Practical Laboratory Medicine 31 (2022) e00282

Contents lists available at ScienceDirect

Practical Laboratory Medicine

journal homepage: www.elsevier.com/locate/plabm

Performance evaluation of the XT MicroSlide assay pairs on the Vitros XT 7600 compared to VITROS single microslide assays on Vitros 5600

Lily Olayinka [a,b], Estella Tam [b], Sridevi Devaraj [a,b,*]

[a] Department of Pathology and Immunology, Baylor College of Medicine, Houston, TX, 77030, USA
[b] Section of Clinical Chemistry, Division of Laboratory Medicine, Department of Pathology, Texas Children's Hospital, Houston, TX, 77030, USA

ARTICLE INFO

Keywords:
Method verification
Performance
Analyte
Assay
Vitros
XT slide

ABSTRACT

Objectives: Pediatric hospitals are always challenged by specimen volumes and thus any innovation in this realm is very welcome. With the introduction of Microslide assay pairs, we aimed to evaluate the analytical performance of the Vitros XT MicroSlide assay pairs on the Vitros XT 7600 compared to single MicroSlides.

Design: Performance characteristics included within-run precision, analytical measurable range, method comparison, and interference verification. We compared six XT MicroSlide pairs on the Vitros XT 7600 with twelve corresponding single slide assays on the Vitros 5600 system.

Results: The XT MicroSlides on Vitros XT 7600 demonstrated excellent precision, equivalent analytical measurable range, and strong method correlation with single slide assays on Vitros 5600 for most of the assays tested. Within-run CVs of the analytes ranged between 0.32% and 2.93% with between-run CV of less than 8.8% and linearity for all analytes was within the manufacturer's specified range. Interference studies showed comparable effects of hemolysis, lipemia, and bilirubin on both instruments.

Conclusions: The XT MicroSlides are comparable to the single MicroSlide assays with improved efficiency, turnaround times and lower sample volumes.

1. Introduction

One of the challenges clinical laboratories servicing pediatric centers face is adequate specimen collection from infants and children in addition to performing multiple tests on the limited sample volume [1]. This challenge is significantly heightened in critically ill patients and in patients for whom phlebotomy is challenging. Diagnostic phlembotomy is a known cause of iatrogenic blood loss which can result in the aggravation of hospital-acquired anemia [2].

Both Vitros 5600 and XT 7600 systems integrate dry chemistry (MicroSlide), wet chemistry and immunoturbidimetry with photometric detection (MicroTip), immunoassays with enhanced chemiluminescence (MicroWell), and photometric measurement of sample quality indices (MicroSensor) into a single analyzer. However, one of the unique features of the Vitros XT 7600 includes the introduction of the Vitros XT Microslide pair technology, which combines two film reagents onto the same slide to minimize sample

* Corresponding author. Baylor College of Medicine, Director, Clinical Chemistry, Texas Children's Hospital, 6621 Fannin St, Houston, TX, 77030, USA.
E-mail address: sxdevara@texaschildrens.org (S. Devaraj).

https://doi.org/10.1016/j.plabm.2022.e00282
Received 7 February 2022; Received in revised form 16 May 2022; Accepted 20 May 2022
Available online 23 May 2022
2352-5517/© 2022 Published by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

volume, improve efficiency and productivity. Currently available Vitros XT MicroSlide pairs include Alanine Aminotransferase/Aspartate Aminotransferase [ALTV-AST], Triglyceride/Cholesterol [TRIG-CHOL], Albumin/Total Protein [ALB-TP], Urea/Creatinine [UREA-CREA], Glucose/Calcium [GLU-CA], Total Bilirubin/Alkaline Phosphatase [TBIL-ALKP]. These MicroSlides are also available as single slides for use on the Vitros XT 7600 and were tested against the paired slides. For this short communication, only the comparison of the paired slides and single slides on 5600 will be discussed. .

In accordance with CLSI guidelines (EP05-A3 and EP-06 A), we evaluated results from XT Microslides for Vitros XT 7600 analyzer compared to the Single MicroSlides assays for Vitros 5600 and report the findings.

2. Materials and methods

Twelve analytes on the XT MicroSlides: ALTV-AST, TRIG-CHOL, ALB-TP, UREA-CREA, GLU-CA, TBIL-ALKP *versus* the Vitros 5600 single slide assays were evaluated using the Vitros XT 7600. The methodology, turnaround time and volume required for the single MicroSlides on Vitros 5600 and XT MicroSlide assays on Vitros XT 7600 are listed in Table 1.

Within-run and Between-run precision was assessed by measurement of ten replicates of two-concentration levels of the manufacturer's quality control material (Performance verifier 1 (PV-1) and Performance verifier 2 (PV-2)) for each analyte. Mean concentration and coefficient of variation (%CV) were calculated for each assay.

Linearity/Analytical Measurable Range (AMR) was carried out with two replicates of a five-level calibrator (Cal Kit 3–1, Cal Kit 3–2, Cal Kit 3–2, PV-1, PV-2), that span across the manufacturer's reportable range for all 12 analytes. The mean of duplicate measurements of each XT Microslide assays was calculated and compared with the expected value from single slides on the Vitros 5600.

Accuracy (method comparison) studies were performed using residual patient serum samples, based on the CLSI EP09-A3 guideline (CLSI guide 2013), except that each sample was measured once due to limited volume. The samples were selected to cover AMR as wide as possible, and twenty-four samples were collected for each assay. Deming regression was used for analysis to calculate an intercept, a slope, and 95% confidence intervals (CI). The percent bias for each assay was compared to the total allowable error (TEa) recommended by Clinical Laboratory Improvement Amendments (CLIA) to assess the clinical significance of the bias [3].

Assay interference was verified by spiking pooled serum samples (12 patients per pool) with different concentrations of hemoglobin (0, 200 mg/dL, 400 mg/dL and 800 mg/dL), bilirubin (0, 15 mg/dL, 30 mg/dL and 60 mg/dL) or intralipid (0, 400 mg/dL, 1000 mg/dL and 3000 mg/dL) using an in-house hemolysate stock and a commercial interference kit for lipemia and icterus (Sun Diagnostics). The concentration corresponded to normal, mild, moderate and significant interference and the hemolysate stock was prepared using the freezing and thawing method [4]. Turnaround time was obtained from the display user interface of each instrument after sample loading and sample volume required for each analyte was obtained from the manufacturer's MicroSlide assay summary.

Statistical analyses for method comparison of the single microslides to the paired slides were performed with EP Evaluator (Burlington, VT USA). The method comparison studies were evaluated using Deming regression, and a Bland-Altman plot was generated to assess bias.

3. Results

The %CVs and 95%CI of within-run and between-run precision were calculated and reported. Within-run %CVs ranged from 0.32% to 2.93%. All assays except total bilirubin met the manufacturer's claimed precision of 1.33%–3.91%. Total bilirubin was observed to exceed the manufacturer's specification at low-level concentration (2.93% vs 2.1%). The between-run precision was 0.7%–8.7%.

Table 1
Sample volume, turnaround time and methodology required for the XT MicroSlides vs. Single slides.

Assays	Single slides (μL)	XT Slides (μL)	Turnaround time (TAT)		Methodology
			5600	XT 7600	
Albumin	5.5	4.2	5 m 3s	5 m 3s	Bromocresol green (BCG)
ALKP	11	5.0	5 m 10s	5 m 10s	*p*-nitrophenyl phosphate substrate
ALTV	7	3.5	5 m 4s	5 m 12s	L-alanine - α-ketoglutarate (P-5-P)
AST	7	3.3	5 m 13s	**ALTV	L-aspartate - α-ketoglutarate (P-5-P)
BUN/Urea	5.5	4.3	5 m 4s	5 m 3s	Colorimetric urease reaction
Calcium	10	3.5	5 m 32s	5 m 31s	Arsenazo III dye
Cholesterol	5.5	3.9	4 m 45s	5 m 3s	Cholesterol oxidase
Creatinine	6	3.2	4 m 54s	**BUN/Urea	Creatine amidinohydrolase
Glucose	6	2.7	5 m 14s	** Calcium	Glucose oxidase
Total Bilirubin	10	5.0	5 m 10s	**ALKP	4-(N-carboxymethylsulfonyl) benzenediazonium hexafluorophosphate
Total Protein	6.5	4.1	5 m 3s	**Albumin	Biuret reaction
Triglycerides	5.5	2.9	5 m 4s	**Cholesterol	L-α-glycerophosphate oxidase

AST- Aspartate Aminotransferase; BUN - Blood Urea Nitrogen.
Total TAT for all assays on 5600: 69 min 2 s.
Total TAT for all assays on XT7600: 31 min 2 s.
Total volume for all assays on 5600: 85.5 μL.
Total volume for all assays on XT 7600: 45.6 μL.
**runs simultaneously.

The AMR of each assay was found to be linear within the manufacturer's specified ranges. The mean slope for all assays ranged from 0.98 to 1.01, with a mean y-intercept ranging from −5.58 to +2.43 and a correlation coefficient from 0.99 to 1.00.

All 12 analytes demonstrated a correlation coefficient >0.95. ALT, CA, CREA, CHOL, and GLU met the set criteria of r = > 0.95 and slopes of 0.9–1.1. Five assays (ALB, ALKP, AST, TP, TRIG and UREA) exhibited slopes <0.9. Percent bias for all the assays were within the desirable specification for total allowable error for each analyte (Table 2).

With regards to total turnaround time (TAT), the XT7600 had a TAT of 31 min and 2 s compared to 69 min 2 s on the 5600. Similarly, the volume of samples utilized by the XT7600 was 53% less than the volume required by the 5600 (Table 1).

All assays were included in the interference studies however, only analytes significantly affected by an interference were reported. The ALB, AST, TBIL and TP assays were affected by mild, moderate and significant hemolysis on the single MicroSlides and XT MicroSlide assays. At mild hemolysis (H-index <250), ALB, AST, TBIL and TP showed a positive bias of 11%, 51%, 1 mg/dL and 11% respectively. Glucose was affected by significant hemolysis with a positive bias of 18%. TP was significantly affected by mild, moderate and significant bilirubin with a positive bias >20%. ALKP was significantly affected by moderate and significant bilirubin with a positive bias >30%. Glucose was affected by intralipid interference on both types of assays with a positive bias of 12–13% (Table 3).

4. Discussion

This report describes the method verification of the XT MicroSlide assays: ALTV-AST, TRIG-CHOL, ALB-TP, UREA-CREA, GLU-CA, TBIL-ALKP on the Vitros XT 7600 digital chemistry analyzer in our pediatric population. The XT Microslide assays demonstrated acceptable intra-assay precision and linearity across clinically relevant concentrations of all 12 analytes.

Analytical performance evaluation is essential when introducing newly developed assays and instruments to the clinical laboratory to replace existing ones. Ensuring that the new instruments and/or assays provide reliable test values comparable to the instruments being replaced is critical.

The XT MicroSlide and single slide assays have multilayered analytical elements coated on a polyester support with similar methodologies for corresponding analytes. Our method comparison studies showed a good correlation between the XT MicroSlides and the single assays. We did observe a small negative percent bias in the XT ALKP and TRIG assays compared to the single slides, −4.83% and −8.42%, respectively.

The sample volume required for all analytes on the XT7600 was significantly lower that the volume required for the analytes on the vitro 5600. Insufficient sample volume is a common cause of incomplete laboratory analysis in the pediatric population and phlebotomy has been identified as a major cause of hospital acquired anemia in hospitalized patients. A prospective multicenter study further reported that blood draws accounted for 73% of daily blood loss which led to blood transfusions in hospitalized pediatric patients [5–8]. Therefore, the development of chemistry analyzers like the XT7600 that can accommodate small sample volumes is advantageous for clinical laboratories that serve special populations like pediatrics. Likewise, the introduction of dual slides that allows simultaneous analysis of two analytes for the time allotted for one single slide significantly improved efficiency.

Table 2
Method comparison of XT MicroSlides vs 5600 single slides.

Assay	Slope (95%CI)	Intercept (95%CI)	Correlation coefficient (R)	TEa	Mean % bias	Within-run % CV	Between-run % CV
Albumin	0.80 (0.70–0.89)	0.84(0.44–1.24)	0.961	10%	0.00	1.11	1.4
ALKP	0.82 (0.75–0.86)	25.82(15.09–36.54)	0.984	30%	−4.63	7.96	3.0
ALTV	0.92 (0.85–0.98)	2.88(1.26–4.51)	0.986	20%	0.83	1.17	1.6
AST	0.81 (0.78–0.84)	7.87(6.61–9.12)	0.997	20%	−0.58	0.94	1.5
BUN/Urea	1.12 (1.03–1.20)	−1.39(-2.68 to −0.11)	0.984	9%/2 mg/dL	0.33	0.73	1.4
Calcium	1.16 (1.06–1.26)	−1.31(-2.28 to −0.34)	0.980	1 mg/dL	0.22	0.43	0.8
Cholesterol	0.96 (0.90–1.01)	6.07(-2.34–14.47)	0.993	10%	−1.42	1.48	1.2
Creatinine	1.02 (0.99–1.04)	−0.03(-0.05 to −0.01)	0.998	15%/0.3 mg/dL	−0.02	0.90	1.4
Glucose	1.01 (0.92–1.10)	−0.69(-7.45 to 6.08)	0.981	10%/6 mg/dL	0.21	0.57	0.7
Total Bilirubin	1.30 (1.20–1.52)	−0.02(-0.32 to −0.10)	0.965	20%/0.4 mg/dL	0.05	2.93	8.7
Total Protein	0.90 (0.78–1.00)	0.85(0.08–1.61)	0.962	10%	0.07	0.75	1.4
Triglycerides	0.86 (0.79–0.92)	12.08(2.29–21.86)	0.983	25%	−8.42	1.44	0.8

CI – Confidence Interval; TEa – total allowable error (CAP); CV – Coefficient of variation; ALKP – Alkaline Phosphatase; ALTV – Alanine Aminotransferase; AST- Aspartate Aminotransferase; BUN Blood Urea Nitrogen.

Table 3
Effect of hemolysis, icterus and lipemia on analytes on the Vitros 5600 vs Vitros XT 7600.

Analyte	Units	Vitros 5600				Vitros XT 7600			
		Baseline	Mild hemolysis	Moderate hemolysis	Significant hemolysis	Baseline values	Mild hemolysis	Moderate hemolysis	Significant hemolysis
Hemolysis index values		<15	209	384	883	<15	166	328	860
ALB	g/dL	3.5 ± 0.0	4.0 ± 0.0	4.5 ± 0.0	5.6 ± 0.1	3.5 ± 0.0	4.0 ± 0.0	4.5 ± 0.0	5.6 ± 0.0
ALKP	U/l	114 ± 1.4	102 ± 1.4	97.5 ± 3.5	91.5 ± 4.9	111.5 ± 1.4	94.5 ± 3.5	91.5 ± 0.7	70.5 ± 7.8
AST	U/l	43.5 ± 0.7	66.0 ± 0.0	89.0 ± 0.0	NR	44.5 ± 0.7	68.5 ± 0.7	91.5 ± 0.7	NR
Glucose	mg/dL	74.0 ± 0.0	75.0 ± 0.0	79.0 ± 0.0	87.0 ± 0.0	73.0 ± 0.0	75.0 ± 0.0	79.0 ± 0.0	88.0 ± 0.0
TBIL	mg/dL	0.6 ± 0.0	1.6 ± 0.0	2.7 ± 0.0	NR	0.6 ± 0.0	1.8 ± 0.1	2.8 ± 0.0	NR
TP	g/dL	6.1 ± 0.1	6.9 ± 0.1	7.7 ± 0.1	9.3 ± 0.0	6.2 ± 0.0	7.0 ± 0.0	7.8 ± 0.1	9.5 ± 0.1
		Baseline	Mild icterus	Moderate icterus	Significant icterus	Baseline	Mild icterus	Moderate icterus	Significant icterus
Icterus index		<2	7	15	>25	<2	5	16	>25
ALKP	U/l	105.0 ± 0.0	127.0 ± 1.4	163.5 ± 3.5	230.0 ± 3.5	106.0 ± 0.0	126.0 ± 1.4	158.0 ± 2.8	209.5 ± 2.8
TP	g/dL	5.5 ± 0.0	5.8 ± 0.0	6.0 ± 0.1	6.5 ± 0.0	5.9 ± 0.0	6.2 ± 0.1	6.4 ± 0.1	6.8 ± 0.0
		Baseline	Mild lipemia	Moderate lipemia	Significant lipemia	Baseline values	Mild lipemia	Moderate lipemia	Significant lipemia
Lipemia index values		<20	36	133	326	<20	43	127	294
Glucose	mg/dL	64.5 ± 0.7	65.5 ± 0.7	67.5 ± 0.7	73.0 ± 0.7	60.5 ± 0.7	61.0 ± 0.0	62.5 ± 0.7	67.5 ± 0.7

Data provided as mean ± SD; ALB – Albumin; ALKP – Alkaline Phosphatase; TBIL - Total bilirubin; TP – Total protein; NR - Not reported.

There is a lack of comparative data for the XT MicroSlides. However, reports from the manufacturer on the analytical performance of the XT MicroSlide assays using sigma metric methodology demonstrated excellent precision and accuracy when evaluated against the CLIA TEa requirement [9]. Similarly, the comparison of within-lab precision for the XT MicroSlides on the XT 7600 with corresponding single test slides on the Vitros 5600 showed that the XT MicroSlides exhibited comparable or improved precision relative to single test slides [10]. The Vitros 5600 and XT 7600 integrated systems also eliminate carryover using disposable versa tips and disposable cuvettes, although this claim was not verified in this study.

ALB, AST, GLU, TBIL, and TP assays were affected by hemolysis. This observation is in congruent with previous findings that hemolysis can increase the serum concentrations of analytes such as AST, LDH and potassium [11]. Although the hemolysis index result for the Vitros XT 7600 was slightly lower than that of the Vitros 5600, the effect of hemolysis on the analytes was similar on both instruments. Glucose was affected by severe lipemia beyond the approved total allowable error recommended by CAP. Of note, the manufacturer does not recommend using grossly lipemic samples with the glucose assay because of lipid interference with assay reactants [12].

There are some limitations in the study which need to be pointed out: i) small number of samples used for comparison (n = 24) due to the pediatric population; ii)dual slides may go against choosing wisely practices; for example cholesterol wouldn't always need to be tested along with TG in all settings.

In summary, the XT MicroSlide assays on the XT 7600 demonstrated comparable analytical measurement ranges, precision, and strong method correlation with the Single MicroSlides for all the assays. It is suitable for use in the pediatric setting as it accommodates lower sample volumes and improved turnaround times without any effect on performance.

Credit author statement

Lily Olayinka: Conceptualization, Methodology, Validation, Writing-Original Draft. Estella Tam: Conceptualization, Methodology, Validation. Sridevi Devaraj: Conceptualization, Writing-Original Draft, Supervision.

Declaration of competing interest

None.

Acknowledgements

LO was supported by the Ching Nan-Ou Fellowship Endowment.

References

[1] S.L. Valentine, S.T. Bateman, Identifying factors to minimize phlebotomy-induced blood loss in the pediatric intensive care unit, Pediatr. Crit. Care Med. (2012), https://doi.org/10.1097/PCC.0b013e318219681d.

[2] P. Helmer, S. Hottenrott, A. Steinisch, D. Röder, J. Schubert, U. Steigerwald, S. Choorapoikayil, P. Meybohm, Avoidable blood loss in critical care and patient blood management: scoping review of diagnostic blood loss, J. Clin. Med. (2022), https://doi.org/10.3390/JCM11020320.

[3] CLIA eCFR :: 42 CFR Part 493 – Laboratory Requirements. https://www.ecfr.gov/current/title-42/chapter-IV/subchapter-G/part-493. Accessed 21 Apr 2022.

[4] Kalaria T, Gill H, Sharrod-Cole H, Ford C, Gama R Conflicting effects of haemolysis on plasma sodium and chloride are due to different haemolysis study protocols: A case for standardisation. https://doi.org/10.1177/00045632211040691.

[5] P.C. Kurniali, S. Curry, K.W. Brennan, K. Velletri, M. Shaik, K.A. Schwartz, E. Mccormack, A Retrospective Study Investigating the Incidence and Predisposing Factors of Hospital-Acquired Anemia, 2014, https://doi.org/10.1155/2014/634582.

[6] D. Zhou, Y.L. Luo, S.H. Luo, M. Feng, M.L. Tang, The effect of diagnostic blood loss on anemia and transfusion among postoperative patients with congenital heart disease in a pediatric intensive care unit, J. Pediatr. Nurs. 38 (2018) 62–67.

[7] N.L. Jackson Chornenki, T.E. James, R. Barty, Y. Liu, B. Rochwerg, N.M. Heddle, D.M. Siegal, Blood loss from laboratory testing, anemia, and red blood cell transfusion in the intensive care unit: a retrospective study, Transfusion 60 (2020) 256–261.

[8] C.E. Counsilman, L.E. Heeger, R. Tan, V. Bekker, J.J. Zwaginga, A.B. te Pas, E. Lopriore, Iatrogenic blood loss in extreme preterm infants due to frequent laboratory tests and procedures, J. Matern. Fetal Neonatal Med. 34 (2021) 2660–2665.

[9] M. Barbero, T. Dimagno, C. Graby, T. Huynh, Sigma metrics for assessing the analytical quality of the new multi-test VITROS® XT chemistry products slides, Clin. Chim. Acta (2019), https://doi.org/10.1016/j.cca.2019.03.1061.

[10] J. Miller, T. Dimagno, Precision OF the new multi-test VITROS® chemistry product slides* ON the vitros XT 7600 integrated system, Clin. Chim. Acta (2019), https://doi.org/10.1016/j.cca.2019.03.1098.

[11] S. Agarwal, G. Vargas, C. Nordstrom, E. Tam, G.J. Buffone, S. Devaraj, Effect of interference from hemolysis, icterus and lipemia on routine pediatric clinical chemistry assays, Clin. Chim. Acta (2015), https://doi.org/10.1016/j.cca.2014.08.008.

[12] Ortho Clinical Diagnostics Technical Documents. https://www.orthoclinicaldiagnostics.com/en-us/home/technical-documents. Accessed 23 Nov 2021.