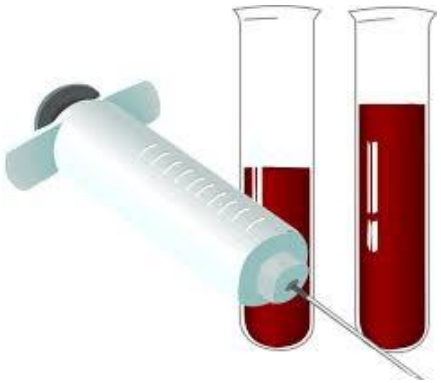
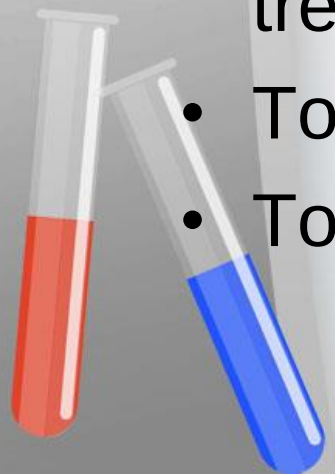
Laboratory Data



Introduction:

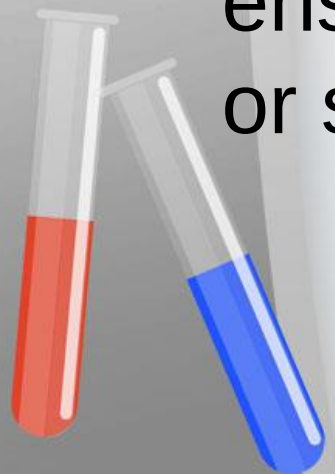
Biochemical and haematological tests provide useful information:

- To confirm or support diagnostic and for screening
- To asses the severity of a disease (prognosis)
- To monitor of disease and its response to treatment
- To monitor appropriate drug dosing
- To help prevent toxic S/E & interactions



Reference ranges

- Results ca be classified as + or –
- A cut-off point
- They represent the test values from 95% of the healthy population (mean ± 2 SD).
- A series of values, is often required to ensure clinical relevance (avoid analytical or sampling errors).



- Specimen commonly analyzed in lab tests are whole blood, serum or plasma, urine & fluid. Reference range for a substance in one body fluid can be different from that in another
- Reference ranges can differ significantly with various measurement methods

Haematology data:

RBC count:

- RBCs are produced in the bone marrow by the process of erythropoiesis. One of the major stimulants of this process is erythropoietin, produced mainly in the kidney.
- The lifespan of a mature red cell is usually about 120 days.

erythroblasts

↓

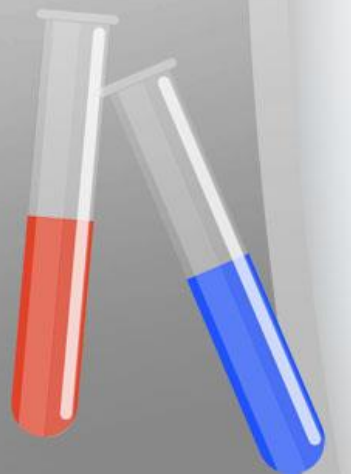
normoblasts (nucleated)

↓

reticulocytes (non-nucleated)

↓

erythrocytes

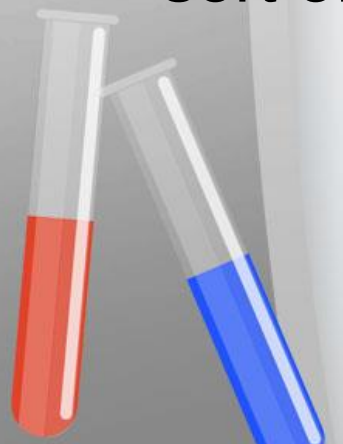


Reticulocytes

- The normal range 0.5% and 1.0% of the total RBC (do not feature significantly in a normal blood profile).
- increased production (reticulocytosis) can be detected in times of rapid red cell regeneration as occurs in response to haemorrhage (may reach 40% of RBC).
- The reticulocyte count may be useful in assessing the response of the marrow to iron, folate or vitamin B12 therapy.

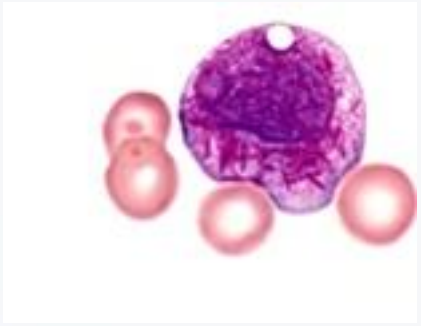
Packed cell volume (PCV)

- The PCV or haematocrit is the ratio of the volume occupied by red cells to the total volume of blood.
- It can be measured by centrifugation of a capillary tube of blood and then expressing the volume of red cells packed in the bottom as a percentage of the total volume.
- The PCV often reflects the RBC (decrease in any sort of anaemia, raised in polycythaemia).



Mean cell volume (MCV)

- The MCV is the average volume of a single red cell. It is measured in femtolitres
- Terms such as ‘microcytic’ and ‘macrocytic’ are descriptive of a low and high MCV, respectively.
- They are useful in the process of identification of various types of anaemias such as caused by iron deficiency (microcytic) or vitamin B12 or folic acid deficiency (megaloblastic or macrocytic).

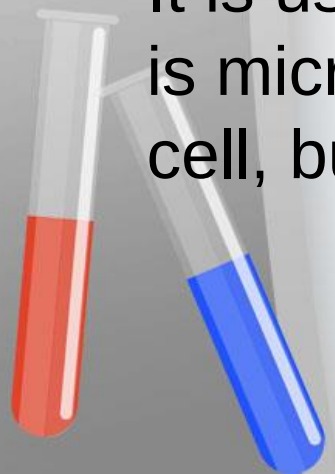


Mean cell haemoglobin (MCH)

- The MCH is the average weight of haemoglobin contained in a red cell. It is measured in picograms and is calculated from the relationship:

$$\text{MCH} = \frac{\text{Haemoglobin}}{\text{RBC}}$$

- It is usually low in iron-deficiency anaemia when there is microcytosis and there is less haemoglobin in each cell, but it may be raised in macrocytic anaemia.

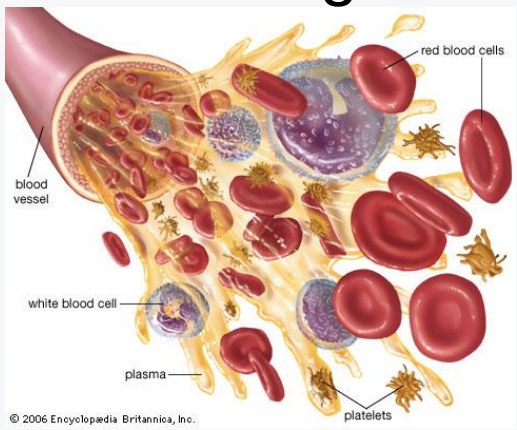


Mean cell haemoglobin concentration (MCHC)

- The MCHC is a measure of the average concentration of haemoglobin in 100 mL of red cells. It is usually expressed as grams per litre but may be reported as a percentage.
- The MCHC will be reported as low in conditions of reduced haemoglobin synthesis, such as in iron-deficiency anaemia. The MCHC can be raised in severe prolonged dehydration.

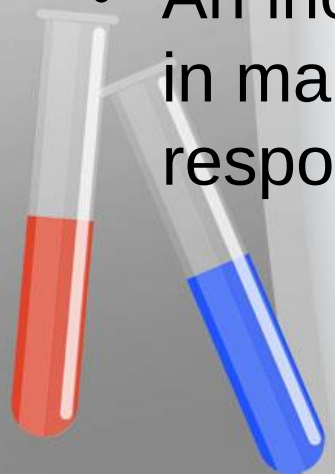
Haemoglobin

- The haemoglobin concentration in men is normally greater than in women (menstrual loss).
- Haemoglobin is most commonly measured to detect anaemia.
- Alterations in the structure of the haemoglobin molecule can be detected by electrophoresis (rare genetic diseases, e.g. HbS (sickle haemoglobin in sickle cell disease)).



Platelets (thrombocytes)

- Platelets are formed in the bone marrow.
- Platelets are normally present in the circulation for 8–12 days.
- A small fall in the platelet count may be seen in pregnancy and following viral infections. Severe thrombocytopenia may result in spontaneous bleeding.
- An increased platelet count (thrombocytosis) occurs in malignancy, inflammatory disease and in response to blood loss.



White blood cell (WBC) count

WBC (leucocytes) are of two types: the granulocytes and the agranular cells. They are made up of various types of cells with different functions.

1- Neutrophils or polymorphonucleocytes (PMNs)

- approximately 40–70% of WBC.
- Their lifespan is 10–20 days.
- The neutrophil count increases in the presence of infection, tissue damage (e.g. infarction) and inflammation (e.g. rheumatoid arthritis, acute gout).
- Neutropenia is associated with malignancy, drug toxicity and viral infections (influenza, and hepatitis).

2- Basophils

- Basophils normally constitute a small proportion of the white cell count.
- basophilia occurs in various malignant.

3- Eosinophils

- Eosinophils constitute normally less than 6% of white cells.
- Their function appears to be concerned with inactivation of mediators released from mast cells (many allergic conditions such as asthma and drug sensitivity reactions).

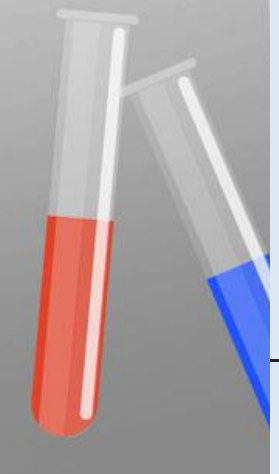
4- Lymphocytes

- Lymphocytes are the second most abundant WBC.
- They are formed in the bone marrow and founded in the spleen and other lymphatic tissue.
- increased particularly in viral infections.

5- Monocytes

- Monocytes are macrophages.
- Their numbers increase in some infections such as typhoid.

Haemoglobin	11.5–16.5 g/dL
Red blood cell (RBC) count	3.8–4.8 × 10^{12}/L
Reticulocyte count	50–100 × 10^{9}/L
Packed cell volume (PCV)	0.36–0.46 L/L
Mean cell volume (MCV)	83–101 fL
Mean cell haemoglobin (MCH)	27–34 pg
Mean cell haemoglobin concentration (MCHC)	31.5–34.5 g/dL
White cell count (WBC)	4.0–11.0 × 10^{9}/L
Differential white cell count:	
Neutrophils (30–75%)	2.0–7.0 × 10^{9}/L
Lymphocytes (5–15%)	1.5–4.0 × 10^{9}/L
Monocytes (2–10%)	0.2–0.8 × 10^{9}/L
Basophils (<1%)	<0.1 × 10^{9}/L
Eosinophils (1–6%)	0.04–0.4 × 10^{9}/L
Platelets	150–450 × 10^{9}/L



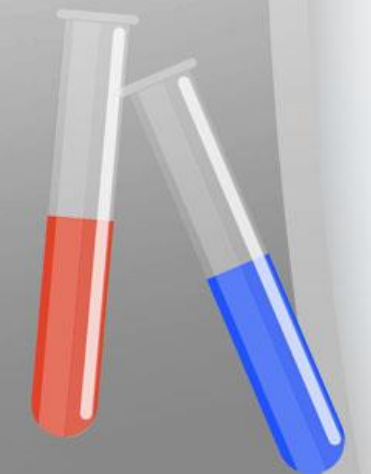
Other blood tests:

Erythrocyte sedimentation rate (ESR)

- The ESR is a measure of the settling rate of red cells in a sample of anticoagulated blood, over a period of 1 h, in a cylindrical tube.
- The normal values do rise with age.
- The test is principally used to monitor inflammatory disease.
- The ESR is non-specific and, therefore, serial tests can be helpful in following the progress of disease, and its response to treatment.

C-reactive protein (CRP)

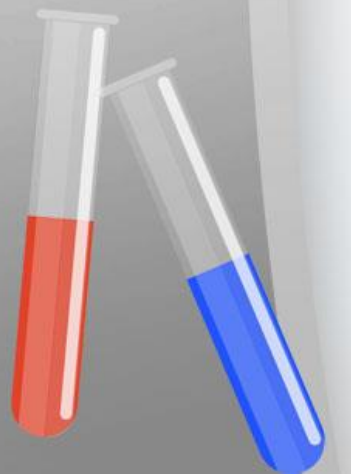
- Production of CRP is rapidly and sensitively upregulated, in hepatocytes, under the control of cytokine (IL-6) originating at the site of pathology.
- CRP values are not diagnostic, it reflects ongoing inflammation or tissue damage more accurately than do other acute-phase parameters such as the ESR



Monitoring anticoagulant therapy

PT

- Measuring the PT is the most commonly used method for monitoring oral anticoagulation therapy.
- The PT is measured by adding calcium and thromboplastin

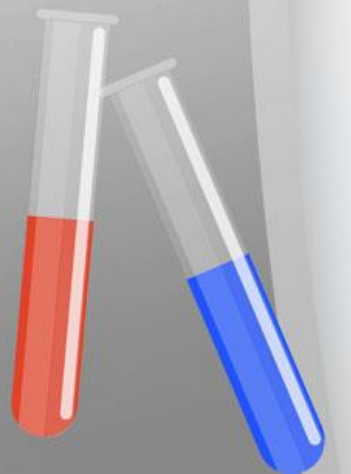


International Normalised Ratio (INR)

•The results of the test are commonly expressed as a ratio of the PT time of the patient compared with that of the normal control. This is known as the INR, a system used to standardise reporting worldwide.

• The most common use of the PT and INR is to monitor oral anticoagulant therapy,

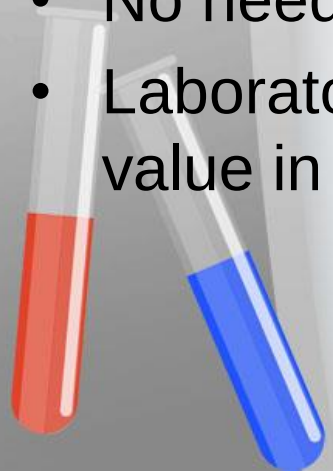
•The target value varies according to the indication for the anticoagulant.



$$\mathrm{INR} = \left\{ \frac{\text{Patient's PT}}{\text{Control PT}} \right\}^{\mathrm{ISI}}.$$

Activated partial thromboplastin time (APTT)

- For monitoring unfractionated heparin therapy.
- A thromboplastic reagent is added to an activator such as activated silicone or kaolin. The mixture of thromboplastin and activator is mixed with citrated plasma to which calcium is added, and the time for the mixture to clot is recorded.
- Low molecular weight heparins are effective and safe for the prevention and treatment of VTE.
- No need to monitor the APTT during treatment.
- Laboratory monitoring of an anti-factor Xa assay may be of value in certain clinical settings (renal insufficiency).



Iron, transferrin and iron binding

- Iron is important in cells producing haemoglobin.
- It is circulating in the serum bounding to transferrin (protein with two iron binding sites).
- Measurement of total iron binding capacity (TIBC), from which the percentage of transferrin saturation with iron may be calculated, saturation of 16% or lower is usually taken to indicate an iron deficiency, as is a raised TIBC of greater than 70 μmol/L.

Ferritin

- Is an iron storage protein found in cell cytosol.
- Serum ferritin measurement is the test of choice in patients suspected of having iron deficiency anaemia.
- Ferritin is an acute-phase protein and levels may be normal or high in the anaemia of chronic disease, such as occurs in rheumatoid arthritis or chronic renal disease.
- Iron overload causes high concentrations of serum ferritin, as can liver disease and some forms of cancer.

Vitamin B12 (cyanocobalamin) and folate

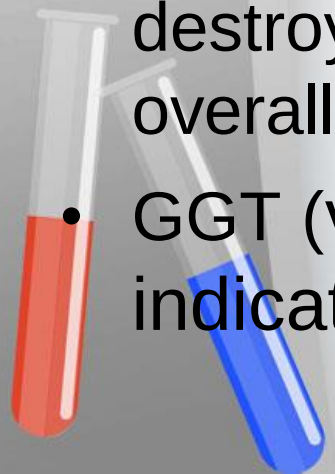
- Deficiency of cobalamin can result both in anaemia, usually macrocytic, and neurological disease.
- Folate deficiency produces anaemia, macrocytosis, depression, dementia and neural tube defects.
- malabsorption of B12 may result from long-term ingestion of antacids such as PPI or H2-receptor antagonists or biguanides (metformin).

Erythrocyte sedimentation rate (ESR)	1–35 mm/h
Ferritin	15–300 µcg/L
Total iron binding capacity (TIBC)	47–70 µmol/L
Serum B_{12}	170–700 ng/L
Red cell folate	160–600 µcg/l
Iron	11–29 µmol/L
Transferrin	1.7–3.4 g/L

Biochemical data

Liver function tests (LFTs)

- Serum albumin levels and prothrombin time (PT) indicate hepatic protein synthesis.
- Albumin has an important role in binding, among others, calcium, bilirubin and many drugs.
- Bilirubin (breakdown product released when RBC are destroyed at the end 120-day lifespan) is a marker of overall liver function.
- GGT (γ-Glutamyl transpeptidase) It is a sensitive indicator of hepatobiliary disease



- Transaminase levels (ALT, AST) indicate hepatocellular injury and death.
- Alkaline phosphatase levels estimate the amount of impedance of bile flow and also reflect bone building or osteoblastic activity

Liver function tests	
Albumin	34–50 g/L
Bilirubin (total)	<19 µmol/L
Enzymes	
Alanine transaminase	<45 U/L
Aspartate transaminase	<35 U/L
Alkaline phosphatase	35–120 U/L
γ-Glutamyl transpeptidase	<70 U/L

Cardiac markers

Troponins (I, T) proteins

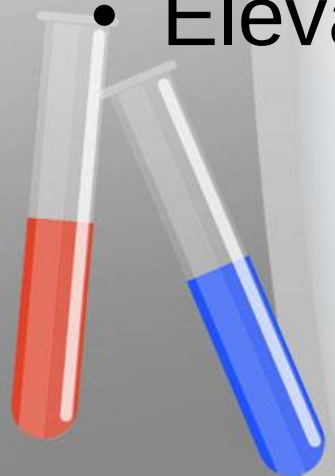
- The preferred biomarker for myocardial necrosis as they have near absolute myocardial tissue specificity as well as high clinical sensitivity.

Creatine kinase (CK)

- An enzyme which is present in relatively high concentrations in heart muscle, skeletal muscle and in brain.
- CK has two protein subunits, M and B, which combine to form three isoenzymes, BB, MM and MB (MB Cardiac tissue).

Lactate dehydrogenase (LD)

- Lactate dehydrogenase has five isoenzymes (LD1–LD5). Total LD activity is rarely measured because of the lack of tissue specificity.
- Elevated following damage



Renal function tests

Creatinine

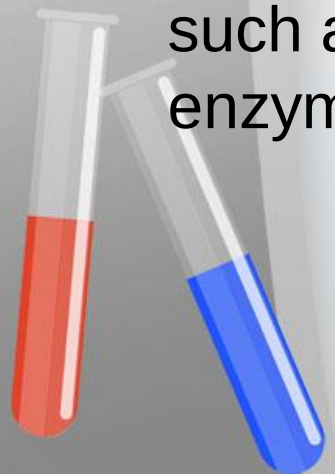
- is a breakdown product of muscle, it is cleared by the kidney, but only reabsorbed & secreted in small amounts
- Creatinine clearance can be used a measure of glomerular filtration rate, and therefore, renal function
- Creatinine levels vary according to a person's size and muscle mass.
- Some drugs including trimethoprim and cimetidine inhibit creatinine secretion, reducing creatinine clearance and elevating serum creatinine without affecting the GFR.

Urea

- Is a breakdown product of protein.
- Most renal disease affect urea excretion, so BUN increase in the blood.
- Patients who are dehydrated can also have abnormal BUN & many drugs affect BUN, mainly by competing with it for renal elimination.
- Serum urea is a less reliable marker of GFR than creatinine.
- Urea levels vary widely with diet.

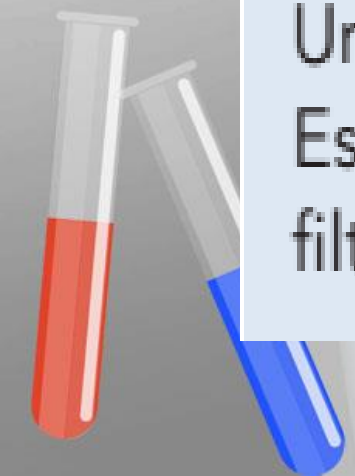
Electrolytes

- They affect a lot of function in the body (e.g renal function), and are affected by many drugs.
- Sodium depletion: dehydration or volume depletion.
- Sodium excess: primary mineralocorticoid excess, hyperaldosteronism.
- Hypokalaemia: Parenteral insulin, laxatives, Mineralocorticoid excess.
- Hyperkalaemia: renal failure, potassium-sparing diuretics such as spironolactone with an angiotensin converting enzyme (ACE) inhibitor.



Urea and electrolytes

Sodium	135–145 mmol/L
Potassium	3.4–5.0 mmol/L
Calcium (total)	2.12–2.60 mmol/L
Calcium (ionised)	1.19–1.37 mmol/L
Phosphate	0.80–1.44 mmol/L
Magnesium	0.7–1.00 mmol/L
Creatinine	75–155 µmol/L
Urea	3.1–7.9 mmol/L
Estimated glomerular filtration rate (eGFR)	≥ 90 ml/min/1.73m^2



Glucose

- Normal ranges for serum glucose concentrations are often quoted as non-fasting (<11.1 mmol/L) or fasting (3.3–6.0mmol/L) concentration ranges.
- Fasting serum glucose levels between 6.1 and 7.0 mmol/L indicate impaired glucose tolerance.
- When symptoms are typical of diabetes, a fasting level above 7.0 mmol/L or a 2 h post-glucose or random serum glucose level ≥11.1 mmol/L is consistent with a diagnosis of diabetes.

Glycated haemoglobin

- Glucose binds to a part of the haemoglobin molecule to form a small glycated fraction.
- Normally, about 5% of haemoglobin is glycated, but this amount is dependent on the average blood glucose concentration over the lifespan of the red cells (about 120 days).
- Measurement of HbA1C is well established as an indicator of chronic glycaemic control in patients with diabetes.

Tumour markers

- They may be detected within malignant cells and metastases.
- only a few markers contribute to the diagnosis of cancer, serial measurements can be useful in assessing the presence of residual disease and response to treatment.

Therapeutic drug monitoring

Drug	Therapeutic level (adults)	Sample carrier	Sample time
Digoxin	1–2 nanomol/L (0.5–2 mcg/L)	SST/PST heparin	6–8 h post-dose
Phenytoin	40–80 mcmol/L (10–20 mg/L)	SST/PST heparin	At any point
Theophylline	55–110 mcmol/L (10–20 mg/L)	PST heparin	Pre-dose
Carbamazepine	35–50 mcmol/L (5.0–12 mg/L)	SST/PST heparin	Pre-dose
Gentamicin	Trough < 2; peak > 5 (t.d.s. dosing)	SST/PST heparin	Pre-dose or 2 h post-dose
Vancomycin	Trough 5–10 mcg/L	SST/PST heparin	Pre-dose
Lithium*	0.5–1.0 mmol/L	SST	12 h post-dose

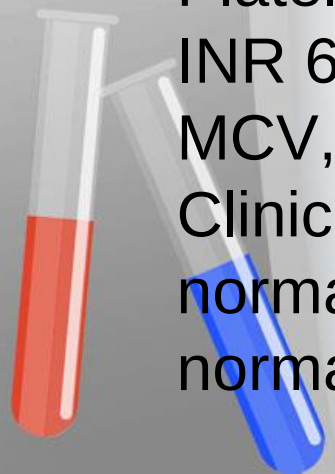
PST, plasma separated tube; SST, serum separated tube.
* Do not use a lithium heparin tube for lithium assays.

Drug	Monitoring
Acetaminophen (>4 g/day)	Hepatic function tests
Aminoglycosides	Serum creatinine, drug levels
Hypoglycemic agents	Blood sugar levels
Antiepileptic agents (older)	Drug levels
Angiotensin-converting enzyme inhibitors	Potassium levels
Antipsychotic agents	Extrapyramidal adverse effects
Appetite stimulants	Weight, appetite
Digoxin	Serum creatinine, drug levels
Diuretic	Potassium levels
Erythropoiesis stimulants	Blood pressure, iron and ferritin levels, complete blood count
Fibrates	Hepatic function test, complete blood count
Iron	Iron and ferritin levels, complete blood count
Lithium	Drug levels
Niacin	Blood sugar levels, hepatic function tests
Statins	Hepatic function tests
Theophylline	Drug levels
Thyroid replacement	Thyroid function tests
Warfarin	Prothrombin time/international normalized ratio

Case 1

A 70-year-old man on a hospital medical ward has a fast pulse rate and falling blood pressure. His recent drug history is warfarin as thrombo-embolic prophylaxis for chronic atrial fibrillation and erythromycin for a recent chest infection. He has vomited a moderate quantity of blood. Haematology results:

Hb 8.8 g/dL
RBC 4.7 × 10^{12}/L
Platelets 570 × 10^{9}/L
INR 6.0
MCV, MCH and the rest of the blood profile are normal
Clinical biochemistry: Urea 11.6 mmol/L, Creatinine is normal and sodium and potassium concentrations are normal.

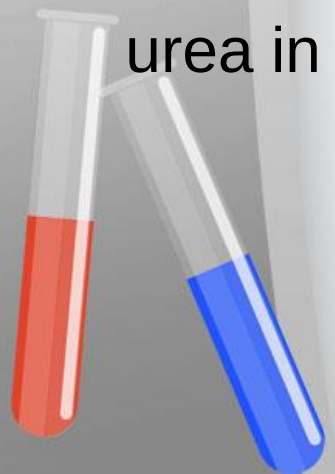


Questions

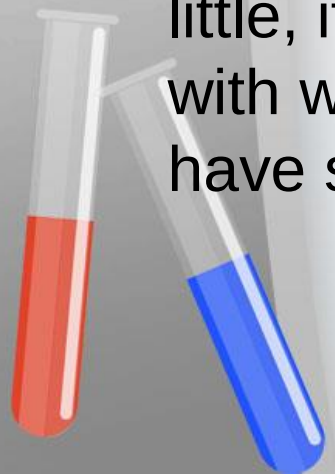
1. What is the cause of this patient's low haemoglobin?
2. What is the likely cause of his raised urea level?
3. What might have contributed to his over-anticoagulation as evidenced by his INR?

Answers

1. The cause of his low haemoglobin is a gastro-intestinal bleed. The picture is one of blood loss, manifested by a loss of red cells and haemoglobin. The red cells are of normal size and colour.
2. A raised urea in the presence of a normal creatinine may signify dehydration or gastro-intestinal bleeding. In this case, given the blood picture, the latter is more likely. Blood in the gastrointestinal tract is a source of protein which will be absorbed into the hepatic portal system and converted to urea in the liver.

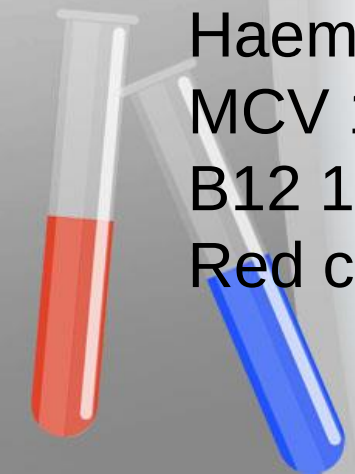


3. Erythromycin inhibits the cytochrome P450 system, particularly the CYP3A4 isoenzyme. CYP1A2 and CYP3A4 are the main enzymes for the inactivation of (R)-warfarin. Erythromycin, therefore, potentiates warfarin's action. The patient has been ill and in hospital and, therefore, his recent intake of vitamin K containing foods, for example, green leafy vegetables may well have been lower than is usual for him. Antibiotics can reduce synthesis of vitamin K by gut bacteria but this has little, if any, effect upon anticoagulation, and interactions with warfarin previously attributed to this mechanism have since been attributed to other modes of interaction.



Case2

An 83-year-old man presents with a few weeks history of tiredness, unsteadiness and an abnormal sensation in both hands and feet. On examination he is ataxic with poor co-ordination due to absent joint position and vibration sense. Clinical diagnosis is of sensory ataxia due to dorsal column pathology. His drug treatment includes long-term metformin for Type 2 diabetes mellitus and lansoprazole. Haematology results show:
Haemoglobin 8.9g/dL
MCV 110fL
B12 128ng/L
Red cell folate 300µcg/L



Questions

1. What is the likely cause of this patient's symptoms and signs?
2. How should his neurological features be investigated?
3. What term describes this type of anaemia?
4. What drug treatment should he receive?

Answers

1. Subacute combined degeneration of the spinal cord and anaemia due to vitamin B12 deficiency.
2. Magnetic Resonance Imaging (MRI) of the spine. This is likely to show signal abnormality in the posterior columns of the spinal cord.
3. Macrocytic anaemia.
4. Parenteral administration, usually by intramuscular injection of vitamin B12. In the UK, several loading doses are given followed by maintenance injections for the patient's lifetime. Treatment with pharmacological doses of oral cyanocobalamin is occasionally given, for example, if the patient has needle phobia or is allergic to the IM B12 preparation