International Journal of Research in Medical Sciences
Maharana SK et al. Int J Res Med Sci. 2024 Jun;12(6):1915-1920
www.msjonline.org
pISSN 2320-6071 | eISSN 2320-6012

DOI: https://dx.doi.org/10.18203/2320-6012.ijrms20241303

Original Research Article

Quantifying the influence of varying centrifugation spin paces and timespans on serum electrolyte dynamics in an Indian clinical laboratory

Sunil K. Maharana*, Shiuli R. Adak, Rupesh K. Dolui

Department of Biochemistry, Murshidabad Medical College and Hospital, Berhampore, Murshidabad, West Bengal, India

Received: 20 April 2024
Accepted: 10 May 2024

***Correspondence:**
Dr. Sunil K. Maharana,
E-mail: docsunilkumarmaharana@gmail.com

ABSTRACT

Background: A reduction in turnaround time at any laboratory is critical for early assessment. Our aim of the study was to evaluate the effects of various spin paces and timespans on certain serum electrolyte concentrations, such as sodium (Na^+), potassium (K^+), and chloride (Cl^-).
Methods: A cross-sectional, observational study was carried out on 66 apparently healthy volunteers. 10 ml of blood was drawn from each and divided into 4 labelled clot vials (2.5 ml in each vial). Concentrations of serum Na^+, K^+, and Cl^- were assessed in 2 groups (1, 2), each with 33 individuals. Group 1 had varying spin pace parameters (1500 rpm, 2500 rpm, 3500 rpm, and 4500 rpm), and group 2 included various timespans (2 min, 5 min, 10 min, and 15 min). The observations were analyzed using statistical package for the social sciences (SPSS). Analysis of variance (ANOVA) was implied along with a post-hoc Tukey test. A p value of <0.05 was considered statistically significant.
Results: Mean concentrations of Na^+, K^+, and Cl^- at different spin paces for a fixed runtime of 2mins had statistically no differences between each other: Na^+ (p=0.978), K^+ (p 0.999), and Cl^- (p=0.997). However, there were statistically significant mean differences at various timespans for Na^+ (p<0.001), K^+ (p<0.001), and Cl^- (p<0.001).
Conclusions: Our study concludes that Na^+, K^+, and Cl^- concentrations were not altered at various spin paces. A timespan of 2 mins at 4500 rpm outperformed the benchmarks without affecting the results, signifying that it can be routinely chosen for estimating serum electrolytes such as Na^+, K^+, and Cl^-, effectively lowering turnaround time.

Keywords: Centrifugation, Serum electrolytes, Turnaround time

INTRODUCTION

Centrifugation is a cardinal process in any clinical laboratory and has been regularly used during pre-analytical phases. With apparent novel advancements in centrifugation devices such as table-top centrifuges, high-speed centrifuges, and ultra-centrifuges, biomaterials of various shapes, sizes, densities, and viscosities can be isolated easily.[1,2] Currently, table-top centrifuges are widely utilized in clinical laboratories. Cellular components of blood are rapidly separated from either plasma or serum by centrifugation at an accelerated relative centrifugal force. Relative centrifugal force (rcf) and rotations per minute (rpm) are calculated as per the rotating radius r (the distance between the axis of the rotation and the base of the container in mm) using the formula given.[3]

$$\text{rcf} = 1{,}118 \times \text{r}\,(\text{RPM}/1000)^2$$

As per the World Health Organisation's (WHO) recommendation, post-plasma coagulation, the samples are to be centrifuged for 10 minutes at a minimum speed of 1500 g.[3] Prolonged and excessive centrifugation may lead to *in vitro* haemolysis, as suggested by a few studies.[4] According to the Clinical and Laboratory Standards

Institute (CLSI), re-centrifugation of specimens for potassium will lead to falsely elevated levels. CLSI does not recommend rimming the tubes, which may have the potential for laboratory-induced haemolysis.[5] Turnaround time (TAT) may be widely defined as; the total time taken from specimen collection to dispatching the reports. This is a critical step for any clinical laboratory, as it reflects the quality and effectiveness. Shorter TATs of stat samples will improve the proficiency of laboratory reports, eventually leading to swift decisions by physicians.[6] Specimen preparation is the leading cause of extended TATs, which leads to dissatisfaction among clinical chemists and physicians. Centrifugation configurations can be altered to optimize the sample flow, effectively reducing TATs without compromising on analytical accuracy or quality. Most of the recent studies have used gel separator tubes, and it is not feasible for all the clinical laboratories because of the higher costs.[7]

Modern studies suggest serum and heparin samples can be centrifuged at higher speeds, such as 3000 x g, for a shorter amount of time, about 5 minutes, without altering the results. However, when using LiHepBar tubes for blood collection, a separate lactate dehydrogenase reference interval was suggested.[8] Novel innovations such as the "axial separation module" with an axial process container were a cost-effective approach with shorter TATs compared to conventional separations.[9] Terumo Venoject II, a plastic-walled blood collection tube with a plasma separator, was evaluated for various modalities, including the utilization of plastic instead of glassware. Plastic-walled tubes were extremely resistant to shear mechanical breakages at higher g-forces.[10]

Our core objective of this study was to evaluate the performance of serum electrolyte concentrations such as sodium (Na^+), potassium (K^+), and chloride (Cl^-) under the influence of various spin paces and at various time spans.

Objective

Aim of the study was to evaluate the effect of various spin paces and timespans using centrifugation on certain serum electrolyte concentrations, such as Na^+, K^+, and Cl^-.

METHODS

It was a cross-sectional, observational study.

This study was carried out at the Central Laboratory, Department of Biochemistry, Murshidabad Medical College and Hospital, West Bengal, India.

The study was carried out from December 2023 to January 2024.

Inclusion criteria

Healthy volunteers from the undergraduate students of our medical college, both males and females.

Exclusion criteria

Samples that were rendered to be haemolysed, lipemic, or icteric were excluded.

Evaluation under varying spin paces, group 1

10 ml of fasting blood was drawn aseptically from apparently 33 healthy undergraduate student volunteers (16 males and 17 females) of our medical college with a proper self-explanatory informed consent, and each sample was divided into 4 equal parts (2.5 ml) decanted in a Haemocheck™ Polymed serum tube, cap colour red, volume 5.0 ml, and kept aside for 10 minutes in upright position at optimum laboratory operating temperature 22°C. Each tube was labelled as 1500 rpm, 2500 rpm, 3500 rpm, and 4500 rpm, respectively.

Each tube was placed within a well-calibrated and balanced REMI-8C centrifugation machine and spined as per the labels at 1500 rpm, 2500 rpm, 3500 rpm, and 4500 rpm for 2 minutes. Serum Na^+, K^+, and Cl^- levels were assessed using a well-calibrated electrolyte analyser, Medica Easylite® PLUS. All the observations were recorded.

Evaluation under varying timespans, group 2

10 ml of fasting blood was drawn aseptically from apparently 33 healthy undergraduate student volunteers (17 males and 16 females) of our medical college with a proper self-explanatory informed consent, and each sample was divided into 4 equal parts (2.5 ml) decanted in a Haemocheck™ Polymed serum tube, cap colour red, volume 5.0 ml, and kept aside for 10 minutes in upright position at optimum laboratory operating temperature 22°C.

Each tube was labelled as 2 mins, 5 mins, 10 mins, and 15 mins respectively. Each tube was placed within a well-calibrated and balanced REMI-8C centrifugation machine and spined at a speed of 4500 rpm as per labelled tube durations such as 2 mins, 5 mins, 10 mins, and 15 mins. Serum Na^+, K^+, and Cl^- levels were assessed using a well-calibrated electrolyte analyser, Medica Easylite® PLUS. All the observations were recorded.

Statistical analysis

Core final data was drafted using IBM statistical package for the social sciences (SPSS) Statistics 20, Ver. 20.0.0.0.155, IBM Corp. 2006, 2011, US. The mean and standard deviation (SD) were calculated using SPSS for groups 1 and 2. For plotting the column scattergram graph, origin 2022 version 9.9 was utilised. The mean differences within various spin paces and for various timespans were analysed with ANOVA implied along with a post-hoc Tukey test for any statistically differential outcomes in serum Na^+, K^+, and Cl^- levels, and a (p value) <0.05 was considered statistically significant.

RESULTS

Mean concentration comparisons of Na^+, K^+, and Cl^- at different spin paces such as 1500 rpm, 2500 rpm, 3500 rpm, and 4500 rpm rendered at a fixed runtime of 2 mins are summarized in Table 1. From group 1, there were no statistical differences between the mean level concentrations of Na^+ (p value 0.978), K^+ (p value 0.999), and Cl^- (p value 0.997) and their respective different spin paces at a fixed runtime of 2 mins. On the contrary, in group 2, there were statistically significant differences between the mean level concentrations of Na^+ (p value <0.001), K^+ (p value <0.001), and Cl^- (p value <0.001) and their respective different timespans, such as 2 mins, 5 mins, 10 mins, and 15 mins summarised in Table 2.

Table 1: Mean comparisons at various spin paces (runtime=2 mins).

Parameters	1500 rpm	2500 rpm	3500 rpm	4500 rpm	ANOVA
Na+ (mean±SD)	139.19±2.60	139.17±2.61	139.39±2.52	139.36±2.50	0.978*
K^+ (mean±SD)	4.105±0.42	4.106±0.41	4.100±0.42	4.101±0.41	0.999*
Cl^- (mean±SD)	105.57±2.88	105.72±2.84	105.63±2.91	105.63±2.90	0.997*

*P value of <0.05 was considered statistically significant

Table 2: Mean comparisons at various timespans (run speed=4500 rpm).

Parameters	2 mins	5 mins	10 mins	15 mins	ANOVA
Na+ (mean±SD)	140.33±2.50	138.25±2.63	135.88±2.80	132.83±3.24	<0.001*
K^+ (mean±SD)	4.00±0.41	4.24±0.42	4.52±0.47	4.93±0.55	<0.001*
Cl^- (mean±SD)	104.6±2.90	106.76±2.96	108.73±3.15	110.74±4.06	<0.001*

*P value of <0.05 was considered statistically significant

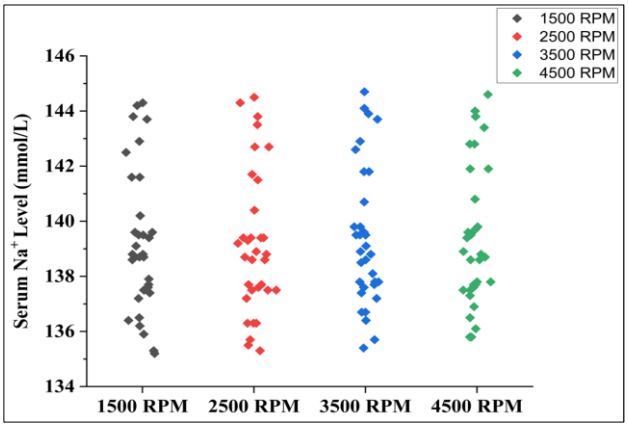

Figure 1: Column scattergram of serum Na^+ levels (mmol/l) at various spin paces (runtime=2 mins).
Various spin paces show no significant concentration differences, RPM=rotations per minute

A post hoc Tukey test was carried out for multiple comparisons, among the serum concentrations of Na+, K+, and Cl- at different spin paces (group 1). From the mean concentrations of serum Na^+ at various spin paces, there were no statistical differences, such as 1500 rpm and 2500 rpm (p value 0.999), 1500 rpm and 3500 rpm (p value 0.988), 1500 rpm and 4500 rpm (p value 0.992), 2500 rpm and 3500 rpm (p value 0.986), 2500 rpm and 4500 rpm (p value 0.990), 3500 rpm and 4500 rpm (p value 0.999), respectively. For the mean concentrations of serum K^+ at various spin paces there were also no statistical differences, such as 1500 rpm and 2500 rpm (p value >0.999), 1500 rpm and 3500 rpm (p value >0.999), 1500 rpm and 4500 rpm (p value >0.999), 2500 rpm and 4500 rpm (p value >0.999), 3500 rpm and 4500 rpm (p value >0.999), 2500 rpm and 3500 rpm (p value >0.999) respectively, and from the mean concentrations of serum Cl^- at various spin paces there were no statistical differences, such as 1500 rpm and 2500 rpm (p value 0.997), 1500 rpm and 3500 rpm (p value >0.999), 1500 rpm and 4500 rpm (p value >0.999), 2500 rpm and 4500 rpm (p value 0.999), 3500 rpm and 4500 rpm (p value >0.999), 2500 rpm and 3500 rpm (p value 0.999).

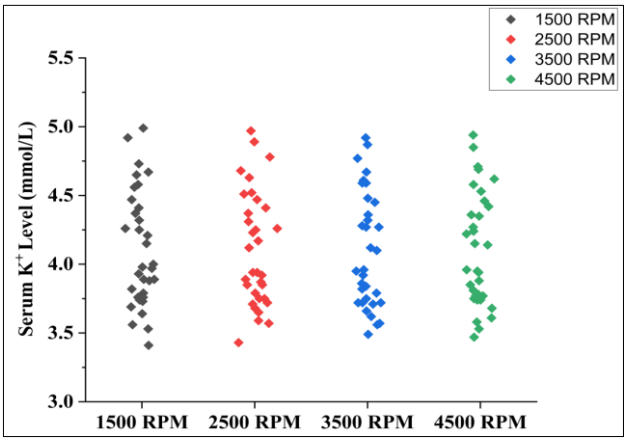

Figure 2: Column scattergram of serum K^+ levels (mmol/l) at various spin paces (runtime=2 mins).
Various spin paces show no significant concentration differences, RPM=rotations per minute

However, when a post hoc Tukey test was carried out for multiple comparisons, among the serum concentrations of Na^+, K^+, and Cl^- at different timespans (group 2), there were distinctive indications such as: from the mean concentrations of serum Na^+, there were statistically significant differences between 2 mins and 5 mins (p value 0.016), 2 mins and 10 mins (p value <0.001), 2 mins and

15 mins (p value <0.001), 5 mins and 10 mins (p value 0.005), 10 mins and 15 mins (p value <0.001), respectively. From the mean concentrations of serum K^+, there were no statistical differences between 2 mins and 5 mins (p value 0.149), 5 mins and 10 mins (p value 0.090). However, there were statistically significant differences between the mean serum K^+ concentrations at 10 mins and 15 mins (p value 0.004), 2 mins and 10 mins (p<0.001), 2 mins and 15 mins (p <0.001). For mean Cl^- concentration levels between 2 mins and 5 mins (p value 0.048), 2 mins and 10 mins (p<0.001), 2 mins and 15 mins (p value <0.001) there were statistically significant differences, but there was no statistical difference between 5 mins and 10 mins (p value 0.078), 10 mins and 15 mins (p value 0.070). Figures 1-3 show column scattergrams of serum Na^+, K^+, and Cl^- levels (mmol/l) at various spin paces (runtime=2 mins). Figures 4-6 show column scattergrams of serum Na^+, K^+, and Cl^- levels (mmol/l) at various timespans (run speed=4500 rpm).

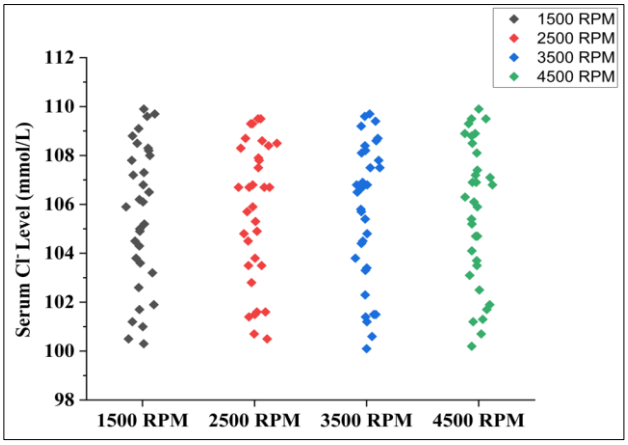

Figure 3: Column scattergram of serum Cl^- levels (mmol/l) at various spin paces (runtime=2 mins).
Various spin paces show no significant concentration differences, RPM=rotations per minute

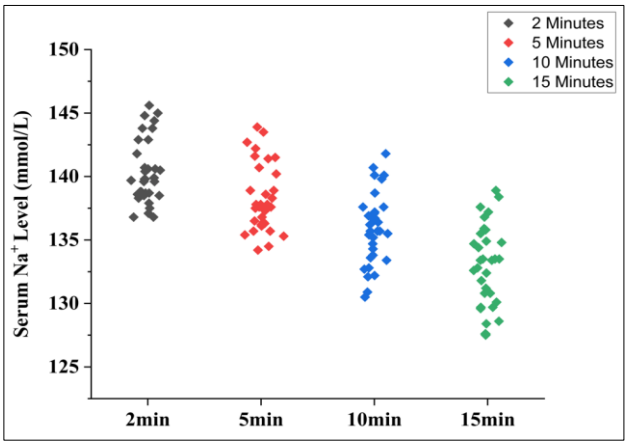

Figure 4: Column scattergram of serum Na^+ levels (mmol/l) at various timespans (run speed=4500 rpm).
Reduction of serum Na^+ levels at variable timespans, RPM=rotations per minute

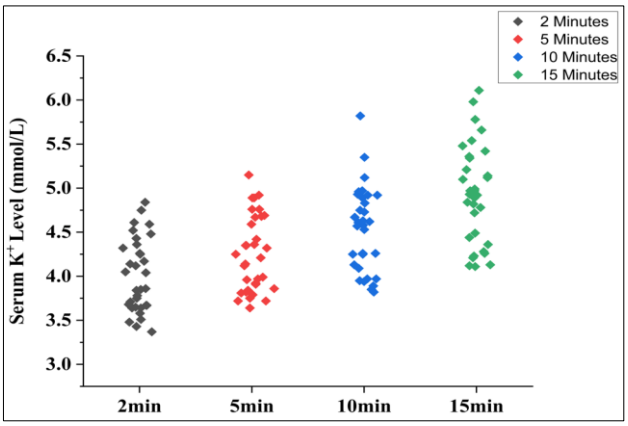

Figure 5: Column scattergram of serum K^+ levels (mmol/l) at various timespans (run speed=4500 rpm).
Serum K^+ levels potentially had significant elevated concentrations at various timespans, RPM=rotations per minute

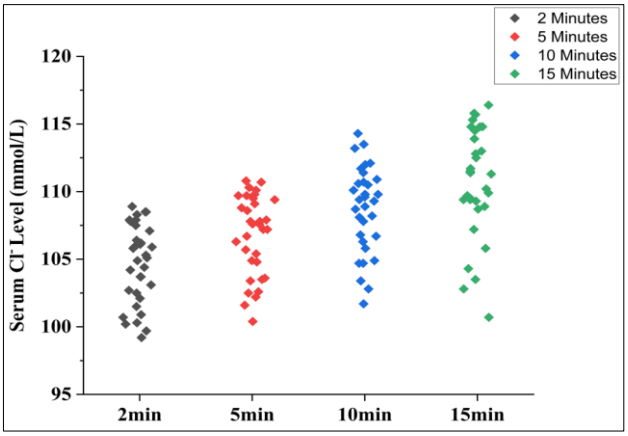

Figure 6: Column scattergram of serum Cl^- levels (mmol/l) at various timespans (run speed=4500 rpm).
Serum Cl^- levels had significant elevated concentrations at various timespans, RPM=rotations per minute

DISCUSSION

Expeditious deployment of accurate and reliable reports should be the prime goal of any distinguished clinical laboratory. A reduction of TATs with several hypothetical dimensions in relation to centrifugation time was suggested, such as increasing the speed, reduction of time, any novel separation techniques, or completely abolishing this preanalytical step altogether.[6] Glassware tubes are highly discouraged and, in general, possess potential risks of breaking at higher speeds inside the centrifugation devices, leading to the exposure of splayed biohazard samples and sharp objects towards the laboratory personnel. On the other hand, plastic-walled collection tubes are extremely shock-resistive at higher centrifugal speeds and have a lesser risk of breakage than conventional glass tubes.[10] In our study, sterile, red-top, plastic vials with a clot activator were effectively utilized for sample collection, serum separation using a well-calibrated balanced centrifuge, and estimation of serum Na^+, K^+, and

Cl^- Levels (mmol/l) using an electrolyte analyzer at various pre-defined conditions. If the samples are centrifuged before the actual complete coagulation sets in, there might be an elevated chance of haemolysis eventually rendering the sample unfit for further analysis.[4] In our experiment, initially all the samples were drawn into the plastic vials and left in an upright position undisturbed for about 10 minutes to induce complete coagulation at the routine laboratory temperature of 22°C, after which they were segregated and centrifuged as per groups. Recent novel innovations, such as the axial separation module, which had a distinct axial process container, effectively reduced the TATs; however, its major drawback was that it could not be run for more than a batch of ten.[9] Majorly, automated specimen logistics have a promising reduction in TATs, such as computer-controlled pneumatic tube modules with a payload capacity of 15 pounds at a speed of 25 feet per second. The use of pneumatic tubes had shown a 25% reduction in TAT for K^+ and haemoglobin. Few commercial laboratories have a track-based system for transporting specimens running at speeds of up to 10 feet per second. However, automation systems cannot be installed at every high-throughput clinical laboratory due to their cost implications. Moreover, these automations are either limited to commercial and privatised laboratories or a few reference labs.[11]

As per our study, the runtime was kept constant at 2 mins for group 1. All the tubes labelled as 1500 rpm, 2500 rpm, 3500 rpm, and 4500 rpm were spined as per their labelling. With analysis, we found that there was no significant variability at different spin paces for all three serum Na^+, K^+, and Cl^- levels (mmol/l). Figures 1-3 show scattergrams plots for all three analytes at different spin paces. The evaluation of group 2 with a consistent 4500 rpm revealed several distinct observations. Serum Na^+ levels (mmol/l) had a significant reduction in concentrations when spun extensively for about 10 or 15 mins. Considering serum K^+ levels (mmol/l) were significantly elevated when the time span was extended for 10 mins and 15 mins. A runtime of 2 mins and 5 mins did not have any concentration variations on serum K^+ levels. Lastly, serum Cl^- levels (mmol/l) had also raised levels of concentration and did not vary much at 10 mins, 15 mins. Figures 4-6 show scattergrams plots for all three analytes at different timespans.

Centrifugation being a key step at the pre-analytical stage, which is carried out routinely, might be a time-consuming process for high throughput clinical laboratories. An overall reduction in centrifugation duration and raising the speed without compromising on the quality and accuracy of the desired analytes might significantly improve the overall turnaround time.

CONCLUSION

Based on our study, we conclude that Na^+, K^+, and Cl^- concentrations were not altered at various spin paces such as 1500 rpm, 2500 rpm, 3500 rpm, and 4500 rpm. A timespan of 2 mins at 4500 rpm outperformed in the benchmarks without affecting the results, signifying that this configuration can be routinely chosen for estimating serum electrolytes such as Na^+, K^+, and Cl^-, effectively lowering turnaround time. However, it should be noted that increased shear spin g-forces may elevate serum K^+ concentrations falsely. A significant reduction in serum Na^+ concentrations was observed when the time span was extended beyond 10-15 mins, and serum Cl^- showed higher concentrations when the time span was extended beyond 10-15 mins.

ACKNOWLEDGEMENTS

Authors would like to thank all the medical laboratory technologists and staff involved. They would also like to thank Dr. Purusottam P., for her unwavering support towards the study.

Funding: No funding sources
Conflict of interest: None declared
Ethical approval: The study was approved by the Institutional Ethics Committee

REFERENCES

1. Allison FI, Ojule AC, Shittu L, Bamigbowu EO. The Effects Of Speed And Duration Of Centrifugation On The Values of Some Commonly Measured Plasma Electrolytes. Eur J Med Health Sci. 2020;2:2.
2. Frei M. Centrifugation Separations. Bio Files. 2012;6(5):6-7.
3. World Health Organization. Use of anticoagulants in diagnostic laboratory investigations. 2002. Available at: https://iris.who.int/handle/10665/65957. Accessed on 09 February 2024.
4. Heireman L, Van Geel P, Musger L, Heylen E, Uyttenbroeck W, Mahieu B. Causes, consequences and management of sample hemolysis in the clinical laboratory. Clin Biochem. 2017;50(18):1317-22.
5. NCCLS. Procedures for the Handling and Processing of Blood Specimens; Approved Guideline—Third Edition. NCCLS document H18-A3. NCCLS, 940 West Valley Road, Suite 1400, Wayne, Pennsylvania 19087-1898 USA. 2004.
6. Lippi G, Salvagno G, Montagnana M, Guidi G. Preparation of a Quality Sample: Effect of Centrifugation Time on Stat Clinical Chemistry Testing. Lab Med. 2007;38:172-6.
7. Tantisaranon P, Dumkengkhachornwong K, Hnoonual A. Influence of reduced centrifugation time on clinical chemistry analytes and literature review. Turk J Biochem. 2023;48(4):376-87.
8. Cadamuro J, Mrazek C, Leichtle AB, Kipman U, Felder TK, Wiedemann H, et al. Influence of centrifugation conditions on the results of 77 routine clinical chemistry analytes using standard vacuum blood collection tubes and the new BD-Barricor tubes. Biochem Med (Zagreb). 2018;28(1):010704.

9. Estey CA, Felder RA. Clinical trials of a novel centrifugation method: Axial separation. Clin Chem. 1996;42:402-29.
10. Landt M, Wilhite TR, Smith CH. A new plastic evacuated tube with plasma separator. J Clin Lab Anal. 1995;9:101-6.
11. Valenstein P. Laboratory turnaround time. Am J Clin Pathol. 1996;105(6):676-88.

Cite this article as: Maharana SK, Adak SR, Dolui RK. Quantifying the influence of varying centrifugation spin paces and timespans on serum electrolyte dynamics in an Indian clinical laboratory. Int J Res Med Sci 2024;12:1915-20.