Pre-analytical issues in Laboratory Medicine

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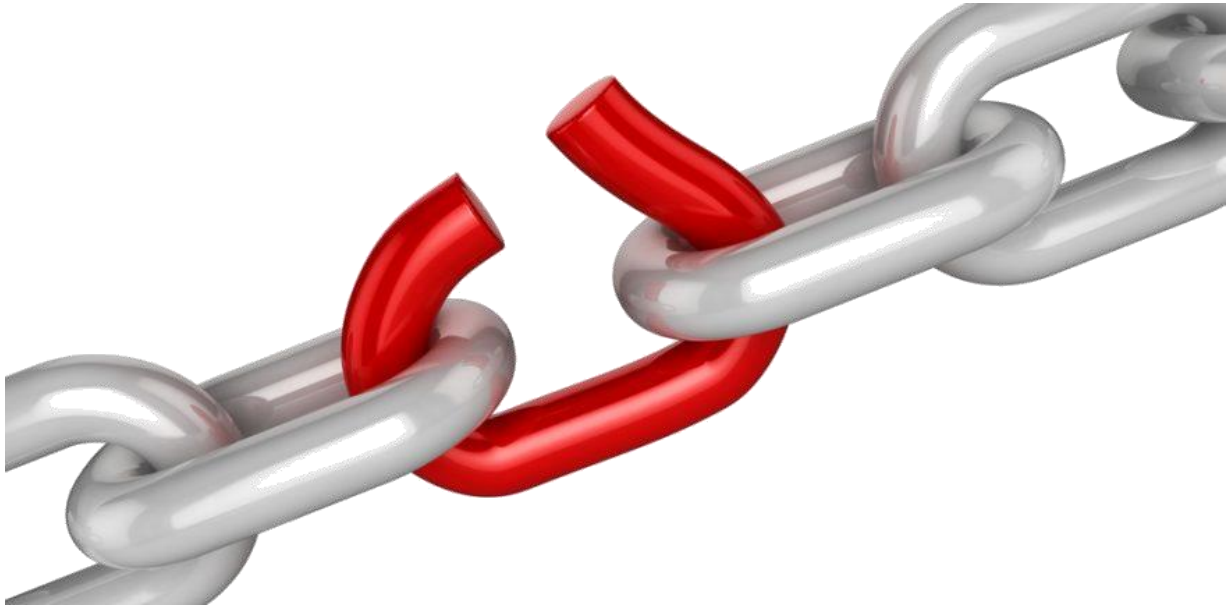
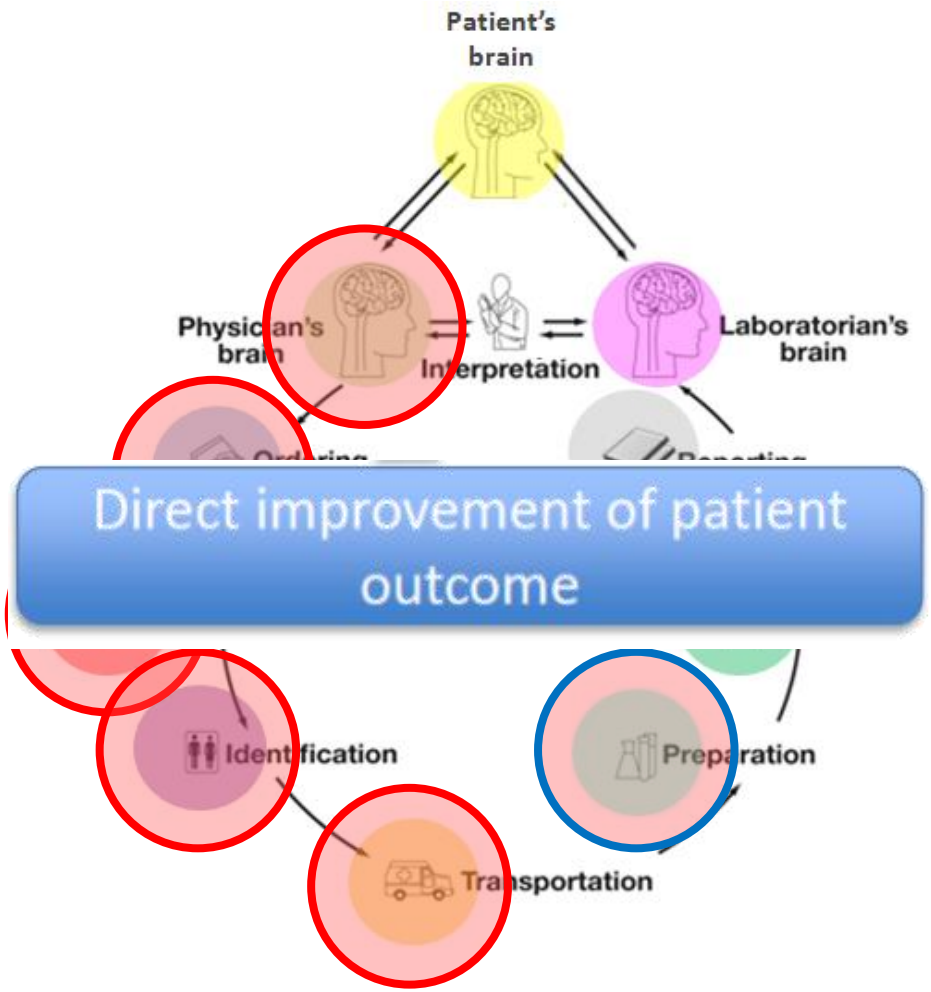
WEQAS Annual Conference 2022
Session: Right patients, right results, right time
Wed 7th December 2022

Talk overview



- TTP and the PP
- Consequences of PAE/PAI
- Types of PAE/PAI
- Monitoring and Reduction of PAE/PAI
- Regional and International initiatives
- PAI cases throughout

TTP and the PP (1)



- aka Brain to brain loop
- 3 Phases
  - Preanalytical Phase
    - Pre-pre-
    - Pre-
  - Analytical Phase
  - Postanalytical Phase
- Errors/issues all phases

Plebani M, Laposata M, Lundberg G. The Brain-to-Brain Loop Concept for Laboratory Testing 4Years After Its Introduction. Am J Clin Pathol 2011;136:829-833

TTP and the PP (2)

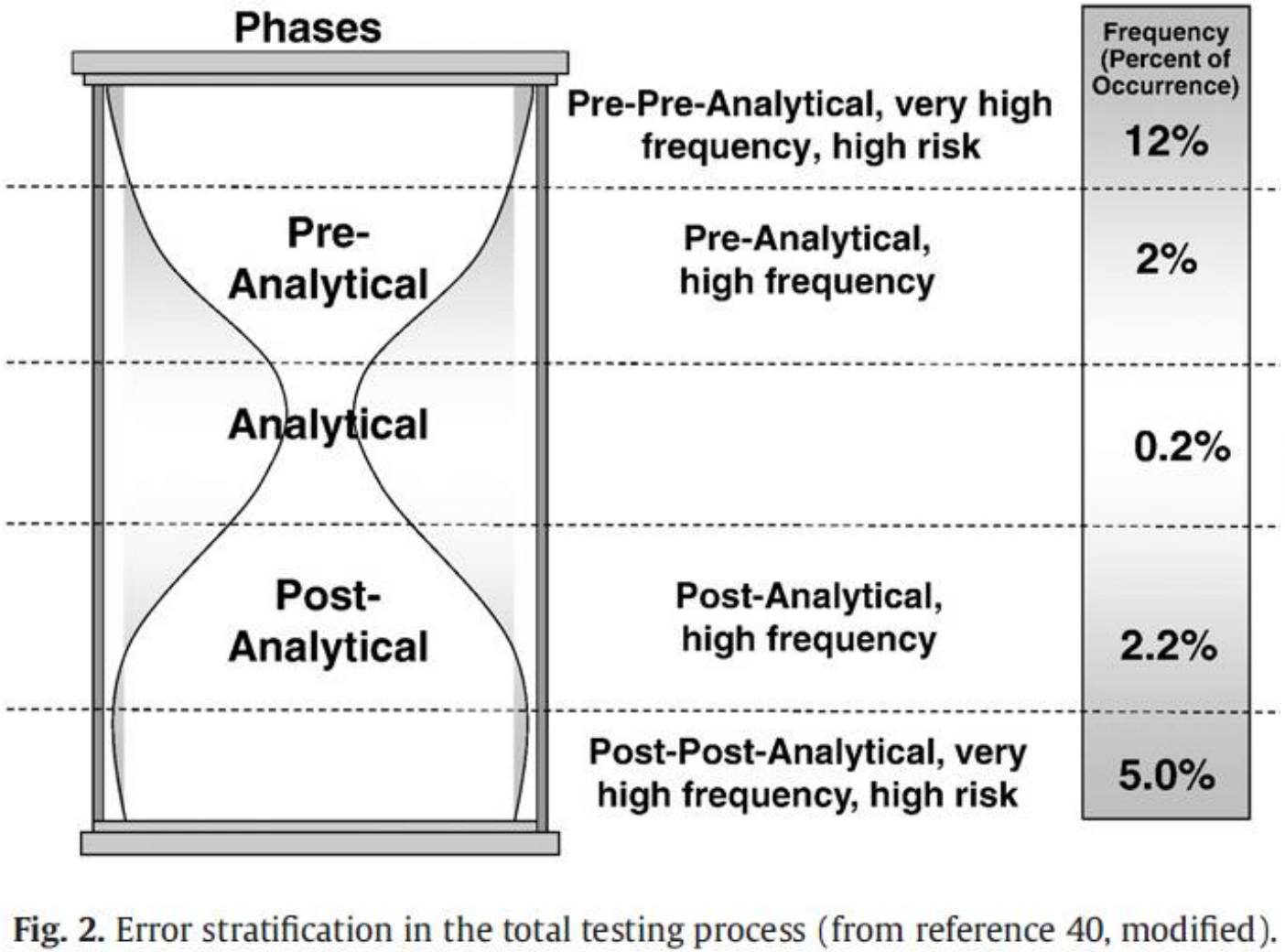
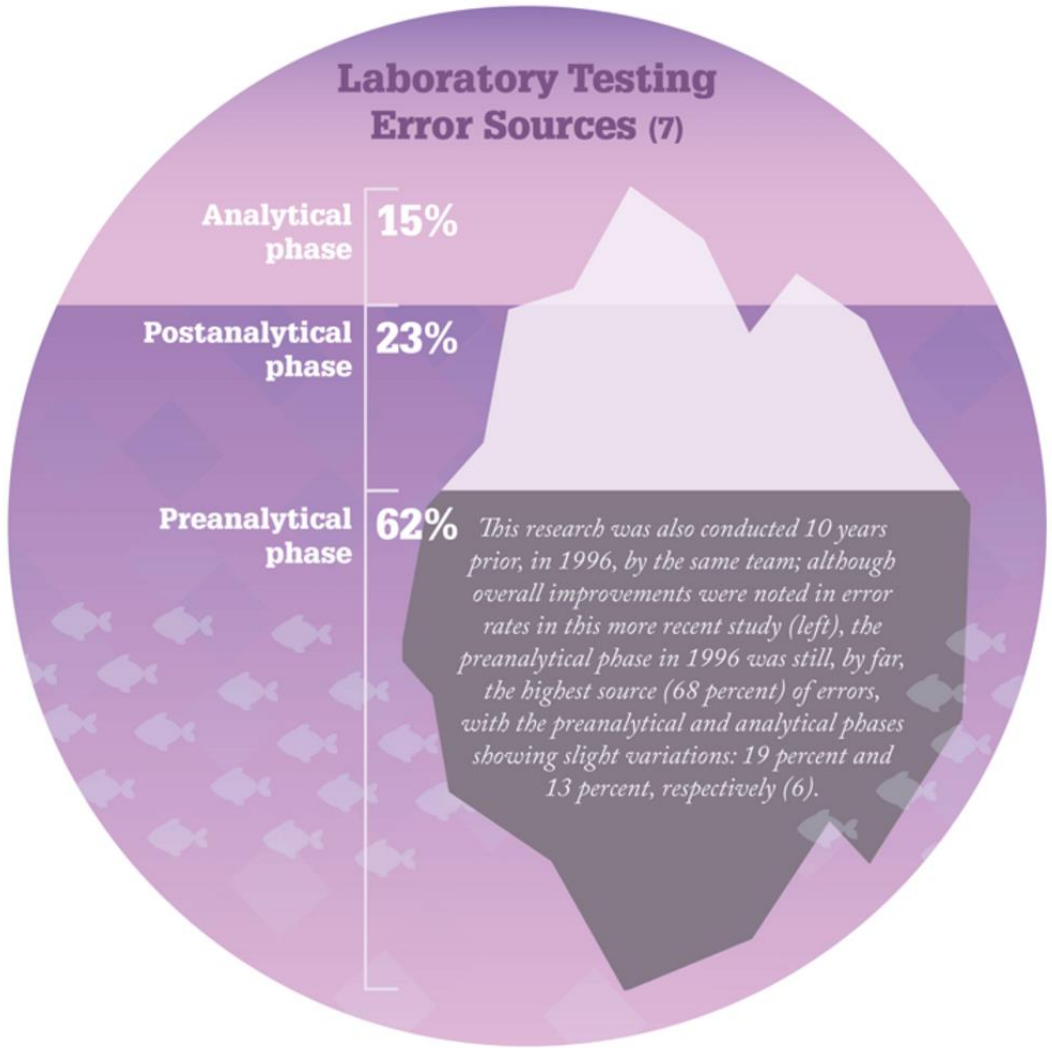


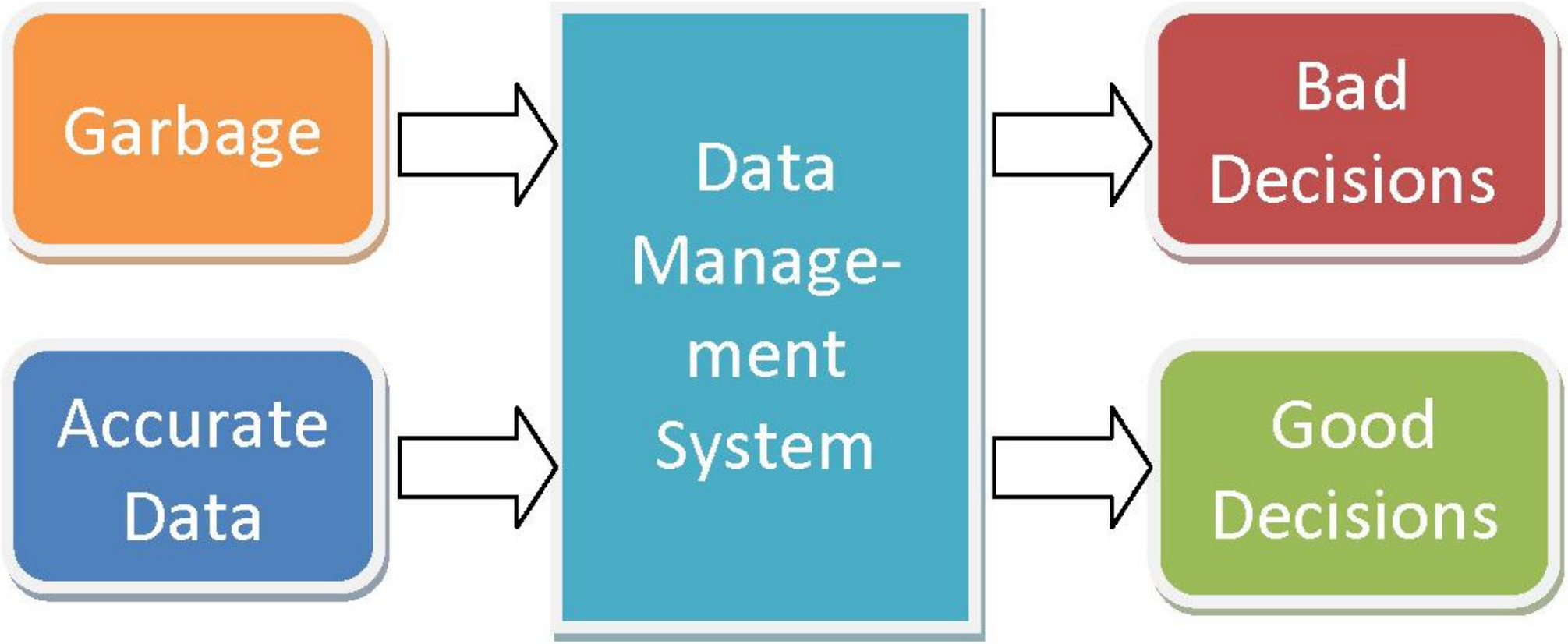
Fig. 2. Error stratification in the total testing process (from reference 40, modified).



Plebani M. Exploring the iceberg of errors in laboratory medicine. Clin Chimica Acta (2009) 16-23

TTP and the PP (3)

Garbage in, garbage out !



Consequences of PAE/PAI (1)

- Incorrect test results (wrong value/patient)
- Incorrect diagnosis (wrong patient/interpretation)
- Unnecessary delays (TTL/results withheld/resample)
- Harm to the patient
- Wasted laboratory time and money
- Wasted hospital beds and staff time

Consequences of PAE/PAI (2)

- Specimen rejection related harm
  - Repeated phlebotomy required in 86.8% of rejected blood specimens
  - Rejected urine specimens required recatheterisation in 13.8% of cases
  - Inconvenience and discomfort for the patient
  - Potential for patient complications
  - Median specimen processing delay was 65 minutes
  - Potential failure to provide adequate care in a timely manner

Karcher DS, et al. Clinical Consequences of Specimen Rejection: A College of American Pathologists Q-Probes Analysis of 78 Clinical Laboratories. Arch Pathol Lab Med. 2014;138:1003-8.

Consequences of PAE/PAI (3)

- Reducing Costs
  - A study was performed in a London teaching hospital
  - Estimated cost of repeating haemolysed specimens, based on an average of 60 admissions per day, was **£4355 per month**, plus additional time and equipment costs.
  - This cost-saving would fund at least one dedicated Emergency Department phlebotomist.

P Jacobs, J Costello, M Beckles. Cost of haemolysis. Ann Clin Biochem. 2012;49(Pt 4):412.

Consequences of PAE/PAI (4)

Barcode read errors Plymouth Hospitals NHS Trust

Consequences of PAE/PAI (5)

Pre-analytical error	Possible consequence	Degree of seriousness
Patient identification	Sample collected from wrong patient	**Mild to Life threatening**
Tube labelling	Wrong patient's blood in tube	**Mild to Life threatening**
Test request management	Incomplete or erroneous test	**Mild to Severe**
Patient rest	↑ or ↓ conc. of analytes	**Mild to Moderate**
Blood tube inversion	No mixing of blood with additive	**None to Moderate**
Vertical tube storage	Incorrect coagulation of serum samples	**None to Mild**

Naz et al. Pak J Med Res. 2012;51:27-30

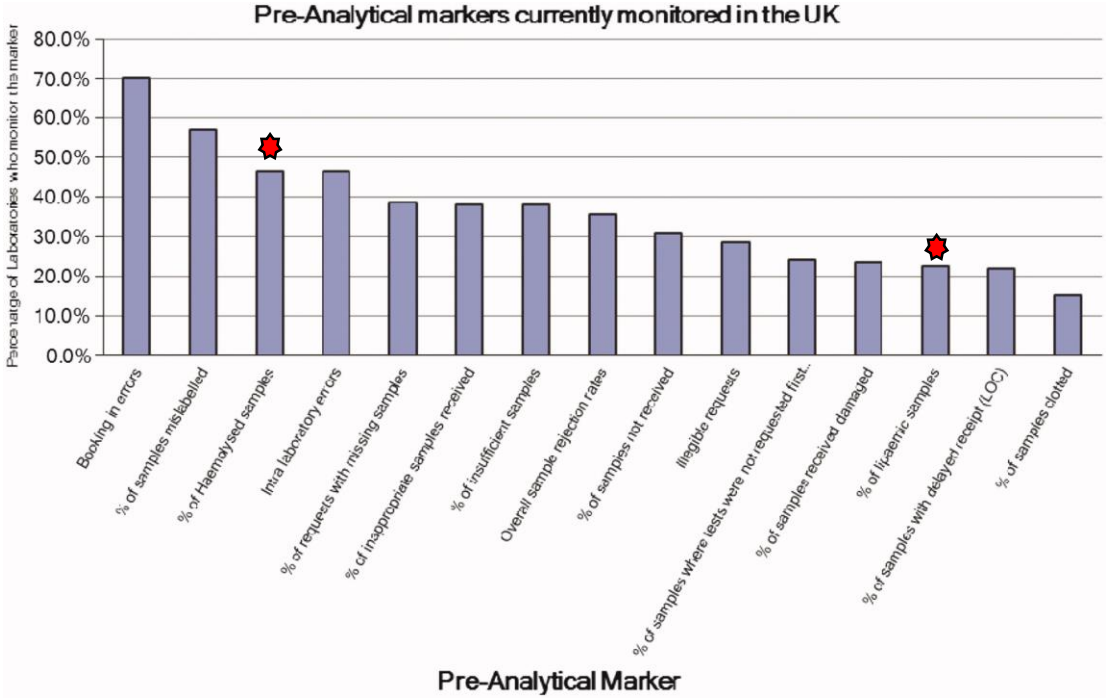
Types of PAE/PAI (1)

- Dozens of steps
- Each can be subdivided
- Can have error at each step
- Each step can be a focus of error reduction

Burrows, J. The Cost of Pre-Analytical Errors in the Context of Inpatient Complete Blood Count Testing at Sunnybrook Health Sciences Centre

Types of PAE/PAI (2)

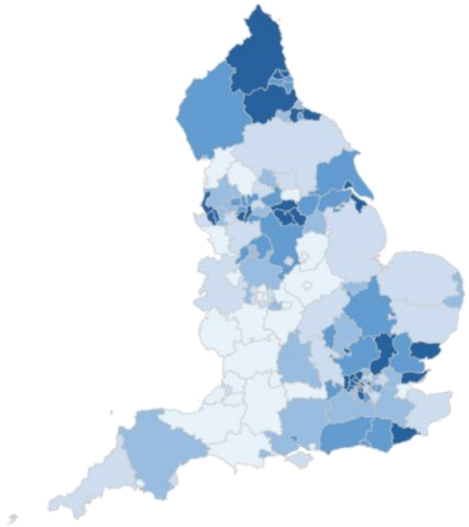
- We will cover
  - Inappropriate test request
  - Patient not appropriately prepared
  - Lack of clinical details with requests
  - Order of draw
  - Haemolysis, Icterus, Lipaemia (HIL)
  - Specimen inappropriately stored

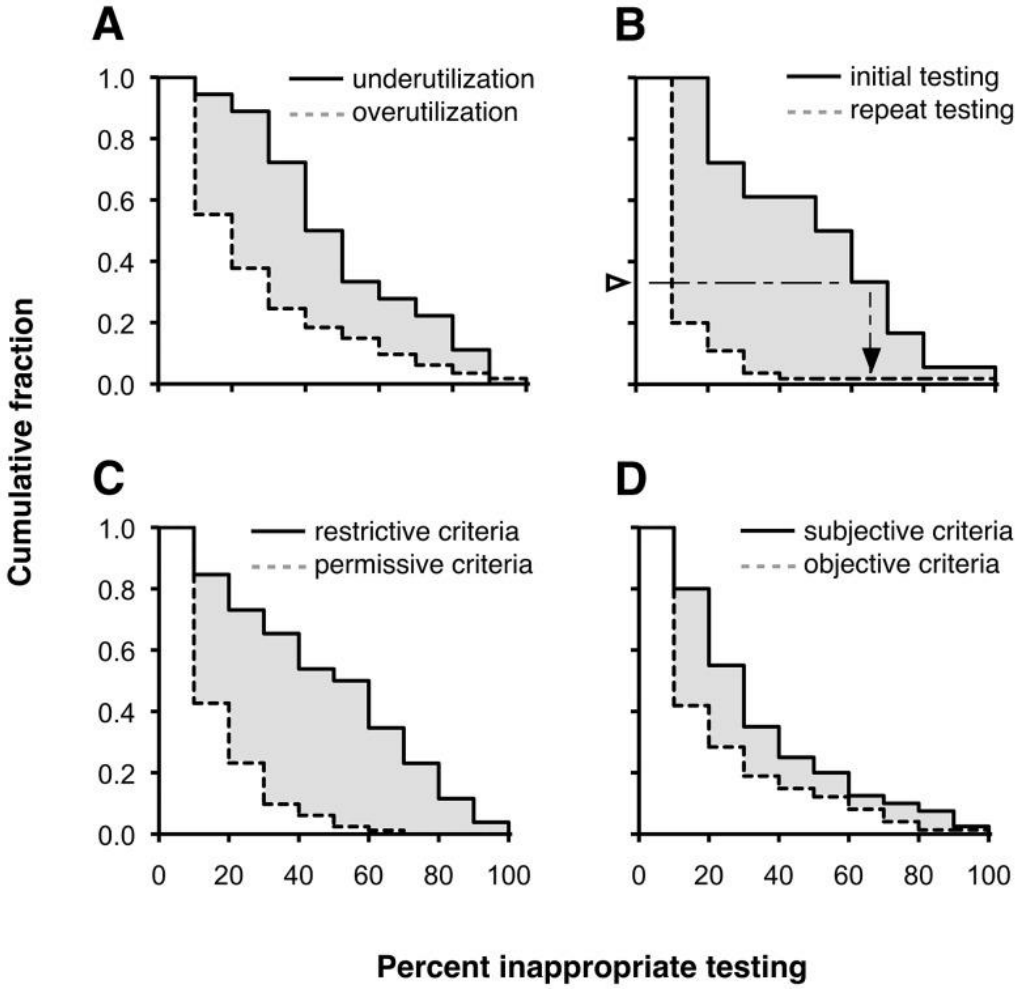


Cornes MP, Atherton J, Pourmahram G, Borthwick H, Kyle B, West J, Costelloe SJ. Monitoring and reporting of preanalytical errors in laboratory medicine: the UK situation Ann Clin Chem epub

Inappropriate test requesting (1)

- Test overutilisation
  - Major problem
  - International issue
- Driven and Enabled by:
  - Lab. automation
  - Poor test panel design
    - “Standard” bloods
  - Electronic requesting
  - Labs failing to ensure appropriate testing
  - High frequency testing is de rigueur in wealthier countries
  - Barrier to getting tested is very low
  - “Wellness” bloods
  - Commercial incentives – private labs – more tests, more profit





PLoS One. 2013 Nov 15;8(11):e78962

Inappropriate test requesting (2)

- **“Rules” for lab test utilisation (Baird)**
  - Rule 1: “If you ask a stupid question, you get a stupid answer”
  - Rule 2: “Laboratory testing is for sick people”
  - Rule 3: “Too many good tests are the same as one bad test”
  - $1 - 0.95^{n}$
    - $1-0.95^{21}$ = ~66%

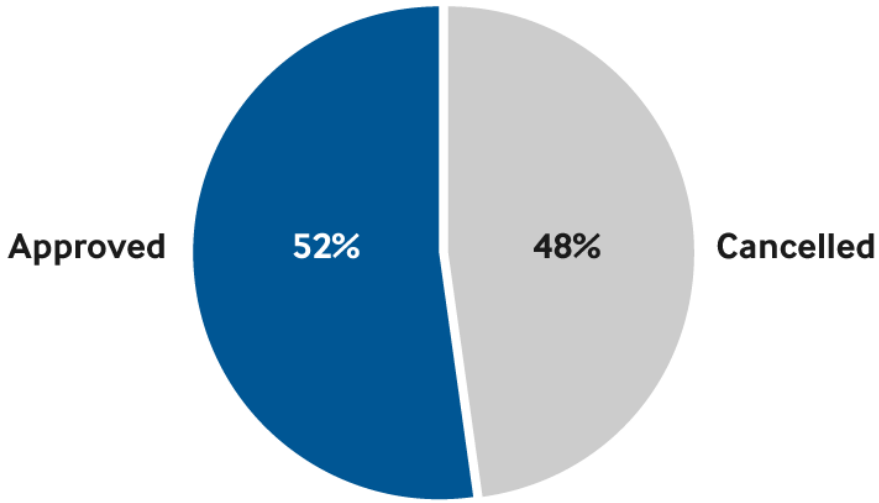
Baird G. Biochem Med (Zagreb) 2014;24(2):223-34

How can laboratories help to improve?

- Vet tests
- Evidence based panels
  - By organ
  - By presentation
  - By payment
- Formulary
- Minimum repeat intervals
- Lab tests online
- Choose Wisely
- Audit under/over utilisation

Outcome of Orders for Restricted Inpatient Tests in 2016

A total of 1,031 orders for tests that were restricted for inpatients were placed in 2016; 48% of these orders were cancelled (gray) and 52% were approved by a laboratory medical director (blue).



Source: Authors

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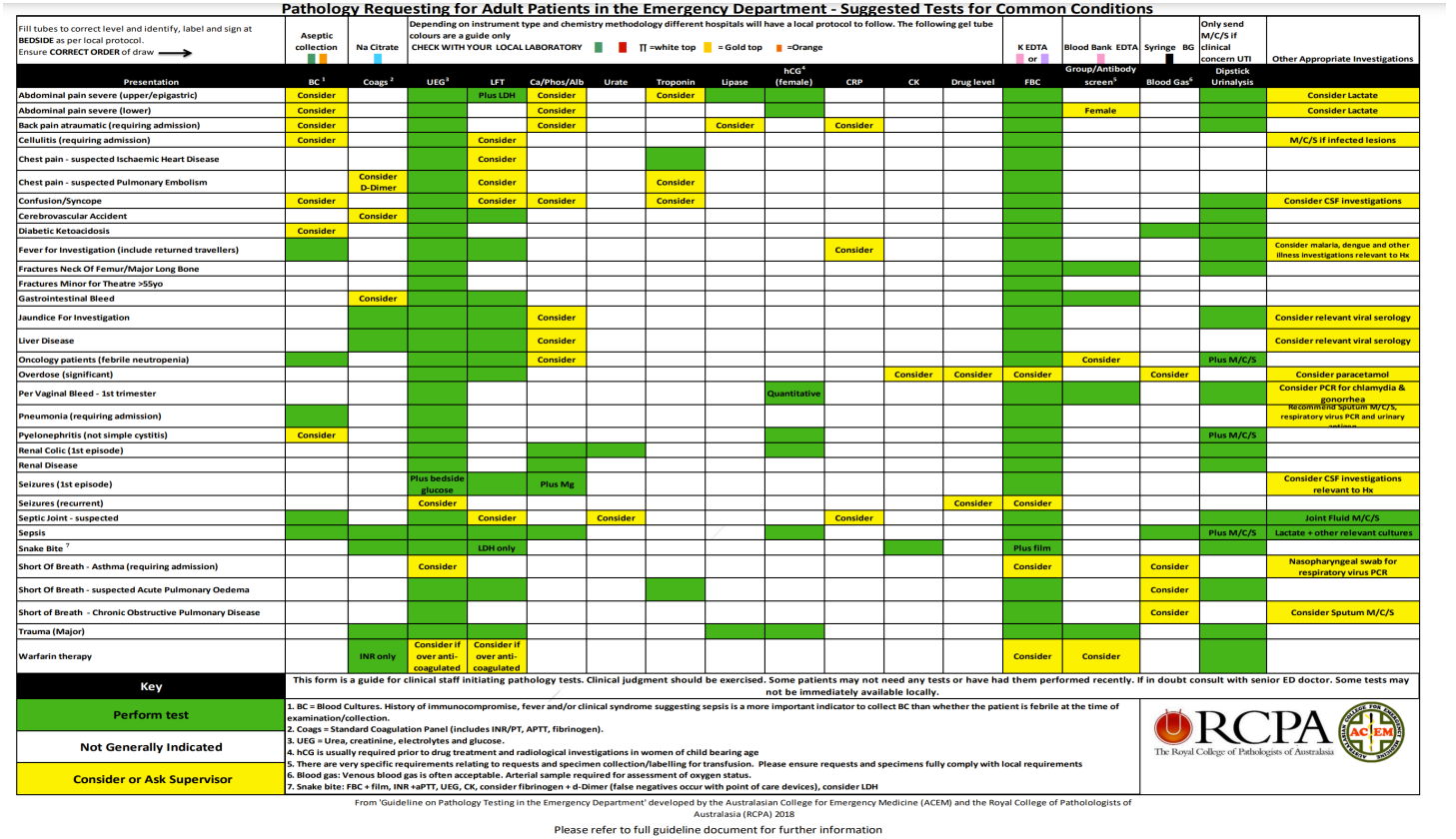

Pathology Requesting for Adult Patients in the Emergency Department - Suggested Tests for Common Conditions

Fill tubes to correct level and identify, label and sign at BEDSIDE as per local protocol. Ensure CORRECT ORDER of draw →

Depending on instrument type and chemistry methodology different hospitals will have a local protocol to follow. The following gel tube colours are a guide only. CHECK WITH YOUR LOCAL LABORATORY ▮ ▮ ▮ =white top ▮ = Gold top ▮ =Orange

Presentation	Aseptic collection: BC [1]	Na Citrate: Coags [2]	UEG [3]	LFT	Ca/Phos/Alb	Urate	Troponin	Lipase	hCG [4] (female)	CRP	CK	Drug level	K EDTA or FBC	Blood Bank EDTA: Group/Antibody screen [5]	Syringe BG: Blood Gas [6]	Only send M/C/S if clinical concern UTI: Dipstick Urinalysis	Other Appropriate Investigations
Abdominal pain severe (upper/epigastric)	Consider			Plus LDH	Consider		Consider										Consider Lactate
Abdominal pain severe (lower)	Consider				Consider									Female			Consider Lactate
Back pain atraumatic (requiring admission)	Consider				Consider			Consider		Consider							
Cellulitis (requiring admission)	Consider			Consider													M/C/S if infected lesions
Chest pain - suspected Ischaemic Heart Disease				Consider													
Chest pain - suspected Pulmonary Embolism		Consider D-Dimer		Consider			Consider										
Confusion/Syncope	Consider			Consider	Consider		Consider										Consider CSF investigations
Cerebrovascular Accident		Consider															
Diabetic Ketoacidosis	Consider																
Fever for Investigation (include returned travellers)										Consider							Consider malaria, dengue and other illness investigations relevant to Hx
Fractures Neck Of Femur/Major Long Bone																	
Fractures Minor for Theatre >55yo																	
Gastrointestinal Bleed		Consider															
Jaundice For Investigation					Consider												Consider relevant viral serology
Liver Disease					Consider												Consider relevant viral serology
Oncology patients (febrile neutropenia)					Consider									Consider		Plus M/C/S	
Overdose (significant)											Consider	Consider	Consider		Consider		Consider paracetamol
Per Vaginal Bleed - 1st trimester									Quantitative								Consider PCR for chlamydia & gonorrhea
Pneumonia (requiring admission)																	Recommend Sputum M/C/S, respiratory virus PCR and urinary antigen
Pyelonephritis (not simple cystitis)	Consider															Plus M/C/S	
Renal Colic (1st episode)																	
Renal Disease																	
Seizures (1st episode)			Plus bedside glucose		Plus Mg												Consider CSF investigations relevant to Hx
Seizures (recurrent)			Consider														
Septic Joint - suspected				Consider		Consider				Consider							Joint Fluid M/C/S
Sepsis																Plus M/C/S	Lactate + other relevant cultures
Snake Bite [7]				LDH only									Plus film				
Short Of Breath - Asthma (requiring admission)			Consider										Consider		Consider		Nasopharyngeal swab for respiratory virus PCR
Short Of Breath - suspected Acute Pulmonary Oedema															Consider		
Short of Breath - Chronic Obstructive Pulmonary Disease															Consider		Consider Sputum M/C/S
Trauma (Major)																	
Warfarin therapy		INR only	Consider if over anti-coagulated	Consider if over anti-coagulated									Consider	Consider			

Key

- Perform test
- Not Generally Indicated
- Consider or Ask Supervisor

This form is a guide for clinical staff initiating pathology tests. Clinical judgment should be exercised. Some patients may not need any tests or have had them performed recently. If in doubt consult with senior ED doctor. Some tests may not be immediately available locally.

1. BC = Blood Cultures. History of immunocompromise, fever and/or clinical syndrome suggesting sepsis is a more important indicator to collect BC than whether the patient is febrile at the time of examination/collection.
2. Coags = Standard Coagulation Panel (includes INR/PT, APTT, fibrinogen).
3. UEG = Urea, creatinine, electrolytes and glucose.
4. hCG is usually required prior to drug treatment and radiological investigations in women of child bearing age
5. There are very specific requirements relating to requests and specimen collection/labelling for transfusion. Please ensure requests and specimens fully comply with local requirements
6. Blood gas: Venous blood gas is often acceptable. Arterial sample required for assessment of oxygen status.
7. Snake bite: FBC + film, INR +aPTT, UEG, CK, consider fibrinogen + d-Dimer (false negatives occur with point of care devices), consider LDH

RCPA The Royal College of Pathologists of Australasia

From 'Guideline on Pathology Testing in the Emergency Department' developed by the Australasian College for Emergency Medicine (ACEM) and the Royal College of Pathologists of Australasia (RCPA) 2018

Please refer to full guideline document for further information

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How can laboratories help to improve?

- Vet tests
- Evidence based panels
  - By organ
  - By presentation
  - By payment
- Prompts @ ordering
- Formulary
- Ban tests
- Add tests (reflex/reflect)
- Minimum repeat intervals
- Lab tests online
- Choose Wisely
- Audit under/over utilisation

Strength	Tool	Target	Strengths	Weaknesses	Example/ References
Strong	**Ban the test**	Obsolete tests, "Quack" testing, Legitimate tests used in inappropriate circumstances	This is the "Nuclear Option", as it ensures a complete cease to ordering	Only useful for tests with broad consensus as to lack of utility, which is unusual. Specific individuals may destroy consensus.	Bleeding time and other "Antiquated" tests (42).
Strong	**Laboratory test formulary**	All tests, especially those with utilization that is recognized, after analytics, to be above what is expected or justifiable.	A uniform policy across a system can be supported by a formulary, in the same way as a pharmacy formulary. Exceptions to formulary can be vetted by a committee or individual tasked with these decisions.	Requires authority and buy-in from multiple factions in a medical system, and likely participation by multiple specialties.	University of Michigan (43).

Biochemia Medica 2014;24(2):223–34

Patient preparation (1)

- Fasting
  - How long?
    - 9-10 hrs fast affects TG, insulin, C-peptide, glucose, Hcy
    - >14 hrs – altered gluconeogenesis – increased TG – AVOID
    - EFLM 12 hrs ± 0.5 hrs
  - Water
    - Can affect results
    - EFLM – patient drinks water as they normally would during fast
  - Poorly standardized – poor evidence base
  - Poorly understood by clinicians/patients
- Caffeine and cigarettes – avoid on morning of sampling
- Alcohol – abstain for 24 hrs

Clinical Chemistry

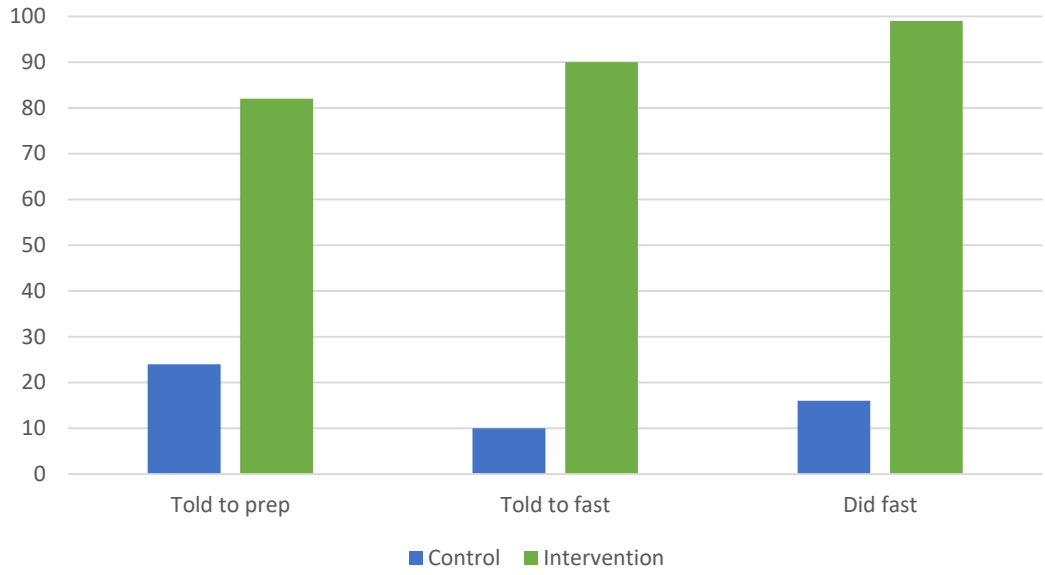
Table 1. Evaluation of articles published in relevant journals in 2002.

Journal	Articles with a group of fasting patients,[a] n	Well-defined fasting, n (%)	Insufficient definition, n (%)	No definition, n (%)
Clinical Chemistry	20	1 (5)	5 (25)	14 (70)
Clinical Chemistry and Laboratory Medicine	24	0 (0)	6 (25)	18 (75)
Scandinavian Journal of Clinical and Laboratory Investigation	18	3 (17)	4 (22)	11 (61)
Diabetes	94	7 (7)	36 (38)	51 (54)

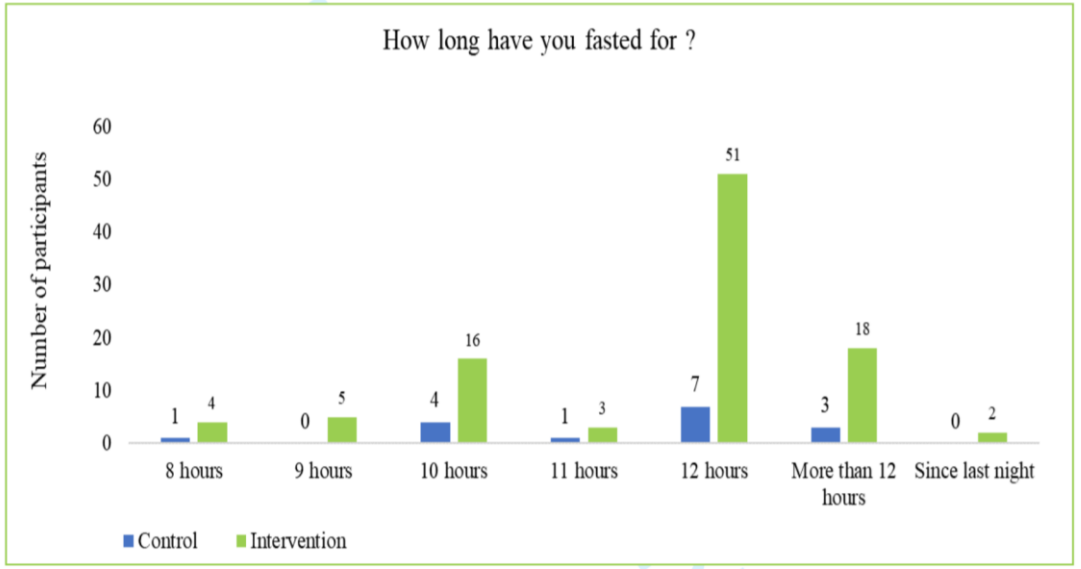
[a] If the term "fasting patient" was used in the Materials and Methods, Results, or Discussion, the publication was considered as using fasting patients.

Patient preparation (2)

- Avoidance of certain foods
  - 5HIAA
    - Banana, Pineapple, Tomato, Plum, Eggplant, Avoca, Kiwi, walnuts
- Physical activity
  - Plasma volume
- Medication
  - Time relative to dose
  - Dose
  - Compliance
- If patient has not prepared
  - Should cancel phlebotomy
    - Risk RIs being incorrect
    - Diagnoses may prove incorrect
    - Patient safety is compromised
    - Rarely in a position to enforce this



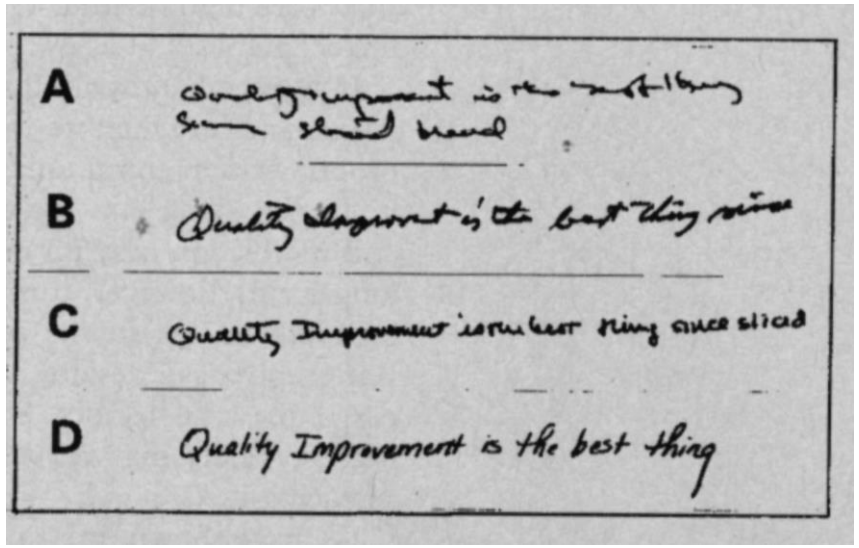
Andrade N., In Press 2022



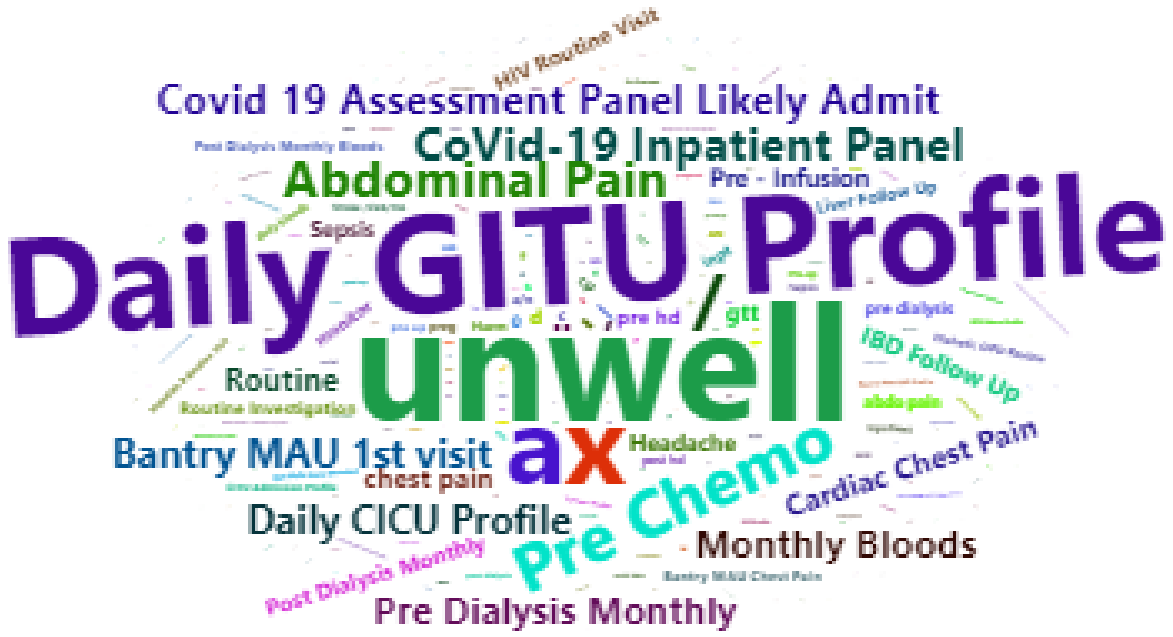
Andrade N., In Press 2022

Clinical details

- Why important?
  - Can define urgency
  - Justify expensive testing
  - Highlight potential interferences
  - Aid interpretation of results
- How can lab.s address?
  - Zero tolerance for certain tests
  - Clinical details associated with profiles
  - User education



BMJ Volume 313, Number 7072



Bransfield, A., In Press 2022

- CUH 2021 – all iCM requests
- 219,434 unique requests (≥1 specimen/request)
- 12,240 unique RFR entries.
- Relevant clinical details in 45,079 (20.5%)
- 65,713 (29.9%) was blank
- 82,585 (37.6%) contained apparently random combinations of letters/numbers/punctuation symbols.
- 26057 (11.9%) non-specific (e.g. ‘Routine’, ‘unwell’, ‘follow-up’).
- ~80% of electronic requests have no informative clinical details, despite RFR being a mandatory

Order of draw (1)

Specimen Volume	Order Of Draw	Closure Colour	Tube Contents	Assays
3ml		Blue	Trisodium Citrate solution	Coagulation Studies
4ml		Red	Separation Gel Clotting Accelerator	Biochemistry Profiles, Viral Studies, Hormone Studies, Immunology, Anti Cardiolipin AB., B12, Folate, Ferritin, RA, Intrinsic Factor AB, Iron Studies, CRP's, TDM (Therapeutic Drug Monitoring), Copper and Zinc levels.
4ml		Red	Clotted (Gel free)	Cryoglobulins, Methotrexate
4ml		Green	Heparin	Chromosomes, Lead Levels, DNA Analysis
3ml		Purple	EDTA	FBC, HBA1C, Hb. Electrophoresis, Maleria Parasites, Sickle Cell, Reticulocyte Count, Coombs Test, Cyclosporin,Tacrolimus ESR, Immunophenotyping, PTH, Cryogobulins
6ml		Pink	EDTA	Crossmatch, Group & Antibody Screen
4ml		Grey	EDTA sodium fluroide	Glucose, Glucose Tolerance,Lactate, Alcohol Levels
9ml		Yellow	ACD-A	HLA Typing

Order of blood draw: Opinion Paper by the European Federation for Clinical Chemistry and Laboratory Medicine (EFLM) Working Group for the Preanalytical Phase (WG-PRE)

Clin Chem Lab Med 2017 Jan 1;55(1):27-31.

Joint EFLM-COLABIOCLI Recommendation for venous blood sampling

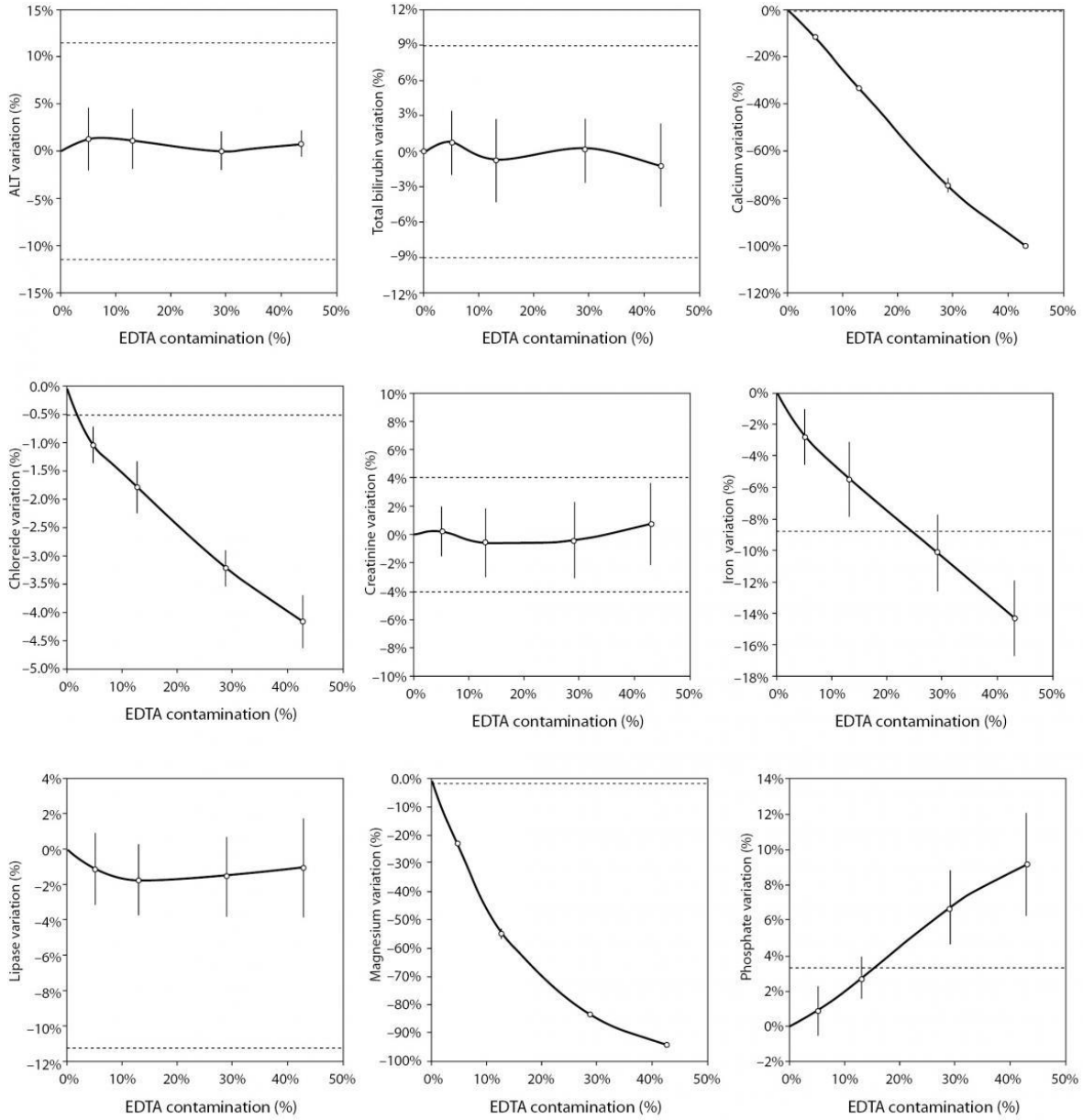
Clin Chem Lab Med 2018 Nov 27;56(12):2015-2038.

The recommended order of draw is as follows:

1. Blood culture tube
2. Citrate tube
3. Plain tube or tube with clot activator
4. Heparin tube
5. EDTA tube
6. Glycolysis inhibitor tube
7. Other tubes

Order of draw (2)

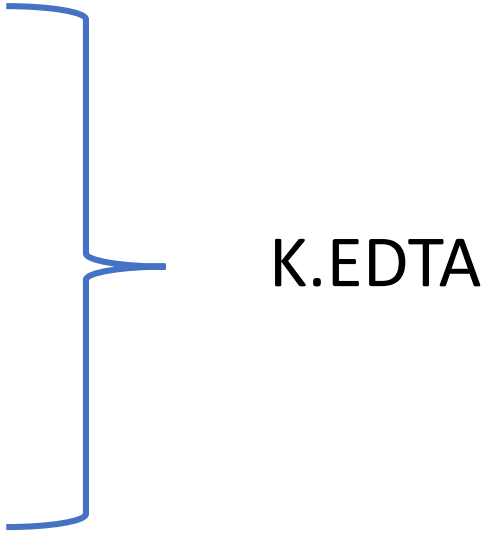
- Important because
  - Tubes contain different additives
    - Procoagulants
      - Serum (clot) tubes
    - Anticoagulants
      - **EDTA**, potassium salt of
      - Flouride **oxalate**
      - Lithium **heparin**
      - Sodium **citrate**
    - Glycolysis inhibitors
      - **Flouride** oxalate
    - Gel/plastic separators



Lima-Oliveira G, Salvagno GL, Danese E, Brocco G, Guidi GC, Lippi G. Contamination of lithium heparin blood by K2-ethylenediaminetetraacetic acid (EDTA): an experimental evaluation. Biochem Med (Zagreb). 2014;24:359-367

Order of draw (3)

- Problems Associated with incorrect Order of Draw
  - Hypernatremia -> Sodium Citrate / Na.EDTA
  - Hyperkalaemia
  - Hypocalcaemia
  - Hypomagnesaemia
  - Low Zinc
  - Low Iron
  - Low ALP



- Poor coagulation -> transfer of anticoagulants
- Dilution effects -> tipping of samples

Order of draw – simple case (1)

28 year old female admitted to A&E with hyperemesis gravidarum at 24/40wks gestation. Bloods taken for haematology & biochemistry

Analyte	Initial Results	Reference Interval
Sodium (mmol/L)	134	136-145
Potassium (mmol/L)	**6.2**	3.5-5.0
Urea (mmol/L)	6.0	2.5-7.5
Creatinine (µmol/L)	70	60-104
Calcium (mmol/L)	**1.50**	2.20-2.60
Albumin (g/L)	39	35-50
Adj. Calcium (mmol/L)	**1.52**	2.20-2.60
ALP (IU/L)	**12**	60-300

Order of draw – simple case (2)

Repeat bloods taken to check low calcium and high potassium

K^+EDTA will cause spurious results.

Analyte	Initial Results	Repeat Results	Reference Interval
Sodium (mmol/L)	134	132	136-145
Potassium (mmol/L)	**6.2**	3.4	3.5-5.0
Urea (mmol/L)	6.0	6.0	2.5-7.5
Creatinine (µmol/L)	70	68	60-104
Calcium (mmol/L)	**1.50**	2.30	2.20-2.60
Albumin (g/L)	39	38	35-50
Adj. Calcium (mmol/L)	**1.52**	2.34	2.20-2.60
ALP (IU/L)	**12**	329	60-300

OOD – is it all a myth? (1)

Ann Clin Biochem. 2008 Nov;45(Pt 6):601-3. doi: 10.1258/acb.2008.007241. Epub 2008 Sep 3.

Spurious hyperkalaemia due to EDTA contamination: common and not always easy to identify

Michael P Cornes [1], Clare Ford, Rousseau Gama

Background: To study the detection and prevalence of spurious hyperkalaemia due to potassium ethylenediaminetetra-acetic acid (kEDTA) contamination

Methods: In a one-month prospective study, serum potassium, zinc, calcium, magnesium concentrations and alkaline phosphatase activity were measured in samples with serum potassium >or=6.0 mmol/L.

Results: Twenty-eight out of 117 hyperkalaemic samples were contaminated with EDTA. Only serum zinc values below the reference range had 100% sensitivity for indicating EDTA contamination, but even at an optimal specificity of 92% at least 12 potentially genuine hyperkalaemic samples would be rejected.

Conclusion: Spurious hyperkalaemia due to kEDTA contamination is common. Gross kEDTA contamination is obvious by marked unexpected hyperkalaemia, hypocalcaemia, hypomagnesaemia and hypozincaemia. Spurious hyperkalaemia due to low concentrations of kEDTA contamination can only be confidently detected by measurement of serum EDTA.

Common ?

Incorrect order of draw of blood samples does not cause potassium EDTA sample contamination

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*Department of Clinical Chemistry, New Cross Hospital; and †Research Institute for Healthcare Sciences, University of Wolverhampton, West Midlands, UK

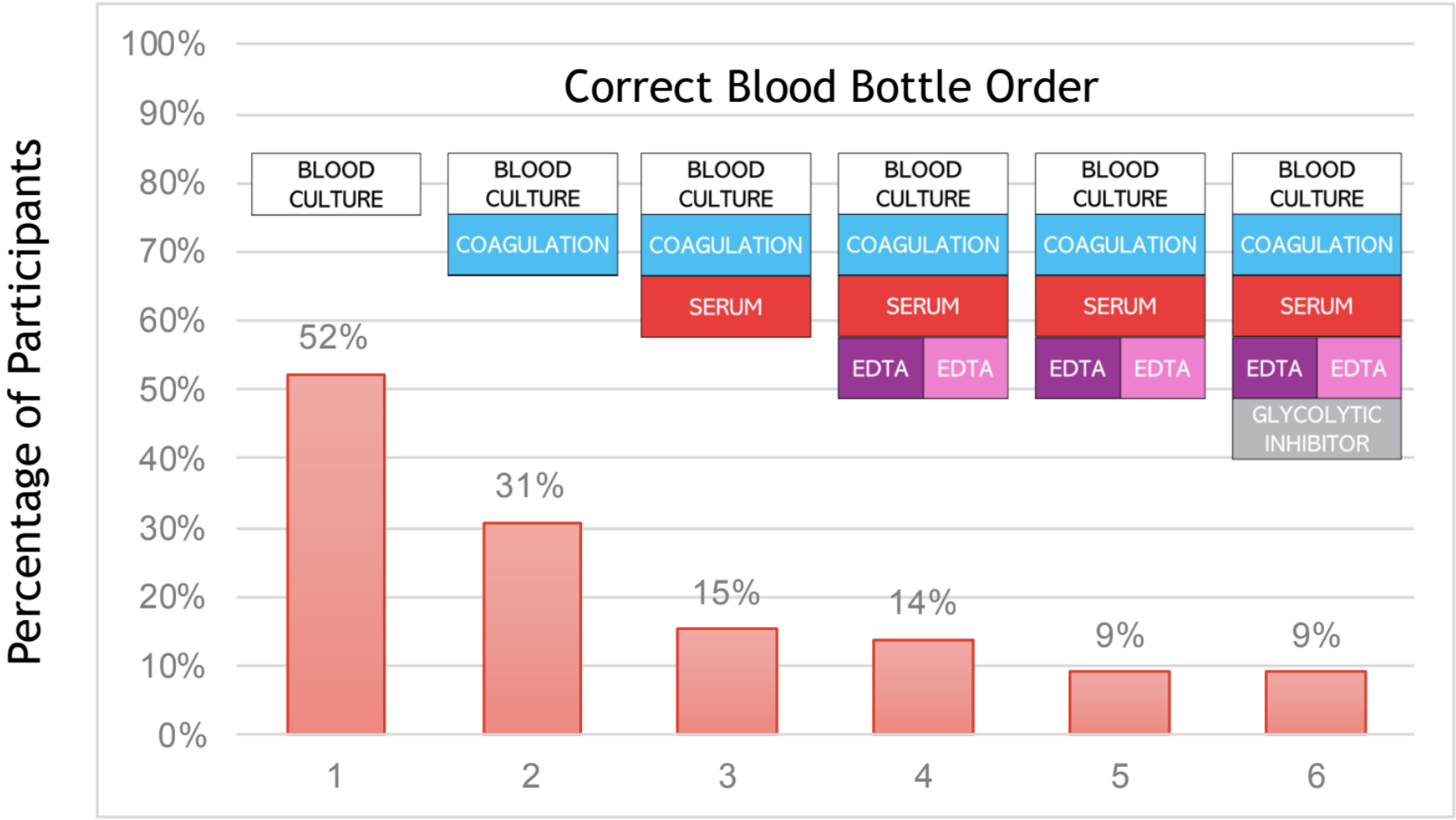
Table 1. Serum analyte concentrations in blood samples collected from 11 subjects before and after collection of the EDTA blood sample.

Analyte	Before EDTA	After EDTA	*P* value
EDTA (mmol/L)	<0.2	<0.2	1
Potassium (mmol/L)	4.2 (0.22)	4.2 (0.29)	0.571
Adjusted calcium (mmol/L)	2.37 (0.0[illegible])	2.39 (0.015)	0.372
Magnesium (mmol/L)	0.8[illegible]	0.83 (0.047)	0.800
Zinc (µmol/L)	1[illegible]	17.4 (6.6)	0.843
Alkaline phosphatase (IU/L)	64.2 (21.8)	65.7 (22.5)	0.872
Creatinine (µmol/L)	79 (11.0)	79 (11.2)	0.955

Results expressed as mean (SD)

OOD not to blame ?

OOD – is it all a myth? (2)



OOD – is it all a myth? (3)

Group No.	First Tube (Control Serum)	Second Tube	Third Tube	Fourth Tube	Fifth Tube
1	 Serum	 EDTA	 Serum		
2	 Serum	 EDTA	 Citrate	 Serum	
3	 Serum	 EDTA	 Sodium Fluoride	 Serum	
4	 Serum	 EDTA	 Sodium Fluoride	 Citrate	 Serum
5	 Serum	 EDTA	 Citrate	 Sodium Fluoride	 Serum

- Trained phlebotomists
- 5 volunteers
- Closed Greiner Vacuette system
- No statically significant differences found between tubes 1 and 5 for:
  - Calcium
  - Magnesium
  - Potassium
  - ALP
  - Iron

OOD – is it all a myth? (4)

- We all see gross EDTA contamination
- Low grade EDTA contamination – unlikely to be “tipping off”
- EDTA contamination due to incorrect OOD not shown experimentally
  - Studies had trained phlebotomist
  - Controlled conditions
  - Same situation if nurse/doctor taking bloods in ED at 2AM?

What are the mechanisms of EDTA contamination.

- 3 possible mechanisms
  1) Direct transfer
     - Easily identified
  2) Backflow due to incorrect order of draw
     - Appears not to be the case under ideal phlebotomy conditions
  3) Syringe needle contamination
     - Best current hypothesis when combined with incorrect order of draw

OOD – is it all a myth? (5)

What is the source of contamination? - Hypothesis

- Hypothesis: Is it is due to syringe transfer?

Table 1 Variation in phlebotomy technique practised in the Majors area of the Emergency Medicine Department

Technique	Number
Cannula with syringe	19 (38%)
Cannula with evacuated tubes and adaptor	21 (42%)
Syringe and needle into vein	7 (14%)
Evacuated tubes system conventionally used	3 (6%)

52% of samples taken with a syringe

Table 2 How blood tubes are filled when they are not the primary receiver of samples

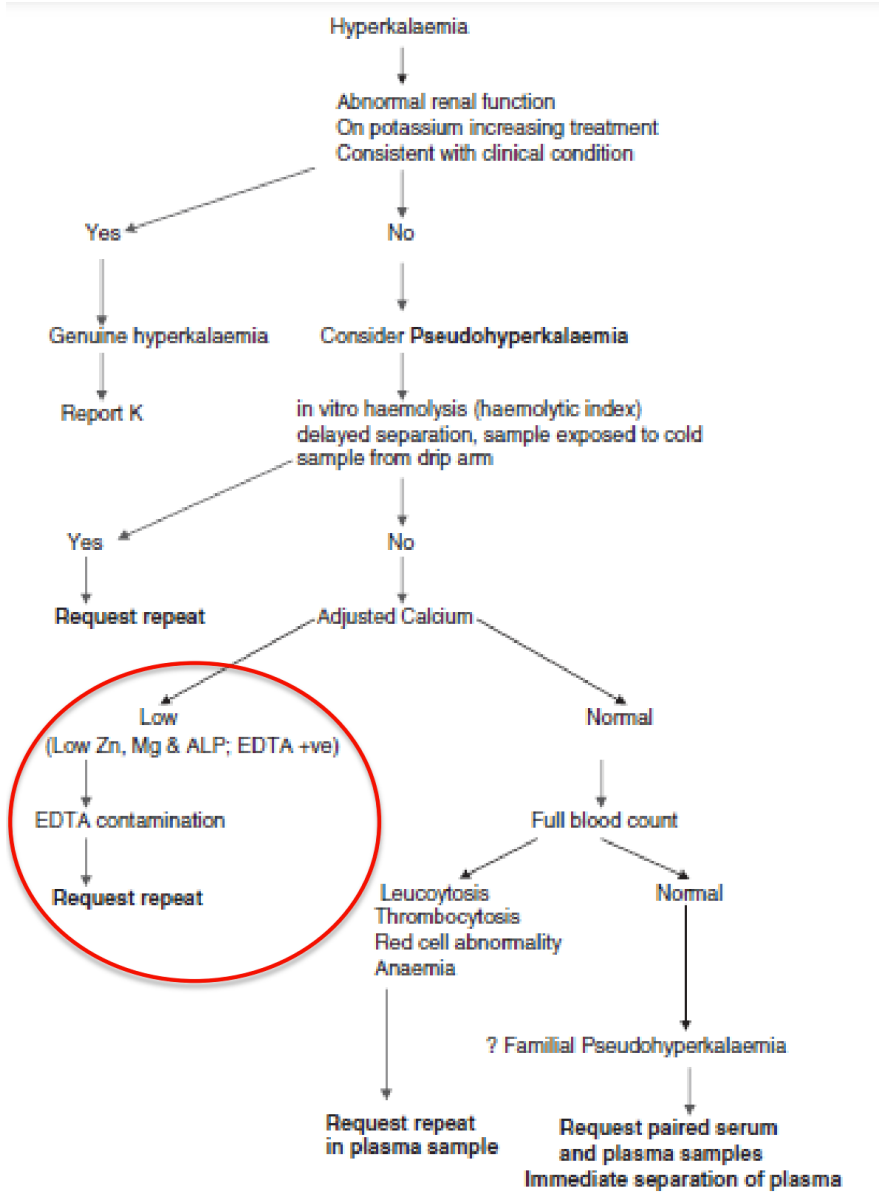
Method of tube filling	Number
Cannula with syringe	
Needle added and then tube cap pierced	14 (74%)
Evacuated tube cap removed	5 (26%)
Syringe and needle into vein	
Needle kept on and tube caps pierced	6 (86%)
Needle removed and evacuated tube cap removed	1 (14%)
Both methods	
Needle piercing of tube cap	20 (77%)
Needle and tube cap removed	6 (23%)

All of these can potentially lead to contamination if an incorrect order of draw is performed.

Ann Clin Biochem. 2011 Nov;48(Pt6):562-5

OOD – is it all a myth? (6)

- OOD not so important with phlebotomists and closed systems?
- Still a good idea to standardise OOD
  - Junior staff, nurses, doctors
  - No "cost" to following OOD
  - Potentially reduce incidence contamination
- Stop "potting off"
  - Laboratory education
- Methods for detecting
  - EDTA assay
  - Algorithms
- N.B. EDTA not only contaminant we see



Clin Chim Acta. 2011 Jan 14;412(1-2):1-6

HIL

- Haemolysis
- Icterus
- Lipaemia
  - Triglyceride
  - Other lipids (e.g. TPN)
- Haemolysis is most common PAE
  - Hb, intracellular
- Icterus
  - Bilirubin

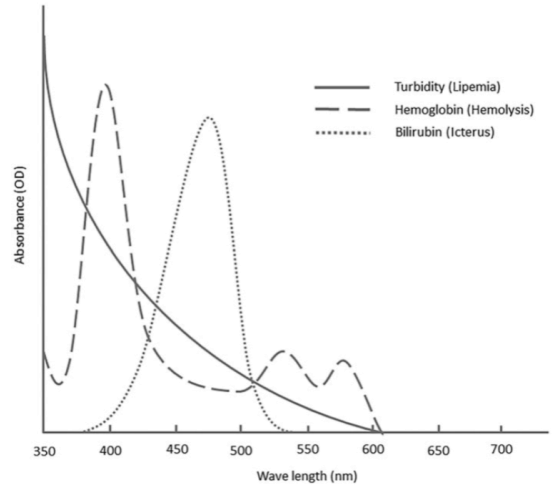
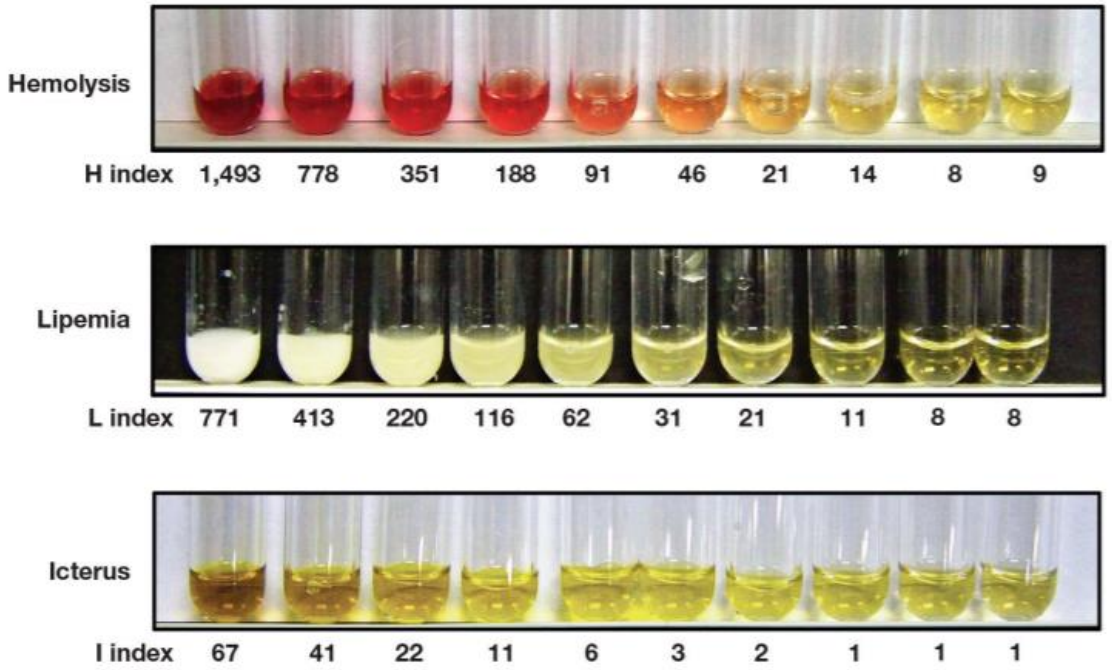


Figure 1. Spectral absorbance of turbidity, icterus and hemolysis (Note: Curves do not reflect the absolute value of HIL extinctions, which are concentration-dependent).



Haemolysis - example

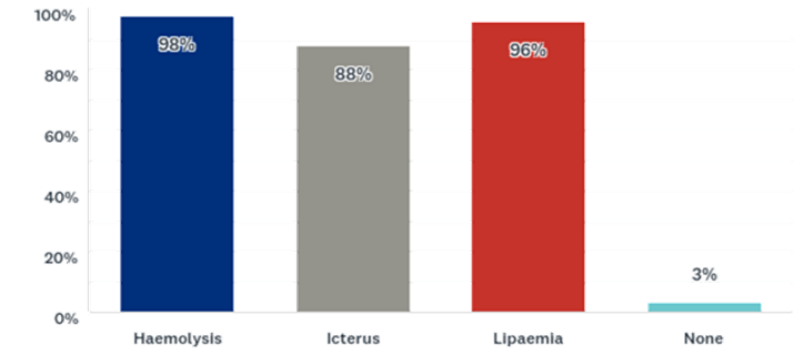
Na	136.9
K	7.25
Urea	1.47
Creatinine	38.26
Chloride	102.5
Albumin	41
ALT	1447.8
AST	513.5
TP	72.78
ALP	91.9
Tbili	17.7

HI
0
1
2
3
4
5

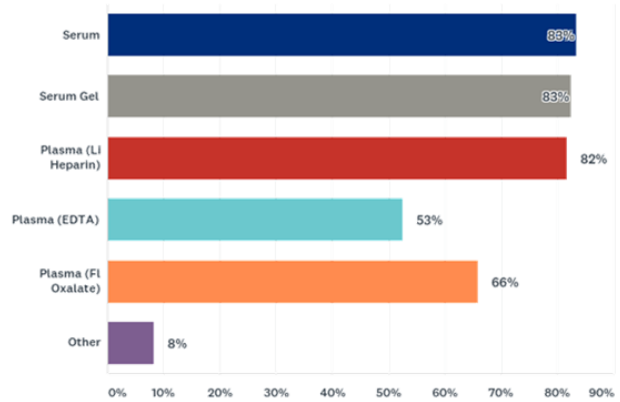
Table 1. Factors most commonly associated with *in vitro* hemolysis.

Phlebotomy	Sample transport	Sample preparation	Sample storage
• Drawing blood from location other than antecubital fossa • Catheter collection (venous or arterial) • Capillary collections • Drawing from hematomas • Blood frothing due to loose connections in blood collection system • Needle gauge too small or too large • Antiseptic used prior to phlebotomy • Tourniquet time • Traumatic draw • Tube underfilling • No mixing or overly vigorous mixing of tube • Collection into tube with excessive suction • Forcing blood into a tube from a syringe	• Origin of specimen (maternity, emergency department, intensive care unit) • Transport modality (e.g. pneumatic tube, porter, courier, drone) • Transport conditions (e.g. time, temperature and humidity) • Number of days at ambient temperature • Frozen in transport	• Time delay before centrifugation • Centrifuge conditions (force, time, temperature) • Poor barrier integrity • Centrifugation before sufficient time to clot has elapsed (serum) • Serum specimen only partly coagulated when centrifuged (e.g. in patients on anticoagulant therapy) • Blood diluted with hypotonic solution	• Specimen re-spun • Storage conditions (temperature and duration)

A. Interferents Monitored



B. Sample types monitored



C. HIL detection methods

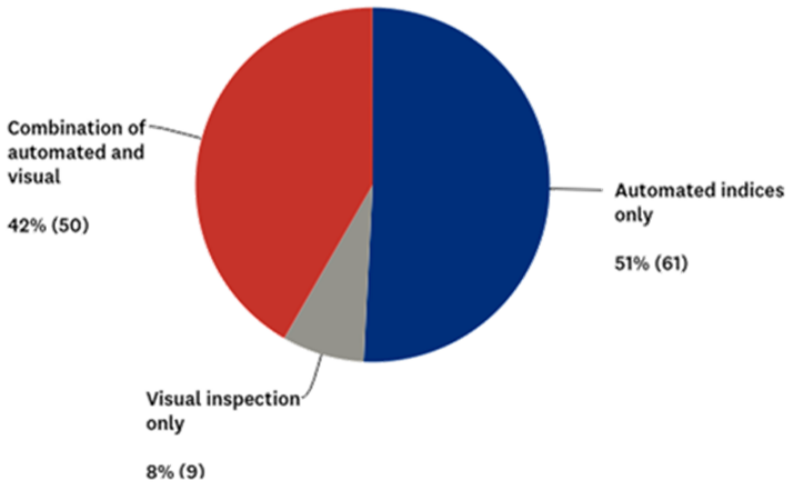


Figure 2: HIL monitoring and detection practices amongst respondents. Figures A, B and C show, respectively, responses to Q10 "*Please indicate which of the following interferents are monitored in your laboratory*"; Q11 "*Please indicate the sample types that are monitored for haemolysis, icterus or lipaemia*"; and Q12 "*How do you monitor haemolysis, icterus and lipaemia in your laboratory?*". RR for Q10 was 123 and for Q11/12 was 120.

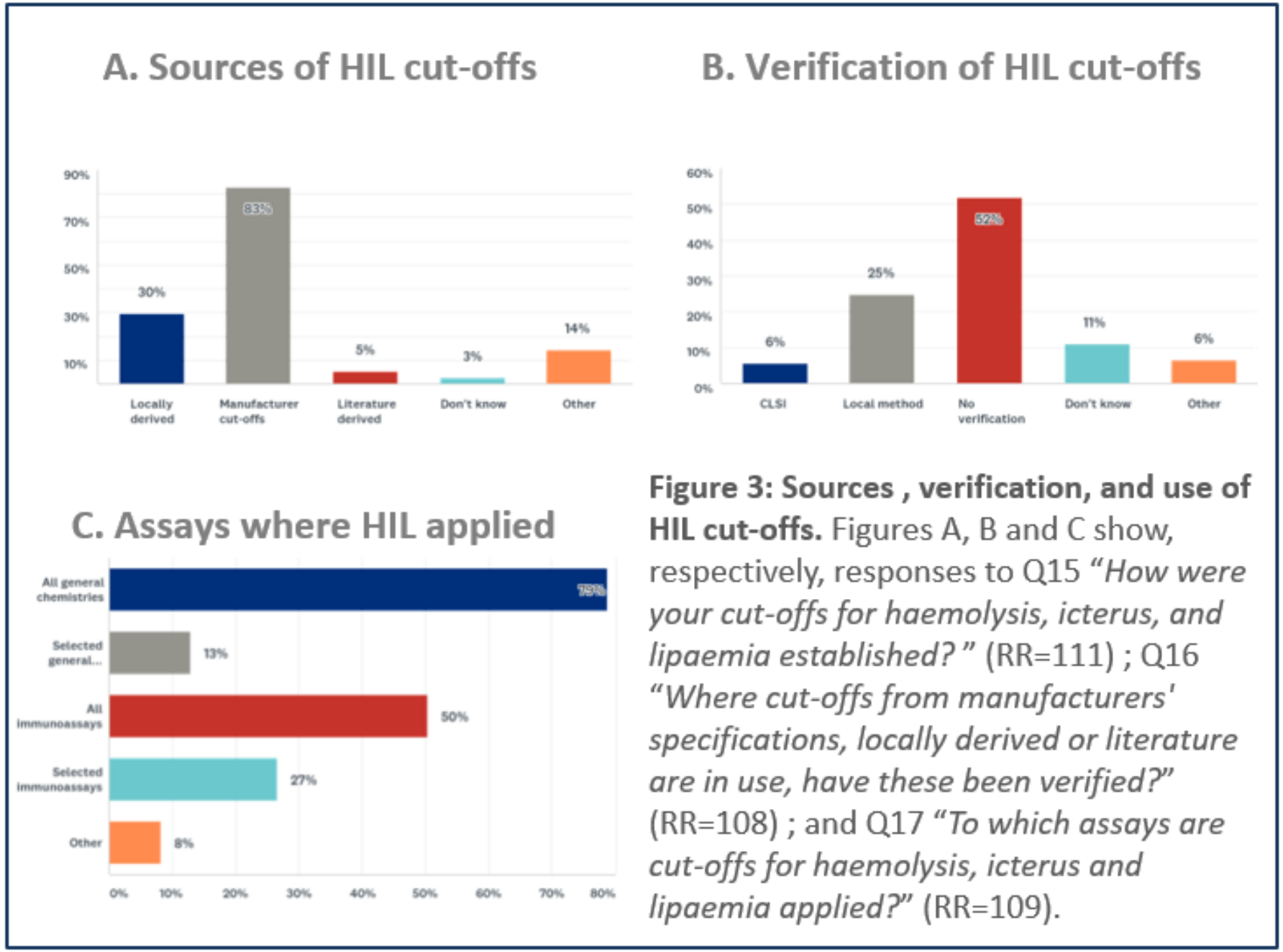


Figure 3: Sources , verification, and use of HIL cut-offs. Figures A, B and C show, respectively, responses to Q15 "*How were your cut-offs for haemolysis, icterus, and lipaemia established?* " (RR=111) ; Q16 "*Where cut-offs from manufacturers' specifications, locally derived or literature are in use, have these been verified?*" (RR=108) ; and Q17 "*To which assays are cut-offs for haemolysis, icterus and lipaemia applied?*" (RR=109).

Monitoring frequency of HIL

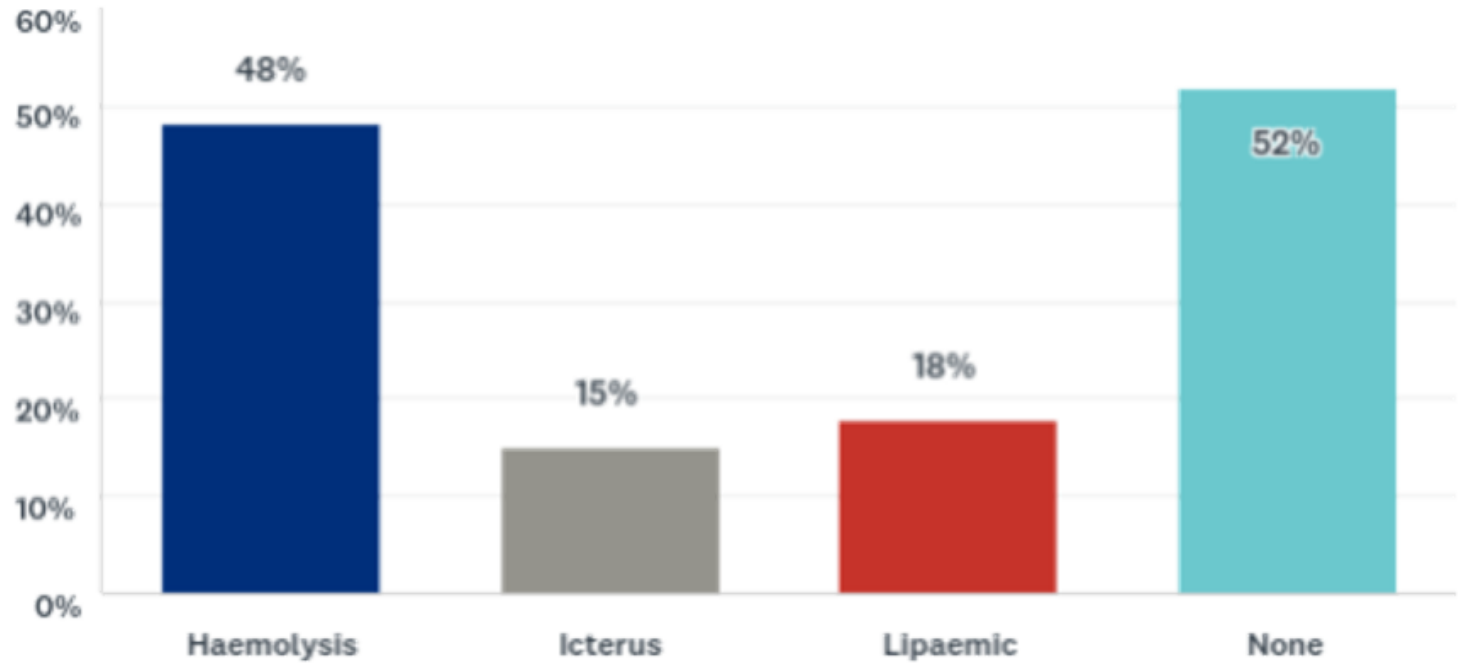


Figure 5: Proportion of responding laboratories who monitor rates of HIL interference. The figure shows responses to Q33 "*Are the proportion of tests affected by haemolysis, icterus and lipaemic interference monitored in your laboratory?*". RR was 112.

Quality assurance of HIL

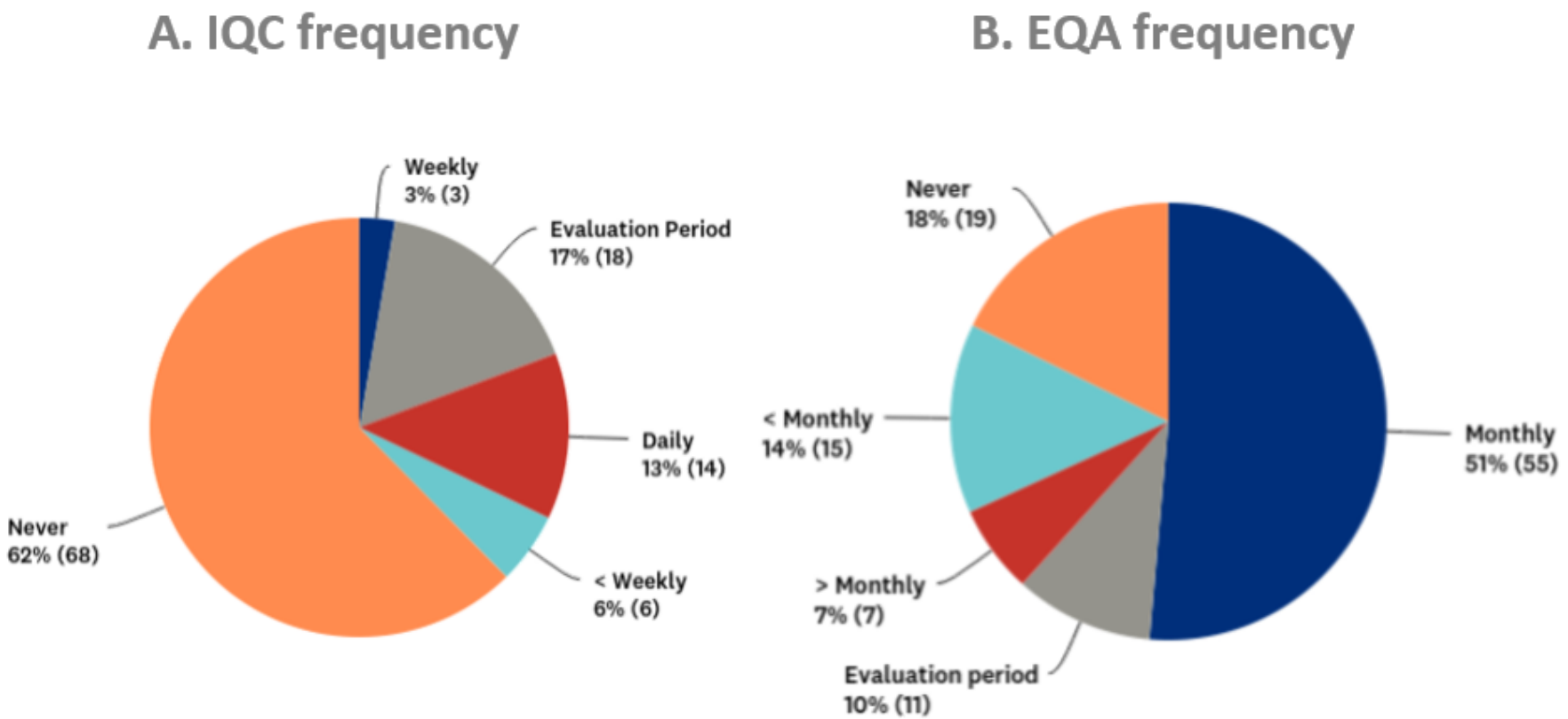


Figure 6: Frequency of quality control procedures for HIL. Figures A and B show, respectively, responses to Q41 "*Frequency of haemolysis, icteric, and lipaemic Internal Quality Control (IQC) procedures*" (RR=109) and Q44 "*Frequency of participation in EQA for haemolysis, icteric, and lipaemic interference*" (RR=107).

HIL: Other laboratory considerations

- Always block result if haemolysed?
  - Ammonia
  - Troponin
  - Give categorical results?
- Humans bad at estimating effects of H
- Need algorithms
- What about POCT?
  - Is frequency of H known?
  - BG analysers
    - K values
    - Bilirubin values
- Detect haemolysis at phlebotomy

Clin Chem Lab Med 2005;43(2):216–220

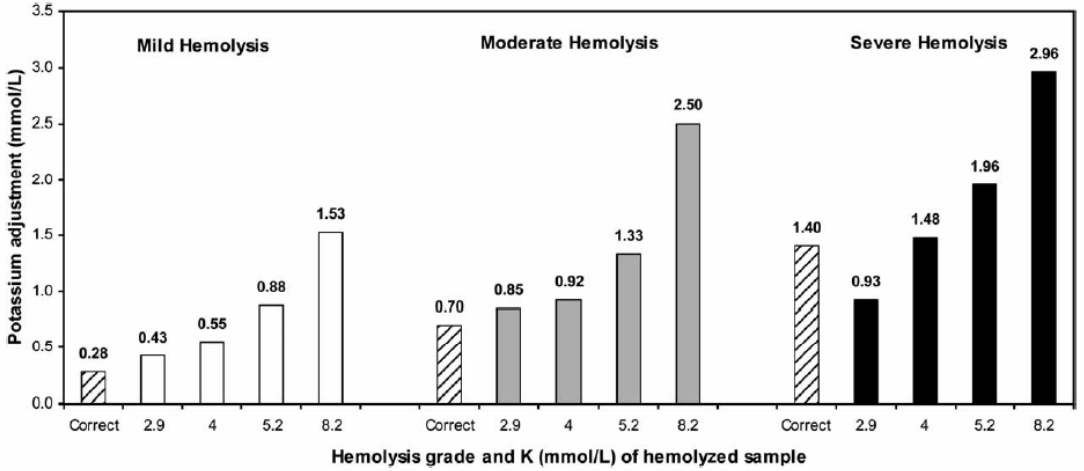


Figure 1 Mean absolute K adjustments (mmol/L) for different hemolysis grades and different "question K" concentrations. The shaded column represents the correct response.

Lippi, G, et al. Clin Chem Lab Med 2018

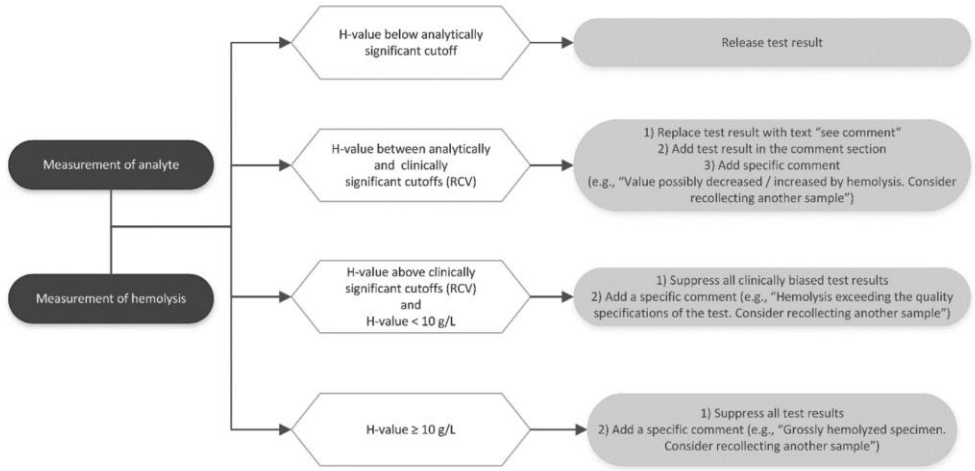


Figure 2: Summary of practical recommendations for managing hemolyzed specimens for clinical chemistry testing.

Case presentation

- Lactate 10.2 mmol/L (RI: 0.5-2.2 mmol/L)
- Levels greater than 4 mmol/L are suggestive of severe sepsis
- HIL not routinely measured for fluoride oxalate on AU5800 at CUH
- Visual inspection
    - grossly icteric
    - Lactate measurement – susceptible to interference from icterus
    - should result be released? Spurious?
- Discussed with Consultant
- HIL detection performed on AU5800 – icteric +1

Assessing HIL by eye

Study by Simundic et al, 2009

Clin Chem Lab Med 2009;47(11):1361–1365 © 2009 by Walter de Gruyter • Berlin • New York. DOI 10.1515/CCLM.2009.306

Comparison of visual vs. automated detection of lipemic, icteric and hemolyzed specimens: can we rely on a human eye?

Table 4 Comparison of visual inspection and LIH Olympus reagent for icterus.

LIH Olympus reagent	Visual inspection		
	0	1	2
0 (n)	1586	41	3
1 (+ and ++)	17	31	11
2 (3+ to 5+)	0	7	8
	1603 (94.1%)	79 (4.6%)	22 (1.3%)

Table 6 κ Coefficients with respective 95% CI for five randomly selected laboratory technicians.

	B κ (95% CI)	C κ (95% CI)	D κ (95% CI)	E κ (95% CI)	F κ (95% CI)	Mean κ
Haemolysis	0.584 (0.295–0.874)	0.726 (0.480–0.972)	0.622 (0.318–0.916)	0.569 (0.268–0.870)	0.583 (0.293–0.874)	0.617 (0.537–0.696)
Lipemia	0.743 (0.470–1.016)	0.643 (0.322–0.964)	0.743 (0.470–1.016)	0.627 (0.292–0.962)	0.732 (0.448–1.016)	0.698 (0.626–0.769)
Icteria	0.655 (0.345–0.965)	0.594 (0.276–0.912)	0.160 (−0.321–0.640)	0.446 (0.098–0.794)	0.527 (0.197–0.857)	0.476 (0.237–0.716)
Mean κ	0.661 (0.463–0.859)	0.654 (0.489–0.820)	0.508 (−0.256–1.273)	0.547 (0.318–0.777)	0.614 (0.351–0.877)	

Best agreement

Least agreement

- Visual vs. automated detection
  - Poor agreement
    - Visual inspection – 101 icteric
    - Automated HIL – 74 icteric
- Poor inter-operator agreement by visual assessment

OUTCOME: Visual assessment is inconsistent & un-reliable

Interference

- Highlights the importance of HIL detection by automated methods
- **What was causing the discolouration of plasma**
- **Could it be interfering with result?**
  - Prompted discussion with clinical colleagues

Case discussion

- Female, 22 yrs, epilepsy, mixed developmental disorder
- Severe aplastic anaemia
- March 2022 - electively admitted to CUH for anti-thymocyte globulin therapy

Haematology	Result	Normal Range
Hb	8.4g/dL	11.7 – 15.9
RBC	$2.76x10^{12}$/L	3.9-5.3
WBC	$1.4x10^{9}$/L	4.4-11.3
Platelets	$26x10^{9}$/L	140-440

Immunophenotyping & Bone Marrow Aspirate
Consistent with aplastic anaemia

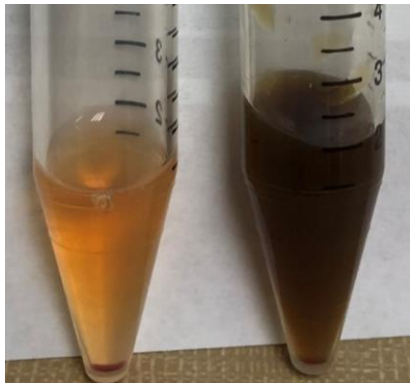
Genetics	
BM Karyotype	46,XX, del(13)(q12q14)[5]/45,XX[25]

- Week 2 – severe neutropenic sepsis – ITU admission

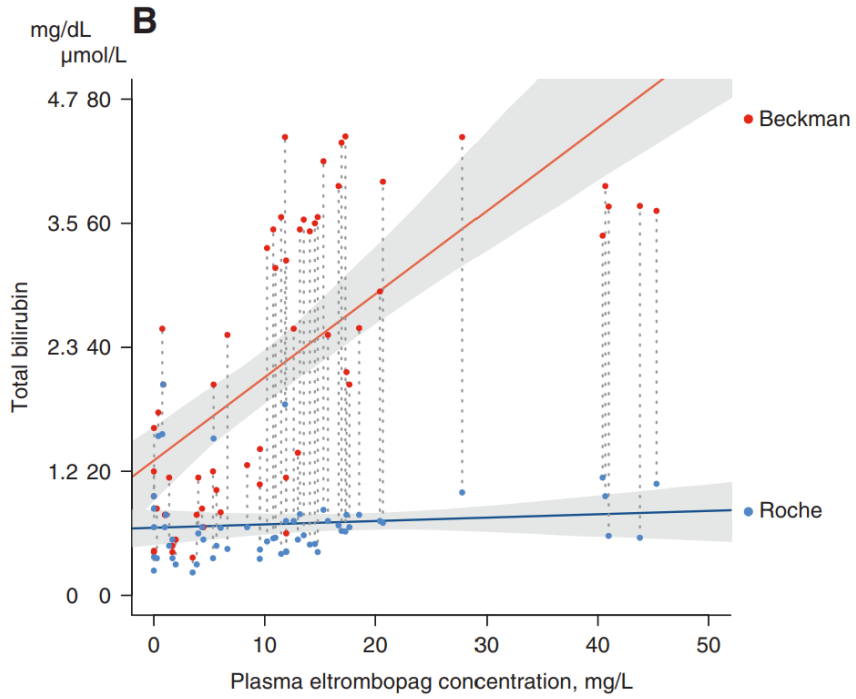
Biochem	Result	Normal Range
Lactate	10.22mmol/L	0.5 – 2.2

Eltrombopag

- Eltrombopag mimics bilirubin
  - Absorbance
    - ~450 nm (Bilirubin)
  - Appearance
    - Causes pH dependent discolouration serum/plasma
  - Causes discrepant inter-analyser bilirubin results
  - Lack of information regarding eltrombopag and lactate
- Likely that Eltrombopag was the cause of discolouration of specimen
- Result released with a cautionary comment

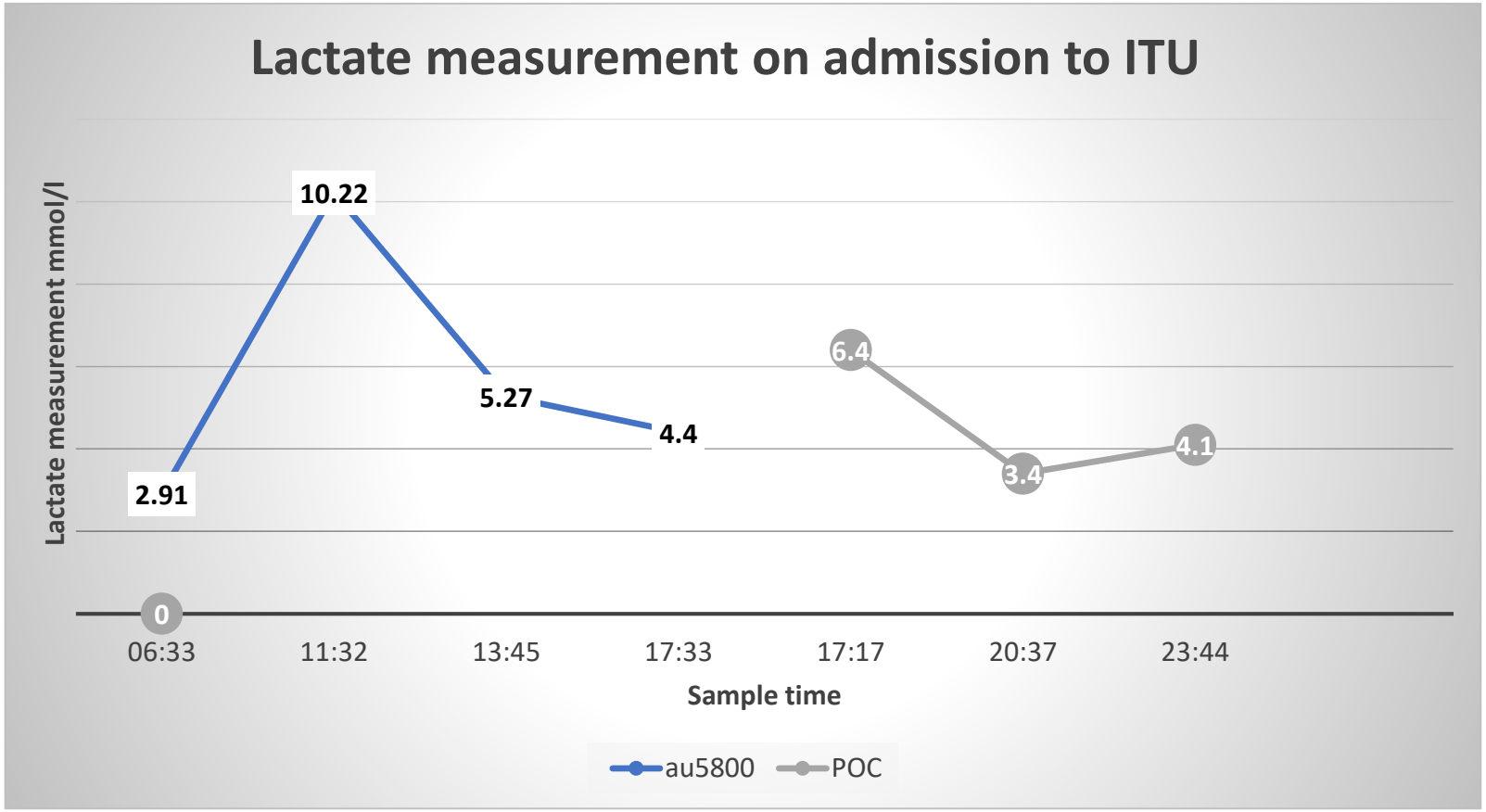


American J Hematol, Volume: 94, Issue: 3, Pages: 394-395, First published: 22 July 2018, DOI: (10.1002/ajh.25169)

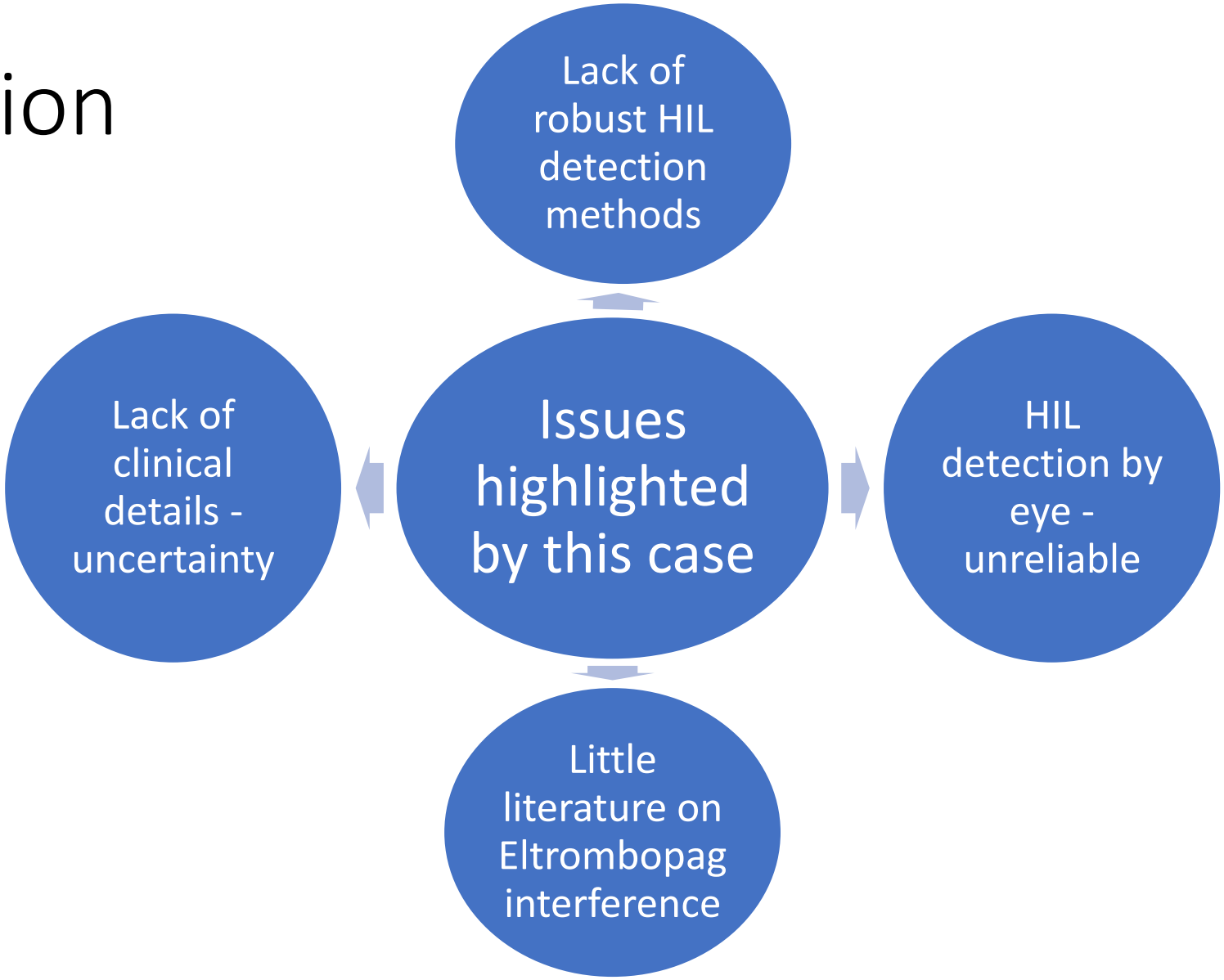


Clin Chem Lab Med 2020; 58(10): 1713–1723

Lactate measurement in patient on Eltrombopag: comparison of AU5800 vs POC



Conclusion


Lack of robust HIL detection methods
Lack of clinical details - uncertainty
Issues highlighted by this case
HIL detection by eye - unreliable
Little literature on Eltrombopag interference

Specimen stability and storage

- Many factors can affect
  - Sample type
  - Temperature
  - Centrifuge conditions
  - Tube type
  - Additives
  - Light exposure
  - Mixing technique
  - Evaporation

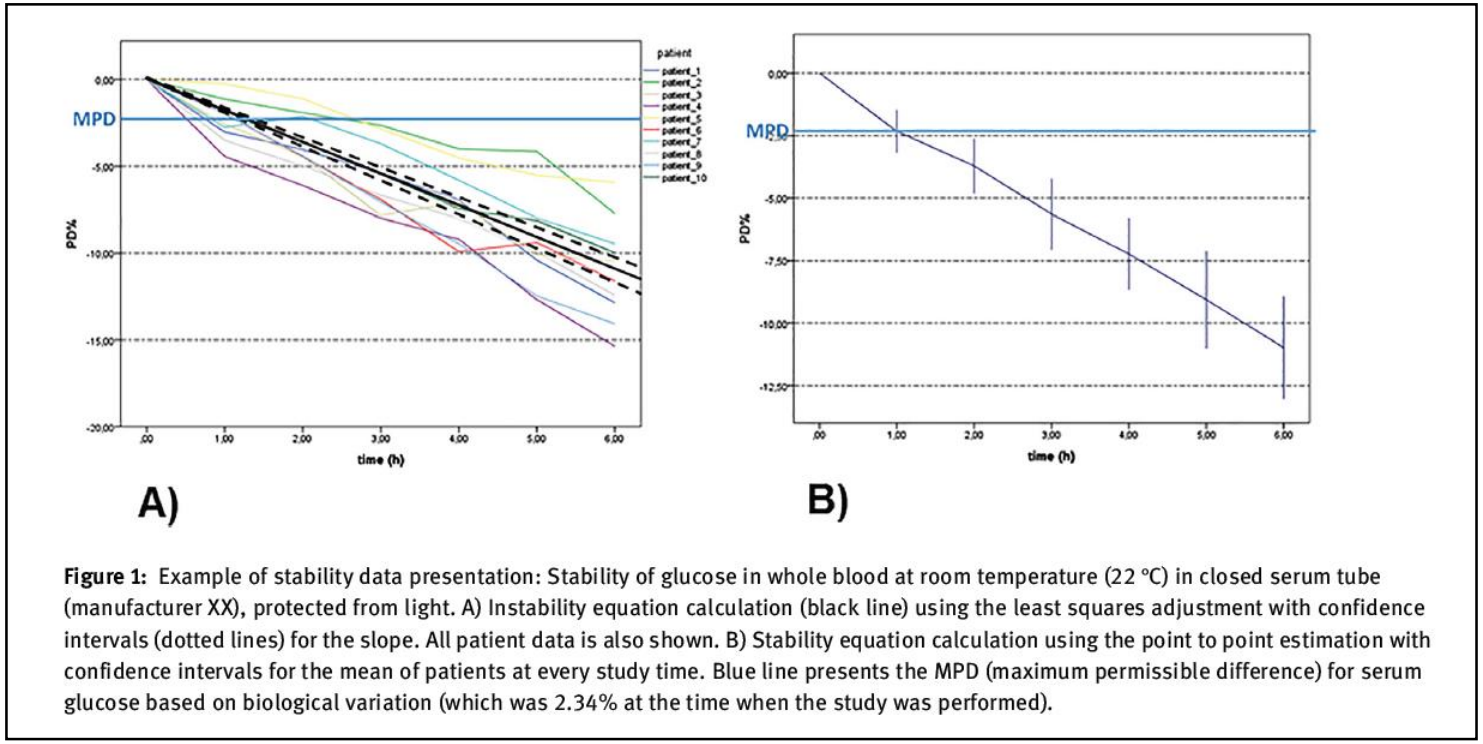


Figure 1: Example of stability data presentation: Stability of glucose in whole blood at room temperature (22 °C) in closed serum tube (manufacturer XX), protected from light. A) Instability equation calculation (black line) using the least squares adjustment with confidence intervals (dotted lines) for the slope. All patient data is also shown. B) Stability equation calculation using the point to point estimation with confidence intervals for the mean of patients at every study time. Blue line presents the MPD (maximum permissible difference) for serum glucose based on biological variation (which was 2.34% at the time when the study was performed).

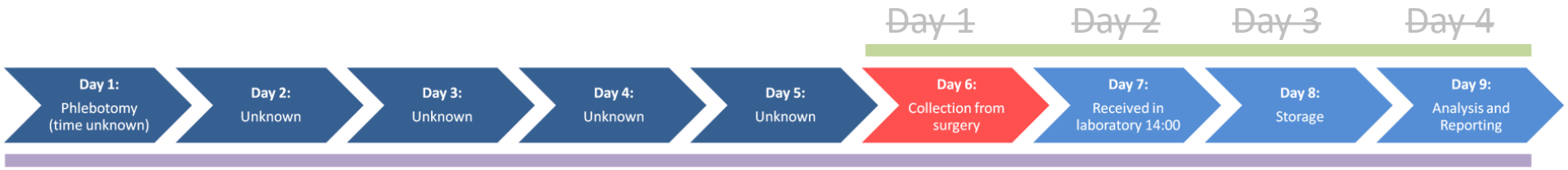
- Each lab. should have stability limits appropriate for their blood tubes, analysers, intended clinical use etc..

Case presentation

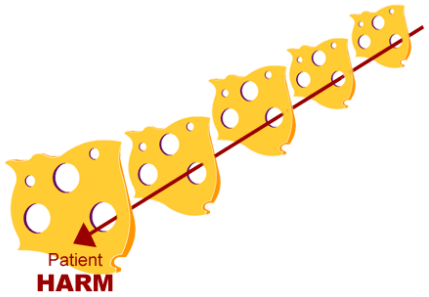
- An unseparated gel serum tube was received from primary care with the following details:
  - 36 year old male
  - Unknown medical history
  - No clinical details
  - Date of collection was previous day (Day 1)
  - No time of collection was given
- Specimen was centrifuged on day of receipt (Day 2)
- Specimen was analysed on a Beckman Coulter AU5800 (Day 4)
- Tests requested included renal profile, liver profile and calcium
- Potassium, AST, and bilirubin results were blocked due to haemolysis (HI = 2)
- Although sodium (119 mmol/L) and calcium (1.15 mmol/L) were significantly lower than previous measurements in this patient and both results were authorised
- Sodium and calcium results breached local critical phoning limits as defined by the Royal College of Pathologists UK

Case presentation (2)

- Low total calcium and sodium results are occasionally observed in cases of delayed separation at CUH
- The General Practitioner (GP) confirmed the request card incorrectly completed
- Phlebotomy was actually 7 days prior to receipt in the laboratory
- “Date of collection” on the request form corresponded with date of collection by the courier from the surgery



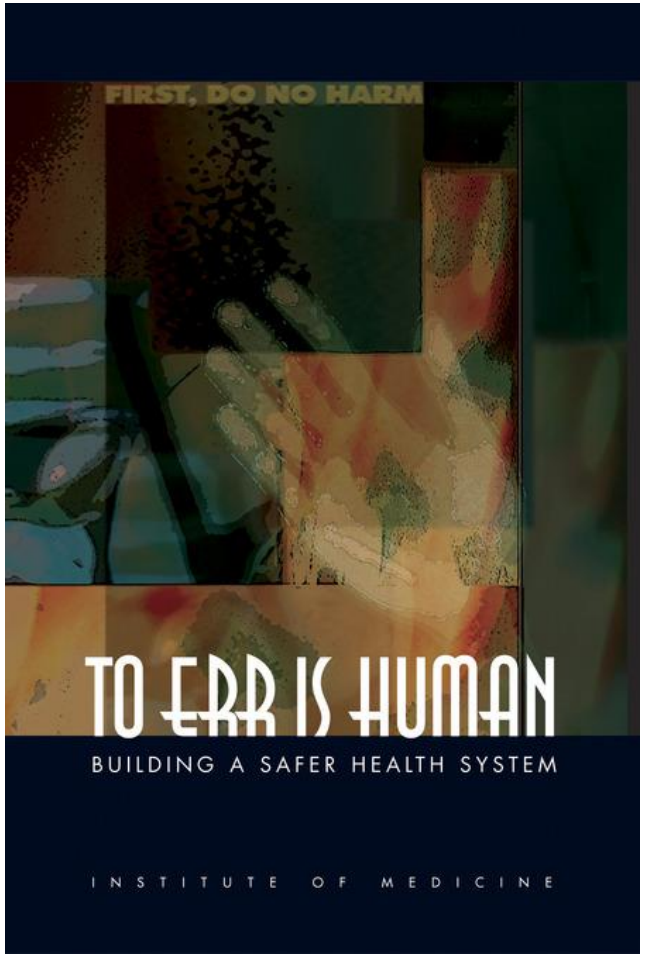
Case presentation (3)



- Several preanalytical errors were at play in this case
  - Incorrect/incomplete date and time of collection
  - Lack of clinician understanding that the date of “collection” applied to date of venesection rather than date of specimen collection by the courier
  - Delayed separation
  - Haemolysis
  - Delayed analysis in the laboratory
  - Lack of robust process to catch potentially spurious calcium and sodium results prior to reporting
- Preanalytical errors converged, leading to confusion in the generation of laboratory results for this patient
- Delayed separation reproducibly causes artefactually low results for total calcium and sodium respectively
- Lack of awareness of these issues by laboratory staff, clinicians or out-of-hours service providers may lead to inappropriate patient admissions

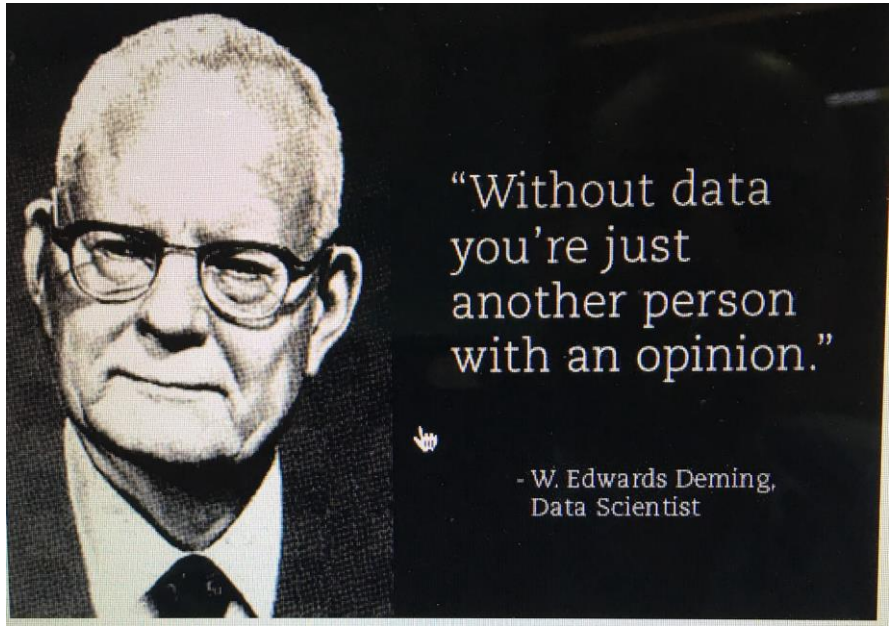
Increased awareness of PAE (2)

- The Institute of Medicine report, *To Err is Human* galvanized a dramatic increase in concern about adverse events and patient safety at an international level.
- ISO15189:2012
  - Lab. scope extends remit of lab. into extra-analytical phases
    - QMS
    - Continual improvement
- 2013 Francis report
- EFLM and IFCC
  - Conferences on Preanalytical Phase
  - IFCC Working Group (WG-LEPS)
  - EFLM Working Group (WG-PRE)
- Australian KIMMS
  - NEQAS style scheme for PAE
- WEQAS EQA schemes for HIL
- NEQAS PrepQ
- ACB SIG



Monitoring and reducing PAE (1)

- Benefits of KPIs in quality management
  - You cannot improve what you don't measure
  - Lab test results are only as good as the condition of the specimen allows Garbage in, garbage out!
  - Ensures the result is connected to the right specimen and patient
  - Ensure quality specimen management for accurate test results
  - Laboratory and patient safety



Monitoring and reducing PAE (3)

- Accreditation and PAE ISO 15189:2012
  - The ISO 15189:2012 standard for laboratory accreditation defines the pre-analytical phase as “steps starting in chronological order, from the clinician's request and including the examination requisition, patient preparation, collection of the primary sample, and transportation to and within the laboratory, and ending when the analytical examination procedure begins”
  - This definition recognizes the need to evaluate, monitor and improve all the procedures and processes in the initial phase of the TTP, including the procedures performed in the so-called “pre-pre-analytical phase”

ISO 15189

- The use of QIs in clinical laboratories to monitor all critical activities of pre-, intra- and post-analytical phases is required

- However, 35% of labs do not routinely monitor any pre-analytical QIs.

Errors that are monitored:

- 80% Haemolysis / Icteric / Lipaemic indices
- 70% Booking-in errors
- 57% Mislabelling errors

Aita et al. Diagnosis 2017;4(4):193-5.

Guidance from EFLM

EFLM Paper

Pieter Vermeersch*, Glynis Frans, Alexander von Meyer, Seán Costelloe, Giuseppe Lippi and Ana-Maria Simundic

How to meet ISO15189:2012 pre-analytical requirements in clinical laboratories? A consensus document by the EFLM WG-PRE

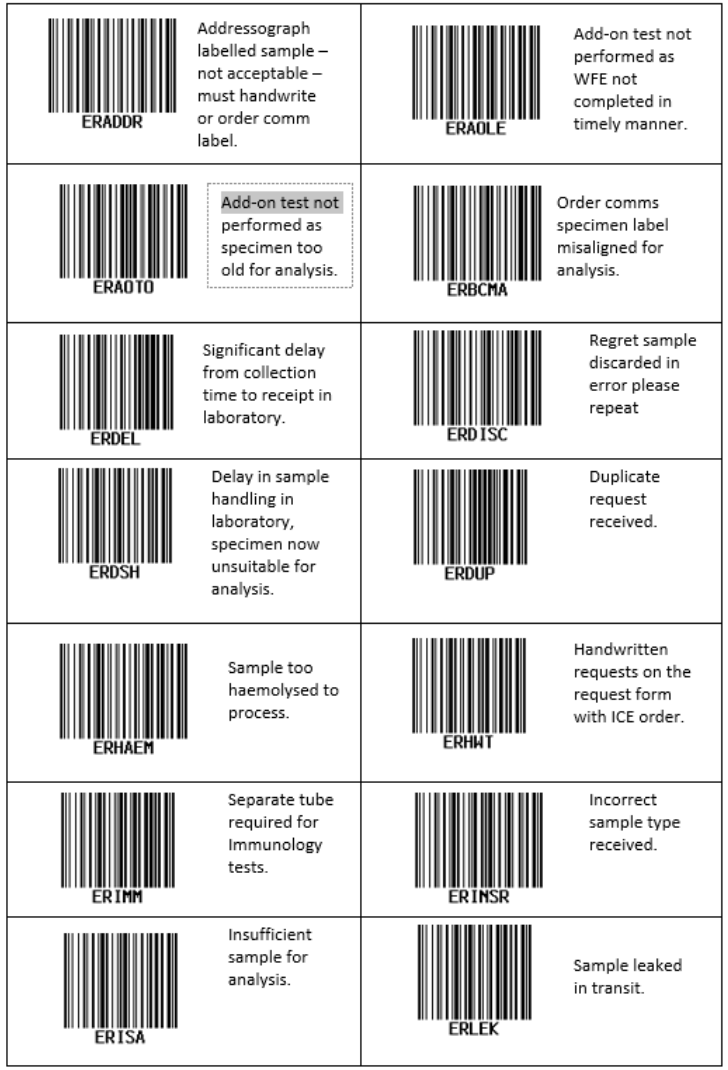
Table 1: ISO15189:2012 requirements and corresponding EFLM WG-PRE recommendations/solutions relating to quality management of the pre-examination phase.

ISO paragraph	Question	ISO requirement	Minimal recommendation	Grade	Best-in-class solution	Grade
4.1.2.4 – Quality objectives and planning	How to define quality objectives and quality indicators for pre-examination processes?	The quality objectives shall be measurable and consistent with the quality policy.	Laboratories should at least monitor one of the following quality indicators: number of misidentification errors, test transcription errors, incorrect sample types, insufficiently filled samples, unsuitable samples, contaminated samples, hemolyzed samples, or clotted samples.	2a	Pre-analytical quality indicators are monitored according to framework provided by the IFCC Model of Quality Pre-analytical Indicators. Laboratories should implement all quality indicators that are relevant for their setting based on risk-assessment. Participation in the IFCC External Quality Assessment program is encouraged.	2a
	How frequent should pre-analytical quality objectives/quality indicators be evaluated?	Planning of the quality management systems is carried out to meet the requirements and the quality objectives.	Yearly.	1	Frequency according to the framework provided by the IFCC Model of Quality Pre-analytical Indicators.	2a

Monitoring and reducing PAE (4)

Developing Indicators

Objective	What are you trying to measure?
Methodology	**1. How to capture the data? – flag data** **2. Who (or what) to capture the data?** **3. How often to capture the data?**
Set Limits	Acceptable, Concern, Unacceptable Critical
Presentation	Graphic or Text
Interpretation	What does it mean? Who's quality does it reflect?
Limitations	Unintended variables or uncontrollable variables
Action Plan	**What will I do if it indicates acceptable performance?** **What will I do if it does not?**
Exit Plan	When can I stop measuring?



Monitoring and reducing PAE (5)

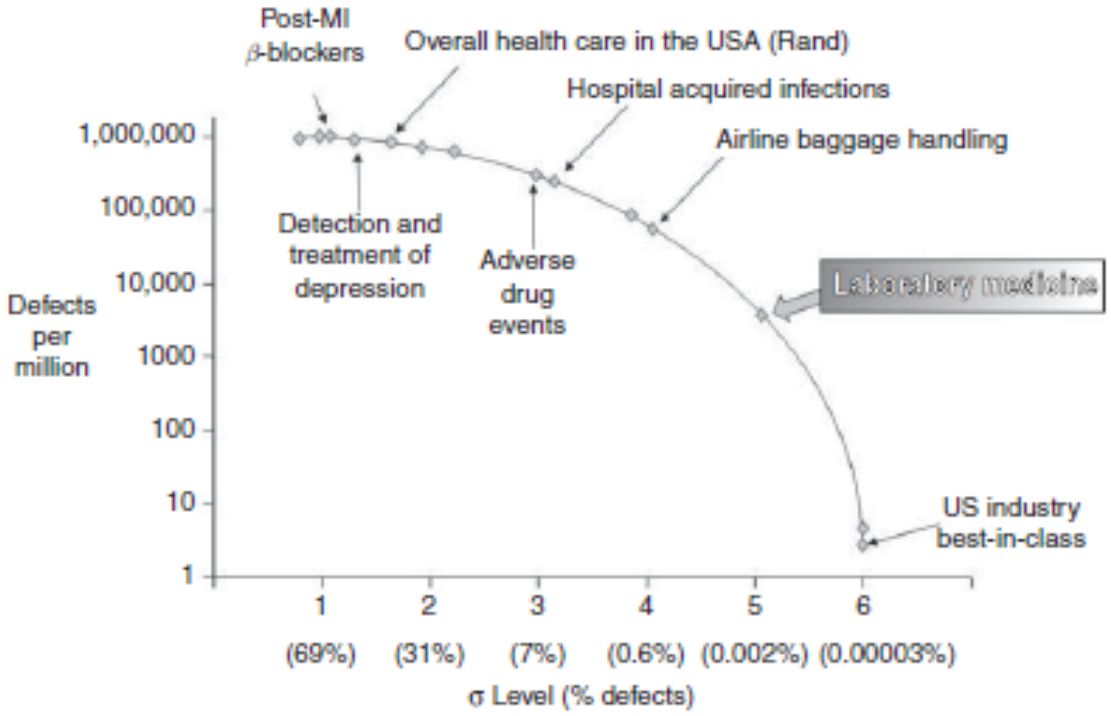
e) Sample collection	
Percentage of "Number of samples collected at inappropriate time / Total number of samples"	2
Percentage of "Number of samples collected with inappropriate sample type / Total number of samples"	1
Percentage of "Number of samples collected in inappropriate container / Total number of samples"	1
Percentage of "Number of samples with insufficient sample volume / Total number of samples"	1
f) Transport of sample	
Percentage of "Number of damaged samples / Total number of samples"	1
Percentage of "Number of samples transported at inappropriate time / Total number of samples for which transport time is checked"	1
Percentage of "Number of samples transported under inappropriate temperature condition / Total number of samples for which the transport temperature is checked"	1
Percentage of "Number of improperly stored samples / Total number of samples"	1
Percentage of "Number of samples lost-not received / Total number of samples"	1
g) Suitability of sample	
Percentage of "Number of samples with inadequate sample-anticoagulant volume ratio / Total number of samples with anticoagulant"	1
Percentage of "Number of hemolyzed samples (hematology) / Total number of samples (hematology)"	1
Percentage of "Number of hemolyzed samples (chemistry) / Total number of samples (chemistry)"	1
Percentage of "Number of clotted samples (hematology) / Total number of samples with anticoagulant (hematology)"	1
Percentage of "Number of clotted samples (chemistry) / Total number of samples with anticoagulant (chemistry)"	1
Percentage of "Number of clotted samples (immunology) / Total number of samples with anticoagulant (immunology)"	1
Percentage of "Number of hemolyzed samples (immunology) / Total number of samples (immunology)"	1
Percentage of "Number of lipemic samples / Total number of samples"	1
Percentage of "Number of unacceptable samples (microbiology) / Total number of samples (microbiology)"	1
Percentage of "Number of contaminated blood cultures / Total number of blood cultures"	1

Plebani M, Sciacovelli L, Aita A, Chiozza ML. Harmonisation of preanalytical quality indicators. Biochemia Medica 2014;24(1):105-13

Monitoring and reducing PAE (6)

Concept of Six Sigma

Sigma	Spelling
7	1 misspelled word in all of the books contained in several large libraries
6	1 misspelled word in all of the books contained in a small library
5	1 misspelled word in a set of encyclopaedias
4	1 misspelled word in a book chapter
3	1.5 misspelled words per page in a book
2	25 misspelled words per page in a book
1	170 misspelled words per page in a book



The detection and prevention of errors in laboratory medicine

Mario Plebani

Monitoring and reducing PAE (7)

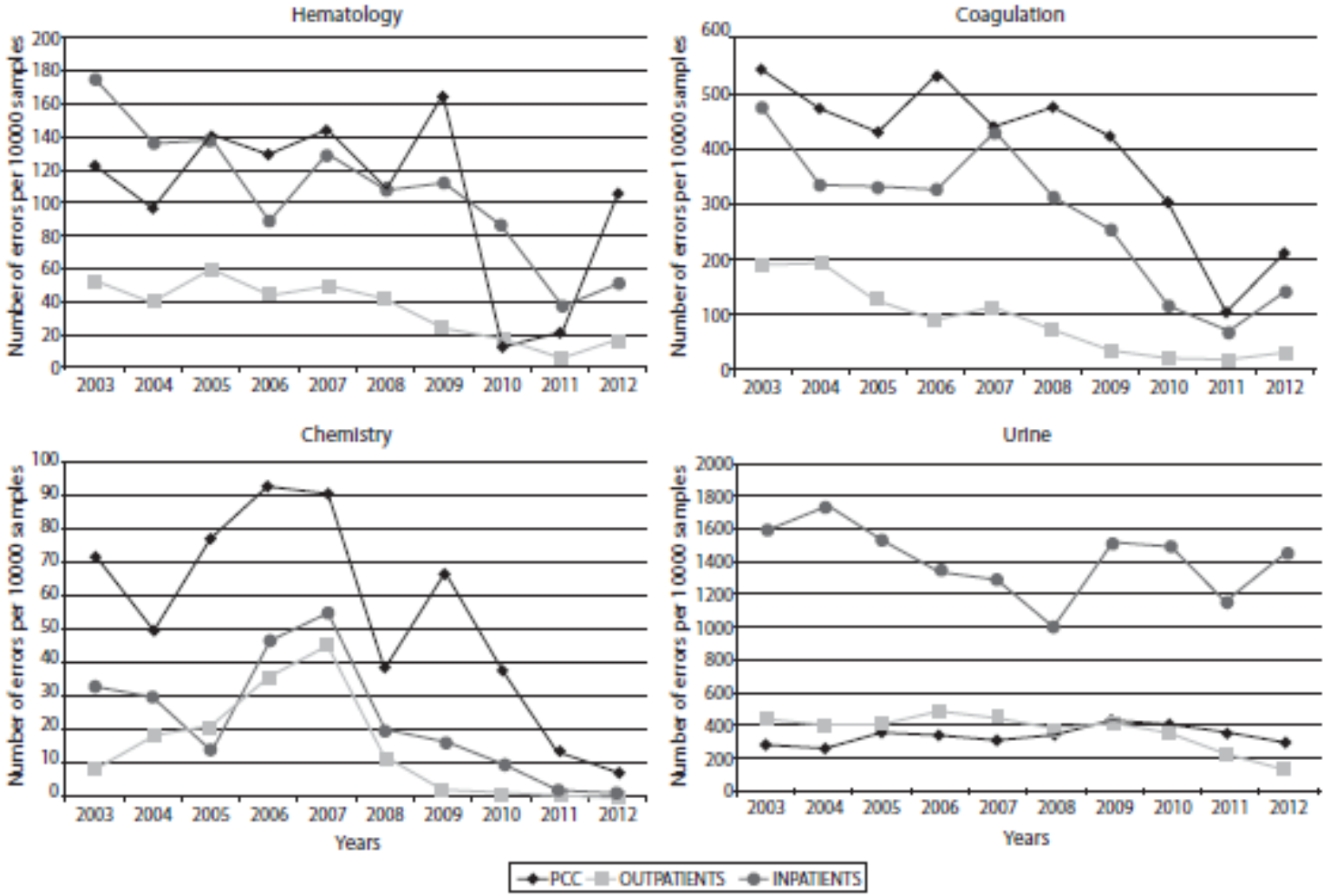


FIGURE 2. Annual global indicator results in every type of patient: Shows the sum of all types of preanalytical errors with respect to every sample collected in inpatients, outpatients and primary care patient's samples.

Salinas M et al. Ten years of preanalytical monitoring and control:Synthetic Balanced Score Card Indicator. Biochemia Medica 2015;25(1):49-56

National, European, International Initiatives

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SAGE journals

Editorial

Management of transgender patients in Laboratory Information Management Systems – Moving on from binary and ternary logic

Seán J Costelloe [1,2] and Sophie Hepburn [1,3]

Research Article

The Association for Clinical Biochemistry & Laboratory Medicine
Better Science, Better Testing, Better Care

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Survey of patient perception of pre-analytical requirements for blood testing in the UK and RoI

S Hepburn[1,2], M Jankute[2,3], MP Cornes[2,4], N Rico Rios[5], A Stretton[2,6] and SJ Costelloe[2,5]

Research Article

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A survey of practice in the management of haemolysis, icterus and lipaemia in blood specimens in the United Kingdom and Republic of Ireland

Seán J. Costelloe[1,2], Natividad Rico Rios[1], Nicola Goulding[1], Hema Mistry[2,3], Adam Stretton[2,4], Barbara De la Salle[2,5], Sophie Hepburn[2,6], Annette Thomas[2,7], Jennifer Atherton[2,8] and Michael Cornes[2,9]

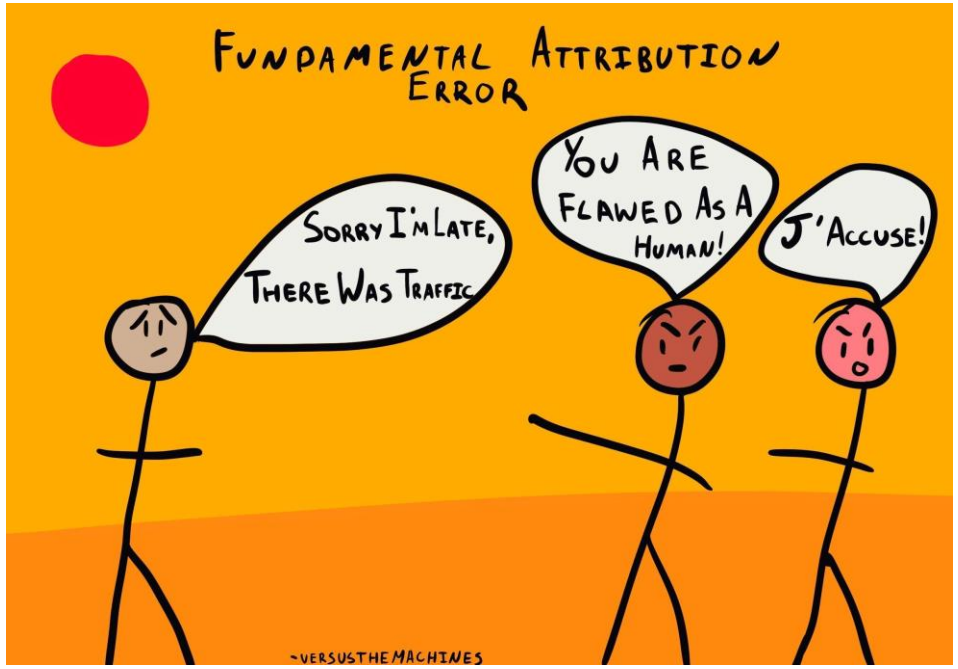
- ACB Preanalytical Special Interest Group
  - Upcoming work
    - Transgender survey
    - ED Panels
    - Multi-centre stability studies
    - Collaboratory with US/Canadian groups
    - Collaboration with GIRFT/EFLM
- EFLM WG-PRE
  - Upcoming work
    - Stability
      - How to perform stability studies
      - How to evaluate stability studies
      - Stability database
      - Urine acidification studies
    - Recommendations tube validation

ACB Preanalytical Special Interest Group

Thoughts on working group

- Work we do
  - Fewer surveys
  - More guidance
- Makeup
  - Excellent expertise
  - Top heavy with consultants?
  - Many projects in pipeline
  - Time to completion is several years
  - Scope for younger members to get involved
  - If interested contact
    - Dr Seán Costelloe: sean.costelloe@hse.ie

Take aways



- ~60% of errors in TTP are in PP
- Does your laboratory give enough time to
  - Recording PAI?
  - Monitoring PAIs (KPIs)?
  - Addressing PAIs?
- Data for most common PAIs (e.g. H) are readily available in lab
- Resources needed – e.g. additional quality managers
- Recommendations and guidelines are available/coming

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Questions?

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