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ORIGINAL RESEARCH ARTICLE

ASSESSMENT OF ACCURACY AND PRECISION STATISTICS IN ROUTINE BIOCHEMISTRY AUTOANALYZER USING INTERNAL QUALITY CONTROL SPECIMENS IN A TERTIARY CARE HOSPITAL LABORATORY

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ABSTRACT

Background: Accuracy and precision are two important yardsticks of a reliable analytical system in the clinical laboratory. The study was designed to determine the accuracy and precision statistics of a routine biochemistry auto-analyzer using two levels of quality control materials and to compare these statistics with the company provided values. The study also aimed to compare the month-wise variations in these statistics.

Methods: It was a cross-sectional study conducted in the Department of Biochemistry at Birat Medical College Teaching Hospital, Nepal. Laboratory data for the months of May–July, 2021 were retrieved from the laboratory information system (LIS). The retrieved data comprised of the results of two levels of quality control specimens run routinely on Beckman Coulter AU480 biochemistry autoanalyzer for most of the biochemical parameters. Accuracy and precision statistics were calculated as mean and coefficient of variation, respectively.

Results: In both levels of control samples, the laboratory determined accuracy statistics were greater in magnitude than the company provided ones for albumin, alkaline phosphatase, aspartate transaminase, creatinine, unsaturated iron binding capacity, urea, direct bilirubin, and amylase; the precision statistics were similarly greater in magnitude for total protein and magnesium. In month-wise comparison of laboratory determined accuracy statistics, the overall mean differences were statistically significant ($p<0.05$) for all parameters except lactate dehydrogenase and magnesium (both levels of control).

Conclusions: The laboratory determined accuracy and precision statistics showed variations from the company provided ones apart from the month-wise variation. Therefore, continuous monitoring of these values is mandatory for ensuring reliable test reports.

INTRODUCTION

Accuracy and precision, two important benchmarks of an efficient analytical testing system in a reliable clinical laboratory, reflect the closeness of the 'test value' of an analyte to its true value and the similarity in the results of an analyte when analyzed repetitively, respectively.[1, 2]

Although every test is desirable to be 100% accurate and precise, deviations emanate from contributions of various factors inherent to the analytical environment.[1, 3] To address this, every clinical laboratory has to set its own sets of accuracy and precision statistics for the instrument systems and the methods employed for the analysis of the biological samples it receives.[3] Control materials are the reference materials with known analyte concentrations and are justly exploited for the purpose. These materials are commercially available in lyophilized forms or as pre-made solutions; can be procured as the first/second/third party controls, or can also be prepared in the laboratory from the pooled serum samples.[4, 5] The Clinical and Laboratory Standards Institute (CLSI) document EP05-A has clearly directed the precision statistics to be set with at least two levels of control specimens. Moreover, CLSI document EP15-A2 has provided the procedures for any laboratory to confirm the precision claims made by any manufacturing company.[3, 6]

The present study was designed to determine the accuracy and precision statistics in Beckman Coulter (AU 480) biochemistry auto-analyzer using two levels of first-party internal quality control materials and to compare these statistics with those provided by the manufacturing company. The study also aimed to compare the month-wise variations in these statistics.

METHODS

It was a quantitative, retrospective observational study, conducted in the Department of Biochemistry at Birat Medical College and Teaching Hospital, Budhiganga, Morang, Nepal in the months of August–October 2021 (three months). After obtaining ethical clearance from the institutional review committee (IRC) of Birat Medical College and Teaching Hospital (BMCTH) (Ref: IRC-PA-168/2078-79), laboratory data for the months of May, June and July, 2021 were retrieved from the

laboratory information system (LIS).

The overall data in the LIS comprised of all the results of the two levels of quality control specimens run routinely on Beckman Coulter AU480 biochemistry autoanalyzer for all the biochemical parameters. The autoanalyzer employed a closed system of utilization of reagents for the assessment of biochemical parameters, wherein, the regents supplied for the Beckman Coulter AU480 by the same manufacturing company were used. From these, the values obtained from the two levels of control samples run daily (both were run together prior to running the routine biological samples usually during the early hours of the day) in the autoanalyzer for the biochemical analytes albumin, alkaline phosphatase (ALP), alanine transaminase (ALT), aspartate transaminase (AST), calcium, creatinine, cholesterol, phosphate, iron, lactate dehydrogenase (LDH), total bilirubin, total protein, triacylglycerol (TAG), unsaturated iron binding capacity (UIBC), urea, uric acid, direct bilirubin, amylase, and magnesium were included in the study. All the values obtained on repeat run of the control samples on a particular day were excluded, including only the initially obtained values.

A non-probability sampling with total enumeration technique was employed to collect the data. The minimum sample size was ascertained in accordance with the mandates of CLSI document EP05-A2 and EP15-A2. CLSI (EP05-A2) directs the precision to be established with at least two levels of quality control specimens with two runs per day for over 20 days. Similarly, CLSI (EP15-A2) mandates the verification of the precision statistics of the manufacturers by running at least two levels of controls with three replicates for over 5 days.[3] In accordance with the above mentioned document, the minimum sample size for each level of control was calculated to be 40 (2 x 20) considering two runs per day for 20 days.

Next, after obtaining the proper gatekeeper consent from the coordinator of Central Clinical Laboratory, BMCTH, the data was retrieved from the Laboratory Information System (LIS) software of Beckman Coulter AU480. Data comprised of the results of Internal Quality Control (QC) samples run daily on the Beckman Coulter (AU480) Biochemistry Autoanalyzer. The QC samples, available in the commercial lyophilized form were in two levels, level 1 and level 2 packed in separate bottles. Level 1 samples consisted of the standard concentration of the analytes, usually around their reference intervals, whereas, level 2 had the concentrations of the analytes, usually above their reference intervals, but within the detection limit for the analytical system. Before running them in the testing system, each level of QC sample was first mixed with a definitive, prescribed volume of distilled water (as directed in the supplementary document supplied along with the lyophilized control specimen, usually making the final volume of 2 mL) so that the resulting solution obtained was a QC sample with serum matrix. Next, the solution was aliquoted into several equal batches in eppendorf tubes and stored in the refrigerator (at temperature range of 2^0C and 8^0C) for routine use. Every day, the QC samples of both levels in the eppendorf tubes were thawed properly before being fed into the autoanalyzer testing system. Thawing was performed at the room temperature, following the guidelines as mentioned in the supplementary document, ensuring that the analytes did not deteriorate as a result of exceedingly high temperature or violent mixing. The thawed QC sample (in a single eppendorf tube) was used only once. The results obtained were automatically stored in the laboratory information system (LIS) software installed in the autoanalyzer testing system.

The process of data retrieval from the LIS of the autoanalyzer testing system was done daily by the principal investigator, when the autoanalyzer was stalled transiently during its routine operation. The values of three months of QC samples run daily were all retrieved without interfering with the routine testing process. The collected data was entered in a specifically designed proforma.

Collected data was entered first in the software, Microsoft Excel – Microsoft Office (2016). After preliminary data cleaning (e.g., removal of outliers), data entry and analysis were performed in the Statistical Package for Social Sciences (SPSS) software Version 16. First, the data was tested for Gaussian distribution followed by the determination of accuracy and precision statistics for the data not significantly deviated from the normal distribution. The mean values of all the data points collected for each level of control specimen of the biochemical parameters were calculated as the accuracy statistics and were compared with those provided by the manufacturing company. Likewise, standard deviation and coefficient of variation of these data points were calculated as the precision statistics. The results were presented in the form of suitable tables.

RESULTS

The accuracy and precision statistics of the biochemistry autoanalyzer Beckman Coulter AU480 were determined for different biochemical parameters by calculating these statistics from the data points retrieved from the laboratory information system. These data points were the values obtained after the control specimen was analysed daily in the biochemistry autoanalyzer.

Table 1 presents the accuracy and precision statistics of the Beckman Coulter AU480 biochemistry autoanalyzer for two levels of control of different biochemical analytes, depicting the statistics calculated from the laboratory determined values along with the company provided statistics. As shown, for both levels of control specimens, the laboratory determined mean values were greater than the company provided values for the biochemical parameters albumin, ALP, AST, creatinine, UIBC, urea, direct bilirubin, and amylase. Likewise, the mean values were less in the laboratory determined results in both control levels for the parameters cholesterol, phosphate, iron, LDH, TAG, uric acid, and magnesium. The coefficient of variation (CV) of the laboratory determined values were greater than the company provided ones in level 1 controls for the parameters cholesterol, iron, LDH, TAG, UIBC, and magnesium, and in levels 2 controls for total protein and magnesium (Table 1).

Table 1: Accuracy (mean) and precision (standard deviation, SD; and coefficient of variation, CV) statistics of the Beckman Coulter AU480 biochemistry autoanalyzer (as determined in the laboratory with the first party internal quality control specimen and as provided by the manufacturing company) for two levels of controls of different biochemical analytes

Analytes	QC Levels	Laboratory Determined				Company Provided		
		N	Mean	SD	CV	Mean	SD	CV
Albumin (g/dL)	QC-1	86	2.5	0.2	8.3%	2.4	0.3	11.4%
	QC-2	86	4.6	0.3	6.6%	4.5	0.5	11.5%
ALP (U/L)	QC-1	86	126.8	13.4	10.6%	119.0	14.8	12.4%
	QC-2	86	498.0	37.9	7.6%	495.0	62.0	12.5%
ALT (U/L)	QC-1	86	46.0	3.3	7.2%	43.6	5.0	11.5%
	QC-2	86	123.5	7.6	6.2%	124.0	14.4	11.6%
AST (U/L)	QC-1	86	56.0	3.3	5.9%	52.5	6.1	11.5%
	QC-2	86	147.4	10.4	7.1%	147.0	17.0	11.6%
Calcium (mg/dL)	QC-1	86	8.9	0.3	3.9%	8.8	0.5	5.5%
	QC-2	86	12.5	0.4	3.5%	12.5	0.7	5.6%
Creatinine (mg/dL)	QC-1	83	1.4	0.1	4.0%	1.2	0.1	11.3%
	QC-2	83	5.3	0.2	3.5%	5.2	0.5	9.7%
Cholesterol (mg/dL)	QC-1	86	149.8	11.2	7.5%	152.0	10.5	6.9%
	QC-2	86	273.8	18.0	6.6%	282.0	20.0	7.1%
Phosphate (mg/dL)	QC-1	86	6.2	0.4	6.9%	6.6	0.6	9.1%
	QC-2	86	10.9	0.6	5.7%	11.6	1.0	8.6%
Iron (µg/dL)	QC-1	85	68.5	6.0	8.8%	69.8	4.9	7.0%
	QC-2	85	210.6	11.1	5.3%	217.0	15.0	6.9%
LDH (U/L)	QC-1	62	146.9	23.5	16.0%	149.0	13.5	9.1%
	QC-2	61	529.4	45.6	8.6%	546.0	49.0	9.0%
Total Bilirubin (mg/dL)	QC-1	86	1.4	0.1	6.6%	1.5	0.2	13.1%
	QC-2	86	6.7	0.4	5.5%	6.7	0.9	13.0%
Total Protein (g/dL)	QC-1	86	3.9	0.3	7.1%	3.9	0.2	5.6%
	QC-2	85	7.4	0.5	6.4%	7.5	0.4	5.3%
TAG (mg/dL)	QC-1	86	141.7	11.5	8.1%	164.0	13.0	7.9%
	QC-2	86	312.2	20.2	6.5%	330.0	29.5	8.9%
UIBC (µg/dL)	QC-1	75	154.5	21.3	13.8%	141.0	14.0	9.9%
	QC-2	76	311.9	26.9	8.6%	293.0	29.5	10.1%
Urea (mg/dL)	QC-1	86	42.4	2.9	6.8%	40.7	4.5	11.1%
	QC-2	86	178.5	11.5	6.4%	173.0	19.0	11.0%
Uric Acid (mg/dL)	QC-1	85	5.9	0.3	4.4%	6.0	0.4	6.7%
	QC-2	85	8.9	0.3	3.7%	9.2	0.6	6.5%
Direct Bilirubin (mg/dL)	QC-1	86	1.2	0.1	7.1%	1.0	0.2	16.0%
	QC-2	86	5.9	0.4	6.6%	5.8	0.8	13.0%
Amylase (U/L)	QC-1	77	102.2	6.9	6.7%	92.6	9.2	10.0%
	QC-2	77	252.3	14.3	5.7%	235.0	23.8	10.1%
Magnesium (mg/dL)	QC-1	75	2.5	0.3	13.2%	2.6	0.2	8.0%
	QC-2	75	3.8	0.4	11.2%	4.0	0.3	8.0%

Table 2 shows the comparison of accuracy and precision statistics in terms of the difference in mean and coefficient of variation between the laboratory determined and company provided values of these statistics. It also presents the total allowable error limits of these biochemical parameters as per the guidelines set by CLIA (Clinical Laboratory Improvement Amendments), CAP (College of American Pathologists) and AAB (American Association of Bioanalysts). As shown, the bias of mean was more than 10% for the parameters creatinine (level 1; +16.7%), TAG (level 1; –13.6%), direct bilirubin (level 1; +20.0%), and amylase (level 1; +10.4%). Likewise, the bias of mean (in percentage and/or values) were within the limits of total allowable errors (TEa) for all the biochemical parameters, except creatinine (level 1; bias: 0.2 mg/dL or +16.7% which is greater than 15% limit, although less than 0.3 mg/dL limit of TEa), urea (level 2; bias: +5.5 mg/dL or +3.2% which is greater than 4.3 mg/dL limit, although less than 9.0% limit of TEa), and direct bilirubin (bias: +0.2 mg/dL or +20% which is equal to the upper limit of 20%, although less than 0.4 mg/dL limit of the TEa).

Regarding the comparison of the precision statistics, the CV of the laboratory determined values was greater than the company provided ones by more than 5% for the parameters, ALT (level 2: –5.4%), LDH (level 1: +6.9%), and magnesium (level 1: +5.2%). Similarly, the difference in CV was less than 5% for the biochemical analytes, cholesterol (level 1: +0.6%), iron (level 1: +1.8%), total protein (level 1: +1.5%, level 2: +1.1%), TAG (level 1: +0.2%), UIBC (level 1: +3.9%), and magnesium (level 2: +3.2%). For all the other parameters, the precision statistics of the laboratory determined values were less than the company provided values, with the magnitude of the difference being more than 5% for AST (level 1: –5.6%), creatinine (level 1: –7.3%, level 2: –6.2%), total bilirubin (level 1: –6.5%, level 2: –7.5%), and direct bilirubin (level 1: –8.9%, level 2: –6.4%) (Table 2).

Table 2: Comparison of the accuracy and precision statistics (mean and coefficient of variation) between laboratory determined values and those provided by the manufacturing company for two levels of controls of different biochemical analytes

Analytes	QC Levels	Result of analysis Vs Base Value (Mean comparison)			Result of analysis Vs Base Value (CV comparison)
		Difference	Percent	TEa Limit (CLIA)	Difference
Albumin (g/dL)	QC-1	+0.1	+4.2%	±10%	–3.1%
	QC-2	+0.1	+2.2%		–4.9%
ALP (U/L)	QC-1	+7.8	+6.6%	±30%	–1.8%
	QC-2	+3.0	+0.6%		–4.9%
ALT (U/L)	QC-1	+2.4	+5.5%	±20%	–4.3%
	QC-2	–0.5	–0.4%		–5.4%
AST (U/L)	QC-1	+3.5	+6.7%	±20%	–5.6%
	QC-2	+0.4	+0.3%		–4.5%
Calcium (mg/dL)	QC-1	+0.1	+1.1%	± 1mg/dL	–1.6%
	QC-2	+0.0	+0.0%		–2.1%
Creatinine (mg/dL)	QC-1	+0.2	+16.7%	± 0.3mg/dL OR ±15% (CAP)	–7.3%
	QC-2	+0.1	+1.9%		–6.2%
Cholesterol (mg/dL)	QC-1	–2.2	–1.4%	±10%	+0.6%
	QC-2	–8.2	–2.9%		–0.5%
Phosphate (mg/dL)	QC-1	–0.4	–6.1%	±10–23% (CAP)	–2.2%
	QC-2	–0.7	–6.0%		–2.9%
Iron (µg/dL)	QC-1	–1.3	–1.9%	±20%	+1.8%
	QC-2	–6.4	–2.9%		–1.6%
LDH (U/L)	QC-1	–2.1	–1.4%	±20%	+6.9%
	QC-2	–16.6	–3.0%		–0.4%
Total Bilirubin (mg/dL)	QC-1	–0.1	–6.7%	±0.4 mg/dL OR ±20% (CAP)	–6.5%
	QC-2	+0.0	+0.0%		–7.5%
Total Protein (g/dL)	QC-1	+0.0	+0.0%	±10%	+1.5%
	QC-2	–0.1	–1.3%		+1.1%
TAG (mg/dL)	QC-1	–22.3	–13.6%	±25%	+0.2%
	QC-2	–17.8	–5.4%		–2.4%
UIBC (µg/dL)	QC-1	+13.5	+9.6%	±25% (AAB)	+3.9%
	QC-2	+18.9	+6.5%		–1.5%
Urea (mg/dL)	QC-1	+1.7	+4.2%	±4.3 mg/dL OR ±9% (CAP)	–4.3%
	QC-2	+5.5	+3.2%		–4.6%
Uric Acid (mg/dL)	QC-1	–0.1	–1.7%	±17%	–2.3%
	QC-2	–0.3	–3.3%		–2.8%
Direct Bilirubin (mg/dL)	QC-1	+0.2	+20.0%	±0.4 mg/dL OR ±20% (CAP)	–8.9%
	QC-2	+0.1	+1.7%		–6.4%
Amylase (U/L)	QC-1	+9.6	+10.4%	±30%	–3.3%
	QC-2	+17.3	+7.4%		–4.4%
Magnesium (mg/dL)	QC-1	–0.1	–3.8%	±25%	+5.2%
	QC-2	–0.2	–5.0%		+3.2%

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TEa: Total Allowable Error
CLIA: CLIA'88, CLIA (Clinical Laboratory Improvement Amendments)
CAP: College of American Pathologists
AAB: American Association of Bioanalysts

Table 3: Month wise comparison of the accuracy and precision statistics of the Beckman Coulter AU480 biochemistry autoanalyzer as determined with the first party internal quality control specimen for two levels of controls for different biochemical analytes

Analytes	QC Levels	May			June			July			ANOVA	
		N	Mean±SD	CV	N	Mean±SD	CV	N	Mean±SD	CV	F	p-value
Albumin (g/dL)	QC-1	28	2.6±0.2	6.3%	29	2.7±0.1	3.7%	29	2.4±0.2	8.1%	35.99	**<0.001***
	QC-2	28	4.7±0.2	3.8%	29	4.9±0.1	2.9%	29	4.3±0.3	6.2%	50.63	**<0.001***
ALP (U/L)	QC-1	28	126.2±12.4	9.8%	29	135.1±12.4	9.2%	29	119.1±10.5	8.8%	13.40	**<0.001***
	QC-2	28	495.0±44.5	9.0%	29	515.3±29.5	5.7%	29	483.7±32.3	6.7%	5.77	**0.005***
ALT (U/L)	QC-1	28	44.3±2.0	4.5%	29	44.3±1.2	2.7%	29	49.4±3.1	6.3%	49.57	**<0.001***
	QC-2	28	119.8±4.0	3.3%	29	118.8±3.6	3.0%	29	131.7±6.5	4.9%	62.77	**<0.001***
AST (U/L)	QC-1	28	56.9±3.1	5.5%	29	53.9±2.6	4.8%	29	57.4±3.1	5.3%	12.27	**<0.001***
	QC-2	28	148.2±15.0	10.1%	29	142.9±5.5	3.9%	29	152.1±5.9	3.9%	6.49	**0.002***
Calcium (mg/dL)	QC-1	28	8.6±0.4	4.3%	29	9.0±0.3	2.8%	29	9.0±0.3	2.9%	13.62	**<0.001***
	QC-2	28	12.2±0.4	3.2%	29	12.5±0.3	2.6%	29	12.8±0.4	3.2%	16.35	**<0.001***
Creatinine (mg/dL)	QC-1	28	1.4±0.1	2.9%	26	1.4±0.1	3.7%	29	1.4±0.1	4.3%	6.78	**0.002***
	QC-2	28	5.2±0.1	2.5%	26	5.3±0.1	1.9%	29	5.3±0.3	4.9%	3.48	**0.036***
Cholesterol (mg/dL)	QC-1	28	150.5±15.8	10.5%	29	143.9±5.7	3.9%	29	155.1±6.5	4.2%	8.68	**<0.001***
	QC-2	28	274.2±23.5	8.6%	29	262.4±7.3	2.8%	29	284.8±11.7	4.1%	14.91	**<0.001***
Phosphate (mg/dL)	QC-1	28	6.1±0.3	5.1%	29	6.6±0.3	4.9%	29	5.9±0.3	4.8%	41.83	**<0.001***
	QC-2	28	10.7±0.6	5.4%	29	11.4±0.3	2.8%	29	10.5±0.5	4.9%	29.79	**<0.001***
Iron (µg/dL)	QC-1	27	71.0±6.5	9.1%	29	65.4±3.8	5.8%	29	69.2±6.3	9.1%	7.19	**0.001***
	QC-2	27	216.6±7.1	3.3%	29	207.8±7.2	3.5%	29	207.8±14.9	7.2%	6.49	**0.002***
LDH (U/L)	QC-1	16	144.8±6.6	4.6%	18	143.7±34.4	23.9%	28	150.0±21.5	14.3%	0.469	**0.628**
	QC-2	16	536.9±21.2	3.9%	17	524.2±69.2	13.2%	28	528.4±38.5	7.3%	0.330	**0.720**
Total Bilirubin (mg/dL)	QC-1	28	1.4±0.1	7.1%	29	1.4±0.1	6.5%	29	1.5±0.1	2.7%	12.32	**<0.001***
	QC-2	28	6.5±0.4	6.2%	29	6.6±0.2	3.2%	29	6.9±0.3	3.8%	20.52	**<0.001***
Total Protein (g/dL)	QC-1	28	3.9±0.3	8.8%	29	4.1±0.3	6.2%	29	3.9±0.2	4.6%	4.94	**0.009***
	QC-2	28	7.3±0.6	8.1%	28	7.6±0.5	6.0%	29	7.3±0.3	3.6%	4.61	**0.013***
TAG (mg/dL)	QC-1	28	149.3±9.9	6.7%	29	139.9±6.5	4.6%	29	136.2±13.2	9.7%	12.19	**<0.001***
	QC-2	28	329.1±14.2	4.3%	29	310.2±8.7	2.8%	29	297.9±21.5	7.2%	28.38	**<0.001***
UIBC (µg/dL)	QC-1	22	167.9±14.4	8.6%	25	165.1±14.0	8.5%	29	135.0±16.0	11.9%	39.99	**<0.001***
	QC-2	21	332.0±16.5	5.0%	24	326.2±15.2	4.7%	30	286.4±18.6	6.5%	57.17	**<0.001***
Urea (mg/dL)	QC-1	28	44.9±2.2	4.9%	29	41.8±2.4	5.6%	29	40.6±2.2	5.5%	28.14	**<0.001***
	QC-2	28	190.8±5.7	3.0%	29	175.2±8.6	4.9%	29	169.9±7.4	4.4%	61.55	**<0.001***
Uric Acid (mg/dL)	QC-1	27	5.9±0.2	3.7%	29	6.0±0.1	2.3%	29	5.6±0.2	4.3%	26.56	**<0.001***
	QC-2	27	9.1±0.2	2.4%	29	9.1±0.1	1.3%	29	8.6±0.3	3.5%	50.28	**<0.001***
Direct Bilirubin (mg/dL)	QC-1	28	1.1±0.1	7.2%	29	1.2±0.1	8.4%	29	1.1±0.1	3.5%	6.32	**<0.001***
	QC-2	28	5.7±0.4	6.5%	29	6.2±0.2	2.9%	29	5.8±0.4	7.1%	15.26	**<0.001***
Amylase (U/L)	QC-1	27	104.3±6.9	6.6%	29	104.1±6.5	6.2%	21	96.8±4.1	4.2%	11.47	**<0.001***
	QC-2	27	259.4±10.7	4.1%	29	255.8±14.9	5.8%	21	238.4±5.5	2.3%	21.83	**<0.001***
Magnesium (mg/dL)	QC-1	18	2.3±0.4	16.2%	28	2.6±0.4	14.9%	29	2.5±0.2	8.0%	2.416	**0.096**
	QC-2	18	3.7±0.5	13.4%	28	3.8±0.5	12.8%	29	3.8±0.3	7.4%	0.875	**0.421**

*: Statistically significant at 95% confidence interval

Table 3 illustrates the comparison of the accuracy and precision statistics of the Beckman Coulter AU480 biochemistry autoanalyzer for two levels of control specimens across the three months (May, June, and July). As shown, the overall comparison of these statistics across the three months shows the differences in mean values to be statistically significant ($p<0.05$) at 95% confidence intervals (CI), for all biochemical analytes, except for LDH (level 1: $p=0.628$ and level 2: $p=0.720$),

and magnesium (level 1: p=0.096 and level 2: p=0.421).

The CV was maximum during the month of May for parameters, ALP (both levels), AST (both levels), calcium (both levels), cholesterol (both levels), phosphate (both levels), iron (level 1), total bilirubin (both levels), total protein (both levels), amylase (level 1) and magnesium (both levels). During July, the CV was the maximum for analytes, albumin (both levels), ALT (both levels), creatinine (both levels), iron (level 2), TAG (both levels), UIBC (both levels), uric acid (both levels), and direct bilirubin (levels 2). Only for LDH, the CV for both levels of controls were the maximum during the month of June (Table 3).

DISCUSSION

The reports generated form a clinical laboratory play a critical role in decision-making in the health care industry, accounting for approximately 3/5th to 4/5th of such decisions. Although frequencies of inaccuracies in the various stages of the overall laboratory testing course have been found to vary greatly, they indisputably call for a conjoint rigorous mechanism of quality management in the clinical laboratory. The cornerstone for ensuring the accuracy and precision in the analytical process, the quality control encompasses two fundamental stratagems, external and internal quality control systems. Whereas the external quality control system is a set of requirements overseen by an outer organization on a somewhat less regular basis (e.g., monthly, or quarterly), the internal quality control system, on the other hand, embraces the daily and uninterrupted scrutiny of the analytical method to ensure that the laboratory reports can be safely issued to the patient parties.[7]

In our study, we have determined the 'accuracy' and 'precision' statistics of Beckman Coulter (AU 480) biochemistry auto-analyzer using the two levels of first-party internal quality control materials, and compared these statistics with those provided by the manufacturing company. Moreover, we have compared the variations of these statistics between the three months, ie., the duration of data collection for our study. Statistically, accuracy is measured as mean and is monitored as bias or the difference between the mean values of the results of samples of known concentrations, or control specimens obtained in a particular laboratory and the mean values of results of the same type of control specimens obtained in some standard reference laboratories. Likewise, precision is measured and monitored using standard deviation (SD) and coefficient of variation (CV) from the data obtained after repeatedly running these specimens. An accurate analytical system ideally has negligible systematic errors. A precise method is reproducible with trivial random errors.[1, 2]

As per the finding of our study, in both the levels of control samples, the laboratory determined mean values were greater than the company provided values for albumin, ALP, AST, creatinine, UIBC, urea, direct bilirubin, and amylase. This implies the presence of positive bias present in these parameters. Likewise, bias was negative in both levels of controls for the parameters cholesterol, phosphate, iron, LDH, TAG, uric acid, and magnesium. On the other hand, the coefficient of variation (CV) of the laboratory determined values were greater than the company provided ones in both levels of controls for the parameters total protein and magnesium, and in control level 1 for the parameters cholesterol, iron, LDH, TAG, UIBC, and magnesium, with a robust amount of precision for the other remaining parameters.

The bias percentage was greater/less than 10% for the parameters creatinine (level 1), TAG (level 1), direct bilirubin (level 1), and amylase (level 1). Furthermore, this bias was within the limits of total allowable errors (TEa) for all the biochemical parameters, except creatinine (level 1), urea (level 2), and direct bilirubin (level 1). In creatinine (level 1), the bias (+0.2 mg/dL or +16.7%) was greater than 15%, although less than 0.3 mg/dL limit of TEa. For urea, (level 2), the bias (+5.5 mg/dL or +3.2%) was greater than 4.3 mg/dL, although less than 9.0% limit of TEa). Finally, for direct bilirubin (level 1), the bias (+0.2 mg/dL or +20%) was equal to the upper limit of 20%, although less than 0.4 mg/dL limit of the TEa. The total allowable error (TEa) that encompasses the range within which the difference between the measured and true value of any biochemical parameter is permitted. Different parameters have been assigned their respective TEa ranges as the guidelines of Clinical Laboratories Improvement Amendment (CLIA).[7] The values of level 1 controls of creatinine and direct bilirubin and the level 2 control of urea exceeding the range of total allowable error point towards the increased frequencies of large random errors in the analysis of these samples apart from the inherent frailties in their normal analytical performance. In any case, these findings necessitate proper troubleshooting methods of the analytical systems in the form of their regular and rational calibrations, proper change of reagents and controls when deemed necessary, and appropriate control of the physical milieu of the laboratory.

In our study, the difference in CV between laboratory determined results and company provided statistics was more than 5% for ALT (level 2), LDH (level 1), and magnesium (level 1), and less than 5% for cholesterol (level 1), iron (level 1), total protein (levels 1 and 2), TAG (level 1), UIBC (level 1), and magnesium (level 2). Likewise, the difference was less than –5% for AST (level 1), creatinine (levels 1 and 2), total bilirubin (levels 1 and 2), and direct bilirubin (levels 1 and 2). Any such difference in CV less than 5% (let alone less than –5% at the extreme) means a decent steadiness of the results of analysis and points towards the strength in terms of precision of the analytical system. However, the difference in CV of more than 5% as seen for level 1 controls of LDH, and magnesium and level 2 control of ALT also warn about the possible disconcerting variations in the results of analysis. As an appreciable deviation of the analytical system's precision statistics with the control sample from the manufacture-provided data, the results point towards the need for further evaluation of the statistics, e.g., using the third-party controls (instead of just the first party ones). Results from the third party control specimens are less likely to subject to variations which are inherent to the analytic systems and controls from the same manufacturing company. To add, a proper manual handling of the control specimens and

analytical system is also indispensable to obtaining less variant results.

Published studies in this subject line have shown varying outcomes. Coudene and associates, in their study assessed 32 biochemical analytes and based on the NCCLS (National Committee for Clinical Laboratory Standards), determined that the precision statistics fell within the reasonable range of acceptance.[8] Miler et al, in their study comparing the accuracy and precision statistics in two analytical systems, and employing the guidelines from the Croatian Society of Medical Biochemists, reported that the findings did not vary significantly in normal levels control samples. However, in low level controls, the bias was approximately 16.5% for direct bilirubin, clearly exceeding the allowable threshold.[9] Biswas et al, in their study evaluated and compared the accuracy and precision statistics of two analytical systems and found that in both the systems, for most of the biochemical parameters, these statistics were within the acceptable limits, except alkaline phosphatase, and TAG.[10]

As observed in the overall comparison of laboratory determined accuracy statistics across the three months, the differences were statistically significant ($p<0.05$) for all biochemical analytes, except for LDH (levels 1 and 2), and magnesium (levels 1 and 2). Results of analysis of both levels of controls for LDH and magnesium, thus can be said to have consistent values across the three months of study. On the other hand, the remaining biochemical parameters showed considerable month-wise variations indicating the effects of extraneous factors on the analysis of these parameters.

The apparent shortcoming of the study stems from the use of the first-party control specimens for determination of accuracy and precision statistics. As these materials are processed from the same raw materials as the calibrators, their utility to monitor the accuracy and precision of a testing system, already calibrated beforehand, might not yield meaningful information. To add, these types of control materials are also subject to significant variations in response to minute alterations in the methods.[5] A third party control sample or a meticulously prepared pooled serum sample could be a viable alternative option for this.

Another drawback of the study is its study design. In a retrospective chart review such as this wherein the data had to be retrieved from the LIS, it was not possible to take into account all the factors that could have impacted the control results. Various modifiable and non-modifiable factors like milieu of the analytical system, consistency and competency of the technicians performing the analyses, flow of samples in the laboratory and most importantly, the regularity of machine maintenance all could have exerted significant variations in the results of the control samples. To this end, a well-planned prospective study with apt documentations of these factors could address this issue suitably, in addition to helping a clinical laboratory be vigilant.

Overall, the accuracy and precision statistics provide the foundation for analytical quality assessment of the laboratory by helping not only in continuous monitoring of the performance of the analytical system but also into deciding which run should be accepted or rejected based on whether the values of quality control materials lie within or outside a particular limit as governed by set mean, standard deviation and coefficient of variation. By routinely running the control materials, against the backdrop of well-established accuracy and precision statistics and finely calibrated analytical systems, proper diagnosis and subsequent correction of the unsound system can be executed efficiently.[2] No to mention, this also forms one of the integral prerequisites for any laboratory before it can apply for its accreditation.[6]

CONCLUSION

In light of the findings suggesting notable variations between the laboratory determined and the company-provided accuracy and precision statistics apart from the significant month-wise variations in laboratory-determined accuracy statistics, it becomes incumbent on a laboratory to keep track of these statistics on a regular basis. Properly ruling out the random errors and troubleshooting the causes of random as well as systematic errors together call for maintaining a robust account of various factors contributing to the inherent variations in the laboratory analytical milieu in order to ensure accurate and precise results in the real biological samples.

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