Aalto University

OPEN ACCESS

Kujala, Johanna; Wester, Niklas; Lohela, Terhi J.; Kurkela, Mika; Backman, Janne T.;
Mikladal, Björn; Laurila, Tomi; Koskinen, Jari; Lilius, Tuomas O.; Kalso, Eija A.

Introduction of an electrochemical point-of-care assay for quantitative determination of
paracetamol in finger-prick capillary whole blood samples

SHORT COMMUNICATION

Introduction of an electrochemical point-of-care assay for quantitative determination of paracetamol in finger-prick capillary whole blood samples

Johanna Kujala[1] | Niklas Wester[2,3,4] | Terhi J. Lohela[5,6,7] | Mika Kurkela[5,7] | Janne T. Backman[5,7] | Björn Mikladal[8] | Tomi Laurila[2,3] | Jari Koskinen[2] | Tuomas O. Lilius[5,7,9] | Eija A. Kalso[5,6]

[1]Department of Emergency Medicine and Services, Helsinki University Hospital and University of Helsinki, Helsinki, Finland

[2]Department of Chemistry and Materials Science, School of Chemical Engineering, Aalto University, Helsinki, Finland

[3]Department of Electrical Engineering and Automation, School of Electrical Engineering, Aalto University, Helsinki, Finland

[4]Fepod Oy Ltd, c/o Terkko Health Hub, Helsinki, Finland

[5]Department of Clinical Pharmacology, Faculty of Medicine, University of Helsinki and Helsinki University Hospital, Helsinki, Finland

[6]Department of Anaesthesiology, Intensive Care and Pain Medicine, Helsinki University Hospital and University of Helsinki, Helsinki, Finland

[7]Individualized Drug Therapy Research Program, Faculty of Medicine, University of Helsinki, Helsinki, Finland

[8]Canatu Oy, Helsinki, Finland

[9]Department of Emergency Medicine and Services, Finnish Poison Information Center, Helsinki University Hospital and University of Helsinki, Helsinki, Finland

Correspondence
Dr. Tuomas Lilius, Department of Emergency Medicine and Services, Finnish Poison Information Center, PO Box 790, FI-00029 HUS, Helsinki, Finland.
Email: tuomas.lilius@hus.fi

Aims: Measuring venous plasma paracetamol concentrations is time- and resource-consuming. We aimed to validate a novel electrochemical point-of-care (POC) assay for rapid paracetamol concentration determinations.

Methods: Twelve healthy volunteers received 1 g oral paracetamol, and its concentrations were analysed 10 times over 12 h for capillary whole blood (POC), venous plasma (high-performance liquid chromatography tandem mass spectrometry (HPLC-MS/MS)), and dried capillary blood (HPLC-MS/MS).

Results: At concentrations >30 μM, POC showed upward biases of 20% (95% limits of agreement [LOA] -22 to 62) and 7% (95% LOA -23 to 38) compared with venous plasma and capillary blood HPLC-MS/MS, respectively. There were no significant differences between mean concentrations for the paracetamol elimination phase.

Conclusions: Upward biases in POC compared with venous plasma HPLC-MS/MS were likely due to higher paracetamol concentrations in capillary blood than in venous plasma and to faulty individual sensors. The novel POC method is a promising tool for paracetamol concentration analysis.

KEYWORDS
acetaminophen, capillary, diagnostics, intoxication, paracetamol, point-of-care, SWCNT

Johanna Kujala and Niklas Wester are co-first authors.

Tuomas O. Lilius and Eija A. Kalso are co-senior authors.

1 | INTRODUCTION

Paracetamol is a widely used over-the-counter medication and it is one of the commonest causes of deliberate or accidental overdose.[1,2] However, information about paracetamol intake can be unreliable and intoxication may be asymptomatic in the early stages. Hence, the diagnosis of intoxication requires rapid concentration measurement.

The severity of poisoning and clinical decision-making about antidote administration are determined using the Rumack–Matthew nomogram plotting serum paracetamol concentration against time from ingestion.[3] Nomogram timelines begin four hours after ingestion, when absorption is considered complete. In standard analyses, paracetamol concentrations are measured in venous plasma or serum samples. Preparation of blood samples is time-consuming and requires skilled professionals, delaying diagnosis and potentially worsening prognosis.

Rapid determination of paracetamol concentration would confirm the intoxication diagnosis and enable early treatment in hospital or even by paramedics. Further, exclusion of paracetamol intoxication in primary care or emergency medicine would avoid unnecessary referral of patients to secondary care, reducing healthcare costs.

Paracetamol can be readily detected with electrochemical methods[4] and it is well suited to point-of-care (POC) analysis, given the availability of inexpensive and easy-to-use portable potentiostats.[5] Moreover, electrochemical test strips enable analysis of small complex biological samples, such as whole blood, with minimal[6] or no sample treatment.[7] Recently, we described the production of single-walled carbon nanotube/Nafion-based test strips (SWCNT/Nafion) with industrial high throughput methods. The test strip showed linearity with $R^2 0.9956$ in concentrations between 0.82 and 2000 μM, indicating good performance also at toxic concentrations.[6]

We used SWCNT/Nafion test strips[6] to construct concentration-time curves for paracetamol in capillary blood samples. We compared these results with plasma and capillary concentrations measured with high-performance liquid chromatography tandem mass spectrometry (HPLC-MS/MS) in 12 healthy volunteers.

Comparison of capillary POC and venous plasma HPLC-MS/MS concentration-time curves was the first step in validating novel POC sensors, as the Rumack-Matthew nomogram is based on plasma or serum measurements. We then compared capillary POC and capillary HPLC-MS/MS ('gold standard') measurements. Finally, we used HPLC-MS/MS to assess paracetamol concentrations in capillary whole blood and venous plasma samples to determine whether capillary blood could be used for concentration analyses.

What is already known about this subject

- Paracetamol intoxication is a common condition in emergency departments and paracetamol concentration measurement is a common analysis.
- Standard paracetamol concentration measurement is time-consuming and requires skilled laboratory personnel.
- A novel point-of-care electrochemical analysis method can be used to detect paracetamol from capillary blood.

What this study adds

- The paracetamol capillary blood concentrations measured with the novel electrochemical 'point-of-care' method correlate with the 'gold standard' HPLC-MS/MS method in capillary blood.
- The pharmacokinetic properties of paracetamol concentrations in capillary blood have previously not been determined. This study shows that paracetamol concentration in capillary blood is higher than in venous plasma in the absorption phase, and that maximum concentration is reached earlier in capillary blood. After 2 h, capillary blood and venous plasma concentrations of paracetamol correlate closely.

2 | METHODS

2.1 | Study participants

After approval of the Helsinki and Uusimaa Hospital District Ethics Committee and the Finnish Medicines Agency, we recruited 12 healthy volunteers of both sexes in accordance with the WMA Helsinki Declaration's ethical principles. All subjects gave written informed consent.

2.2 | Study setting and sample collection

Volunteers ingested 1 g of paracetamol (Para-Tabs® 1 g, Orion Pharma, Finland) with 200 mL of water after overnight fasting.

They had standardized meals four and eight hours after the paracetamol.

Paired finger-prick and venous blood samples were collected prior to paracetamol administration, followed by sampling at timepoints 0.5; 1.0; 1.5; 2.0; 3.0; 4.0; 6.0; 8.0 and 12 h.

Venous blood samples were centrifuged to plasma and frozen at −80°C for later HPLC-MS/MS analysis. Capillary finger-prick samples were collected for fast electrochemical POC analysis, and duplicate capillary blood samples with the VAMS Mitra® Cartridge Blood Sampling Device (Neoteryx, CA, USA) for later HPLC-MS/MS analysis. Detailed description of analytical procedures can be found in the Supporting Information.

2.3 | POC analysis

POC sensor strips were calibrated for paracetamol in 10 mM phosphate-buffered saline (PBS) solution (pH 7.4) and in plasma.

All electrochemical measurements were carried out with differential pulse voltammetry (DPV), using PalmSens4 portable potentiostat (PalmSens BV, the Netherlands) connected to a personal computer.

POC analysis of capillary whole blood was performed immediately after sample collection. Prior to analysis, 20 μL of capillary blood was diluted with 20 μL PBS in order to obtain a sufficient amount of sample. The sample was transferred onto the test strip and the measurement script was run. The script included an automated incubation time of 2.5 min followed by a 30 s DPV scan. The full protocol, including background scan, sample dilution, incubation time

and sample analysis, could be performed in less than 5 min. Each determination was carried out with a new disposable sensor.

2.4 | Pharmacokinetic and statistical analyses

The pharmacokinetic parameters were calculated using Phoenix WinNonlin v. 6.4 (Certara, USA).

The pharmacokinetic results are expressed as geometric means with geometric coefficients of variation (CV) or geometric mean ratios with 90% confidence intervals (CIs), as recommended for pharmacological comparisons.[8]

Statistical analysis and method comparisons with Bland–Altman plot and Passing-Bablok were performed with user-written *blandaltman* and *agree* commands in Stata version 15.1/MP2 (StataCorp, College Station, TX, USA).[9,10]

3 | RESULTS

3.1 | Participants

Twelve healthy subjects aged 18–28 years, nine male, three female, European origin, were recruited; BMIs were 21–28 kg/m^2 (median 23); all completed. During analysis, we suspected two subjects of violating the protocol: initial plasma and capillary samples of one contained paracetamol in HPLC-MS/MS, indicating prior use; blood samples from another appeared turbid, suggesting lack of fasting. These unconfirmed protocol violations did not result in exclusion, because

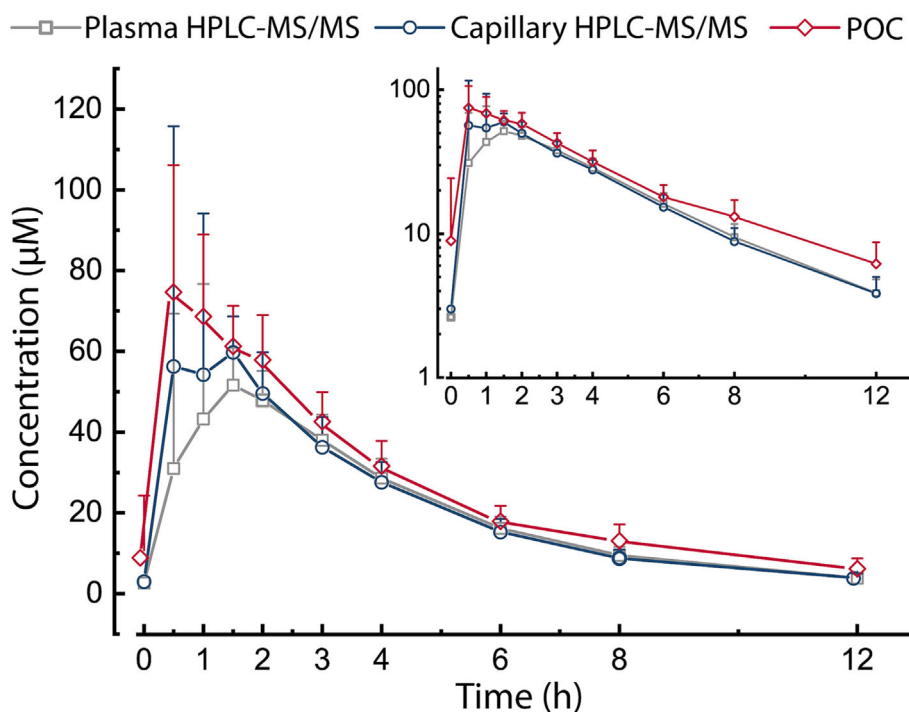**FIGURE 1** Mean concentration-time curves for paracetamol in venous plasma (grey squares) and in capillary whole blood (blue circles), measured with high-performance liquid chromatography mass-spectrometry (HPLC-MS/MS), and in capillary whole blood, measured with electrochemical point-of-care (POC) sensors (red diamonds). Geometric means with 90% CIs are presented. Insets show the same data on a semilogarithmic scale.

the main focus was on comparing two methods of analysis rather than paracetamol kinetics. We also show the main analysis excluding these individuals in Supporting Information.

3.2 | Comparison of paracetamol concentrations in capillary POC, capillary HPLC-MS/MS and venous plasma HPLC-MS/MS

Paracetamol concentrations were highest in capillary POC samples and higher in capillary samples (POC and HPLC-MS/MS) than in venous plasma HPLC-MS/MS, particularly during the first two hours (Figure 1). Thereafter, the capillary blood and plasma concentrations analysed with HPLC-MS/MS aligned fully while POC values remained somewhat higher.

Individual concentration-time curves are presented in Figure S1. Absorption was delayed in Subject 10 who may not have fasted as required; Subject 3 had 3 μM paracetamol in plasma HPLC-MS/MS prior to paracetamol administration. Exclusion of these two participants had little effect on the overall results. Only the range of t_{max} narrowed, likely due to omitting Subject 10 (Tables S1 and S2 and Tables 1 and 2, respectively).

The C_{max} and t_{max} of capillary POC and HPLC-MS/MS were similar, but the AUC_{0-last} was 17% higher in POC than with HPLC-MS/MS (Table 1, Figure 1). Similarly, the AUC_{0-last} was 26% higher in POC and 8% higher in capillary HPLC-MS/MS than in venous plasma HPLC-MS/MS (Table 2). Mean C_{max} values were 55% and 50% higher in capillary POC and HPLC-MS/MS than in venous plasma, respectively; t_{max} was reached earlier (0.5 h) in capillary blood than in venous plasma (1 h) (Table 2).

Comparison of POC calibration in plasma and PBS with capillary HPLC-MS/MS is presented in Table S3.

3.3 | Comparison of methods

We conducted Bland–Altman and Passing-Bablok analyses to compare the capillary POC method with HPLC-MS/MS in venous plasma and capillary samples (Figures S2–S4).

Capillary POC showed an upward bias of 23% (95% limits of agreement [LOA] −79 to 124) compared with capillary HPLC-MS/MS (Figure S2A). The bias was reduced to 7% when analysis was limited to plasma concentrations ≥30 μM (Figure S2B). The Passing-Bablok slope was 1.04 (95% CI 0.98–1.10) (Figure S2C) with four datapoints showing greater than 20 μM differences between the two methods (Figure S2D).

Capillary POC showed a 30% upward bias (95% LOA −73 to 133) compared with venous plasma HPLC-MS/MS (Figure S3A). At plasma concentrations ≥30 μM, the upward bias and LOA decreased to 20% (95% LOA −22 to 62) (Figure S3B). Similarly, the slope of the Passing–Bablok regression line including all datapoints was 1.23 (95% CI 1.16–1.29) (Figure S3C). Scattering of POC concentrations from venous measurements with HPLC-MS/MS is shown in the Passing-Bablok residual plot (Figure S3D).

Finally, capillary HPLC-MS/MS showed an 8% upward bias (95% LOA −44 to 61) compared with venous HPLC-MS/MS (Figure S4A), which increased to 12% (95% LOA −30 to 55) at plasma concentrations ≥30 μM (Figure S4B). The Passing–Bablok slope was 1.09 with a y-axis intercept at −0.8 (Figure S4C) and higher scattering in higher concentrations in the residual plot (Figure S4D).

4 | DISCUSSION

In the process of developing POC test strips, this is the first *in vivo* study, a mandatory step in the method validation. Here, we measured paracetamol in healthy volunteers in the presence of its metabolites, improving on the previous *in vitro* study using a similar method,[6] providing concentration results in 5 min in a clinical study setting.

POC correlated well with capillary HPLC-MS/MS with an upward bias of 7% at plasma concentrations >30 μM. The upward bias of POC in the low concentration range <30 μM is not clinically relevant. When the POC method is compared with venous plasma HPLC-MS/MS, higher capillary concentrations in the absorption phase led to overestimation of paracetamol concentrations. Individual outliers, particularly in the low concentration range (<30 μM), also caused overestimation of paracetamol concentrations. This upward bias is

TABLE 1 Pharmacokinetic parameters of paracetamol in capillary whole blood measured with high-performance liquid chromatography mass spectrometry (HPLC/MS–MS) and with an electrochemical point-of-care (POC) sensor in 12 healthy participants.

Parameter	Capillary HPLC-MS/MS (control)	Capillary POC
C_{max} (μmol/L)	98.67 (31)	101.73 (21)
Ratio-to-control	1	1.03 (90% CI 0.94–1.13) $P = 0.5513$
t_{max} (h)	0.5 (range 0.5–2)	0.5 (range 0.5–2) $P = 1.0$
AUC_{0-last} (h μmol/L)	283.41 (30)	330.60 (29)
Ratio-to-control	1	1.17 (90% CI 1.10–1.23) $P = 0.0004$

Note: Data were presented as geometric means with geometric coefficients of variation (as percentage), except t_{max} for which median and range are shown; t_{max} was compared using Wilcoxon matched-pairs signed-ranks tests between capillary POC and capillary HPLC-MS/MS (control). Other pharmacokinetic variables were compared with paired *t*-tests on log-transformed data. Geometric mean ratios between capillary POC and capillary HPLC-MS/MS were shown with 90% CI. 90% CIs and *P*-values were shown for differences between capillary POC and capillary HPLC-MS/MS sample (control).
Abbreviations: AUC, area under concentration-time curve; CI, confidence interval; C_{max}, maximum concentration; t_{max}, time to reach maximum concentration.

TABLE 2 Pharmacokinetic parameters of paracetamol in venous plasma (control) and in capillary whole blood measured with high-performance liquid chromatography mass spectrometry (HPLC/MS-MS) and in capillary whole blood measured with an electrochemical point-of-care (POC) sensor in 12 healthy participants.

Parameter	Plasma HPLC-MS/MS (control)	Capillary HPLC-MS/MS	Capillary POC
C_{max} (μmol/L)	65.61 (27)	98.67 (31)	101.73 (21)
Ratio-to-control	1	1.50 (90% CI 1.36–1.66) $P < 0.0001$	1.55 (90% CI 1.42–1.69) $P < 0.0001$
t_{max} (h)	1 (range 0.5–2)	0.5 (range 0.5–2) $p = 0.0265$	0.5 (range 0.5–2) $p = 0.0265$
AUC_{0-last} (h μmol/L)	262.21 (26)	283.41 (30)	330.60 (29)
Ratio-to-control	1	1.08 (90% CI 1.04–1.11) $P = 0.0006$	1.26 (90% CI 1.19–1.34) $P < 0.0001$

Note: Data were presented as geometric means with geometric coefficients of variation (as percentage), except t_{max} for which median and range are shown; t_{max} was compared using Wilcoxon matched-pairs signed-ranks tests between capillary HPLC-MS/MS or capillary POC and venous plasma HPLC-MS/MS (control). Other pharmacokinetic variables were compared with paired *t*-tests on log-transformed data. Geometric mean ratios between capillary HPLC-MS/MS or capillary POC and venous plasma HPLC-MS/MS (control) were shown with 90% CI. 90% CIs and *P*-values are shown for differences between the respective test and venous plasma HPLC-MS/MS sample (control).
Abbreviations: AUC, area under concentration-time curve; CI, confidence interval; C_{max}, maximum concentration; t_{max}, time to reach maximum concentration.

likely caused by electrochemically active endogenous interferents or faulty electrodes (outliers in Figure S1 showed significantly larger upward biases than other POC results, suggesting faults). Paracetamol's metabolites are unlikely to explain the bias during absorption as they would not yet have been formed.

Capillary kinetics of paracetamol must be studied before introducing finger-prick sampling into clinical use. The capillary blood and venous plasma concentrations agreed almost completely during the elimination phase (Figures 1 and S1), but the capillary concentrations were higher in the absorption phase and t_{max} was reached earlier (Table 2). This may not have clinical relevance, as in intoxication suspicions the paracetamol concentrations must be assessed after four hours from ingestion.[1,2]

Published studies are somewhat contradictory about paracetamol's pharmacokinetics in capillary blood. Our results agree with Mohammed et al, who compared paracetamol concentrations in capillary whole blood and venous samples in 12 adult males after a single 1 g dose of oral paracetamol.[11] Contrary to the finding by us and Mohammed et al., Rittau et al. found the AUC to be significantly higher in venous plasma than in capillary plasma or saliva.[12] In that study, capillary whole blood samples were not used, differing from the studies by Mohammed et al. and us. In addition, Rittau et al.'s study was performed in a steady state dosing protocol and it did not require fasting. Food intake is known to affect paracetamol's pharmacokinetics.[13]

Our findings and those of Mohammed et al. suggest rapid distribution of paracetamol to capillary circulation during absorption, followed by levelling of the difference between capillary and plasma concentrations. This may be a result of capillary blood comprising venous and arterial blood mixed with intra- and intercellular fluid.[14]

A large proportion of arterial blood in this mixture facilitates transportation of the drug to the capillary during the distribution phase.

POC electrode sensitivity and selectivity may vary due to defects or non-uniformity in permselective coatings, changing permeability of paracetamol and its interferents. Electrochemically active interferents, such as uric or ascorbic acids, can cause upward bias, while plasma proteins may passivate electrodes, causing downward bias. The latter was not observed in the clinically used concentration range (Figure S3B), indicating absence of large defects that allow permeation of proteins. Upward is preferred to downward bias as false negatives are highly undesirable. Upward biases are most likely caused by small defects in the permselective layer of individual sensors, allowing increased passage of interferents.[15]

As the experimental setup did not enable repeated analysis in case of faulty sensors, we performed only one measurement per timepoint. The test strips were still prototypes and quality control for mass production was not yet available at the time of the study. Further research is ongoing to better understand sources of sensor variability, improve the manufacturing process, and establish quality control methods for ensuring sensor reliability. Due to large errors in measurement, we conducted crude Bland–Altman analyses without adjusting for clustering at the level of participants, which influences the limits of agreement.

Combined with our previous *in vitro* studies, this study suggests that the novel electrochemical POC method is promising as a screening test for paracetamol. Simple sample processing and delivery of results in minutes suggests that this could be a welcome method in primary health care and emergency units. Further research is needed to assess quantitative performance of the sensor at toxic concentrations.

Indeed, we are currently pursuing a study using samples from intoxicated individuals to further develop this method for improving patient safety and controlling healthcare costs.

AUTHOR CONTRIBUTIONS

Johanna Kujala and Niklas Wester: conception and design of the study, acquisition, analysis and interpretation of data, and drafting and approval of the manuscript. Terhi J. Lohela: analysis and interpretation of data, drafting and approval of the manuscript. Mika Kurkela: acquisition of data, drafting and approval the manuscript. Janne T. Backman, Tomi Laurila, Jari Koskinen, Tuomas O. Lilius, Eija A. Kalso: conception and design of the study, supervising the study, analysis and interpretation of data, drafting and approval of the manuscript. Björn Mikladal: acquisition of data, approval of manuscript.

CONFLICT OF INTEREST STATEMENT

EK has received fees for lectures or advisory board membership or related to the current study from Orion Pharma, GSK and Pfizer. NW is one of the founders, owners, and board member of FEPOD Oy Ltd that is currently commercializing the POC sensor.

DATA AVAILABILITY STATEMENT

The data are available from the corresponding author.

ORCID

Johanna Kujala https://orcid.org/0000-0001-8779-1914
Niklas Wester https://orcid.org/0000-0002-7937-9011
Terhi J. Lohela https://orcid.org/0000-0002-6596-1780
Mika Kurkela https://orcid.org/0000-0002-4153-4143
Janne T. Backman https://orcid.org/0000-0002-9577-2788
Björn Mikladal https://orcid.org/0000-0003-4242-1180
Tomi Laurila https://orcid.org/0000-0002-1252-8764
Jari Koskinen https://orcid.org/0000-0002-8907-6743
Tuomas O. Lilius https://orcid.org/0000-0002-8361-3683
Eija A. Kalso https://orcid.org/0000-0002-4899-605X

REFERENCES

1. Seifert SA, Kirschner RI, Martin TG, Schrader RM, Karowski K, Anaradian PC. Acetaminophen concentrations prior to 4 hours of ingestion: impact on diagnostic decision-making and treatment. *Clin Toxicol (15563650)*. 2015;53(7):2-623. doi:10.3109/15563650.2015.1059942
2. Froberg BA, King KJ, Kurera TD, et al. Negative predictive value of acetaminophen concentrations within four hours of ingestion. *Acad Emerg Med*. 2013;20(10):1072-1075. doi:10.1111/acem.12222
3. Rumack BH, Matthew H. Acetaminophen poisoning and toxicity. *Pediatrics*. 1975;55(6):871-876. doi:10.1542/peds.55.6.871
4. Miner DJ, Rice JR, Riggin RM, Kissinger PT. Voltammetry of acetaminophen and its metabolites. *Anal Chem*. 1981;53(14):2258-2263. doi:10.1021/ac00237a029
5. Ning Q, Feng S, Cheng Y, Li T, Cui D, Wang K. Point-of-care biochemical assays using electrochemical technologies: approaches, applications, and opportunities. *Microchim Acta*. 2022;189(8):310. doi:10.1007/s00604-022-05425-z
6. Wester N, Mikladal BF, Varjos I, et al. Disposable Nafion-coated single-walled carbon nanotube test strip for electrochemical quantitative determination of acetaminophen in a finger-prick whole blood sample. *Anal Chem*. 2020;92(19):13017-13024. doi:10.1021/acs.analchem.0c01857
7. Verrinder E, Wester N, Leppänen E, et al. Electrochemical detection of morphine in untreated human capillary whole blood. *ACS Omega*. 2021;6(17):11563-11569. doi:10.1021/acsomega.1c00773
8. Clinical Pharmacology & Therapeutics Editorial Team. Statistical guide for clinical pharmacology & therapeutics. *Clin Pharmacol Therapeut*. 2010;88(2):150-152. doi:10.1038/clpt.2010.113
9. metodo.uab.cat/stata/agree.ado. Accessed September 15, 2022. http://metodo.uab.cat/stata/agree.ado
10. Chatfield M. BLANDALTMAN: Stata module to create Bland-Altman plots featuring differences or %differences or ratios, with options to add a variety of lines and intervals. Statistical Software Components, 2022. Accessed November 14, 2022. https://ideas.repec.org/c/boc/bocode/s459040.html
11. Mohammed BS, Cameron GA, Cameron L, Hawksworth GH, Helms PJ, McLay JS. Can finger-prick sampling replace venous sampling to determine the pharmacokinetic profile of oral paracetamol? *Br J Clin Pharmacol*. 2010;70(1):52-56. doi:10.1111/j.1365-2125.2010.03668.x
12. Rittau AM, McLachlan AJ. Investigating paracetamol pharmacokinetics using venous and capillary blood and saliva sampling. *J Pharm Pharmacol*. 2012;64(5):705-711. doi:10.1111/j.2042-7158.2012.01459.x
13. Moore RA, Derry S, Wiffen PJ, Straube S. Effects of food on pharmacokinetics of immediate release oral formulations of aspirin, dipyrone, paracetamol and NSAIDs – a systematic review. *Br J Clin Pharmacol*. 2015;80(3):381-388. doi:10.1111/bcp.12628
14. Merton G, Jones K, Lee M, Johnston A, Holt DW. Accuracy of cyclosporin measurements made in capillary blood samples obtained by skin puncture. *Ther Drug Monit*. 2000;22(5):594-598. doi:10.1097/00007691-200010000-00015
15. Leppänen E, Peltonen A, Seitsonen J, Koskinen J, Laurila T. Effect of thickness and additional elements on the filtering properties of a thin Nafion layer. *J Electroanal Chem*. 2019;843:12-21. doi:10.1016/j.jelechem.2019.05.002

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.