

Thesis Title	Comparative Study of Bilirubin Interference on Serum Creatinine Assays by the Kinetic Jaffé Reaction With and Without the Precipitation Technique to the Enzymatic Method
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ABSTRACT

The measurement of serum creatinine provides an useful information for renal function test. However, using the kinetic Jaffé reaction for determining of creatinine are subjected to the negative bilirubin interference in jaundice specimens. The aim of this study was to compare bilirubin interference on serum creatinine assays by 5 kinetic Jaffé automated methods, with and without deproteinization, to the dry chemistry reference method. The kinetic Jaffé automated methods were the CR-T and CREA by the Synchron CX5, the CREAT and CRE by the Axon and the CREA by the Selectra. The CRSC by the Kodak Ektachem was used as a reference method.

The negative interference on creatinine value was demonstrated in both the bilirubin added pooled samples and the icteric patient samples using the kinetic Jaffé methods performed on the Synchron CX5 without serum

blank correction (CREA), the Axon (CREA and CRE) and the Selectra (CREA). Highest degree of the negative bilirubin interference was found in the CREA by the Selectra especially in the icteric patient sera with elevated bilirubin value. In contrast, the kinetic Jaffé reaction with serum blank correction (CR-T) by the Synchron CX5 was unaffected from bilirubin interference only in bilirubin added pooled sera; however, significant positive bilirubin interference was observed in the icteric patient sera ($p < 0.05$). Significant positive bias was also found on serum creatinine values assayed by the CRSC (Kodak Ektachem) in bilirubin added pooled sera compared to the based line.

Attempt to eliminate the negative bilirubin interference in all kinetic Jaffé automated methods was performed by incorporation of acid deproteinization prior to the kinetic Jaffé reaction. Different concentrations of trichloroacetic acid (TCA), 0.37 mol/L (ppt1) and 0.55 mol/L (ppt2) were used. In order to evaluate acid deproteinization technique, the results were compared to the Kodak Ektachem CRSC single-slide method. Using TCA of 0.37 mol/L (ppt1) gave a significant difference ($p < 0.05$) of creatinine value performed by the CREAT and CRE (Axon) method. TCA of 0.55 mol/L could correct bilirubin interference on creatinine value and gave no significant difference on the result compared to the reference method. ($p > 0.05$)

The precision of all automated kinetic Jaffé methods with and without deproteinization was studied. The CREA(Selectra) was found the most precise method with $CV = 0.8-2.1\%$. The other methods were less precise than CREA (Selectra) as increase in %CV as follow, CREAT and CRE by the Axon ($CV=0.4-11.5\%$ and $0.8-12.1\%$) and CR-T and CREA by

the Synchron CX5 (CV=1.3-25.0% and 1.8-19.2%). However, using TCA, 0.55 mol/L (ppt2) could improve poor precision in the CR-T (CX5) and CREA (CX5) methods.

In conclusion, introducing of TCA, 0.55 mol/L as acid precipitant prior to the automated kinetic Jaffé reactions was the best approach to overcome bilirubin interference from bilirubin in added pooled sera as well as in the icteric patient sera. In this study, we recommend acid deproteinization technique as a reference method for studying the effect of bilirubin interference on serum creatinine instead of CRSC Kodak Ektachem. Since deproteinization is very simple and convenience for all laboratory. No additional instruments are required as well.