Clinical Biochemistry xxx (xxxx) xxx

Contents lists available at ScienceDirect

Clinical Biochemistry

journal homepage: www.elsevier.com/locate/clinbiochem

Autoverification-based algorithms to detect preanalytical errors: Two examples

Ruhan Wei, William Légaré, Adam J. McShane *

Department of Laboratory Medicine, Cleveland Clinic, Cleveland, OH, USA

ARTICLE INFO

Keywords:
Autoverification
Preanalytical errors
Pseudohypoglycemia
Pseudohyperkalemia

ABSTRACT

The preanalytical phase of testing accounts for the majority of the errors. Software-based quality rules, such as autoverification, can assist in preanalytical error detection; therefore, preventing erroneous results from being reported. Two autoverification rules, turbidity/lipemia, and pseudohypoglycemia/pseudohyperkalemia alarms, are highlighted. Increased sample turbidity may arise from several causes outside of lipemia. Typically, this can be resolved by clarifying the sample with standard centrifugation. Truly lipemic specimens typically require higher centrifugation speeds and greater centrifugation time. At our facility, 96% of direct bilirubin (DBIL), 95% of aspartate transaminase (AST), and 98% of alanine transaminase (ALT) turbidity/lipemia alarms were found to be from sample turbidity versus lipemia. Secondly, a pseudohypoglycemia/pseudohyperkalemia rule was employed for specimens with delayed separation from cellular material. Of the total potassium results >6.0 mmol/L or glucose results <40 mg/dL (2.2 mmol/L), 30% and 50% respectively were noted to have delayed sample separation.

1. Introduction

Laboratory testing comprises the majority of the information in the electronic medical record [1]. Accurate laboratory test results are essential for high-quality patient care. In general, laboratory testing includes three phases: the preanalytical, the analytical, and the post-analytical. Although laboratory-related errors could happen at any of these phases, most of them occur outside of the actual laboratory. Recent studies have shown that approximately 46%-68% of laboratory-related errors occur in the preanalytical phase of laboratory testing [2,3].

One common preanalytical error noted in our clinical biochemistry laboratory is sample turbidity not due to lipemia. This can be produced from suspended cells or debris in the specimen, potentially from rough transportation or poor centrifugation. Another common preanalytical error is delayed separation of the sample from the cellular material. Potassium (K) cellular leakage and *in vitro* glycolysis may arise leading to falsely-low glucose (Glu, pseudohypoglycemia) or falsely-high potassium (pseudohyperkalemia) concentrations.

One way of detecting the preanalytical errors is through autoverification. Autoverification is a process where software-based algorithms perform actions on laboratory results without laboratory staff intervention [4]. The algorithms are built utilizing laboratory-defined acceptance parameters. Results are automatically released if they fall within the predetermined acceptance parameters. If results fail to meet the criteria, they will be reviewed by the laboratory staff before reporting.

At our institution, autoverification rules are used to assess the aforementioned preanalytical errors amongst others. Automated lipemia/turbidity indices (LI) are utilized to distinguish lipemia or turbidity of the specimen [5]. If the index is above the designated threshold, the specimen will be aliquoted and centrifuged to remove turbidity (false lipemia) or, if the LI was not lowered below the threshold, centrifuged at high-speed to remove lipemia (true lipemia). The second preanalytical error is detected by monitoring the specimen's Glu and K concentrations, as the extreme values may be due to nonpathological reasons such as delayed separation from the cells. If the specimen has a critically low Glu concentration (<40 mg/dL; <2.2 mmol/L) and/or a critically high K concentration (>6.0 mmol/L) and is received as whole blood and the time is greater than 4 h from collection, the order will be canceled with an explanatory comment. This study aims to 1) Determine the frequency of true and false lipemia in 3 common analytes with relatively low LI thresholds (AST, ALT, and DBIL), 2) Evaluate the operational efficiency of the aforementioned lipemia workflow, and 3) Determine the frequency of pseudohypoglycemia/pseudohyperkalemia.

* Corresponding author at: Department of Laboratory Medicine, Cleveland Clinic, 9500 Euclid Ave., L11, Cleveland, OH 44195, USA.
E-mail address: mcshana@ccf.org (A.J. McShane).

https://doi.org/10.1016/j.clinbiochem.2022.06.010
Received 30 April 2022; Received in revised form 21 June 2022; Accepted 25 June 2022
Available online 30 June 2022

2. Material and methods

2.1. Data collection

De-identified outpatient data for DBIL, AST, and ALT with their corresponding serum indices was collected for 1 month and data for K and Glu was collected for 1 year from the middleware repository (Data Innovations). Analyses for DBIL, AST, ALT, and Glu were performed on a cobas c702 chemistry analyzer (Roche Diagnostics). K analyses were performed on a cobas ISE module (Roche Diagnostics). The autoverification rules are maintained in the middleware.

2.2. Autoverification rules

2.2.1. Lipemia

ALT and AST samples with a LI beyond the analyte specific threshold were held from autoverification in the middleware. DBIL samples greater than the limit of quantitation (0.2 mg/dL; 3.4 μmol/L) and with a LI beyond the analyte specific threshold were held from autoverification in the middleware. Medical laboratory scientists were prompted to mitigate the turbidity/lipemia.

2.2.2. Pseudohypoglycemia and pseudohyperkalemia

Glu samples with a collection time greater than 4 h and results less than 40 mg/dL (2.2 mmol/L) were held from autoverification in the middleware. K samples with a collection time greater than 4 h and results greater than 6.0 mmol/L were held from autoverification in the middleware. Medical laboratory scientists were prompted to evaluate if the specimen was received as whole blood or poorly separated from cellular material.

2.3. Turbidity versus lipemia

Samples with an initial LI greater than their corresponding threshold but dropped below the threshold after routine centrifugation (6 min at 2000 RCF) were classified as false lipemia. Samples that subsequently required high-speed centrifugation (30 min at 21,000 RCF) to clarify were classified as true lipemia. Samples were aliquoted before any repeat centrifugation. Only DBIL results above the limit of quantitation (0.2 mg/dL; 3.4 μmol/L) were included in the analysis due to lipemia/turbidity interference causing false elevation.

2.4. Pseudohyperkalemia and pseudohypoglycemia

Specimens received separated from cellular material with Glu concentration less than 40 mg/dL (2.2 mmol/L) were classified as critical hypoglycemia, and specimens with K concentration greater than 6.0 mmol/L were classified as critical hyperkalemia. Specimens with a critically low Glu or a critically high K concentration received as whole blood and greater than 4 h from the time of collection were classified as suspected pseudohypoglycemia or pseudohyperkalemia, respectively. These were identified via a standard comment in the middleware repository. Total hypoglycemia was calculated as the sum of the pseudohypoglycemia and critical hypoglycemia, and total hyperkalemia was calculated as the sum of the pseudohyperkalemia and critical hyperkalemia. Rejected specimens for other concerns, such as hemolysis, are not included in the data.

2.5. Data analysis

Data analysis was performed using Microsoft Excel. This study was approved by the Cleveland Clinic Institutional Review Board as an exempt study.

3. Results and discussion

3.1. Turbidity versus lipemia

Inadequate time of blood sampling after the meal, intravenous lipid infusions, and type 2 diabetes are some of the most common reasons for lipemia [6,7]. LI is designed as a photometric estimation of specimen interference from lipids. The semi-quantitative analysis is based on the ratio of sample absorbance at various wavelengths multiplied by a correction factor [8]. Because of the testing mechanism, the assay is affected by other entities within the specimen that also absorbs light with similar wavelengths, such as suspended cells, debris, paraproteins, and dyes [9–11]. Therefore, turbidity from improper transportation or sample processing can cause a high LI without the sample being lipemic. DBIL, AST, and ALT are photometric, high-volume tests with relatively low LI thresholds. Consequently, they are more susceptible to this pre-analytical error.

In one month, there were 2,348, 33,674, and 34,184 results released for DBIL, AST, and ALT, respectively (Table 1). Out of these results, 96% of the DBIL and 99% of the AST and ALT were without turbidity/lipemia holds in the middleware. Therefore, only 4% of the DBIL and 1% of the AST and ALT required laboratory staff intervention for elevated LI. Upon investigation of the specimens held for turbidity/lipemia, 4% of the DBIL, 5% of the AST, and 2% of the ALT were true lipemia (i.e., required high-speed centrifugation to clarify), whereas the rest of the specimens were false lipemia (i.e., clarified with routine centrifugation). A total of 44 h were spent investigating the suboptimal specimens every month assuming approximately 3 mins and 6 mins to handle a false lipemic and a true lipemic specimen, respectively. Efforts, such as proper transportation, storage, and centrifugation, to reduce false lipemia would increase the efficiency of the performing laboratory [12]. If the autoverification rule was not implemented and every specimen required a visual inspection (around 0.5 min), 588 h would be needed. Visually inspecting every sample is impractical at a large academic health center, not to mention that visual estimation of the incidence and amount of turbidity is highly unreliable [13]. This adds a burden to the laboratory; it also jeopardizes the accuracy of laboratory results and the quality of patient care.

3.2. Pseudohyperkalemia and pseudohypoglycemia

At our institution, outpatient test volumes for K and Glu were 49,074 and 48,811 per month. Around 0.14% of K and 0.06% of Glu were released as critical values each month. Critical results are considered life-threatening and require immediate notification. Without the implementation of a pseudohyperkalemia/pseudohypoglycemia autoverification rule, this would increase to 0.20% for K and 0.12% for Glu (Fig. 1). A 43% increase in potassium and a 50% increase in glucose critical value notifications would be required. Assuming 5 mins to notify a single provider, this would result in 5 additional hours of notifications per month. Patient care will be adversely affected if the patient receives the improper medical intervention based on poor quality laboratory results. Also, the patient may be unnecessarily burdened to seek immediate medical attention causing undue stress to the patient and the healthcare system.

3.3. Limitations

The pseudohypoglycemia and pseudohyperkalemia rules as outlined would not hold specimen results with falsely-low glucose values or falsely-high potassium values if not critical. If the laboratory is able to incorporate cellular material separation status into the autoverification rules, this would provide an opportunity to prevent further erroneous results from being released.

False-lipemia and true-lipemia were classified via centrifugal force needed to clear the specimen. This study assumes a short duration,

Table 1
One month of DBIL, AST, and ALT specimens with LI quality rule alarms. True lipemia specimens required high-speed centrifugation to clarify where false lipemia specimens were cleared with conventional centrifugation. Note: DBIL data only includes specimens above the limit of quantitation (0.2 mg/dL; 3.4 μmol/L).

	Total	Above Lipemia Index Threshold	Above Lipemia Index Threshold %	True Lipemia	False Lipemia	% True Lipemia	% False Lipemia
DBIL	2348	91	4%	4	87	4%	96%
AST	33,674	380	1%	19	361	5%	95%
ALT	34,184	384	1%	9	375	2%	98%

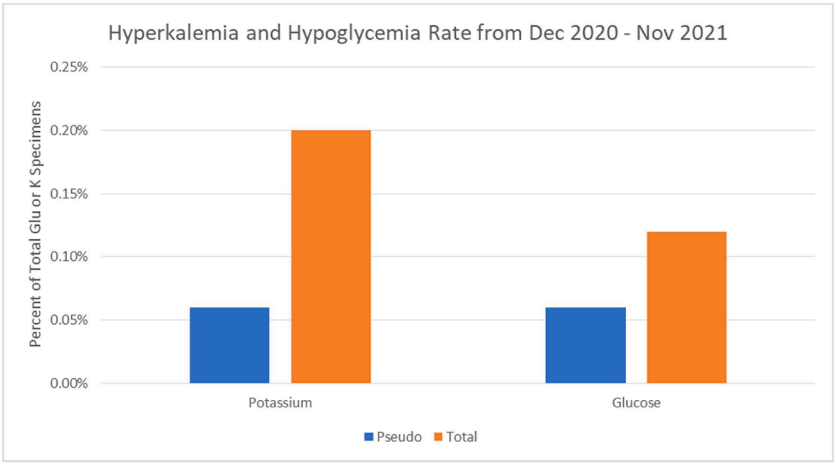

Fig. 1. Percent of pseudohyperkalemia and pseudohypoglycemia alarms and all >6.0 mmol/L potassium and <40 mg/dL (2.2 mmol/L) glucose results normalized to the total number of K or Glu results.

conventional centrifugation would be unable to remove lipemia where higher speed centrifugation is typically required [14].

4. Conclusion

The preanalytical phase is a significant component of laboratory testing but remains a major source of laboratory-related errors. A turbidity/lipemia algorithm noted the overwhelming majority of LI alarms were from sample turbidity unrelated to lipemia. Reduction in preanalytical sample turbidity would be of benefit to the laboratory. Further, a pseudohypoglycemia/pseudohyperkalemia rule prevented the release of results with prolonged contact with cellular material. The implementation of quality-based autoverification rules may minimize the potential errors and therefore improve the value of laboratory results.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

References

[1] M.J. Hallworth, The "70% claim": what is the evidence base? Ann. Clin. Biochem. 48 (Pt 6) (2011) 487–488, https://doi.org/10.1258/ACB.2011.011177.
[2] G. Lippi, J.J. Chance, S. Church, P. Dazzi, R. Fontana, D. Giavarina, K. Grankvist, W. Huisman, T. Kouri, V. Palicka, M. Plebani, V. Puro, G.L. Salvagno, S. Sandberg, K. Sikaris, I. Watson, A.K. Stankovic, A.-M. Simundic, Preanalytical quality improvement: from dream to reality, Clin. Chem. Lab Med. 49 (7) (2011), https://doi.org/10.1515/CCLM.2011.600.
[3] M. Plebani, Errors in clinical laboratories or errors in laboratory medicine? Clin. Chem. Lab Med. 44 (6) (2006) 750–759, https://doi.org/10.1515/CCLM.2006.123.
[4] Riben M. Laboratory Automation and Middleware. *Surg. Pathol. Clin.* 2015;8(2): 175-186.
[5] Y.Z. Zheng, R.W. Pearce, A.J. McShane, Lipemia Interference for ALT and AST: effect from native lipid and commercial lipid emulsion-supplemented samples, J. Appl. Lab Med. 5 (4) (2020) 817–819, https://doi.org/10.1093/JALM/JFAA025.
[6] N. Nikolac, Lipemia: causes, interference mechanisms, detection and management, Biochem. Med. 24 (1) (2014) 57–67, https://doi.org/10.11613/BM.2014.008.
[7] S. Mainali, S.R. Davis, M.D. Krasowski, Frequency and causes of lipemia interference of clinical chemistry laboratory tests, Pract. Lab Med. 8 (2017) 1–9, https://doi.org/10.1016/J.PLABM.2017.02.001.
[8] Z. Shajani-Yi, J.H. Nichols, Preanalytical variation, in: W. Clarke, M.A. Marzinke (Eds.), Contemporary Practice in Clinical Chemistry, 4th ed., Elsevier, Cambridge, 2020, p. 9.
[9] E. Fliser, K. Jerkovič, T. Vidovič, M. Gorenjak, Original papers Investigation of unusual high serum indices for lipemia in clear serum samples on Siemens analysers Dimension, Biochem. Med. 22 (3) (2012) 352–362.
[10] I. Munnix, M. Raijmakers, W. Oosterhuis, H. Kleinveld, Detection of a monoclonal gammopathy by lipemia-index measurement, Ned. Tijdschr Klin. Chem. Labgeneesk. 34 (4) (2009) 248–249.
[11] D. Darby, C. Broomhead, Interference with serum indices measurement, but not chemical analysis, on the Roche Modular by Patent Blue V, Ann. Clin. Biochem 45 (3) (2008) 289–292.
[12] G. Lippi, G. Banfi, S. Church, et al., Preanalytical quality improvement. in pursuit of harmony, on behalf of European Federation for Clinical Chemistry and Laboratory Medicine (EFLM) Working group for Preanalytical Phase (WG-PRE), Clin. Chem. Lab Med. 53 (3) (2015) 357–370, https://doi.org/10.1515/CCLM-2014-1051/PDF.
[13] M. Glick, K.W. Ryder, S. Glick, J.R. Woods, Unreliable visual estimation of the incidence and amount of turbidity, hemolysis, and icterus in serum from hospitalized patients, Clin. Chem. 35 (5) (1989) 837–839.
[14] M.-J. Castro-Castro, B. Candás-Estébanez, M. Esteban-Salán, P. Calmarza, T. Arrobas-Velilla, C. Romero-Román, M. Pocoví-Mieras, J.-Á. Aguilar-Doreste, Removing lipemia in serum/plasma samples: a multicenter study, Ann. Lab Med. 38 (6) (2018) 518–523.