Coagulation testing in common clinical scenarios

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'Routine' coagulation tests may help to confirm and identify the cause of a bleeding disorder, but coagulation results should always be interpreted within the individual patient context and with knowledge of the testing limitations.

Routine coagulation tests incorporate an activated partial thromboplastin time (APTT) and prothrombin time (PT). They provide insight into the clotting pathways of a patient. However, when to order them and what to do with an abnormal result, especially with many patients now being prescribed nonvitamin K oral anticoagulants (NOACs; also known as direct oral anticoagulants [DOACs]), is increasingly challenging. The utility and limitations of routine coagulation testing are explored in this article through three scenarios that focus on patients who are concerned about their bleeding risk.

Haemostasis overview

Haemostasis represents the delicate balance between maintaining circulatory flow within the vasculature and the need to cease bleeding at the site of any vascular injury. It comprises the complex interplay between blood vessels, platelets, coagulation factors and the fibrinolytic system. Damage to a vessel triggers recruitment of von Willebrand factor (VWF), which facilitates platelet attachment and activation. This process is known as primary haemostasis. Secondary haemostasis represents the stepwise activation of coagulation proteins leading to fibrin production, which reinforces the initial platelet 'plug' (Figure 1).

KEY POINTS

- A thorough medical history is the mainstay of assessing patients for underlying bleeding disorders.
- Standardised bleeding questionnaires are helpful and reduce reliance on patient interpretation.
- If a bleeding disorder is suspected, initial screening tests include a full blood count, blood film, coagulation profile (activated partial thromboplastin time and prothrombin time) and fibrinogen level.
- Routine coagulation testing is not required in patients who are taking a nonvitamin K oral anticoagulant; however, it is important to note that if testing is required these medications have an impact on the results.
- Preoperative coagulation tests are unnecessary unless there is concern about a bleeding disorder.

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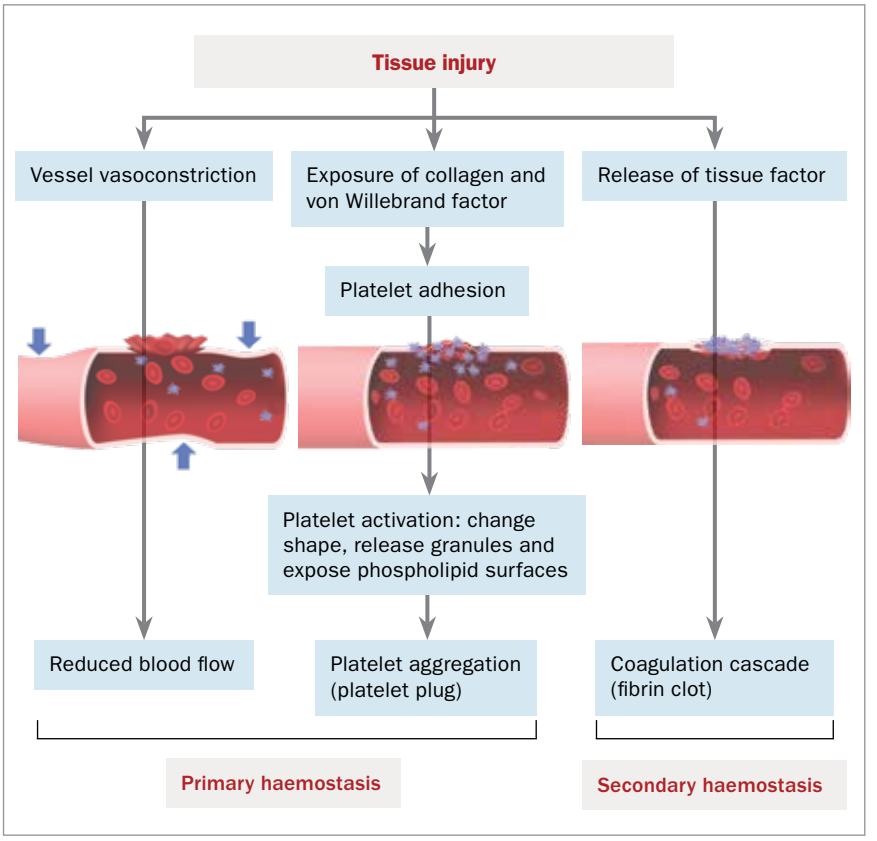

Figure 1. Overview of haemostasis.

These processes are balanced with endogenous anticoagulants (protein C, protein S and antithrombin) and fibrinolytic pathways. Protein C inactivates clotting factors V and VIII and protein S potentiates protein C activity. Antithrombin's main role is the inactivation of thrombin (factor II) and factor X. The fibrinolytic system acts to contain the clot to the site of injury and then dissolve the clot as the wound heals. If this delicate haemostatic system becomes unbalanced, it can lead to either thrombosis or haemorrhage.[1]

Routine coagulation tests – APTT and PT

Routine coagulation testing is largely designed to detect specific coagulation factor deficiencies and should not be considered a measure of overall haemostasis as it excludes several key components. Although inherently integrated in vivo, the clotting cascade is traditionally depicted as consisting of intrinsic (contact) and extrinsic (tissue factor) pathways (Figure 2). In vitro, these pathways are represented by the APTT and PT coagulation tests, respectively. To ensure an accurate and useful result, the blood sample needs to be collected in the correct tube, filled to the correct volume (the sodium citrate in the tube is measured to provide a specific ratio to blood) and transported to the laboratory in a timely manner with the clinical indication for testing and the patient's current anticoagulation therapy, if any, recorded on the request form.

For the APTT, the sample is centrifuged to obtain platelet-poor plasma and then combined with phospholipid, a contact activator (kaolin, silica or ellagic acid) and calcium (this reverses the effect of citrate and enables the blood to clot again). The test result is the time (in seconds) from the addition of calcium to the formation of a fibrin clot. The APTT test is sensitive to the presence and activities of factors II, V, VIII, IX, X, XI, XII and fibrinogen (factor I). It is important to discuss with your local laboratory whether their APTT reagent is sensitive or insensitive to the presence of a lupus anticoagulant.

The PT represents the time taken for the blood to clot after platelet-poor plasma is combined with thromboplastin (tissue factor and phospholipid) and calcium. The PT is sensitive to the presence and activity of factors II, V, VII, X and fibrinogen (Figure 1). The international normalised ratio (INR) is derived from the PT and adjusts for differences in the PT reagents over time and between laboratories (thereby allowing monitoring of patients regardless of where testing is performed). Most laboratories report both PT and INR values whenever a PT test is performed; however, the INR is applicable only for patients taking warfarin.

Causes of abnormal PT and APTT are outlined in Table 1. NOACs have intentionally been excluded here and are discussed below.

Scenario 1

A young woman presents to your practice. She describes her periods as heavy and has noticed a few bruises on her arms and legs and cannot remember how she got them. She has occasional nosebleeds in winter.

Minor bleeding, although sometimes alarming, is common in otherwise healthy people.[2] A thorough history should include the type of bleeding, when it was first noticed (e.g. menorrhagia since menarche is suggestive of a possible bleeding disorder), the frequency and the spontaneous or provoked nature of the bleeding including any relationship with haemostatic challenges (including trauma, surgery, dental extractions and pregnancy). Disorders of primary haemostasis are primarily due to abnormal levels and/or function of both VWF and platelets. They tend to manifest as mucocutaneous bleeding.

Secondary haemostatic defects are primarily due to abnormal levels and/or function of clotting factors and tend to manifest as bleeding in joints or deep tissues.[3]

Standardised bleeding questionnaires have been generated to help assess bleeding symptoms using clear objective criteria to reduce reliance on patient interpretation. Haematologists commonly use the International Society on Thrombosis and Haemostasis Bleeding Assessment Tool in clinical practice.[4] This assessment tool covers all areas of haemostasis in bleeding disorders such as bruising, epistaxis, dental extraction, menses, childbirth, muscle haematomas and gastrointestinal bleeding. The patient's score increases if their bleeding has required medical consultation and further increases if intervention was required, particularly blood transfusion.

A review of all medications including over-the-counter medications, especially anti-inflammatories, and herbal supplements should be obtained. Family history is also vital in the assessment of a potential bleeding disorder. Finally, clinical examination for any bleeding signs (such as petechiae, ecchymoses and haemarthroses), joint hyperflexibility (associated with connective tissue disorders), stigmata of chronic liver disease and any evidence of underlying congenital abnormalities should be performed.[5]

If a bleeding disorder is considered possible, initial screening laboratory tests include a full blood count, blood film, coagulation profile (APTT and PT) and fibrinogen level. The FBC and film should identify thrombocytopenia as well as any abnormal platelet morphology, which may suggest a specific diagnosis. For example, the inherited platelet function disorder Bernard Soulier syndrome is characterised by thrombocytopenia and large platelets from childhood. Renal and liver function tests are also recommended, as renal failure can cause acquired platelet dysfunction and liver failure reduces the production of all procoagulant and anticoagulant proteins except factor VIII and VWF.

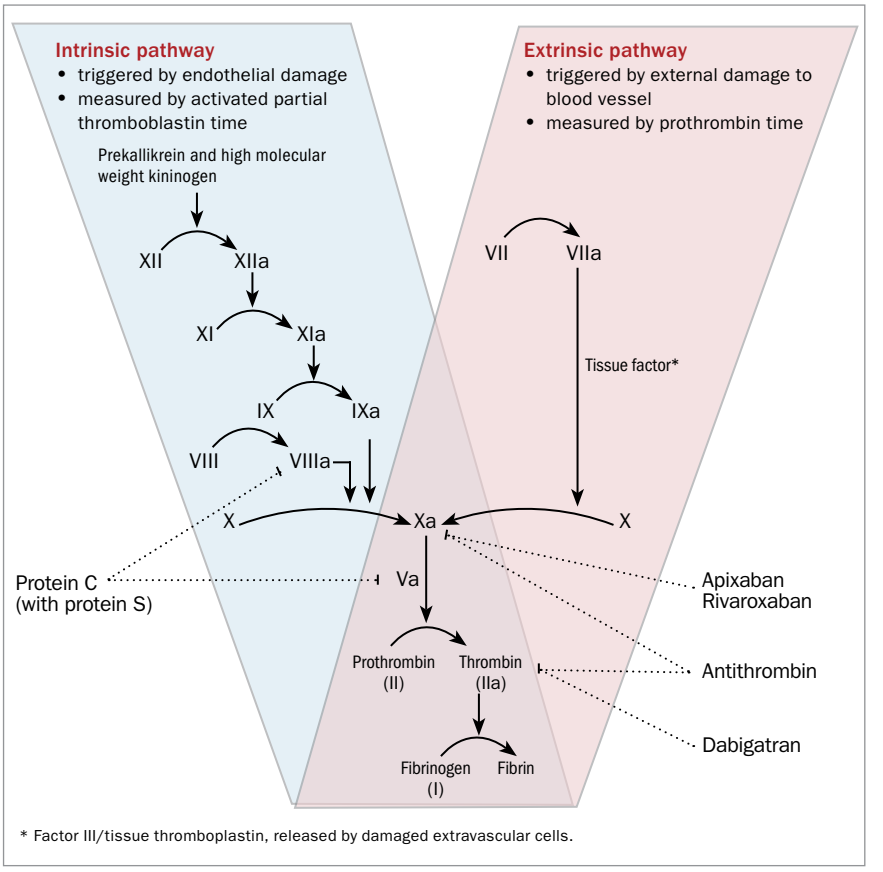

Figure 2. Coagulation cascade showing inhibition of coagulation by endogenous anticoagulants and nonvitamin K oral anticoagulants.

If the patient has a prolonged PT and/or APTT, the next step we recommend when investigating for a bleeding disorder is ordering a mixing study. This involves combining 50% of the patient's plasma with 50% normal plasma and repeating the test. If the cause for the test prolongation is deficiency in one or more clotting factors, they will reach sufficient levels by adding in the normal plasma and hence the mixed sample will provide a normal result. If this is the case, the next step is to quantify the patient's factor levels (i.e. factors VIII, IX, XI and XII for APTT) that contributed to the original abnormal test result. A mixing study that does not correct implies the presence of a factor inhibitor. If the patient is not taking an anticoagulant and testing is negative for lupus anticoagulant (which acts as an inhibitor in vitro but leads to thrombosis, not bleeding, in vivo), we would recommend discussion with a haematologist, as a factor inhibitor can represent a life-threatening condition.

If the patient's symptoms are suggestive of a primary haemostatic defect and/or the factor VIII level is decreased, we also recommend assessing for von Willebrand disease. This comprises a panel of tests that assess the level and function of the patient's VWF. It is worth noting that VWF levels fluctuate with inflammation, infection, pregnancy, hormonal therapy, cancer and acute stress. Therefore, if clinical suspicion is high, the tests should be repeated.[6]

All patients with an identified bleeding disorder should be referred to a haematologist for ongoing management. It is not

TABLE 1. CAUSES OF ABNORMAL PROTHROMBIN TIME (PT) AND ACTIVATED PARTIAL THROMBOPLASTIN TIME (APTT)

Laboratory finding	Cause	Notes
Prolonged APTT	Deficiency of factors VIII, IX, XI	• In general, the factor level must be less than 20 to 40% of normal before the APTT is prolonged. These conditions are all clinically significant and include haemophilia A (factor VIII), haemophilia B (factor IX) and haemophilia C (factor XI) • Von Willebrand factor is the carrier protein for factor VIII, so some types of von Willebrand disease can also lead to low factor VIII levels and a prolonged APTT
	Deficiency of factor XII	• Not associated with clinical bleeding
	Acquired factor inhibitors	• Usually develop against factor VIII. Can present with severe clinical bleeding
	Lupus anticoagulant	• Only if laboratory uses APTT reagent sensitive to the presence of a lupus anticoagulant • Although lupus anticoagulant prolongs APTT in vitro, it causes thrombotic tendency in vivo
	Heparin therapy	• Usually performed on hospital inpatients with clear therapeutic ranges
Prolonged PT	Warfarin	• Interferes with vitamin K dependent modifications of factors II, VII, IX and X. Can prolong APTT as well, particularly if a patient is over-warfarinised
	Factor VII deficiency	• Rare
Prolonged APTT and PT	Vitamin K deficiency	• Associated with malnutrition and fat malabsorption syndromes
	Liver disease	• Can reduce production of procoagulant and anticoagulant factors; therefore, PT/APTT do not always accurately reflect the in vivo haemostatic picture
	Disseminated intravascular coagulation	• Depletes clotting factors
	Massive transfusion	• Dilutional coagulopathy can develop
	Thrombolysis therapy	
	Combined factor deficiencies and common factor deficiencies (factor X, factor V, factor II, fibrinogen)	• May be congenital or acquired
Short APTT	High factor VIII levels	• Acute-phase response
	Difficult collection	• Can activate clotting factors

uncommon that, despite these screening investigations, a cause is not immediately apparent. If suspicion remains high based on the patient's personal and/or family history, a haematology referral should be considered. Further investigations may include platelet function analysis and more detailed coagulation testing, which are beyond the scope of this article.

Scenario 2

A 65-year-old patient recently changed from warfarin to apixaban for atrial fibrillation. He liked knowing his INR and asks if you can check this new medication 'just to be sure'.

The NOACs currently include the direct thrombin inhibitor dabigatran and the activated factor X (factor Xa) inhibitors apixaban and rivaroxaban. Clinical trials and real-world data show that these medications have similar or superior efficacy and safety profiles to vitamin K antagonists. Combined with the fact that they do not require routine laboratory monitoring, they have understandably revolutionised the management of atrial fibrillation and venous thromboembolism.[7,8]

However, there are disadvantages that should be acknowledged. These medications are more expensive, and a missed dose can result in a lack of appropriate anticoagulation for 12 to 24 hours (depending on the medication). Although dabigatran may be reversed with idarucizumab, there is no reversal agent available in Australia for apixaban or rivaroxaban. In addition, they should not be used in patients with prosthetic heart valves, antiphospholipid syndrome, severe renal impairment or severe liver disease, or who are pregnant or breastfeeding. Although there is accumulating evidence for the efficacy of these drugs in malignancy-associated venous thromboembolism and heparin-induced thrombocytopenia, we recommend discussion with a haematologist before prescribing in these contexts.[9,10]

NOACs can cause confusion when routine coagulation tests are ordered. Importantly, although none of the NOACs

TABLE 2. EFFECTS OF ANTICOAGULANTS ON COAGULATION TEST RESULTS

Medication	Mechanism of action	Test result if anticoagulant present	Test to quantify drug (if needed)
Heparin	Enhances antithrombin effect on thrombin and factor Xa	Prolongs APTT	APTT or drug-specific anti-Xa assay
Low-molecular-weight heparin	Enhances antithrombin effect on factor Xa	May prolong APTT but cannot be reliably excluded even with normal PT/APTT	Drug-specific anti-Xa assay
Apixaban	Factor Xa inhibitor	May prolong PT but cannot be reliably excluded even with normal PT/APTT	Drug-specific anti-Xa assay
Rivaroxaban	Factor Xa inhibitor	Prolongs PT (with sensitive reagent)	Drug-specific anti-Xa assay
Dabigatran	Direct thrombin inhibitor	Prolongs TT* and APTT	Dilute TT

Abbreviations: APTT = activated partial thromboplastin time; factor Xa = activated factor X; NOAC = nonvitamin K oral anticoagulant; PT = prothrombin time; TT = thrombin time.
* TT measures the time taken after addition of exogenous thrombin for conversion of fibrinogen to fibrin, the final step in clot formation, common to the intrinsic and extrinsic pathways.

are monitored with the traditional PT/INR or APTT, they will affect these tests in various ways (Table 2). The only way to ascertain a drug level is by performing a drug-specific assay, and discussion with the testing laboratory should take place before requesting this.

NOACs can also have a significant impact on more specialised coagulation testing including factor levels, protein C, protein S, antithrombin and lupus anticoagulant testing (depending on the method used). We strongly recommend discussion with your laboratory before ordering these tests on a patient taking a NOAC to avoid a misdiagnosis.

We do not routinely perform any laboratory studies to monitor patients who

TABLE 3. TIMING OF NOAC CESSATION BEFORE SURGERY*[15]

Anticoagulant	Renal function (creatinine clearance)	Timing of last dose before surgery if low bleeding risk	Timing of last dose before surgery if high bleeding risk
Dabigatran	≥80 mL/min	24 hours	48 hours
	50 to 79 mL/min	24 to 48 hours	48 to 72 hours
	30 to 49 mL/min	48 to 72 hours	96 hours
Apixaban	>50 mL/min	24 hours	48 to 72 hours
	30 to 50 mL/min	48 hours	72 hours
Rivaroxaban	>50 mL/min	24 hours	48 to 72 hours
	30 to 50 mL/min	48 hours	72 hours

Abbreviation: NOAC = nonvitamin K oral anticoagulant (also known as direct oral anticoagulant [DOAC]).
* Bridging therapy is not required.

are taking NOACs. We also endeavour to order all necessary specialised testing before these drugs are started. In our experience, the most common real-world scenarios in which drug levels are ordered are when an urgent surgery or invasive procedure is required (to ensure the drug level is low/undetectable), when therapy appears to have failed, or in the case of an overdose.

Scenario 3

A 78-year-old woman presents to you before her knee replacement. She remembers bleeding when she had her tonsils removed as a child and wants to discuss the upcoming surgery.

Routine coagulation tests are frequently performed in the preoperative setting in the belief that they may identify patients who have bleeding disorders and thereby reduce the risk of perioperative bleeding. However, indiscriminate screening will only very rarely identify previously undetected individuals. In contrast, prolongation of the APTT may be due to mild factor XII deficiency, high molecular weight kininogen deficiency, prekallikrein deficiency, a collection issue or the presence of a lupus anticoagulant, none of which are associated with a bleeding phenotype. Also, defining the normal range as the mean plus two standard deviations in healthy subjects means that 2.5% of normal individuals will yield a prolonged result. Therefore, if coagulation testing were performed on every patient preoperatively, it would be likely to lead to unnecessary testing, patient concern, increased costs and procedural delay without any clinical relevance.[11]

It is also dangerous to presume that all bleeding disorders will be detected by PT and APTT. Conditions that may not be identified include von Willebrand disease, platelet function disorders, factor XIII deficiency, connective tissue disorders and fibrinolytic pathway deficiencies. The British Committee for Standards in Haematology guideline therefore recommends against ordering coagulation tests to predict perioperative bleeding risk before surgery or other invasive procedures in patients without any bleeding history.[12] This recommendation is also shared by the guidelines of the European Society of Anaesthesiology and the French Society of Anaesthesia and Intensive Care.[13,14]

We therefore advocate that a bleeding history including any evidence of excessive post-traumatic or postsurgical bleeding, family history and medication review be undertaken in all patients before surgery or invasive procedures. Only then, if there is concern for a bleeding disorder, should testing incorporating a coagulation profile (APTT and PT) and fibrinogen level be done. Preoperative interventions may then include antifibrinolytics such as tranexamic acid, platelets or clotting factor replacement depending on the underlying condition detected.

If a patient requires urgent surgery, concern for a potential bleeding disorder should not prevent lifesaving procedures. If surgery can be safely delayed until further work-up can be done, this is preferable. Other acute conditions that can influence haemostasis and warrant preoperative coagulation testing include liver disease, sepsis, diffuse intravascular coagulation and poor nutritional states (which can lead to vitamin K deficiency).

In summary, in the absence of a definitive bleeding history we do not recommend routine coagulation testing before surgeries/procedures. We also do not recommend routinely checking NOAC levels before elective surgeries. Rather, we recommend withholding the medication beforehand, basing the timing of cessation on the bleeding risk of the surgery, the half-life of the drug and the renal function of the patient (Table 3).

Conclusion

A thorough medical history remains the mainstay of assessing patients preoperatively and for possible bleeding disorders. If required, judicious coagulation testing can then provide significant insight into any underlying primary or secondary haemostatic condition. Routine testing for patients on NOACs is not required; however, these medications can influence the results. Therefore, coagulation results should always be interpreted within the individualised clinical context and with knowledge of the testing limitations. MT

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