Critical Care

CORRESPONDENCE

Open Access

The novel biomarker t^6A accurately identified septic patients at admission but failed to predict outcome

Marcin F. Osuchowski[1*], Barbara Adamik[2], Waldemar Gozdzik[2], Tomasz Skalec[2], Daniel Mascher[3], Heinz Redl[1], Johannes Zipperle[1], Gerhard Fritsch[1,4], Wolfgang Voelckel[4], Martin S. Winkler[5], Onnen Moerer[5], Helmut Schütz[6,7] and Hermann Mascher[3]

Sepsis is a life-threatening condition caused by the body's extreme response to infection [1]. Early diagnosis of sepsis is crucial for improving patient outcomes, yet current diagnostic methods including microbiological cultures are delayed and frequently inconclusive. This has driven the search for novel biomarkers and detection systems capable of recognizing sepsis more rapidly and accurately [2]. Procalcitonin (PCT) is the most widely used biomarker for sepsis detection but its use is limited as circulating PCT concentration is influenced by noninfectious inflammation (e.g. trauma, surgery).

Nucleoside modifications are a hallmark of the post-transcriptional processing of transfer ribonucleic acid (tRNA) that generate multiple structurally modified nucleosides [3]. One of such tRNA-modified nucleosides, N6-threonylcarbamoyladenosine (t^6A), is critical for ensuring efficient protein synthesis in health and disease [4]. We discovered that circulating t^6A possesses a diagnostic potential in sepsis. Consequently, we evaluated the diagnostic accuracy of t^6A in differentiating bacterial sepsis and COVID-19 from two different non-septic patient cohorts: i) patients undergoing elective coronary artery bypass graft (CABG) surgery and ii) severe polytrauma patients. In all cohorts, t^6A diagnostic accuracy was compared to PCT. Additionally, we tested t^6A potential to predict death/survival in patients with sepsis.

This multicenter retrospective observational study analyzed plasma samples from four cohorts (Supplementary Table 1). The study included 81 patients with bacterial sepsis (cohort 1) and 49 patients with severe COVID-19 infection (cohort 2) diagnosed upon ICU/Emergency admission , 87 patients undergoing elective CABG surgery (cohort 3) and 64 severe (Injury Severity Score > 15) polytrauma patients (cohort 4). Sepsis in cohort 1 was defined according to the Sepsis-3 criteria, and all patients received treatment aligned with the Surviving Sepsis Campaign guidelines. In COVID-19 patients, SARS-CoV-2 infection was confirmed by molecular test. The CABG and polytrauma groups were used as non-septic comparators to assess t^6A's diagnostic specificity and accuracy against patients with sepsis and those with COVID-19. We compared blood samples collected at admission for the sepsis and COVID-19 cohorts, to samples collected 24 h post-surgery (cohort 3) and trauma (cohort 4). Plasma t^6A concentrations were measured

*Correspondence:
Marcin F. Osuchowski
marcin.osuchowski@trauma.lbg.ac.at
[1] Ludwig Boltzmann Institute for Traumatology, The Research Center in Cooperation With AUVA, Donaueschingenstraße 13, 1200 Vienna, Austria
[2] Clinical Department of Anesthesiology and Intensive Therapy, Wroclaw Medical University, Wroclaw, Poland
[3] Pharm-Analyt Labor, Baden, Austria
[4] Department of Anesthesiology and Intensive Care Medicine, AUVA Trauma Center Salzburg, Salzburg, Austria
[5] Department of Anesthesiology, University Medical Center Göttingen, Göttingen, Germany
[6] BEBAC, Vienna, Austria
[7] Center for Medical Data Science of the Medical University of Vienna, Vienna, Austria

using tandem mass spectrometry with stable isotope internal standardization. PCT was measured using standard Brahms PCT luminescence immunoassay. Statistical analyses of the receiver operating characteristic (ROC) curves were carried out with a total of 50,000 stratified bootstrap samples to estimate the 95% confidence interval (CI) of the area under the curve (AUC) of the ROC curve and its optimal threshold. The AUCs were compared via De Long's two-sided test [5].

t^6A demonstrated outstanding diagnostic accuracy for sepsis at the ICU/Emergency admission. In separately evaluated exploratory and validation CABG cohorts, t^6A consistently showed greater accuracy (AUC > 90%) compared to PCT (AUC 80–88%). t^6A accuracy against the pooled CABG patients (cohort 3) achieved an AUC of 95% (vs. 88% for PCT, $p < 0.05$) (Fig. 1A). Against the polytrauma group, which represented a more clinically relevant comparator, t^6A again outperformed PCT; its AUC was 97% compared to 88% for PCT ($p < 0.05$) (Fig. 1B). In the COVID-19 cohort, t^6A also displayed excellent diagnostic performance when SARS-CoV-2-infected patients were compared to CABG (AUC 87%, Fig. 1C) and polytrauma (AUC 93%, Fig. 1D) patients. PCT measurement was not differentiating (AUC 50–52%, $p > 0.05$). Notably, the optimal diagnostic threshold for t^6A was nearly identical (3% deviation) against CABG (40.0 ng/mL) and polytrauma (38.8 ng/mL) comparators; it varied by 22% for PCT (2.04 and 1.6 ng/mL).

Despite its strong diagnostic performance, t^6A showed a limited utility for predicting sepsis outcomes. At the ICU/Emergency admission, t^6A concentrations exhibited a considerable overlap between survivors and

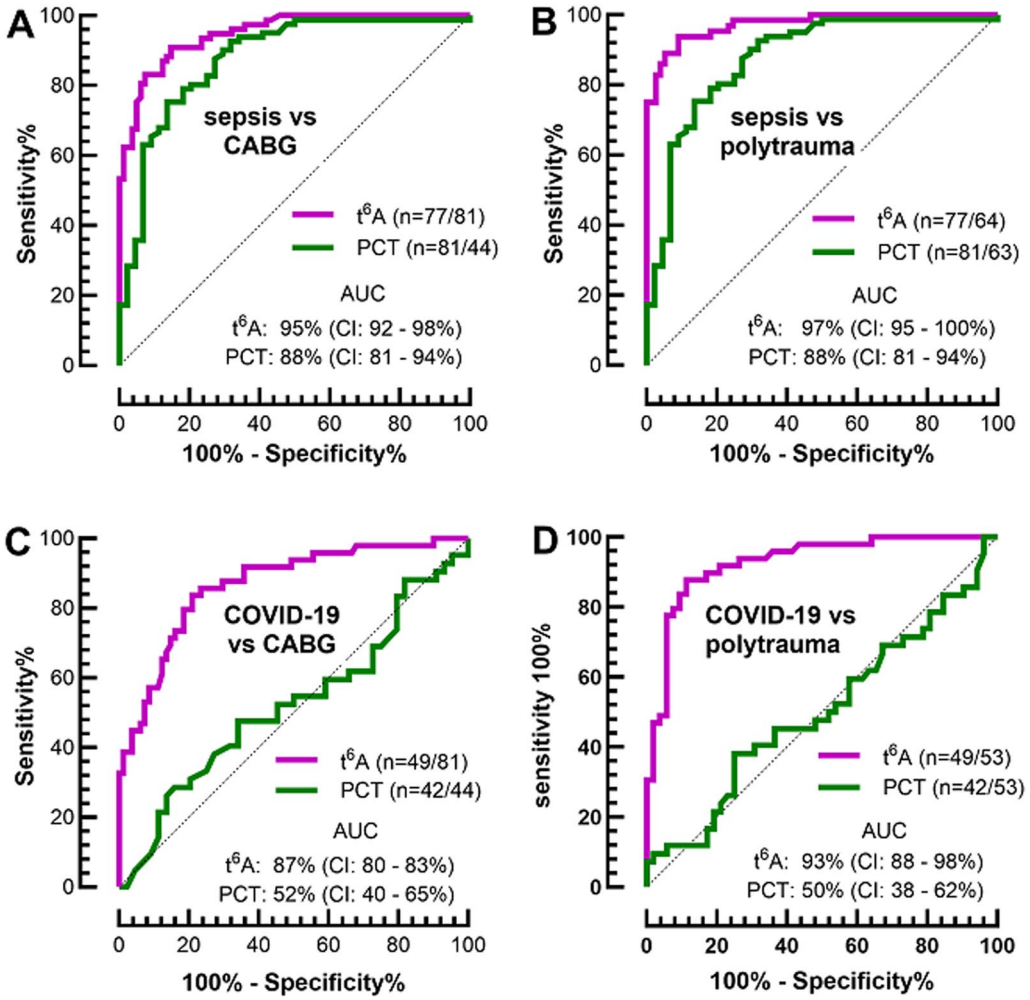

Fig. 1 Diagnostic performance of t^6A and PCT for detecting bacterial sepsis (**A**, **B**) and COVID-19 (**C**, **D**) at the ICU/Emergency admission. n: the first number in parentheses defines the number of sepsis or COVID-19 patients; the second number indicates the number of CABG or polytrauma patients. CI: 95% Confidence Intervals

non-survivors and failed to show any protracted post-admission separation, resulting in an AUC of only 62%. PCT concentrations demonstrated a similar overlap dynamics and performed only modestly better, with an AUC of 72%, suggesting that while t^6A is highly effective for identifying sepsis, it lacks prognostic value. This discrepancy highlights the biomarker's primary utility in diagnosis rather than outcome prediction.

This is the first report regarding the potential utility of t^6A as a diagnostic biomarker in patients with sepsis. We show t^6A as a highly accurate and reliable biomarker for early sepsis diagnosis at the ICU/Emergency admission. t^6A outperformed PCT in differentiating septic patients from non-septic controls including CABG and polytrauma cohorts. Insensitivity of t^6A to sterile inflammation positions it as a promising diagnostic tool for clinical use. However, the lack of a commercially available assay for rapid t^6A measurement limits its utility and this deficiency needs to be addressed. Further studies are required to evaluate t^6A under different ICU conditions, including localized (non-sepsis) infections and extracorporeal therapies, to confirm its broader clinical applicability.

Abbreviations

AUC	Area under the curve
CABG	Coronary artery bypass graft surgery
COVID-19	Coronavirus disease 2019
CI	Confidence interval
ICU	Intensive care unit
ISS	Injury severity score
PCT	Procalcitonin
ROC	Receiver-operating characteristic
t^6A	*N^6*-Threonylcarbamoyladenosine
tRNA	Transfer ribonucleic acid

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s13054-025-05354-2.

Supplementary file 1.

Acknowledgements
Not applicable.

Authors' contribution
MFO and HM conceived of the study and MFO wrote the text. MFO, BA, WG, WV and HM developed the study design. DM, HM, MFO, TS, JZ, OM and GF performed research analysis. HS, BA, JZ and MFO performed data analysis. All authors helped with data interpretation, editing of the manuscript, and read and approved the final text.

Funding
Not applicable.

Data availability
The patient datasets generated and/or analyzed during the current study are available from the corresponding author upon a reasonable request.

Availability of data and materials
The patient datasets generated and/or analyzed during the current study are available from the corresponding author upon a reasonable request.

Declarations

Ethics approval and consent to participate
The study including patients in the cohorts 1 (sepsis) and 3 (CABG) was approved by the Bioethical Committee of Wroclaw Medical University on November 29, 2018 (approval no. 710/2018). The study including patients in the cohort 3 (COVID-19) was approved by the Bioethical Committee of the University Medical Center Göttingen on June 27, 2018 (SeptImmun Study No. 24/4/19Ü). The study including patients in the cohort 4 (polytrauma) was approved by the Bioethical Committee of Allgemeine Unfallversicherungsanstalt on September 15, 2016 (PRIME study No. 09/2016). This research was conducted in accordance with the Helsinki Declaration of 1975, as revised in 2008.

Consent for publication
Not applicable.

Competing interests
Prof. Mascher is a consultant at pharm-analyt Labor, Baden, Austria, and holds patent applications for the use of t^6A to monitor sepsis conditions. The other authors report no competing interest.

Received: 12 February 2025 Accepted: 4 March 2025
Published online: 20 March 2025

References

1. Vincent JL, Jones G, David S, et al. Frequency and mortality of septic shock in Europe and North America: a systematic review and meta-analysis. Crit Care. 2019;23(1):196.
2. Liu C, Song X, Liu J, et al. Consistency between metagenomic next-generation sequencing versus traditional microbiological tests for infective disease: systemic review and meta-analysis. Crit Care. 2025;29(1):55.
3. Boccaletto P, Machnicka MA, Purta E, et al. MODOMICS: a database of RNA modification pathways. 2017 update. Nucleic Acids Res. 2018;46(D1):D303–7.
4. Chujo T, Tomizawa K. Human transfer RNA modopathies: diseases caused by aberrations in transfer RNA modifications. FEBS J. 2021;288(24):7096–122.
5. DeLong ER, DeLong DM, Clarke-Pearson DL. Comparing the areas under two or more correlated receiver operating characteristic curves: a nonparametric approach. Biometrics. 1988;44(3):837–45.

Publisher's Note