature management.

[a]Data missing for two patients, b Data missing for 13 patients, c Data missing for nine patients

Table 2. NfL concentrations with Siemens Atellica IM NfL method at ICU admission and 24, 48 and 72 h after cardiac arrest for patients with good outcome (CPC 1–2) and for those with poor outcome (CPC 3–5)

NfL median concentration ng/L (IQR)				Cutoff with 99% specificity	
Time	CPC 1–2	CPC 3–5	p-value	NfL ng/L	Sensitivity
ICU admission	15 (11–19)	20 (14–33)	0.0009		
24 h	17 (11–28)	694 (135–1169)	<0.0001	112	0.76
48 h	21 (15–32)	1704 (583–3408)	<0.0001	229	0.86
72 h	23 (15–35)	1461 (688–2755)	<0.0001	331	0.85

NfL, Neurofilament light; ICU, intensive care unit; CPC Cerebral Performance Category, IQR Interquartile range (1st and 3rd quartiles)

Figures captions

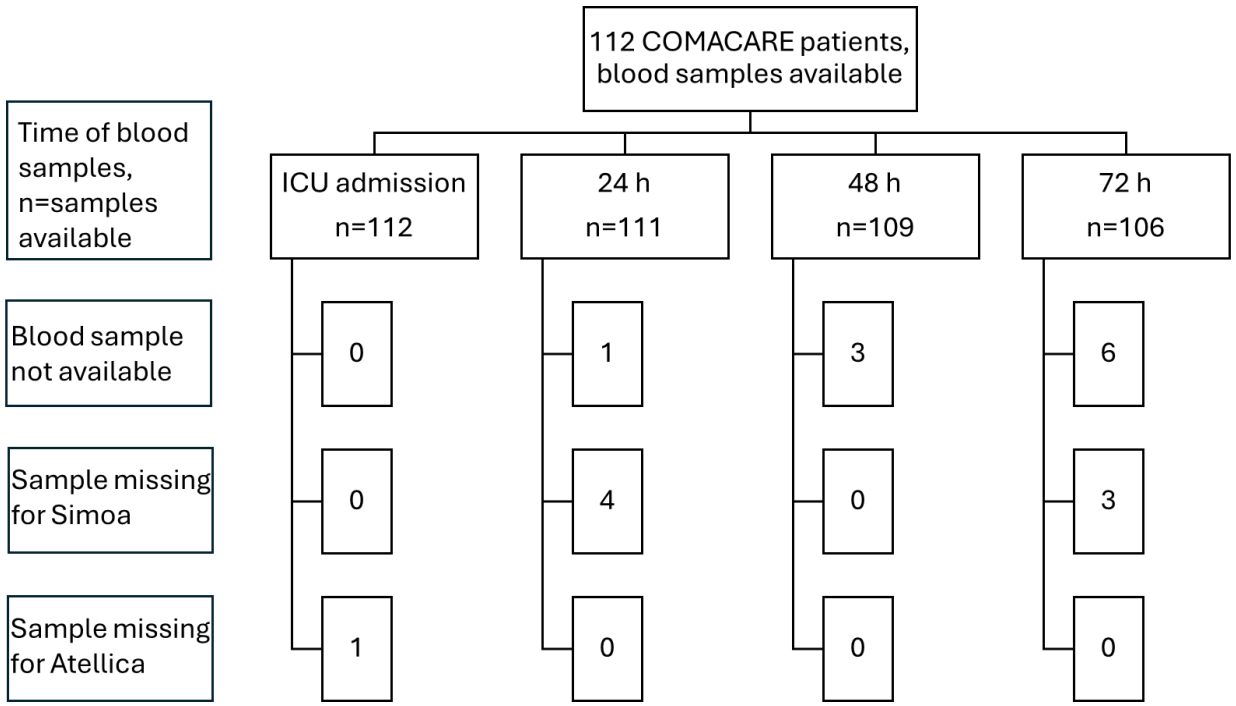

Figure 1. Flow chart of the study population. The sample set consisted of 112 COMACARE-trial patients who had blood samples available.

ICU, intensive care unit; CPC, Cerebral Performance Category

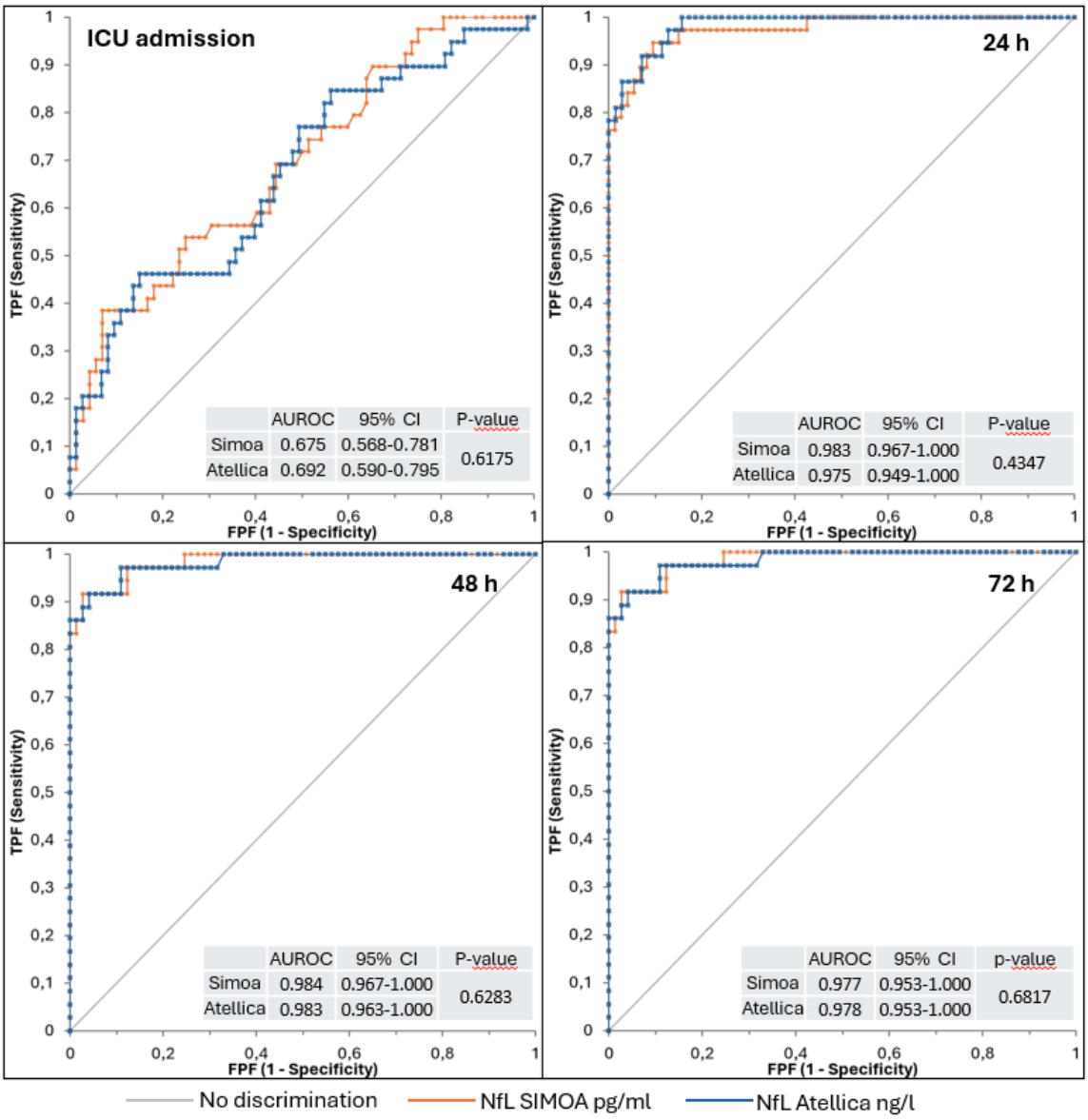

Figure 2. Comparison of Atellica and Simoa receiver operating characteristic (ROC) curves for NfL at intensive care unit (ICU) admission, 24, 48, and 72 h after cardiac arrest.

Note: The upper limit of the 95% confidence interval was capped at 1.00, as the AUROC cannot exceed this theoretical maximum despite the statistical method producing a value slightly above it.

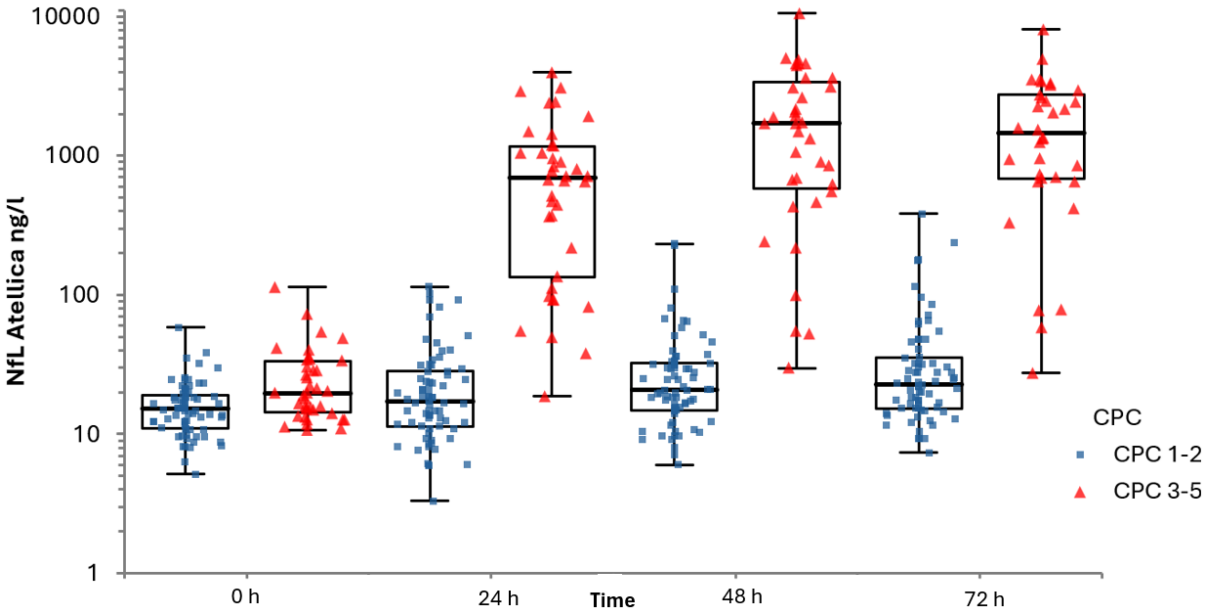

Figure 3. Box plots presenting NfL concentrations measured with Siemens Atellica IM 1600 analyzer at intensive care unit admission (0 h), 24, 48, 72 h after cardiac arrest for patients with good outcome (Cerebral Performance Category (CPC) 1–2 and those with poor outcome (CPC 3–5) with a 10-based logarithmic scale.

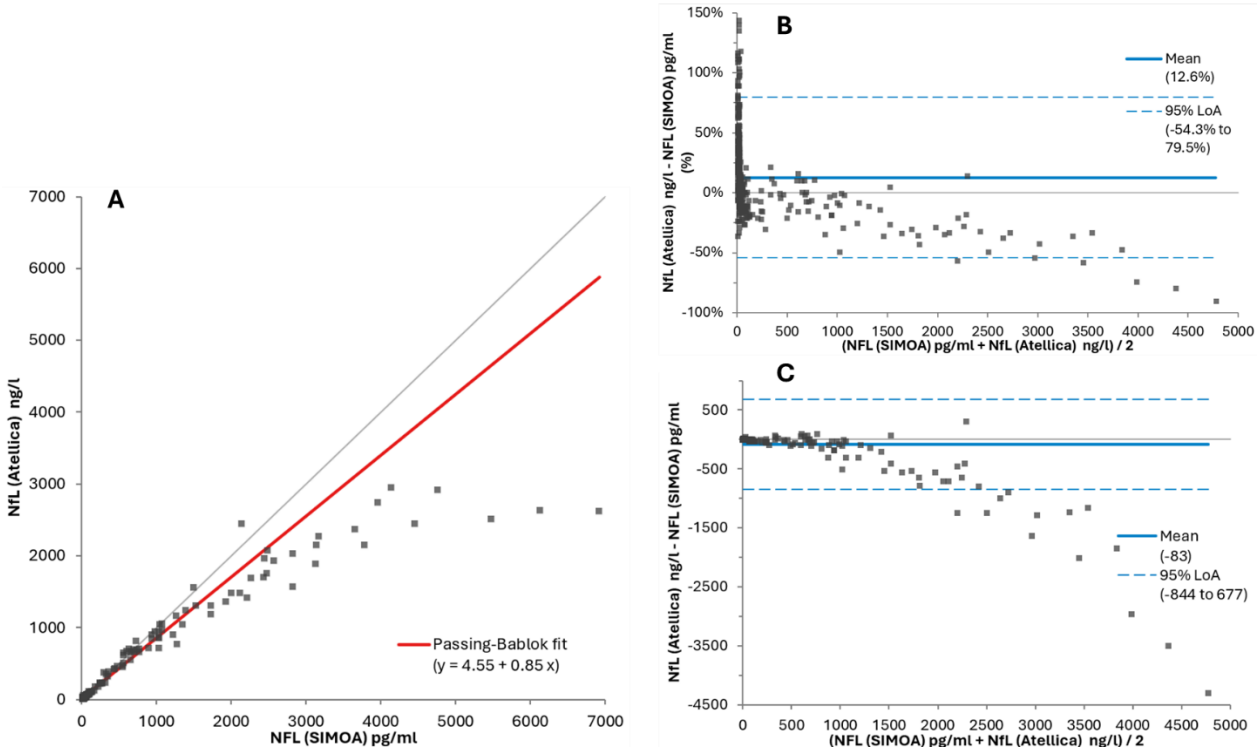

Figure 4. Passing-Bablok regression analysis (A) and Bland-Altman agreements analysis for relative (B) and constant (C) bias of COMACARE trial samples between Atellica IM NfL and Quanterix Simoa NfL for results within extended measuring range e.g. <3000 ng/L. n=411.