Haemolysed or Clotted samples – Preventable Causes of Unsuitable Samples

Haemolysed samples

Haemolysis = release of haemoglobin and other intracellular components from red blood cells (RBC's) into plasma.

Haemolysis index is reported in g/L.

Haemolysed samples are the commonest cause for unsuitable for analysis samples. Haemolysis interferes with testing in many ways:

Release of cellular content - Potassium, Folate, Magnesium, Phosphate, LDH, AST

Release of enzymes that degrade analytes - Insulin, Troponin T

Release of enzymes that interfere with assay - CK

Optical and or chemical interference – large list of analytes.

Inhibition of reactions – PCR based assays on viral and bacterial DNA and RNA

Measuring haemolysis

In the past, samples were visually checked for haemolysis after centrifugation (pink or red serum/plasma). Grossly haemolysed samples were quickly identified and requestors were informed that a recollect was required, however visual assessment missed lesser degrees of haemolysis.

Centrifugation is now automated; scientists do not look at the samples. Haemoglobin is measured by the main biochemistry analyser at the same time as the tests requested. Identification of haemolysed samples is now more reliable but occurs much later.

Not all assays are equally sensitive to interference by haemoglobin. The haemoglobin result is used in an assay specific way to determine whether to report the result or. When haemoglobin is > 10 g/L (= grossly haemolysed), no tests are reported.

Causes of haemolysis

The laboratory does not haemolyse samples. It is possible to avoid most of the haemolysed samples received by the laboratory. The sample can be haemolysed at any step from sample collection to receipt in the laboratory.

The risk of haemolysis increases with:

• Collection using a syringe.

Pulling or pushing on the plunger exerts pressure on RBC's, causing RBC's to break.

• Collecting through an IV catheter.

The risk for haemolysis is significantly larger compared with collecting in to a vacutainer through a straight needle. Usually a syringe is used.

• Collecting from sites other than "antecubital area"

The antecubital area veins are larger, making it easy to access, are less likely to collapse and allows use of larger needles. Haemolysis is more common with difficult collects – veins on other sites are smaller and/or fragile and therefore easily traumatized.

• Excessive shaking of the tube.

Samples should be inverted gently 10 times to mix with additives. Shaking will damage RBC's.

• Needle size.

There is a lack of good evidence in literature but it is claimed that using a large needle (< 21 gauge) causes haemolysis, due to rapid entry of blood in the tube. The RBC's are traumatised by hitting the bottom of the tube. With small needles (> 21 gauge), the shear stress upon entering the vacuum tube breaks the RBC's.

• Capillary collects

The pressure applied by squeezing can cause extreme hydraulic pressure in capillaries, breaking RBC's

• Transport (rare cause) by Lamson

Acceleration may traumatise red cells. There is more "sloshing" when tubes are incompletely filled. Some patients have more "fragile" RBC's, if samples are repeatedly haemolysed, delivery by foot should be considered.

• Other causes include – extended tourniquet time (more than 2 mins), not allowing the antiseptic used for collection to dry, transport conditions (time, temperature), centrifugation speed, re-spun of samples. Taking blood from a bruise and the patient pumping can cause haemolysis.

Most causes of haemolysis are preventable

Suboptimal sample collection technique causes the majority of haemolysed samples. Training is a "quality improvement" that benefits all, with no capital spending required. Haemolysed samples requiring recollection waste the time of the collector, is unpleasant for the patient and delays the management of the patient.

A small % of patients have haemolytic conditions. This can be challenging for all involved in testing.

The Laboratory Quality Manager monitors haemolysis rates from various areas and takes action as and when required.

Clotted samples

Causes include:-

- Blood slow to fill the tube
- Prolonged use of a tourniquet

- Samples incompletely mixed
- Syringe collect and slow transfer of sample
- Not enough blood in the tube.

Haematology rejects clotted samples because all results are unreliable.

Biochemistry – clots are not always detected and can cause falsely low or high results.

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