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Evaluation of the Analytical Performance Creatinine_3 Assay on the Atellica CH and CI Analyzers

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Background

Creatinine, a nitrogenous waste product found in blood, originates from normal muscle turnover and digestion of food protein. In the body, creatinine is excreted by the kidneys. The circulating concentration of creatinine is influenced by many factors such as age, sex, and weight. Increased concentrations of serum creatinine may indicate that kidneys are unable to filter creatinine due to acute or chronic kidney disease. The estimated glomerular filtration rate (eGFR) is a measure of how well kidneys function to remove waste products. The eGFR is used to determine the severity (stage) of chronic kidney disease and is calculated using serum creatinine concentrations, age, and sex in most equations. A value <60 mL/min/1.73 m^2 may indicate that kidneys are not functioning normally, and a value of <15 mL/min/1.73 m^2 is indicative of kidney failure.[1-4]

The Atellica CH Creatinine_3 (Crea3) assay measures creatinine in human serum, plasma (lithium heparin, dipotassium EDTA, sodium heparin), and urine. The assay aids diagnosis and treatment of renal diseases, and monitoring of renal dialysis in kidney failure patients.

Objective

To evaluate the analytical performance of the Atellica CH Creatinine_3 (Crea3)[5] assay on the Atellica CH and CI[6] Analyzers with respect to precision, method comparison, detection capability, linearity, and specimen equivalency.

Materials and Methods

Principles of the Procedure

Creatinine reacts with picrate in an alkaline medium to produce a red chromophore creatinine picrate complex. The rate of complex formation is measured at 505/571 nm and is proportional to creatinine concentration. The Atellica CH Crea3 assay (modified Jaffe method) minimizes bilirubin interference, and intercept correction. Because non-specific serum/plasma protein interactions with this reagent have been found to produce a positive bias of approximately 0.3 mg/dL (26.5 µmol/L), serum/plasma measurements are automatically corrected by subtracting 0.3 mg/dL (26.5 µmol/L) from each result.[5]

Precision (CLSI EP05-A3)

Repeatability and Within-Laboratory Precision

- Serum: Native and diluted human serum, native human serum spiked with Creatinine – anhydrous, ≥98% (Sigma Aldrich), QC Liquid Assayed Multiqual (Bio-Rad).
- Urine: Native human urine, QC Liquichek Urine Chemistry Control (Bio-Rad).
- Samples were tested in duplicate/run, two runs/day, 20 days (n=80/sample), three reagent lots, one analyzer per system.
- Data were analyzed using a nested, two factor (days and runs nested within days) ANOVA model.

Reproducibility

- Five replicates/sample, one run/day, 5 days, three lots, three analyzers (n=225 measurements/sample).
- Replicates were analyzed cross-nested with runs nested in days, days nested in lots, and lots nested in systems. The following components of precision were calculated: repeatability, between-day, between-lot, between-instrument, and reproducibility (total).

Method Comparison (CLSI EP09c-ED3)

- Individual serum and urine samples were tested on the Atellica CH system with the Atellica CH Crea3 (test) assay and the Atellica CH Crea_2[7] (predicate) assay. In addition, the Atellica CH Crea3 assay analytical performance was compared on the Atellica CH and CI Analyzers. Where necessary, samples were spiked with concentrated Creatinine – anhydrous, ≥98% (Sigma Aldrich) to cover the assay measuring interval. A single replicate was processed for each sample with three reagent lots. Slope, Y-intercept, and correlation results were calculated by Deming regression.

Detection Capability (CLSI EP17-A2)

LoB: Highest measurement result that is likely to be observed on a blank sample.

- Four blank samples (Atellica CH Diluent), five replicates/sample, one run/day, 3 test days, one analyzer, three reagent lots (total n=60 measurements/reagent lot).
- LoB was calculated non-parametrically at the 95th percentile. For each lot, the rank position at the 95th percentile was determined as: Rank position = 0.5 + (n x 0.95), where n is the total number of replicates. The largest LoB calculated among the lots was the assay's LoB.

LoD: Lowest concentration detectable with 95% probability.

- Five low samples of native human serum and urine (diluted with Atellica CH Diluent) were processed on three reagent lots for 3 days, five replicates/sample, (total n=75 measurements/lot).
- LoD is taken as the maximum value of all lots tested. LoD was calculated parametrically using the pooled within-lab standard deviation (SD_{WL}) for all samples from a given reagent lot using the equation: **$LoD = LoB + c_p SD_{WL}$**, where c_p is a multiplier used to give 95th percentile of a normal distribution with probability of Type II error 0.05 (β= 0.05).

LoQ: LoQ defined in this study is the lowest amount of a measurand in a sample that can be quantitatively determined with stated total allowable error.

- Five low samples of native human serum or urine (diluted with Atellica CH Diluent) were processed; three reagent lots, 3 days, five replicates/day, one instrument (total = 75 measurements/reagent lot).
- Data analysis involved parametric method LoQ. Samples were prepared at approximately 0.15 mg/dL (serum) and 1.75 mg/dL (urine). For each reagent lot, the mean, SD, and Total Error of each low-level sample was determined. The target concentration value was determined via the predicate assay Atellica CH Creatinine_2 (Crea_2). Total Error was determined via the Westgard Model equation: $Westgard\ Model: TE = |Bias| + 2s$. The concentration of the lowest level sample that meets the Total Error goal, with all higher samples also meeting the Total Error goal, was taken as the LoQ for the lot. The largest LoQ calculated among the lots was the assay's LoQ.

Linearity (CLSI EP06-ED2)

- Samples: A dilution series with nine levels was prepared by mixing high (spiked with Creatinine – anhydrous, ≥98% (Sigma Aldrich)) and low (diluted with Atellica CH Diluent) sample pools.
- Five replicates/level, one test day/lot, three reagent lots (n=5/level/lot).
- Expected values were calculated from the measurand concentrations of the low and high samples. Bias was calculated for each sample as the difference between the mean observed value and the value predicted by the linear regression model (weighted least squares regression of observed mean (y) versus expected value (x)).

Specimen equivalency (CLSI EP09C-ED3)

- Fifty matched serum and plasma (sodium heparin, lithium Heparin, and dipotassium EDTA) samples were processed with n=1 replicate and one reagent lot. Samples were spiked with creatinine (anhydrous, ≥98%) to span the assay measuring interval. Slope, Y-intercept, and correlation (r) results were generated using a Deming regression.

Results

The following results are representative of the performance of the assay.

Precision

Table 1. Precision for the Atellica CH Crea3 Assay on the Atellica CH Analyzer.

Specimen Type	n[a] = 80 Mean mg/dL (µmol/L)	Repeatability		Within-laboratory	
		SD[b] mg/dL (µmol/L)	CV[c] (%)	SD mg/dL (µmol/L)	CV (%)
Serum 1	0.38 (34)	0.006 (0.5)	1.6	0.012 (1.1)	3.2
Serum 2	0.73 (65)	0.023 (2.0)	3.2	0.029 (2.6)	4.0
Serum 3	0.73 (65)	0.006 (0.5)	0.8	0.019 (1.7)	2.6
Serum 4	1.18 (104)	0.007 (0.6)	0.6	0.019 (1.7)	1.6
Serum QC 1	1.85 (164)	0.007 (0.6)	0.4	0.024 (2.1)	1.3
Serum QC 2	6.21 (549)	0.011 (1.0)	0.2	0.067 (5.9)	1.1
Serum 5	17.39 (1537)	0.035 (3.1)	0.2	0.189 (16.7)	1.1
Serum 6	28.54 (2523)	0.056 (5.0)	0.2	0.317 (28.0)	1.1
Urine 1	56.74 (5016)	0.102 (9.0)	0.2	0.746 (65.9)	1.3
Urine QC 1	135.80 (12005)	0.206 (18.2)	0.2	1.601 (141.5)	1.2
Urine 2	195.79 (17308)	0.253 (22.4)	0.1	2.376 (210.0)	1.2

[a]n: number of measurements; [b]SD: standard deviation; [c]CV: coefficient of variation.

Across the sample interval, serum repeatability CV was ≤3.2% and within laboratory CV was ≤4.0%; urine repeatability was ≤0.2% and within laboratory CV was ≤1.3%. Reproducibility CVs for serum were 0.9–5.0% at 0.40-28.76 mg/dL (35–2542 µmol/L) and for urine 1.4–1.6% at 57.23–199.45 mg/dL (5059–17,631 µmol/L) (data not shown).

Table 2. Precision for the Atellica CH Crea3 Assay on the Atellica CI Analyzer.

Specimen Type	n[a] = 80 Mean mg/dL (µmol/L)	Repeatability		Within-laboratory	
		SD[b] mg/dL (µmol/L)	CV[c] (%)	SD mg/dL (µmol/L)	CV (%)
Serum 1	0.40 (35)	0.010 (0.9)	2.5	0.012 (1.1)	3.0
Serum 2	0.75 (66)	0.030 (2.7)	4.0	0.030 (2.7)	4.0
Serum 3	0.72 (64)	0.010 (0.9)	1.4	0.023 (2.0)	3.2
Serum 4	1.21 (107)	0.010 (0.9)	0.8	0.017 (1.5)	1.4
Serum QC 1	1.89 (167)	0.016 (1.4)	0.8	0.026 (2.3)	1.4
Serum QC 2	6.29 (556)	0.038 (3.4)	0.6	0.055 (4.9)	0.9
Serum 5	17.57 (1553)	0.064 (5.7)	0.4	0.122 (10.8)	0.7
Serum 6	28.75 (2542)	0.137 (12.1)	0.5	0.229 (20.2)	0.8
Urine 1	56.62 (5005)	0.246 (21.7)	0.4	0.610 (53.9)	1.1
Urine QC 1	135.92 (12,015)	0.416 (36.8)	0.3	0.963 (85.1)	0.7
Urine 2	195.39 (17,272)	0.729 (64.4)	0.4	1.896 (167.6)	1.0

[a]n: number of measurements; [b]SD: standard deviation; [c]CV: coefficient of variation.

Across the sample interval, serum repeatability CV was ≤4.0% and within laboratory CV was ≤4.0%; urine repeatability was ≤0.4% and within laboratory CV was ≤1.1%. Reproducibility CVs for serum were 0.6–12.2% at 0.41–29.00 mg/dL (36–2564 µmol/L) and for urine 1.2–1.4% at 56.68–196.76 mg/dL (5011–17394 µmol/L) (data not shown).

Method Comparison

Table 3. Performance of the Atellica CH Crea3 assay on the Atellica CH Analyzer compared with the performance of the comparison assay, Crea_2 (x), on the Atellica CH Analyzer and the performance of the Atellica CH Crea3 assay on the Atellica CI Analyzer compared with the performance of the comparison assay, Atellica CH Crea3 (x), on the Atellica CH Analyzer.

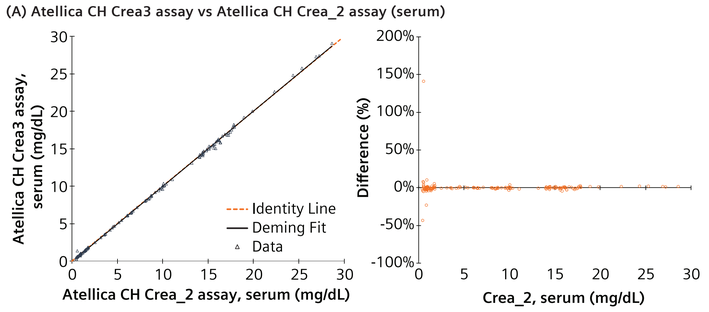
Specimen Type	Comparison Assay (x)	Candidate Assay and Analyzer (y)	n	r	Regression Equation	Sample Range
Serum	Atellica CH Crea_2	Atellica CH Crea3 on Atellica CH	151	1.000	y = 1.00x – 0.04 mg/dL (y = 1.00x – 4 µmol/L)	0.44–28.64 mg/dL (39–2532 µmol/L)
Urine	Atellica CH Crea_2	Atellica CH Crea3 on Atellica CH	113	1.000	y = 1.00x + 0.14 mg/dL (y = 1.00x + 12 µmol/L)	12.60–237.06 mg/dL (1114–20,956 µmol/L)
Serum	Atellica CH Crea3	Atellica CH Crea3 on Atellica CI	151	1.000	y = 1.00x + 0.02 mg/dL (y = 1.00x + 2 µmol/L)	0.46–28.89 mg/dL (41–2,554 µmol/L)
Urine	Atellica CH Crea3	Atellica CH Crea3 on Atellica CI	113	1.000	y = 0.97x + 0.22 mg/dL (y = 0.97x + 19 µmol/L)	12.50–235.17 mg/dL (1105–20,789 µmol/L)

n: number of samples; r: correlation coefficient.

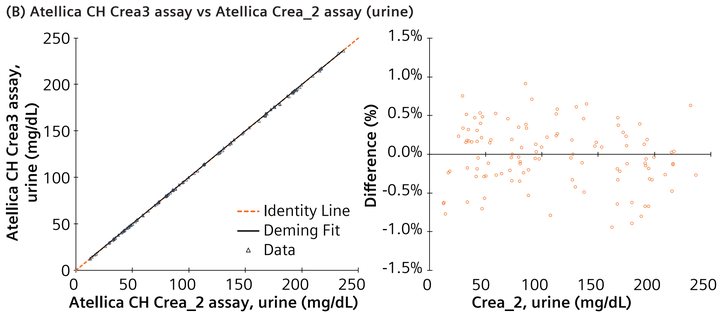
The design requirements for method comparison were met for Crea3 assay on the Atellica CH and CI Analyzers. When analyzed by regression, the Crea3 assay on the Atellica CH Analyzer vs the Crea_2 assay on the Atellica CH Analyzer and the Crea3 assay on the Atellica CH Analyzer vs the Crea3 assay on the Atellica CI Analyzer, recovered samples spanning the measuring interval, with a slope of 1.00 ± 0.05 and a correlation coefficient ≥ 0.950 (r).

Deming fit and percent difference plots for the comparisons in Table 3 are shown in Figure 1.

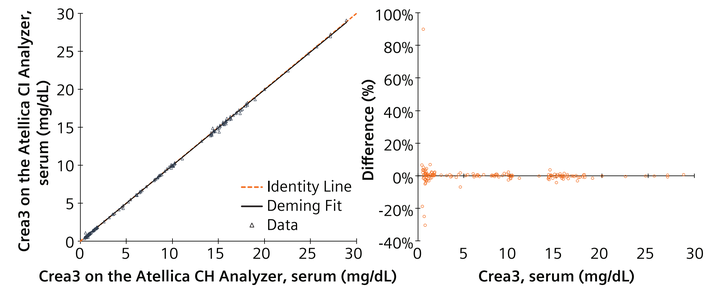
(A) Atellica CH Crea3 assay vs Atellica CH Crea_2 assay (serum)



(B) Atellica CH Crea3 assay vs Atellica Crea_2 assay (urine)



(C) Atellica CH Crea3 assay on Atellica CH Analyzer vs Atellica CI Analyzer (serum)



(D) Atellica CH Crea3 assay on Atellica CH Analyzer vs Atellica CI Analyzer (urine)

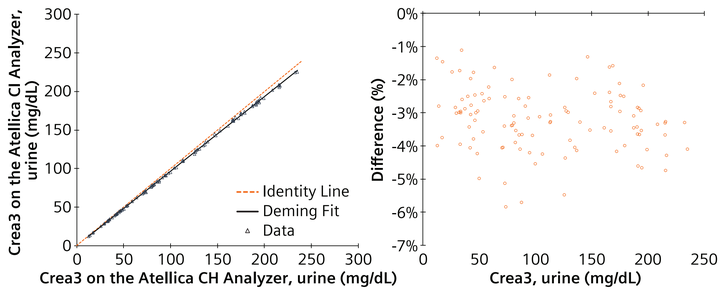


Figure 1. Deming regression and difference plots for the Atellica CH Crea3 assay vs Atellica CH Crea_2 assay for serum (A) and urine (B) samples, and for the Atellica CH Crea3 assay on the Atellica CH vs Atellica CI Analyzers for serum (C) and urine (D) samples.

Detection Capability

Table 4. LoB, LoD, and LoQ for the Atellica CH Crea3 assay on both the Atellica CH and CI Analyzers.

Specimen Type	LoB and LoD Total Measurements per Reagent Lot	LoB Reported	LoD Reported	LoQ Total Replicates per Reagent Lot	LoQ Reported
Serum	LoB 60 LoD 75	0.05 mg/dL (4 µmol/L)	0.10 mg/dL (9 µmol/L)	75	0.15 mg/dL (13 µmol/L)
Urine	LoB 60 LoD 75	0.50 mg/dL (44 µmol/L)	1.00 mg/dL (88 µmol/L)	75	3.00 mg/dL (265 µmol/L)

Linearity

Table 5. Linearity interval for the Atellica CH Crea3 assay on both the Atellica CH and CI Analyzers.

Specimen Type	# of Sample Levels	Linearity Interval Reported
Serum	9	0.15 mg/dL (13 µmol/L) to 30.00 mg/dL (2652 µmol/L)
Urine	9	3.00 mg/dL (265 µmol/L) to 245.00 mg/dL (21,658 µmol/L)

The Crea3 assay is linear on the Atellica CH and CI Analyzers across the intervals indicated.

Table 6. Deming regression results for plasma and serum specimen equivalency on the Atellica CH and CI Analyzers.

Sample (y)	Reference sample (x)	Regression Equation	Sample Interval	n	Correlation Coefficient (r)
Sodium heparin plasma	Serum	y = 1.00x + 0.00 mg/dL (y = 1.00x + 0 µmol/L)	0.60–27.26 mg/dL (53–2410 µmol/L	50	0.999
Lithium heparin plasma	Serum	y = 0.99x + 0.06 mg/dL (y = 0.99x + 5 µmol/L)	0.60–27.26 mg/dL (53–2410 µmol/L)	50	0.999
Dipotassium EDTA	Serum	y = 0.98x + 0.04 mg/dL (y = 0.98x + 4 µmol/L)	0.60–27.26 mg/dL (53–2410 µmol/L)	50	0.998

Specimen equivalency for sodium heparin-, lithium heparin-, and dipotassium EDTA plasma vs. serum yielded slopes 0.98–1.00 and intercepts 0.00–0.06 mg/dL (0–5 µmol/L). Results are transferable to the Atellica CI Analyzer. Agreement of the specimen types may vary depending on the study design and sample population used.

Conclusion

The Atellica CH Crea3 assay demonstrated acceptable analytical performance and specimen equivalency for both the Atellica CH and CI Analyzers; Method comparison results for the Crea3 and Crea_2 assays on the Atellica CH Analyzer were similar for the same specimen type and similar for the Crea3 assay on the Atellica CH and CI Analyzers.

References

1. Levin A, Ahmed SB, Carrero JJ, et al. Executive summary of the KDIGO 2024 Clinical Practice Guideline for the Evaluation and Management of CKD: known knowns and known unknowns. Kidney Int 2024;105:684-701.
2. Iatridi F, Carrero JJ, Gall EC, et al. KDIGO 2024 Clinical Practice Guideline for the Evaluation and Management of CKD in Children and Adults: a commentary from the European Renal Best Practice (ERBP). Nephrol Dial Transplant 2025;40:273-282.
3. Cavalier E, Zima T, Datta P, et al. Recommendations for European laboratories based on the KDIGO 2024 Clinical Practice Guideline for the Evaluation and Management of CKD. Clin Chem Lab Med 2025;63:525-534.
4. Biljak VR, Honović L, Matica J, et al. The role of laboratory testing in detection and classification of CKD: national recommendations. Biochem Med (Zagreb) 2017;27:153-176.
5. Creatinine_3 assay. Atellica CH Analyzer. 11537275_EN Rev. 01, 2025-01.
6. Technical specifications. Atellica CI Analyzer. CLS-23-3123-76. QR700003930. 12-2023.
7. Creatinine_2 assay. Atellica CH Analyzer. 11110159_EN Rev. 04, 2019-07.

Data/some data first presented at Worldlab IFCC 2025

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