Barw Medical Journal | 2024;2(3):20-25
Journal home page: https://barw.krd/index.php/BMJ

Original Article

Variation in Parameters of Complete Blood Count Using Ethylene Diamine Tetra Acetic Acid (EDTA), Sodium Citrate and Lithium Heparin: A Cross-Sectional Study

Sharaza Qadir Omer[1], Rawezh Q. Salih[2*], Shvan Luqman Ezzat[2], Dahat A. Hussein[2], Ayman M. Mustafa[1], Aga Khdir Hamad[3], Hawnaz Salih Abdullah[2], Bayan Aziz Saeed[2], Shahid Salah Rashid[4], Sangar Jum'a[5], Mohammed Hassan Sofi[2]

1. *Smart Health Tower, Madam Mitterrand Street, Sulaymaniyah, Kurdistan, Iraq*
2. *Department of Biology, College of Science, University of Sulaimani, Sulaymaniyah, Kurdistan, Iraq*
3. *Medical Laboratory Science, College of Health Science, University of Duhok, Duhok, Kurdistan, Iraq*
4. *Department of Biology, College of Science, Koya University, Erbil, Kurdistan, Iraq*
5. *Department of Chemistry, College of Science, University of Sulaimani, Sulaymaniyah, Kurdistan, Iraq*

[*] ***Corresponding author***: Rawezh.mohammed@univsul.edu.iq *(R.Q. Salih). Azadi Street, House no. 8, Zip code: 46001 Sulaymaniyah, Kurdistan, Iraq*

Keywords:
Complete blood count
EDTA
Sodium citrate
Lithium heparin
Parameters

Received: April 7, 2024
Revised: May 26, 2024
Accepted: June 10, 2024
First Published: June 20, 2024

Citation: Omer SQ, Salih RQ, Ezzat SL, Hussein DA, Mustafa AM, Hamad AK, et al. Variation in Parameters of Complete Blood Count Using Ethylene Diamine Tetra Acetic Acid (EDTA), Sodium Citrate and Lithium Heparin: A Cross-Sectional Study. Barw Medical Journal. 2024;2(3):20-25.
https://doi.org/10.58742/bmj.v2i2.90

Abstract

Introduction

The primary anticoagulants utilized in hematology are ethylene diamine tetra acetic acid (EDTA), lithium heparin, and citrate. Although various studies have investigated their impact on complete blood count (CBC) in various animals, limited data are available concerning humans. This study explores the differences in CBC parameters when using these anticoagulants.

Methods

This study was a cross-sectional study comprising 250 participants who underwent CBC tests with different anticoagulants. Blood samples were examined using the Medonic M51. Statistical analyses, including one-way ANOVA, intra-class correlation coefficient, and concordance correlation coefficient, were employed as applicable, with significance level set at p-values <0.05.

Results

One hundred eight participants (43.2%) were male, and 142 (56.8%) were female, with a mean age of 40.88±17.06 years ranging from 7 to 91 years. Comparing K2EDTA with lithium heparin, comparable values were found in 14 out of 23(60.87%) CBC parameters, while citrate showed similar results in 13(56.52%) parameters. Using K2EDTA as the standard, citrate showed perfect or substantial agreement in assessing 9 out of 23 CBC parameters (39.13%). Similarly, Lithium heparin showed perfect or substantial agreement in determining 9 out of 23(39.13%) parameters. Compared to K2EDTA, lithium heparin exhibited high precision and accuracy in estimating 13 out of 23(56.52%) CBC parameters. In contrast, citrate was accurate in 9 out of 23(39.13%) parameters.

Conclusion

Using citrate instead of K2EDTA for CBC estimation may yield inaccurate outcomes, whereas lithium heparin could serve as an alternative anticoagulant, requiring careful monitoring.

1. Introduction

The complete blood count (CBC) is a commonly conducted test in medical labs involving the analysis of red blood cells (RBCs), white blood cells (WBCs), and platelets (PLTs), along with hemoglobin (HGB) concentration and cell size. These parameters play a role in the diagnosis and monitoring of various health conditions. Hence, precise measurement is essential for effective patient care [1]. It serves as a diagnostic tool to assess human health, helping to detect congenital abnormalities and functional changes caused by exposure to potential diseases. Additionally, CBC results can indicate various health conditions, such as infections with elevated WBCs, leukemia with abnormal WBC counts, anemia with low HGB levels, and liver cirrhosis with reduced PLT counts [2].

Various factors significantly influence the outcomes of hematological analyses, particularly CBC results. These factors include the method and procedure used for sampling and collection of blood, techniques employed, storage conditions of the samples, and the selection of anticoagulants. Each of these elements can have a notable impact on the accuracy and reliability of the CBC findings [3]. Thoughtful consideration in choosing the appropriate anticoagulant is essential because of its notable impact on the outcomes of CBC tests. In the field of clinical hematology, a range of anticoagulants is employed for both the collection of blood samples and standard laboratory analyses. Some of the frequently utilized anticoagulants include the sodium and potassium salts of ethylene diamine tetra acetic acid (EDTA), citrate, sodium and lithium heparin, as well as oxalates [4].

The National Committee for Clinical Laboratory Standards has endorsed EDTA for CBC tests primarily due to its ability to preserve cellular structures effectively. Additionally, they recommend the use of dipotassium EDTA salt over other sodium and potassium salts of EDTA as the anticoagulant of choice for CBC tests [5]. In addition to EDTA, other anticoagulants like sodium citrate and heparin are utilized for CBC tests due to their ability to slow or prevent the clotting of blood [6].

Most existing genuine literature primarily examines various anticoagulant effects on CBC results or its components across various animal species [7]. The current study, however, seeks to assess differences in CBC parameters among humans by using different anticoagulants, dipotassium EDTA (K2EDTA), sodium citrate, and lithium heparin, to evaluate their efficacy.

2. Methods

2.1 Study design and population

This is a multicentered prospective study that included children diagnosed with viral meningitis who visited pediatric clinics throughout Sulaimani, Halabja, Kalar, and Ranya from May 2023 to June 2023. The ethics committee of the College of Medicine at the University of Sulaimani granted ethical approval (Decree No. 45).

2.2 Eligible criteria

The study focused on patients visiting clinics for various health conditions, including both genders. Those who declined to give consent were not included in the study.

2.3 Determination of the Sample Size

The sample size was calculated using G*Power statistic 3.1.9.7, employing linear multiple regression with a two-tailed approach. With an effective sample size of 0.35, α error probability of 0.01, a statistical power of 0.99, and a predictor value of 1, the minimum sample size required was determined to be 158. Hence, a sample size of 250 was used for each comparison of CBC parameters among the three anticoagulants.

2.4 Sample and data collection

Trained healthcare professionals conducted data collection and obtained blood samples from participants. Blood samples were drawn using a sterile syringe and needle, extracting 5 millilitres (5ml) from either the median cubital vein or a prominent forearm vein. Subsequently, 1.8 ml of the whole blood was transferred into a sodium citrate tube, while 1.6 ml was dispensed into tubes containing K2EDTA and lithium heparin. The tubes were gently mixed by inverting them approximately seven times to ensure thorough mixing of blood and anticoagulants.

Each participant's CBC was examined using the Medonic M51 automated hematology analyzer from Boule Medical AB. This analysis occurred within 3 to 6 hours of sample collection. Tube specifications are outlined (Table 1). The

Table 1: Characteristics of Laboratory Test Tubes Employed for CBC Assessment.

Tube details	EDTA	Heparin	Citrate
Type of tube	K2EDTA	Lithium (vacuum blood collection tube)	PT Tube (Sodium citrate)
Dimension	13 x 75 mm		13 x 75 mm
Storage	5- 25°C	13 x 75 mm	5-25°C
Year of expiry	31-3-2025	5-25°C	19-12-2025
Tube capacity (volume)	5 ml	24-11-2027	5ml
Required volume	3 ml		1.8ml
Tube material	Plastic	5 ml	glass
Manufacturer	Vacutest kima sri	3ml	MR+
Origin/country	Italy	glass	China
Anticoagulant type & concentration	5.4 mg	MR+	3.2%

evaluation encompassed diverse hematological parameters, including WBC, %Neutrophil(%Neu), %Lymphocyte(%Lymph), %Monocyte(%Mon), %Eosinophil(%Eos), %Basophil(%Bas), Neutrophil (Neu), Lymphocyte (Lymph0, Monocyte (Mon),

Eosinophil (Eos), Bas, RBC, HGB, Hematocrit (HCT), Mean Corpuscular Volume (MCV), Mean Corpuscular Hemoglobin Concentration (MCHC), Red cell Distribution width (RDW-SD), PLT, Mean Platelet volume, Platelet Distribution Width (PDW), Platelet Crit (PCT), and Platelet Larger Cell Ratio. Additionally, age and gender data were collected for each participant.

2.5 Statistical Analysis

Before statistical analysis, the collected data underwent initial processing. Initially, it was inputted into Microsoft Excel 2019. Subsequently, rigorous checks were performed to ensure both consistency and completeness. Once validated, the data were transferred to Statistical Package for the Social Sciences software (SPSS) 25.0 and MedCalc version 20 for further statistical analysis.

The consistency of results obtained from the three anticoagulants was assessed using Intra-class correlation coefficient (ICC) analysis. ICC values were interpreted as follows: below 0.50, indicating poor consistency; between 0.50 and 0.75, indicating moderate consistency; between 0.75 and 0.90, indicating good consistency; and above 0.90, indicating excellent consistency. Furthermore, a significance level of $p < 0.05$ was considered for all statistical comparisons. The study utilized the one-way ANOVA test to examine differences among CBC parameters within and between samples collected using K2EDTA, sodium citrate, and lithium heparin.

The concordance correlation coefficient (CCC) was used to assess the agreement among the anticoagulants, with K2EDTA as the reference standard. CCC values were interpreted as follows: 0.99 or higher indicated almost perfect agreement, 0.95 to 0.99 suggested significant agreement, 0.90 to 0.95 indicated moderate agreement, and below 0.90 indicated poor agreement [8].

3. Results

One hundred eight participants (43.2%) were male, and 142 (56.8%) were female. The average age of these participants was 40.88 ± 17.06 years (7-91). Concerning the consistency of CBC results with lithium heparin, K2EDTA, and sodium citrate, remarkable consistency was observed in estimating WBC, %Neu, %Lymph, %Eos, Neu, Eos, RBC, HGB, HCT, MCV, MCH, RDW-SD, and PDW (Table 2).

In the comparison among K2EDTA, sodium citrate, and lithium heparin for estimating variation in CBC parameters, no significant differences were observed in median values for WBC, %Lymph, %Eos, Neu, Lymph, Eos, MCV, MCH, and RDW-SD across these anticoagulants (Table 3).

Similar results were observed when examining variations in CBC parameter estimation using different anticoagulated blood samples. For K2EDTA-sodium citrate, median values for WBC, %Neu, %Lymph, %Eos, %Bas, Neu, Lymph, Eos, Bas, MCV, MCH, MCHC, and PDW were comparable with a p-value ≥ 0.05. Likewise, K2EDTA-lithium heparin comparisons showed similar results for median WBC, %Lymph, %Eos, Neu, Lymph, Eos, RBC, HGB, HCT, MCV, MCH, RDW-SD, PDW, and Platelet Larger Cell Ratio with a p-value ≥ 0.05. Comparing sodium citrate-lithium heparin results indicated nonsignificant differences in median WBC, %Lymph, %Eos, Lymph, Eos, MCV, MCH, RDW-SD, PLT, and PCT (Table 4)

When compared to K2EDTA as the standard, sodium citrate displayed perfect agreement in assessing %Lymph, MCV, and MCH (CCC = 0.990), with significant agreement in determining WBC, %Neu, %Eos, Neu, Lymph, and Eos (CCC between 0.95 and 0.99). Conversely, lithium heparin showed perfect agreement in determining RBC (CCC=0.994), HCT (CCC= 0.993), MCV (CCC=0.995), and MCH (CCC=0.994), with substantial agreement in other parameters such as WBC, %Lymph, Neu, HGB, and RDW-SD (CCC between 0.95 and 0.99). Poor agreement was observed in other parameters with CCC<0.90. Comparatively, when compared with K2EDTA, heparin demonstrated high precision and accuracy in estimating 13 out of 23(56.52%) CBC parameters namely WBC, %Neu, %Lymph, %Eos, Neu, Eos, RBC, HGB, HCT, MCV, MCH, MCHC, RDW-SD, while citrate was highly precise and accurate in 9 out of 23(39.13%) parameters (Table 5).

Table 2. Consistency of CBC Findings with K2EDTA, Lithium Heparin, and Sodium Citrate

CBC parameters	Intra-class correlation coefficient	Confidence interval 95%	
		Lower	Upper
WBC	0.991	0.975	0.996
%Neu	0.969	0.908	0.985
%Lymph	0.988	0.986	0.991
%Mon	0.573	0.147	0.762
%Eos	0.979	0.969	0.985
%Bas	0.810	0.752	0.854
Neu	0.990	0.976	0.995
Lymph	0.810	0.764	0.847
Mon	0.599	0.237	0.766
Eos	0.979	0.974	0.984
Bas	0.626	0.537	0.701
RBC	0.926	0.281	0.978
HGB	0.934	0.313	0.981
HCT	0.925	0.274	0.978
MCV	0.999	0.996	0.999
MCH	0.998	0.995	0.999
MCHC	0.911	0.877	0.934
RDW-SD	0.965	0.953	0.974
PLT	0.713	0.136	0.876
MPV	0.934	0.878	0.960
PDW	0.942	0.909	0.961
PCT	0.660	0.081	0.847
PLCR	0.946	0.897	0.968

Table 3. Differences in CBC Parameters with Various Anticoagulants.

CBC parameters	Lithium Heparin Median (Min-Max)	Sodium Citrate Median (Min-Max)	K2EDTA Median (Min-Max)	P-value
WBC	7.91(0.27-25.27)	7.48(0.20-21.92)	8.02(0.25-23.95)	0.063
%Neu	62.90(31.70-95.90)	59.55(27.00-93.40)	59(25.80-93.20)	<0.001
%Lymph	30.70(3.00-61.50)	31.20(3.40-63.80)	31.70(3.50-66.80)	0.736
%Mon	1.70(0.00-17.70)	5.80(0.20-11.60)	6.50(0.40-13.10)	<0.001
%Eos	2.30(0.00-13.70)	2.50(0.10-13.10)	2.10(0.10-13.90)	0.095
%Bas	0.70(0.10-1.60)	0.50(0.10-1.60)	0.50(0.10-1.60)	<0.001
Neu	4.78(1.70-22.48)	4.35(1.64-19.66)	4.62(1.81-21.78)	0.070
Lymph	2.40(0.46-20.20)	2.27(0.45-7.08)	2.45(052-8.01)	0.088
Mon	0.14(0.00-1.28)	0.39(0.02-2.20)	0.48(0.03-1.89)	<0.001
Eos	0.18(0.00-1.21)	0.18(0.01-1.09)	0.16(0.01-1.22)	0.432
Bas	0.05(0.01-0.18)	0.04(0.01-0.50)	0.04(0.01-0.11)	0.001
RBC	4.97(3.03-6.82)	4.50(2.80-6.14)	4.97(3.18-6.83)	<0.001
HGB	13.60(7.20-18.20)	12.20(6.20-16.70)	13.60(6.80-18.10)	<0.001
HCT	41.25(23.80-52.90)	37.30(21.30-51.00)	41.5(22.90-53.60)	<0.001
MCV	84.25(49.40-98.40)	84(50.10-98.20)	84.70(49.40-99.40)	0.674
MCH	28.10(14.90-31.70)	27.80(15.40-31.40)	27.90(14.80-32.00)	0.556
MCHC	33.25(27.60-35.60)	33(22.10-34.90)	32.90(27.10-35.90)	0.001
RDW-SD	43.80(35.50-65.60)	43.50(10.20-65.60)	44.25(35.90-66.70)	0.069
PLT	169(18-506)	175(17-487)	260(23-588)	<0.001
MPV	9.30(7.20-11.20)	8.90(7.10-11.10)	9.10(7.30-11.10)	<0.001
PDW	12(8.20-21.70)	11.20(7.60-20.60)	11.60(8.10-20.00)	<0.001
PCT	0.16(0.02-0.38)	0.16(0.02-0.39)	0.24(0.02-0.52)	<0.001
PLCR	31.80(0.90-52.80)	28.90(16.00-50.00)	30.45(16.60-48.20)	<0.001

4. Discussion

The choice of anticoagulants and the duration of sample storage are pivotal factors in laboratory blood sample analysis. It's widely acknowledged that these variables significantly impact blood parameters independently [9]. In a study involving 55 healthy individuals, it was shown that there was strong consistency in estimating 3 out of 14(21.43%) CBC parameters, %Lymph, MCV, MCH across three different anticoagulants: K3EDTA, sodium citrate, and lithium heparin [3]. The current study found strong consistency in estimating 13 out of 23(56.52%) parameters among lithium heparin, K2EDTA, and sodium citrate.

Several studies shed light on notable findings in examining the variation in CBC parameters across different anticoagulants. In a study involving 30 clinically healthy dogs of various breeds, no difference was observed statistically between sodium citrate and K3EDTA in 50% of CBC parameters, including HGB, HCT, PLT, and PCT [10]. Similarly, in another study on healthy humans, no statistically significant distinction was found in 35.7% of CBC parameters, including MCV, MCHC, MCH, %Lymph, and %Neu, among K3EDTA, citrate, and lithium heparin [3]. In the current study, which compares K2EDTA, sodium citrate, and lithium heparin, it was observed that 39.13% of CBC parameters showed no significant variation among these anticoagulants. However, significant variations were observed in other CBC parameters, emphasizing the importance of careful anticoagulant selection, particularly in clinical scenarios where precise and consistent CBC measurements are essential for accurate diagnosis and monitoring of conditions [11].

Various studies investigated the variation in CBC parameters when using different anticoagulants. In a study involving 50 healthy dogs, no comparable results were found between EDTA and sodium citrate for all CBC parameters, suggesting that citrate may lead to less accurate results than EDTA [12]. Similarly, in another study of 55 healthy humans, comparing K3EDTA and heparin revealed significant variation in 9 out of 14(64.29%) CBC parameters. However, comparing results between heparin and K3EDTA revealed significant variations in only three parameters (21.43%), including PLT, Mon, and %Mon [3]. In the current study, comparing K2EDTA and lithium heparin revealed comparable results in 14 out of 23(60.87%) CBC parameters, while comparing K2EDTA and citrate revealed comparable results in 13 out of 23(56.52%) CBC parameters.

When comparing PLT levels between blood samples treated with K2EDTA, sodium citrate, and lithium heparin, a notable decrease in PLT counts in samples observed treated with sodium citrate and lithium heparin compared to those treated with EDTA. In other studies that involved patients with pseudo thrombocytopenia, PLT was found to be higher among those treated with sodium citrate than EDTA [13-15]. According to a study, reduced PLT count might result from the potent activation of platelets by citrate in ill animals, forming small platelet aggregates. [16].

Regarding RBC indices, different studies suggested lower HGB and HCT in citrated blood samples compared to EDTA [3, 9]. Similarly, the current study observed the same trend. Reduced HGB levels in the EDTA-treated blood sample may be attributed to the dilutional effect of sodium citrate and the diminished capacity of HGB to undergo oxidation in citrate [3].

Different studies indicated low WBC, MCV, and MCHC in sodium citrate-treated blood compared to those treated with

Barw Medical Journal | 2024;2(3):20-25 | https://doi.org/10.58742/bmj.v2i2.90

Table 4. Differences in CBC estimation with results using two Anticoagulants.

CBC parameters	Sodium Citrate Median (Min-Max)	Lithium Heparin Median (Min-Max)	P-value	K2EDTA Median (Min-Max)	Lithium Heparin Median (Min-Max)	P-value	K2EDTA Median (Min-Max)	Sodium Citrate Median (Min-Max)	P-value
WBC	7.48(0.20-21.92)	7.91(0.27-25.27)	0.162	8.02(0.25-23.95)	7.91(0.27-25.27)	0.923	8.02(0.25-23.95)	7.48(0.20-21.92)	0.071
%Neu	59.55(27.00-93.40)	62.90(31.70-95.90)	0.001	59(25.80-93.20)	62.90(31.70-95.90)	<0.001	59(25.80-93.20)	59.55(27.00-93.40)	0.800
%Lymph	31.20(3.40-63.80)	30.70(3.00-61.50)	0.869	31.70(3.50-66.80)	30.70(3.00-61.50)	0.722	31.70(3.50-66.80)	31.20(3.40-63.80)	0.962
%Mon	5.80(0.20-11.60)	1.70(0.00-17.70)	<0.001	6.50(0.40-13.10)	1.70(0.00-17.70)	<0.001	6.50(0.40-13.10)	5.80(0.20-11.60)	<0.001
%Eos	2.50(0.10-13.10)	2.30(0.00-13.70)	0.534	2.10(0.10-13.90)	2.30(0.00-13.70)	0.509	2.10(0.10-13.90)	2.50(0.10-13.10)	0.076
%Bas	0.50(0.10-1.60)	0.70(0.10-1.60)	0.012	0.50(0.10-1.60)	0.70(0.10-1.60)	<0.001	0.50(0.10-1.60)	0.50(0.10-1.60)	0.172
Neu	4.35(1.64-19.66)	4.78(1.70-22.48)	0.05	4.62(1.81-21.78)	4.78(1.70-22.48)	0.532	4.62(1.81-21.78)	4.35(1.64-19.66)	0.430
Lymph	2.27(0.45-7.08)	2.40(0.46-20.20)	0.147	2.45(052-8.01)	2.40(0.46-20.20)	0.997	2.45(052-8.01)	2.27(0.45-7.08)	0.126
Mon	0.39(0.02-2.20)	0.14(0.00-1.28)	<0.001	0.48(0.03-1.89)	0.14(0.00-1.28)	<0.001	0.48(0.03-1.89)	0.39(0.02-2.20)	<0.001
Eos	0.18(0.01-1.09)	0.18(0.00-1.21)	0.998	0.16(0.01-1.22)	0.18(0.00-1.21)	0.520	0.16(0.01-1.22)	0.18(0.01-1.09)	0.484
Bas	0.04(0.01-0.50)	0.05(0.01-0.18)	0.016	0.04(0.01-0.11)	0.05(0.01-0.18)	0.001	0.04(0.01-0.11)	0.04(0.01-0.50)	0.654
RBC	4.50(2.80-6.14)	4.97(3.03-6.82)	<0.001	4.97(3.18-6.83)	4.97(3.03-6.82)	0.978	4.97(3.18-6.83)	4.50(2.80-6.14)	<0.001
HGB	12.20(6.20-16.70)	13.60(7.20-18.20)	<0.001	13.60(6.80-18.10)	13.60(7.20-18.20)	0.961	13.60(6.80-18.10)	12.20(6.20-16.70)	<0.001
HCT	37.30(21.30-51.00)	41.25(23.80-52.90)	<0.001	41.5(22.90-53.60)	41.25(23.80-52.90)	0.835	41.5(22.90-53.60)	37.30(21.30-51.00)	<0.001
MCV	84(50.10-98.20)	84.25(49.40-98.40)	0.939	84.70(49.40-99.40)	84.25(49.40-98.40)	0.850	84.70(49.40-99.40)	84(50.10-98.20)	0.653
MCH	27.80(15.40-31.40)	28.10(14.90-31.70)	0.525	27.90(14.80-32.00)	28.10(14.90-31.70)	0.852	27.90(14.80-32.00)	27.80(15.40-31.40)	0.850
MCHC	33(22.10-34.90)	33.25(27.60-35.60)	0.004	32.90(27.10-35.90)	33.25(27.60-35.60)	0.001	32.90(27.10-35.90)	33(22.10-34.90)	0.959
RDW-SD	43.50(10.20-65.60)	43.80(35.50-65.60)	0.637	44.25(35.90-66.70)	43.80(35.50-65.60)	0.346	44.25(35.90-66.70)	43.50(10.20-65.60)	0.05
PLT	175(17-487)	169(18-506)	0.407	260(23-588)	169(18-506)	<0.001	260(23-588)	175(17-487)	<0.001
MPV	8.90(7.10-11.10)	9.30(7.20-11.20)	<0.001	9.10(7.30-11.10)	9.30(7.20-11.20)	0.045	9.10(7.30-11.10)	8.90(7.10-11.10)	0.022
PDW	11.20(7.60-20.60)	12(8.20-21.70)	<0.001	11.60(8.10-20.00)	12(8.20-21.70)	0.070	11.60(8.10-20.00)	11.20(7.60-20.60)	0.194
PCT	0.16(0.02-0.39)	0.16(0.02-0.38)	0.969	0.24(0.02-0.52)	0.16(0.02-0.38)	<0.001	0.24(0.02-0.52)	0.16(0.02-0.39)	<0.001
PLCR	28.90(16.00-50.00)	31.80(0.90-52.80)	<0.001	30.45(16.60-48.20)	31.80(0.90-52.80)	0.089	30.45(16.60-48.20)	28.90(16.00-50.00)	0.05

Table 5. Concordance Correlation Coefficient (CCC) for Assessing Agreement Levels between K2EDTA with Sodium Citrate and Lithium Heparin.

CBC parameters	K2EDTA-Citrate	Pearson ρ (precision)	Bias correction factor (accuracy)	K2EDTA-Lithium heparin	Pearson ρ (precision)	Bias correction factor (accuracy)
WBC	0.969(0.962- 0.975)	0.991	0.978	0.988(0.985 - 0.991)	0.990	0.999
%Neu	0.975(0.968 -0.980)	0.976	0.999	0.879(0.851 - 0.902)	0.941	0.934
%Lymph	0.990(0.988 - 0.993)	0.991	0.999	0.954(0.942 - 0.964)	0.957	0.997
%Mon	0.621(0.54 - 0.687)	0.666	0.932	0.190(0.134- 0.245)	0.428	0.443
%Eos	0.955(0.943 - 0.965)	0.972	0.983	0.932(0.915 - 0.947)	0.941	0.991
%Bas	0.661(0.587 - 0.725)	0.673	0.983	0.547(0.462 - 0.623)	0.599	0.914
Neu	0.983(0.979 - 0.987)	0.992	0.991	0.978(0.973 - 0.983)	0.983	0.995
Lymph	0.961(0.952- 0.968)	0.988	0.972	0.504(0.421 - 0.579)	0.566	0.890
Mon	0.568(0.485 - 0.641)	0.619	0.917	0.231(0.170 - 0.290)	0.471	0.490
Eos	0.958(0.947 - 0.967)	0.964	0.994	0.925(0.906 -0.941)	0.932	0.992
Bas	0.341(0.248 - 0.428)	0.406	0.840	0.525(0.439 - 0.601)	0.584	0.899
RBC	0.726(0.687- 0.760)	0.988	0.734	0.994(0.992 - 0.995)	0.994	0.999
HGB	0.742(0.705 - 0.775)	0.988	0.751	0.977(0.970 - 0.982)	0.979	0.998
HCT	0.716(0.677- 0.751)	0.989	0.723	0.993(0.991 - 0.995)	0.994	0.999
MCV	0.996(0.995- 0.997)	0.999	0.997	0.995(0.994 - 0.997)	0.997	0.999
MCH	0.994(0.992 -0.995)	0.995	0.999	0.994(0.993 - 0.995)	0.995	0.999
MCHC	0.0804(0.063 - 0.098)	0.630	0.128	0.876(0.846 - 0.900)	0.926	0.946
RDW-SD	0.861(0.828- 0.889)	0.887	0.971	0.986(0.982 - 0.989)	0.995	0.991
PLT	0.444(0.389 - 0.497)	0.836	0.532	0.363(0.305 - 0.418)	0.722	0.503
MPV	0.859(0.824- 0.888)	0.880	0.977	0.852(0.815 -0.882)	0.876	0.973
PDW	0.878(0.847 - 0.903)	0.894	0.982	0.850(0.813 - 0.880)	0.87	0.972
PCT	0.369(0.317 - 0.418)	0.817	0.451	0.313(0.256 - 0.368)	0.655	0.478
PLCR	0.879(0.848- 0.904)	0.897	0.980	0.838(0.798- 0.871)	0.854	0.982

EDTA [12,17]. The current study observed non-significant low levels of WBC, MCV, and MCHC.

The current study assessed the agreement levels among different anticoagulants, with K2EDTA serving as the standard. Lithium heparin exhibited perfect or substantial agreement in determining 9 out of 23 CBC parameters (39.13%), which included RBC, HCT, MCV, MCH, WBC, %Lymph, Neu, HGB, and RDW-SD.

This percentage was higher than the literature, where heparin showed perfect or significant agreement in evaluating 4 out of 14 CBC parameters (28.57%), namely RBC, HGB, HCT, and MCH [3]. One limitation of the current study warrants consideration: the cross-sectional design restricts the ability to establish causality or observe changes over time.

5. Conclusion

Using citrate instead of K2EDTA for CBC estimation may yield inaccurate outcomes, whereas lithium heparin could serve as an alternative anticoagulant, requiring careful monitoring.

Declarations

Conflicts of interest: The author(s) have no conflicts of interest to disclose.

Ethical approval: Not applicable.

Patient consent (participation and publication)**:** Not applicable.

Source of Funding: Star Lab Company.

Role of Funder: The funder remained independent, refraining from involvement in data collection, analysis, or result formulation, ensuring unbiased research free from external influence.

Acknowledgements: None to be declared.

Authors' contributions: RQS was a major contributor to the conception of the study and the literature search for related studies. SQO, AMM, SLE, and DAH were involved in the literature review, manuscript writing, and data analysis and interpretation. AKH, HAS, BAS and SSR Literature review, final approval of the manuscript, and processing of the tables. SJ, MHS, and HSA were involved in the literature review, the study's design, and the manuscript's critical revision. RQS and AMM Confirmation of the authenticity of all the raw data All authors approved the final version of the manuscript.

Use of AI: AI was not used in the drafting of the manuscript, the production of graphical elements, or the collection and analysis of data.

Data availability statement: Not applicable.

References

1. Doeleman MJ, Esseveld A, Huisman A, de Roock S, Tiel Groenestege WM. Stability and comparison of complete blood count parameters between capillary and venous blood samples. International Journal of Laboratory Hematology. 2023;45(5):659-67. doi:10.1111/ijlh.14080
2. Etim NN, Williams ME, Akpabio U, Offiong EE. Haematological parameters and factors affecting their values. Agricultural science. 2014;2(1):37-47. doi:N/A
3. Akorsu EE, Adjabeng LB, Sulleymana MA, Kwadzokpui PK. Variations in the full blood count parameters among apparently healthy humans in the Ho municipality using ethylenediamine tetraacetic acid (EDTA), sodium citrate and lithium heparin anticoagulants: A laboratory-based cross-sectional analytical study. Heliyon. 2023;9(6). doi:10.1016/j.heliyon.2023.e17311
4. Walencik J, Witeska M. The effects of anticoagulants on hematological indices and blood cell morphology of common carp (Cyprinus carpio L.). Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology. 2007;146(3):331-5. doi:10.1016/j.cbpc.2007.04.004
5. Nemec A, Drobnič–Košorok M, Butinar J. The effect of high anticoagulant k3-edta concentration on complete blood count and white blood cell differential counts in healthy beagle dogs. Slovenian Veterinary Research. 2005;42(3/4):65-70. doi:N/A
6. Weiss DJ, Tvedten H. The complete blood count, bone marrow examination, and blood banking: general comments and selected techniques. InSmall animal clinical diagnosis by laboratory methods 2012(pp. 12-37). WB Saunders. doi:10.1016/B978-1-4377-0657-4.00002-8
7. Muhialdeen AS, Ahmed JO, Baba HO, Abdullah IY, Hassan HA, Najar KA, et al. Kscien's List; A New Strategy to Discourage Predatory Journals and Publishers (Second Version). Barw Medical Journal.2023;1(1):24-26. doi:10.58742/bmj.v1i1.14
8. Lawrence I, Lin K. A concordance correlation coefficient to evaluate reproducibility. Biometrics. 1989:255-68. doi:10.2307/2532051
9. Faggio C, Arfuso F, Piccione G, Zumbo A, Fazio F. Effect of three different anticoagulants and storage time on haematological parameters of Mugil cephalus (Linneaus, 1758). Turkish Journal of Fisheries and Aquatic Sciences. 2014;14(3):615-21. doi:10.4194/1303-2712-v14_3_03
10. Zmigrodzka M, Winnicka A, Guzera M. Comparison of the influence of ADTA-K3 and sodium citrate on haematology analysis in healthy dogs. Polish journal of veterinary sciences. 2012;15(2). doi:10.2478/v10181-012-0059-6
11. Gunawardena D, Jayaweera S, Madhubhashini G, Lokumarakkala DD, Senanayake SJ. Reliability of parameters of complete blood count with different storage conditions. Journal of clinical laboratory analysis. 2017;31(2): e22042. doi:10.1002/jcla.22042
12. Stokol T, Erb HN. A comparison of platelet parameters in EDTA-and citrate-anticoagulated blood in dogs. Veterinary Clinical Pathology. 2007 Jun;36(2):148-54. doi:10.1111/j.1939-165X.2007.tb00201.x
13. Lin J, Luo Y, Yao S, Yan M, Li J, Ouyang W, Kuang M. Discovery and correction of spurious low platelet counts due to EDTA-dependent pseudothrombocytopenia. Journal of clinical laboratory analysis. 2015;29(5):419-26. doi:10.1002/jcla.21818
14. Zhang L, Xu J, Gao L, Pan S. Spurious thrombocytopenia in automated platelet count. Laboratory medicine. 2018;49(2):130-3. doi:10.1093/labmed/lmx081
15. Dima F, Salvagno GL, Danese E, Veneri D, Lippi G. An unusual case of sodium citrate-dependent artifactual platelet count. Interventional Medicine and Applied Science. 2020;11(3):193-6. doi:10.1556/1646.11.2019.24
16. McShine RL, Das PC, Smit Sibinga CT, Brozović B. Effect of EDTA on platelet count and other platelet parameters in blood and blood components collected with CPDA-1. Vox sanguinis. 1991;61(2):84-9. doi:10.1159/000461330
17. Sharif MT, Mahabadi MA, Moshfeghi S, Sharifi H, Hoseini SM, Alavi SM. Artifactual changes in hematological variables in equine blood samples stored at different temperatures and various anticoagulants. Comparative Clinical Pathology. 2012; 21:449-52. doi:10.1007/s00580-010-1116-0