

Precision and bias evaluation of three different triglyceride assays: Discrepant bias per the CLSI EP15-A3 and EP09-A3 guidelines

Hatice Bozkurt Yavuz*, Soycan Tüner

Department of Clinical Biochemistry, Faculty of Medicine, Uşak University, Türkiye

ABSTRACT

Background: Besides its clinical importance, triglyceride (TG) measurement plays a critical role in calculating Low-Density Lipoprotein Cholesterol (LDL-C). Therefore, it is important to highlight any biases when changing methods. The Clinical Laboratory Standards Institute (CLSI) EP09-A3 recommends evaluating bias between two methods using fresh samples, while EP15-A3 suggests verifying precision and estimating bias, with quality control (QC) material as an option for samples.

Methods: The performance of three different TG reagents (Abbott Triglyceride, Abbott Triglyceride-2, and Archem Triglyceride) with EP09-A3 and EP15-A3 and the effects of TG results on LDL-C calculations were evaluated.

Results: Notable inconsistencies were observed between biases from patient samples and QC materials. The lowest QC measurements and the highest patient results were both seen with Abbott TG. While median bias values were within acceptable limits, there were discrepancies in slope and intercept values between TG reagents. The bias in LDL-C values calculated using Friedewald's and Sampson's equation was acceptable, but the Martin-Hopkins equation showed even less bias. The Abbott TG reagent fully met the imprecision criteria, the Abbott TG 2 reagent partially but clinically acceptably met them, whereas the Archem TG reagent, despite having imprecision values below the optimum CV, failed to meet the manufacturer's claims.

Conclusions: This discrepancy in bias between patient samples and QC materials underscores the challenge of control material commutability. Laboratories should assess bias when switching methods using the practical EP09-A3 approach, and perform an EP15-A3 verification study before introducing any new kit into routine use.

Keywords: bias, CLSI EP09-A3, CLSI EP15-A3, triglyceride, verification

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INTRODUCTION

Fasting serum triglyceride (TG) levels of 150 mg/dL or higher are linked to an increased risk of cardiovascular disease, while extremely high levels (500 mg/dL or above) also elevate the risk of pancreatitis [1]. Besides its own clinical importance, TG plays a crucial role in the calculation of Low Density Lipoprotein - Cholesterol (LDL-C) [2]. Friedewald, Martin-Hopkins and Sampson formula are commonly used equations for calculating LDL cholesterol [2-4]. Both the Friedewald and Martin-Hopkins formulas are not considered valid when triglyceride levels exceed 400 mg/dL [5]. In contrast, the Sampson equation remains accurate at triglyceride concentrations up to approximately 800 mg/dL [4]. TG can be measured using different methods in laboratories, with the enzymatic method being a commonly used one, both Abbott and Archem reagents perform measurements using this method [6-8]. Given the importance of accurate

TG measurements, it is essential to assess the performance of different reagents used in enzymatic methods to ensure reliable results. The Clinical & Laboratory Standards Institute (CLSI) recommends conducting a bias study according to the CLSI EP09-A3 guideline (Measurement Procedure Comparison and Bias Estimation Using Patient Samples; Approved guideline) to detect any bias between two methods in laboratories [9]. The CLSI EP15-A3 guideline (User Verification of Precision and Estimation of Bias; Approved guideline) serves as a useful diagnostic tool for investigating and verifying method precision performance [10]. In this framework, EP15-A3 is used for user verification and complements, rather than replaces, patient-sample-based method comparison performed according to the EP09-A3 guideline. Consequently, some differences between the results obtained using the EP09-A3 and EP15-A3 guidelines may be expected, particularly when non-commutable control or reference materials are involved. In Conformité Européenne (CE)-

* Corresponding author: Hatice Bozkurt Yavuz, Department of Clinical Biochemistry, Faculty of Medicine, Uşak University, Turkey. E-mail: haticebozkurtyavuz@gmail.com

certified products, comparisons are made using the current method [11]. The aim of this study is to determine both the precision of three different reagents used on the same analyzer and whether there is any bias between them. Additionally, if a bias is present, the study aims to evaluate its impact on the calculated LDL-C results. Furthermore, the bias results obtained using different methods from two guidelines will also be compared.

METHODS

This prospective method comparison study was conducted in the Medical Biochemistry Laboratory of Uşak Training and Research Hospital. The study used anonymized routine laboratory data; however ethical approval was obtained from the local Ethics Committee in accordance with institutional and national regulations. The study was designed and performed in accordance with the Clinical and Laboratory Standards Institute (CLSI) guidelines, specifically EP15-A3 for verification of precision and bias, and EP09-A3 for method comparison and bias estimation. The study was approved by the local Ethics Committee (455-455-02).

Reagents

In the study, three different TG reagents were compared: Abbott Triglyceride reagent (601664N23), Abbott Triglyceride 2 reagent (56477UD00), and Archem Triglycerides reagent (ref: 06T85-35). Abbott Architect C16000 analyzer (Abbott Diagnostics, IL, USA) was used and quality control protocols were followed for each reagent, the assays were tested according to the instrument operation instructions. All reagents had CE certification [6-8]. Technopath internal quality control (IQC) materials (LOT numbers: 13910231 and 13910232; Tipperary, Ireland) were used for active evaluation of analytical quality control, Westgard multirule quality control procedures were routinely applied in our laboratory.

Implementation of the CLSI EP09-A3 guideline

According to the CLSI EP09-A3 guideline, the bias study should be conducted by the end-user laboratory using at least 40 fresh samples [9]. The study involved the use of leftover patient serum samples. The leftover patient samples were all de-identified during the collection. In this context, during the specified dates we used 50 samples which also had results of total cholesterol (TC), High Density Lipoprotein cholesterol (HDL-C). This sample size exceeds the minimum requirement recommended by the CLSI EP09-A3 guideline for method comparison studies. Samples identified as having interference from hemolysis and icterus indices according to the triglyceride kit inserts were excluded from the study. Passing-Bablok regression analyses were used to calculate the intercept and slope values. Also, the allowable bias percentage was set at 9.6% for TG and 5.4% for LDL-C [12]. Desirable

bias specifications were applied, as they are commonly used in method comparison studies to account for systematic differences between analytical methods. Passing Bablok fits were generated using Analyze-it® for Microsoft Excel (Analyze-it Software, Ltd., Leeds, UK).

LDL-C calculation

Of the 50 samples, 40 with TG levels <400 mg/dL were included in LDL-C calculations using the Friedewald and Martin-Hopkins equations, while 48 samples with TG levels <800 mg/dL were used for the Sampson equation. LDL-C was calculated using the following equations, with all lipid parameters expressed in mg/dL: Friedewald equation ($LDL-C = TC - [HDL-C + TG/5]$); Martin-Hopkins equation ($LDL-C = TC - [HDL-C + TG/adjustable\ factor]$); and Sampson equation ($LDL-C = 0.948 \times TC - 0.971 \times HDL-C - 0.302 \times TG + 6.3$) [2-4]. TC and HDL-C values were retrieved from the laboratory information system. Bias between calculated LDL-C results was evaluated using Passing-Bablok regression analysis.

Implementation of the CLSI EP15-A3 guideline

The CLSI EP15-A3 guideline was used for both user verification of precision and estimation of bias. Technopath internal quality control (IQC) materials (LOT numbers: 13910231 and 13910232; Tipperary, Ireland) were used for EP15 A3 [13]. The IQC material was tested five times a day for five days between October 21 and October 25, 2024. Within-subject variation (CV_i) was obtained from the European Federation of Clinical Chemistry and Laboratory Medicine (EFLM) Biological Variation (BV) database [12], and the optimal analytical coefficient of variation (CV_A) value was set as 25 percent of the CV_i ($19.8/4 = 4.95$) [14]. This approach was used to ensure stricter control of analytical repeatability in triglyceride measurements. The results were also compared with the values provided by the claims of the manufacturers on reagent assay sheets. According to the recommendations of the guideline, Grubbs' test was applied for outlier exclusion with the equation of Grubbs' limits = mean $\pm G \times$ standard deviation, ($G=3.135$ for 5 run and 25 total number of results) [10]. Within-laboratory imprecision was assessed using one-way ANOVA. Statistical analyses were performed using Analyze-it® for Microsoft Excel.

For the ANOVA calculations, between-run and within-run sums of squares were computed. These values were then divided by their respective degrees of freedom—4 for between-run and 20 for within-run—to obtain the mean square (MS) estimates. The variation between runs (VB) was calculated as (MS between-run – MS within-run) / the average number of results per run (n_0). In accordance with the guideline, when MS between-run was less than MS within-run, VB was set to zero. As de-

fined in the guideline, the MS within-run term was taken as the within-run variance component (VW). The user estimate for repeatability(SR) was calculated as $SR=VW$, while the between-run standard deviation (SB) was obtained as $SB=VVB$. The user estimate for within-laboratory imprecision (SWL) was subsequently derived using the formula $SWL = \sqrt{VB+VW}$.

To calculate the Upper Verification Limit (UVL), the manufacturer’s within-run and within-laboratory precision claims were used. According to the EP15-A3, the ratio of the manufacturer’s within-laboratory CV% to the within-run CV% was computed to obtain the p (rho) value. The df WL corresponding to this rho value was selected from the table of the guideline, and the appropriate F factor was obtained based on this df and the number of samples in the study. Finally, the UVL was calculated using the formula $UVL = F \times \text{manufacturer within-laboratory CV\%}$.

RESULTS

Results per the CLSI EP09-A3 guideline

The median (IQR) results for fresh patient samples measured with Abbott TG, Abbott TG 2, and Archem TG reagents were found to be 220 (198.75), 219.5 (210.25), and 214 (204.75), respectively. Table 1 presents the findings of the CLSI EP09-A3 guideline for TG, summarizing the bias evaluations for different reagents. All TG median bias values were below the desirable bias value. However, only the Passing-Bablok regression between Abbott TG and Archem TG 2 showed that the 95% confidence interval of the slope included 1, indicating no proportional bias, while the 95% confidence interval of the intercept between Abbott TG 2 and Archem TG included 0, indicating no constant bias.

Results of LDL-C calculation

When impact of different TG reagents were evaluated using the Friedewald formula, median bias values remained below the desirable limit. Consistent slope and intercept were observed between LDL-C Friedewald (Abbott TG) and (Archem), whereas LDL-C (Abbott TG 2) and (Archem) showed consistency only in slope. For LDL-C calculated with the Martin–Hopkins formula, all bias values were below the desirable threshold, and in every pairwise comparison the 95% CIs of the slope and intercept included 1 and 0, respectively. Similarly, for the Sampson formula, all bias values were below the desirable bias. Slope and intercept confidence intervals included 1 and 0 for comparisons between Abbott TG and Archem TG reagents, but not for comparisons between the two Abbott reagents. In the comparison between Abbott TG 2 and Archem TG, only the slope CI included 1. The findings are summarized in Table 2.

Results per the CLSI EP15-A3 guideline

The results of the Grubbs test and the values to be excluded, along with the ANOVA test results, are summarized in Table 3. According to the results of the Grubbs test, one high level internal quality control result from each of the two test kits (Abbott TG and Abbott TG2) was found to be slightly above the upper acceptable limit. However, based on the recommendations of the relevant guideline, no identifiable source of error could be determined. In line with the guideline, and since no extreme outliers were detected, these results were included in the calculations of CV and other performance parameters. CV_A values of reagents and the claims of imprecision as CV% with upper verification limits are shown in Table 3. Observed bias and verification limits according to the EP15-A3 were shown in Table 4. Passing Bablok fit illustrating the pairwise com-

Table 1. Pairwise comparison of the three triglyceride reagents, according to the CLSI EP 09-A3 guideline

Compared TG reagents	Median difference (95% CI)	Passing-Bablok slope (95% CI)	Passing-Bablok intercept (95% CI)
Abbott TG vs Abbott TG2	1.307 (0.159; 2.454)	1.082 (1.068; 1.096)	-16.045 (-18.903; -12.714)
Abbott TG vs Archem TG	-3.149 (-4.483; -1.814)	1.015 (0.997; 1.039)	- 9.824 (-13.683; -6.846)
Abbott TG2 vs Archem TG	-3.559 (-4.286; -2.832)	0.942 (0.923; 0.965)	3.482 (-0.018; 6.346)

The comparison of the three TG reagents was performed on 48 samples (n=48). TG were measured in mg/dL. Abbreviations: CI – confidence interval, PB – Passing-Bablok (regression), TG – triglycerides.

Table 2. Pairwise comparison of estimated LDL-C, using the three triglyceride reagents

Compared TG reagents	LDL-C formula	Median difference (95% CI)	PB slope (95% CI)	PB intercept (95% CI)
Abbott TG vs Abbott TG2	Friedewald	0.335 (0.093; 0.576)	1.018 (1.008; 1.029)	-1.708 (-2.912; -0.575)
	Martin–Hopkins	0.303 (0.155; 0.452)	1.006 (0.998; 1.015)	-0.239 (-1.315; 0.576)
	Sampson	0.247 (0.080; 0.413)	1.013 (1.001; 1.026)	-1.968 (-3.155; -0.536)
Abbott TG vs Archem TG	Friedewald	1.297 (0.911; 1.683)	1.008 (1.000; 1.019)	0.509 (-0.644; 1.500)
	Martin–Hopkins	0.826 (0.609; 1.044)	1.006 (1.000; 1.017)	0.253 (-1.150; 0.950)
	Sampson	1.065 (0.826; 1.304)	1.008 (1.000; 1.018)	0.162 (-0.744; 1.100)
Abbott TG2 vs Archem TG	Friedewald	0.882 (0.363; 1.402)	0.994 (0.984; 1.000)	1.724 (0.900; 2.859)
	Martin–Hopkins	0.461 (0.219; 0.703)	1.001 (0.995; 1.012)	0.416 (-0.768; 1.170)
	Sampson	0.756 (0.443; 1.070)	0.997 (0.988; 1.006)	1.367 (0.372; 2.579)

The three triglyceride reagents were compared pairwise. Each comparison was performed using the three most commonly applied LDL-C estimation formulas: Friedewald, Martin–Hopkins, and Sampson. The number of samples included was n = 40 for the Friedewald and Martin–Hopkins calculations, and n = 48 for the Sampson calculation. TG and LDL-C were measured in mg/dL. Abbreviations: CI – confidence interval, LDL-C – low-density lipoprotein cholesterol, PB – Passing-Bablok (regression), TG – triglycerides.

parisons of all three kits are presented as Figure 1. In the analysis performed with the high-level internal quality control of the Abbott TG reagent—and similarly for the Abbott TG 2 reagent at both control levels—the MS between run component was found to be lower than the MS within-run component. Therefore, VB and consequently the between-run imprecision were set to zero, indicating that the entire within-laboratory imprecision originated from the within-run component. At the low concentration of the Abbott TG reagent, the between-run imprecision was also minimal, and the Abbott TG reagent successfully met the allowable imprecision criteria at both concentration levels. For the Abbott TG 2 reagent, the high-level control (131 mg/dL) met the verification criteria, whereas the low-level control (77 mg/dL) exhibited within-laboratory imprecision above the UVL and therefore failed to meet the required limit. Upon evaluation of the Archem TG reagent results, both the within-run and within-laboratory CV

values were found to be higher compared with the Abbott reagents. Moreover, these CV values exceeded the manufacturer's claimed imprecision limits. Therefore, the reagent did not meet the allowable imprecision criteria.

Abbott TG reagent fully met the verification criteria and was classified under Scenario 1A of the CLSI EP15-A3 guide. Abbott TG 2 reagent demonstrated a Scenario 1B pattern, partially meeting the criteria; however, the acceptable level corresponded to the concentration range most frequently encountered in clinical practice, so routine use of the test was continued. In contrast, for the Archem TG reagent, although the observed imprecision values at both levels were below the optimum CV threshold (4.95%), it exceeded the manufacturer's imprecision claims at both levels and was classified under Scenario 3 (several estimates failed). Consequently, the manufacturer was contacted, routine use of the test was suspended until the issue is resolved, and a more comprehensive evaluation following the CLSI EP05 has been considered.

Table 3. Precision verification results for the three triglyceride reagents according to the CLSI EP15-A3

	Abbott TG		Abbott TG2		Archem	
	77 mg/dL	131 mg/dL	77 mg/dL	131 mg/dL	77 mg/dL	131 mg/dL
Lowest result (mg/dL)	73	123	77	129	60	123
Highest result (mg/dL)	82	135	89	141	98	147
Grubbs' lower limit (mg/dL)	72.3	118.7	71.1	123.3	59.2	113.0
Grubbs higher limit (mg/dL)	84.5	134.2	90.4	140.9	108.7	156.0
Between-run sum of squares	17.20	14.16	6.16	19.44	342.16	411.44
Between-run mean squares	4.30	3.54	1.54	4.86	85.54	102.86
Within-run sum of squares	74.80	134.00	220.40	170.40	1154.80	718.80
Within-run mean squares	3.74	6.70	11.02	8.52	57.74	35.94
Within-run variance (Vw)	3.74	6.70	11.02	8.52	57.74	35.94
Between-run variance (Vb)	0.11	0.00	0.00	0.00	5.56	13.38
Within-run SD (SR)	1.93	2.59	3.32	2.92	7.60	5.99
Between-run SD (SB)	0.33	0.00	0.00	0.00	2.36	3.66
Within-laboratory SD (SWL)	1.96	2.59	3.32	2.92	7.96	7.02
Grand mean	78.40	126.44	80.76	132.08	83.96	134.48
Within-run CV (%)	2.47	2.05	4.11	2.21	9.05	4.46
Between-run CV (%)	0.43	0.00	0.00	0.00	2.81	2.72
Within-laboratory CV (%)	2.50	2.05	4.11	2.21	9.48	5.22
Claim of manufacturer within-run CV	0.80	0.70	1.10	0.80	1.41	1.68
Claim of manufacturer within-laboratory CV	1.88	1.75	2.00	1.70	1.85	2.75
p (rho)	2.35	2.49	1.82	2.13	1.31	1.64
df WL for Upper verification limit	6	5	7	6	12	8
F for Upper verification limit	1.55	1.6	1.51	1.55	1.39	1.48
UVL for within-laboratory CV	2.91	2.79	3.02	2.64	2.57	4.07
Fail/pass	Pass	Pass	Fail	Pass	Fail	Fail

Abbreviations: CV – coefficient of variation, df – degrees of freedom, F – factor for determining upper verification limit, SD – Standard deviation, TG – triglycerides, UVL – upper verification limit.

Table 4. Bias results obtained by internal quality control samples with the CLSI EP15-A3 guideline

TG reagent	IQC level	IQC concentration	Mean of measurements	Observed bias	UVL	LVL	Allowed bias*
Abbott TG	Low	77.00	78.40	1.40	78.45	75.55	7.39
	High	131.00	126.44	-4.56	132.32	129.69	12.58
Abbott TG 2	Low	77.00	80.76	3.76	77.87	76.13	7.39
	High	131.00	132.08	1.08	132.54	129.46	12.58
Archem	Low	77.00	83.96	6.96	84.36	69.64	7.39
	High	131.00	134.48	3.48	138.09	123.91	12.58

*Allowed bias was calculated using the 9.6% bias value for TG, provided by the European Federation of Clinical Chemistry and Laboratory Medicine (EFLM) biological variation database. All data presented in the table are expressed as mg/dL. Abbreviations: IQC – internal quality control, LVL – lower verification limit, TG – triglycerides, UVL – upper verification limit.

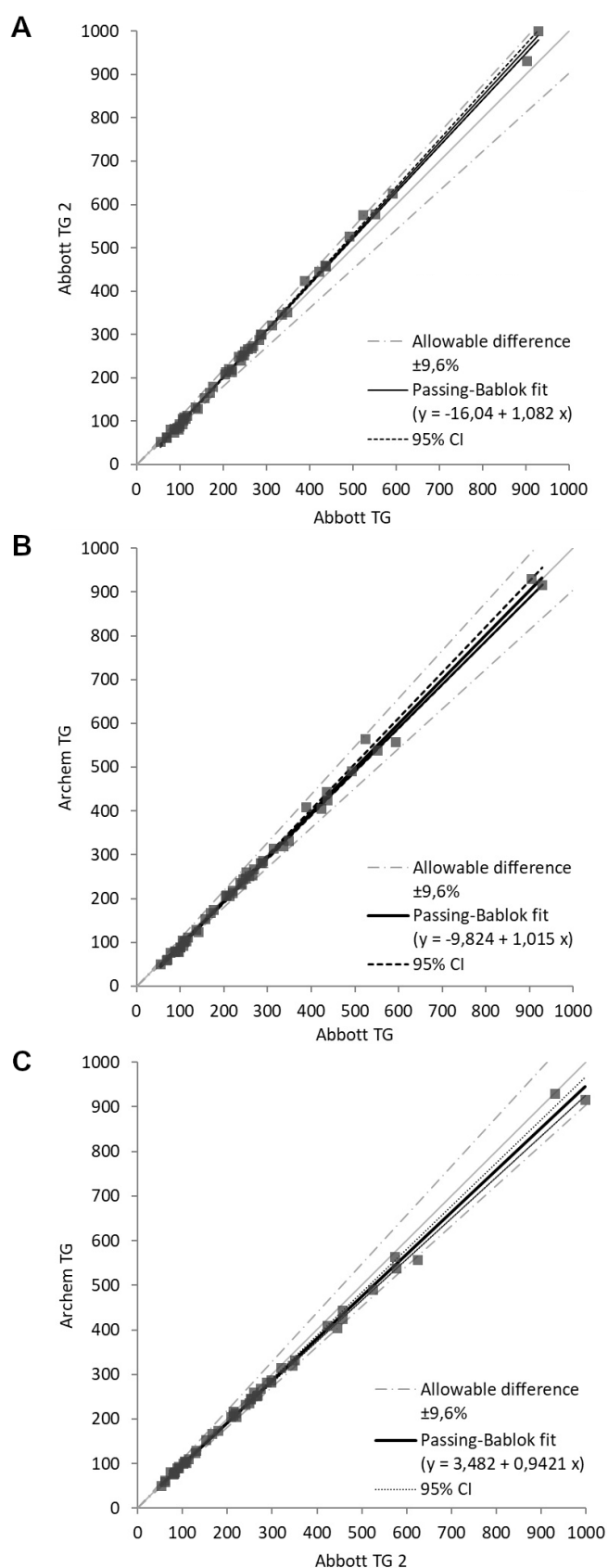


Figure 1. Passing–Bablok regression plots illustrating the method comparison between triglyceride reagents (A) Abbott TG 2 vs Abbott TG (reference method); (B) Archem TG vs Abbott TG (reference method); (C) Archem TG vs Abbott TG 2 (reference method). Method comparison was performed on 50 samples.

DISCUSSION

In the stage of our study conducted according to the EP09 guideline, although the differences are within an acceptable range, the patient results measured with the Archem Triglycerides reagent were found to be lower compared to those measured with both Abbott TG and Abbott TG 2 reagents. In the stage of our study carried out according to the CLSI EP15 guideline, when measurements were made using the same IQC material, Abbott Triglyceride was the only reagent that showed a negative bias compared to the IQC material. In other words, the bias observed in the study of patient samples and the study of control material did not align in the same direction. Since the analytical results of fresh patient samples and commutable samples are alike, commutable samples can serve as substitutes for fresh patient samples in performance assessments of measurement procedures [14]. However, the limitations of quality control materials include noncommutability and matrix effect [15]. From this perspective, the observed findings primarily reflect the limitations related to the commutability of control materials rather than an inconsistency between the CLSI EP09 and EP15 guidelines. Therefore, while the CLSI EP15 guideline provides a practical approach by offering opportunities for both imprecision and bias assessment, we recommend performing bias studies using fresh serum, as outlined in the EP09 guideline, when comparing new and old methods/analysers/reagents in order to ensure commutability. We believe that the assignment of different mean values for different instruments/reagents using the same quality control materials also supports our recommendation. In our study, different target values were assigned by manufacturers to the same IQC sample for the reagents used. The manufacturer provided a higher mean value for the low-level IQC material and a lower mean value for the high-level IQC material for the Archem reagent compared to the Abbott reagents [13,16]. However, these values did not fully align with our measured values either.

Relying on the difference between a single test result and a reference or assigned value to determine bias is a misconception in medical laboratories [14]. A single-bias value does not reflect the quality of measurement and it is not recommended for use in calculations such as the sigma metric and measurement uncertainty [14,17]. However, our study demonstrated that in bias calculation, not only the use of single or multiple measurement results matters, but also the type of material used plays a significant role.

The fact that the intercept value does not include 0 within its 95% confidence interval and the slope value does not include 1 within its 95% confidence interval

is interpreted as evidence of a statistically significant difference between the two reagents [18]. However, a pre-established maximum acceptable difference value should be used in the bias assessment [19]. In our study, we used the current desirable bias value from the EFLM BV database, and the difference between all reagents were below this value.

Although the use of 50 patient samples meets the minimum requirements of the CLSI EP09-A3 guideline for method comparison, this sample size may limit the precision of Passing–Bablok regression estimates. Therefore, the width of confidence intervals should be interpreted with this consideration in mind. The primary aim of this study was to support practical laboratory decision-making rather than high-precision method standardization.

In the comparison of TG2 vs. TG conducted by Abbott using the CLSI EP09 guideline, the slope was found to be 0.99 and the intercept 1.41 according to the manufacturer [7]. These slope and intercept values are not in the same direction as our findings. Archem reagent also includes information about a comparison but does not give information about the comparative method [8].

Method changes may be significant when evaluating the results of monitored patients. The significance of the difference between two consecutive results can be determined based on the reference change value (RCV). When calculating the RCV, only the analytical CV and CV_i are used, and bias does not have a place in the formula due to its nature [20]. In the presence of a significant bias between two methods, the difference between two results may exceed the RCV value, potentially leading to incorrect assessments. Therefore, we recommend calculating the bias when method/analyser/reagent changes are made in the laboratory. If the bias is found to be significant, corrective-preventive actions can be implemented within the laboratory. Additionally, for tests like TG and LDL-C, where clinical decision limits are used instead of reference ranges, greater attention should be paid to whether there is a significant bias between methods.

In the part of the study conducted using the CLSI EP15-A3 guideline, the obtained CV% values were higher than the manufacturer's claim of within-run imprecision and the upper verification limit. The likely reason for this could be that the manufacturer's claim is very low. In the EFLM BV database, even the optimal analytical CV is calculated as 4.95%. In the repeatability studies conducted and reported by the manufacturer, the lowest claimed CV is 0.7% for Abbott Triglyceride and the highest is 2.75% for Archem Triglycerides reagent.

Verification continues to be a significant challenge for clinical laboratories [21]; however, careful adherence to CLSI guidelines demonstrates that these processes are

both feasible and essential for reliable method implementation. We believe that, in addition to their clinical significance, tests like TG used in calculations should also be evaluated for the impact of bias between methods on the calculated tests. In our study, LDL-C values were calculated using the Sampson's, Friedewald and Martin-Hopkins equations. Unlike Friedewald's and Sampson's fixed TG denominator, Martin-Hopkins uses a variable factor [3,4]. This likely reduces bias, as the factor increases with TG, minimizing its relative impact on the calculation. In the Sampson equation, the TG coefficient appears to be higher than in the Friedewald formula; however, once the additional variables and constants in the model are taken into account, the actual contribution of TG seems to become attenuated. Changes in the TG assay have the least impact on LDL-C calculations when using the Martin–Hopkins formula, the greatest impact with the Friedewald formula, and an intermediate effect with the Sampson equation. From an analytical perspective, these findings indicate that the Martin–Hopkins equation offers greater robustness against triglyceride-related analytical bias compared with equations using fixed TG denominators. Accordingly, the present analysis is intended to evaluate the analytical propagation of TG bias into calculated LDL-C values, rather than to assess diagnostic reclassification or changes in clinical risk categorization.

This study has limitations. First, the evaluation was conducted in a single laboratory setting, which may limit the generalizability of the findings to other platforms or routine clinical environments. Second, although both patient samples and quality control materials were assessed in accordance with the CLSI EP09-A3 and EP15-A3 recommendations, the number of concentration levels and the study duration were limited to routine verification conditions.

CONCLUSIONS

Bias assessment is crucial when evaluating reagent performance. The fact that IQC results are high relative to the target value does not necessarily indicate a positive bias in patient results, and vice versa. For method changes, we recommend using the EP09 guideline to assess bias rather than relying solely on IQC monitoring. While the Abbott TG reagent met the verification standards and the Abbott TG 2 reagent met them in part, the Archem TG reagent—despite having imprecision values below the optimum CV—did not comply with the manufacturer's imprecision specifications. In addition, an EP15-A3–based verification study should be performed before introducing a new triglyceride reagent into routine use to ensure that precision and bias are acceptable under local laboratory conditions.

ABBREVIATIONS

BV – biological variation
 CLSI – Clinical Laboratory Standards Institute
 CV_A – analytical coefficient of variation
 CV_I – within-subject variation
 HDL-C – High-Density Lipoprotein Cholesterol
 IQC – internal quality control
 LDL-C – Low-Density Lipoprotein Cholesterol
 MS – mean square
 SB – between-run standard deviation
 SR – repeatability
 SWL – within-laboratory imprecision
 TG – Triglyceride
 UVL – Upper Verification Limit
 VB – variation between runs
 VW – within-run variance component

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AUTHORS' CONTRIBUTION

HBV: investigation, data curation, formal analysis, visualization, writing.

ST: investigation, data curation, formal analysis, writing.

All authors have read and approved the final version of the manuscript.

CONFLICT OF INTEREST

None to declare.

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REFERENCES

- Oh RC, Trivette ET, Westerfield KL. Management of Hypertriglyceridemia: Common Questions and Answers. *Am Fam Physician*. 2020 Sep 15;102(6):347-354.
- Romaszko J, Gromadziński L, Buciński A. Friedewald formula may be used to calculate nonHDL-C from LDL-C and TG. *Front Med (Lausanne)*. 2023 Sep 14;10:1247126. DOI: 10.3389/fmed.2023.1247126
- Ginsberg HN, Rosenson RS, Hovingh GK, Letierce A, Samuel R, Poulouin Y, et al. LDL-C calculated by Friedewald, Martin-Hopkins, or NIH equation 2 versus beta-quantification: pooled alirocumab trials. *J Lipid Res*. 2022 Jan;63(1):100148. DOI: 10.1016/j.jlr.2021.100148
- Sampson M, Ling C, Sun Q, Harb R, Ashmaig M, Warnick R, et al. A New Equation for Calculation of Low-Density Lipoprotein Cholesterol in Patients With Normolipidemia and/or Hypertriglyceridemia. *JAMA Cardiol*. 2020 May 1;5(5):540-548. DOI: 10.1001/jamacardio.2020.0013
- Ertürk Zararsız G, Bolat S, Cephe A, Kochan N, Yerlitaş S, Doğan HO, et al. Validation of Friedewald, Martin-Hopkins and Sampson low-density lipoprotein cholesterol equations. *PLoS One*. 2022 May 13;17(5):e0263860. DOI: 10.1371/journal.pone.0263860
- Abbott Triglyceride for use with Architect reagent user manual, Revision: June 2018. Abbott Laboratories Abbott Park, IL 60064 USA.
- Abbott Triglyceride 2 for use with Architect reagent user manual, Revision: March 2021. Abbott Ireland Diagnostics Division Lisnamuck, Longford Co. Longford Ireland.
- Archem Sağlık Sanayi ve Ticaret AŞ. Archem Diagnostics Triglycerides reagent: user manual. Revision February 2022. Istanbul, Turkey.
- Clinical and Laboratory Standards Institute. EP09-A3 Measurement Procedure Comparison and Bias Estimation Using Patient Samples; Approved Guideline- Third Edition. Wayne, PA: CLSI, 2013 .
- Clinical and Laboratory Standards Institute. User verification of precision and estimation of bias; Approved guideline - 3rd ed. CLSI document EP15-A3. Wayne, PA: CLSI, 2014.
- Pum J. A practical guide to validation and verification of analytical methods in the clinical laboratory. *Adv Clin Chem*. 2019;90:215-281. DOI: 10.1016/bs.acc.2019.01.006
- Aarsand AK, Fernandez-Calle P, Webster C, Coskun A, Gonzalez-Lao E, Diaz-Garzon J, et al. The EFLM Biological Variation Database [Internet]. Available from: <https://biologicalvariation.eu/> Accessed November 30, 2024.
- Technopath Clinical Diagnostics Multichem S Plus (Assayed), July 2023. Techno-Path Manufacturing Ltd. Fort Henry Business Park, Ballina Co. Tipperary Ireland.
- Coskun A. Bias in Laboratory Medicine: The Dark Side of the Moon. *Ann Lab Med*. 2024 Jan 1;44(1):6-20. doi: 10.3343/alm.2024.44.1.6. DOI: 10.3343/alm.2024.44.1.6
- Yang X, Chen Q, Pan Z, Cheng J, Zheng W, Liang Y, et al. Application of Patient-Based Real-Time Quality Control Based on Artificial Intelligence Monitoring Platform in Continuously Quality Risk Monitoring of Down Syndrome Serum Screening. *J Clin Lab Anal*. 2024 Mar;38(5):e25019. DOI: 10.1002/jcla.25019
- Archem Sağlık Sanayi ve Tic A.Ş. Multichem S Plus for ARCHITECT Chemistry Systems. Ref No: 05P78-10/11. Technopath Lot No: 13910231-2. Istanbul, Turkey.
- Bozkurt Yavuz H, Karahan S, Yaman H, Örem A, Aliyazıcıoğlu Y. Two approaches for measurement uncertainty estimation: which role for bias? Complete blood count experience. *Turkish Journal of Biochemistry*. 2021;46(6): 639-645. DOI: 10.1515/tjb-2020-0357
- Ozcelik F, Yiginer O, Serdar MA, Kurt İ, Öztosun M, Arslan E, et al. Comparison of three methods for measurement of HbA1c. *Turk J Biochem*. 2010;35(4):344-349.
- Giavarina D. Understanding Bland Altman analysis. *Biochem Med (Zagreb)*. 2015 Jun 5;25(2):141-51. DOI: 10.11613/BM.2015.015
- Fokkema MR, Herrmann Z, Muskiet FA, Moecks J. Reference change values for brain natriuretic peptides revisited. *Clin Chem*. 2006 Aug;52(8):1602-3. DOI: 10.1373/clinchem.2006.069369
- Serdar CC, Cihan M, Yücel D, Serdar MA. Sample size, power and effect size revisited: simplified and practical approaches in pre-clinical, clinical and laboratory studies. *Biochem Med (Zagreb)*. 2021 Feb 15;31(1):0105 DOI: 10.11613/BM.2021.010502

