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Review Article

A narrative review on laboratory investigations of serum creatinine and solutions to problems therein

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Abstract
Introduction: It is of utmost importance to assess renal function accurately for the proper management of renal failure. Serum creatinine level is used to calculate the creatinine clearance which is used to measure the glomerular filtration rate (GFR) and in grading different stages of chronic kidney disease (CKD). Therefore, the accurate measurement of serum creatinine is of great importance. This review aimed to explain the different methods used in serum creatinine determination and solutions to problems in some of the methods. Methods: A narrative review was conducted with 28 articles searched through the Web searching engine Google Scholar and academic research databases using keywords creatinine, isotope dilution mass spectrometry, high performance liquid chromatography (HPLC), Jaffe reaction, creatinase enzyme and bilirubin. Results: There are different methods available in the determination of serum creatinine level in the medical laboratory sector including isotope dilution mass spectrometry, HPLC, Jaffe reaction and creatinase enzymatic method. Conclusion: Jaffe reaction is the most widely used method to determine serum creatinine levels worldwide. However, this method has several drawbacks and corrective methods are adopted.

Keywords: Creatinine, Isotope dilution mass spectrometry, High Performance Liquid Chromatography, Jaffe reaction, Creatinase method, Bilirubin

Introduction

The main source of energy for most of the tissues is adenosine triphosphate (ATP) and adenosine diphosphate (ADP). However, this is insufficient for high and fluctuating energy requirements of tissues such as skeletal muscle, cardiac muscle, brain, retina, and spermatozoa. Creatine phosphate is a high-energy phosphate molecule that is metabolized to produce energy in such demanding tissues and cells. Creatine phosphate is converted to its anhydride creatinine. It is a non-enzymatic spontaneous reaction. Creatinine is excreted solely through the kidneys. Creatinine is filtered through the Bowman's capsule and is not reabsorbed by the convoluted tubules. Although a small amount of creatinine is secreted from the convoluted tubules into the filtrate, it is considered negligible [1]. Therefore, serum creatinine measurement is used as a marker of renal function

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and in calculating the glomerular filtration rate (GFR). GFR is important in assessing renal function and is used in grading different stages of chronic kidney disease (CKD). Therefore, the accurate measurement of serum creatinine is of great importance. The most common test method for serum creatinine estimation is the Jaffe reaction method as it is simple and cost-effective. However, this method has several major drawbacks due to interferences from endogenous substances such as bilirubin, ketone bodies, proteins, glucose and exogenous substances including drugs [2]. Hence, the objective of this review article was to explain the methods available in the determination of serum creatinine and solutions to problems in some of the methods.

Methodology

The specific area of the review article was determined and the literature survey was conducted by using the Web Search engine "Google Scholar", Scopus, Web of Science, PubMed and ScienceDirect. The keywords creatinine, isotope dilution mass spectrometry, high performance liquid chromatography (HPLC), Jaffe reaction, creatinase enzyme and bilirubin were used to search the relevant literature. The literature mainly included books and journal articles. The selected literature was read thoroughly and evaluated. The literature was organized by developing headings and sub-headings and the review article was structured accordingly. Data for the review were collected from October 2017 to February 2018. The literature with methods related to serum creatinine determination was included in the study. Out of 35 articles selected, seven were excluded as they were inappropriate.

Results and Discussion

Reference ranges of serum creatinine

The serum creatinine value differs according to muscle mass and males have higher serum creatinine levels than females. Similarly, the muscle mass goes down as one age, causing the serum creatinine values to drop [3]. Table 1 indicates how the reference range of serum creatinine varies with the age [4].

Table 1: Reference ranges of serum creatinine with age [4]

Age	Creatinine (mg/dL)	Creatinine (mmol/L)
Cord	0.6-1.2	53-106
Newborn	0.3-1.0	27-88
Infant	0.2-0.4	18-35
Child	0.3-0.7	27-62
Adolescent	0.5-1.0	44-88
Adult, male	0.7-1.3	62-115
Adult, female	0.6-1.1	53-97

Importance of serum creatinine determination in clinical diagnosis

Since creatinine is neither secreted nor reabsorbed by the renal tubules, the total amount of serum creatinine is excreted through renal filtration. As a result, the impaired renal function causes serum creatinine concentration to increase and it can be used as a biomarker for renal damage [4, 5].

Serum creatinine level is used to calculate creatinine clearance. Creatinine clearance can be used as a measure of the GFR. GFR is considered to be the best indicator of kidney function and it is used as a measure of renal function as well as to grade different stages of CKD [6]. Therefore, the accurate measurement of serum creatinine levels is extremely important. The most consistent and clinically relevant findings are an increase in the serum creatinine concentration and a decrease in the renal clearance of creatinine with the progression of renal disease. Both the serum concentration of creatinine and creatinine clearance have been and still are widely used as

markers of renal function, in particular of the GFR [7]. Errors in serum creatinine measurement can lead to misclassification of renal impairment which can have serious consequences [8]. Serum creatinine is also used to calculate the urea to creatinine ratio [9].

Laboratory methods used in the estimation of serum creatinine

Different methods have been used to estimate serum creatinine such as isotope dilution mass spectrometry method [10-12], HPLC method [13-15], creatinase enzymatic method [16,17], and Jaffe reaction method [16-18]. Out of all the methods, creatinase enzymatic method and Jaffe reaction methods have been used in clinical settings [16-18] while the HPLC and mass spectroscopic methods are used mainly for research purposes [13].

Isotope dilution mass spectrometry (IDMS) method

Isotope dilution mass spectrometry (IDMS) method is the gold standard method for the determination of serum creatinine levels [10,11]. In this method, a fixed amount of labelled creatinine is added to the serum specimen and the labelled and non-labelled creatinine are equilibrated at room temperature. The substances are then absorbed into an ion exchange material. After washing with water, the creatinine and the labelled internal standard are eluted with an ammonia solution and the isolated creatinine is then reacted to form trimethylsilyl derivatives. The reaction products are injected into a fused silica capillary column with combined Gas chromatography-mass spectrometry (GC-MS) with the m/z values of 332 and 329 which are characteristic of molecular masses of the labelled and non-labelled analyte and they are monitored continuously during gas chromatography. The analytical results are calculated from the isotope ratios determined in the serum specimen and a series of standards containing defined mixtures of the labelled and the non-labelled analyte [12]. This method is not readily available in most of the clinical chemistry laboratories due to economic and technical constraints [10].

High performance liquid chromatography (HPLC) method

The HPLC method is frequently used in research studies related to serum creatinine [13]. This provides a more specific method to separate and determine serum creatinine. Although both reverse-phase and ion-exchange techniques are used in HPLC, the ion-exchange method is the widely used separation technique in creatinine determinations [14]. Such HPLC method is based on a strong cation-exchange column using a dual buffer system. At pH 4.68, the majority of creatinine is present in cationic form and is retained by the resin. By increasing the pH to 7.1, it can be eluted and subsequently quantified. Cimetidine is used as the internal standard in this method. After separation, ultra-violet detection is used to quantify the creatinine level [15]. However, this method is also not used routinely due to economic and technical constraints similar to the IDMS method [13].

Creatinase enzymatic method

The creatinase method is an enzymatic method used to measure serum creatinine levels. It is a more frequently used technique in serum creatinine determinations and is more specific [16]. This enzymatic assay for creatinine involves a series of coupled enzymatic reactions including creatininase enzymatic conversion of creatinine into the product creatine which is converted to sarcosine by creatine amidinohydrolase (creatinase) followed by oxidation of sarcosine by sarcosine oxidase producing hydrogen peroxide [17]. The concentration of hydrogen peroxide produced is then measured spectrophotometrically.

The enzymatic assay shows improved specificity and requires a smaller serum volume. It does not

interfere with glucose, acetoacetate and cefoxitin. However, bilirubin has been reported to cause a slight negative interference which depends on the bilirubin and creatinine concentrations [17]. This problem has been largely overcome in the current assays, which uses a more efficient hydrogen peroxide acceptor triiodo-hydroxy-benzoic acid and includes potassium ferrocyanide and detergents to reduce bilirubin interference further [11]. The disadvantages of this method are greater cost and shorter shelf life of the creatinase enzyme [17].

Jaffe reaction method

Jaffe reaction method is the most widely used laboratory test method in the estimation of serum creatinine. It is simple and cost effective. In the Jaffe reaction, alkaline picric acid reacts with creatinine to produce a red-coloured complex. The absorbance of this complex is measured at a specific wavelength of 490-510 nm [18,19]. There are three minor variations in the Jaffe reaction namely kinetic Jaffe, fixed time Jaffe and the end-point Jaffe [16].

In the end-point Jaffe method, picric acid in an alkaline medium reacts with creatinine to form an orange-coloured complex with the alkaline picrate. Upon addition of acid reagent in the colourimetric end-point procedure, the orange-coloured complex formed above gets decolourized because of the pH change. The extent of decolourization is directly proportional to the creatinine concentration in the serum specimen where the colour change between the first and second steps is calculated. The drawback of Jaffe's end-point reaction is the interference due to non-specific substances such as proteins, ascorbic acid and keto-acids [16]. In the fixed time Jaffe method, picric acid in an alkaline medium reacts with creatinine to form an orange-coloured complex with the alkaline picrate. The intensity of the colour formed during the fixed time is directly proportional to the amount of creatinine present in the serum specimen [16]. In the kinetic method, before the picrate creatinine complex formation is monitored it minimizes interference from the fast-reacting substances such as keto-acids. Hence, subsequent measurements are done up to 120 seconds and largely refer to true creatinine values only. Other advantages of this kinetic reaction method include no deproteinization, providing results rapidly and the requirement of low serum volume [16].

Interferences in the Jaffe reaction method

All Jaffe reactions as a whole have one significant drawback due to lack of specificity [8]. It can give erroneous readings in the presence of interfering substances such as proteins, ketones and keto-acidosis, glucose, intra-lipids, and drugs such as cephalosporin. High protein concentrations tend to give a positive bias while low protein concentrations give a negative bias. Similarly, glucose has been found to cause a positive bias in the Jaffe reaction [20]. Depending upon the time of measurement, the composition of alkaline picrate and the acetoacetate level in the test sample may cause either a positive or negative interference in the kinetic Jaffé method [21]. The kinetic method can correct interferences from slow-reacting non-creatinine chromogens such as glucose, acetone and ascorbic acid. However, fast reacting substances such as α-keto compounds and cephalosporin antibiotics give positive interference [2].

Out of these interferences, bilirubin interference is of utmost clinical significance. In the alkaline medium, bilirubin is oxidized into biliverdin which shows a major decrease in the change in absorbance as measured at 510 nm. In the initial part of the reaction, bilirubin absorbs light around 500 nm which is the wavelength used in creatinine assay. As the reaction proceeds, creatinine reacts with picrate and the absorbance is increased, but as bilirubin is converted to biliverdin there is a decline in absorbance. However, the net change in

absorbance is decreased resulting in an underestimation of creatinine level [9]. Due to this interference, increased bilirubin leads to an incorrect assessment of renal function. Therefore, accurate assessment of renal function in hyperbilirubinaemic patients, in certain clinical conditions such as hepatorenal failure, multiple-organ failure, neonatal jaundice with impaired renal function, and high bilirubin coexist with the renal disease would be a problem [19]. Serum creatinine is also used to monitor the effect of nephrotoxic drugs and adjust the dose of drugs that are excreted through the kidney such as aminoglycosides. The falsely low creatinine values due to bilirubin interference may mislead clinicians in prescribing the correct dose of the medicine [2].

Several studies have been carried out to analyze this interference. Srisawasdi *et al.* [22] has conducted a broad study in which spikes pooled serum of three different creatinine concentrations with different bilirubin concentrations to compare the level of interference. Each serum specimen has been tested using two kinetic Jaffe methods and one enzymatic method. The findings of this research indicated that there is a significant difference between the mean creatinine values of one of the kinetic Jaffe methods and the enzymatic method and no such difference between the other kinetic Jaffe methods even though both methods were based on the same principle.

Similarly, a study carried out by Dimeski *et al.* [23] has compared the bilirubin interference in two Beckman automated machines using the same kinetic Jaffe principle. The authors have used the HPLC technique as a reference. Their findings showed that one machine had a significant difference in HPLC value while the other one does not. The authors have attributed this discrepancy to the different incubation temperatures used in two machines suggesting that higher temperatures minimize interference by completing the oxidation of bilirubin before the formation of the picric acid complex.

A study conducted by Weber *et al.* [16] investigated several interfering substances for creatinine measurement using the kinetic Jaffe reaction in comparison with the enzymatic method. The interference caused by bilirubin has been tested using two methods. First, the pooled patient serum had been spiked with different concentrations of bilirubin. The kinetic Jaffe reaction was used to estimate the creatinine level both before the addition of bilirubin and after the addition of bilirubin. Then, it had been calculated the average assay value for bilirubin interference as a percentage of the original concentration of creatinine and plotted against the bilirubin concentration. The authors had used these values to calculate the factor of interference. This gives the deviation of creatinine result in micromole/L per mmol of interference per liter. The results showed that the creatinase enzymatic method is more desirable as the Jaffe method shows a negative bias.

Marakala *et al.* [17] have performed a comparison between the kinetic Jaffe method and the enzymatic method in assessing creatinine values in the presence of bilirubin interference. The researchers collected normal and icteric serum specimens and the serum specimens had been tested by using the Jaffe method and creatinine enzymatic method. This had been followed by a comparison of mean values of creatinine level. The mean difference between the two methods in the test group has shown a higher variance than the control group although it is statistically non-significant.

Owen and Keevil [24] conducted an *in-vitro* study with the preparation of a concentration gradient of creatinine solution in phosphate buffered saline with 40g/L bovine serum albumin. These prepared serum specimens had been then spiked with

unconjugated bilirubin. In the second phase of the study, the icteric serum specimens with creatinine concentrations <150micromole/L had been analyzed. The bilirubin concentration of each specimen had been recorded and all the creatinine estimations had been performed by using the rate blanked compensated Jaffe method and creatinine plus enzymatic method.

In the compensated method, positive interference given by Jaffe-like chromogen can be easily corrected by subtracting 0.3 mg/dL which is an arithmetic compensation. The interference given by bilirubin can be eliminated by rate blanking which corrects the rate of change in absorbance by bilirubin from the absorbance change by the Jaffe reaction. Compensated rate-blanked Jaffe kinetic assay employs the above two major corrections. In this study, liquid chromatography-tandem mass spectrometry was used as the reference method. The results indicated that there is no significant difference in either method when compared to the reference. This may be due to the use of rate blanked compensated Jaffe method. The authors stated that unconjugated bilirubin has a slightly greater tendency of interfering than conjugated bilirubin because it is easier to oxidize unconjugated bilirubin into biliverdin than conjugated bilirubin [25].

Corrective methods adopted for the Jaffe reaction

There are several methods that have been adopted for the Jaffe reaction to overcome the interference caused by bilirubin. This involves the usage of sodium hydroxide, trichloroacetic acid, potassium ferricyanide, sodium dodecyl sulphate and Fullers Earth (FulE) [16].

Pre-incubation with sodium hydroxide

Sodium hydroxide (NaOH) acts as an oxidizing agent and converts bilirubin into biliverdin thus reducing the interference of bilirubin. Several studies have been carried out to find out the effect of NaOH in this reaction [2,10,18,19]. A study has been carried out to compare the creatinine values measured by the Jaffe kinetic method before and after incubation with NaOH, using serum specimens collected from both jaundice patients and non-jaundiced individuals. The creatinine value obtained by pre-incubation with NaOH was found to be higher than creatinine obtained without pre-incubation [19].

Another study carried out by Vaishya *et al.* [10] has compared the interference of bilirubin after pre-incubation of serum specimens with three different concentrations of NaOH in the Jaffe reaction. In this study, it had been used serum specimens with both high and low creatinine concentrations and bilirubin concentrations vary from normal to abnormal. The serum specimens had been pre-incubated in three different NaOH concentrations for five minutes prior to carrying out the kinetic Jaffe and creatinine enzymatic methods. The findings showed that when NaOH concentration increases, the gap between Jaffe and enzymatic methods is decreased [10]. These results support the findings of Chaudhary *et al.* [19].

Another study has been carried out to compare the values of the Jaffe kinetic method of three analytical kits. In this study, it has been used bilirubin spiked pooled serum and creatinine before and after the treatment with NaOH. The results showed that creatinine levels are higher in NaOH treated samples compared to non-treated samples [18].

Further, deproteinization can remove both positive and negative bilirubin interferences on creatinine obtained from all kinetic Jaffe methods because bilirubin is precipitated with the precipitant and it does not interfere with the reaction with alkaline picrate. The removal of bilirubin along with albumin can be performed either by filtration through a membrane layer or by manual acid

deproteinization [2]. Tungstic acid, sulfosalicylic acid and trichloroacetic acid can be used for acidification prior to the Jaffe reaction [26].

Trichloroacetic acid treatment

A number of studies have been carried out regarding the effect of trichloroacetic acid on bilirubin interference [2,18,20,22,26]. Srisawadi *et al.* [22] showed that there is a significant difference in the serum creatinine values with high bilirubin before and after the addition of trichloroacetic acid.

These results are supported by the study carried out by Lolekha and Sritong [26]. The study highlighted that there is a significant bilirubin interference in three kinetic Jaffe assays prior to trichloroacetic acid treatment and the enzymatic assay method. It is mentioned that the bilirubin interference increases linearly with the bilirubin concentration. The authors recommended this technique as one of the best approaches to correct all forms of bilirubin in serum creatinine [26].

The results of another study conducted to measure the creatinine values of pooled serum using the Jaffe reaction has indicated an elevation of creatinine value after treatment with trichloroacetic acid. This also indicated that trichloroacetic acid treatment corrects the bilirubin interference in the Jaffe reaction [18].

The advantage of using trichloroacetic acid is that it removes all forms of bilirubin interferences on serum creatinine [2]. However, this can be time consuming, less comfortable and error prone due to the dilution step [20].

Potassium ferricyanide treatment

Potassium ferricyanide is an oxidant that prevents bilirubin interference by the oxidation of bilirubin to biliverdin before the alkaline picrate solution is added to the serum sample [27]. Srisawasdi *et al.* [22] have carried out the same kinetic Jaffe principle using two separate analytical methods. The results suggested that ferricyanide improves the specificity of the reaction. Lolekha *et al.* [2] conducted a similar study which supported the observations made by Srisawasdi *et al.* [22]. Further, the authors have mentioned that the addition of potassium ferricyanide resulted in a false positive creatinine value [2]. This may be due to increased protein interference caused by potassium ferricyanide. A study conducted by O'Leary [27] has showed that the addition of potassium ferricyanide gives similar results to that of the rate blanked method. On the downside, potassium ferricyanide is an unstable working reagent

Addition of sodium dodecyl sulphate

Sodium dodecyl sulphate (SDS) promotes the release of bilirubin from albumin and allows complete bilirubin oxidation to biliverdin by alkaline buffer [2]. Srisawasdi *et al.* [22] and Lolekha *et al.* [26] have carried out similar studies comparing the bilirubin interference before and after the addition of SDS. Both studies have shown that SDS minimizes bilirubin interference [22,26]. They also mentioned that the action of SDS is more potent than that of potassium ferricyanide. As a disadvantage, SDS tends to precipitate at low temperatures [27].

Addition of Fullers Earth (FulE)

The addition of FulE is also a lesser-known technique for removing bilirubin interference. FulE adsorbs all the bilirubin thereby removing any interference. Weber *et al.* [16] have shown that this reduces the negative bias.

Comparison of different laboratory test methods used in serum creatinine estimation

Since the IDSM method is the gold standard, all other methods are required to be standardized using the IDSM method [28]. The IDSM method is superior to other tests with respect to sensitivity, specificity, and stability. Despite the significant

advantages of this method, it is not readily available in most of the clinical chemistry laboratories due to economic and technical constraints [10]. HPLC is frequently used as a reference method for research purposes in creatinine estimation [13]. Hence, it is high in accuracy and precision. However, similar to IDSM, the cost per test to measure serum creatinine using the HPLC method is extremely high. The cost per test in spectrophotometric methods such as creatinase and Jaffe methods is significantly lower. Both methods require a low serum specimen volume and can be carried out using a spectrophotometer as well as automated machines. When comparing the two spectrophotometric methods, the enzymatic method is more specific [16]. The creatinase reagent has a shorter shelf life when compared with the Jaffe reaction [17]. Both methods have interferences. The interferences seen in the Jaffe reaction are more significant. The interferences in the creatinase enzymatic method have been minimized by modifications done to the test kit [11]. While modifications can be performed to reduce the interferences to the Jaffe method, they are not incorporated into the test kit. The medical laboratory scientist must perform additional steps manually which can be time-consuming and tedious considering that serum creatinine is a very frequently requesting test. Although the creatinase method is clearly more beneficial, the creatinase enzymatic method is expensive when compared to the cost of the Jaffe method [17].

Conclusions

Isotope dilution mass spectrometry is the gold standard method in the determination of serum creatinine levels whereas HPLC is frequently used as a reference method for research purposes. Creatinase enzymatic method is a more frequently used technique and the Jaffe reaction is the most widely used laboratory test method in the estimation of serum creatinine. The major drawback of the Jaffe reaction is the lack of specificity due to interfering substances. The corrective methods to improve the specificity of the Jaffe reaction include pre-incubation with sodium hydroxide, trichloroacetic acid and potassium ferricyanide treatment, in addition to sodium dodecyl sulphate and Fullers Earth. The corrective methods to improve the specificity of the Jaffe reaction include pre-incubation with sodium hydroxide, trichloroacetic acid and potassium ferricyanide treatment, in addition of sodium dodecyl sulphate and FulE.

References

1. Vasudevan, D.M., Sreekumari, S., and Vaidyanathan, K. Textbook of Biochemistry for medical Students. 7th ed. Jaypee Brothers Medical Publishers (P) Ltd, 2013.
2. Lolekha, P.H., Jaruthunyaluck, S. and Srisawasdi, P. Deproteinization of serum: Another best approach to eliminate all forms of bilirubin interference on serum creatinine by the kinetic Jaffe reaction. Journal of Clinical Laboratory Analysis. 2001;15(3):116–121. DOI: https://doi.org/10.1002/jcla.1013.
3. Mason, D.L., and Assimon, M.M. Chronic kidney disease. Renal resource centre. 2013.
4. Fishbach, F., and Dunning, M. A. Manual of laboratory and diagnostic tests. 7th ed. Lippincott Williams & Wilkins Publishers, 2003.
5. Spencer, K. Analytical reviews in clinical biochemistry: the estimation of creatinine. Annals of Clinical Biochemistry.1986;23:1–25. DOI: https://doi.org/10.1177/000456328602300101.
6. Levey, A.S., Coresh, J., Bolton, K., Culleton, B., Harvey, K.S., TIkizler, T., et al. K/DOQI clinical practice guidelines for chronic kidney disease: Evaluation, classification, and stratification. American Journal of Kidney Diseases. 2002;39(2): S1-266.
7. Wyss, M., and Kaddurah-Daouk R. Creatine and creatinine metabolism. Physiological

Reviews. 2000;80(3):1107–1213.
8. Schmidt, R.L., Straseski, J.A., Raphael, K.L., Adams, A.H. and Lehman, C.M. A risk assessment of the Jaffe vs enzymatic method for creatinine measurement in an outpatient population. PLoS One. 2015;24(10(11)]. DOI: https://doi.org/10.1371/journal.pone.0143205.
9. Kroll, M.H., and Mccudden, C.R. Endogenous interferences in clinical laboratory tests. Walter de Gruyter GmbH; 2013.
10. Vaishya, R., Arora, S., Singh, B., Mallika, V., and Arora, S. Modification of Jaffe's kinetic method decreases bilirubin interference: A preliminary report. Indian Journal of Clinical Biochemistry. 2010;25(1):64–66.
11. Peake, M., and Whiting, M. Measurement of serum creatinine-current status and future goals. The Clinical Biochemistry Reviews 2006;27(4):173–184.
12. Siekmann, L., Kruse, R., and Röhle, G. Clinical Chemistry. Isotope dilution — mass spectrometry as a reference method. Springer, 1989.
13. Boot, S., LaRoche, N., and Legg, E.F. Elimination of bilirubin interference in creatinine assays by routine techniques: comparisons with a high performance liquid chromatography method. Annals of Clinical Biochemistry. 1994;31:262-266. DOI: https://doi.org/10.1177/000456329403100308.
14. Guy, J.M., and Legg, E.F. An improved cation exchange HPLC method for the measurement of serum creatinine. Annals of Clinical Biochemistry. 1990;27:223–226. DOI: https://doi.org/10.1177/000456329002700307.
15. Tsikas, D. Simplified HPLC method for urinary and circulating creatinine. Clinical Chemistry. 2004 1;50(1):201–203. DOI:https://doi.org/10.1373/clinchem.2003.024141.
16. Weber, J.A., and Van Zanten, A.P. Interferences in current methods for measurements of creatinine. Clinical Chemistry. 1991;37(5):695–700. DOI: https://doi.org/10.1093/clinchem/37.5.695.
17. Marakala, V., Avinash, S.S., Shivashankara, A.R., Malathi, M., and Kumar, A. Serum creatinine assay: Enzymatic vs kinetic Jaffe'S method. Journal of Evolution of Medical and Dental Sciences. 2012;1(4):328–334.
18. Nigam, P.K. Bilirubin interference in serum creatinine estimation by Jaffe's kinetic method and its rectification in three different kits. Indian Journal of Clinical Biochemistry. 2016;31(2):237–239. DOI: 10.1007/s12291-015-0545-6.
19. Chaudhary, S.S., Shah J. P., and Mahato, R. V. Interference of bilirubin in serum creatinine value measurement by Jaffe's kinetic method. Annals of Clinical Chemistry and Laboratory Medicine. 2015;1(1):25-28.
20. den Elzen, W.P.J., Cobbaert, C.M., Klein Gunnewiek, J.M.T., Bakkeren, D.L., van Berkel, M., Frasa, M.A.M., et al. Glucose and total protein: unacceptable interference on Jaffe creatinine assays in patients. Clinical Chemistry and Laboratory Medicine. 2018; 56(8): e185–e187. DOI: https://doi.org/10.1515/cclm-2017-1170.
21. Blass, K.G., and Ng, D.S.K. Reactivity of acetoacetate with alkaline picrate : An interference of the Jaffe reaction. Clinical Biochemistry. 1988;21:39–47. DOI: https://doi.org/10.1016/S0009-9120(88)80110-3.
22. Srisawasdi, P., Chaichanajarernkul, U., Teerakanjana, N., Vanavanan, S., and Kroll, M.H. Exogenous interferences with Jaffe creatinine assays: Addition of sodium dodecyl sulfate to reagent eliminates bilirubin and total protein interference with Jaffe methods. Journal of Clinical Laboratory Analysis. 2010;24(3):123–133. DOI: https://doi.org/10.1002/jcla.20350.
23. Dimeski, G., McWhinney, B., Jones, B., Mason, R., and Carter, A. Extent of bilirubin interference with Beckman creatinine

methods. Annals of Clinical Biochemistry. 2008;45(1):91–92. DOI: https://doi.org/10.1258/acb.2007.007079.

24. Owen, L.J., and Keevil, B.G. Does bilirubin cause interference in Roche creatinine methods? Clinical Chemistry.7;53(2):370–371. DOI: https://doi.org/10.1373/clinchem.2006.075846.
25. Chung, H., Chun, S., and Min, W. Creatinine determination with minimized interference. Journal of Laboratory Medicine and Quality Assurance. 2008;30:229–231.
26. Lolekha, P.H., and Sritong, N. Comparison of techniques for minimizing interference of bilirubin on serum creatinine determined by the kinetic Jaffé reaction. Journal of Clinical Laboratory Analysis. 1994;8(6):391–399. DOI: https://doi.org/10.1002/jcla.1860080609
27. O'Leary, N., Pembroke, A., and Duggan, P.F. A simplified procedure for eliminating the negative interference of bilirubin in the Jaffe reaction for creatinine. Clinical Chemistry. 1992;38(9):1749–1751. DOI: https://doi.org/10.1093/clinchem/38.9.1749.
28. Moore, J.F., and Sharer, J.D. Methods for quantitative creatinine determination. Current Protocols in Human Genetics. 2017. DOI: https://doi.org/10.1002/cphg.38.