Coagulation Testing
at the Point of Care

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Coagulation Testing

- Monitoring hemostasis

Bleeding  Clotting
What is Heparin?

- Glucopolysaccharide
- MW range: 6,000 - 25,000 daltons
- Only ~1/3 molecules active
  - Must contain specific sequence of glucosaccharides to function
Heparin Effects on Coagulation

12 → Prekallikrein → Kallikrein

11 → Heparin + AT

9 → Heparin + AT

8 → Heparin + AT

7 → Plasminogen Converted to Plasmin

10 → Fibrinogen

5 → Thrombin

2 → CLOT (Fibrin)

FDP

D-dimers

Modified from Utley. Vol.1, 1982
Why Monitor Heparin?

- Potency varies by manufacturer
  - Potency varies by lot
- Dose response varies by patient
  - Half life ranges from 60 - 120 minutes
  - Non-specific binding
- Functions by accelerating action of antithrombin
  - Antithrombin level critical for appropriate response
How to Monitor Heparin?

- Laboratory measures of activity
  - a Factor Xa
  - a Factor IIa (thrombin)
  - No clear correlation between heparin activity and patient outcome
  - TAT generally too long for peri-procedural use

- Viscoelastography
  - TEG / ROTEM
  - Reflects entire coagulation process
  - Requires interpretation
  - TAT generally too long for peri-procedural use

- ACT
What is an ACT?

- Modified Lee-White clotting time
  - Add blood to glass tube, shake
    - Place in heat block
    - Visual clot detection

- First described in 1966 by Hattersley
  - Activated Clotting Time
    - Add blood to glass tube with dirt, shake
      - Diatomaceous earth activator
      - Place in heat block
      - Visual clot detection
    - Proposed for both screening for coagulation defects and for heparin monitoring
Activated Clotting Time

Extrinsic Pathway

Intrinsic Pathway

Common Pathway

CLOT

Extrinsic Pathway
Why do we use an ACT?

- **Point of Care**
  - Immediate turn around
  - Rapidly adjust anticoagulant dosing as needed

- **Literature supports use of ACT**
  - Poor correlation between ACT & heparin level (1981)
  - Hemochron and HemoTec clinically different (1988)
  - Differences ignored by clinicians, yet...

- **Improved clinical outcome with ACT use**
  - Reviewed: 2007 NACB Laboratory medicine practice guideline for point of care coagulation testing
Why do ACTs Differ?

- Activator
  - diatomaceous earth; kaolin; glass beads; thromboplastin; combinations

- Sample measurement
  - Manual; automated

- Sample mixing
  - Manual; automated; physical; chemical

- Endpoint detection
  - Clot; surrogate marker

- By design!
HEMOCHRONOMETER

- Later - HEMOCHRON
- Add blood to tube, shake
  - Manual sample treatment
- Place in test well
  - Automated heating
  - Mechanical, objective fibrin clot detection
- Two different activators
  - CA510 (later FTCA510)
    - Diatomaceous earth
    - P214 glass bead
Two assays for separate uses
1980’s

- HemoTec ACT (later Medtronics ACTII)
  - Add blood to dual cartridge
    - Liquid kaolin activator
  - Place in instrument
    - Automated mixing
- Results don’t match Hemochron
1990’s

- Microsample ACTs - Hemochron Jr
  - Add blood to sample well, press start
    - Automated sample measurement
    - Automated mixing
    - Objective clot detection

- Results still don’t match
Abbott Point of Care - i-STAT

- Thrombin detection
  - Synthetic thrombin substrate
  - Electro-active compound formed, detected amperometrically
  - Clotting time reported
- First non-mechanical clot detection
- Direct chemical assessment of the appearance of active thrombin
Where is an ACT Used?

- Cardiac surgery
  - Recommended as 1° method in AmSECT guidelines
- Percutaneous coronary intervention (PCI)
- Interventional cardiology
- ECMO
- Critical care
- Interventional radiology
- Electrophysiology
- Vascular surgery
- etc.
Dosing & Target Times

- “Standard” target times
  - Most developed with manual ACT
  - Suggested due to high variability
  - No evidence for optimal ACT targets

- Drug defined targets
  - GPIIb/IIIa Inhibitors; Angiomax
  - Drug manufacturer defines ACT target
    - Does not specify ACT type
    - Ignores “off-label” indications
How to Compare ACTs?

- **Clinical Correlation**
  - In clinical setting to be used
    - Do not compare in CVOR to change in cath lab
  - Data MUST span current target times
  - Correlation coefficient
    - $R \geq 0.88$

**CORRELATE DOES NOT MEAN MATCH**
Clinical Comparison

- Data used to predict new target time
- Clinical agreement determined from predicted target time
  - Only method of value in ECMO, sheath pull
    - Range of values too small for correlation analysis
Evaluate Clinical Agreement

- CVOR example

<table>
<thead>
<tr>
<th>Current</th>
<th>New</th>
<th>N</th>
<th>%</th>
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<tbody>
<tr>
<td>≥ 480</td>
<td>≥ 520</td>
<td>72</td>
<td>34%</td>
</tr>
<tr>
<td>≥ 480</td>
<td>&lt; 520</td>
<td>19</td>
<td>9%</td>
</tr>
<tr>
<td>&lt; 480</td>
<td>≥ 520</td>
<td>7</td>
<td>3%</td>
</tr>
<tr>
<td>&lt;480</td>
<td>&lt;520</td>
<td>117</td>
<td>54%</td>
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</table>

- 88% agreement
  - 21 of 26 discrepancies
    - Current value within 10% of 480
  - 5 of 26 discrepancies
    - New leads to additional heparin given
Help clinician overcome differences

- Source:
  - Reagent differences
  - Technology differences
  - No standardization

Alter target times to Maintain clinical protocols
Coagulation Testing

Intrinsic Pathway

Extrinsic Pathway

Common Pathway

X → Xa

II → IIa (thrombin)

LMWH & DXaI

Hirudin & DTI

Monitor with ACT / aPTT

Monitor with PT

Monitor with ???

HEPARIN

WARFARIN

CLOT
ACT versus aPTT

- **ACT**
  - Activated clotting time
  - POC Only
  - Low, moderate or high dose heparin
    - System dependent

- **aPTT**
  - Activated partial thromboplastin time
  - Laboratory or POC
  - Low dose heparin only
    - System dependent upper limit
Where is an aPTT Used?

- Critical care
  - Heparin drip maintenance
- Unusual, but possible:
  - Interventional radiology
  - Electrophysiology
  - Vascular surgery
  - ECMO
- Any low dose heparin application
aPTT test methods

- **Standard Laboratory**
  - Platelet Poor Plasma
  - Sodium Citrate Anticoagulant
  - Dilution in testing
  - Variable Preanalytical Delay
  - Instruments
  - Reagents

- **Point of Care**
  - Whole Blood
  - No Added Anticoagulant
  - No Dilution
  - No Preanalytical Delay
  - Instruments
  - Reagents
$y = 0.737x + 22.2$

$R = 0.920$

Correlate Does Not Mean Match
Extrinsic Pathway

Common Pathway

Intrinsic Pathway

HEPARIN

WARFARIN

Monitor with ACT / aPTT

Monitor with PT

Monitor with ???

CLOT

II

IIa (thrombin)

X

Xa

LMWH & DXa1

Hirudin & DTI
## Heparin versus Warfarin

<table>
<thead>
<tr>
<th>Drug</th>
<th>Mechanism of Action</th>
<th>Cofactor</th>
<th>Monitor</th>
<th>Effective</th>
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<tbody>
<tr>
<td>Heparin</td>
<td>Direct thrombin inhibition</td>
<td>Anti-thrombin</td>
<td>aPTT ACT</td>
<td>Immediate</td>
</tr>
<tr>
<td>Warfarin</td>
<td>Decrease factor production</td>
<td>Vitamin K</td>
<td>PT</td>
<td>3-5 day delay</td>
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</table>
What is Warfarin?

- Rat poison
- Cause of “sweet clover disease”
- Orally active anticoagulant
Warfarin Effects on Coagulation

Anticoagulant action of warfarin: Slow onset

1. KO-reductase — warfarin sensitive
2. K-reductase — relatively warfarin resistant

Why Monitor Warfarin?

- Potency may vary by manufacturer
- Dose response varies by patient
  - Dietary interactions
  - Life-style influences
- Functions by decreasing production of Vitamin K dependent clotting factors in liver
  - Delayed onset of anticoagulation
How to monitor warfarin?

Quick, et. al., 1937 – Prothrombin Time

- Combine thromboplastin, calcium and patient plasma
  - Measures activity of factors I, II, V, VII, X

40 – 50 years pass

- Thromboplastin isolated from:
  - Different species: pig; cow; human; etc.
  - Different organs: brain; thymus; lung; etc.
- All yield different results
  - Results vary by instrument system in use
    - Manual tilt tube “gold standard”
    - Fibrometer; automated coagulation systems
- PT ratios adopted to determine therapeutic range
1983 – WHO and ISTH recommend the use of the INR to standardize PT result reporting

**International Normalized Ratio (INR)**

- **ISI** = international Sensitivity Index
- INR target ranges are specified by patient populations, e.g.,
  - DVT, Afib, Atrial MHV: INR= 2.0 - 3.0
  - Mitral mechanical heart valve: INR= 2.5 – 3.5
  - Individual variation

\[
INR = \left( \frac{PT_{\text{patient}}}{PT_{\text{meannormal}}} \right)^{ISI}
\]
Key variables

- **ISI**
  - Initially determined by reagent manufacturer
  - Traceable to IRP
    - International Reference thromboplastin Preparation
  - WHO defined process
    - Calibration up to INR = 4.5
    - manual tilt tube method reference
  - Local calibrations can be performed to determine the instrument specific ISI\(^1\)

- **Mean normal PT**
  - The mean normal PT should be determined for each new batch of thromboplastin with the same instrument used to assay the PT\(^1\)

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**Effect of Local Calibration**

- Local calibration may introduce variability

> Same sample yields different results depending on calibration method

POC Calibration

- Manufacturer assigns ISI and mean normal PT (MNPT)
  - Lot specific
- Traceable to IRP
  - Often through secondary standard
- Cannot be changed by end user
  - Does not vary by location of testing
Will POC Results Match the Lab?

but it WILL Correlate
Why not?

- **Point of Care**
  - Whole Blood
  - No Added Anticoagulant
  - No Dilution
  - No Preanalytical Delay
  > Reagent
  > Instrument
  > Clot detection

- **Laboratory**
  - Platelet Poor Plasma
  - Sodium Citrate Anticoagulant
  - 1:9 Dilution
  - Variable Preanalytical Delay

<table>
<thead>
<tr>
<th>Thromboplastin</th>
<th>Analyzer</th>
<th>calibration</th>
<th>Thromboplastin</th>
<th>Analyzer</th>
<th>calibration</th>
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<tbody>
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<td>Innovin</td>
<td>CA1500</td>
<td>Local vs rTF/95</td>
<td>HepatoQuick</td>
<td>STA-R</td>
<td>Manufacturer</td>
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<tr>
<td>Recombiplastin</td>
<td>MLA1800</td>
<td>Local vs rTF/95</td>
<td>Thrombotest</td>
<td>KC10</td>
<td>Local vs OBT/79</td>
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<tr>
<td>Neoplastin Plus</td>
<td>STA-R</td>
<td>Manufacturer</td>
<td>Thromboplastin C Plus</td>
<td>CA1500</td>
<td>Manufacturer</td>
</tr>
</tbody>
</table>
Expectations Lab to Lab

- 10 OAT patients across 7 analyzer/reagent combinations

Expectations POC to lab

- 36 patients over 4 visits each
  - 3 POC; 1 lab
  
Variability of Lab INR

- Observed:
  - ± 0.4 at INR = 2.0
  - ± 0.8 at INR = 3.0
  - ± 1.2 at INR = 4.0

- Standardization as with glucose is unlikely
  - discrete analyte to be tested
  - versus a biologic process

Patient Management

1. Understand limitations in the INR
   > Whenever a patient undergoes duplicate testing on different systems, there is the potential for disagreement

2. Attempt to have patients managed with a consistent methodology

How to Compare INR Results

- Lower dose?
- Keep same dose?
- Raise Dose?
- Test Again?
- Test more often?
Why perform POC PT?

- Results Available While Patient is Present
  - Improved Anticoagulation Management
  - Improved Standard of Care
  - Staff Efficiency

- Immediate Retesting (if needed)
  - Fingerstick Sampling
**LIMITATION!!!!!!**

- INR was developed to monitor effect of vitamin K antagonists (warfarin, others)
- INR is inappropriate scale for monitoring coagulopathies
- Most POC PT/INR tests cleared ONLY for monitoring patients receiving oral anticoagulation therapy such as Coumadin or warfarin.
POC Coagulation Testing

- Monitoring hemostasis

Bleeding

Clotting
QUESTIONS?