Haematology

Dr Tom Bate

Again!
Basic sciences to underpin anaesthetic practice

Learning Outcomes:

➢ To gain a good understanding of human anatomy relevant to the safe practice of anaesthesia at basic level and to support progress to intermediate level training
➢ To acquire a sound understanding of human physiology, biochemistry and pharmacology, and to be able to apply this to clinical practice at a basic level and to support progress to intermediate training.
➢ To gain a good understanding of the basic principles of physics and clinical measurement; emphasis is on the function of monitoring equipment, equipment safety, and measurement techniques.

NB: All competencies annotated with the letter ‘E’ can be examined in any of the components of the Primary examination identified in the FRCA examination blueprint on page B-99 or in the Final examination identified in the Final FRCA blueprint on page C72 of Annex C.
### What you need to know

| PB_BK_17  | Blood: physical properties, components, functions | A,C,E | 1 |
| PB_BK_18  | Red blood cells: production and turnover, haematins, haemoglobin and its variants including abnormal haemoglobins eg thalassaemia, HbS | A,C,E | 1 |
| PB_BK_19  | Anaemia: acute and chronic adaptations – Iron absorption, transportation, metabolism | A,C,E | 1 |
| PB_BK_20  | Polycythaemia: causes and implications | A,C,E | 1 |
| PB_BK_21  | Blood groups: ABO, Rhesus, others | A,C,E | 1 |
| PB_BK_22  | Transfusion reactions; rhesus incompatibility | A,C,E | 1 |
| PB_BK_23  | Haemostasis and coagulation, fibrinolysis – including abnormalities, congenital and acquired | A,C,E | 1 |
| PB_BK_24  | Alternative oxygen carrying solutions | A,C,E | 1 |
| PB_BK_25  | White blood cells: types, origins, characteristics, turnover | A,C,E | 1 |
| PR_BK_49  | Antiplatelet agents. Perioperative management of antiplatelet medication | A,C,E | 1,2 |
| PR_BK_50  | Pro-coagulants: Drugs. Individual factor concentrates; multi-factor preparations including FFP; vitamin K | A,C,E | 1,2 |
| PR_BK_51  | Colloids, including blood and blood products: Composition of preparations; safe use and avoidance of errors | A,C,E | 1,2 |
Blood Groups

- ~30 blood group systems recognised (ISBT)
  - ABO, Rhesus, Lewis, Kell, Duffy, Kidd
  - MNS system, Kell system
- The ABO antigens are the most abundant on the red cell membrane and the most important in transfusion medicine
- Serum contains antibodies which are IgM molecules which rapidly agglutinate cells which contain the antigens they are directed against
Inheritance of Blood Groups

- Chromosome 9q34
- 3 alleles A, B, O
- A and B are co-dominant over O
- O produces no antigens and is recessive
- 4 phenotypes
  - A (Genotypes AA, AO)
  - B (Genotypes BB, BO)
  - AB (Genotype AB)
  - O (Genotype OO)
<table>
<thead>
<tr>
<th>Blood Group (UK incidence)</th>
<th>Antigens on RBC</th>
<th>Antibodies in Serum</th>
</tr>
</thead>
<tbody>
<tr>
<td>A (42%)</td>
<td>A</td>
<td>Anti-B</td>
</tr>
<tr>
<td>B (8%)</td>
<td>B</td>
<td>Anti-A</td>
</tr>
<tr>
<td>AB (3%) Universal Recipient</td>
<td>A &amp; B</td>
<td>Neither</td>
</tr>
<tr>
<td>O (47%) Universal Donor</td>
<td>Neither</td>
<td>Anti-A &amp; Anti-B</td>
</tr>
</tbody>
</table>
Rhesus Blood Group System

- ‘Rhesus positive’ refers to cells that carry the D antigen

- Unlike ABO system there are no ‘naturally occurring’ antibodies. Antibodies are only produced on exposure to the appropriate Rh antigens (i.e. blood transfusion or foeto-maternal haemorrhage)

- Inherited according to simple Mendelian genetics
<table>
<thead>
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<th></th>
<th>5g/dl</th>
<th>Essential</th>
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<tbody>
<tr>
<td></td>
<td>&lt;7</td>
<td>Transfuse</td>
</tr>
<tr>
<td></td>
<td>8 – 10</td>
<td>Safe</td>
</tr>
<tr>
<td></td>
<td>&gt;10</td>
<td>No transfusion</td>
</tr>
</tbody>
</table>

Symptomatic | Transfuse
Haemoglobin

- Normal Haemoglobin
  - Tetramer consisting of 4 globin chains, each combined with one haem moiety
  - Each molecule contains 2α chains and 2 non-α chains

Structure of haemoglobin

Each erythrocyte (RBC) contains ~270 million haemoglobin molecules
Haemoglobin

- RBC normally contain 3 types of Hb
  - Hb-A (95%) $\alpha_2\beta_2$
  - Hb-A$_2$ (2.5-3.5%) $\alpha_2\delta_2$
  - Hb-F (<2%) $\alpha_2\gamma_2$ : protects against SCD
Sickle Cell Disease

- Single base mutation: glutamine for valine at position 6 of the $\beta$ globin chain
- HbS has normal $O_2$ carrying capacity but with deoxygenation: polymerises - crystal-like rods - inflexible sickle-shaped erythrocytes
- Haemolysis and organ damage
- HbAS; HbSS; HbSC/HbS$\beta$-thal
Sickle Cell Disease

- Low Hb (6-9)
- Blood film: reticulocytes, sickle and target cells
- Screening tests
- Electrophoresis
- Exchange transfusions (<30% HbS)

**AVOID**
- Dehydration
- Hypoxia
- Hypothermia
- Acidosis

**PROVIDE**
- Pain control (RA)
- Antibiotics
Thalassaemias

- Partial or complete failure of synthesis of one or more of the globin chains, with overproduction of the normal chains
- Defective haemoglobinisation of the RBC and haemolysis
- β-Thal Major – Severe anaemia due to haemolysis, iron overload
- β-Thal Minor – Clinically asymptomatic
The Bit You have all been waiting for!
Why does a snowman not bleed to death when stabbed in the eye??
Haemostasis & Coagulation

- Overall haemostatic strategy:
  - Constriction of damage vessel
  - Plug the hole (platelets)
  - Consolidate the plug (generation of thrombin)
  - Dissolve the plug after healing
Primary Haemostasis

- Vascular trauma
  - Vasoconstriction and exposure of subendothelium
- Platelet activation and adhesion to collagen
- Platelet shape change and granule release leading to ↑ local concentrations of:
  - platelet activators
  - coagulation factors
  - vasoconstrictors
- Primary plug formed and blood loss stopped
Secondary Haemostasis

- The classical coagulation pathway
- The cell based model of coagulation
Secondary Haemostasis

- The classical coagulation pathway is a proteolytic cascade
- Each enzyme of the pathway is present in the plasma in its inactive form
- The end result of the clotting cascade is the production of thrombin (from prothrombin) for the conversion of fibrinogen to fibrin

3 phases:

- Intrinsic pathway
- Extrinsic pathway
- Final common pathway
Intrinsic Pathway

- Activated when blood comes into contact with sub-endothelial connective tissues (collagen)

- Quantitatively more important than extrinsic pathway, but slower
Intrinsic Pathway

- Ultimately activates factor X, which is the first molecule of the common pathway.
- Factor X is activated by a complex of molecules containing factor IXa, factor VIII, calcium and phospholipid (from the platelet surface, where the reaction takes place).
- Precise role of factor VIII not understood, but essential (Haemophiliacs).
- Factor VIII is modified by thrombin, which greatly enhances factor VIII activity, promoting factor X activation.
Extrinsic Pathway

- Provides a very rapid response to tissue injury, producing factor Xa almost instantaneously
- The main function of the extrinsic pathway is to augment the function of the intrinsic pathway
- Components unique to extrinsic pathway are tissue factor (or factor III) and factor VII
Extrinsic Pathway

- Tissue factor (FIII) is a transmembrane protein found in vesicles below the cell surface in most human cells. It is relocated to the surface after vascular damage where it binds rapidly to factor VII.

- FVIIa forms a complex with FIII (TF), calcium and phospholipid which then activates FX.
End result is the production of thrombin (from prothrombin) for the conversion of fibrinogen (F1) to fibrin.

Exposure of fibrinogen to thrombin results in rapid proteolysis of fibrinogen and the release of fibrinopeptide A.

A second peptide, fibrinopeptide B, is then cleaved by thrombin and the fibrin monomers formed polymerise spontaneously to form an insoluble gel.
Common Pathway

- The polymerised fibrin, held together by non-covalent and electrostatic forces, is stabilised by factor XIIIa (produced by the action of thrombin on factor XIII).

- These insoluble fibrin molecules aggregate, together with aggregated platelets, block the damaged blood vessel, and prevent further bleeding.
Coagulation Screening

Intrinsic Pathway - Partial Thromboplastin Time (PTT) (25-35 secs)

Extrinsic Pathway - Prothrombin Time (PT) (11-15 secs)

Common Pathway - PTT & PT
Inhibitors of Coagulation

- Thrombin is a very potent enzyme and various inhibitory agents limit its function to prevent uncontrolled and widespread intravascular coagulation.
- Antithrombin III and Protein C (& cofactor Protein S)
- Liver removes and degrades circulating activated clotting factors.
- Heparin acts by binding to antithrombin III, greatly accelerating inactivation of FXa and thrombin.
Fibrinolysis

- Once haemostasis is restored and the tissue is repaired, the clot must be removed by the fibrinolytic pathway.

- The enzyme plasmin is the end product of this pathway.

- Activators found in vascular endothelium, tissues and other sites convert the proenzyme plasminogen to the active enzyme plasmin.

- Tissue plasminogen activator (tPA) is the most important activator.

- Plasmin digests fibrin and fibrinogen to form Fibrin Degradation Products (FDPs).
The Fibrinolytic Pathway

Tissue plasminogen activator

Factor XIIa kallikrein

Plasminogen

Plasmin

Streptokinase

Fibrin

Protein C

Plasminogen activator inhibitor-1

α2-antiplasmin

Fibrin degradation products

Green arrows indicate activation. Black arrows indicate inhibition.
Vitamin K

- Factors II, VII, IX, and X are structurally similar – serine proteases
- Synthesised in the liver as inert precursors
- Converted to biologically functioning molecules by vitamin K dependent enzyme gamma-decarboxylase

Precursor → Gamma-carboxylase → Functional clotting factor

Reduced Vitamin K → Vitamin K Reductase → Oxidised Vitamin K

WARFARIN
TEG - The Future

WELCOME TO THE WORLD OF AUTOMATED MODULAR THROMBOELASTOGRAPHY
SO SIMPLE!

OR IS IT?

Figure 2 - Thromboelastograph Components
WHAT SORT OF SAUSAGE!

A: normal

B: prolonged (anticoagulation and factor deficiency)

C: decreased maximum amplitude (thrombocytopenia and platelet function inhibitors)

D: fibrinolysis

E: hypercoagulability

F: disseminated intravascular coagulation (DIC)

G: end-stage DIC (hypocoagulation)
The Cell Based Model

- Although the classical model of coagulation supports lab tests of coagulation disorders, it does not adequately explain the mechanisms leading to haemostasis \textit{in vivo}.

- A cell-based model of haemostasis has been developed to replace the classical model of the coagulation cascade.

- This model suggests that haemostasis occurs on different cell surfaces in three overlapping steps:
  - 1. Initiation
  - 2. Amplification
  - 3. Propagation
The Cell Based Model

- Initiation occurs on a tissue factor-bearing cell
- In the amplification phase, platelets and co-factors are activated to prepare for large-scale thrombin generation
- Finally, propagation occurs on the surface of platelets, and results in the propagation of large amounts of thrombin
1. Initiation phase

Injury of vessels wall leads to contact between blood and subendothelial cells

Tissue factor (TF) is exposed and binds to FVIIa or FVII which is subsequently converted to FVIIa

The complex between TF and FVIIa activates FIX and FX

FXa binds to FVa on the cell surface
The FXa/FVa complex converts small amounts of prothrombin into thrombin.

The small amount of thrombin generated activates FVIII, FV, FXI, and platelets locally. FXIa converts FIX to FIXa.

Activated platelets bind FVa, FVIIa, and FIXa.
3. Propagation phase

The FVIIIa/FIXa complex activates FX on the surfaces of activated platelets.

FXa in association with FVa converts large amounts of prothrombin into thrombin creating a “thrombin burst”.

The “thrombin burst” leads to the formation of a stable fibrin clot.
Haemostatic drug therapy

- Coagulation factor concentrates: ‘beriplex’
- Antifibrinolytics: tranexamic acid, aminocaprioc acid, aprotonin
- Desmopressin (Haemophilia A, vWD, uraemic patients)
- rFVII (Novoseven), ?the ultimate haemostatic drug!!!!
PCC (‘Beriplex’)

- II, VII, IX, X
- Admin over 5-10 mins
- Correct INR within 5 mins
- 15-25u/kg

- Elective: stop warfarin for 4 days
- 24hrs: PO vitamin K
- 6hrs: IV vitamin K
- Immediate: PCC
NovoSeven® Mode of Action
Eptacog alfa (activated)

Tissue factor (TF)/FVIIa, or TF/rFVIIa interaction, is necessary to initiate haemostasis.

At pharmacological concentrations rFVIIa directly activates FX on the surface of locally activated platelets. This activation will initiate the "thrombin burst" independently of FVIII and FIX. This step is independent of TF.

The thrombin burst leads to the formation of a stable clot.
# NovoSeven

<table>
<thead>
<tr>
<th>Presentation</th>
<th>Powder for injection with solvent</th>
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<tbody>
<tr>
<td><strong>Uses</strong></td>
<td>Patients with haemophilia and congenital FVII deficiency</td>
</tr>
<tr>
<td><strong>Doses</strong></td>
<td>Haemophilia: 90 mcg/kg&lt;br&gt;FVIIa deficiency: 15–30mcg/kg every 4–6hrs</td>
</tr>
<tr>
<td><strong>Precautions</strong></td>
<td>Close collaboration with haematologist</td>
</tr>
<tr>
<td><strong>Side-effects</strong></td>
<td>MI, arterial, venous thrombotic events</td>
</tr>
<tr>
<td><strong>Cost</strong></td>
<td>1.2 mg £664, 2.4mg £1329, 4.8mg £2658</td>
</tr>
</tbody>
</table>
ANY QUESTIONS
THANK YOU

- SCTG primary FRCA
- VIVA day
- Jan 7th 2013
- Brighton