

## INFLUENCE OF HEMOLYSIS ON ROUTINE CLINICAL CHEMISTRY TESTING

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### Abstract

While analytical standards have been developed by established quality control criteria, there has been paucity in the development of standards for the preanalytical phase<sup>1,4</sup>. Every laboratory should have a strategy for recognizing preanalytical errors. In this study there was a consistent trend towards overestimation of creatinine, Creatine kinase and potassium but glucose in the analytes was decreased significantly.

### Introduction

Preanalytical factors are the main source of variation in clinical chemistry testing and among the major determinants of Preanalytical variability; sample hemolysis can exert a strong influence on result reliability<sup>2,3</sup>. Hemolytic samples are a rather common and unfavorable occurrence in laboratory practice, as they are often considered unsuitable for routine testing due to biological and analytical interference. However, definitive indications on the analytical and clinical management of hemolyzed specimens are currently lacking. Therefore, the present investigation evaluated the influence of in vitro blood cell lysis on routine clinical chemistry testing.

### Methods

Five aliquots, prepared by serial dilutions of homologous hemolyzed samples collected from 12 different subjects and containing a final concentration of serum hemoglobin ranging from below 6.0, 6-10 more than 10 g/L, were tested for the most common clinical chemistry analytes. Lysis was achieved by subjecting whole blood to an overnight freeze-thaw cycle. From each of them Serum glucose, creatinine, creatine kinase and potassium were estimated. Results were presented in tables and graphs.

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### Results

Hemolysis interference appeared to be almost approximately linearly dependent on the final concentration of blood-cell lysate in the specimen. This generated a consistent trend towards overestimation of creatinine (Table-I, Graph-1) creatine kinase (CK) (Table-II, Graph-2) and potassium (Table-III, Graph-3) whereas mean value of glucose were substantially decreased (Table-IV, Graph-4)

### Discussion

Only recently recommendations have been published regarding the quality of samples including the definition of the optimal sample size, the use of anticoagulants and stabilizers, stability criteria regarding transport and storage and handling of hemolytic, lipemic and icteric samples<sup>5,7</sup>.

Hemolysis may interfere with chemistry tests by the following mechanisms:

1. Increased absorbance: Released hemoglobin increases absorbance in the spectral range<sup>2,3</sup>.
2. Inhibition of reactions: Released hemoglobin can directly inhibit chemical reactions.
3. Analytes release: Release of analytes found in high concentrations in red blood cells will falsely elevating the values of these analytes.
4. Enzyme release: Release of enzymes which participate in chemical reactions, e.g. Creatine kinase<sup>4</sup>.
5. Water release: Release of red blood cell water dilutes analytes<sup>8</sup>.

### Conclusion

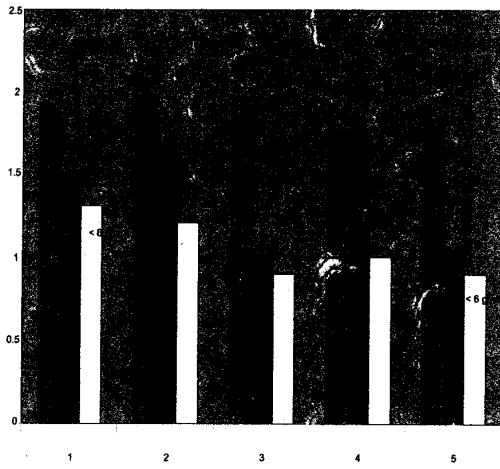
If hemolysis and blood cell lysis result from an in vitro cause, it may be suggested that the most convenient corrective solution might be quantification of free hemoglobin, alerting the clinicians and sample recollection. The rather heterogeneous and unpredictable response to hemolysis observed for several parameters prevented the adoption of reliable statistic corrective measures for results on the basis of the degree of hemolysis.

### Recommendation

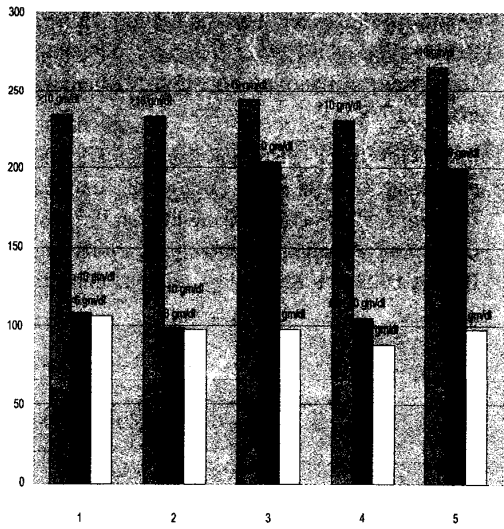
Though recommendations have been documented regarding the quality of samples including hemolysis, 9 but establishment of a quality manual addressing preanalytical variables is a prerequisite

for implementing measures to recognize and control this crucial component of laboratory quality, which cannot be detected by traditional analytical quality control procedures. From this study it may be suggested a standard operating procedure must be prepared during blood sample collection to prevent hemolysis.

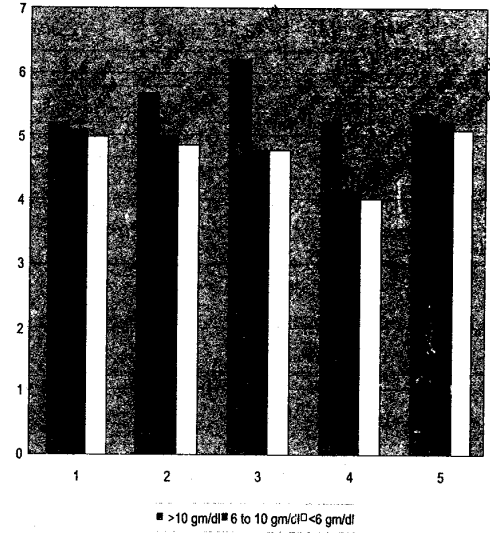
Graph-1: Creatinine in analytes in mg/dl



Graph-2: Change in creatine kinase in U/L



Graph-3: Potassium in MEQ/L



Graph-4: Change Of Glucose In Analytes

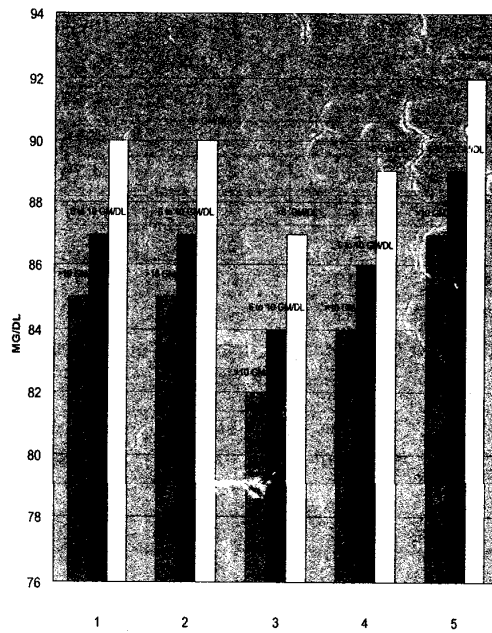


Table-I: Creatinine mg/dl analytes in different concentration of Hb

Creatinine	Hb conc >10 gm/dl	Hb conc 6 to 10 gm/dl	Hb conc <6 gm/dl
1	1.89	1.42	1.3
2	1.98	1.66	1.2
3	1.92	1.01	0.89
4	1.78	1.76	0.99
5	1.88	1.01	0.89
Mean	1.89	1.65	1.054

Table-II: Creatine Kinase U/L in analytes in different concentration of Hb

Creatine Kinase	Hb conc >10 gm/dl	Hb conc 6 to 10 gm/dl	Hb conc <6 gm/dl
1	234	108	106
2	233	99	97
3	244	204	97
4	231	105	88
5	265	201	97
Mean	241.4	143.4	97

Table-III: K<sup>+</sup> concentration Meq/L in analytes in different concentration of Hb

Potassium	Hb conc >10 gm/dl	Hb conc 6 to 10 gm/dl	Hb conc <6 gm/dl
1	5.2	5.1	4.99
2	5.66	5	4.86
3	6.2	4.78	4.79
4	5.22	4.15	4.01
5	5.31	5.24	5.1
Mean	5.518	4.854	4.75

Table-IV: Glucose mg/dl in analytes in different concentration of Hb

Glucose	Hb conc >10 gm/dl	Hb conc 6 to 10 gm/dl	Hb conc <6 gm/dl
1	85	87	90
2	85	87	90
3	82	84	87
4	84	86	89
5	87	89	92
Mean	84.6	86.6	89.6

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