Abnormal Basic Coagulation Testing

Laboratory Testing Algorithms

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Global Coagulation Testing

- No single global laboratory test
- Bleeding history is the strongest predictor of bleeding risk for any procedure
- Testing performed for:
  - Screening
  - Monitoring anticoagulant therapy
  - Guide for component therapy
  - Examination of components of coagulation
  - “for the record”, “medicolegal”, “routine”
- These assays examine components of coagulation
  - Prothrombin Time (PT): Extrinsic and Common Pathway
  - Activated Partial Thromboplastin Time (aPTT): Intrinsic and Common Pathway
  - Fibrinogen
- Problems
  - Not predictive
  - Not completely standardized
  - Artifacts
  - Misleading
  - False sense of security
  - Pursuit of clinically irrelevant abnormal tests

Coagulation Cascade

A Little Simpler

Lee-White Clotting Time

- 1912 - Roger Lee & Paul Duncan White
  - formulated and developed the ‘Lee-White’ clotting time.
- Historically used to monitor heparin
- Poorly standardized
  - Activator
  - Calcium
  - Phospholipid

Prothrombin Time

- Platelet Poor Plasma
- Citrated 9:1; 3.2%
- Thromboplastin
  - Phospholipid
  - Source of tissue factor (e.g. rabbit brain)
  - Ca++
- Time to clot detection (seconds)
- Sensitive to factor VII, but also V and X
- Standardization
  - INR & ISI (Prior Lecture)
**aPTT**
- Platelet Poor Plasma
- Citrated 9:1 3.2%
- Activator (e.g. silica, kaolin)
- Partial Thromboplastin
  - No source of tissue factor
  - Phospholipid
  - Ca\(^{+2}\)
- Time to Clot Detection (seconds)
- Need to determine therapeutic range for heparin (Prior Lecture)
- aPTT can be shortened due to elevated FVIII as an acute phase reactant

**Fibrinogen**
- Plasma diluted
- High concentration of thrombin (IIa)
- Calibrators plotted against log(TT)
- Will not differentiate hypofibrinogenemia from dysfibrinogenemia

**Hemorrhagic Diseases with Normal PT and/or aPTT**
- Mild von Willebrand Disease
- Mild Hemophilia
- Platelet Dysfunction
- α2-antiplasmin deficiency
- Dysfibrinogenemia
- Monoclonal Gammopathy
- Factor XIII deficiency
- Vascular or connective tissue abnormalities

**Abnormal Coagulation Tests with No Bleeding**
- Factor XII Deficiency
- Prekallikrein Deficiency
- High molecular weight kininogen deficiency
- Mild VII deficiency (e.g. heterozygotes)
- Lupus Anticoagulants

**Preanalytical Variable and Spurious Results**

<table>
<thead>
<tr>
<th>Variable</th>
<th>Explanation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Anticoagulant</td>
<td>3.2% vs. 3.8% Citrate</td>
</tr>
<tr>
<td>Under or over fill</td>
<td>Whole Blood to Anticoagulant Ratio (9:1) altered</td>
</tr>
<tr>
<td>Hematocrit</td>
<td>Plasma to Anticoagulant Ratio (9:1) altered</td>
</tr>
<tr>
<td></td>
<td>Low Hct: too little anticoagulant</td>
</tr>
<tr>
<td></td>
<td>High Hct: too much anticoagulant</td>
</tr>
<tr>
<td>Order of fill</td>
<td>EDTA should be tube drawn after coag  EDTA can lead to over-binding of calcium</td>
</tr>
<tr>
<td>Transport</td>
<td>PF4 release over time neutralizes heparin</td>
</tr>
<tr>
<td></td>
<td>Labile factors decrease with time at room temperature</td>
</tr>
<tr>
<td>Phlebotomy</td>
<td>Waste Tube Prior to Drawing Coag Studies</td>
</tr>
<tr>
<td></td>
<td>Thromboplastin release with phospholany</td>
</tr>
<tr>
<td>Clot</td>
<td>Loss of factors to form clot (lengthen time)</td>
</tr>
<tr>
<td></td>
<td>Clot can interfere with clot detection (shorten time)</td>
</tr>
</tbody>
</table>
Causes of abnormal PT only

- Factor Deficiency
  - Factor VII
  - Common Pathway (II, V, X)
- Warfarin Ingestion
- Liver Dysfunction
- Vitamin K Deficiency
- Disseminated Intravascular Coagulation
- Lupus Anticoagulant

Congenital Factor VII Deficiency

- AR disorder 1 in 500,000
- 50% no function, 50% no antigen
- Presentation varies widely depending on level of expression
- <10% factor activity increases likelihood of bleeding
- Bruising, epistaxis, soft tissue hemorrhage, menorrhagia, post-partum bleeding
- <1% have severe bleeding
- CNS hemorrhage during delivery, hemorrhages
- Association with aplastic anemia, homocystinuria, Dubin-Johnson, Rotor Syndrome, Gilbert Syndrome
- Treat → Novoseven (20-30 μg/kg), Prothrombin Concentrates, Plasma

Causes of Elevated aPTT only

- Heparin, DTI
- Factor Deficiencies
  - HMWK and PK
  - Factors XII, XI, IX, VIII
  - Common Pathway (X, V, II, I)
  - Usually found with elevation of aPTT
- Lupus Anticoagulant
- Specific Inhibitors (e.g. Factor VIII inhibitors)
- Possible warfarin, liver dysfunction, DIC
Thrombin Time

- Hypofibrinogenemia
- Dysfibrinogenemia
- Heparin (very sensitive!!!)
- Fibrin Degradation Products
- High [Immunoglobulins]
- Anti-bovine thrombin antibodies if bovine thrombin used

Reptilase Time

- Bothrops atrox
- Hypo/Dysfibrinogenemia (Another lecture!)
  - Congenital
    - Can’t convert fibrinogen to fibrin
    - Abnormal fibrinopeptide release
    - Fibrin polymerization defect
    - Abnormal stabilization
    - Resistance to fibrin lysis
  - Afibrinogenemia
    - Mutation in any of the three chains
  - Presentation varies from no complications to hemorrhagic and thrombotic complications

Mixing Study

- Hemophilia A and B and C (another lecture!)
  - Hemophilia A
    - 1/5,000 live male births
    - XLR Factor VIII Deficiency (Mild, Moderate or Severe)
    - Classically intraarticular, soft tissue and CNS bleeding (2-8%)
    - 80-85% of hemophilia cases
    - Replace with Recombinant Factor VIII
  - Hemophilia B
    - 1/30,000 live male births
    - XLR Factor IX Deficiency
    - Replace with Recombinant Factor IX
  - Factor XI (Hemophilia C)
    - AR; mild or no bleeding tendency
    - Levels do not correlate with bleeding
    - Spontaneous bleeding is not a feature

Other Factor Deficiencies with no bleeding

- Prekallikrein
  - AR
  - May be associated with thromboembolism
- HMWK
  - AR
  - No bleeding abnormalities
- Factor XII (Hageman Factor)
  - AR and at times AD
  - Homozygotes have no activity
  - Heterozygotes 20-60%
  - Do not experience bleeding
  - Reported associations with spontaneous abortion, premature delivery, arterial and venous thrombosis, MI, PE
Lupus Anticoagulants (Another lecture!)

- Antibodies to phospholipids, phospholipid bound proteins (e.g. beta-2-glycoprotein I)
- Interfere with coagulation assay, prolonging the tests but a misnomer because they cause thrombosis
- Criteria
  1. Two prolonged phospholipid-dependent screening tests
     1. aPTT, DRVVT, KCT, dPT (TTI)
  2. Mixing study shows circulating anticoagulant
  3. Confirmatory test is positive
  4. Demonstrated twice at least 6 weeks apart

Factor VIII Inhibitor

- Alloantibodies developing in hemophiliacs
  - 15 to 35% of patients
  - Active bleeding does not subside with factor VIII replacement
  - Bypassing agents, Novoseven
  - Immune tolerance induction

- Acquired Inhibitors (Non-hemophilia)
  - Bleeding Manifestations are usually severe
  - Soft tissue bleeding (e.g. intramuscular), GI or urinary bleeding more common than intraarticular bleeding
  - 8-22% mortality, usually within weeks after presentation
  - No concomitant disease can be found in 50%
  - Remainder have connective tissue disease, IBD, malignancy, dermatologic disorders
  - Treat with immunosuppressant and Novoseven
**Von Willebrand Disease (Another lecture!)**

- Types 1, 2A, B, N, M, 2 pseudo
- If moderate to severe can present with bleeding in childhood or young adulthood
- Male and females equally affected
- Platelet Type Bleeding
  - Bruising
  - Epistaxis, oral bleeding
  - Menorrhagia
  - GI bleeding
- Laboratory Tests:
  - Bleeding Time/PFA-100
  - vWF Antigen
  - vWF Ristocetin Cofactor
  - Factor VIII Activity
  - Ristocetin Induced Platelet Aggregation
  - Multimer Analysis
- Treatment
  - DDAVP
  - Humate-P

**Causes of isolated aPTT increase in hospital**

- >50% of cases due to LAC
- No cause found in >30%
- Factor Deficiencies and combined factor deficiencies
- Low Numbers of:
  - vWD
  - Factor inhibitors

**Elevated PT and aPTT**

- Intrinsic + Extrinsic Pathway
- Intrinsic + Common Pathway
- Extrinsic + Common Pathway
- Extrinsic + Intrinsic + Common Pathway
- Common Pathway Only
  - Factors X, V, II

**Causes of PT and aPTT elevation together**

- Supertherapeutic Warfarin
- Supertherapeutic Heparin
- Direct Thrombin Inhibitors
- Multiple Factor Deficiencies
- Liver Disease
- Disseminated Intravascular Coagulation (can be shortened due to activated II and X, elevated FVIII)
- Afibrinogenemia or Hypofibrinogenemia
- Congenital Dysfibrinogenemia
- Lupus Anticoagulant (e.g. against prothrombin(II))
- Specific Inhibitor (multiple factors)