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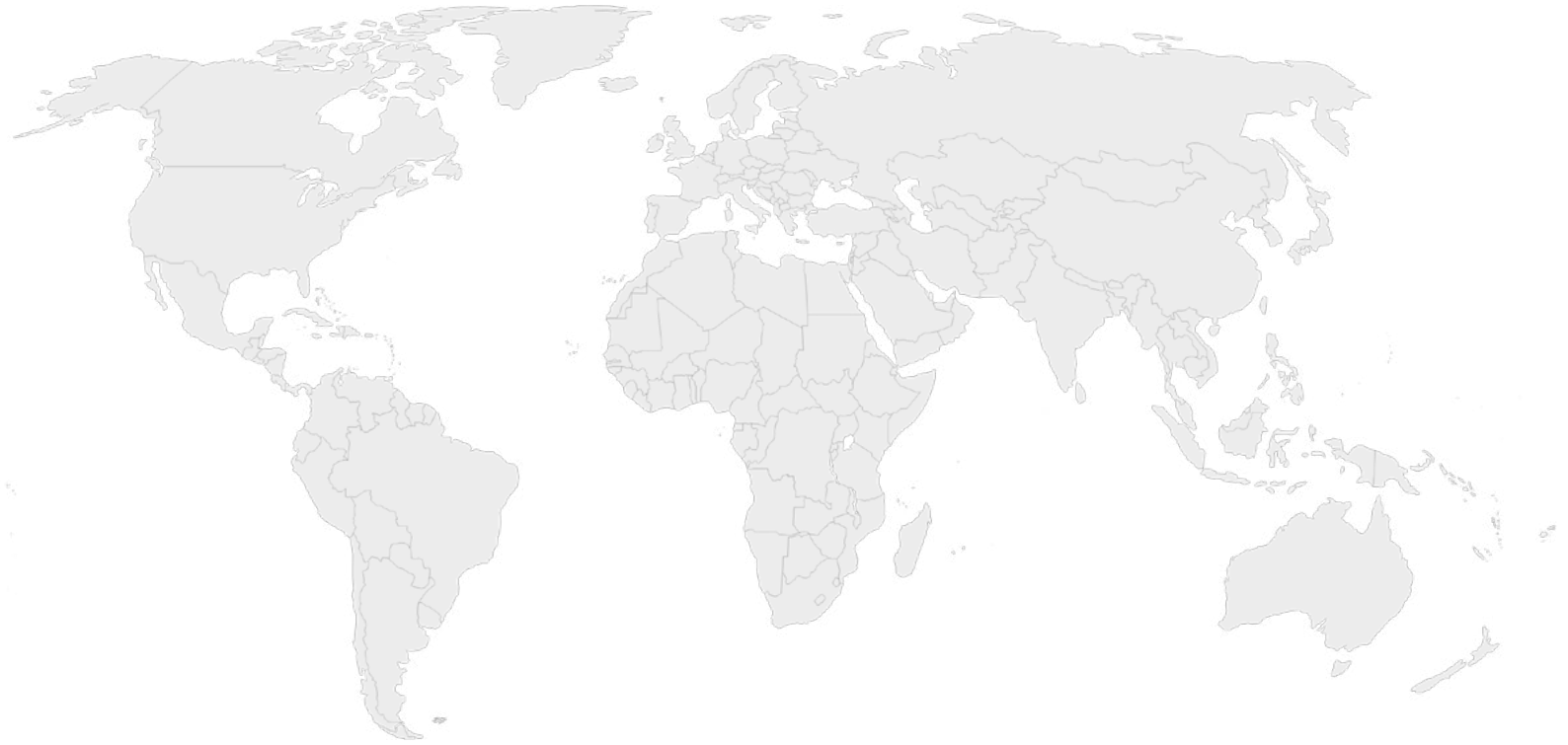

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In this issue

Original Article

Optimization of the diagnostic capacity of traditional biomarkers in muscle damage and its use in the diagnosis of dermatomyositis and polymyositis

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Keywords

creatine kinase, aldolase, muscle damage, dermatomyositis, polymyositis

Abstract

BACKGROUND-AIM

Creatine kinase (CK) and aldolase are markers traditionally used in the study of muscle damage (MD). As CK determination is more specific to muscle damage, the demand for both determinations in routine laboratory tests would entail an extra cost.

METHODS

Retrospective observational study conducted between 2019-2020. CK and aldolase concentrations from 218 patients were studied.

ROC curves were analyzed for CK and aldolase for muscle damage detection. Cut-off values were selected for both strategies. Specifity of CK and aldolase for dermatomyositis or polymyositis diagnosis in our population was studied using the McNemar's test.

RESULTS

The area under the ROC curve (AUC) for total CK was 0.716 (95%CI: 0.651-0.775), for CK in males it was 0.703 (95%CI: 0.592-0.799), and for CK in females was 0.719 (95%CI: 0.636-0.793). For aldolase, AUC was 0.505 (95%CI: 0.437-0.573). Optimized cut-off points for each determination were: 112 U/L for CK in men, with a sensitivity of 73.9% (95%CI: 51.6-89.8) and a specificity of 49.2% (95%CI: 35.9-62.5); 88 U/L for CK in women, with a sensitivity of 75.0% (95%CI: 57.8-87.9) and specificity of 50.5% (95%CI: 40.4-60.6); and 5.6 U/L for aldolase, with a sensitivity of 61.0% (95%CI: 53.2-68.8) and a specificity of 38.8% (95%CI: 26.5-52.6).

Regarding the individuals diagnosed with dermatomyositis or polymyositis, 66.7% and 44.4% of them were correctly classified as pathological by CK and aldolase results, respectively. McNemar's test did not reveal significant differences.

CONCLUSION

The determination of CK offers a better diagnostic performance of MD and, in addition, does not present significant differences regarding the determination of aldolase in cases of polymyositis and dermatomyositis. Therefore, the single determination of CK would be sufficient for MD screening.

INTRODUCTION

The evaluation of serum muscle enzymes is common in the study of patients presenting muscle weakness or myalgias in whom muscle myopathy is suspected. These enzymes can be elevated in inflammatory myopathies (such as polymyositis and dermatomyositis), infectious myopathies, dystrophinopathies, rabdomyolysis, and metabolic myopathies, among others [1,2]. Myositis is described as any condition that causes inflammation in skeletal muscles. Its main symptom is muscle weakness, whereas muscular pain is not always present. It can be caused by different entities including infections, autoimmune diseases, and muscle injury [3]. In this context, polymyositis is a rare inflammatory disease that causes muscle weakness, thereby affecting both sides of the body. Having this disease can make it difficult to climb stairs, stand after sitting, lift, or reach for places above your head. Commonly, it affects adults between 30 and 50 years and is more common in African than in Caucasian population. In addition, women are affected more often than men. Signs and symptoms usually appear gradually, over weeks or months. Risk of polymyositis is higher in case of lupus, rheumatoid arthritis, scleroderma, or Sjögren's syndrome [4-6]. While polymyositis has no cure, treatment ranging from medications to physical therapy can improve muscle strength and function [7]. Dermatomyositis, however, as well as being a rare inflammatory disease characterized by muscle weakness, also causes a particular skin rash. It can affect adults and children. In adults it occurs between the age of 45 and 65 years. In children, it usually appears between 5 and 15 years old [4,5,6]. Dermatomyositis also affects women more than men. It has no cure, but there may be periods when symptoms improve. Meanwhile, treatment can eliminate the skin rash and help regain muscle strength and function [7]. Recently, dysregulation in microRNA which normally regulates the immune system has been found in cases of myositis, but further research is needed in order to understand the role of microRNAs in these cases [8].

In clinical laboratories, aldolase and creatine kinase (CK) are two enzymes whose analytical determination has the study of muscle damage as its main usefulness [1]. Aldolase is an enzyme that acts on glucose, allowing energy to be obtained. It is distributed throughout the body, mainly in muscle tissue. The main clinical use of aldolase determination is in the diagnosis and monitoring of musculoskeletal diseases [9,10]. CK is an enzyme located in the inner mitochondrial membrane, myofibrils, and cytoplasm of myocytes. It is involved in the storage and transfer of energy and is the most widely used enzyme for diagnosing and tracking muscle diseases. Its highest serum concentrations are found in response to muscle damage for which it is the most sensitive marker [10,11]. However, regarding polymyositis and dermatomyositis, an increase in aldolase concentration has been described with no increase in CK concentration [12,13,14]. Myoglobin is a protein that is also released in muscle damage together with CK. However, it has a short half-life and its concentrations start to decrease when CK concentrations are still elevated, thus it is less sensitive if the muscle damage is not recent. Also, myoglobin is most commonly used to study the risk of acute renal injury since high concentrations of this protein can exceed the protein-binding capacity of the plasma causing nephrotoxicity and renal failure. On top of that, determination of this protein is more expensive than other enzymes that appear to be more informative, like CK. That is why it is not a commonly used determination for muscle damage study in clinical laboratories [15,16]. Currently, it is common practice for clinicians to ask for the simultaneous measurement of these two biomarkers in cases of suspected muscle damage. Nevertheless, in our experience, aldolase and CK appear elevated when there is muscle damage. Hence, as CK determination is more specific to muscle damage, the demand for both determinations in routine laboratory tests would entail an extra cost [17,18]. The aim of this study was to optimize the decision limits for CK and aldolase as muscle damage biomarkers in our laboratory, as well as to compare their diagnostic accuracy for this kind of pathologies. Based on the results, a study of the economic cost derived from an incorrect demand for these markers was also carried out.

MATERIAL AND METHODS

A retrospective observational study was performed at Hospital Universitari Son Espases (Palma de Mallorca, Spain), a tertiary level hospital that provides healthcare for an approximate population of 325,000 individuals. The assessed period was between January 2019 to December 2020. Analytical data were obtained from the laboratory information system Gestlab (Indra, Spain), and the clinical information (diagnostic data) was extracted from the hospital information system Millenium (Cerner Corporation), after obtaining approval from the Ethics Board of our institution (Research Ethics Committee of the Balearic Islands (IB 5121/23 PI)). This study is in compliance with the ethical principles for medical research involving human subjects, in accordance with the Declaration of Helsinki. The current decision limits (upper reference limits) in our hospital for serum CK and Aldolase were provided by the manufacturers, Abbott Diagnostics (USA) and BioSystems (Spain), respectively: CK (Male (M) 200 U/L, Female (F) 168 U/mL); Aldolase (7 U/L). Serum CK was quantified by spectrophotometry on the Alinity c platform (Abbott Diagnostics, USA). Long-term imprecision (6 months) was satisfactory, with coefficients of variation (CVs) of 2.5% at 78 U/L, and 3.5% at 660 U/L; whereas aldolase enzyme levels were quantified by spectrophotometry using the BA25 equipment (BioSystems, Spain) with an analytical coefficient of variation of 4.8% (concentration similar to the current decision limit).

Inclusion criteria

Inpatient laboratory requests were considered if they included simultaneous CK and aldolase determinations. Only the first request for each individual with both biomarkers was included in the study. Results were considered pathological when the clinical information reflected an entity of muscle damage as the final diagnosis for that episode.

Diagnostic comparison: CK vs Aldolase

For each individual, age and sex were recorded, and different groups were established according to the diagnosis reflected on the medical records: individuals with non-muscle damage versus individuals with reported muscle damage. In order to determine which diagnostic strategy had a better discriminating power for muscle damage, a receiving operating characteristics (ROC) curve analysis was performed and the area under the curve (AUC) was quantified. Based on the results, cut-off values (for CK (by sex) and aldolase) were selected for the maximization of the diagnostic performance. Cut-off values with maximal discrimination power were used to calculate Cohen´s Kappa index and the odds ratio (OR) for muscle damage. Once cut-off points were optimized, we checked whether aldolase was the more specific enzyme for dermatomyositis or polymyositis diagnosis in our population, as described in the literature. In order to verify this fact, we evaluated the proportion of well-classified (dermatomyositis/polymyositis versus non-muscle damage) individuals according to CK and aldolase concentrations.

Economic implications

Using only one enzyme, either CK or aldolase when muscle damage is suspected, would lead to economic savings. Evaluation of such savings was also performed retrospectively, considering a unit price of 2 euro for each determination (CK and aldolase).

Statistical Analysis

AUCs were compared using the Bamber methodology. Comparison of proportions for related samples was performed using the McNemar test. SPSS v.24 (IBM Corporation, USA) and MedCalc v.19.3 (Belgium) software were used for all calculations. Statistical significance was set at 0.05.

RESULTS

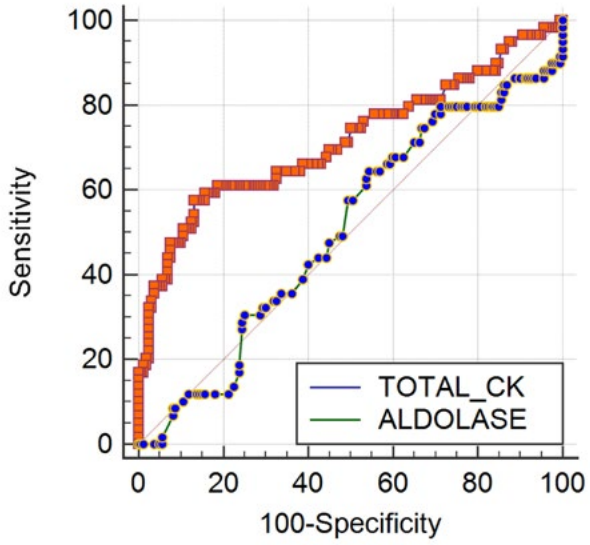
During the period studied, in a total of 1502 individuals, CK and aldolase determinations for muscle damage were simultaneously requested, 218 of whom were inpatients. Sixty-eight of them (31%) were considered pathological in view of the final diagnosis reflected in their medical records. The diagnosis included mainly: myopathy (15%), myelitis (9%), dermatomyositis and polymyositis (26%), Behçet syndrome (7%), myalgia (14%), rheumatoid arthritis (12%), physical exercise (1.5%), Kawasaki syndrome (1.5%), ELA (1,5%), chronic inflammatory demyelinating polyneuropathy (1.5%), seizures (1.5%), and Duchenne syndrome (1.5%). Anthropometric and biochemical features are described in Table 1. The area under the ROC curve (AUC) for total CK was 0.716 (95% CI: 0.651-0.775). However, by sex, for CK in males it was 0.703 (95% CI: 0.592-0.799), whereas AUC for CK in females was 0.719 (95% CI: 0.636-0.793). For aldolase, AUC was 0.505 (95% CI: 0.437-0.573). Comparison of ROC curves of total CK and aldolase is shown in Figure 1. In addition, statistically significant differences were found between CK (male and female) and aldolase AUCs (p-value<0.01). Optimized cut-off points for each determination were: 112 U/L for CK in men, with a sensitivity of 73.9% (95% CI: 51.6-89.8) and a specificity of 49.2% (95% CI: 35.9-62.5); 88 U/L for CK in women, with a sensitivity of 75% (95% CI: 57.8-87.9) and specificity of 50.5% (95% CI: 40.4-60.6); and 5.6 U/L for aldolase, with a sensitivity of 61% (95% CI: 53.2-68.8) and a specificity of 38.8% (95% CI: 26.5-52.6). Upon applying the optimized cut-off points of CK and aldolase, Cohen's Kappa index for women was 0.51, while for men it was 0.19, for any diagnosis of muscle damage. Similarly, the following odds ratios were obtained for general muscle damage: CK (M) 2.7 (95% CI: 1.0 – 7.8); CK (F) 3.1 (95% CI: 1.4 – 7.1); aldolase 1.0 (95% CI: 0.5 – 1.8). After studying individuals diagnosed with dermatomyositis or polymyositis (26% pathological diagnosis), and in consideration of the CK results, 66.7% (12/18) presented a value higher than the established cut-off value and, therefore, were correctly classified as pathological. However, regarding aldolase results, only 44.4% (8/18) were correctly classified as pathological. Despite these differences, McNemar's test did not reveal significant differences between both biomarkers (p-value = 0.344). Over the two years of the study, a total of 1502 aldolase determinations were co-processed with CK, amounting to a total cost of 3004 euro for this extra determination.

Table 1: Descriptive statistics of the studied population

Diagnosis	Sex (Male/Female)	Age: Median (years, IQR*)	Median CK (U/L, IQR)	Median Aldolase (U/L, IQR)
Pathological (31%)	22/46	42 (13-53)	122 (82-318)	6.3 (4.6-8)
Non Pathological (69%)	58/92	11 (6-21)	94 (67-142)	6.1 (4.6-8.7)

*IQR: interquartile range

Figure 1: ROC curves (total CK and aldolase) for diagnosis of muscle damage



DISCUSSION

CK and aldolase are two analytical determinations that are used for the diagnostic study of muscle damage, in which an increase in these biomarkers can occur. It is described that the only cases in which an elevation of the enzyme aldolase could occur without elevation of the enzyme CK are those of dermatomyositis and polymyositis [12-14]. Currently, it is common to simultaneously determine both biomarkers to study muscle damage, which could lead to unnecessary health expenditure if both determinations offer the same information. According to the literature, this happens only in the specific cases of dermatomyositis and polymyositis, the incidence of which is not very high – presenting 4.7-5.2/106/year for dermatomyositis and 3.7-4.1/106/year for polymyositis in Spain, in general [19]. To study the diagnostic capacity of both biomarkers, 218 patients were studied, from whom the ROC curves of CK, separated by sex, and aldolase were analyzed. Analysis of the ROC curve of both biomarkers indicated that the determination of aldolase for muscle damage offered no information and, further, the diagnostic performance for muscle damage of CK was significantly higher than the diagnostic performance of aldolase. This fact is also highlighted through the odds ratio obtained for both biomarkers. Furthermore, according to international recommendations [20], each laboratory should establish its own reference values and decision limits. In this way, new cut-off values of both determinations were obtained. These decision limits were used to study the concordance of both biomarkers for muscle damage. However, the fact that the cut-off point obtained for CK is around the median of the normal value, highlights both, that the reference values provided by the manufacturer are not well characterized or do not suit to our population, so it would be necessary to further study and properly characterize our population with good reference ranges. And that, even if CK seems to be a better biomarker for the study of muscle damage than aldolase, it is far from being an ideal biomarker due to its lack in distinguishing between pathological and non-pathological patients.

Finally, since in the cases of dermatomyositis and polymyositis an elevation of aldolase has been described without the presence of CK elevation, the differences between both markers were evaluated for 18 cases of dermatomyositis and polymyositis. Positive results were observed in 12 of the 18 cases for CK, representing 66.7% of cases, and in 8 for aldolase, representing 44.4%. No significant differences were found, in our population, between the measurement of CK and aldolase for the described cases of polymyositis and dermatomyositis. These results show that, in our population, the use of aldolase for detection of cases of dermatomyositis and polymyositis does not offer more information than the use of CK. However, it would be necessary to continue the study by increasing the number of cases to confirm these results. Globally, this study has some limitations, mainly related to its retrospective nature and the confidence in the records obtained from the LIS and hospital information system. Besides, the number of cases studied was low, especially in the cases of dermatomyositis and polymyositis. From an economic point of view, the single request of CK determination in case of diagnostic suspicion of muscle damage would be enough and would lead to financial savings. It is important to highlight that the study was carried out during a period corresponding to the COVID-19 pandemic, with a greater expense being likely in non-pandemic operating conditions of the laboratory.

CONCLUSION

The results obtained in this study indicate that the determination of CK offers a better diagnostic performance of muscle damage and, in addition, does not present significant differences regarding the determination of aldolase in cases of polymyositis and dermatomyositis, in which an increase in aldolase without an increase in CK has been described. Therefore, the single determination of CK would be sufficient for muscle damage screening and would mean a decrease in health expenditure. Based on the 'right care' philosophy, clinical laboratories need to offer not only true results, but also become a cornerstone in the optimization of resources.

Research Funding

This study has not received any type of public or private funding.

Author contributions

Sara Sanchez Asis: Conceptualization, Investigation, Methodology, Software, Supervision, Validation, Visualization, Data curation, Formal analysis, Writing original draft - review & editing. **María Cristina Gomez Cobo:** Formal analysis. **David Ramos Chavarino:** Formal analysis. **Beatriz Garcia Garcia:** Formal analysis. **Isabel Llompart Alabern:** Supervision, Validation. **Jose Antonio Delgado Rodriguez:** Investigation, Methodology, Supervision, Validation, Visualization, Writing - review & editing.

Conflict of interest

All authors declare no conflicts of interest.

Ethics approval and consent to participate

The study was approved by the Ethics Board of our institution [Research Ethics Committee of the Balearic Islands (IB 5121/23 PI). This study is in compliance with the ethical principles for medical research involving human subjects, in accordance with the Declaration of Helsinki.

Consent for publication

Consent to submit has been received explicitly from all co-authors, as well as from the responsible authorities. Authors whose names appear on the submission have contributed sufficiently to the scientific work and therefore share collective responsibility and accountability for the results.

Data availability

This is a study performed at Hospital Universitario Son Espases (Palma de Mallorca, Spain). Analytical data were obtained from the laboratory information system GestLab (Indra, Spain), and the clinical information was extracted from the hospital information system Millennium (Cerner Corporation, USA).

Acknowledgements

Not applicable

References

1. Brancaccio P, Lippi G, Maffulli N. Biochemical markers of muscular damage. Clin Chem Lab Med. 2010;48(6):757–67.
2. Baig S, Paik JJ. Inflammatory muscle disease - An update. Best Pract Res Clin Rheumatol. 2020;34(1):101484.
3. Milone M. Diagnosis and Management of Immune-Mediated Myopathies. Mayo Clinic Proceedings. 2017;92(5):826–37.
4. Dalakas MC, Hohlfeld R. Polymyositis and dermatomyositis. Lancet. 2003;362(9388):971–82.
5. Bohan A, Peter JB. Polymyositis and dermatomyositis. N Engl J Med. 1975;292(8):403–7.
6. Yang S-H, Chang C, Lian Z-X. Polymyositis and dermatomyositis - challenges in diagnosis and management. J Transl Autoimmun. 2019;2(100018):100018.
7. Sasaki H, Kohsaka H. Current diagnosis and treatment of polymyositis and dermatomyositis. Mod Rheumatol. 2018;28(6):913–21.
8. Parkes JE, Thoma A, Lightfoot AP, Day PJ, Chinoy H, Lamb JA. MicroRNA and mRNA profiling in the idiopathic inflammatory myopathies. BMC rheumatol. 2020;4(1):25.
9. Dalby AR, Tolan DR, Littlechild JA. The structure of human liver fructose-1,6-bisphosphate aldolase. Acta Crystallogr D Biol Crystallogr. 2001;57(11):1526–33.
10. Panteghini M, Bais R. Serum enzymes. Tietz Textbook of Clinical Chemistry and Molecular Diagnostics. 6ª Edition Elsevier. 2017.
11. Bong SM, Moon JH, Nam KH, Lee KS, Chi YM, Hwang KY. Structural studies of human brain-type creatine kinase complexed with the ADP-Mg2+-NO3- -creatine transition-state analogue complex. FEBS Lett. 2008;582(28):3959–65.

12. Nozaki K, Pestronk A. High aldolase with normal creatine kinase in serum predicts a myopathy with perimysial pathology. J Neurol Neurosurg Psychiatry. 2009;80(8):904–8.
13. Mercado U. Dermatomyositis with normal creatine kinase and elevated aldolase levels. J Rheumatol. 2002;29:2242–3.
14. Carter JD, Kanit KS, Vasey FB, Valeriano-Marcet J. Dermatomyositis with normal creatine kinase and elevaded aldolase levels. J Rheumatol. 2001;28:2366–7.
15. B.M.I. Cabral, S.N. Edding and J.P. Portocarrero et al., Rhabdomyolysis, Disease-a-Month. 2020;66(8):101015.
16. Schifman RB, Luevano DR. Value and Use of Urinalysis for Myoglobinuria. Arch Pathol Amp Lab Med. 2019; 143(11):1378-81.
17. Mammen A. Autoimmune muscle disease. Handb Clin Neurol. 2016;133:467-84.
18. Nagaraju K, Gladue HS, Lundberg IE. Inflammatory diseases of muscle and other myopathies. In: Firestein GS, Budd RC, Gabriel SE, McInnes IB, O'Dell JR, editors. Kelley and Firestein's Textbook of Rheumatology. 10th Edition Elsevier. 2017:1461–88.
19. Vargas-Leguás H, Selva-O'Callaghan A, Campins-Martí M, Hermosilla Pérez E, Grau-Junyent JM, Martínez Gómez X, et al. Polimiositis y dermatomiositis: incidencia en España (1997-2004). Med Clin (Barc). 2007;129(19):721–4.
20. CLSI EP 28-A3C (Formerly C 28-A3). Defining, Establishing, and Verifying Reference Intervals in the Clinical Laboratory; Approved Guideline. 3rd Edition. 2010.

Original Article

Validation of Becton Dickinson Barricor™ tubes for use with Abbott Alinity™ and Siemens Atellica® highly sensitive cardiac troponin I measuring systems

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Keywords

troponin I, hemolysis, clinical validation, lithium heparin plasma, sample quality, turnaround time.

Abstract

BACKGROUND: BD Barricor™ tubes have been proposed to decrease laboratory turnaround time (TAT). We analytically validated and then clinically verified these tubes for use with Abbott Alinity™ and Siemens Atellica® highly sensitive cardiac troponin I (hs-cTnI) assays.

METHODS: hs-cTnI measurements were undertaken in paired Barricor™ and in-use PSTII™ tubes on both systems. 359 matched samples with hs-cTnI levels between 3 and 15,000 ng/L (Atellica® values) were used to assess the hemolysis rate and make method comparisons. 599 paired patient samples were collected on emergency department (ED) admission to compare the performance of the rapid acute myocardial infarction (AMI) rule-out strategy based on hs-cTnI concentrations lower than recommended thresholds (<4 ng/L Alinity™; <5 ng/L Atellica®) when different tubes and systems were employed.

RESULTS: No between-tube differences in hemolysis rate were seen when free hemoglobin concentrations in plasma samples were ≥0.25 g/L, even if PSTII™ showed a significant increase of hemolysis rate vs. Barricor™ (31% vs. 22%, p=0.007) when a lower cut-off for hemolysis (≥0.11 g/L) was employed on the Atellica® detection system. The alternate use of these tubes did not influence the hs-cTnI results obtained from either of the two assays, which remained markedly biased (~40%) irrespective of the tube used. The expected optimal ability of very low hs-cTnI values on ED admission for ruling out AMI was confirmed by using both systems regardless of the tube type.

CONCLUSIONS: Barricor™ and PSTII™ tubes can provide analytically equivalent hs-cTnI results when used on either Alinity™ or Atellica® hs-cTnI assays.

Introduction

According to current clinical recommendations [1,2], cardiac troponins I or T are the preferred biomarkers for the detection of myocardial injury and key diagnostic elements in diagnosing acute myocardial infarction (AMI), especially in non-ST-segment elevation myocardial infarction (NSTEMI) [2]. As such, an accurate quantification of these biomarkers, through a detailed knowledge of the preanalytical, analytical, and clinical performance of available assays, is crucial in avoiding erroneous results, potentially leading to wrong diagnosis and inappropriate management of patients with suspected AMI [3].

For measuring cardiac troponin I, several highly sensitive immunoassays are now marketed, which are available on fully automated, high-throughput platforms. Among others, the Alinity™ i STAT High Sensitive Troponin-I and the Atellica® IM High-Sensitivity Troponin I assays have an analytical turnaround time (TAT) <15 min. TAT is an important indicator of laboratory service performance [3,4] and a lag ≤60 min from the time of receipt of blood tubes in the central laboratory to troponin result reporting to clinical wards has been recommended. Meeting appropriate TAT to ensure timeliness in reporting troponin results is a prerequisite for the implementation of fast-track algorithms recommended in clinical guidelines [2]. Furthermore, a rapid TAT for troponin testing can facilitate early diagnosis, timely initiation of treatment, and improved patient outcomes [5,7]. The use of plasma allows for faster blood sample processing compared to serum as the clotting time is eliminated. Therefore, plasma providing tubes are widely used for troponin measurements in emergency departments (ED) [5]. In 2016, Becton Dickinson (BD) introduced a novel type of lithium heparin tube which contains a mechanical separator of blood cells (Barricor™ Lithium Heparin Plasma Blood Collection Tubes) as opposed to classical gel separation, e.g., in Plasma Separator Tubes II (PSTII™). Importantly, Barricor™ tubes require a shorter centrifugation time than PSTII™ tubes (3 minutes as opposed to 10 minutes), potentially offering a further effective reduction of TAT when used in acute clinical setting [6,7]. Although Barricor™ tubes are available since a few years, data evaluating the Barricor™ tube as an alternate sample type for cardiac troponin I measurements are still limited [8,10]. Consequently, in this study we validated the use of Barricor™ tubes on the Alinity™ and Atellica® highly sensitive cardiac troponin I (hs-cTnI) measuring systems, by comparing them with in-use PSTII™ tubes.

Our study sought to evaluate: a) the frequency of hemolysis in using these two tubes, as quantified by the hemolysis index (HI) on both platforms; b) the impact, if any, of the tubes on the assay comparison; and c) the influence of the tubes, if any, on rapid AMI rule-out strategy employing recommended cut offs of hs-cTnI assays.

Materials and Methods

Analytical study design

Sample collection and processing

359 Barricor™-PSTII™ lithium heparin paired samples were collected from hospitalized patients with routinely ordered hs-cTnI testing. All study participants provided informed, written consent prior to adding the Barricor™ tube to PSTII™ tube needed for hs-cTnI measurements. No exclusion criteria, other than insufficient samples (blood volume <3 mL) or with troponin concentrations <3 ng/L, i.e., the lower limit of measurement range, were applied. Blood from each individual was collected into a 3 mL Barricor™ tube (ref. 365044) and then in a 3 mL PSTII™ tube (ref. 367374). Following blood drawing, tubes were gently inverted 4-5 times before immediate transfer to the laboratory, where they were centrifuged according to vendor recommendations, i.e., at 4000g for 3 min for Barricor™, using a dedicated swing bucket DASH Apex 6 Compact STAT centrifuge (Drucker Diagnostics), and at 2000g for 10 min for PSTII™ using a swing bucket Eppendorf centrifuge 5702, respectively. HI measurements and hs-cTnI testing of plasma samples were performed within 30 min following centrifugation. The study was conducted from June 2019 to March 2021.

Characteristics of hs-cTnI assays and HI measurement systems

The hs-cTnI measuring interval was 3 to 50,000 ng/L for Alinity™ i and 3 to 25,000 ng/L for Atellica® IM, respectively. According to the IFCC recommendations to use whole numbers (no decimals) for hs-cTnI reporting in clinical practice [11], all values were rounded up or down to the nearest whole number. The lower end of the measuring interval was defined by the limit of quantitation for Atellica®, rounded to the smallest integer common on both systems. The overall 99th percentile URLs were 26 ng/L and 45 ng/L for Alinity™ and Atellica®, respectively. The respective HI were measured on the Alinity™ c and Atellica® CH. The performance of these photometric determinations has been previously described in detail [12-13]. It should be noted that Alinity™ c permits a quantitatively accurate estimate of free hemoglobin (fHb) concentrations in plasma, while in the Atellica® CH the quantitative results are bucketed into index intervals to report in qualitative terms. Based on previous experiences establishing 0.25 and 1.00 g/L of fHb as the clinically most important thresholds for hemolysis interference [14], we used for Alinity™ the corresponding HI of 25 and 100, and for Atellica® the index ranges of 1 (0.11-1.30 g/L fHb) and 2 (1.31-2.49 g/L fHb) to establish the hemolysis rates by using the two evaluated tubes.

Method comparison studies

The between-assay comparisons using the same tube and the between-tube intra-assay comparisons were carried out using the same 359 matched samples having hs-cTnI concentrations covering the range between 3 and 15,000 ng/L (Atellica® values). To highlight correlation results in the most important clinical range, comparisons were also done on a subgroup of 300 paired samples with hs-cTnI ranging from 3 to 300 ng/L, a value previously identified as threshold for immediate rule-in at patient admission when using Abbott Architect™ platform [15]. All hs-cTnI measurements were performed in duplicate and the mean value was calculated. Method comparison studies were undertaken in compliance with CLSI EP09-A3 standards [16].

Clinical study design

Study population and blood sampling

We prospectively enrolled 599 unselected patients admitted to the ED with chest pain of possible cardiac origin and suspected AMI and with pain onset within the last 6 h. Paired Barricor™ and PSTII™ samples were collected at patient presentation and processed promptly. Testing of paired plasma samples from each patient was performed firstly on the routinely employed Atellica® and shortly after on the Alinity™ measuring system.

AMI rule-out strategy on ED admission and study endpoint

An hs-cTnI-based AMI rule-out strategy using previously recommended thresholds (<4 ng/L for Alinity™ [2], and <5 ng/L for Atellica® [17], was employed using single sample results obtained from these two types of primary tubes. The primary endpoint was to compare the performance of the aforementioned strategy to rule-out AMI using Barricor™ and PSTII™ tubes.

AMI adjudication

After review of relevant clinical information and the standards of the University Hospital No.1 in Bydgoszcz, cases were adjudicated for AMI (including type 1 and 2) following the Fourth Universal Definition of AMI consensus recommendations [1]. The adjudicators (cardiologists EL and MJ) were blinded to the investigational Alinity™ and Atellica® hs-cTnI. The hs-cTnI Atellica® PSTII™ results were available to the adjudicators during the hospitalization period of the patient.

Compliance with ethical standards

In compliance with the ethical principles for medical research involving human subjects the study protocol was approved by the Bioethics Committee of the Collegium Medicum in Bydgoszcz, Nicolaus Copernicus University in Torun, Poland in agreement with the Helsinki declaration on ethical standards [No. 402/2019]. All patients provided written informed consent for enrollment in the study.

Statistical analysis

Categorical variables were compared using the chi-square test and their percentage share in the entire group of results was determined. The compliance with the normal distribution was checked using the Shapiro-Wilk W test. Normally distributed values were presented as mean and standard deviation (SD). Values whose distribution deviated from normal were presented as medians with the 25th and 75th percentiles.

Depending on whether a given distribution met the criteria of a normal distribution, the obtained values were compared using the Student's t-test or, if these conditions were not met, the U-Mann-Whitney test. Comparison of hs-cTnI concentrations in BD Barricor™ and BD PSTII™ tubes on Alinity i and Atellica IM analyzers was made using Deming regression analysis with the determination of the Pearson linear correlation coefficient. Scatter plots of hs-cTnI concentrations measured by the Alinity™ and Atellica® measuring systems were generated, slopes and intercepts [with corresponding 95% confidence intervals (CI) were estimated, and between-assay percentage differences calculated. Diagnostic sensitivities and negative predictive values (NPV), with corresponding CIs, were calculated to examine the diagnostic performance of AMI rule-out strategy for both the hs-cTnI Alinity™ and Atellica® assays and using the two different tubes. Differences in the proportion of results obtained with both tubes were compared. A p value <0.05 was considered statistically significant. All statistical analyzes were performed using MedCalc v.20.023 software (MedCalc Software, Ostend, Belgium).

Results

Comparisons of hemolysis rates using Barricor™ and PSTII™ tubes, as automatically detected by the Alinity™ c and Atellica® CH systems, are shown in Table 1. No between-tube differences were seen in the hemolysis rate on either platform when a medium degree of hemolysis, defined as fHb ≥1.00 g/L, was detected. The impact of different thresholds in detecting low degree hemolysis on these two platforms, i.e., Alinity™ ≥0.25 g/L and Atellica® ≥0.11 g/L, may explain the significant increase in the percentage of hemolyzed samples detected by Atellica® compared to Alinity™ when using PSTII™ tubes (31% vs. 22%, p=0.011). It is indeed possible that some part of samples reported with HI ≥1 on Atellica® were not detected as hemolyzed on Alinity™, as an fHb range between 0.11 and 0.25 g/L approached the threshold for a low degree of hemolysis on Alinity™. The same observation may explain significant increases in low-degree hemolysis rate (31% vs. 22%, p=0.007) on Atellica® when PSTII™ tubes were compared with Barricor™ tubes, indicating that PSTII™ may increase the number of samples displaying relatively low fHb (between 0.11 to 0.25 g/L). A direct relationship of hemolysis indices of the two blood collection tubes and the two analytical systems is presented in Supplementary Figure 1.

Table 1: Hemolysis rates in 359 paired BarricorTM and PSTIITM lithium heparin plasma samples as detected by automatic hemolysis index on the two measuring systems. Chi-square test was used for comparisons.

	Low-degree hemolysis*	P value between tubes	P value between platforms
Alinity™ Barricor™	19%	0.229	0.307†
Alinity™ PSTII™	22%		
Atellica® Barricor™	22%	0.007	0.011§
Atellica® PSTII™	31%		
	Medium-degree hemolysis†		
Alinity™ Barricor™	5%	0.613	0.247†
Alinity™ PSTII™	6%		
Atellica® Barricor™	3%	0.327	0.494§
Atellica® PSTII™	5%		

* H-index ≥0.25 g/L for Alinity™ and ≥1 g/L (quantitative values in the range 0.11-1.30 g/L) for Atellica®.
† H-index ≥1 g/L for Alinity™ and ≥2 g/L (quantitative values in the range 1.31-2.49 g/L) for Atellica®.
‡ Differences between Alinity™ Barricor™ and Atellica® Barricor™
§ Differences between Alinity™ PSTII™ and Atellica® PSTII™

Comparisons between plasma samples obtained from the two types of tubes run on either the Alinity™ or the Atellica® hs-cTnI measuring systems are shown in Supplemental Figure 2. The regression equations revealed near equivalence between tubes, showing that the alternate use of the two types of tubes did not influence hs-cTnI results obtained by each of the two measuring systems. Supplemental Figure 3 shows the between-assay comparisons using the same tube. Regression analyses remained the same regardless of tube type employed for obtaining plasma. As expected, the two hs-cTnI systems showed non-comparable results, with Alinity™ giving hs-cTnI results markedly lower than Atellica®. Slopes and intercepts for both comparisons indicated both constant and proportional difference. Difference plots confirmed the substantial between-assay bias, in average ranging from 38% to 40%, that was however unaffected by the employed type of tube (Figure 1). We also compared distributions of Barricor™ and PSTII™ paired samples according to the specific categories on either system: <4ng/L Alinity™, <5ng/L Atellica®, between these low values and the assay-specific 99th percentiles, and >99th percentiles which showed no statistically significant differences between tubes regardless of the system used (Supplemental Table 1). A rapid AMI rule-out strategy using the two hs-cTnI measuring systems and the two types of tubes was applied to 599 patients admitted to ED with suspected AMI. Baseline characteristics of these patients and corresponding hs-cTnI concentrations measured using both systems and tubes are shown in Table 2. The average age in patients with suspected AMI was 68.7 years and the majority of them were men. Patients finally diagnosed with AMI were of similar age to patients with AMI excluded. However, AMI was diagnosed significantly more frequently in men compared to women. The average age of women was 71.9 ± 11.7 years, while that of men was 66.8 ± 11.8 years and the observed difference was statistically significant (P <0.001). The median hs-cTnI concentrations in the study group did not differ statistically significantly between both tubes on the same analyzer. Statistically significant differences were observed between median hs-cTnI concentrations obtained in the same tubes using two measurement systems. As expected, median hs-cTnI concentrations were statistically significantly higher in patients with confirmed AMI compared to patients with AMI excluded.

Table 2: Baseline characteristics of patients with suspected AMI and corresponding hs-cTn concentrations.

	All patients (n=599)	Non-AMI patients (n-530)	AMI patients (n=69)	p value
Age [years]	68.7 ± 12.0	68.7 ± 12.0	68.5 ± 11.9	0.988
Sex [female]	227 (38%)	209 (39%)	18 (26%)	<0.001
Alinity I BD Barricor™ hs-cTnI [ng/L]	10 (5-33)	9 (5-26)	41 (13-428)	<0.001
Alinity i BD PSTII™ hs-cTnI [ng/L]	10 (5-33)	9 (5-26)	39 (11-406)	<0.001
Atellica IM BD Barricor™ hs-cTnI [ng/L]	15 (8-52)	14 (8-41)	86 (24-747)	<0.001
Atellica IM BD PSTII™ hs-cTnI [ng/L]	15 (8-52)	13 (7-40)	88 (23-751)	<0.001

Using this approach, excellent sensitivities and NPV were obtained, irrespective of the tube type employed (Table 3).

Table 3: Diagnostic performance of acute myocardial infarction rule-out strategy using recommended cut-offs for hs-cTnI on Alinity™ (<4 ng/L) and Atellica® (<5 ng/L) measuring systems with Barricor™ and PSTII™ tubes. 95% confidence intervals in parentheses.

	Alinity™ Barricor™	Alinity™ PSTII™	Atellica® Barricor™	Atellica® PSTII™
Sensitivity	100% (95-100)	100% (95-100)	99% (92-100)	99% (92-100)
Negative Predictive Value	100% (94-100)	100% (93-100)	98% (90-100)	98% (90-100)

Sixty-nine (12%) patients were finally diagnosed with AMI. In reporting hs-cTnI values lower than the recommended cut-offs for the AMI rule-out procedure, we found overall agreement between both hs-cTnI measuring systems and both tube types employed (Supplemental Table 2). However, it should be noted that whilst Alinity™ did not show any false negative results (i.e., AMI patients with hs-cTnI <4 ng/L on ED admission), Atellica® displayed two false negative results in two different AMI patients, with both showing a hs-cTnI value of 4 ng/L, one when using the Barricor™ tube, the second the PSTII™.

Discussion

In this study, we successfully performed an analytical validation and clinical verification of BD Barricor™ tubes for use on both the Alinity™ and the Atellica® hs-cTnI measuring systems in comparison with the in-use PSTII™ tubes. Our results clearly demonstrate that BD Barricor™ tubes displayed an acceptable analytical and clinical performance on both hs-cTnI measuring systems and that they are fit for purpose in an emergency setting for patients presenting with chest pain. Initially, we assessed the effect of Barricor™ tubes on the incidence of hemolysis. In vitro hemolysis is an undesirable, though relatively common problem, which may adversely affect patient management. Our data revealed that the incidence of a medium degree of hemolysis, as defined by a fHb concentration of ≥1.00 g/L, detected in our setting in approximately 5% of plasma samples, was independent of the blood collection tubes used. On the other hand, PSTII™ showed a significant increase in rate of hemolysis when compared with Barricor™ tubes if a lower cut-off for hemolysis (≥0.11 g/L) was employed on the Atellica® CH detection system, demonstrating a slightly better quality of plasma being obtained from Barricor™ tubes and indicating that PSTII™ may increase the number of samples showing a very low hemolysis degree, which were however still within the physiological fHb range [18]. Other authors also noted a significantly lower frequency of hemolysis and a better quality of plasma in Barricor™ when compared to PSTII™ tubes, even if different centrifugation protocols for Barricor™ tubes were employed in the various studies [6, 7, 19, 20]. Our results showed that significant between-tube differences occurred where HI was relatively low. Increases in hemolysis severity cancelled out these differences. Nevertheless, interference thresholds for HI differed slightly in employed analyzers, further highlighting the need for establishing standardized and universally accepted

criteria for detecting and reporting HI among manufacturers as well as defining significant assay interference thresholds according to both the analytical criteria and clinical relevance. Our study is the first to demonstrate that hs-cTnI results obtained in both BD lithium heparin tubes fully agreed across the measuring range within the same system (Alinity™ or Atellica®). Dupuy et al. previously compared highly sensitive cardiac troponin T (hs-cTnT) measurements in both lithium heparin tubes, revealing only a negligible difference between the tubes [21]. Although these data were limited by the small sample size (samples were collected from only 9 patients and 5 healthy individuals) and the narrow range of evaluated hs-cTnT values (3-159 ng/L), those authors concluded that the use of Barricor™ tubes with a shorter centrifugation time did not affect hs-cTnT measurements, suggesting that both Barricor™ and PSTII™ tubes can be used interchangeably [21]. Our results confirmed that both tubes can also provide analytically equivalent results when used on either of the evaluated hs-cTnI measuring systems. As demonstrated in previous studies [22], marked differences in hs-cTnI concentrations between the two systems were observed even when using the same sample tube. The lack of both a commutable reference material and the different antibody configuration of assays may explain these differences. While Atellica® hs-cTnI is traceable to an internal standard manufactured using human heart homogenate, AlinityTM hs-cTnI is believed traceable to National Institute of Standards and Technology SRM 2921 through an alignment to the ArchitectTM assay, even though specific information on traceability implementation and the assessment of SRM 2921 commutability is not available [23]. It is however noteworthy, that when between-assay comparisons were focused on the 3 to 300 ng/L range, i.e., the hs-cTnI values having the most clinically important role in classifying patients with suspected AMI and in which assay harmonization is most desirable, the intercept, indicating the existence of a constant bias due to the different selectivity of antibody sandwiches in the two assays, was reduced to 5 ng/L. This supports the concept that differences between hs-cTnI assays could be markedly reduced by the availability of a commutable reference material utilized as a common calibrator in commercial systems [24]. Finally, in employing an AMI rapid single-measurement rule-out strategy using hs-cTnI with assay-specific cut-offs, we have clinically validated Barricor™ tubes [2,17]. With this approach, we showed that the rule-out ability for both evaluated hs-cTnI systems was excellent, with high NPV irrespective of the employed tube type. Sensitivities and NPVs found in this study corresponded with those found in previous reports using hs-cTnI assays, in which the safety and clinical efficacy of early AMI rule-out strategies using marker concentrations near to the assay limit of detection were evaluated [25, 27]. As recently highlighted in guidelines released by the UK National Institute for Health and Care Excellence (NICE) [28], no specific diagnostic accuracy evidence has been published to date for AlinityTM hs-cTnI. In selecting the AlinityTM cut-off for the optimal ruling out of AMI in this study, we followed the NICE suggestion in using the recommended ArchitectTM cut-off, as Alinity™ uses the same method principle and reagents as an alternative version of the test, the only marked difference being that they are run on different analyzers [28]. With accord to this approach, several studies previously evaluated the diagnostic performance of very low ArchitectTM hs-cTnI concentrations on ED admission, with their results being consistent with our Alinity™ data [29,31]. Only three studies evaluating AMI rule-out power at patient ED admission have been published [17,22,32]. Our study obtained the same sensitivity figure (99%) as Sandoval et al. [17], and that approached (98%) by Chapman et al. [32], with both studies using the same 5 ng/L cut-off. Our NPV was slightly lower possibly influenced by differences in cohort patient numbers and recruitment protocols. It has been shown that the NPV may be higher in enrolled populations that have a higher prevalence of non-ischemic myocardial injury [33]. Although AMI rule-out performance may vary in principle among measuring systems [34], our study showed that the rule-out strategy based on a single sample with very low hs-cTnI concentrations measured at ED admission did not alter its outcome whichever of the two tubes were employed. Nevertheless, we observed two false negative hs-cTnI results with using Atellica® measuring system. Two hypothesis may explain this undesirable situation. Importantly, is the possible variability of hs-cTnI measurements due to the imprecision of the assay at these very low concentrations. Similarly, rounding results to the smallest integer may introduce a bias of an estimator. There are several limitations which should be acknowledged. Firstly, we did not provide a detailed characteristics of patients with suspected AMI as we focused specifically on the rapid rule-out strategy in non-selected ED patients with suspected AMI which may limit the generalizability of our findings. Secondly, our study is limited by the lack of information on a 30-day risk of major adverse cardiovascular events (MACE). Furthermore, the quality of plasma obtained using BD Barricor tubes has not been evaluated by plasma residual cells. In conclusion, we demonstrated that Barricor™ tubes performed equally well, both analytically and clinically, when compared with PSTII™ tubes. Assuming a potential reduction in laboratory TAT, without impairment of the quality of laboratory service, Barricor™ tubes may provide an advantage which is of particular interest in hs-cTnI testing where a more expeditious availability of results has a central clinical role.

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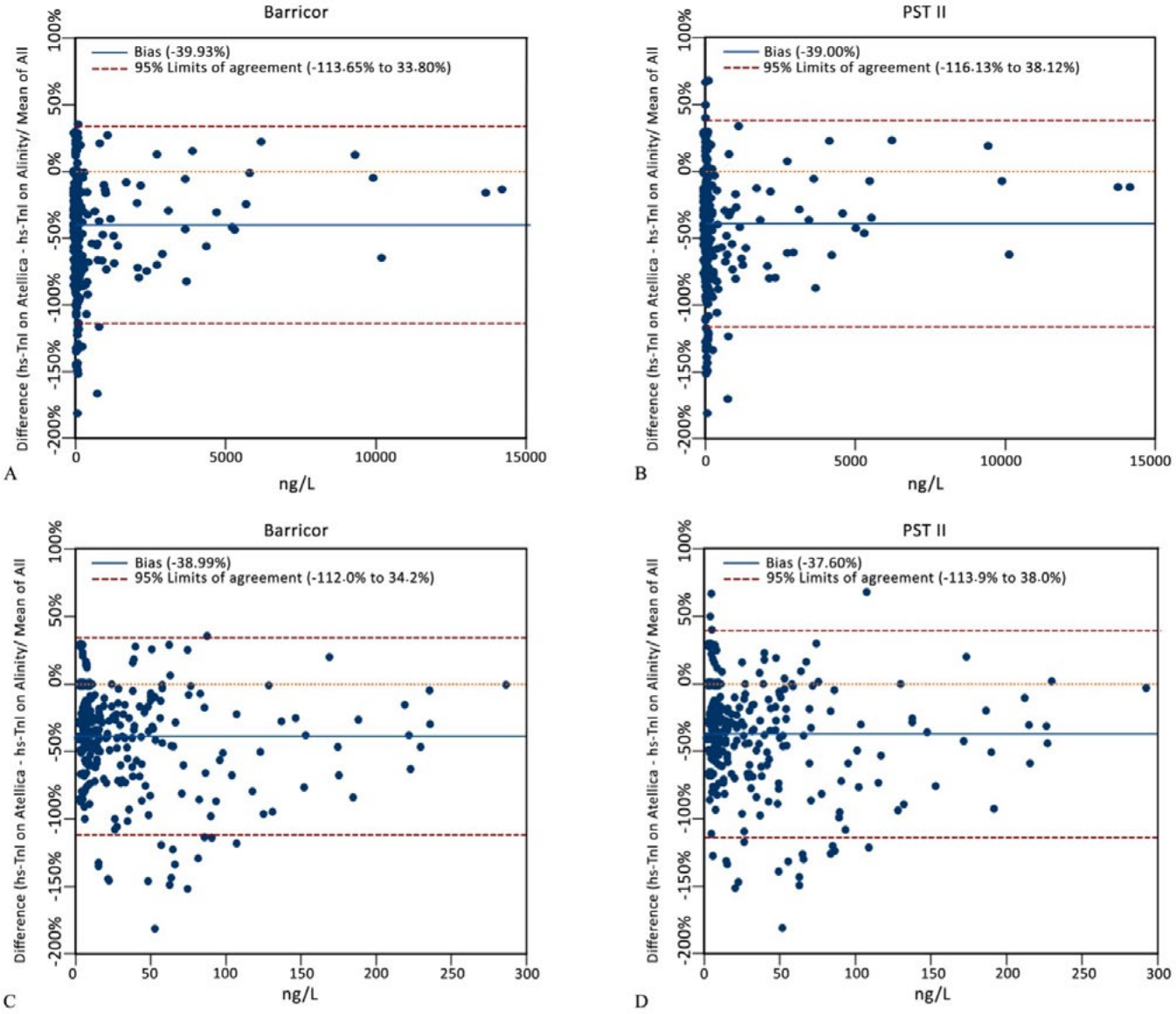
All authors declare that they have no conflicts of interest.

Figure 1: Difference plots for hs-cTnI results: (A) in PSTII™ tubes on the Alinity™ and Atellica® systems for hs-cTnI in the range 3 to 15,000 ng/L; (B) in Barricor™ tubes on the Alinity™ and Atellica® systems for hs-cTnI in the range 3 to 15,000 ng/L; (C) in PSTII™ tubes on the Alinity™ and Atellica systems for hs-cTnI in the range <300 ng/L; (D) in Barricor™ tubes on the Alinity™ and Atellica® systems for hs-cTnI in the range <300 ng/L.



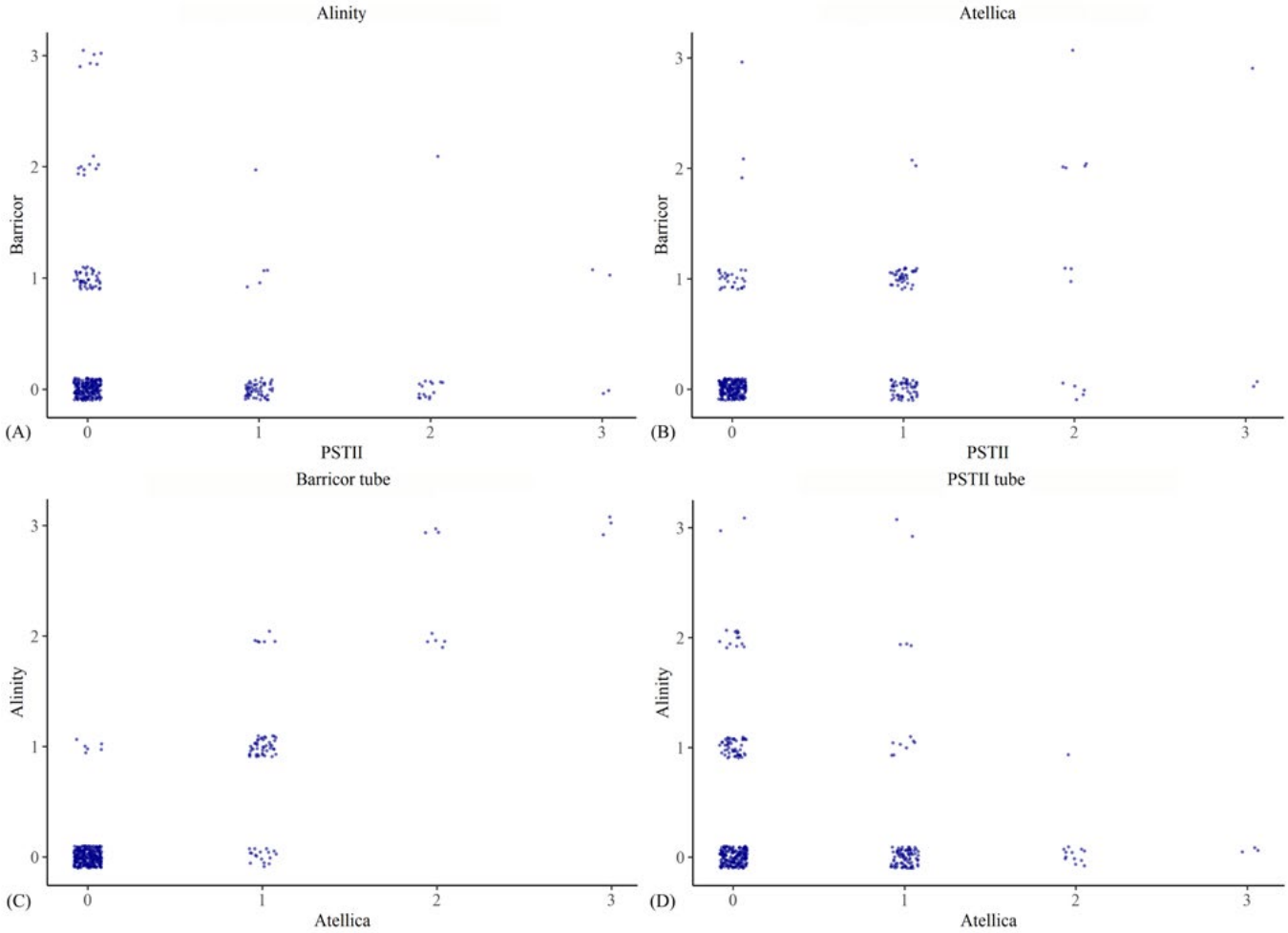
Supplemental Table 1: Distributions of Barricor and PSTII paired samples according to the categories on either system: <4ng/L Alinity, <5ng/L Atellica, between these low values and the assay-specific 99th percentiles and > 99th percentiles. Chi-square test was used for comparisons.

	Barricor™ No. of samples	**PSTII™ No. of samples**	**P value for comparisons between tubes**
Alinity™ <4 ng/L	37	29	0.301
Atellica® <5 ng/L	37	37	1.000
Alinity™ 4 ng/L – 26 ng/L	181	185	0.765
Atellica® 5 ng/L – 45 ng/L	179	179	1.000
Alinity™ >26 ng/L	141	145	0.760
Atellica® >45 ng/L	143	143	1.000

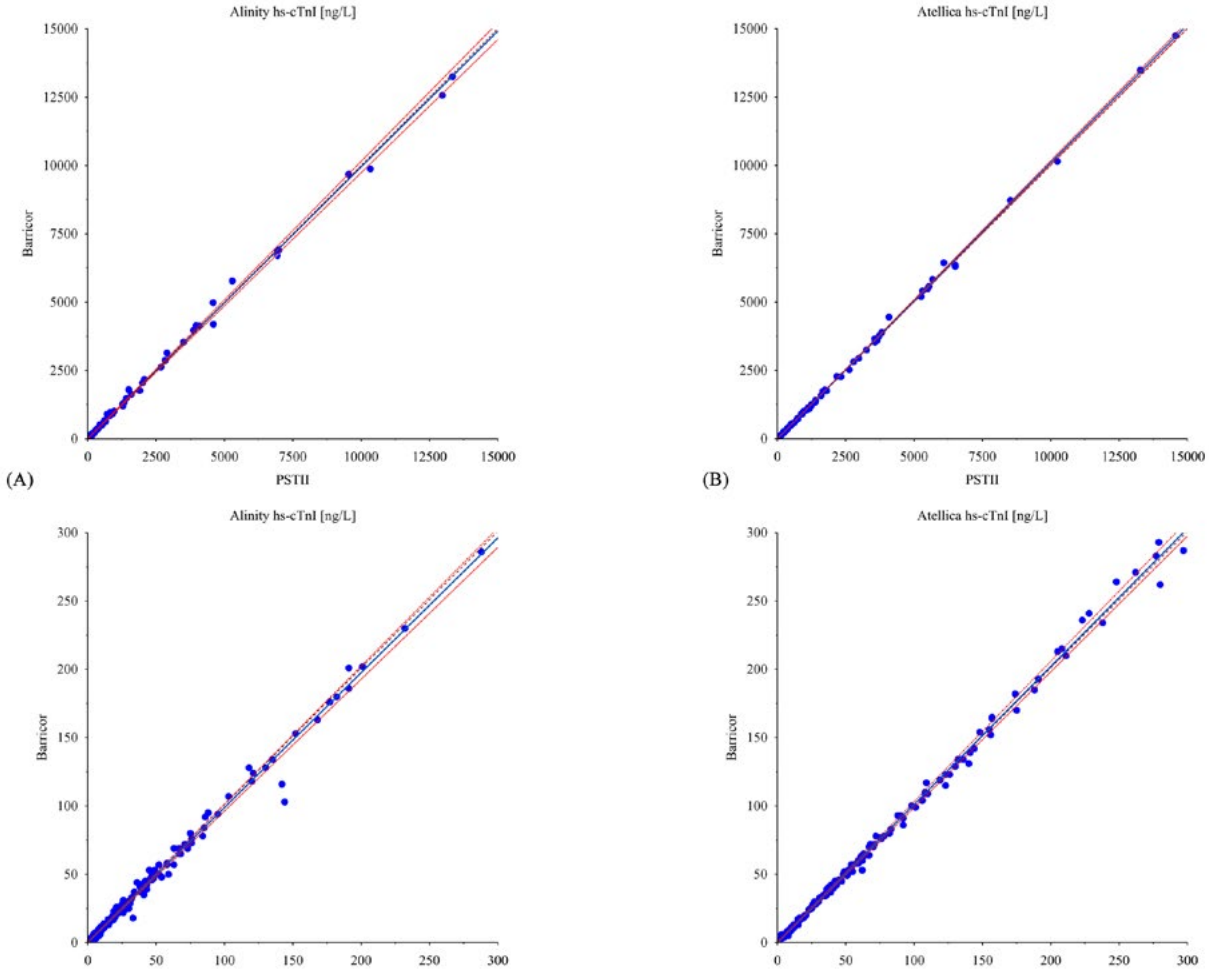
Supplemental Table 2: Paired comparisons between both measuring systems and the two tube types regarding the number and proportion of hs-cTnI results <4 ng/L for Alinity and <5 ng/L for Atellica. Chi-square test was used for comparisons.

System and tube type	**System and tube type**	**P value**
Alinity™ Barricor™ vs. Alinity™ PSTII™	29	0.301
Alinity™ Barricor™ vs. Atellica® Barricor™	37	1.000
Alinity™ Barricor™ vs. Atellica® PSTII™	185	0.765
Alinity™ PSTII™ vs. Atellica® Barricor™	179	1.000
Alinity™ PSTII™ – Atellica® PSTII™	145	0.760
Atellica™ Barricor™ vs. Atellica® PSTII™	143	1.000

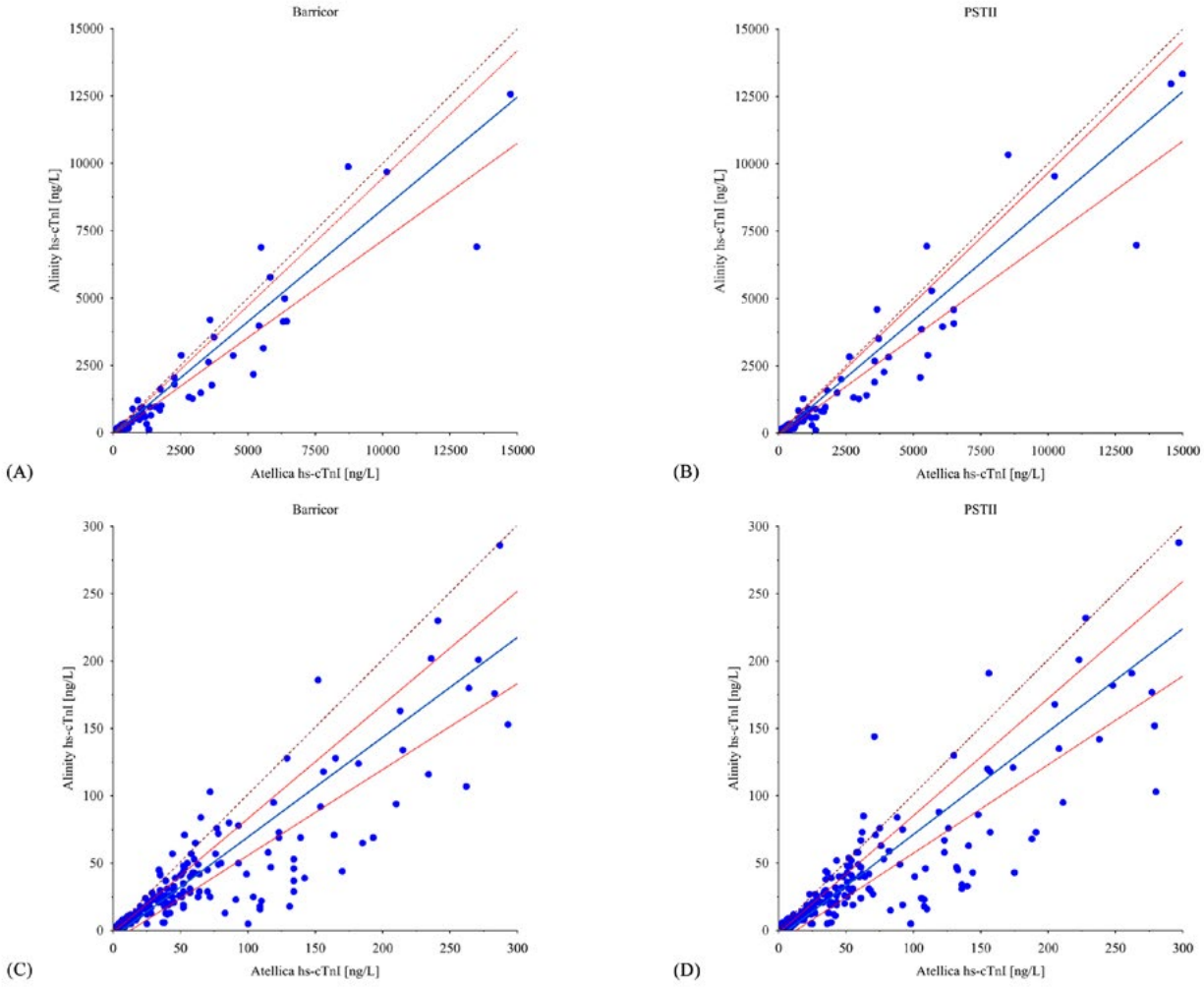
Supplemental Figure 1: Comparisons of hemolysis indices in the Barricor™ vs. PSTII™ tubes employing each of the two evaluated measuring systems:
(A) Comparison between Barricor™ tubes and PSTII™ tubes on the Alinity™; rho = -0.11; CI: -0.18, -0.01; p = 0.032
(B) Comparison between Barricor™ tubes and PSTII™ tubes on the Atellica®; rho = 0.38; CI: 0.29, 0.48; p < 0.001
(C) Comparison between AlinityTM and Atellica® systems in BarricorTM tubes; rho = 0.82; CI: 0.73, 0.87; p < 0.001
(D) Comparison between AlinityTM and Atellica® systems in PSTIITM tubes; rho = -0.15; CI: -0.22, -0.05; p = 0.004



Supplemental Figure 2: Regression analyses of comparisons of hs-cTnI measurements in Barricor™ vs. PSTII™ tubes employing each of the two evaluated measuring systems:
(A) Comparison on the Alinity™ on all 359 samples; regression equation: Barricor™ = 0.99 (CI: 0.97-1.01) PSTII™ + 6 (2-10) ng/L; r=0.99.
(B) Comparison on the Atellica® on all 359 samples; regression equation: Barricor™ = 1.01 (CI: 1.00-1.02) PSTII™ − 1 (−3 to 1) ng/L; r=0.99.
(C) Comparison on the Alinity™ on 300 samples with hs-cTnI <300 ng/L; regression equation: Barricor™ = 0.99 (CI: 0.96-1.01) PSTII™ + 0.2 (−0.04 to 0.5) ng/L; r=0.99.
(D) Comparison on the Atellica® on 300 samples with hs-cTnI <300 ng/L; regression equation: Barricor™ = 1.01 (CI: 0.99-1.03) PSTII™ + 0.1 (−0.3 to 0.4) ng/L. r=0.99.
Dashed line corresponds to the identity line. Continuous red lines correspond to the 95% confidence intervals of the regression line.



Supplemental Figure 3: Regression analyses of comparisons of hs-cTnI results obtained by the two evaluated measuring systems in the same type of tube:
(A) Comparison between Alinity™ and Atellica® systems in Barricor™ tubes on all 359 samples; regression equation: Alinity™ = 0.83 (CI: 0.72-0.95) Atellica® − 48 (−76 to −19) ng/L; r=0.96.
(B) Comparison between Alinity™ and Atellica® systems in PSTII™ tubes on all 359 samples; regression equation: Alinity™ = 0.85 (CI: 0.73-0.97) Atellica® − 56 (−87 to −24) ng/L; r=0.96.
(C) Comparison between Alinity™ and Atellica® systems in Barricor™ tubes on 300 samples with hs-cTnI <300 ng/L; regression equation: Alinity™= 0.74 (CI: 0.64-0.84) Atellica® − 5 (−8 to −1) ng/L; r=0.89.
(D) Comparison between Alinity™ and Atellica® systems in PSTII™ tubes on 300 samples with hs-cTnI <300 ng/L; regression equation: Alinity™ = 0.76 (CI: 0.66-0.87) Atellica® − 5 (−8 to −2) ng/L; r=0.88.
Dashed line corresponds to the identity line. Continuous red lines correspond to the 95% confidence intervals of the regression line.



References

1. Thygesen K, Alpert JS, Jaffe AS, Chaitman BR, Bax JJ, Morrow DA, et al. Fourth universal definition of myocardial infarction (2018). Circulation 2018;138:e618–51.
2. Collet J-P, Thiele H, Barbato E, Barthélémy O, Bauersachs J, Bhatt DL, et al. 2020 ESC Guidelines for the management of acute coronary syndromes in patients presenting without persistent ST-segment elevation. Eur Heart J 2021;42:1289–367.
3. Krintus M, Panteghini M. Laboratory-related issues in the measurement of cardiac troponins with highly sensitive assays. Clin Chem Lab Med 2020;58:1773–83.
4. Ferraro S, Dolci A, Panteghini M. Fast track protocols using highly sensitive troponin assays for ruling out and ruling in non-ST elevation acute coronary syndrome. Clin Chem Lab Med 2017;55:1683–9.
5. Backus BE, Body R, Weinstock MB. Troponin Testing in the Emergency Department-When 2 Become 1. JAMA Netw Open. 2021; 1;4:e210329.
6. Ramakers C. BD Vacutainer® Barricor tube in the emergency department: reduced hemolysis rates using partial draw tubes with reduced vacuum. Clin Chem Lab Med 2018;56:e31–2.
7. Badiou S, Vuillot O, Bargnoux AS, Kuster N, Lefebvre S, Sebbane M, et al. Improved quality of samples and laboratory turnaround time using 3.5 mL low vacuum BD Vacutainer® Barricor tubes in the emergency department. Pract Lab Med 2019;16:e00128
8. Füzéry AK, Raizman JE, Goudreau BL, Moses K, Niemann K, Park J, Cembrowski GS. The BD Barricor blood collection tube is an acceptable and robust alternative to the PST for use with the Beckman AccuTnI+3 assay. Clin Biochem 2017;50:851-7.
9. Raizman JE, Tsui AKY, Goudreau BL, Füzéry AK, Estey M, Sadrzadeh H, Higgins T. Multi-platform analytical evaluation of the Beckman Coulter Access high-sensitivity troponin I assay across different laboratory sites using Barricor plasma. Clin Biochem. 2020;78:25-31.
10. Cembrowski G, Qiu Y, Sherazi A, Shea JL. Retrospective analysis of intra-patient laborato-ry variation demonstrates that the BD Vacutainer® Barricor™ blood collection tube reduces troponin variation. Clin Biochem 2023;114:24-29.
11. Barth JH, Panteghini M, Bunk DM, Christenson RH, Katrukha A, Noble JE, et al.; IFCC Working Group on Standardization of Cardiac Troponin I. Recommendation to harmonize the units for reporting cardiac troponin results. Clin Chim Acta 2014;432:166.
12. Capoferri A, Aloisio E, Pasqualetti S, Panteghini M. More about the random uncertainty of photometric determination of hemolysis index on the Abbott AlinityTM c platform. Clin Biochem 2022;105-106:94-5.
13. Snyder JT, Garcia E, Davis J, Cheek J. Performance evaluation of the Atellica CH analyzer HIL alert feature. 72nd AACC Annual Scientific Meeting Abstracts, Abstract B-239, 2020:S144.
14. Pasqualetti S, Szőke D, Panteghini M. Heparinate but not serum tubes are susceptible to hemolysis by pneumatic tube transportation. Clin Chem Lab Med 2016;54:785–9.
15. Boeddinghaus J, Nestelberger T, Badertscher P, Twerenbold R, Fitze B, Wussler D, et al.; APACE Investigators. Predicting acute myocardial infarction with a single blood draw. Clin Chem 2019;65:437-50.
16. CLSI Measurement Procedure Comparison and Bias Estimation Using Patient Samples; Approved Guideline–Third Edition. CLSI document EP09-A3. Wayne, PA. Clinical and Laboratory Standards Institute; 2013
17. Sandoval Y, Nowak R, deFilippi CR, Christenson RH, Peacock WF, McCord J, et al. Myocardial infarction risk stratification with a single measurement of high-sensitivity troponin I. J Am Coll Cardiol 2019;74:271-82.
18. Gils C, Sandberg MB, Nybo M. Verification of the hemolysis index measurement: imprecision, accuracy, measuring range, reference interval and impact of implementing analytically and clinically derived sample rejection criteria. Scand J Clin Lab Invest 2020;80:580-9.
19. Gawria G, Tillmar L, Landberg E. A comparison of stability of chemical analytes in plasma from the BD Vacutainer® Barricor™ tube with mechanical separator versus tubes containing gel separator. J Clin Lab Anal 2020;34:e23060.
20. 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;28:010704.
21. Dupuy AM, Badiou S, Daubin D, Bargnoux AS, Magnan C, Klouche K, Cristol JP. Comparison of Barricor™ vs. lithium heparin tubes for selected routine biochemical analytes and evaluation of post centrifugation stability. Biochem Med (Zagreb) 2018;28:020902.
22. Sörensen NA, Goßling A, Neumann JT, Hartikainen TS, Haller PM, Scharlemann L, et al. Diagnostic validation of a high-sensitivity cardiac troponin I assay. Clin Chem 2021;67:1230-9.
23. Sharrod-Cole H, Ford C. Multicenter evaluation of a high-sensitivity troponin I assay and verification of an early rule-out algorithm. J Appl Lab Med 2019;4:95-100.
24. Tate JR, Bunk DM, Christenson RH, Barth JH, Katrukha A, Noble JE, et al; IFCC Working Group on Standardization of Cardiac Troponin I. Evaluation of standardization capability of current cardiac troponin I assays by a correlation study: results of an IFCC pilot project. Clin Chem Lab Med 2015;53:677-90.
25. Carlton EW, Ingram J, Taylor H, Glynn J, Kandiyali R, Campbell S, et al. Limit of detection of troponin discharge strategy versus usual care: randomised controlled trial. Heart 2020;106:1586–94.

26. Anand A, Lee KK, Chapman AR, Ferry AV, Adamson PD, Strachan FE, et al. High-sensitivity cardiac troponin on presentation to rule out myocardial infarction: A stepped-wedge cluster randomized controlled trial. Circulation 2021;143:2214–24.
27. Chiang CH, Chiang CH, Lee GH, Gi WT, Wu YK, Huang SS, et al. Safety and efficacy of the European Society of Cardiology 0/1-hour algorithm for diagnosis of myocardial infarction: systematic review and meta-analysis. Heart 2020;106:985-91.
28. National Institute for Health and Care Excellence. High-sensitivity troponin tests for the early rule out of NSTEMI. Diagnostics guidance [DG40]. Published: 26 August 2020.
29. Boeddinghaus J, Nestelberger T, Twerenbold R, Wildi K, Badertscher P, Cupa J, et al. Direct comparison of 4 very early rule-out strategies for acute myocardial infarction using high sensitivity cardiac troponin I. Circulation 2017;135:1597–611.
30. Sandoval Y, Smith SW, Love SA, Sexter A, Schulz K, Apple FS. Single high-sensitivity cardiac troponin I to rule out acute myocardial infarction. Am J Med 2017;130:1076–83. e1.
31. Shortt C, Ma J, Clayton N, Sherbino J, Whitlock R, Pare G, et al. Rule-in and rule-out of myocardial infarction using cardiac troponin and glycemic biomarkers in patients with symptoms suggestive of acute coronary syndrome. Clin Chem 2017;63:403–14.
32. Chapman AR, Fujisawa T, Lee KK, Andrews JP, Anand A, Sandeman D, et al. Novel high-sensitivity cardiac troponin I assay in patients with suspected acute coronary syndrome. Heart 2019;105:616–22.
33. Lowry MTH, Anand A, Mills NL. Implementing an early rule-out pathway for acute myocardial infarction in clinical practice. Heart 2021;107:1912-9.
34. Karády J, Mayrhofer T, Ferencik M, Nagurney JT, Udelson JE, Kammerlander AA, et al. Discordance of high-sensitivity troponin assays in patients with suspected acute coronary syndromes. J Am Coll Cardiol 2021;77:1487-99.

Original Article

Artificial Intelligence – Perception of Clinical Laboratories' Technical Staff a Nationwide Multicentre Survey in Pakistan

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Keywords

Artificial intelligence, Clinical laboratory, Pakistan, Pathology, AI tools, Survey

Abstract

Introduction

As Artificial Intelligence (AI) technology continues to assimilate into various industries, there is a huge scope in the healthcare industry specifically in clinical laboratories. The perspective of the laboratory professionals can give valuable insight on the ideal path to take for AI implementation.

Methods

The study utilized a cross-sectional survey design and was conducted at the section of Chemical Pathology, Department of Pathology and Laboratory Medicine, the Aga Khan University (AKU), Karachi, Pakistan in collaboration with Consultant Pathologists of 9 clinical laboratories associated with teaching hospitals across Pakistan from October-November 2023. The survey was for a duration of 2 weeks and was circulated to all working laboratory technical staff after informed consent.

Results

A total of 351 responses were received, of which 342 (male=146, female=196) responses were recorded after exclusion. Respondents ranged from technologists, faculty, residents, and coordinators, and were from different sections (chemical pathology, microbiology, haematology, histopathology, POCT). Out of the total 312 (91.2%) of respondents stated that they were at least somewhat familiar with AI technology. Experts in AI were only 2.0% (n=7) of all respondents, but 90% (n=6) of these were < 30 years old. 76.3% (n=261) of the respondents felt the need to implement more AI technology in the laboratories, with time saving (26.1%) and improving performances of tests (17.7%) cited to be the greatest benefits of AI. Security concerns (n=144) and a fear of decreasing personal touch (n=143) were the main concerns of the respondents while the younger employees had an increased fear of losing their jobs. 76.3% were in favour of an increase in AI usage in the laboratories.

Conclusion

This study highlights a favourable perspective among laboratory professionals, acknowledging the potential of AI to enhance both the efficiency and quality of laboratory practices. However, it underscores the importance of addressing their concerns in the thoughtful implementation of this emerging technology.

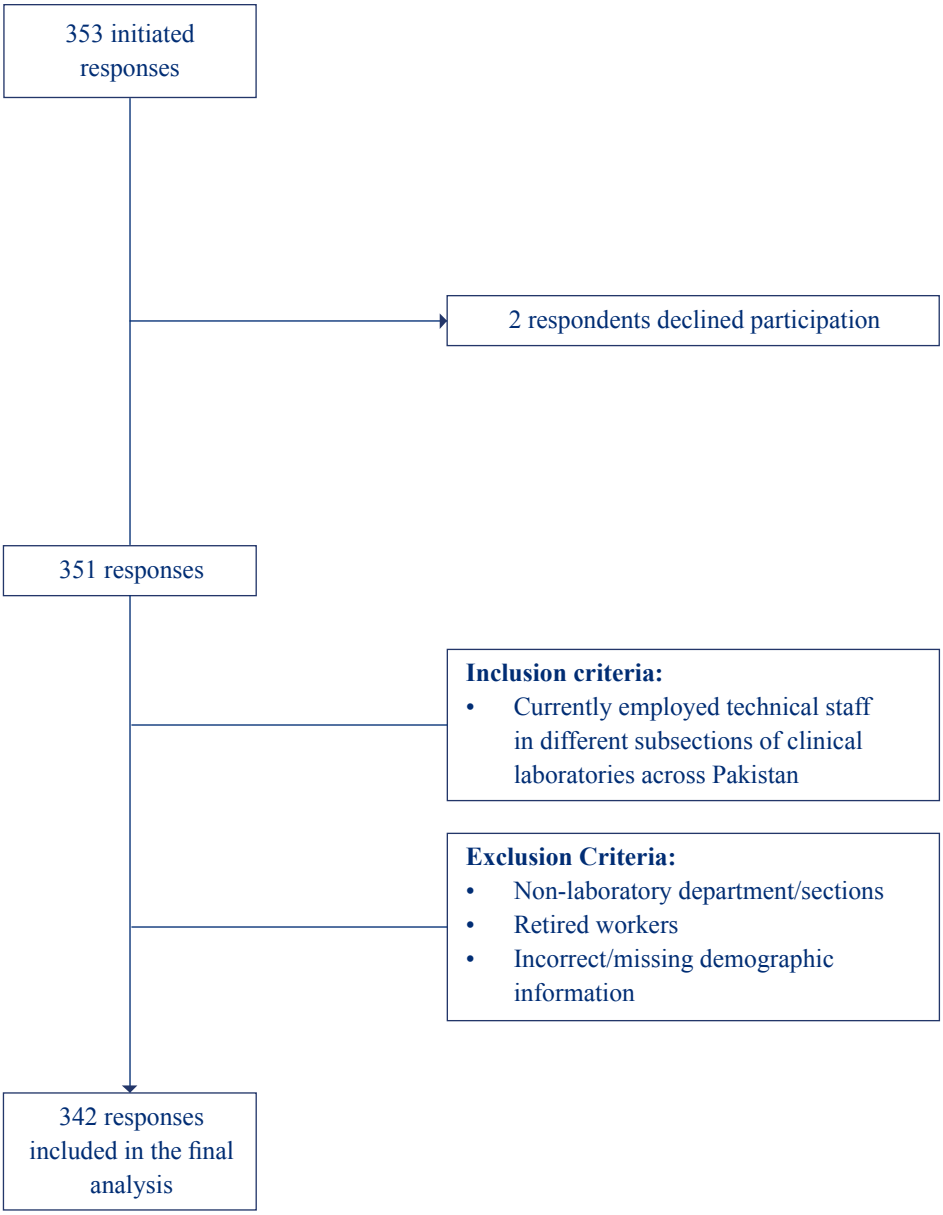
Introduction

The evolution of Artificial Intelligence (AI) has led to the improvement of various industries including the healthcare industry. The integration of AI technology in healthcare is at the forefront of the current era due to the potential benefits that it can provide [1]. Within the complex ecosystem of healthcare, laboratory medicine remains the domain with the most to gain from AI integration [2, 3]. The potential to streamline laboratory processes, enhance diagnostic accuracy, and improve decision-making is huge and this possibility granted by AI can make a compelling case to invest in AI implementation [4]. AI is gaining popularity amongst clinical laboratories in Pakistan, in line with the global landscape. Recent attempts at automation have increased efficiency and accuracy of lab processes and AI is expected to usher in a new wave of improvements [5, 6]. Few studies have been conducted in other countries to include employee attitudes on AI yet the knowledge, attitudes, and experiences of the workers in Pakistan regarding AI remain an underexplored area [7, 8]. While the utility of AI is undeniable, and its benefits are evidently seen in present implementations in other industries, there are many differing perspectives to it. While some people accept this technology as a powerful tool that can do wonders in the laboratory environment, others show concerns for the technology. Besides advancing at a frighteningly fast pace without proper regulations, the big talking point is the fear of job displacement as employees feel threatened with being made redundant as AI technology advances. Other issues also arise from not being familiar with the technology, hence accuracy and safety might not be trusted. Addressing these concerns would be vital in resolving the most effective integration strategy for Pakistan's clinical laboratories. Before AI gets through implementation in clinical labs, there is a dire need to provide a comprehensive baseline exploration of the perceptions and insights of professionals in the clinical laboratory sector across Pakistan, examining their familiarity with AI, opinions on its impact, fears surrounding the increased use of AI, and recommendations for effective integration [9]. The objective of this research was to identify the knowledge gaps and establish a baseline on the attitudes and expertise of these professionals, by surveying the laboratory professionals in institutions across the country. By gathering a diverse range of opinions from different specialties and positions we can uncover the AI knowledge landscape in the clinical laboratories and utilize this to provide valuable insights for leaders of healthcare institutions and policymakers [10]. Action plans to facilitate deeper understanding of the role and proper integration of AI technology can take into account the current state of AI awareness and utilization.

Materials and Methods

A cross-sectional survey was conducted by the section of Chemical Pathology, Department of Pathology and Laboratory Medicine, the Aga Khan University, Karachi after approval from the ethical review committee (AKU- 2023-9228-26528). The study was undertaken in compliance with the ethical principles for medical research involving human subjects, in accordance with the Declaration of Helsinki. A previously validated and published tool by Ardon O et al was used with some modifications according to local context [7]. The survey was filled in by two Consultant Pathologists and a senior technologist as a pilot to locally validate the questionnaire for understanding of language and content. The survey was designed and circulated via Google Forms link to the lead Pathologists of ten major clinical laboratories across Pakistan who in turn dispersed the survey amongst the employees of the labs and to other labs outside the initial ten using WhatsApp and Email. The participation was entirely voluntary and anonymized, and respondents were asked to give consent before attempting the questions. Moreover, for further convenience QR code of the survey link was also generated and hardcopy was used to ensure that people with limited access to WhatsApp or Email can utilized the web version via direct link. The survey consisted of three sections, first was the general information and consent; then the demographic section with eight questions, and a section with seven questions related to AI. The sample size was calculated prior to the dissemination of the survey. An open EPI calculator at 90% confidence interval was used which yielded a sample of 174. This sample size was calculated on the assumption that 20% of participants possess some knowledge and awareness of AI. However, we targeted maximum responses achieved during the defined timeframe. The survey accepted responses from October-November 2023. 353 people attempted the survey; after the inclusion/exclusion criteria was met, 342 responses from current laboratory professionals were included in the final analysis (Figure 1). The data was analysed to reveal the differences in the demographic groups, and associations between the groups and their AI opinions using the chi-square test of independence. The Excel (Microsoft Corporation, 2018) and Stata (Stata Corp, College Station, Tx) software were used for data collection and analysis.

Figure 1: Flowchart of data collection showing the final number of responses included in the study.



Results

Out of the 10 clinical laboratories contacted, 9 laboratories collaborated in the project. The 342 responses came from a wide range of demographics, including those from different ages, genders, and sections. Most respondents were from the age group of 30-49 years old (n=194) while the split between male and female was 42.7% and 57.3%, respectively. A map of Pakistan showing the distribution of responses are shown in Figure 1. Participants were from seven sections of laboratory medicine services with the highest number from Chemical Pathology (52.3%), followed by Microbiology (18.4%), Haematology (17.3%), Histopathology (6.1%), Molecular Pathology (4.1%), Point of Care Test (POCT) (0.9%) and Immunology (0.6%). The survey received responses from a multitude of positions with the greatest number coming from the technologists (31.9%), and faculty members (23.7%). Thirty eight percent of respondents had achieved FCPS, MPhil, or PHD level education, 30.4% had studied until Bachelor's, 19.9% Master's and 11.1% MBBS. Two (0.6%) of the respondents reported having completed the Diploma of Medical Laboratory Services (DMLS) degree. 46.8% of the respondents had less than 5 years of experience, and the number of respondents lowered as experience level increased.

Figure 2: Map of Pakistan showing the cities from which the responses came, along with the frequencies of responses.

Out of the respondents, 91.2% reported being at least somewhat familiar with AI technology (Figure 3). While experts at AI, most of these were from the less than 30 years old, indicating correlation between different age groups and the level of familiarity ($p=0.016$). Among various positions, lab coordinators had the highest familiarity levels. 85.7% of coordinators were experts or very familiar with AI technology. There was a significant difference between the specialities regarding their familiarity to AI ($p=0.001$) with Chemical Pathology and Haematology superseding other sections. There was also significant difference between genders ($p<0.001$). All 7 of the experts identified as male, while more females were unfamiliar or somewhat familiar with AI (80.8% females to 63.6% of males).

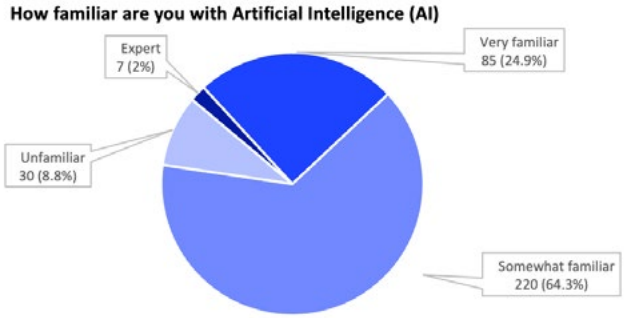


Figure 3: Repsonses to the question, "How familiar are you with AI?"

When asked if they have encountered any AI applications (Figure 4), the participants responded with a majority (58.2%) yes, while 34.8% had no exposure, and 7.0% were unsure if they had. The more experienced the respondents, the more likely they were to have encounter AI technology ($p=0.042$).

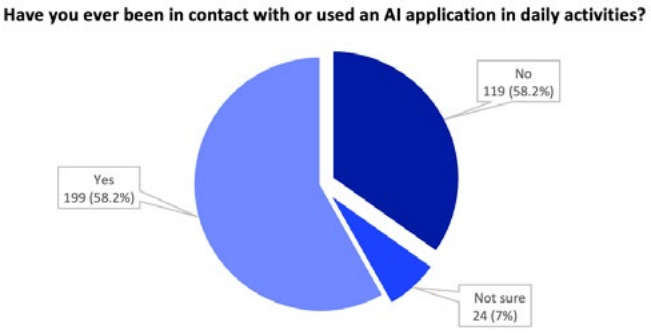


Figure 4: Repsonses to the question, "Have you ever been in contact with, or used an AI application in daily activities?"

Participants were asked for examples of AI tools that they had used (Table 1), the two main tools were ChatGPT (58.7%) and Google Bard (13.0%). Other tools reported were Quillbot, Grammarly, Scite, Perplexity, etc. Younger people were observed to utilise more AI tools (9 for <30 compared to 4 for >=50 from the list of tools acquired) and use them at a higher frequency. The positions with the most varied AI use were the faculty (8 tools) and technologists (7 tools). Similarly, respondents having completed their FCPS/PhD/MPhil level education reported the greatest AI use at 9 tools. There was no significant observation difference between the genders or specialties. The respondents were questioned about the proposed uses of AI (Table 2). The responses were fairly evenly distributed with time saving (26.1%) being the most useful benefit of AI, followed by increased performance of tests (17.7%) and prevention of workplace errors (16.4%). There were no significant associations between their responses and the demographics of the respondents.

Table 1: AI Tools Used

Frequencies of AI tools used	Frequency (n)	Percentage (%)
ChatGPT	172	58.7
Google Bard	38	13.0
Quillbot	4	1.4
Grammarly	4	1.4
Bing	3	1.0
Copy.ai	3	1.0
Perplexity	2	0.7
Google Lens	2	0.7
SnapChat	2	0.7
Tome	2	0.7
Scite	1	0.3
Others	8	2.7
Not Reported	52	17.7
Total	**293**	**100**

The respondents were questioned about the proposed uses of AI (Table 2). The responses were fairly evenly distributed with time saving (26.1%) being the most useful benefit of AI, followed by increased performance of tests (17.7%) and prevention of workplace errors (16.4%). There were no significant associations between their responses and the demographics of the respondents.

Table 2: Responses to the question "If you could use AI to help you perform your job, what would you like it to accomplish?".

If you could use AI to help you perform your job, what would you like to accomplish?	Frequency (n)	Percentage (%)
Time Saving	265	26.1
Test Performance	179	17.7
Reduce Errors	166	16.4
Drafting letters	147	14.5
Increase Objectivity	117	11.5
Reduce Repetition	117	11.5
Unfamiliar with AI	18	1.8
Others	5	0.5
Total	**1014**	**100**

The participants' main concerns about AI (Table 3) were of the security especially with regards to patient information (23.4%), and a decrease in hands-on work (23.2%). The fear of losing jobs was higher in younger professionals (20.5% for <30 vs 6.3% for >50). Similarly, job security was not as much of a concern for those with over 20 years of experience. However, more (12.5%) respondents with 20+ years' experience stated that they were unfamiliar with AI than the other respondents. Male respondents (26.4%) were more concerned about the learning curve than females (14.3%). There were no significant differences between ages, positions, or specialities.

Table 3: Responses to the question "What concerns do you have about AI?".

What concerns do you have about AI?	Frequency (n)	Percentage (%)
Security concern	144	23.4
Decreased personal element	143	23.2
Big learning curve	104	16.9
Fear of losing jobs	96	15.6
Unsure of new technology capabilities	93	15.1
Unfamiliar with AI (Unable to answer)	32	5.2
Others	4	0.6
Total	**616**	**100**

Respondents were asked about their opinion on using AI technology in the laboratory setting (Figure 5). 76.3% of the respondents agreed with the use of more AI tools in the laboratory, while 12.2% disagreed. Less experienced respondents were slightly more supportive of AI technology use, while men (10.1% strongly disagree) were more hesitant to accept AI than women (2.9% strongly disagree). When asked about the areas in which AI would be most beneficial (Table 4), the responses were distributed fairly evenly. Data analysis (20.5%) and scientific research (19.7%) were the sectors most often chosen. 17.3% respondents believed AI would benefit in education. Error detection (16.8%), results verification (14.0%), and customer care (10.9%) followed. There was no difference in the distribution of responses by gender, age, education, specialty, or position.

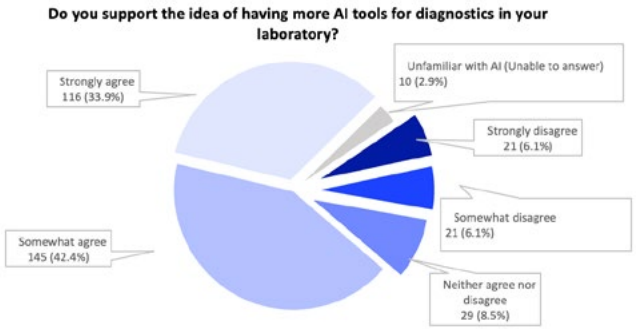


Figure 5: Repsonses to the question, "Do you support the idea of having more AI tools for diagnostics in your laboratory?"

Table 4: Responses to the question "What areas could most benefit from AI implementation?".

What areas could most benefit from AI implementation??	Frequency (n)	Percentage (%)
Data analysis	231	20.5
Scientific research	222	19.7
Education	195	17.3
Error Detection	190	16.8
Results verification and reporting	158	14.0
Customer care	123	10.9
Unfamiliar with AI (Unable to answer)	10	0.9
Total	**1129**	**100**

Discussion

A significant amount of feedback from participants (342 responses) was received for this survey, which aimed to highlight the level of expertise, knowledge, concerns, and interest among Pakistani clinical laboratory professionals in the field of AI and its applications in laboratory medicine. Web-based surveys have previously demonstrated advantages over traditional approaches, particularly for health social science researchers [11, 12]. Furthermore, since every person surveyed used WhatsApp or email for work-related purposes regularly, secondly the QR code availability ensured that the representativeness bias which could exist in a web-based survey could be ruled out. Most respondents supported the utility of AI-augmented diagnostic tools despite worries about job loss, and about 91.2% responded that they were somewhat familiar with AI. Respondents acknowledged that AI could boost productivity and decrease errors. Individuals with advanced degrees shown higher levels of knowledge and interaction with AI, whereas younger persons showed higher levels of familiarity with the technology. The AI tools are starting to be used in diagnostic labs [13, 14]. The usage of AI tools reported fell under two categories, laboratory use and professional use. AI has been applied to the prediction of errors in genetic variants and phenotypes, infectious diseases, cervical cancer categorization in cytology specimens, histology, and so on [15, 16]. AI also has the potential to develop algorithms to use diagnostic tests more judiciously thus conserving the resources and time [17]. From the results, a conclusion was formed that the participants are not fully aware of more advanced AI tools that could be beneficial in research. Increase in familiarity with tools such as Trinka and Consensus, among others, could boost the efficiency and level of research being conducted inside the clinical laboratories. Literature review revealed few surveys evaluating knowledge, attitude, and practice of AI amongst medical professionals in Pakistan, but they were targeted towards physicians and students [18, 19]. However, from clinical laboratories perspective, where AI is booming globally, there was no baseline data available from the region. Our study was different from other surveys in that we polled a large sample of laboratory workers, whereas earlier surveys were restricted to medical professionals. AI is likely to have wide-ranging implications on all members of the workforce, both technical staff and Consultant Pathologists. Therefore, it is critical to comprehend the beliefs and attitudes of non-physicians. Our results reveal that while general laboratory workers are enthusiastic about AI, they nevertheless have some of the same concerns as physicians. Data security concerns, lack of personal element and fear of losing job were the major concerns recorded. Male respondents seemed to be more wary of AI technology than females. The fear of losing job was more in the younger group i.e., less than 30 years. Moreover, it was the more experienced age group i.e., greater than 30 years that were less supportive of having more AI technology in the laboratory setting. Comparing Ardon O et al to these results, there was an overall support of AI from both studies' participants although there was a greater number of neutral laboratory professionals in the US study (30% neither agreed or disagreed). They had similar responses for the uses of AI, with time saving and reduction in errors the top 2 options in both studies. Moreover, the areas of AI implementation were also consistent with this result's findings. The main concern from the US based study was the fear of losing jobs, while the findings of this study show that it is a concern, it is less than security and human personalization. Another study conducted of labs across Italy (n=227) showed a much higher rate of AI support (95% expressed interest in learning about AI technology) although current AI knowledge was still low (15% very familiar, 5% expert). These comparisons reveal that the general situation, whether in the US, Pakistan, or a European country, is still the same with much improvement to be made in terms of AI implementation in the laboratory [20]. From future laboratory management perspective, to decrease resistance towards adoption of AI, there is a dire need to propagate that AI does not necessarily cause employment losses, much like other disruptive technologies. Instead, AI eliminates the laborious parts of work and increases efficiency in the laboratory environment [21]. The strengths of this study were a sizable sample size that included a range of job responsibilities, work settings, and educational backgrounds. Prior research has concentrated on specific tasks, like image processing, or on limited populations, such only physicians. Given the wide-ranging consequences of AI, it is critical to comprehend the opinions and attitudes of everyone who could be impacted. The successful creation and application of AI tools can be aided by this understanding. However, from limitation perspective, the results only reflect the Pakistani large clinical laboratories affiliated with teaching institutes and housing all sections of Pathology, despite the high number of responses indicating a strong adherence to the questionnaire, this still represents a very small portion of the estimated more than 500 smaller clinical laboratories in Pakistan. Secondly, rather than being measured, the results were self-reported. For example, rather than performing a formal examination, we asked respondents about their judgement regarding their degree of knowledge. Finally, the survey was quite short. Because of the workforce's time demands, we were worried that a lengthy survey might result in a poor response rate. In conclusion, a positive trend towards increased familiarity with AI in this low-resource context is revealed by the survey on the attitudes, knowledge, and practises of AI among laboratory professionals throughout Pakistan. The highly engaged online poll demonstrated the extensive usage of different AI tools like ChatGPT, demonstrating an increasing adoption of AI technologies. But the report also points out significant gaps, especially in the area of digital pathology, where further AI integration is desperately needed. Though it was well received, others expressed worries about possible data security risks, a perceived lack of personal touch, and the possibility of losing one's job. The results can aid proper management and strategic planning in clinical laboratories for near future in the country, the challenges can be mitigated, paving the way for increased efficiency and advancements in clinical laboratory practices

through AI integration. However, thorough validations are necessary before practical adoption of AI tools in clinical laboratory practices.

Disclosure Statement

The authors declare that (s)he has no relevant or material financial interests that relate to the research described in this paper.

References

1. Davenport T, Kalakota R. The potential for artificial intelligence in healthcare. Future Healthc J. 2019;6(2):94-8.
2. Wilson S, Steele S, Adeli K. Innovative technological advancements in laboratory medicine: Predicting the lab of the future. Biotechnol Biotechnol. Equip. 2022;36(S1): S5-S17.
3. Tran NK, Albahra S, May L, Waldman S, Crabtree S, Bainbridge S, et al. Evolving applications of Artificial Intelligence and Machine Learning in infectious diseases testing. Clin Chem. 2021;68(1):125-33.
4. Rifai N, Topol E, Chan E, Lo YM, Wittwer CT. Disruptive innovation in laboratory medicine. Clin Chem. 2015;61(9):1129-32.
5. Lippi G, Da Rin G. Advantages and limitations of total laboratory automation: A personal overview. Clin Chem Lab Med. 2019;57(6):802-11.
6. Brown AS, Badrick T. The next wave of innovation in laboratory automation: Systems for auto-verification, quality control and specimen quality assurance. Clin Chem Lab Med. 2023;61(1):37-43.
7. Ardon O, Schmidt RL. Clinical laboratory employees' attitudes toward Artificial Intelligence. Lab Med. 2020;51(6):649-54.
8. Al Hadithy ZA, Al Lawati A, Al-Zadjali R, Al Sinawi H. Knowledge, attitudes, and perceptions of Artificial Intelligence in healthcare among medical students at Sultan Qaboos University. Cureus. 2023;15(9):e44887.
9. Han C, Rundo L, Murao K, Nemoto T, Nakayama H. Bridging the gap between AI and healthcare sides: Towards developing clinically relevant AI-powered diagnosis systems. Artif Intell Appl Innov. 2020;584:320-33.
10. Paranjape K, Schinkel M, Hammer RD, Schouten B, Nannan Panday RS, Elbers PWG, et al. The value of Artificial Intelligence in laboratory medicine: Current opinions and barriers to implementation. Am J Clin Pathol. 2020;155(6):823-31.
11. Fleming, CM, Bowden, M. Web-based surveys as an alternative to traditional mail methods. J Environ Manage. 2009;90:284–92.
12. Wright, KB. Web-based survey methodology. In: Liamputtong, P, editor. Handbook of research methods in health social sciences. Singapore: Springer; 2019:1339–52.
13. Haymond S, McCudden C. Rise of the machines: Artificial Intelligence and the clinical laboratory. J Appl Lab Med. 2021;6(6):1640-54.
14. Naugler C, Church DL. Automation and artificial intelligence in the clinical laboratory. Crit Rev Clin Lab Sci. 2019;56(2):98-110.
15. Alyafeai Z, Ghouti L. A fully automated deep learning pipeline for cervical cancer classification. Expert Syst Appl 2020;141:112951.
16. Landau MS, Pantanowitz L. Artificial Intelligence in cytopathology: A review of the literature and overview of commercial landscape. J Am Soc Cytopathol. 2019;8(4):230-241.
17. Wen X, Leng P, Wang J, Yang G, Zu R, Jia X et al. Clinlabomics: Leveraging clinical laboratory data by data mining strategies. BMC Bioinform. 2022;23(1):1-20.
18. Ahmed Z, Bhinder KK, Tariq A, Tahir MJ, Mehmood Q, Tabassum MS et al. Knowledge, attitude, and practice of artificial intelligence among doctors and medical students in Pakistan: A cross-sectional online survey. Ann Med Surg. 2022;76:103493.
19. Akhtar H, Faheem S, Qasim R, Irfan F, Tanwir A, Hammad H, Hirani S. Assessment of knowledge and perceptions of Artificial Intelligence among dental students of Karachi, Pakistan. A multi-center survey. Pak Oral Dent J. 2022;42(4):193-9.
20. Bellini C, Padoan A, Carobene A, Guerranti R. A survey on Artificial Intelligence and Big Data utilisation in Italian clinical laboratories. Clin Chem Lab Med. 2022;60(12):2017-2026.
21. Cadamuro J. Disruption vs. evolution in laboratory medicine. Current challenges and possible strategies, making laboratories and the laboratory specialist profession fit for the future. Clin Chem Lab Med. 2023;61(4):558-66.

Rapid Communication

Effectiveness of cord blood as a strategy to rule out conjugated hyperbilirubinemia

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Keywords
biliary atresia, conjugated bilirubin, cord blood

Abstract

Early detection of biliary atresia is crucial for timely intervention and improved outcomes in infants with elevated conjugated bilirubin levels. This study aims to investigate the viability of cord blood gas analysis as a novel method for assessing conjugated bilirubin levels. Infants with high heel stick levels also showed elevated cord blood bilirubin levels, indicating that cord blood testing could replace the need for repeat heel stick tests, especially benefiting low birth weight infants. Ongoing research, including larger cohorts and alternative bilirubin measurement methods, will further validate this innovative screening approach. Infants with biliary atresia have high conjugated bilirubin levels at birth. As a result, infants can be screened with newborn conjugated bilirubin measurements, to allow for early detection, timely treatment, and the best chances of delaying or even avoiding the need for a liver transplant [1]. An important limitation of screening, however, is that infants must undergo a separate blood test. To overcome this limitation, we investigated whether conjugated bilirubin measurements from cord blood could be useful.

We compared conjugated bilirubin from 2 sample types, from infants born at Texas Children's Hospital between October 2022-March 2023. The first sample type was from umbilical venous cord blood, collected in a heparin syringe for cord gas measurements. Leftover samples were stored in the dark at 4°C until time of analysis, ranging from 4-48 hours after collection. The second sample type was heel-stick blood, obtained in the first 60 hours of life as part of routine newborn clinical care. Conjugated bilirubin in cord blood (plasma) and heel stick (serum) was measured with the neonatal bilirubin spectrophotometric method (BuBC) using the Vitros 7600 system, and levels >0.2 mg/dL were considered high. There were 3361 births during the study period, including 3295 infants (98.0%) who had conjugated bilirubin measured by heel stick. These included 3278 infants (99.5%) with normal heel stick levels of <0.1 mg/dL, 12 infants (0.4%) with normal heel stick levels of 0.1-0.2 mg/dL, and 5 infants (0.2%) with high heel stick levels of >0.2 mg/dL (Figure1). Of the infants with heel stick levels of <0.1 mg/dL, 184 infants had cord blood available for measurement; all these infants had cord blood levels of 0.0 mg/dL. Of the infants with heel stick

levels of 0.1-0.2 mg/dL, 9 infants had cord blood available for measurement including one with transient alterations in liver function and a high cord blood level of 0.3 mg/dL (Table 1). Finally, of the infants with heel stick levels of >0.2 mg/dL, 3 infants had cord blood available for measurement. All had high conjugated bilirubin levels in cord blood, including one premature infant with an omphalocele and 2 infants with maternal antibodies directed against red blood cells. These preliminary results show that in most cases conjugated bilirubin levels of cord blood (collected at birth) and heel stick (collected in the first 60 hours of life) samples correlate perfectly and are 0.0 mg/dL. Thus, the negative predictive value of a cord blood bilirubin of 0.0 mg/dL in our study was 99.8%. Only 7 out of the high heel stick Bc level infants would need to be re-screened using this strategy. In addition, cord blood conjugated bilirubin levels that are elevated in infants with high heel stick levels perhaps reflect disease processes that have already started before birth. Future larger studies are needed to include patients with biliary atresia, compare levels from umbilical veins and arteries, and in addition, test the commonly-used diazo method which measures "direct bilirubin." Such studies will indicate that another blood draw or heel stick is unnecessary for most infants with a cord blood Bc of 0.0 mg/dL and will be important, especially in low birth weight infants.

AUTHOR'S DISCLOSURE

SH is a member of a Data Safety Monitoring Board coordinated by Syneos Health..

ETHICAL APPROVAL AND COMPLIANCE

This single-center study was conducted in strict adherence to the ethical guidelines for medical research involving human subjects. The study protocol received approval from the Institutional Review Board (IRB) at Baylor College of Medicine.

Figure 1: Patient flow

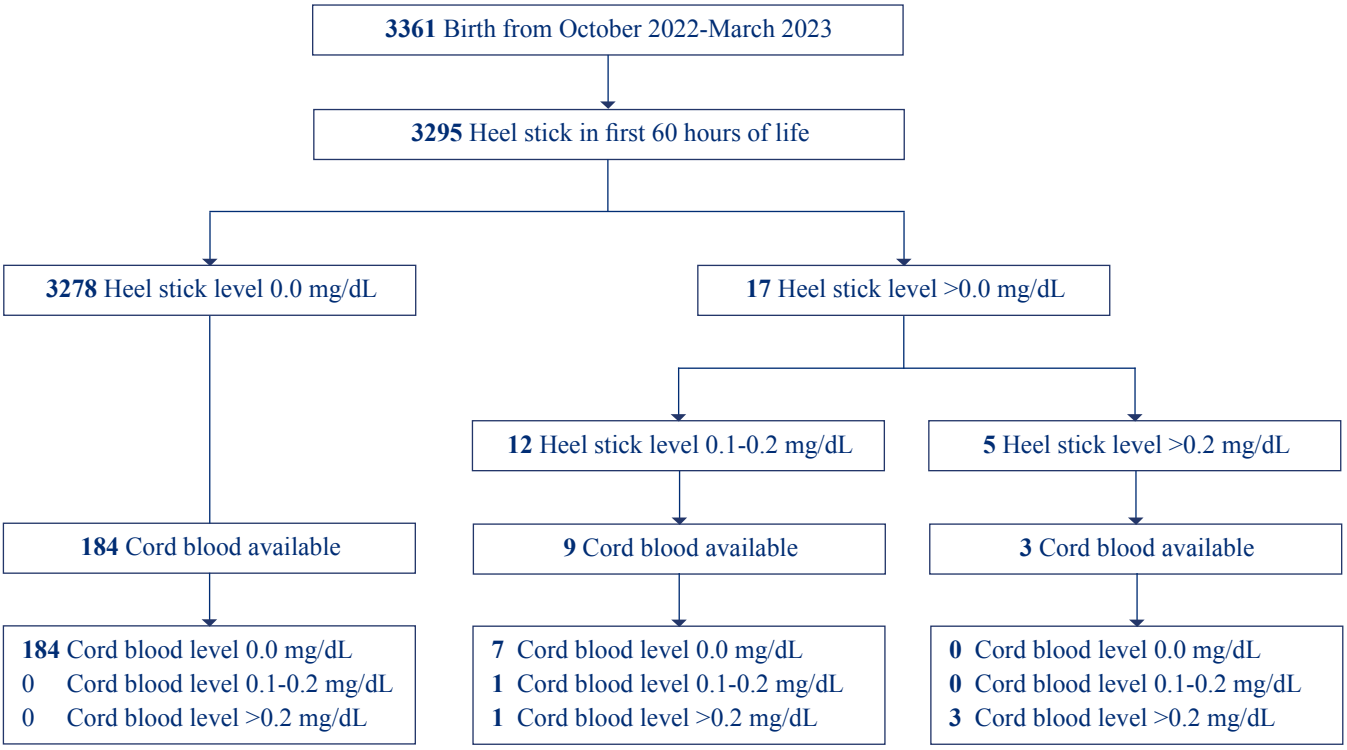

Table 1: Conjugated bilirubin levels in cord blood from infants with heel stick levels of >0.0 mg/dL

Gestational age	Additional birth factors	Cord blood level (mg/dL)	Heel stick level (mg/dL)†	Hospital length of stay
Patient heel stick Bc 0.1-0.2 mg/dL (n=12)				
33 weeks	None	*	0.1	>3 months
Term	None	*	0.1	<2 days
30 weeks	Anti-A isoimmunization	0.0	0.1	>1 month
30 weeks	Congenital anomalies	0.0	0.1	Deceased <14 days
31 weeks	Twin B	0.0	0.1	>2 months
34 weeks	Congenital anomalies	0.0	0.1	Deceased <14 days
Term	Anti-A isoimmunization	0.0	0.1	<2 days
30 weeks	Alteration in liver function	0.3	0.1	>3 months
Term	None	*	0.2	<2 days
31 weeks	Twin A	0.0	0.2	>2 months
Term	Concern for sepsis	0.0	0.2	Deceased <14 days
Term	None	0.1	0.2	<2 days
Patient heel stick Bc ≥0.2 mg/dL (n=5)				
29 weeks	Twin to twin transfusion syndrome	*	0.3	Deceased <14 days
Term	Anti-A isoimmunization	0.4	0.3	<2 days
26 weeks	Myelomeningocele	*	0.4	>4 months
35 weeks	Prematurity, omphalocele	0.3	1.3	>7 months
34 weeks	Anti-Rh isoimmunization	0.9	9.0	<1 month

* Cord blood not available

† Drawn in first 60 hours of life

REFERENCE

1. Harpavat S. et al. Diagnostic Yield of Newborn Screening for Biliary Atresia Using Direct or Conjugated Bilirubin Measurements. JAMA 2020;323:1141-1150

Review Article

Utilizing Data Analytics And Business Intelligence Tools In Laboratory Workflow

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Keywords

Benchmarking, Big Data, Business, Data Analytic, Quality Indicators,

Abstract

A business intelligence (BI) tool in a laboratory workflow offers various benefits, including data consolidation, real-time monitoring, process optimization, cost analysis, performance benchmarking (quality indicators), predictive analytics, compliance reporting, and decision support. These tools improve operational efficiency, quality control, inventory management, cost analysis, and clinical decision-making. This write up reveals the workflow process and implementation of BI in a private hospital laboratory. By identifying challenges and overcoming them, laboratories can utilize the power of BI and analytics solutions to accelerate healthcare performance, lower costs, and improve care quality. We used navify (Viewics) as a BI platform which relies on an infinity data warehouse for analytics and dashboards. We applied it for pre-analytic, analytic and post-analytic phases in laboratory. We conclude, digitalization is crucial for innovation and competitiveness, enhancing productivity, efficiency, and flexibility in future laboratories.

Introduction

It has been accepted that digitalization is a powerful engine for economic growth in today world [1]. Data analytics are becoming an increasingly important part of daily operations and quality improvement efforts within clinical laboratories. Business intelligence (BI) dashboards are one of the key components of data analytics since they provide decision makers timely access to summarized analyses and visualizations [2]. Dashboards typically display the status of key performance indicators (KPIs) and other metrics or summary statistics on a single screen, providing information for specific objectives at a glance. These BI dashboards can enhance healthcare organizations' financial and operational performance and quality of patient care. These BI tools allow us to remove manual steps done using Excel worksheets, for example data handling, using excel formulas and generating visual graphical works. Today what leadership roles needs are to: (a) determine clinical need and strategic direction for local environments, (b) ensure technology solutions are cost-effective, safe and reliable, (c) assume the business acumen to market, negotiate and manage change in services, (d) expect understanding of the clinical bioinformatics that underpin genomics, health information science (data mining and health economics) and physical sciences (e) expect

knowledge and skills in the provision of direct clinical care in the face of staffing shortfalls experienced by many healthcare systems and (f) enhance their communication and interactive skills. In growing their leadership contribution, a partnership approach in education and training across healthcare divides, in conjunction with the diagnostics and/or information technology industries, through integrated professional organization approaches, joint approaches with academia and policy related healthcare organizations are recommended [3,4]. Either we can built our own dashboard using open-source programming languages like R or Python programming language or can use available laboratory tailored BI tools from vendors [5,6]. There are multiple options available in market to use variety of user-friendly BI tools such as Microsoft Excel and Power BI, GraphPad, Tableau, and tools specific to one's EHR such as SlicerDicer that could be utilized for a variety of data analyses, reporting, and visualization purposes. But to have specific lab related BI tools which increases the collaboration with information technology, applications development, and business intelligence teams would be a valuable resource to help meet the data analytics needs of one's clinical laboratory. We have introduced the modern-day digitalization tool in our work area to monitor laboratory effectiveness and efficiency. This digital tool named navify (Viewics) works on Roche Platform covering the major bulk test menu in the laboratory, is a data analytics platform that offers dashboards and insights using Tableau. This is a web-based tool connected with hospital laboratory information system (LIS) via middleware Infinity provided by Roche, which is a lab IT solution that aids in managing workflow and data [7]. This paper delves into the multifaceted challenges faced by private laboratories in the Middle East when implementing BI and analytics solutions within their workflows. Through an in-depth literature review along with our personal experience, this study aims to shed light on the intricate interplay between the unique characteristics of laboratory operations and the complexities of implementing data-driven technologies. By identifying these challenges, laboratory professionals, IT stakeholders, and decision-makers can gain a comprehensive understanding of the potential bottlenecks and obstacles that might impede successful adoption. The subsequent sections of this paper will scrutinize key steps, encompassing issues related to data quality, integration with existing systems, privacy and security concerns, organizational change management, and the scarcity of domain-specific expertise [3,8]. By dissecting these challenges, this study seeks to provide insights that can guide laboratories in the Middle East toward informed decision-making and effective implementation of BI and analytics solutions.

How we started our journey

We, The International Medical Center (IMC) is a 300-bed tertiary care hospital located in Jeddah, Kingdom of Saudi Arabia. Built in 2005, IMC covers more than 30 specialties. Highly qualified staff, paired with the state-of-the-art facilities, support IMC's strategic pillars-wellness, quality, patient experience, digitization, people, medical and finance. To maintain its ranking among the region's best healthcare providers and to align with global healthcare practices, IMC has focused on creating a digitalization purpose-driven culture. This digitization empowers IMC to provide excellent care, improve patient experience, and optimize operations. Together, these factors provide a holistic approach to healing patients. Digitization requires adopting a digital mindset that recognizes the ongoing and fluctuating nature of the journey. IMC first created a 3-year digital transformation roadmap in 2019 that included 55 initiatives. The roadmap allows for additional initiatives to meet market trends, business requirements and ongoing institutional needs. In November 2020, we have implemented the use of navify (Viewics) to monitor our laboratory KPIs. Specific utilities of a BI tool in our laboratory workflow includes data consolidation using Infinity (Roche middleware). This middleware solution describe a software that functioned as a mediator between laboratory analyzers and the laboratory information system (LIS) [9]. This allows for a comprehensive view of laboratory operations and facilitates data-driven decision-making. By analyzing historical data and trends, BI tools can identify inefficiencies and difficulties in laboratory workflows. This insight enables process optimization, resulting in reduced turnaround times, improved productivity, and better resource allocation [10]. Here we choose data intensive dashboard (navify analytics for core lab) for monitoring Pre-analytic Volume, Analytic Turnaround Time, Post-Analytic Workload Analysis and Instrument Utilization. A dashboard represents a customized graphical view of some subset of the underlying preferred dataset(s). Vendor-specific middleware and commercial analytics platforms build dashboards and share them with business users; either individually, website or as part of an application. If a business user is given permissions to the report, they can build their own dashboards too. Laboratory dashboard (Figure 1) provides you with the overview needed as a laboratory director or as a facility manager. Gathering in one central point all the data on every division of the laboratory phases will be of great help to run it smoothly, giving you big picture of the facility. These serves as quality indicators (QIs). Indeed, QIs are improving tools for measuring the quality of selected aspects of care by comparing them against defined criteria [11,12]. This promotes accountability, help in decision makings to set priorities and thus help in comparison to be made between timelines and setting effective interventions.

Pre-Analytic Monitoring

Major bulk of laboratory errors occur at pre-analytic stage of almost 70% or more [13,14]. Organizations use different options for pre-analytic solutions to overcome this greatest challenge to laboratory professionals. We are using Roche solutions and observing the workload on navify such as Order Volume, Sample Volume and Count of tests by test (Figure 2). Usually pre-analytic phase has always been an area of focused for improvement in sample collection such as sample rejection due hemolysis, lipemia, wrong barcode label, specimen lost, etc [15]. Other than

that most striking QI would be workload analysis of phlebotomy done. Good laboratory practice includes balance requirements of staff to adequately provide patient services [16]. Laboratory testing generally begins with phlebotomy tasks. One of the obvious challenge that laboratory face and should handle in effective manner is deploying phlebotomy services considering the peaks and valley of patient influx. By having volume data of pre-analytic we have been able in removing excess staff and thus improving cost reduction. Other than that we have been able to correct appropriate work force required on specific day time and area has been corrected and the service outcomes improved. The patient's prediction for improved staffing is one of the striking feature for the one who likes to use pre-analytic QI dashboard [17]. This helps us implementing the concept of Lean and Six Sigma process improvement at pre-analytic phase of laboratory. We implemented methodical modifications to inpatient and outpatient phlebotomy services achieving higher patient satisfaction. This resulted due to improvement in the timeliness of specimen collections and shortening waiting time for patients. By this we have been able to improve motivation for work and decrease the absenteeism rate among the staff. Other than that by monitoring 'Change in Sample Volume and Number of Individual Tests' (Figure 2) is a useful tool. Although laboratory services only account for 5 percent of a hospital's budget, they influence 60 to 70% of all important decisions, including patient admittance and discharge [18]. This BI data determining the true potential cost savings or new net revenue from projects under consideration by the hospital system. The other utility of pre-analytic monitoring, we are exploring is blood concentrations of various analytes change during the course of the day. These cyclical variations can be significant, so the timing of sample collection should be strictly controlled. Chemistry analytes such as hormones levels are affected by circadian variation [19]. With the help of navify we are able to monitor timing of sample collection of such tests, e.g. TSH, cortisol, testosterone, etc. With such informative data, we are able to guide our physicians and patients.

Analytic Monitoring:

BI tools are often a good choice for a laboratory to start when approaching an analytic problem, as it is easier to understand visualized data. By using navify we have been able to monitor our laboratory turnaround time (TAT), a promising quality indicator for analytic business tools. TAT is one of the most important measurable tools of laboratory service and is always been used as a key performance indicator of laboratory performance. By monitoring TAT one can organize workflow in the laboratory [20]. According to the International Organization for Standardization (ISO) guidelines, each laboratory shall establish turnaround times for each of its tests that reflect clinical needs in consultation of physicians, and shall periodically evaluate whether or not it is meeting the established turnaround times. TAT prolongation can be minimized by checking the outliers and reason behind them. Increase TAT would cause delay in diagnosis and treatment of patients. Holland et al. claim that there was 43% treatment delay and 61% increased length of stay in the emergency department due to raised TAT [21]. Other effects of delay TAT might be increase in workload due to reorder of tests by physicians as STAT, thus increasing the cost burden of health care. Therefore strict monitoring of TAT is required for effective laboratory management in addition to focused business QI. We have been able to monitor TAT using navify (Figure 3). Total laboratory automation improves the efficiency of the laboratory. Reporting improvement in laboratory productivity leading to decreased laboratory workforce has been recently claimed by Al Naam et al [22]. As less workforce is required to operate TLA, this reduce labor work is transferred to handle problems in rectifying outliers in pre and post-analytic area, so consequently improving total TAT. This has also been achieved using this BI in our laboratory work. In addition to just simple monitoring of TAT, we have been able to use monitoring, change in TAT and outliers for different tests such as cardiac markers, renal functions tests, liver function tests, hormones, etc. This gives us an edge on having additional information to improve our business strategies as we monitored change and reason behind of improvement or decline in our system. To cope up with high volume complexed tests, clinical laboratories in advance era equipped with high throughput auto-analyzers and even total laboratory automation. By using highly sophisticated automated analyzers, massive amount of laboratory data are generated. Mostly the auto-validation rules are set in to cope up with high demand results within TAT [23]. Auto-validation rules are designed based on auto-release range using clinical decision points, analytical measurement ranges, delta checks and dilution rules. The use of interactive dashboard for auto-validation allows visual display and enables the data-driven decision-making process ease for us. Monthly more than 95% results are auto-validated based on our laboratory set rules (Figure 3). This improves our business goal by providing timely results to ordering physicians. We found auto-validation as another machine learning tools which improves our experience of analytic throughput by auto-validation and increase reliability that have been traditionally subjective when performed by humans.

Post-Analytic Monitoring

Post-analytics phase is the end stage of testing process, in which not only test results are finalized with reference ranges but also other important aspect of laboratory management are dealt with [24]. What important aspects other than TAT we focused are instrument utilization, improved supply chain, workload monitoring by instruments, by weekdays and by hours and supply chain. All the data from instrument log files and infinity (middleware) is used to gather these important information.

The scope of service of a modern day laboratory is most likely dependent upon availability of highly sophisticated and complexed laboratory instrument. To achieve our business goal, one should know about instrument utilization. This data about work quantification have substantial impact on how laboratories

should manage business. The insufficient visibility into instrument availability and utilization is a challenge for many laboratory that is faced nowadays due to lack of data utilization. We have been able to overcome this problem by using navify. We have different analyzers placed at different locations connected to navify digital solutions and are monitoring the utilization by workload balance ratio (Figure 4). This process mining enables us to review preventive maintenance policies and processes during machine downtime as suggested by Tsai et al. [25]. By using the factual BI software, laboratory can collect and analyze real-time data about the performance of their laboratory in areas such as operations and finances, as well as personnel management. It is obvious to use laboratory instrument and staff efficiently to keep our business goals alive and prosperous. For this simulation tool in healthcare is applied by different researchers [26]. Lote et al. focused to increase resource utilization by having laboratory personnel use resources more evenly [27]. Bottlenecks in the existing and potential configurations of laboratory tests were assessed by Kadi et al. [28]. In order to determine which resources had the highest utilization, the writers calculated each resource's utilization. We have been able to deploy staff according to much needed area as navify give us true insights to define staffing schedules to cover demand from pre to post analytic phases (Figure 4). Getting this level of insight in such an intuitive way allows us to redirect resources where they are most needed and optimize areas that are not performing well to ensure the best return on investment possible. We found sample workload that is sample throughput is measurable business intelligence tool [29]. The maximum throughput of each resource must be known in order to calculate the maximum throughput of a laboratory; however, it might not be able to gauge staff work pace when faced with their maximum workload. The goal is to maximize throughput in general. A higher throughput corresponds to a higher production rate, implying that more samples were processed in the laboratory throughout a given time period. Laboratories with a suitably high maximum throughput can handle peak demand and, in the event of machine breakdown, can quickly process delayed samples when the laboratory is operating again or using backup laboratory analyzers. More obvious advantage for this BI tool is that laboratories may elect to accept more samples, such as those from other laboratories and even clinical trials. Any clinical laboratory can increase its capacity to do more with less and do it effectively, giving more value for the money spent on healthcare, by rethinking workflow and procedures. Things to focus on data for workload completion. Once we have data, we can easily correct causes of outliers. This definitely boost productivity, efficiency and quality in a clinical lab. And this feature has been the consistently used by our laboratory management.

What Additional Features can benefit - Recommendations

Although we have successfully implemented the policy of 'Reporting of Critical Values' in our hospital but our wish list for improving dashboard for BI includes addition of data for critical laboratory result. Informing of critical laboratory value in timely manner always affect patient treatment and hospital stay [24,30]. All laboratories must follow strict guidelines for reporting critical values, as mandated by the international and local accreditation agencies such as College of American Pathologists (CAP) [31]. This includes implementing a strong quality assurance system. Laboratory critical value is always has been listed and agreed depending upon a number of variables, including variations in staffing, equipment, patient demographic and clinical demand in a hospital. Laboratory professionals working together with other stakeholders in the delivery of healthcare has been able to develop hospital wide critical laboratory values. Laboratory supply chain management is so crucial for critical ill patient that if not managed correctly will lead to disaster for hospital management. Focusing on unforeseen problems in maintaining and delivering laboratory supplies can be easily handled by using valuable analytics. Having the analytics available both descriptive and predictive models can be applied enhancing the decisions for negotiating prices, reducing the variation in supplies, and optimizing the ordering process as a whole. This will keep the supply chain well-organized from end to end and avoids bottlenecks. McHugh has claimed reduce in waste and reduce inventor expenses by almost 8 percent which gives more focused understanding of operations [32]. Using the artificial intelligence or predictive analytics features such as forecasts or value comparisons can give data alerts using the database available. By creating this future prediction dashboard will explore available data and predict business future. By this feature one will be able to view data from the past, present, and future on a single screen.

How BI will impact laboratories in future, concluding remarks

In summary, a business intelligence tool (BI) in laboratory workflow offers numerous utilities that improve our operational efficiency, quality control, inventory management, cost analysis, decision support, and compliance reporting. It empowers laboratory managers and personnel with valuable insights to enhance performance, optimize processes, and deliver better patient care. This monitoring bring insights about the interrelationships among BI value to our diagnostic laboratory. It has been shown by Phillips-Wren G and McKniff S et al. that "how the choice of visualization of workflow and operational processes impacts decisions to embrace real-time, big data technology" [33]. The findings help IMC Lab to focus on what causes the benefits related to BI implementation that lead to greater competitive advantage. As laboratories grapple with the imperative to advance their workflows through data-driven strategies, a nuanced comprehension of the challenges they encounter is pivotal. By navigating these hurdles, laboratories can harness the transformative potential of BI and analytics solutions to expedite their progression towards efficiency, innovation, and excellence. This Digital transformation is an opportunity to accelerate healthcare performance by lowering cost and

improving quality of care [4,34]. We found navify as one of the ideal laboratory BI platform to achieve our business targets. This and other BI tools in markets as procedure of digitalization is to be seen as required to pursue innovation and remain competitive. In laboratories of the future, laboratory workflows will be more productive, efficient, and flexible due to these BI tools. True laboratory BI is definitely understanding the required data in a way that it brings solution to our laboratory business.

Declarations

Funding

No funding was required or done.

Ethical approval

The study has been approved by Institutional Review Board, International Medical Center, Research Center, Jeddah, Kingdom of Saudi Arabia (IRB approval # 2023-09-226). However, because of the nature of the study (retrospective review of records), informed consent was not required from the study subjects. The need for informed consent was waived by an IRB.

Consent to Publish

Not applicable.

Availability of Data and Materials

Data is available on request.

List of Abbreviations

Business intelligence (BI);
College of American Pathologists (CAP);
International Medical Center (IMC);
International Organization for Standardization (ISO);
Key performance indicators (KPIs);
Laboratory information system (LIS);
Laboratory turnaround time (TAT)
Quality indicators (QIs)

Competing interests

The authors declare that they have no competing interests.

Authors' Contributions

IM: Protocol development, gaining ethical approval, manuscript writing.

FJD: Record review and data collection, data analysis, manuscript writing.

All authors reviewed and edited the manuscript and approved the final version of the manuscript to be published.

Figure 1: Analytic Dashboard For Core Lab

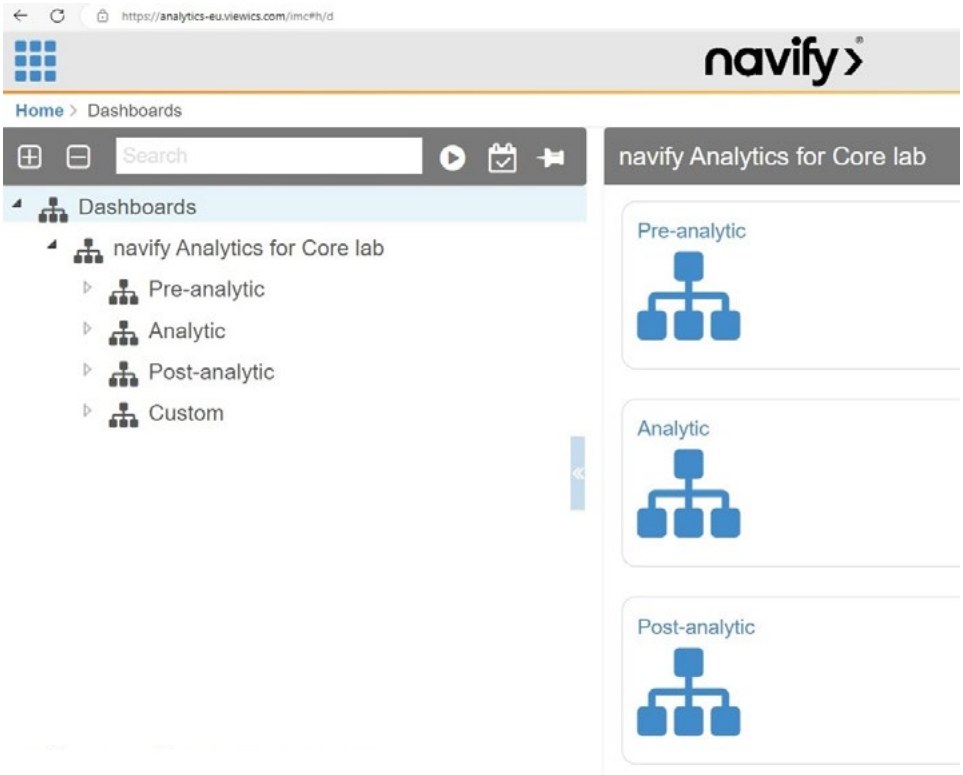
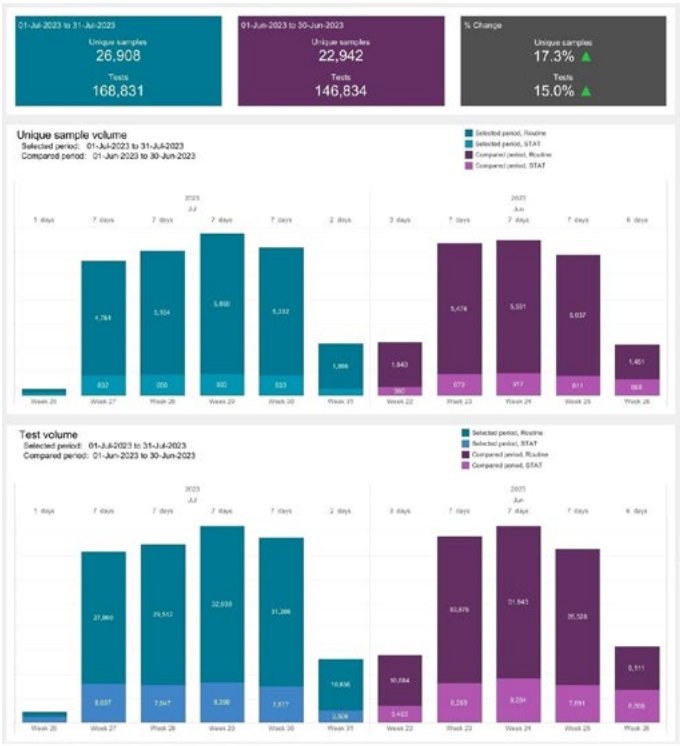
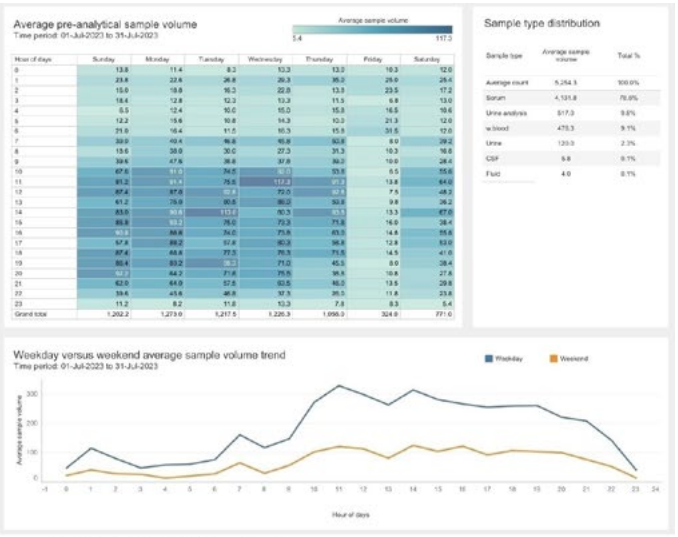


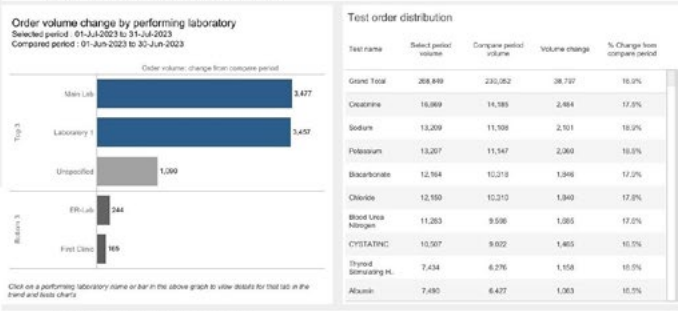
Figure 2: Pre-Analytic Monitoring - Change in Order, Sample And Test Volume
a) Pre-Analytic Sample & Test Volume Percent Change in Orders
b) Pre-Analytic Sample Volume By Hours Of Days
c) Order Volume Change By Laboratory Site



a) Pre-Analytic Sample & Test Volume Percent Change in Orders



b) Pre-Analytic Sample Volume By Hours Of Days



c) Order Volume Change By Laboratory Site

Figure 3: Analytic Monitoring - Turnaround Time and Auto-Validation
a) Monthly Turnaround Time Distribution
b) Yearly Turnaround Distribution
c) Auto-Validated Test Volume Per Sample
d) Analytic Volume Percent Change Turnaround Time For Cardiac Markers

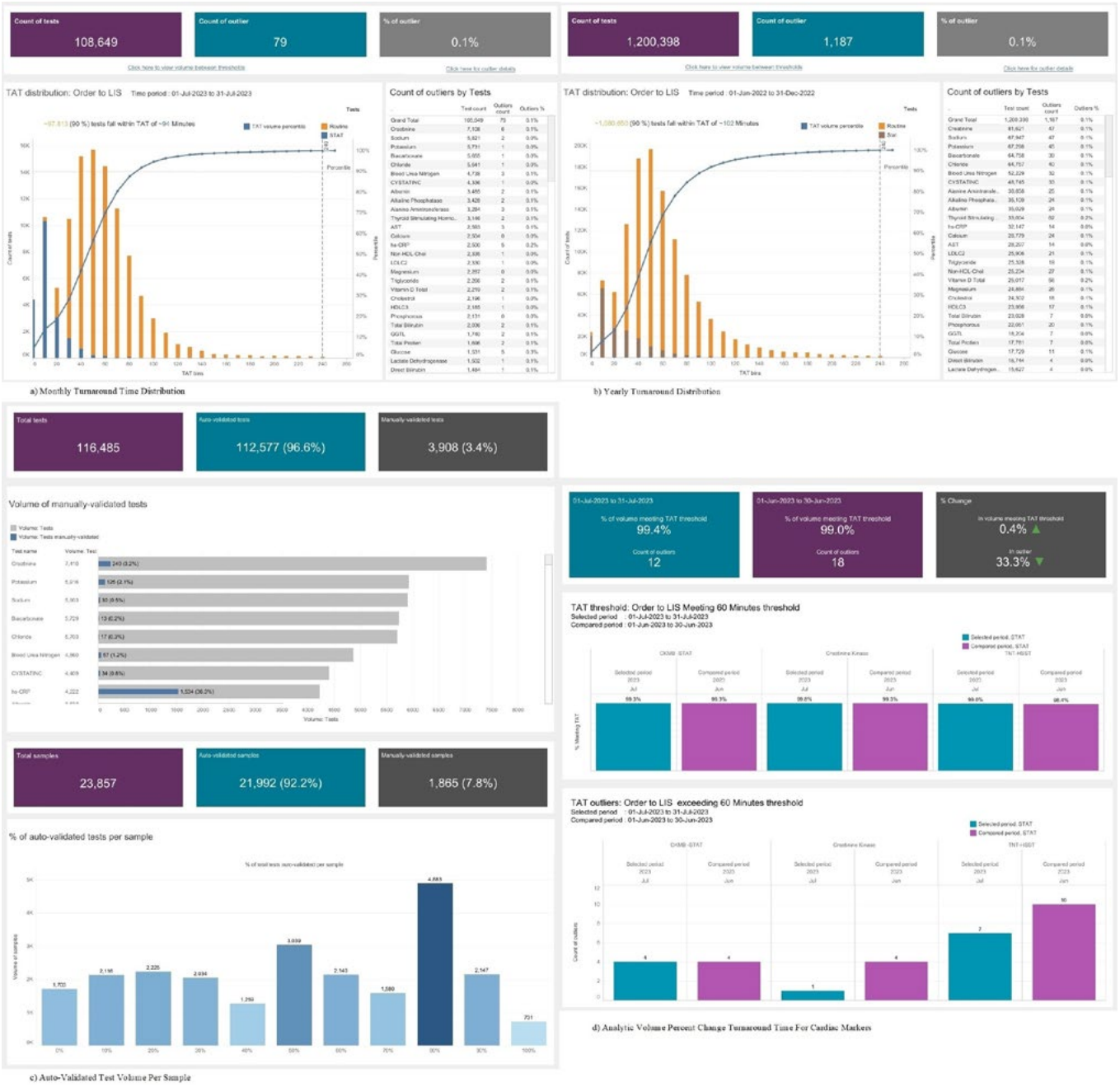


Figure 4: Post-Analytic Monitoring - Workload
a) Analytic Workload Balance Daily Ratio By Instruments
b) Instrument Utilization Percent For Each Instrument In Lab
c) Sample Workloads By Days And Hours On Different Lab Instruments
d) Monitoring Daily Workload Completion
e) Test Analyzed By Days And Hours
f) Test Analyzed On Different Analyzer With Number Of Sample Analyzed And Test Result
g) Test Rerun Overview

References

1. Konopik J, Blunck D. Development of an Evidence-Based Conceptual Model of the Health Care Sector Under Digital Transformation: Integrative Review. J Med Internet Res. 2023;25:e41512.
2. Alghamdi NA, Al-Baity HH. Augmented Analytics Driven by AI: A Digital Transformation beyond Business Intelligence. Sensors (Basel). 2022;22(20):8071.
3. Shirts BH, Jackson BR, Baird GS, Baron JM, Clements B, Grisson R, et al. Clinical laboratory analytics: Challenges and promise for an emerging discipline. J Pathol Inform. 2015;6:9.
4. Alami H, Gagnon MP, Fortin JP. Digital health and the challenge of health systems transformation. mHealth. 2017;3:31.
5. Haymond S. Create laboratory business intelligence dashboards for free using R: A tutorial using the flexdashboard package. J Mass Spectrom Adv Clin Lab. 2022;23:39–43.
6. Cotten SW. Comparing Commercial, Vendor-Specific vs Open-Source Business Intelligence Dashboard Solutions. J Appl Lab Med. 2023;8(1):223–5.
7. Riben M. Laboratory Automation and Middleware. Surg Pathol Clin. 2015;8(2):175–86.
8. Patel JA, Sharma P. Online Analytical Processing for Business Intelligence in Big Data. Big Data. 2020;8(6):501–18.
9. Bagwell H, Coughlin B, Jarvis D. Middleware services may be the key to its return on investment. MLO Med Lab Obs. 2008;40(5):38, 40–1.
10. Loewen L, Roudsari A. Evidence for Busines Intelligence in Health Care: A Literature Review. Stud Health Technol Inform. 2017;235:579–83.
11. Barth JH. Clinical quality indicators in laboratory medicine. Ann Clin Biochem Int J Lab Med. 2012;49(1):9–16.
12. Sciacovelli L, Padoan A, Aita A, Basso D, Plebani M. Quality indicators in laboratory medicine: state-of-the-art, quality specifications and future strategies. Clin Chem Lab Med CCLM. 2023;61(4):688–95.
13. Plebani M, Sciacovelli L, Aita A, Chiozza ML. Harmonization of pre-analytical quality indicators. Biochem Medica. 2014;24(1):105–13.
14. Plebani M. Exploring the iceberg of errors in laboratory medicine. Clin Chim Acta. 2009;404(1):16–23.
15. Lippi G, Von Meyer A, Cadamuro J, Simundic AM. Blood sample quality. Diagnosis. 2019;6(1):25–31.
16. 16. Novis DA, Long T, Blond B, Tworek J, Talbert ML. Phlebotomy Staffing. Arch Pathol Lab Med. 2022;146(6):686–90.
17. Mijailovic AS, Tanasijevic MJ, Goonan EM, Le RD, Baum JM, Melanson SEF. Optimizing Outpatient Phlebotomy Staffing: Tools to Assess Staffing Needs and Monitor Effectiveness. Arch Pathol Lab Med. 2014;138(7):929–35.
18. Forsman RW. Why is the laboratory an afterthought for managed care organizations? Clin Chem. 1996 ;42(5):813–6.
19. Gamble KL, Berry R, Frank SJ, Young ME. Circadian clock control of endocrine factors. Nat Rev Endocrinol. 2014;10(8):466–75.
20. Inal TC, Goruroglu Ozturk O, Kibar F, Cetiner S, Matyar S, Daglioglu G, et al. Lean six sigma methodologies improve clinical laboratory efficiency and reduce turnaround times. J Clin Lab Anal. 2018;32(1):e22180.
21. Holland LL, Smith LL, Blick KE. Reducing laboratory turnaround time outliers can reduce emergency department patient length of stay: an 11-hospital study. Am J Clin Pathol. 2005;124(5):672–4.
22. Al Naam YA, Elsafi S, Al Jahdali MH, Al Shaman RS, Al-Qurouni BH, Al Zahrani EM. The Impact of Total Automaton on the Clinical Laboratory Workforce: A Case Study. J Healthc Leadersh. 2022;14:55–62.
23. Krasowski MD, Davis SR, Drees D, Morris C, Kulhavy J, Crone C, et al. Autoverification in a core clinical chemistry laboratory at an academic medical center. J Pathol Inform. 2014;5(1):13.
24. Lenicek Krleza J, Honovic L, Vlasic Tanaskovic J, Podolar S, Rimac V, Jokic A. Post-analytical laboratory work: national recommendations from the Working Group for Post-analytics on behalf of the Croatian Society of Medical Biochemistry and Laboratory Medicine. Biochem Medica. 2019;29(2):020502.
25. Tsai ER, Tintu AN, Boucherie RJ, de Rijke YB, Schotman HHM, Demirtas D. Characterization of Laboratory Flow and Performance for Process Improvements via Application of Process Mining. Appl Clin Inform. 2023;14(1):144–52.
26. Yang T, Wang TK, Li VC, Su CL. The optimization of total laboratory automation by simulation of a pull-strategy. J Med Syst. 2015;39(1):162.
27. Lote R, Williams EJ, Ülgen OM. Simulation Of Medical Laboratory Operations To Achieve Optimal Resource Allocation. In: ECMS 2009 Proceedings edited by J Otamendi, A Bargiela, J L Montes, L M Doncel Pedrera [Internet]. ECMS; 2009 [accessed: 15/02/2024]. p. 249–55. Available from: https://www.scs-europe.net/dlib/2009/2009-0249.htm
28. Kadı D, Kuvvetli Y, Çolak S. Performance analysis of a university hospital blood laboratory via discrete event simulation. SIMULATION. 2016;92(5):473-84.
29. Modrak V, Soltysova Z, Bednar S. Performance Evaluation of Layout Designs by Throughput Rate and Operational Complexity. Procedia CIRP. 2017;62:175–80.
30. Lippi G, Mattiuzzi C. Critical laboratory values communication: summary recommendations from available guidelines. Ann Transl Med. 2016;4(20):400–400.

31. AlSadah K, S El-Masry O, Alzahrani F, Alomar A, Ghany MA. Reporting Clinical Laboratory Critical Values: A Focus On The Recommendations Of The American College Of Pathologists. J Ayub Med Coll Abbottabad JAMC. 2019;31(4):612–8.
32. 32. McHugh TM. Supply chain management in the clinical laboratory. Clin Leadersh Manag Rev J CLMA. 2006;20(1):E4.
33. 33. Phillips-Wren G, McKniff S. Overcoming Resistance to Big Data and Operational Changes Through Interactive Data Visualization. Big Data. 2020;8(6):528–39.
34. 34. Herrmann M, Boehme P, Mondritzki T, Ehlers JP, Kavadias S, Truebel H. Digital Transformation and Disruption of the Health Care Sector: Internet-Based Observational Study. J Med Internet Res. 2018;20(3):e104.

Review Article

Triglyceride-Glucose Index As A Biomarker Of Insulin Resistance, Diabetes Mellitus, Metabolic Syndrome, And Cardiovascular Disease: A Review

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Cardiovascular, diabetes, insulin resistance, TyG index

Abstract

The triglyceride-glucose (TyG) index is one of the parameters that have been used in assessing insulin resistance. Triglycerides and fasting blood glucose are two low-cost, common laboratory indicators that are used to compute the TyG index. This article reviews the link between the TyG index and several aspects concerning insulin resistance-related disorders and cardiovascular disease, as well as the use of various TyG index cutoffs in the above conditions with sensitivity and specificity, respectively, in various populations in the world.

Introduction

The triglyceride and glucose index or TyG index is a parameter that has been widely used recently in various reports and research in the field of medical laboratories concerning insulin resistance-related disorders and cardiovascular disease [1,2]. The calculation of TyG index is easy and inexpensive to do because it only requires the results of fasting blood triglycerides and glucose, which are routinely examined both in hospitals and clinical laboratories [3].

The lipoproteins that have the highest triglyceride content include chylomicron and very low-density lipoprotein (VLDL). Hyperglycemic conditions and insulin resistance trigger increased VLDL production and the release of chylomicron, which cause serum triglyceride levels to rise in these subjects [4]. Insulin resistance in the liver causes liver metabolism disruptions in regulating blood glucose levels. In insulin resistance and diabetes mellitus, hepatic glycogenesis decreases, while increased hepatic glyconeogenesis leads to increased hepatic glucose production, so blood glucose levels increase [5]. The association of the increased of triglycerides and glucose levels in insulin resistance is the basis for the use of both (indicated in the TyG index) to assess various abnormalities related to insulin sensitivity disorders. The TyG index is measured using the formula: TyG index = Ln (fasting triglycerides [mg/dL] x fasting glucose [mg/dL])/2 or TyG index = Ln (fasting triglycerides [mmol/L] x 88.57 x fasting glucose [mmol/L] x 18)/2) [6].

The role of TyG index in assessing insulin resistance

Insulin resistance is one condition that precedes the occurrence of diabetes. Among patients with insulin resistance, blood insulin production in normal amounts cannot optimally trigger the transfer of glucose from within the blood to peripheral tissues, including muscle and fat tissues, so blood glucose levels tend to rise. To maintain normal blood glucose levels, more insulin production is needed due to insulin resistance [7,8]. When it comes to diagnosing insulin resistance, the hyperinsulinemic-euglycemic clamp (HEC) method is regarded as the best, but this technique is rather complicated, less practical, and quite expensive. Some easier-to-do tests have been used as alternative tests to diagnose insulin resistance condition including the homeostatic model assessment for insulin resistance or HOMA-IR, the Matsuda index, and other tests [7]. The TyG index is a fairly easy and affordable test to be conducted. Various studies have shown its usefulness in assessing insulin resistance [1]. Guerrero-Romero et al. reported that TyG Index showed an excellent association with HEC gold standard method in diagnosing insulin resistance (area under the curve (AUC) = 0.858), with TyG index cutoff 4.68 having a sensitivity of 96.5% and a specificity of 85% in estimating insulin resistance in the adult population [6]. Several investigations have been carried out to evaluate the TyG index's ability to measure insulin resistance based on HOMA-IR because the HEC method is quite difficult to do. Aman et al. showed that the TyG index was associated with HOMA-IR (r = 0.436) and could be used to predict insulin resistance using a cutoff of 4.66 (sensitivity 86.2%, specificity 44.1%) in an adult male population of non-diabetic mellitus in Indonesia [9]. A study conducted in the adult population of Venezuela showed that TyG index with 4.49 cutoff had specificity of 0.821 and sensitivity of 0.826 in determining insulin resistance (this study used a cutoff HOMA-IR >2 to diagnose insulin resistance) [10]. A study in the 2170 population of Xinjiang Kazakh in China demonstrated that TyG and the body mass index (BMI) had the closest relationship with the incidence of insulin resistance (HOMA-IR > 3.45 (75 percentile)) [11]. The TyG index may also predict the occurrence of insulin resistance in children. A study conducted on 915 school-age children in Argentina showed that the TyG index correlates with HOMA-IR (r = 0.34), but its ability to predict insulin resistance is generally lower than that reported in adults (AUC = 0.65, cutoff TyG Index = 8.00, sensitivity 0.62, specificity 0.62) [12]. A study among teenagers aged 10-19 in South Korea showed the 8.26-cutoff TyG index could predict the occurrence of insulin resistance (HOMA-IR >95th percentile) with an AUC value of 0.723 (sensitivity 66.45% and specificity 65.56%) [13]. In the population of women with polycystic ovarian syndrome (PCOS), HOMA-IR and TyG index were also shown to be significantly correlated (r = 0.515). By using HOMA-IR cutoff > 2.5 as having insulin resistance, the occurrence of insulin resistance in PCOS subjects can be predicted by TyG index (AUC = 0.781, the cutoff TyG Index = 8.51, with specificity of 87% and sensitivity of 63.2%) [14]. Table 1 provides an overview of the TyG index's role in predicting the occurrence of insulin resistance.

The role of TyG index in assessing diabetes mellitus

TyG index has also been extensively studied concerning diabetes mellitus. One of the risk factors for type 2 diabetes is insulin resistance. Hyperglycemia due to abnormalities in insulin secretion or insulin resistance in peripheral tissues is a hallmark of diabetes mellitus [15]. Various reports describe the association between TyG index and various aspects of diabetes mellitus. A study of 2,900 subjects undergoing medical check-ups in Korea showed that the TyG index might be used as a marker in assessing risk of developing diabetes. Those with TyG index >8.97 (quartile 4) had a hazard ratio of having diabetes 5.65 times higher than those with a TyG index <8.21 (quartile 1) [16)]. A study conducted on 140 type 2 diabetes subjects in India showed that TyG index with a cutoff of > 15.50 could be used to predict poor glycemic control (HbA1c >7%) with an AUC = 0.802 [17]. Another study performed in 914 subjects, including normoglycemic, prediabetic, and diabetic patients, in China showed that TyG Index can be applied for assessing the function of pancreatic β cells. The TyG index value had negative correlation with pancreatic β cells function in the three groups above. The cutoff value of TyG index 9.08 can be used to determine the occurrence of early β phase cell dysfunction, whereas the cutoff value of 9.20 can be utilized to evaluate advanced β-phase cell dysfunction [18]. TyG index is reported having a stronger predictor value than HOMA-IR in diagnosing type 2 diabetes among adolescents and children subjects in Korea (AUC 0.839 versus 0.645) [19]. Interestingly, the TyG Index can also be used for assessing macrovascular complications in type 2 diabetes subjects using the 9.31 cutoff (AUC = 0.702, sensitivity 59%, specificity 74%) [20]. A report of 157 type 2 diabetes subjects conducted in Harbin, China, revealed that the TyG index had association with incidence of mild cognitive impairment in people suffering from type 2 diabetes. The TyG index 9.45 cutoff (AUC = 0.79) can be used to diagnose slight cognitive impairment in those subjects, with 69% sensitivity and 80% specificity [21]. Table 2 provides an overview of the TyG index's role in relation to diabetes mellitus.

The role of the TyG index in assessing metabolic syndrome

Metabolic syndrome, formerly known as syndrome X, is a disorder marked by insulin resistance, impaired glucose and lipid metabolism, and elevated blood pressure linked to a higher risk of cardiovascular disease [22,23]. There are several criteria for establishing metabolic syndrome diagnosis, including those proposed by the American Heart Association (AHA), International Diabetes Federation (IDF), Adult Treatment Panel III (ATP III), European Group for Study of Insulin Resistance (EGIR), and World Health Organization (WHO), which generally involve measuring fasting glucose, triglycerides, high-density lipoprotein (HDL) levels, blood pressure, and waist circumference [22]. The prevalence of metabolic syndrome is increasing worldwide, along with the increasing incidence of overweight and obesity [23].

A study involving a large population (298,652 subjects) in Wuhu, China, reported the greater the TyG index quartile, the greater the percentage of population suffering from metabolic syndrome. The TyG index has a higher AUC (AUC = 0.89) than triglycerides (AUC = 0.77) and fasting glucose (AUC = 0.81) in diagnosing metabolic syndrome. The best TyG index cutoff value in determining metabolic syndrome is 8.85, with 81% sensitivity and 91% specificity [24]. Another report from China that conducted a study of 30,291 subjects found that TyG index has a better AUC value than metabolic score for IR (METS-IR) and ratio of triglyceride/HDL in diagnosing metabolic syndrome. In male subjects, the 8.81 TyG index cutoff (AUC = 0.863) had 77.47% sensitivity and 83.55% specificity in determining metabolic syndrome, while in women, a cutoff of 8.73 (AUC = 0.867) had 71.49% sensitivity and 88.57% specificity in determining metabolic syndrome [25]. Similar findings were reported in the Argentina population, which found the TyG index had a higher AUC value than triglyceride/HDL ratio to determine the occurrence of metabolic syndrome (AUC = 0.88 vs. 0.85) with a TyG index cutoff value similar to the population in China of 8.80 in males (sensitivity 84%, specificity 82%) and 8.70 in females (sensitivity 72%, specificity 91%) [26]. A meta-analysis and systematic review was conducted to assess accuracy of TyG index in determining metabolic syndrome in adults, which analyzed 13 reports with a total of 49,325 subjects, reported that the summary receiver operating characteristic (ROC) curve in male had AUC of 0.90 (79% specificity, 82% sensitivity) and in female had AUC of 0.87 (85% specificity, 81% sensitivity), so that TyG index showed the capability to determine the occurrence of metabolic syndrome with high accuracy, although the determination of TyG index cutoff value for each population needs further investigation [3]. Table 3 provides an overview of the TyG index's role in identifying the presence of metabolic syndrome.

The role of TyG index in cardiovascular disorders

Globally, cardiovascular disease is the primary cause of mortality. The incidence of cardiovascular disease increased by almost 100% from 1990 (estimated at 271 million cases) to 2019 (estimated at 523 million cases). Deaths from cardiovascular disease increased by nearly 50% (18.6 million) in 2019 compared to 1990 (12.1 million). Ischemic heart disease is the primary mortality cause from cardiovascular disease, which accounts for nearly 50% of deaths [27]. The number of deaths from ischemic heart disease worldwide in 2021 is estimated at 9,440,00 deaths [28]. Countries with the largest number of deaths from cardiovascular disease include China, India, Russia, the United States, and Indonesia [27]. The TyG index also plays a role in assessing incidence of cardiovascular disorders. Yoon et al. who conducted a study on more than 9000 Korean children and adolescents, reported the TyG index is linked with various cardiometabolic variables including systole and diastole blood pressure, glucose, triglycerides, and waist circumference while it is negatively associated with HDL [19]. Fiorentino et al. conducted a study on 631 normoglycemic, prediabetes, and diabetes subjects, finding that subclinical vascular damage characterized by vascular atherosclerosis and vascular stiffness deals with various markers of insulin sensitivity, including the TyG index. TyG index with cutoff 9.19 can be used to assess the presence of vascular atherosclerosis (AUC = 0.739, sensitivity 82.5%, specificity 59.2%), and cutoff 8.99 can be used to assess vascular stiffness (AUC = 0.579, sensitivity 74.4%, specificity 41.7%) [29]. The TyG index had association with mortality, both during treatment and during follow-up, in patients with cardiovascular disease. Zhai et al. in China conducted a study on 4839 hospitalized heart disease subjects with critical condition. The higher the TyG index quartile, the higher the mortality rate during treatment (12.1% in quartile 4 with a TyG index >9.37 vs. 5.3% in quartile 1 with a TyG index <8.51), and the longer the intensive care unit stay period [30]. Jin et al. performed an investigation on 3,745 subjects with stable coronary artery disease in China and followed them up for 36 months. They found that TyG index had positive association with cardiovascular events (mortality and myocardial infarction) during the monitoring period. Patients in highest quartile (TyG index >9.17) experienced higher risk of cardiovascular events than subjects in quartiles 3, 2, and 1 (20.3% vs. 17.5% vs. 12.8% vs. 16.1%) [31]. In healthy adults, the TyG index may also be used to predict the occurrence of cardiovascular disease. Cho et al. conducted a study using data from more than 6 million healthy young Korean adults, conducted 7.4 years of median time of monitoring, and found that the higher the TyG index, the greater the hazard of stroke, myocardial infarction, and mortality. Subjects in quartile 4 had a 25.8% myocardial infarction risk and 15.1% mortality, higher than quartile 1 [32]. TyG index had association with mortality in subjects with acute decompensated heart failure. Huang et al. conducted a study on 932 acutely decompensated heart failure patients hospitalized in China. During the 478-day (median) follow-up, subjects with the top tertile (TyG index >9.32) got a 2.31 times higher cardiovascular-related death risk compared to subjects with the bottom tertile (TyG index <8.83) [33]. Table 4 provides an overview of the TyG index's role in relation to cardiovascular disease.

Conclusions

The TyG index is a low-cost insulin resistance parameter and widely studied in several populations around the world concerning insulin resistance-related disorders and cardiovascular disease. The TyG index correlates well with the HEC method of assessing insulin resistance. The TyG index may be used in predicting the occurrence of insulin resistance, both in pediatric and adult populations. The TyG index could also be used in predicting the occurrence of diabetes mellitus, assess poor glycemic control in diabetes, assess diabetes complications, and assess the function of pancreatic β cells. The TyG index can also be applied to assess the presence of metabolic syndrome in various populations around the world. The TyG index also had significant association with cardiometabolic risk factors, vascular disorders, cardiovascular

events, and mortality and can be used as a cardiovascular disorder predictor. In the studies above, various TyG cutoff values were found that vary depending on population and conditions, so further study is needed to determine the TyG cutoff index that can be used universally in these conditions. The limitation of this review is that most of the data obtained comes from Asian populations and data from the Caucasian race is still limited . In general, high TyG index value is linked to the occurrence of insulin resistance, diabetes mellitus, metabolic syndrome, and cardiovascular disease risk, as well as the occurrence of various complications related to the above conditions.

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CONFLICTS OF INTEREST
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Table 1: The association between TyG index and insulin resistance

Reference	Year	Place, Country	Population	Age (years)	Methods	AUC	TyG index cutoff	Sensitivity	Specificity
6	2010	Guadalajara, Mexico	32 type 2 diabetes and 67 healthy subjects	39.9±9.30	Comparation between TyG index with euglycemic-hyperinsulinemic clamp as the gold standard	0.858	4.68	0.965	0.85
9	2021	Makassar, Indonesia	88 subjects without diabetes	51.15±6.83	Comparation between TyG index with HOMA-IR. HOMA-IR cutoff >2.24 (tertile 3) used to define insulin resistance	0.701	4.66	0.862	0.441
10	2018	Maracaibo, Venezuela	2004 subjects >18 years consist of 1050 female and 954 male	39.6±15.3	TyG index was compared with HOMA2IR. HOMA2IR >2 was used to define insulin resistance	0.889 (all subjects) 0.903 (male) 0.871 (female)	4.49 4.51 4.45	0.826 0.872 0.803	0.821 0.831 0.806
12	2022	Argentina	915 school-age children	9.24±2.17	Comparation between TyG index with HOMA-IR. HOMA-IR >3rd quartile (cutoff not mentioned) was used to define insulin resistance	0.65	8.00	0.62	0.62
13	2021	South Korea	3728 young subjects	14.56±0.06	Comparation between TyG index with HOMA-IR. HOMA-IR >95 percentile was used to define insulin resistance	0.723 (all subjects) 0.756 (male) 0.680 (female)	8.26 8.17 8.26	0.664 0.766 0.644	0.655 0.604 0.635

14	2022	Shantou, China	175 female subjects consist of 114 PCOS and 61 control subjects	29 (mean)	Comparation between TyG index with HOMA-IR. HOMA-IR>2.5 was used to define insulin resistance	0.781	8.51	0.632	0.87

Table 2: The role of TyG index concerning diabetes mellitus condition

Reference	Year	Place, Country	Population, Method	Age (years)	TyG index cutoff	Important Finding
16	2016	Seoul, Korea	2900 nondiabetic adults enrolled in the study and followed for 4 years. The baseline of the TyG index was documented, its association with the occurrence of diabetes after 4 years was assessed	44.3±6.5	8.97	Those with an initial TyG index >8.97 (quartile 4) had a 5.65 times hazard ratio having diabetes within 4 years compared to subjects with TyG index <8.21 (quartile 1)
17	2021	Puducherry, India	140 type 2 diabetes mellitus subjects were recruited, the link between TG index and poor glycemic control (HbA1c >7%) was measured	51.2±9.2	15.50	TyG index >15.5 (AUC 0.806) could be used to predict poor glycemic control among type 2 diabetes mellitus patients
18	2022	Changsha, China	914 participants undergoing medical check-ups consisting of 315 diabetes mellitus patients, 276 normoglycemic, and 323 impaired glucose tolerance (IGT) subjects were recruited and the association between TyG index with β cell dysfunction was measured	44.68±9.30 (NGT), 48.46±9.14 (IGT), 50.92±9.47 (DM)	9.08 and 9.20	TyG index >9.08 (AUC 0.68, sensitivity 0.76, specificity 0.53) could be used to predict early-phase β cell dysfunction while TyG index >9.2 (AUC 0.74, sensitivity 0.76, specificity 0.62) could be used to predict late-phase-β cell dysfunction
19	2022	South Korea	170 adolescents and children with overweight and obesity were recruited. TyG index and HOMA-IR ability to predict the occurrence of type 2 diabetes mellitus was compared	11.34±3.24	(-)	TyG index had a better capability compared to HOMA-IR in predicting the occurrence of type 2 diabetes mellitus (AUC = 0.839 vs 0.645)
20	2022	Zhejiang, China	858 type 2 diabetes mellitus subjects were recruited in the retrospective research	67.13±11.07	9.31	TyG index >9.31 could be used to predict macrovascular complications among type 2 diabetes mellitus subjects (AUC 0.702, sensitivity 0.59, specificity 0.74)
21	2022	Harbin, China	517 type 2 diabetes patients were recruited. TyG index was used to predict mild cognitive impairment among the subjects	58 (median)	9.45	TyG index >9.45 (AUC 0.79) had 0.69 sensitivity and 0.80 specificity in predicting mild cognitive impairment in type 2 diabetes patients

Table 3: The association between TyG index and metabolic syndrome

Reference	Year	Place, Country	Population	Age (years)	AUC	TyG Index Cutoff To Define Metabolic Syndrome	Sensitivity	Specificity
24	2022	Wuhu, Anhui, China	298,652 subjects who came for medical check-ups	47.08±12.94	0.89	8.85	0.81	0.91
25	2019	China	30,291 adult subjects. Metabolic syndrome was defined according to harmonized IDF criteria	43.26±13.66	0.863 (male),	8.81	0.774	0.835
					0.867 (female)	8.73	0.714	0.885
26	2014	Bahia Blanca, Buenos Aires, Argentina	525 participants with metabolic syndrome (89 subjects) and without metabolic syndrome (436 subjects)	45 (median, metabolic syndrome), 33 (median, without metabolic syndrome)	0.88	8.80 (male)	0.84	0.82
						8.70 (female)	0.72	0.91
3	2022	(-)	A systematic review of 13 researches involving 49,325 subjects	(-)	0.90 (male)	(-)	0.82	0.79
					0.87 (female)	(-)	0.81	0.85

Table 4: The association between TvG index concerning cardiovascular disorders

Reference	Year	Place, Country	Population	Age (years)	TyG Index	Important Finding
29	2019	Rome and Cantazaro, Italia	631 adults consisting of normoglycemic, prediabetes, and diabetes subjects were recruited in the research. The TyG index was used in determining subclinical vascular damage	39.6±10.7	9.19	TyG Index >9.192 (AUC 0.739) could be used to predict vascular atherosclerosis with 82.5% sensitivity and 59.9% specificity.
					8.99	TyG Index >8.987 (AUC 0.579) could be used to predict increased vascular stiffness with a sensitivity of 74.4% and specificity of 41.7%.
30	2022	China	4,839 heart disease subjects with critical conditions were studied. The relationship of TyG index with mortality while hospitalized was assessed	65.2±13.8	9.37	Patients in highest quartile (TyG index >9.37) had a higher mortality rate than patients in quartile 1 (TyG index <8.51) with mortality 12.1% vs. 5.3% (OR 1.83, 95% CI = 1.27-2.64)
31	2018	China	3,745 subjects with stable coronary artery disease were recruited in the study and were followed up for 3 years. Initial TyG index and cardiovascular events were analyzed	59.5 (mean)	9.17	Subjects in quartile 4 (TyG index >9.17) experienced the highest mortality rate compared to those in quartile 3, 2 and 1 (20.3% vs. 17.5% vs. 12.8% vs. 16.1%)
32	2022	South Korea	6,675,424 healthy subjects joined the national health program and a mean of 7.4 years follow up period performed. The initial TyG index was used in predicting cardiovascular disease and mortality	20-39 (range)	9.42 (quartile 4, male), 8.71 (quartile 4, female)	Subjects in quartile 4 had a 25.8% higher risk of having myocardial infarction and a 15.1% higher risk of mortality compared to those in quartile 1
33	2022	Nanjing, Jiangsu, China	932 subjects with acute decompensated heart failure were followed up (median 478 days)	70 (median)	9.32	Subjects in tertile 3 (TyG index >9.32) had 2.31 times higher cardiovascular-related death risk compared to subjects compared to subjects in tertile 1 (TyG index <8.83)

REFERENCES

1. Sanchez-Garcia A, Rodriguez-Gutierrez R, Mancillas-Adame L, Gonzalez-Nava V, Gonzalez-Colmonero AD, Solis CR, et al. Diagnostic accuracy of the triglyceride and glucose index for insulin resistance: a systematic review. Int J Endocrinol. Volume 2020, Article ID 4678526.
2. da Silva A, Caldas APS, Rocha DMUP, Bressan J. Triglyceride-glucose index predicts independently type 2 diabetes mellitus risk: a systematic review and meta-analysis of cohort studies. Prim Care Diabetes. 2020;14:584-593.
3. Nabipoorahrafi SA, Seyedi SA, Rabizadeh S, Ebrahimi M, Ranjbar SA, Reyhan SK, et al. The accuracy of triglyceride-glucose (tyg) index for the screening of metabolic syndrome in adults: a systematic review and meta-analysis. Nutr Metab Cardiovasc Dis. 2022;32:2677-2688.
4. Laufs U, Parhofer KG, Ginsberg HN, Hegele RA. Clinical review on triglycerides. Eur Heart J. 2020;41:99-109.
5. Jiang S, Young JL, Wang K, Qian Y, Cai L. Diabetic-induced alterations in hepatic glucose and lipid metabolism: the role of type 1 and type 2 diabetes mellitus (review). Mol Med Rep. 2020;22:603-611.
6. Guerrero-Romero F, Simental-Mendia LE, Gonzalez-Ortiz M, Martinez-Abundis E, Ramos-Zavala MG, Hernandez-Gonzalez SO, et al. The product of triglycerides and glucose, a simple measure of insulin sensitivity. Comparison with the euglycemic-hyperinsulinemic clamp. J Clin Endocrinol Metab. 2010;95(7):3347-3351.
7. Li M, Chi X, Wang Y, Setrerrahmane S, Xie W, Xu H. Trends in insulin resistance: insights into mechanism and therapeutic strategy. Signal Transduct Target Ther. 2022;7:216.
8. Petersen MC, Shulman GI. Mechanism of insulin action and insulin resistance. Physiol Rev. 2018;98:2133-2223.
9. Aman M, Resnawita D, Rasyid H, Kasim H, Bakri S, Umar H, et al. The concordance of triglyceride glucose index (tyg index) and homeostatic model assessment for insulin resistance (homa-ir) in non-diabetic subjects of adult indonesian males. Clin Epidemiol Glob Health. 2021;9:227-230.
10. Salazar J, Bermudez V, Calvo M, Olivar LC, Luzardo E, Navarro C, et al. Optimal cutoff for the evaluation of insulin resistance through triglyceride-glucose index: a cross-sectional study in a venezuelan population. F1000Research. 2018;6:133.
11. Yu L, Li Y, Ma R, Guo H, Zhang X, Yan Y, et al. Construction of a personalized insulin resistance risk assessment tool in xinjiang kazakhs based on lipid- and obesity-related indices. Risk Manag Healthc Policy. 2022;15:631-641.
12. Hirschler V, Molinari C, Edit S, Miorin C, Bocco P, Guntsche Z, et al. Ability of tyg index as a marker of insulin resistance in argentinean school children. Front Pediatr. 2022;10:885242.
13. Song K, Park G, Lee HS, Choi Y, Oh JS, Choi HS, et al. Prediction of insulin resistance by modified triglyceride glucose indices in youth. Life. 2021;11(4):286.
14. Zheng Y, Yin G, Chen F, Lin L, Chen Y. Evaluation of triglyceride glucose index and homeostasis model of insulin resistance in patients with polycystic ovary syndrome. Int J Womens Health. 2022;14:1821-2829.
15. Galicia-Garcia U, Benito-Vicente A, Jebari S, Larrea-Sebal A, Siddiqi H, Uribe KP, et al. Pathophysiology of type 2 diabetes mellitus. Int J Mol Sci. 2020;21:6275.
16. Lee DY, Lee ES, Kim JH, Park SU, Park C-Y, Oh K-W, et al. Predictive value of triglyceride glucose index for the risk of incident diabetes: a 4-year retrospective longitudinal study. PLoS ONE. 2016. 11(9):e0163465.
17. Selvi NMK, Nandhini S, Sakthivadivel V, Lokesh S, Srinivasan AR, Sumathi S. Association of triglyceride-glucose index (tyg index) with hba1c and insulin resistance in type 2 diabetes mellitus. Maedica A Journal of Science. 2021;16(3):375-381.
18. Chen Z, Wen J. Elevated triglyceride-glucose (tyg) index predicts impaired islet β-cell function: a hospital-based cross-sectional study. Front Endocrinol. 2022;13:973655.
19. Yoon JS, Lee HJ, Jeong HR, Shim YS, Kang MJ, Hwang IT. Triglyceride glucose index is superior biomarker for predicting type 2 diabetes mellitus in children and adolescents. Endocr J. 2022;69(5):559-565.
20. Yao H, Sun Z, Yuan W, Shao C, Cai H, Li L, et al. Relationship between the triglyceride-glucose index and type 2 diabetic macroangiopathy: a single-center retrospective analysis. Diabetes Metab Syndr Obes. 2022;15:3483-3497.
21. Tong X-W, Zhang Y-T, Yu Z-W, Pu S-D, Li X, Xu Y-X, et al. Triglyceride glucose index is related with the risk of mild cognitive impairment in type 2 diabetes. Diabetes Metab Syndr Obes. 2022;15:3577-3587.
22. Fahed G, Aoun L, Zerdan MB, Allam S, Zerdan MB, Bouferraa Y, et al. Metabolic syndrome: updates on pathophysiology and management in 2021. Int J Mol Sci. 2022;23:786.
23. Rochlani Y, Pothineni NV, Kovelamudi S, Mehta JL. Metabolic syndrome: pathophysiology, management, and modulation by natural compounds. Ther Adv Cardiovasc Dis. 2017;11(8):215-225.
24. Jiang M, Li X, Wu H, Su F, Cao L, Ren X, et al. Triglyceride-glucose index for the diagnosis of metabolic syndrome: a cross-sectional study of 298,652 individuals receiving a health check-up in China. Int J Endocrinol. 2022:3583603.
25. Yu X, Wang L, Zhang W, Ming J, Jia A, Xu S, et al. Fasting triglycerides and glucose index is more suitable for the identification of metabolically unhealthy individuals in the chinese adults population: a nationwide study. J Diabetes Investig. 2019;10(4):1050-1058.
26. Unger G, Benozzi SF, Perruzza F, Pennacchiotti GL. Triglycerides and glucose index: a useful indicator of insulin resistance. Endocrinol Nutr. 2014;61:533-540.

27. Roth GA, Mensah GA, Johnson CO, Addoorato G, Ammirati E, Baddour LM, et al. Global burden of cardiovascular diseases and risk factors, 1990-2019. JACC. 2020;76(25):2982-3021.
28. Vaduganathan M, Mensah GA, Turco JV, Fuster V, Roth GA. The global burden of cardiovascular diseases and risk. JACC. 2022;80(25):2361-2371.
29. Fiorentino TV, Marini MA, Succurro E, Andreozzi F, Sesti G. Relationships of surrogate indexes of insulin resistance with insulin sensitivity assessed by euglycemic hyperinsulinemic clamp and subclinical vascular damage. BMJ Open Diab Res Care. 2019;7(1):e000911.
30. Zhai G, Wang J, Liu Y, Zhou Y. Triglyceride-glucose index linked to in-hospital mortality in critically ill patients with heart disease. Res Cardiovasc Med. 2022;23(8):263.
31. Jin J-L, Cao Y-X, Wu L-G, You X-D, Guo Y-L, Wu Na-Q, et al. Triglyceride glucose index for predicting cardiovascular outcomes in patients with coronary artery disease. J Thorac Dis. 2018;10(11):6137-6146.
32. Cho YK, Han KD, Kim HS, Jung CH, Park JY, Lee WJ. Triglyceride-glucose index is a useful marker for predicting future cardiovascular disease and mortality in young korean adults: a nationwide population-based cohort study. J Lipid Atheroscler. 2022;11(2):178-186.
33. Huang R, Wang Z, Chen J, Bao X, Xu N, Guo S, et al. Prognostic value of triglyceride glucose (tyg) index in patients with acute decompensated heart failure. Cardiovasc Diabetol. 2022;21:88.

Case Report

Autoimmune Hemolytic Anemia as Paraneoplastic Phenomenon in Hodgkins Lymphoma in children – a rare occurrence

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Keywords

Autoimmune Hemolytic Anemia(AIHA), Hodgkin Lymphoma , paraneoplastic phenomenon, autoantibodies.

INTRODUCTION

Autoimmune disorders have been described in children with Hodgkin Lymphoma including autoimmune haemolytic anaemia [AIHA], autoimmune neutropenia, and immune thrombocytopenic purpura [ITP]. The mechanism of AIHA's association with Hodgkin's Lymphoma is not very clear. It has been thought to be due to the autoantibodies produced by tumour cells or as a Para neoplastic phenomenon, or immunity to tumour cells may cross-react with antigens on the red cells [1]. The association of AIHA with lymph proliferative disorders has been well recognized however its association with Hodgkin Lymphoma is rarely reported particularly in children [2]. Eisner et al first reported the association of Hodgkin's disease with immune haemolytic anaemia in 1967 [3].

In this case report, we describe the presentation of AIHA, subsequently diagnosed with Hodgkin Lymphoma.

CLINICAL-DIAGNOSTIC CASE

An 11-year-old boy presented with complaints of right-sided neck swelling and intermittent fever for 2 weeks. The child was well 2 weeks ago with no other significant complaints. On examination, multiple enlarged matted cervical lymph nodes were noted on the right side of the neck measuring approximately 5x3cm, which was non-tender and firm in consistency. A few isolated lymph nodes were also palpable on the left cervical region. No hepatosplenomegaly was noted. The rest of the examination was unremarkable. Baseline investigations done showed haemoglobin 6.3g/dl, mean corpuscular volume 95.4fl, mean corpuscular haemoglobin 22.2pg, mean corpuscular haemoglobin concentration 30.2g/dl, total white blood cell count 4.65 x 10^9/L with neutrophils-55.1% and lymphocytes-32.7%, platelet count 386 x 10^9/L and erythrocyte sedimentation rate-65mm/hour [shown in Table 1]. The basic biochemical analysis was also within the normal limits. Viral serologies were negative for HIV, HBsAg, and HCV. Computed tomography [CT] scan neck showed right cervical and axillary lymphadenopathy. PET CT revealed multiple FDG avid enlarged lymph nodes in the right upper, middle & lower deep cervical, right posterior

triangle, right axillary, right infraclavicular, and splenic hilum region with the largest node measuring 3.1x3cm and smallest measuring 1.5x1.5cm, suggestive of Lymphoma-LUGANO stage III. We planned for an excision biopsy of the right cervical lymph node along with bone marrow aspiration and biopsy to confirm the diagnosis. When cross matching was done before excision biopsy, there was a difficulty recognized in identifying the compatible packed red cell units. Further autoimmune workup was started, direct coombs test [DCT] was positive, peripheral smear showed microcytic hypochromic anaemia, reticulocyte count was 2.3%, there was mild indirect hyperbilirubinemia and serology for antinuclear antibodies was negative. The coagulation profile was within normal limits. The least incompatible blood unit was transfused immediately after the diagnostic procedure was completed under the cover of intravenous methylprednisolone. Histopathological examination of the right cervical lymph node excision specimen depicted effaced architecture of the node consisting of scattered large, atypical cells with nucleate and mononuclear forms resembling Reed Sternberg [RS] cells in a background of paucity of inflammatory cells composed of lymphocytes, plasma cells and occasional neutrophils with diffuse fibrosis [Figure 1, 2]. These typical morphological features are suggestive of classic Hodgkin Lymphoma – Lymphocyte depleted type, further immunohistochemical [IHC] markers were done to confirm the same and to check for the EBV association. By IHC, the large atypical cells were positive for CD15, CD30, EBV-LMP, weak positive for PAX-5, negative for CD45 and the background T & B lymphocytes were positive for CD3 and CD20 respectively which confirmed the above findings. Bone marrow examination was also normal. With this clinical presentation, laboratory investigations and histopathological findings were consistent with that of classic Hodgkin Lymphoma presenting with autoimmune haemolytic anaemia as an association. The patient was started on chemotherapy OEPA [vincristine, etoposide, doxorubicin, and Prednisolone] as per Euronet protocol and he showed a dramatic clinical response with a significant reduction in the size of nodes and improvement in haemoglobin after 1st cycle of chemotherapy.

DISCUSSION

Association of Lymphomas with syndromes of immune dysregulation and B-cell immunodeficiency are well known. Autoimmune manifestations and paraneoplastic features have been described with Hodgkin's Lymphoma commonly, however, are rare in children [2]. Levine et al in their retrospective study of adults with Hodgkin's Lymphoma in the records of 71 cases found Direct Coombs test was positive in seven who had advanced disease of which four had mixed cellularity and three had nodular sclerosis. Coombs test was positive at the time of initial diagnosis in three and at the time of relapse in others [4]. No specific studies in children for incidence are found except for anecdotal case reports.AIHA in Hodgkin's Lymphoma may present concurrently at the time of initial diagnosis of Hodgkin's Lymphoma, during the disease, or rarely can be preceding the diagnosis of Hodgkin's Lymphoma/ relapse as well [5, 6].

Chu observed AIHA in three children with Hodgkin's disease over 7 years [7]. Sierra found that advanced Hodgkin disease particularly nodular sclerosis or mixed cellularity types have a stronger association with AIHA [8]. Apart from autoimmune haemolytic anaemia various immunologic abnormalities, including autoimmune hepatitis, Hashimoto's thyroiditis, and ITP have been reported mostly in adults [9]. We have published a rare association of Hodgkin's Lymphoma with nephrotic syndrome presenting concurrently at the time of diagnosis in an adolescent boy [10]. In rare situations, Hodgkin's Lymphoma associated with AIHA may be Direct antiglobulin test negative possibly due to very low levels of IgG or low affinity bound IgG antibodies which might have been washed away during pre-test processing, or non-IgG antibodies like IgA or IgM [10]. The symptoms of AIHA a not directly related to Hodgkin lymphoma and it gets resolved after the treatment of Hodgkin lymphoma. Hence AIHA is considered a Para neoplastic phenomenon in Hodgkin's lymphoma. Treatment of Hodgkin's Lymphoma is the mainstay of therapy for associated AIHA. Anaemia or other autoimmune cytopenias usually responds to the management of the underlying malignancy and in our case, steroids being the backbone of management of Hodgkin's lymphoma, was useful for both.

TAKE HOME MESSAGES/LEARNING POINTS

Though rare, awareness of the association between Hodgkin lymphoma and autoimmune haemolytic anaemia or other autoimmune manifestations in paediatric practice is crucial since recognition of this association is the key to appropriately evaluating before starting steroids. Steroids mask the diagnosis and may lead to inadvertent delays and significantly affect the outcomes. Clinical examination for lymphadenopathy and organomegaly and appropriate investigations like chest X-ray, abdominal sonogram, or bone marrow studies should be considered before starting a patient on steroids.

Disclosures

This complies with the ethical principles for medical research involving human subjects, by the Declaration of Helsinki.

Conflicts of Interests

None

Author Contribution

JS and SK collected the data and prepared the initial draft. DJ and SG contributed to the review, editing, and final proofing of the draft. All the authors have checked the final manuscript and accepted it.

Table 1: Salient laboratory features at diagnosis

Hemogram	**At diagnosis**	**Reference intervals**
Hemoglobulin	6.3 g/dl	11.5-15.5 g/dl
Mean corpuscular Volume (MCV) Mean Corpuscular Hemoglobin (MCH) Mean Corpuscular HB Concentration (MCHC)	95.4 fl 22.2 pg 30.2 g/dl	77-95 fL 25-33 pg 31-37 g/dl
WBC and differentials	4.65 x 109/L (P : 55.1 % ; L : 32.7 %)	4.5-13.5 x 10^9/L (P : 50-60 % ; L : 24-54 %)
PLT	386 x 10^9/L	150-450 x 109/L
Peripheral smear	Macrocytic anemia; no blasts	Normal study
Reticulocyte count	3.6%	0.5% - 2.5%
Direct Coomb's test	POSITIVE	Negative
CRP ESR	0.8 U/L 65mm/hr	<0.8 U/L 4-10mm/hr
LDH Uric acid	670 U/L 5.6mg/dl	208-378 U/L 3.5- 7.2 mg/dl
Liver function tests	SGPT – 55U/L Albumin – 3.1g/dl Globulin – 3 g/dl Bilirubin - 2.4/ 1.6 mg/dl (total/indirect)	SGPT – 13 - 45 U/L Albumin – 3.5 -5.2 g/dl Globulin – 2 - 3.5 g/dl Bilirubin - 0.3 -1.2mg/dl (total/indirect)
Creatinine	0.4 mg/dl	0.3-0.77 mg/dl

References

1. Burley NB, Dy PS, Hande S, Kalantri S, Mohindroo C, Miller K. Autoimmune Hemolytic Anemia as Presenting Symptom of Hodgkin Lymphoma. Case Rep Hematol. 2021:5580823.
2. Ertem M, Uysal Z, Yavuz G, Gözdaşoğlu S. Immune thrombocytopenia and hemolytic anemia as a presenting manifestation of Hodgkin disease. Pediatr Hematol Oncol. 2000;17(2):181-5.
3. Eisner E, Ley AB, Mayer K. Coombs'-positive hemolytic anemia in Hodgkin's disease. Ann Intern Med. 1967;66(2):258-73.
4. Levine AM, Thornton P, Forman SJ, Van Hale P, Holdorf D, Rouault CL, Powars D, Feinstein DI, Lukes RJ. Positive Coombs test in Hodgkin's disease: significance and implications. Blood. 1980;55(4):607-11.
5. Lechner K., Chen Y.A. Paraneoplastic autoimmune cytopenias in Hodgkin Lymphoma. Leuk Lymphoma. 2010;51:469–474.
6. Gomes MM, Oliva T, Pinto A. Autoimmune Hemolytic Anemia and Hodgkin's Disease: An Unusual Pediatric Association. Case Rep Pediatr. 2016;2016:4598587.
7. Chu JY. Autoimmune hemolytic anemia in childhood Hodgkin's disease. Am J Pediatr Hematol Oncol. 1982;4(2):125-8.
8. Sierra RD. Coombs-positive hemolytic anemia in Hodgkin's disease: case presentation and review of the literature. Mil Med. 1991;156(12):691-2.
9. Saitoh T, Matsushima T, Saito Y, Yamane A, Yokohoma A, Irisawa H et al. Hodgkin Lymphoma presenting with various immunologic abnormalities, including autoimmune hepatitis, Hashimoto's thyroiditis, autoimmune hemolytic anemia, and immune thrombocytopenia. Clin Lymphoma Myeloma. 2008;8(1):62-4.
10. Padmanaban PD, Jayaraman D, Shanmugam SG, Geminiganesan S 2022, Nephrotic syndrome and Hodgkins Lymphoma - an unusual association, eJIFCC. 33:3, 262-267.

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