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Original Article

Performance Comparison of Two Different Branded Blood Collection Tubes Used in Biochemistry Laboratory

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A B S T R A C T

Background and Aims: Blood collection tubes (BCT) with gel separators are diagnostic devices that can affect all laboratory processes. In this study, the aim was to compare the performance of a new brand of BCT with the sample tubes that are well-known and widely used in the market.

Materials and Methods: Blood samples were taken from 50 healthy volunteers to two brands of BCTs (KWS and BD Vacutainer SST II Advance) by providing standard conditions. The nine most requested test parameters were determined and analyzed simultaneously. The results were analyzed using the SPSS 21 software.

Results: There was no significant difference in the results of most tests of the new brand tubes compared to the Becton-Dickinson brand tube, and the bias calculated for the tests except calcium and potassium was detected to be lower than the desirable bias.

Conclusion: It was determined that the test parameters analyzed in both tubes were in statistical agreement. The percentage of bias value of calcium and potassium tests in KWS branded tubes is higher than the desired value. In addition, constant and proportional errors were determined for the calcium test. These test parameters should be analyzed in larger patient groups. Therefore, KWS tubes can be accepted for routine laboratory operations.

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Introduction

The most important criterion used in evaluating the performance of the clinical biochemistry laboratory is to give the right result from the right patient at the right time in a single analysis. The accuracy of a laboratory test is affected by many factors. The quality of the result is mostly affected by the factors belonging to the pre-analytical period [1]. Taking the blood samples into the appropriate container with the appropriate technique, transferring them to the laboratory, and the preparatory procedures made before the analysis and storage conditions of samples are the main steps of the pre-analytical phase. The number of studies evaluating the pre-analytical phase is very few compared to the analytical phase. However, since blood collection tubes (BCT) are accepted as an *in vitro* diagnostic device, performance evaluation should be performed for test result quality. Plastic serum separator tubes (SSTs) are mostly used in routine analyses. SSTs include a gel barrier moving to the serum/clot interface [2].

Ideally desired in SST is the rapid formation of clots, a clear separation from the serum after centrifugation, not detecting gel and fibrin particles in the serum, and not mixing the separated serum with the clot again [3]. Spontaneous and complete clotting occurs within 30 to 60 minutes at room temperature (20 to 25 °C). Blood specimens for serum samples should be adequately clotted before centrifugation. Storage conditions are significant to ensure the stability of specific analytes. At 2 to 8 °C, significant stability of separated serum has been observed for 2 to 14 days [4]. Since different manufacturers produce plastic tubes by providing components such as a surfactant, stopper, separating gel, and clot activator in different ways and with different materials, the performance evaluation of the tubes must be performed.

This study aimed to compare the performance of KWS (Shijiazhuang Kang Weishi Medical Instrument Co, LTD, China) branded tubes with the Becton-Dickinson Vacutainer SST II Advance (Becton Dickinson and Company, USA) tubes that are well-known and widely used in the market. Clinical and Laboratory Standards Institute (CLSI) document EP28A3c directives were followed for collecting, processing, and storing samples in the laboratory [5]. When technical and clinically valid indicators for local validation of the European Federation of Clinical Chemistry and Laboratory Medicine (EFLM), the Working Group for Preanalytical Phase (WG-PRE) proposed by the consensus protocol were used (6).

Materials and Methods

The CLSI-EP28-A3c guide was used for consideration of pre-analytical factors. According to this guide, subject preparation, specimen collection, and specimen handling conditions are determined. The guideline "EFLM WG-Preanalytical phase opinion paper: local validation of blood collection tubes in clinical laboratories" published by the EFLM recommends using at least 20 blood samples in such comparison studies. Accordingly, the study included 50 randomly selected healthy

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volunteers, aged 18-70, without any diagnosed disease and not using regular medication. To compare different levels of test parameters, 12 hours of fasting condition was not set. Venous blood samples were taken from the antecubital vein using a vacuum tube holder (sitting position) and a 21G needle into two branded vacuum sample tubes containing a gel separator. A tourniquet was applied during the phlebotomy, and the tourniquet time was less than 1 minute. The volume of a blood sample taken into each tube was 5 ml. The same brand needle and tube holder were used for all blood collection procedures, and the tube sequence was randomly arranged in the collection process. All samples were transported to the laboratory by the staff in a vertical position in the carrier containers and sample carrying cases. The sample tubes were kept at room temperature for 30 minutes and centrifuged at 4000 rpm for 15 minutes. These tubes were primarily evaluated for the quality indicators specified in the CLSI and EFLM guidelines. These criteria are [6]; Finding deformity arising from production in the tubes, insufficient filling due to inadequate tube vacuum effect, incompatibility between the tube and blood collection adapter, sample leakage in tubes due to reasons such as breaking after centrifuge and opening the tube cap, hemolysis, clot, gel residue, foreign material in the centrifuged tube. Tubes with any of these criteria were excluded.

Subsequently, the nine test parameters (glucose, creatinine, calcium (Ca^{+2}), triglyceride, aspartate aminotransferase (AST), potassium (K^{+}), thyroid stimulating hormone (TSH), free T4 (FT4), ferritin) and "hemolysis icter lipemia index" were analyzed simultaneously. The selected tests are the most requested ones according to the last year's data. Samples with hemolysis icter lipemia index results outside the acceptable range were not included in the study. The analyzes were performed with Abbott Architect c16000 (Abbott Diagnostic, Lake Forest, IL, USA) and Siemens Advia Centaur XP (Siemens Healthcare Diagnostics, Munich, Germany) analyzers, whose internal quality control studies were appropriate. All studies were performed with the same lot number kits to prevent Lot to Lot variation. Deviation values measured in the tests' internal and external quality programs were within acceptable limits. The brands, lot numbers, and code names used in the study of BCTs are as follows; BD Vacutainer SST II Advance, Becton Dickinson and Company Made in the UK, Lot: 9171639 (Coded as BD for analyzes), and KWS, Shijiazhuang Kang Weishi Medical Instrument Co., LTD Lot: 20190602, Made in PRC (Coded as KWS for analyses). The Ethical Committee and Institutional Review Board of Usak University Faculty of Medicine, where the study was conducted, approved the study design (22.11.2019; approval number: 238-05). This study was conducted by the principles of the Helsinki Declaration.

Statistical Analysis

Statistical analyses were performed using the SPSS software version 21. Then, the assay results were named BD and KWS, and the Kolmogorov-Smirnov test was used to check the normality of all variables. Mean and standard deviation were calculated for variables

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with normal distribution, and median and interquartile range values were calculated for those with the abnormal distribution. Since the test results were not normally disturbed, non-parametric tests were conducted to compare these parameters. For each test parameter, pairwise comparisons were performed with the Mann–Whitney U test. A p-value of less than 0.05 was considered to show a statistically significant result. Bland-Altman Plots and Passing-Bablok Regression analysis were used for each test parameter to evaluate the association. In addition, BD results were accepted as the reference value (Vr), and the Bias(%) values were calculated with the formula "% Bias= Measured value-Vr/ Vr x100". These values are compared with the desirable inaccuracy specification (B%) values calculated by Ricos et al. and published on the Westgard QC website (www.westgard.com) [7].

Results

The gender distribution of the sampled volunteers is 36 males and 14 females, and the average age was calculated as 44.8 ± 13.5 (min:20, max:67). The descriptive statistical data of all test parameters according to tube brands are shown in Table 1. Only AST results of KWS were found to be significantly different from BD ($p < 0.001$). There was no significant difference between the results of the other 8 test parameters. Bland Altman Plots indicate the compatibility of BD results with KWS results in pairwise comparisons (Figure 1). The horizontal axis consists of the mean values of the BD and KWS measurement results, assuming a constant error rate in the BD results. The vertical axis shows the ratio of the two measurement results to the average value. The ± 1.96 SD lines in the graphs represent the 95% confidence interval (%95 CI). The Mean line represents the average of the differences in between.

The fact that this value is close to zero indicates that the results agree. Accordingly, the average difference value for the AST test in the KWS tube is higher than other tests (3%). However, it is seen in Figure 1 that only 2 results for the AST test are outside the 95% confidence interval.

In addition to the Bland-Altman plots, Passing-Bablok Regression analysis was applied to the obtained data. Accordingly, when the regression equation ($y = a + bx$) of each test is examined, it is observed that the constant error (a) and the proportional error (b) are negligible values in all tests except calcium, and the Spearman correlation coefficients are close to 1. For Ca^{+2} test, "y = 1.187 + 0.875x" for the KWS results. This equation determines a constant and proportional error for the calcium test. The correlation constant of the Ca^{+2} test was also calculated as the lowest value among all the tests (0.854). Studies with larger populations and different calcium levels may be required.

Table 2 visualizes the comparison of the calculated bias with the desirable bias. Ca^{+2} Bias% (1.59%) and K^{+} Bias% (1.82) are higher than B%.

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Table 1. Descriptive statistical data and comparison of mean values by Mann-Whitney U test

	BD		KWS		Mann-Whitney U test
	Median	**IQR****	**Median**	**IQR**	**(p-values)**
Aspartate aminotransferase (IU/L)	17.5	15-21	17.0	14.0-20.0	*0.000*
Creatinine (mg/dL)	0.89	0.79-1.02	0.89	0.79-1.03	*0.139*
Potassium (mEq/L)	4.2	4.1-4.4	4.2	4.1-4.4	*0.537*
Trigliceryde (mg/dL)	146.5	113-226	147	112-228	*0.526*
Calcium (mg/dL)	9.3	9.1-9.6	9.4	9.2-9.6	*0.128*
Thyroid stimulating hormone (mIU/L)	1.54	1.0-2.0	1.54	0.98-1.98	*0.472*
Free T4 (ng/dL)	1.13	1.03-1.21	1.12	0.99-1.18	*0.238*
Ferritin (ug/L)	48.6	29.9-80.1	47.5	29.1-82.2	*0.270*
Glucose (mg/dL)	97	87-112	97	88-114	*0.201*

*BD: Measurement results of BD brand tubes, KWS: Measurement results of KWS brand tubes
** Inter Quartile Range

Table 2. Comparison of Desirable Bias (B%) and Bias% values

	Desirable Bias	**Bias* BD vs. KWS**
Aspartate aminotransferase (IU/L)	6,50	4,06
Glucose (mg/dL)	2,34	1,84
Creatinine (mg/dL)	3,96	1,53
Potassium (mEq/L)	1,81	1,82
Triglyceride (mg/dL)	9,57	2,17
Calcium (mg/dL)	0,82	**1,59**
Thyroid stimulating hormone (mIU/L)	7,8	2,36
Free T4 (ng/dL)	3,3	3,16
Ferritin (ng/mL)	5,2	2,14

* ([Avg KWS - AvgBD]/AvgBD)x100

Discussion

The most common sample used in the biochemistry laboratory is serum. There are many BCT brands available in the market with different separation methods. The most commonly used barrier material is separator gels. A new BCT brand intended for use in the laboratory should be subject to verification studies to maintain the reliability of test results. Pre-analytical errors account for 70% of all laboratory errors. Sources of error in the pre-analytical phase are very diverse and often unavoidable [8]. Errors that may arise from blood collection tubes produce predictable and preventable results in the pre-analytical stage. The components of the blood collection tube should not interact with the analytes present in the blood and should not affect the accuracy of the result.

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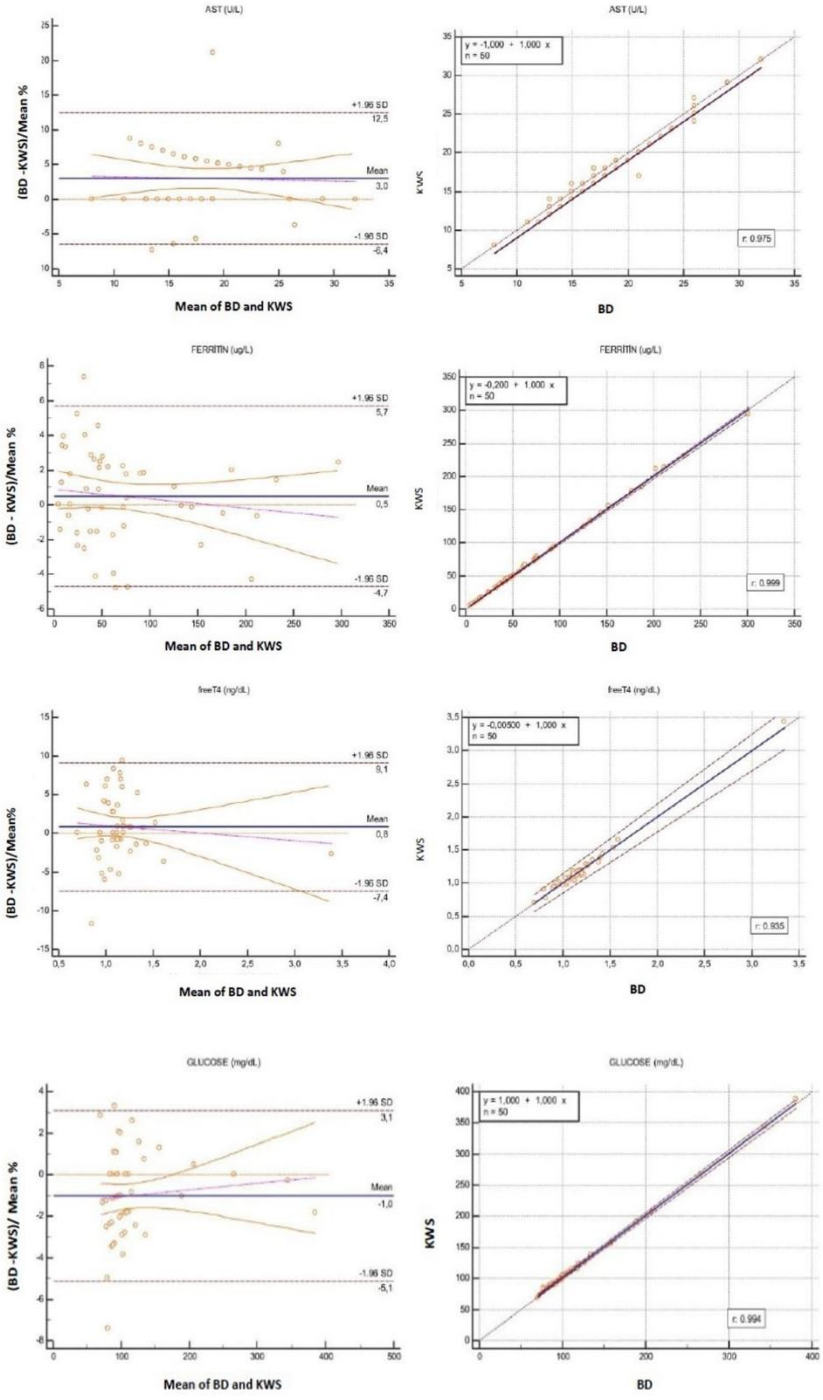

Fig. 1. Bland-Altman difference plots and Passing-Bablok regression analysis for the nine biochemical analytes obtained with Becton-Dickinson SST II Advance (BD), KWS (KWS) Tubes

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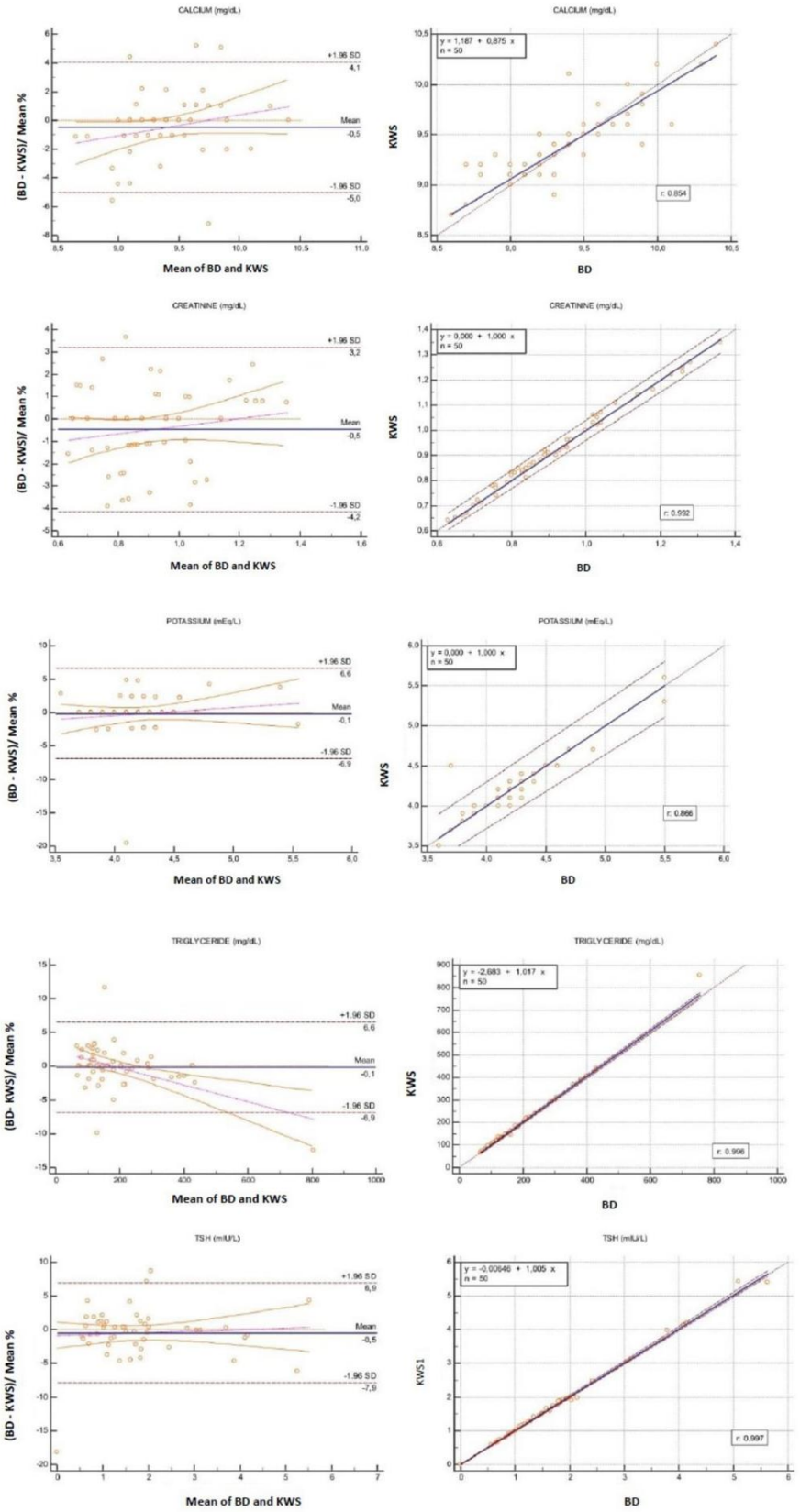

Fig. 1. Bland-Altman difference plots and Passing-Bablok regression analysis for the nine biochemical analytes obtained with Becton-Dickinson SST II Advance (BD), KWS (KWS) Tubes (Continued)

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Laboratory professionals can use the CLSI-EP28-A3c (5) and EFLM-WG-PRE [6] guidelines for local technical validation. These guidelines can give an idea of whether the tube brand evaluated meets the quality requirements of the current lot-numbered product line.

This study aimed to compare two different branded tubes, one is newly produced (KWS, Shijiazhuang Kang Weishi Medical Instrument Co., LTD, China), and the other is widely used worldwide (Becton Dickinson SST II Advance, USA). The comparison study analyzed the nine most requested test parameters in blood samples. According to the comparison results, compatible results were obtained between the tube brands. Although there is a significant difference in the Mann-Whitney U test for the AST test, the median values of the two groups are close to each other (17.5 IU/L and 17 IU/L). The Bias values of serum Ca^{+2} and K^{+} tests are higher than the expected bias. Furthermore, the correlation coefficient of the calcium test is the lowest in the Passing-Bablok analysis. It has been concluded that analysis with larger sample groups is necessary for the calcium test.

The values used in the Bias calculation were calculated as the average of only two measurements for each test parameter. Calculating the mean value with more within-run measurements would be more accurate. It can be considered a limitation of the bias comparison in the study.

Conclusion

Physical examination of the KWS brand tubes revealed no problem with the analyzed lot number. With statistical comparisons, acceptable results were obtained except for Ca^{+2} and K^{+} tests. If monitoring continues for these two tests, KWS tubes are admissible for the routine process. Such verification studies should be carried out regularly with tube series with different lot numbers. In this way, the stability of the production quality will be tested.

Conflict of Interest

The authors have no conflict of interest.

Acknowledgement

Not applicable.

References

[1]. Lippi G, Chance JJ, Church S, Dazzi P, Fontana R, Giavarina D, et al. Pre-analytical quality improvement from dream to reality. Clin Chem Lab Med. 2011; 49(7): 1113-126.

[2]. Bowen R, Kim S, Sattayapiwat A, Esguerra V, Zare R. Performance of chemically modified plastic blood collection tubes. Clinical Biochemistry 2016; 49: 90-99.

[3]. Landt M, Wilhite TR, Smith CH. A new plastic evacuated tube with seperator. J Clin Lab Anal 1995; 9(2): 101-106.

[4]. Clinical and Laboratory Standards Institute (CLSI) document GP44-A4 Procedures for the Handling and Processing of Blood Specimens for Common Laboratory Tests; Approved Guideline, 4th ed, 2010; p. 30.

[5]. Clinical and Laboratory Standards Institute (CLSI) document EP28-A3c. Defining, Establishing, and Verifying Reference Intervals in the Clinical Laboratory; Approved Guideline, 3th ed, 2010; p. 28.

[DOR: 20.1001.1.24233706.2022.9.3.8.2]

[DOI: 10.18502/ijml.v9i3.10906]

[6]. Lippi G, Cornes MP, Grankvist K, Nybo M, Simundic AM, on behalf of the Working Group for Preanalytical Phase (WG-PRE), European Federation of Clinical Chemistry and Laboratory Medicine (EFLM). EFLM WG-Preanalytical phase opinion paper: local validation of blood collection tubes in clinical laboratories. Clin Chem Lab Med 2016; 54(5): 755-60.

[7]. Ricos C, Alvarez V, Cava F, Garcia-Lario JV, Hernandez A, Jimenez CV, et al. Current databases on biologic variation: pros, cons and progress. Scand J Clin Lab Invest. 1999; 59: 491-500.

[8]. Lippi G, Chance JJ, Church S, Dazzi P, Fontana R, Giavarina D, et al. Pre-analytical quality improvement: from dream to reality. Clin Chem Lab Med. 2011; 49(7): 1113-126.

[DOR: 20.1001.1.24233706.2022.9.3.8.2]

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