Preanalytical cases for educational purposes

From the Nordic Scientific Working Group on Preanalytics

Case #1

- A 56-year-old man
- Admitted to the ED with alcohol-induced severe acute pancreatitis
- Blood tests were ordered
- Test results:
  - Blood glucose, 15.4 mmol/L
- Clinicians refuse to take action since there were no signs of hyperglycaemia
- Second blood sample drawn and transported to the laboratory
- Test results:
  - Blood glucose, 4.9 mmol/L

Anaesthesia 2013, 68, 1179–1187

Accidental hypoglycaemia caused by an arterial flush drug error: a case report and contributory causes analysis

K. J. Gupta and T. M. Cook

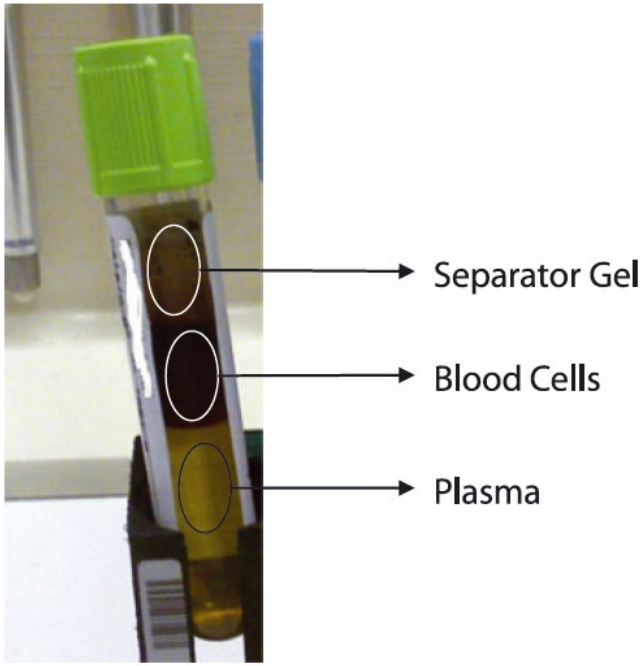
- First blood sample was drawn from **glucose-contaminated arterial blood line**, without appropriate flushing.

- **How do we avoid this??**

- **Education**
- **Education**
- **Education**

Case #2

- 50-year-old man
- Admitted to the ED for acute myocardial infarction
- Subjected to percutaneous coronary intervention (PCI)
- First blood sample drawn from femoral artery and transported to the laboratory
- Apperance of sample after standard centrifugation:



Biochemia Medica 2016;26(3):444–50

Abnormal gel flotation caused by contrast media during adrenal vein sampling

Gabriel Lima-Oliveira*[1], Giuseppe Lippi[2], Gian Luca Salvagno[1], Matteo Gelati[1], Antonella Bassi[1], Alberto Contro[3], Francesca Pizzolo[4], Gian Cesare Guidi[1]

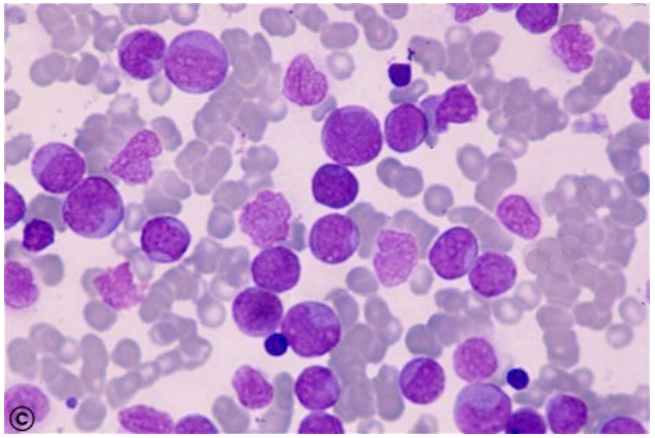
• Specific gravity of serum and plasma is 1.026-1.031 g/cm^3, and that of the clot is 1.092-1.095.
• Specific gravity of separator gels should be 1.03-1.09 g/cm^3 to permit its suitable positioning after centrifugation.
• The interfering substance was a **tri-iodinated nonionic water-soluble contrast dye**, 140 ml of which were administered to the patient before coronary revascularization

• **How do we avoid this??**

Case #3

- 72-year-old man
- Admitted to the ED for fatigue and dizziness lasting for days
- First blood sample drawn and transported to the laboratory by the PTS
- Test results:
  - WBC, 75 x 10^9/L
  - Hb, 5.0 mmol/L
  - Plasma potassium, 5.7 mmol/L
- No signs or symptoms of hyperkalemia
- Second blood sample drawn 45 after admission and manaully transported to the Lab
- Test results:
  - WBC, 78 x 10^9/L
  - Hb, 5.0 mmol/L
  - Plasma potassium, 3.9 mmol/L

- The patient is diagnosed with acute myeloid leukemia.
- Fragile **neoplastic leukocytes are injuried or destroyed** during PTS transportation, releasing potassium in the surrounding plasma.



Clinical Chemistry 64:5
782–790 (2018)

Mini-Review

Blood Sample Transportation by Pneumatic Transportation Systems: A Systematic Literature Review

Mads Nybo,[1*] Merete E. Lund,[2] Kjell Titlestad,[2] and Christian U. Maegaard[3]

Case #4

- 66-year-old man
- Hospitalized for colorectal cancer
- Routine (morning) blood sample drawn and transported to the laboratory
- Test results:
  - Creatinine, 82 µmol/L
  - Hb, 7.5 mmol/L
  - Plasma potassium, 17.2 mmol/L
  - Serum calcium, not measurable
- Second blood sample drawn after 1 h admission
- Test results:
  - Creatinine, 81 µmol/L
  - Hb, 7.6 mmol/L
  - Plasma potassium, 3.7 mmol/L
  - Serum calcium, 2.5 mmol/L

- Four blood samples were planned to be collected:
    - 1 EDTA blood tube
    - 1 Sodium citrate blood tube
    - 2 Serum blood tubes
- Blood stopped during collection of the fourth blood tube, leaving the tube almost empty.
- The nurse **pour some EDTA blood** into the serum blood tube.

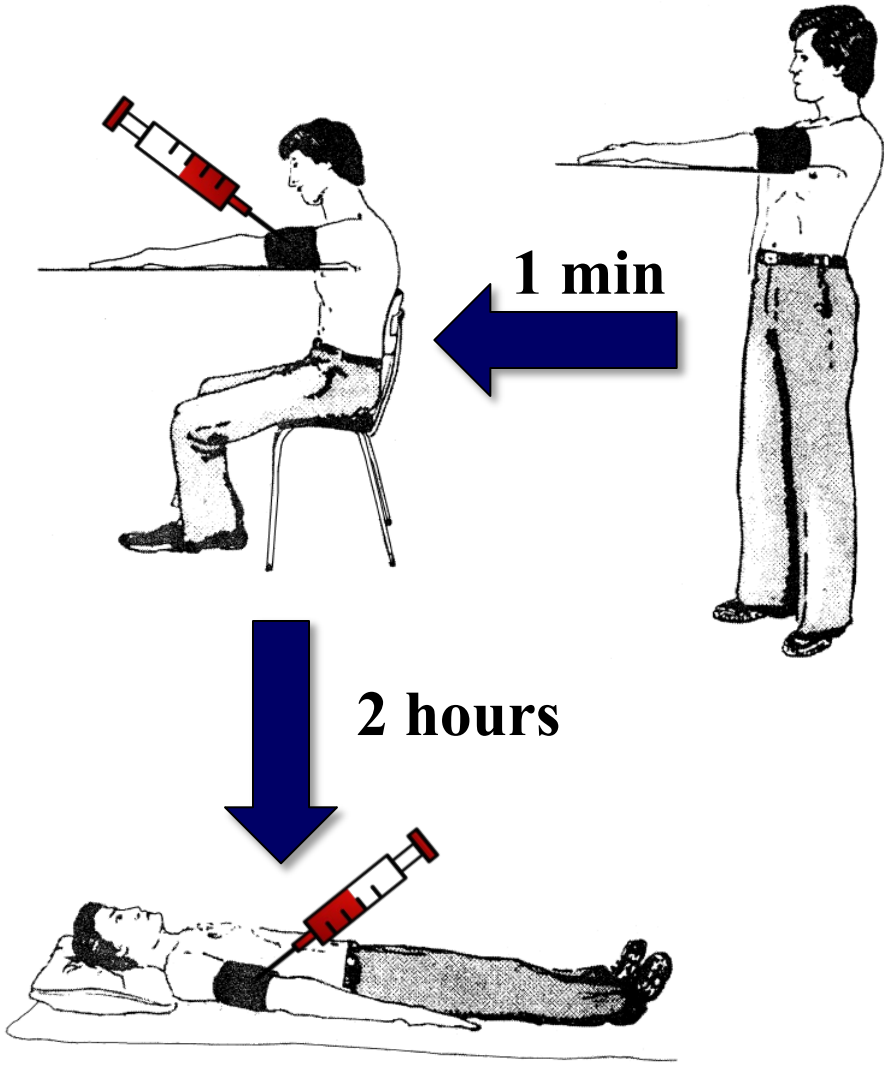
Case #5

- 55-year-old women
- Admitted to the ED at 1 AM for acute gastrointestinal pain lasting for 5 hours
- Blood sample immediately drawn upon arrival and transported to the laboratory
- Test results:
  - CRP, 1.2 mg/L
  - Hb, 7.3 mmol/L
  - WBC, 3.5 x 109/L
- Patient managed with "watch-and-wait" approach, waiting for ultrasound
- Second blood sample drawn after 2 h, in the ED observation unit
- Test results:
  - CRP, 1.0 mg/L
  - Hb, 6.6 mmol/L
  - WBC, 3.0 x 109/L
- Ultrasound negative, no clinical signs of bleeding, no other signs or symptoms

Scientific Event 2017

Tallinn 2017

- First blood sample drawn with only 1 min of stay in seated position
- Second blood samples drawn in supine position
- Plasma volume **changes up to 20% shifting from standing to supine position**



Case #6

- A 71-year-old man
- Admitted with left anterior cerebral artery hemorrhagic stroke
- Blood tests were never ordered
- Test results:
  - Procalcitonin, 4.4 ng/mL
  - CRP, 13.3 mg/L
  - WBC, 13.5 x 109/L
- Clinicians refuse to take action and order lab tests on this patient
- Blood sample drawn and transported to the laboratory
- Test results:
  - Procalcitonin, Not requested
  - CRP, 0.3 mg/L
  - WBC, 9.4 x 109/L

Bucurescu S, J Neurol Neurophysiol 2013, 4:5

Pre-analytical Laboratory Error in a Stroke Patient due to Blood Collection from another Stroke Patient: A Case Report

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Neurology at Klinikum Ansbach, Escherichstr. 1, 91522 Ansbach, Germany

• First blood sample drawn from **another 75-year-old same gender patient with right middle cerebral artery ischemic stroke, with a similar family name**, who was transferred the same day to the intensive care unit due to a nosocomial infection

Case #7

- A 5 months old infant hospitalized with lung dysfunction due to prematurity
- Routine zinc measurement reveals unexpected elevated zinc concentration: 20.2 µmol/L (ref. 10.0-19.0 µmol/L) compared to 11.6 µmol/L five days earlier
- When repeated some days later the zinc concentration are further increased to 42.4 µmol/L
- No clinical signs of increased zinc concentration
- Medication and nutrition supplements reveals no relevant zinc content

Pre-analytical mysteries

Elevated zinc concentrations in a 5 months old infant: A case report

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- The blood sample was obtained by capillary sampling
- The mother had applied vitamin E ointment containing zinc oxide at the infant's left heel
- A capillary sample obtained from the right heel revealed a totally normal zinc concentration
- Preanalytical contamination with ointments must be considered in unexpected measurements from capillary blood
- Ask the parents!
- Avoid unnecessary testing!!

Case #8

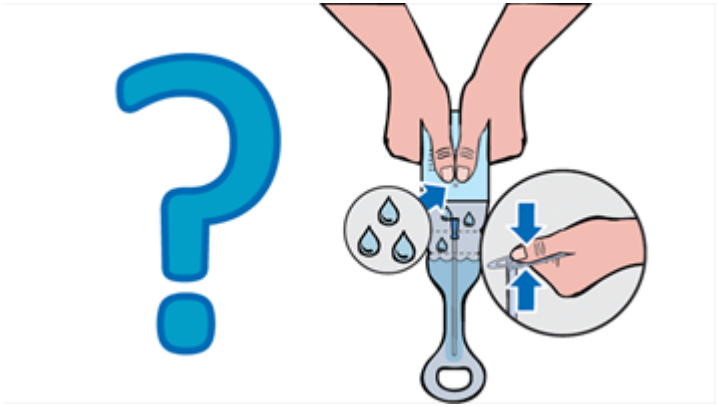
- 61-year old man with chronic kidney disease
- Undergoing maintenance haemodialysis
- Blood tests ordered:
- Test results:
  - Sodium, 182 mmol/L
  - Potassium, 4.8 mmol/L
  - Chloride, 87 mmol/L
- Second blood sample immediately drawn and transported to the laboratory
- Test results:
  - Sodium, 139 mmol/L
  - Potassium, 4.6 mmol/L
  - Chloride, 88 mmol/L

Biochemia Medica 2016;26(2):260–3

Pseudohypernatremia secondary to trisodium citrate (Citra-Lock™)

Janice Milliere[1], Daryl Corriveau[1], Malvinder S. Parmar*[1,2]

• First blood sample **contaminated during collection with trisodium citrate, a catheter-lock solution, commonly used in dialysis units** to maintain patency of dialysis catheters.



Case #9

10 months old boy admitted with hemoglobin 4.2 mmol/L and rectal bleeding

Coagulation parameters from Sysmex CS5100:

aPTT	> 300 seconds	(22 – 28 seconds)
INR	1.1	(normal)
Fibrinogen	5.5 µmol/L	(5.5 – 11.5 µmol/L)

Hemophilia A or B? Von Willebrand's disease? Heparinised sample?

KF VIII	1.81
KF IX	1.22
aPTT (STAR)	34 seconds

Interference in Coagulation Testing: Focus on Spurious Hemolysis, Icterus, and Lipemia

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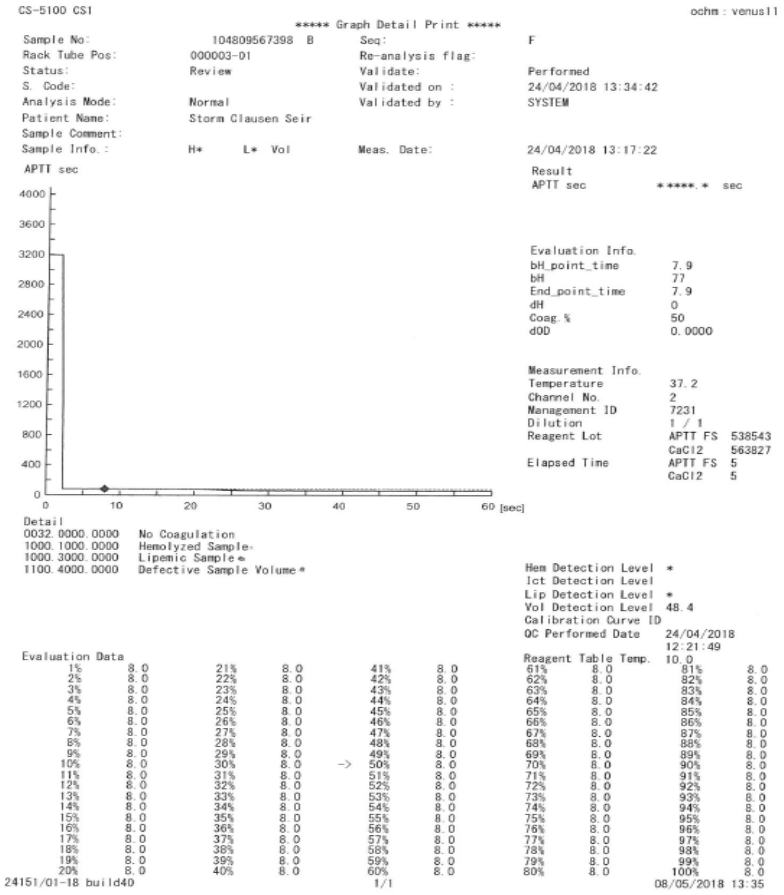
Semin Thromb Hemost 2013;39:258–266.

- **The blood sample was highly lipemic, but no one saw this in the automated solution**
- The curve from the CS5100 indicated “No coagulation”, which was interpreted as endless clotting time and an aPTT > 300 sec.
- If the curve had been inspected properly, the cause would have been obvious
- **Be aware of the “automated” interpretation algorithms**
- **Always inspect samples with unexpected results!**



Case #10

Two patients in the same room had their blood count and CRP – samples taken during the morning rounds. After the results were visible in the LIS system the nurse feared that the samples / responses were interchanged. The responses were compared to the patients' previous results and it was noted that for the other patient the CRP level had risen dramatically at the same time as the other patient's CRP had decreased strongly. New samples were drawn from both patients at the 11 o'clock round. The results of these samples confirmed that the previous samples were mixed up.

- The phlebotomist / nurse had left the samples unlabeled on the desk and worked on both patients at the same time.
- Mixing up the samples caused extra cost to the ward due to repeated samples.
- The other patient could not be discharged on time and the other patient received delayed care
- The staff had to do extra work and the incident caused bad publicity to the ward.

Case #11

A patient was on its way to surgery and had a nametag put on as alway. The phlebotomist had removed the nametag due to a troublesome phlebotomy and had to use the patient's both hands. Removal of the nametag was not informed to anyone and had been noticed only in the OR. At this stage the patient had already received premedications and was not able to inform on his/her ID number. New nametag was promised to be inserted in the OR.

- At this stage the patient had already received premedications and was not able to inform on his/her ID number.
- For several patient groups the nametag is the only unequivocal way of identifying the patient
- Could have serious effects if patient identified wrong in the OR

Case #12

A STAT Creatinine sample had been requested from a patient and the response should have come within one hour. The patient, a child was anesthesized for a MRI but the investigation had to be stopped since the creatinine response was delayed.

- The sample had been lost somewhere and had not been taken to the analyzer for analysis.
- The patient was subjected to prolonged anesthesia time.

Case #13

43-year-old man with known T2 Diabetes mellitus admitted to the ED with suspected diabetic ketoacidiosis. Severe metabolic acidosis was confirmed by the blood gas analyses:

- Arterial blood gas:
  ↓↓ pH 6,82
  ↓↓ Base excess -28.2 mmol/L (± 3)
  ↑ Anion gap 24.4 mmol/L (6–17)
  ↑ Glucose 33 mmol/L
  (↑) Creatinine 112 µmol/L
  (↑) Lactate 5,0 mmol/L (0-2,5)

- Venous blood test:
  ↓↓ Hb 5,0 g/dL (prehospital Hb was 17, later 12-13)
  ↑ Glucose 12,9 mmol/L (with the comment: lipemic sample, may give falsely elevated result)
  ↓↓ Creatinine 32 µmol/L
  ↑↑ Triglycerides 39,4 mmol/L
  ↑↑ P-Osmolality 637 (280–300)
  ↑↑ S-Phosphate 12,3 mmol/L (0,75–1,65)

Questions:
1. Why was Hb low, and why was creatinine and glucose lower in the venous sample than in the arterial blood gas?
2. Explanations for the elevated osmolality and phosphate?
3. Can lipemia explain these results?

1. Dilution of the venous blood sample from intravenous solution.
2. Contamination with the intravenous solution Tribonat
3. Many analyses are affected by lipemia, but not those mentioned above

The patient was treated with Tribonat, an intravenous solution used for treatment of metabolic acidosis. Tribonat contains hypertonic electrolyte concentrations with an osmolality of around 800 mosmol/kg and high phosphate concentration of 20 mmol/L. The venous blood sample was drawn from the same arm as the i.v infusion, which resulted in a ≈ 3:1- dilution of the blood sample with Tribonat, (roughly estimated from the differences in Hb, glucose and creatinine results). Sample dilution with Tribonat explains the low Hb, creatinine and glucose results in the venous sample, and also the highly elevated osmolality and phosphate concentration.

What about lipemia? Several other tests of this patient were difficult to interpret because of the lipemia, which added to the confusion of the blood test results. In fact, the triglycerides were also falsely low because of dilution, probably the real concentration of triglycerides was more than 100 mmol/L

Case #14

Two identical twins girls (22 years of age) were referred to genetic screening for familiar hypercholesterolemia. Their mother has a disease-causing variant in the LDL receptor gene resulting in familiar hypercholesterolemia. The twin girls had no clinical characteristics of cholesterol deposits but their LDL levels (6.5 and 6.8 mmol/L) were greater than the diagnostic criteria for definite familiar hypercholesterolemia (> 4.9 mmol/L).

The genetic screening of the twins girls revealed that one girl had no variants in the LDL receptor gene whereas the other girl had a heterozygous disease-causing variant in the LDL receptor gene.

- Some developing twin embryos may already have genetic differences, but in most cases, a pair of identical twins share the same DNA when they split. It therefore seems highly unlikely that the twins girls have genetic differences in the gene that codes for the LDL receptor.

- Based on suspicion of identification error, another blood sample procedure was performed from both girls and the genetic testing was repeated. This revealed a heterozygous disease-causing variant in the LDL receptor gene in both girls. We concluded that an identification error had occurred during the first phlebotomy procedure with the one twin girl who had no variants in the LDL receptor gene. Luckily, they were twins. Otherwise, we would not have detected the error. This would have caused a missed diagnosis of familiar hypercholesterolemia and potentially life-saving medical treatment.

Case #15

All chromatograms from our Tosoh G8 instrument (HPLC method) was visually inspected due to an outlier in the intern quality control programme. One of the chromatograms had an extra shoulder which was not seen or described before in the laboratory. The HbA1c value in the sample was 58.3 mmol/L. After consulting the laboratory doctor and clinical chemists, we decided to reanalyse the sample on an Afinion instrument (boronate affinity method) which revealed a HbA1c value of 30,0 mmol/L. Due to this discrepancy we measured fasting blood glucose and fructosamine which were normal. Gene sequencing did not reveal any suspicion of hemoglobinopathy. The patient had been on antidiabetic medication for several years. We could not find any immediate explanation for the different HbA1c results. We were given the permission to look into the patients medical journal. His HbA1c values were consistently elevated to levels compatible with diabetes mellitus for the last 5-6 years. We noticed that the patient had had several episodes of aspirin misuse.

- We assumed that the patient had falsely elevated HbA1c when measured with Tosoh G8 (HPLC method) as acetylated hemoglobin might be identified as glycated hemolgobin when measured with ion exchange HPLC and thus may falsely increase HbA1c. We tested this assumption and find that aspirin concentration of 7.0 mg/L did not have any significant effect on HbAc1 concentreations when comparing to samples without aspirin. Adding higher concentrations of aspirin (326 and 978 mg/L) increased HbA1c results in a dose-dependent manner with HPLC method.
- There was no difference between HbA1c measurements obtained with Afinion in samples with the highest aspirin concentration and Tosoh G8 in samples without aspirin.
- We concluded, that the patient did not have diabetes mellitus. He was "just" poisoned by aspirin. This case highlights a pre-pre- and preanalytical problem that is not paid much attention by neither laboratory nor clinical personal.

Case #16

Biotin case
A 27-year-old young man was referred to the Department of Endocrinology due to treatment-resistant blood pressure of 147/101 mmHg.

Aldosterone and renin were measured twice a few weeks apart. Aldosterone was measured at 3495 pmol/L and <102 pmol/L (cut-off <1197 pmol/L), and Renin at 4.9 kIU/L and 17.5 kIU/L, respectively (reference interval 5.3 – 99.1 kIU/L).
The first test result was compatible with primary hyperaldosteronism and the second was not.

- It emerged that the patient had taken biotin supplements before the first sample was taken, but not before the second sample. Biotin interference could be the reason for the surprising changes in results over a short period of time.
- Aldosterone was repeated at the primary laboratory (iSYS) and the sample was sent for control measurement in another laboratory, which uses a radioimmunoassay (RIA) in which biotin is not included in the assay. Aldosterone was measured at <102 pmol/L with iSYS and at 206 pmol/l with RIA. Previous method comparisons have shown that RIA measures about twice as high concentrations as iSYS.
- The patient in the case ended up being diagnosed with essential hypertension.
- Biotin's influence on iSYS assays was further investigated. It was confirmed that biotin gives falsely elevated values of aldosterone, even at concentrations of biotin corresponding to food supplements, and that renin measurement will be affected in a falsely low direction, but to a lesser extent
- Knudsen CS, Adelborg K, Søndergaard E, Parkner T. Biotin interference for aldosterone, renin, insulin-like growth factor 1, growth hormone and bone alkaline phosphatase. SJCLI. 2022;82:6-11.

Case #17

One of the elderly care wards in Uppsala had an unusual high proportion of patients with hyperpotassemia. Approximately half the patients in the ward were treated with resonium to reduce the hyperpotassemia. This was considered strange, but there were no hemolysis flags for the samples.

On closer inspection it was found out that the staff at the ward used to place the tubes in a refrigerator to preserve the samples better instead of storing the tubes at room temperature.

- The sodium-potassium pump requires energy and probably operates best at 37 C. At room temperature the sodium-potassium pump is slightly less effective and the function is even worse at +4 C. Storing samples at +4 C will lead to increased plasma potassium values.
- If the samples are stored at 37 C the values are slightly lower than tubes stored at room temperature.
- Identifying the problem and information on proper handling of tubes for potassium request solved the problem and the number of patients on resonium decreased.

Case #18

Ammonia (NH_3) is produced by cells throughout the body, especially the intestines, liver, and kidneys. Reference interval 11-48 umol/L for adults 18-50 years of age. Inborn errors of the amino acid metabolism (IEM) play an important role in the development of hyperammonemia (HA) in newborns. IEM is a heterogeneous group of disorders with complex clinical manifestations. Most patients with inherited HA exhibit non-specific symptoms, such as poor feeding, lethargy, dyspnea, or hypothermia, which may progress to convulsions or coma.

Uppsala received a request for ammonia testing on a newborn boy from another hospital. The ammonia value was approximately 400 umol/L. The result was reported back to the pediatric department at that hospital. This strengthened their suspicion of an IEM and they started a IEM investigation and at the same time the boy was put on a protein restricted diet. Over the next months we received several follow up samples that all had similar high ammonia values as the first one despite further reductions in the protein intake. After approximately 6 months the boy was transferred to the Pediatric University Hospital here in Uppsala. One of the first investigations that the pediatricians performed was retesting for ammonia. The sample sent was within the reference value.

- Measurement of ammonia is problematic as it rapidly increases with storage in whole blood and also increases with storage in separated plasma. Thus, we initially considered this as the cause of the false hyperammonemia.
- Approximately three months later the pediatricians at the hospital where the boy was originally treated called and said that they had observed that the tubes that they had been using had a small text saying that it was ammonia heparin in the tubes!
- We normally use sodium or lithium heparin tubes and the region has never purchased ammonia heparin tubes. Where the tubes in the pediatric ward came from is unknown.

Case #19

To our central laboratory, we received a Li-heparin vacutainer tube for analysis of various common parameters, including potassium. The result: >13 mmol/L.

- When contacting the physician responsible, the head of a medicine department, they explain taking blood from a "purple tube" (a K-EDTA tube) and poured into the Li-hep tube we received.
- The reasoning: "You and your laboratory's bureaucratic propensity for artificial rules make our work more difficult. I just assumed that the color of the stopper serves to facilitate the sorting of the tubes when it arrives at your lab, or something similar."
- We explained that the tubes contain different additives, and that the purple one in particular contains potassium.
- This case illustrates, not only an example of a common source of preanalytical error that the person responsible for may not always admit as readily as this time, but it also illustrates lack of knowledge related to sampling instructions.

Case #20

Multiple times potassium could not be measured reliably from a Li-heparin plasma because of in vitro hemolysis.

- The patient had immunoglobulins attaching to their red cells that caused the cells to be very fragile.
- Serum is not commonly used for potassium in our laboratory, but we tried it as all Li-heparin plasma samples had too much hemolysis.
- In the serum sample there was no significant hemolysis.