An Approach to bleeding

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Death in the Emergency

First 24 hr bleeding is an Independent Predictor

- Mortality
- ICU Admission
- ICU LOSS

(After Controlling for ISS; GCS; Age; Gender; Anemia; Shock; Lactate; Base Deficit; Shock Index HR/SBP>0.6)

ICU mortality

Cook et al. Critical Care 2001; 5(6):368-375
Which bleeding disorders?

Bleeding frequency and bleeding recurrence is more common in inherited bleeding disorders than acquired bleeding disorders.
In this talk:

- Mechanism of haemostasis
- Aetiology of inherited bleeding
- Evaluation of inherited bleeding
- Principles of management of bleeding
Pathophysiology of bleeding

Any bleeding is a direct consequence of one or more of these three factors:

- Platelets
- Plasma proteins
- Blood vessel
Haemostatic balance

- Platelet
  - Plasma proteins
  - Coagulation
  - Blood vessel
  - Plasma proteins
  - Fibrinolysis
Vessel injury

Endothelial cell

Platelet

Sub Endothelial cell
Vasoconstriction
Endothelial cell

Platelet plug

Endothelial cell

TF

TF

TF

TF
Outline

- Mechanism of haemostasis
- **Aetiology of bleeding**
- Evaluation of bleeding
- Management of bleeding patient
Blood vessel abnormalities

Altered vessel wall
- Kasabach Merrit
- Hereditary haemorrhagic telangiectasia

Connective tissue
- Marfan syndrome
- Ehlers Danlos
- Pseudoxanthanoma
- Scleroderma

Congenital blood vessel abnormalities are very rare
Inherited platelet disorders

- Adhesion
- Aggregation
- Signal transduction
- Granular secretion
- Cytoskeletal changes
<table>
<thead>
<tr>
<th>Category</th>
<th>Disorders/Conditions</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adhesion</td>
<td>• Bernard Soulier</td>
</tr>
<tr>
<td>Aggregation</td>
<td>• Glanzman’s thrombasthenia</td>
</tr>
<tr>
<td>Signal transduction</td>
<td>• TS deficiency, cyclooxygenase</td>
</tr>
<tr>
<td>Granules</td>
<td>• Hemansky pudlac; GPS</td>
</tr>
<tr>
<td>Cytoskeletal</td>
<td>• Wiskott Aldrich</td>
</tr>
<tr>
<td>Primary secretion</td>
<td>• TVA2, ADP, Epinephrine</td>
</tr>
<tr>
<td>Production</td>
<td>• MYH9; TWAR</td>
</tr>
</tbody>
</table>
## Congenital Plasma proteins

<table>
<thead>
<tr>
<th>Plasma protein</th>
<th>Inheritance</th>
<th>Prevalence</th>
</tr>
</thead>
<tbody>
<tr>
<td>Factor I</td>
<td>AR/AD</td>
<td>Rare</td>
</tr>
<tr>
<td>Factor II</td>
<td>AD</td>
<td>Rare (extremely)</td>
</tr>
<tr>
<td>Factor V</td>
<td>AR</td>
<td>1 /1000 000 births</td>
</tr>
<tr>
<td>Factor VII</td>
<td>AR</td>
<td>1/500 000 births</td>
</tr>
<tr>
<td>Factor VIII</td>
<td>X linked</td>
<td>1/10 000 male births</td>
</tr>
<tr>
<td>Factor IX</td>
<td>X linked</td>
<td>1/60 000 male births</td>
</tr>
<tr>
<td>Factor X</td>
<td>AR</td>
<td>1/500 000 births</td>
</tr>
<tr>
<td>Factor XI</td>
<td>AD</td>
<td>4% AJ, rare otherwise</td>
</tr>
<tr>
<td>Factor XIII</td>
<td>AR/AD</td>
<td>Rare</td>
</tr>
<tr>
<td>vWF</td>
<td>AR/AD</td>
<td>1/100</td>
</tr>
</tbody>
</table>
Congenital Plasma proteins

vWF → Von Willebrand Disease

FVIII → Haemophilia A

FIX → Haemophilia B
Outline

- Pathophysiology of haemostasis
- Aetiology of bleeding
- Evaluation of bleeding
- Management of bleeding
Bleeding evaluation

- History
- Physical examination
- Screening tests
- Confirmatory tests
Bleeding evaluation

**History**
- Main differential diagnosis
- Congenital or acquired
- Drugs
Bleeding evaluation

History

- Type of bleed
- Extent of bleed
- Site
- Amount
- Spontaneous or induced
- Immediate or delayed
Skin bleed
Joint bleed
Mucous membrane bleed
Traumatic bleed
Bleeding evaluation

Examination

- Pallor, jaundice
- Petechiae, purpura, ecchymoses,
- Haemarthrosis, haematomas
- Melaena, haematochezia
- Neuropathy, blindness, respiratory compromise
Who to test?

- Definite family history
- Spontaneous bleeding
- Induced uncontrolled bleeding
- Life threatening bleed
- Minor recurrent bleed
- Prior to major surgery
- Prior to biopsy or invasive procedure
Von Willebrand Disease
Von Willebrand disease (vWD)

**Definition**

- Commonest inherited heterogeneous group of bleeding disorders
- Due to mutation in vWF gene
- Resulting in qualitative and/or quantitative deficiency of Von Willebrand factor
- May or may not present with bleeding
vWD - Prevalence

Estimated frequency
- 1 in 100
- No race, sex or age predilection

Accurate prevalence estimation precluded
- Variable expressivity
- Reduced penetrance
- New mutations
vWD - Aetiology

vWF gene mutation

Quantitative vWF abnormalities
- Partial or Complete

Qualitative vWF abnormalities
- Structure Function
## vWD - Classification

<table>
<thead>
<tr>
<th>Abnormality</th>
<th>vWD Type</th>
</tr>
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<tbody>
<tr>
<td><strong>Quantitative</strong></td>
<td></td>
</tr>
<tr>
<td>Partial absence</td>
<td>1</td>
</tr>
<tr>
<td>Total absence</td>
<td>3</td>
</tr>
<tr>
<td><strong>Structure</strong></td>
<td></td>
</tr>
<tr>
<td>Abn assembly/secretion</td>
<td>2A1</td>
</tr>
<tr>
<td>Multimer proteolysis</td>
<td>2A2</td>
</tr>
<tr>
<td><strong>Function</strong></td>
<td></td>
</tr>
<tr>
<td>↑binding to platelets</td>
<td>2B</td>
</tr>
<tr>
<td>↓binding to platelets</td>
<td>2M</td>
</tr>
<tr>
<td>↓binding to FVIII</td>
<td>2N</td>
</tr>
<tr>
<td>Type</td>
<td>Frequency</td>
</tr>
<tr>
<td>--------</td>
<td>-----------</td>
</tr>
<tr>
<td>Type 1</td>
<td>70-80%</td>
</tr>
<tr>
<td>Type 2</td>
<td>20-30%</td>
</tr>
<tr>
<td>Type 3</td>
<td>&lt;5%</td>
</tr>
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</table>
vWD- clinical features

<table>
<thead>
<tr>
<th>Symptom</th>
<th>Frequency (%)</th>
</tr>
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<tbody>
<tr>
<td>Epistaxis</td>
<td>60</td>
</tr>
<tr>
<td>Dental surgery</td>
<td>50</td>
</tr>
<tr>
<td>Easy bruising</td>
<td>40</td>
</tr>
<tr>
<td>Menorrhagia</td>
<td>35</td>
</tr>
<tr>
<td>Gum bleeding</td>
<td>35</td>
</tr>
<tr>
<td>Post[artum</td>
<td>25</td>
</tr>
<tr>
<td>Postoperative</td>
<td>20</td>
</tr>
<tr>
<td>GIT bleeding</td>
<td>10</td>
</tr>
</tbody>
</table>
Tests to establish the diagnosis
1- vWF antigen
2- vWF:R:Co
3- Factor VIII
4- PTT
5- Bleeding time

Tests to subtype vWD
1- RIPA
2- Multimer analysis
3- vWF- Factor VIII binding (Type 2N)
## Diagnostic Typing

<table>
<thead>
<tr>
<th></th>
<th>Type 1</th>
<th>Type 2</th>
<th>Type 3</th>
</tr>
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<tbody>
<tr>
<td>vWF-Ag</td>
<td>40-60</td>
<td>N</td>
<td>0</td>
</tr>
<tr>
<td>vWF-Activity</td>
<td>decreased</td>
<td>N</td>
<td>decreased</td>
</tr>
<tr>
<td>Factor VIII</td>
<td>40-60</td>
<td>N</td>
<td>2-3%</td>
</tr>
<tr>
<td>PTT</td>
<td>Normal</td>
<td>N</td>
<td>Prolonged</td>
</tr>
<tr>
<td>Bleeding time</td>
<td>Prolonged</td>
<td>N</td>
<td>Prolonged</td>
</tr>
</tbody>
</table>
## Diagnostic subtyping

<table>
<thead>
<tr>
<th></th>
<th>HMW MULTIMER</th>
<th>RI PA</th>
<th>F VII</th>
</tr>
</thead>
<tbody>
<tr>
<td>2A</td>
<td>Absent</td>
<td>Decreased</td>
<td>Normal</td>
</tr>
<tr>
<td>2B</td>
<td>Normal</td>
<td>Increased</td>
<td>Normal</td>
</tr>
<tr>
<td>2M</td>
<td>Normal</td>
<td>Decreased</td>
<td>Normal</td>
</tr>
<tr>
<td>2N</td>
<td>Normal</td>
<td>Normal</td>
<td>Reduced</td>
</tr>
</tbody>
</table>
Treatment of vWD

**Non replacement therapy**
- DDAVP
- Antifibrinolytic therapy
- Oestrogen therapy

**Replacement therapy**
- vWF plasma concentrate
- Cryoprecipitate
- Recombinant vWF
The Haemophiliias
Factor level

- Factor <1% - Severe
- Factor 2-5% - Moderate
- Factor >5-50% - Mild
- Factor >50% - Normal

Note

- Biochemical-clinical phenotypic discrepancy
Clinical features

Haemorrhagic manifestation
- **Severe** - Bleed spontaneously
- **Moderate** - Bleed with minor trauma
- **Mild** - Bleed with surgery or trauma

Sites of bleeding
- **Joints** - knees > ankles > elbows > shoulder > wrist > hips
- **Muscles** - Psoas muscle, quadratus,
- **Mucous membranes** - oral, nasal, GIT, GUT
- **Organ systems** - GUT, GIT, CNS,
Laboratory Diagnosis

Screening tests
- INR
- PTT
- Platelet count
- Hess test /bleeding time

Confirmatory tests
- Correction studies
- Factor assay
- Inhibitor assay
- Genetic family studies
<table>
<thead>
<tr>
<th>Differential diagnosis</th>
<th>Coagulation defect</th>
<th>Plt/ capillary defect</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Bleeding onset</strong></td>
<td>Spontaneous</td>
<td>Induced</td>
</tr>
<tr>
<td><strong>Site of bleeding</strong></td>
<td>Joints/muscles</td>
<td>Skin/ mucous</td>
</tr>
<tr>
<td><strong>Type of bleeding</strong></td>
<td>Haemarthrosis/ haematoma</td>
<td>Petechiae/echymoses/ purpura</td>
</tr>
<tr>
<td><strong>Time to onset</strong></td>
<td>Delayed</td>
<td>Immediate</td>
</tr>
<tr>
<td><strong>INR/ PTT</strong></td>
<td>Prolonged</td>
<td>Normal</td>
</tr>
<tr>
<td><strong>Bleeding time</strong></td>
<td>Normal</td>
<td>Prolonged</td>
</tr>
<tr>
<td><strong>Platelet count/ fx</strong></td>
<td>Normal</td>
<td>Abnormal</td>
</tr>
</tbody>
</table>
Principles of Management

- Treat bleeds with specific product
- Treat bleeds early
- If in doubt, treat
- Treat veins with care
- Avoid product causing platelet dysfunction
- Avoid intramuscular injections
- Do not aspirate joints before treatment
- Apply local measures
- Multidisciplinary approach
Advances in management of bleeding disorders
Standard of care is replacement of missing clotting factor
Standard of care in haemophilia is replacement of missing clotting factor.
### Remarkable progress in haemophilia bleed management therapies

<table>
<thead>
<tr>
<th>Year</th>
<th>Event</th>
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<tbody>
<tr>
<td>1944</td>
<td>1st use of whole blood</td>
</tr>
<tr>
<td>1955</td>
<td>1st use of Fresh Frozen Plasma used</td>
</tr>
<tr>
<td>1964</td>
<td>1st use of Cryoprecipitate used</td>
</tr>
<tr>
<td>1980</td>
<td>1st use of Plasma derived Clotting factor</td>
</tr>
<tr>
<td>1990</td>
<td>1st use of recombinant Clotting Factor</td>
</tr>
<tr>
<td>1998</td>
<td>1st use of extended half life clotting factors</td>
</tr>
<tr>
<td>2014</td>
<td>1st use of non-replacement therapies</td>
</tr>
</tbody>
</table>
CFC replacement approaches

**New CFC replacement therapies**
- Fusion technology
- PEGylation technology
- Sequence modification CFC

**Non CFC replacement therapies**
- Anti-TFPI
- AT3 RNAi
- Anti IXa/X
- Gene therapy

CFC- Clotting factor concentrate; AT3- antithrombin 3; TFPI-tissue factor pathway inhibitor; RNAi – interfering ribonucleic acid
CFC replacement technologies

**Fusion technology**
1. FC fusion
2. Albumin fusion
3. CTP fusion
4. PSA fusion

**PEGylation technology**
1. Site directed
2. Random
3. GlycoPEGylation

**Sequence modification**
1. Heavy+light chain fusion

**Ongoing**
- Mahlangu et al 2014
- Powell et al 2013
- Santogostino 2016
- Shah et al 2016
- Konkle et al 2015
- Giangrande 2016
- Young et al 2016

**Completed**
- Mahlangu 2016

**Products**
- FVIII-FC
- FIX-FC
- FVIII-FP
- FIX-FP
- FVII-FP
- FVII-CTP
- Bax-826
- Bay-94 9027
- Bax-855
- N8-GP
- N9-GP
- SingleChain FVIII
- Mahlangu 2016
Non-replacement therapies in haemophilia

- Anti-TFPI
- AT3 RNAi
- Anti IXa/X
- Gene therapy

Non CFC replacement therapies
Non-replacement therapies in haemophilia

- Anti-TFPI
- AT3 RNAi
- Anti IXa/X
- Gene therapy
Rationale for anti-TFPI use in haemophilia

Rationale for anti-TFPI use in haemophilia

Broze GJ, Jr., Girad TJ. *Front Biosci (Landmark Ed)* 2012; 17: 262–80.]
Anti-TFPI Clinical studies

PEG-Aptamer
- Bax 499
- Phase I/II
- Increased TFPI level and decreased thrombin
- Stopped due to excessive bleeding
Phase 1 anti-TFPI study

Safety and pharmacokinetics of anti-TFPI antibody (concizumab) in healthy volunteers and patients with hemophilia: a randomized first human dose trial


*Katharine Dormandy Haemophilia Centre and Thrombosis Unit, Royal Free Hospital, London, UK; †Novo Nordisk A/S, Søborg, Denmark; ‡Copenhagen University, Copenhagen, Denmark; §Division of Hematology, University Hospital, Zurich, Switzerland; ¶University Department of Haematology, Manchester Royal Infirmary, Manchester, UK; **Haemophilia Centre, National Blood Centre, Kuala Lumpur, Malaysia; ††Department of Internal Medicine—Angiology, Haemostasis and Coagulation disorders, Vivantes Hospital im Friedrichshain, Berlin, Germany; †††Division of Haematology and Haemostasis, Department of Medicine 1, Medical University of Vienna, Vienna, Austria; §§Imperial College London, Hammersmith Hospital, London, UK; ¶¶Department of Molecular Medicine and Haematology, Faculty of Health Sciences, University of the Witwatersrand and NHLS, Johannesburg, South Africa; ***Zentrum für Innere Medizin, Med. Klinik III, Hämobilie-Zentrum, Frankfurt/M, Germany; †††Thrombosis and Haemostasis Unit, Department of Haematology, Rigshospitalet, Copenhagen, Denmark; ‡‡‡Haematology Department, Hospital Universitario La Paz, Madrid, Spain; and §§§Division of Hematology, Department of Medicine, Faculty of Medicine, Ramathibodi Hospital, Bangkok, Thailand
Pharmacokinetics sc vs iv

Non-linear pharmacokinetics

Safety profile

- No SAE
- No thrombosis or vascular events
- No allergy
- No anti-concizumab antibodies
- No inhibitors
- No clinically relevant changes in
  - TT, aPTT, fibrinogen, antithrombin
- Dose dependent changes in
  - Ddimers
  - Prothrombin fragment 1+2
Non-replacement therapies in haemophilia

- Anti-TFPI
- AT3 RNAi
- Anti IXa/X
- Gene therapy

Non CFC replacement therapies
Haemostasis in normal and PWH

AT role

FVIIIa → FX → FVa → Prothrombin → Thrombin → Fibrinogen → Fibrin → Blood clot

FV → FVa

FIXa → FXa

FVIIa → FX

AT3 RNAi Mechanism of action

Ragni M NEJM 2015; 373(4): 389-390
Phase 1 Results: Dose dependent response

Mean maximal AT lowering of $87 \pm 1\%$ at 80 mg fixed dose

AT after monthly dosing of ALN

Pasi et al. WFH congress 2016, Orlando
Summary of Median ABR (All Cohorts, n=17)

Pasi et al. WFH congress 2016, Orlando
Safety profile based on current data

- Fitusiran generally well tolerated in hemophilia A and B patients with and without inhibitor

- No SAEs related to study drug;

- No thromboembolic events

- 11 (35%) patients reported mild drug-related ISRs
  - Mostly pain and/or erythema at the injection site

- AEs (excluding injection site reactions(ISRs)) in ≥10% of patients:
  - upper respiratory tract infection (10%) and arthralgia (10%);
  - majority mild or moderate in severity
  - 1 discontinuation due to AE; event resolved in this patient with symptomatic management
Summary on ALN-RNAi

- Dose-dependent AT lowering and thrombin generation increase achieved, with once-monthly subcutaneous dose regimen; fixed 80 mg dose provides consistent AT lowering >75%
- Evidence of clinical activity and potential correction of hemophilia phenotype in non-inhibitor patients
- In exploratory post-hoc analysis in monthly dose cohorts, fitusiran achieved median ABR = 0, with
  - 53% patients bleed-free and
  - 82% patients experiencing zero spontaneous bleeds
Non-replacement therapies in haemophilia

- Anti-TFPI
- AT3 RNAi
- Anti IXa/X
- Gene therapy

Non CFC replacement therapies
Plasma ACE910 concentration

A

B
Emicizumab was associated with neither serious adverse events nor clinically relevant coagulation abnormalities.

Plasma concentrations of emicizumab increased in a dose-dependent manner.

Activated partial-thromboplastin times remained short throughout the study.

The median annualized bleeding rates in cohorts 1, 2, and 3 decreased from 32.5 to 4.4, 18.3 to 0.0, and 15.2 to 0.0, respectively.

- There was no bleeding in 8 of 11 patients with factor VIII inhibitors (73%) and in 5 of 7 patients without factor VIII inhibitors (71%). Episodic use of clotting factors to control bleeding was reduced. Antibodies to emicizumab did not develop.
Scepticism about gene therapy

Gene therapy deserves a fresh chance

Initial interest in gene therapy waned after the technology failed to live up to expectation. Progress made since has received little attention, but suggests that the pervading sense of disillusionment is misplaced.
First successful FIX gene therapy in man

- Phase 1
- 6 patients, 2 in each of 3 dose escalations
- Durable FIX levels 1–6%
- 2/6 transient ALT elevation

Nathwani, et al. NEJM 2011;365;2357
Haemophilia B longer term follow up data

- Long-Term Safety and Efficacy of Factor IX Gene Therapy in Hemophilia B.
- Long-term follow up of original 6 patients
  - Each of 3 dose escalations
- Plus 4 additional subjects
  - at high dose: $2 \times 10^{12}$ vector genomes/kg
- Long term FIX levels 1–6% – median 3.2 years follow up

Nathwani, et al. *NEJM* 2014;371:21
FVIII gene therapy?

- Bigger protein than FIX- Packaging more complex

- Platelet directed FVIII gene therapy
  - Animal model, lentiviral

- Hybrid porcine / human sequence
  - Lentiviral


 BMN 270: AAV 5-factor VIII vector
  - Trial due to start Q2/3 2015
  - First in man phase 1 FVIII trials

 UCL/St Jude (Nathwani/Davidoff)
  - Imminent
Summary

- Significant progress has been made in the development of therapies for haemophilia.
- Extended half life products promise to make prophylaxis a reality.
- Evolving alternative therapies are beginning to emerge.
- All therapies have limitations in that they may not apply to all patients with haemophilia.
- Gene therapy remains the choice option for cure of haemophilia.
Thank You!