The effect of the direct oral anticoagulants (DOACs) on haemostasis tests.

Emmanuel J Favaloro, Haematology, ICPMR, Pathology West, Sydney Centres for Thrombosis and Haemostasis, Westmead Hospital

(with assistance from plethora of others including the RCPA Haematology QAP)

emmanuel.favaloro@health.nsw.gov.au
February, 2016
Background publications available from speaker:

Direct oral anticoagulants (DOACs)

- Main agents:
  - Dabigatran (etexilate; Pradaxa; Boehringer Ingelheim Pty Ltd): Direct thrombin inhibitor
  - Rivaroxaban (Xarelto; Bayer Ltd): Direct Factor Xa inhibitor
  - Apixaban (Eliquis; Bristol-Myers Squibb Pharmaceuticals): Direct Factor Xa inhibitor
  - (Edoxaban (Savaysa, Daiichi Sankyo): Direct Factor Xa inhibitor;
  - Others coming)
Direct oral anticoagulants

- Terminology/acronyms:
  - New or ‘novel’ oral anticoagulants (NOACs)
  - Eventually not so new or novel, so ‘direct’ oral anticoagulants coined (DOACs)
  - Push for return/continuation of ‘NOACs’ (non-vitamin K antagonist oral anticoagulants).
  - Not-so-new oral anticoagulants (NOACs)
  - Others... TSOCs (target specific), ......
  - DOACs recently endorsed by ISTH (by one vote!)
Direct oral anticoagulants

- ‘General’ indications – summarised – adults (dabigatran, apixaban, rivaroxaban):
  - Prevention of venous thromboembolic events (VTE) (major orthopaedic surgery of the lower limb)
  - Prevention of stroke and systemic embolism in non-valvular atrial fibrillation (and at least one additional risk factor for stroke).
  - Treatment of deep vein thrombosis (DVT) and pulmonary embolism (PE)
  - Prevention of recurrent DVT and PE
DOAC usage – an increasing trend:


Laboratory testing – a changing landscape

Item 65120: Prothrombin time (including INR where appropriate), activated partial thromboplastin time, thrombin time (including test for the presence of heparin), test for factor XIII deficiency (qualitative), Echis test, Stypven test, reptilase time, fibrinogen, or 1 of fibrinogen degradation products, fibrin monomer or D-dimer

Laboratory testing – a changing landscape
Direct oral anticoagulants

Contact (Intrinsic) pathway
- Contact activation
  - FXII
  - FXIIa
  - FXI
  - FIX
  - FIXa
  - FX
  - FVIIIa

Tissue Factor (Extrinsic) pathway
- Vascular injury-trauma
  - FXIIa
  - FVIIa
  - FX
  - FVII

- rivaroxaban
- apixaban

- dabigatran
dabigatran - the basics

Summary
- Affects TT > APTT > PT
- Affects dRVVT > APTT
- Some reagent variability
rivaroxaban - the basics

Summary

- Affects PT > APTT
- Affects dRVVT \( \sim \) PT
- Some reagent variability
apixaban - the basics

Summary
- Hardly affects PT, APTT
- Does affect dRVVT
- Some reagent variability
DOACs – The basics - summary

- Dabigatran can be accurately measured using dTT/DTI methods (commercial and in house)
- Apixaban & rivaroxaban can be accurately measured using anti-Xa methods (using specific drug as calibrator)
- Low level sensitivity issues (20-25ng/mL for all assays)
- High level sensitivity issues (dilute samples for accurate reading)
- For routine PT & APTT assays:
  - Dabigatran APTT > PT
  - Rivaroxaban PT > APTT
  - Apixaban – neither unless sensitive reagent
  - Generally, dRVVT more sensitive than either PT or APTT
DOACs – a summary of additional findings (data not shown)

- Although dabigatran can be accurately measured using dTT/DTI methods, apixaban & rivaroxaban do not affect dTT/DTI assays

- Although apixaban & rivaroxaban can be accurately measured using anti-Xa methods, dabigatran does not affect anti-Xa assays

- Some other tests not affected by DOACs:
  - Fibrinogen (except for some assays (IL Fib C) for higher levels of dabigatran)
  - Protein S antigen levels (free Protein S)
  - Protein C by chromogenic assay

- Many other tests affected by DOACs:
  - Include factor assays, Protein S clot based assays, APCR assays, lupus anticoagulants, antithrombin (selective)
DOACs – Antithrombin

- Dabigatran affects antithrombin assays using an anti-IIa method, but not an anti-Xa method
- Apixaban & rivaroxaban affect antithrombin assays using an anti-Xa method, but not an anti-IIa method
DOACs – factors –

- All factor assays potentially affected
- May occur if clinician wants factor assays for ‘unexpectedly’ raised APTT and/or PT
DOACs – LA

- All DOACs affect LA
- Raises both dRVVT screen and confirm, as well as (potentially) SCT/APTT, etc
- Potential false positive LA with dabigatran & rivaroxaban
- Potential false negative LA
DOACs – Factor inhibitors

- All DOACs affect factor assays
- Behave ‘like’ inhibitors
- Potential false positive factor inhibitor

- 14-03b outlier - reported 0.5BU/mL FVIII inhibitor = 'detected'. Pool of normal plasma; no inhibitor present; = “false positive” (n=1/51 = <2% ‘error rate’)
- 14-03a - dabigatran (~800ng/mL) sample; = “false positive” (n=49/51 = >97% ‘error rate’)
DOACs – Factor inhibitors and beyond

- INH15-03a = normal pooled plasma spiked with rivaroxaban (~638ng/mL)
- All method median result = 0.5 BU
- Range of reported 'inhibitor' levels undetectable to 3 BU
- Interpretations = inhibitor detected (46%) vs not detected (54%)
DOACs – RCPA QAP Module G

- 3 years worth of dabigatran surveys
  - Very reproducible findings with dTT/DTI assays
  - High concordance of medians of samples repeat tested in separate surveys
  - Moderate CVs; very few outliers; ?Revise ALP
  - Interesting mix of ‘LL/ML/HL’ with ‘detected’

- 2 years worth of apixaban & rivaroxaban surveys
  - Very reproducible findings with anti-Xa assays
  - Moderate CVs; very few outliers; ?Revise ALP
  - High concordance of medians for rivaroxaban samples repeat tested in separate surveys
  - Have not been able as yet to assess concordance from medians of apixaban samples repeat tested in separate surveys
  - Interesting mix of ‘LL/ML/HL’ with ‘detected’

- Any problems or concerns?
  - Contact the RCPA QAP!
Simple clinical view of hemostasis

Activation of platelets / coagulation factors

Lots of exciting biochemistry

“CLOT”

Simple clinical view of thrombosis

Over activation of platelets / coagulation factors

Too much exciting biochemistry

“Thrombosis”
Simple clinical view of anti-coagulant / anti-platelet therapy & monitoring:

Activation of platelets / coagulation factors

Drugs $\rightarrow$ exciting biochemistry is inhibited

$\rightarrow$

“Thrombosis” is inhibited

(easy for lab to “measure” decreased “CLOT” formation or “platelet activity”)

Do not undertake testing for:
- Factors
- LA
- ‘Thrombophilia tests’
- ‘inhibitor assays’
### Testing DOACs at Westmead - summary


**Table: Coagulation test Effect of**

<table>
<thead>
<tr>
<th>Coagulation test</th>
<th>Anti-Xa DOACs (Rivaroxaban)</th>
<th>Anti-Xa DOACs (Apixaban)</th>
<th>Anti-IIa DOACs (Dabigatran)</th>
<th>VKAs</th>
<th>UH</th>
<th>LMWH</th>
</tr>
</thead>
<tbody>
<tr>
<td>PT/INR</td>
<td>++</td>
<td>(+)</td>
<td>+</td>
<td>++</td>
<td>-/+</td>
<td>-</td>
</tr>
<tr>
<td>aPTT</td>
<td>+</td>
<td>(+)</td>
<td>++</td>
<td>+</td>
<td>++</td>
<td>-/+</td>
</tr>
<tr>
<td>TT</td>
<td>-</td>
<td>-</td>
<td>++</td>
<td>-</td>
<td>++</td>
<td>+/-</td>
</tr>
<tr>
<td>Anti-Xa</td>
<td>++</td>
<td>++</td>
<td>-</td>
<td>-</td>
<td>++</td>
<td>++</td>
</tr>
<tr>
<td>dRVVT</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>-/+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Factor assays</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>++/-</td>
<td>++/-</td>
<td>-/+</td>
</tr>
<tr>
<td>LA</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>++/-</td>
<td>-/+</td>
<td>-</td>
</tr>
<tr>
<td>PC, PS, AT</td>
<td>-/+</td>
<td>-/+</td>
<td>-/+</td>
<td>-/+</td>
<td>-/+</td>
<td>-/+</td>
</tr>
<tr>
<td>Inhib. assays</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>-/+</td>
<td>-/+</td>
<td>-/+</td>
</tr>
</tbody>
</table>

**Diagram:**

- Basic coagulation panel: PT, APTT, TT
-Optional (if available): dRVVT (using ‘LA confirm reagent’)
- Consider timing of tests in relation to last dose of DOAC
- Consider/assess specific individual test sensitivities to each DOAC
- Significant dabigatran & rivaroxaban activity can be excluded.
- PT prolonged > APTT prolonged TT normal
- Suggestive of rivaroxaban effect (exclude warfarin effect, liver disease, etc)
- Confirmatory assay: anti-Xa using specific rivaroxaban assay/standard
- dRVVT normal
- Significant rivaroxaban activity can be excluded.
- PT prolonged > APTT prolonged TT normal
- Suggestive of dabigatran effect (exclude heparin effect, low fibrinogen, etc)
- Confirmatory assay: Hemoclot or dTT using specific dabigatran assay/standard
- Anti-Xa using specific apixaban assay/standard; if no activity, assess for differential diagnosis
- dRVVT normal
- Significant apixaban activity can be excluded.
Acknowledgments

- Meeting organisers
- RCPAQAP Haematology, especially Roslyn Bonar
- Diagnostic Haemostasis laboratory staff
  - Soma Mohammed
  - Jane McDonald
  - Ella Grezchnik
  - Monica Ahuja
  - Shabana Azimulla
  - Yifang Zhang

Haemostasis = Love
Everybody talks about it, nobody understands it.