

Prothrombin time is the time required for the plasma to clot after an excess of thromboplastin and an optimal concentration of calcium have been added.

The PT used to determine the clotting tendency of blood, in the measure of warfarin dosage, liver damage, and vitamin K status.

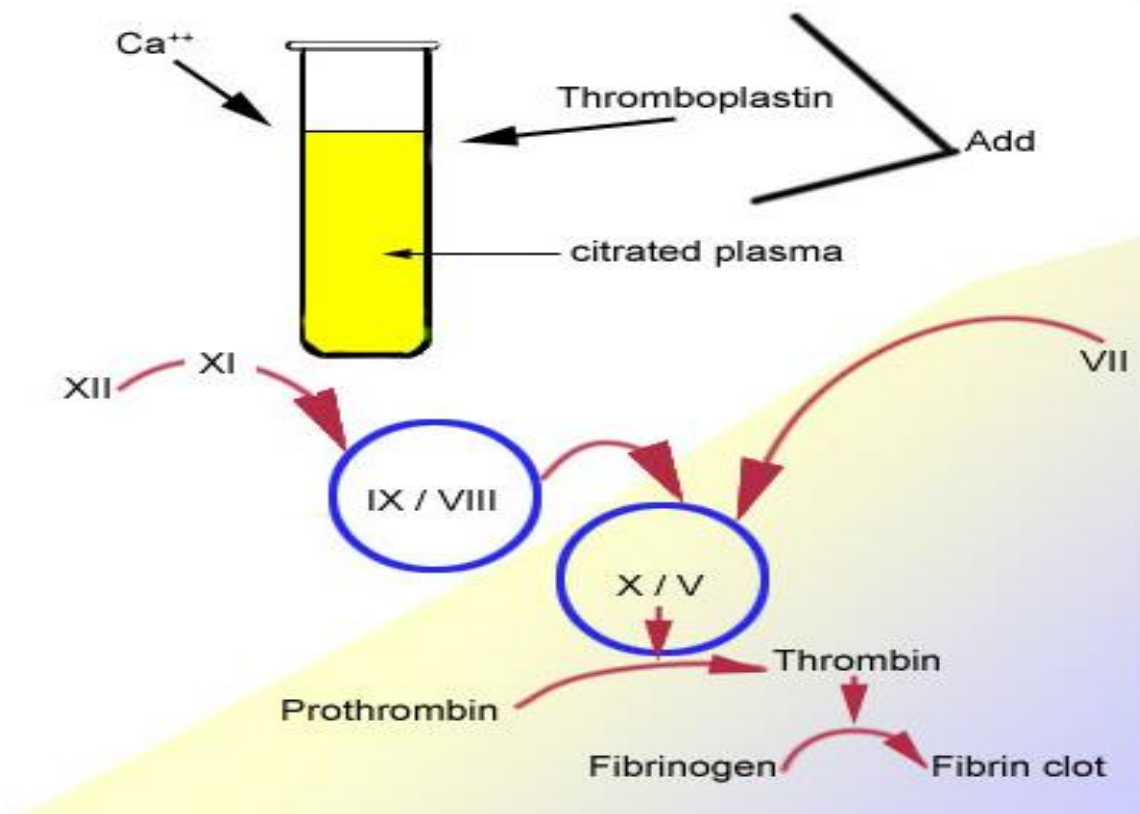
The diagram illustrates the blood coagulation cascade, divided into three main phases: **Initiation**, **Amplification**, and **Propagation**.

- Initiation:**
 - Extrinsic Pathway:** Tissue factor and FVII activate FVII to FVIIa. FVIIa then forms the **TF-VIIa complex** with Tissue factor.
 - Intrinsic Pathway:** High MW kininogen is converted to Bradykinin by Prekallikrein/kallikrein. FXII is converted to FXIIa, which then activates FXI to FXIa. FXIa activates FIX to FIXa. FIXa, along with FVIIIa (formed from FVIII by FIIa), forms the **"Tenase complex"**.
- Amplification:**
 - The **TF-VIIa complex** and the **"Tenase complex"** both activate FX to FXa.
 - FXa, along with FV and FIIa (formed from FII by FXa), forms the **"Prothrombinase complex"**.
 - The **"Prothrombinase complex"** converts FII (Prothrombin) to FIIa (Thrombin).
- Propagation:**
 - FIIa (Thrombin) converts Fibrinogen to Soluble fibrin.
 - Soluble fibrin, along with FXIIIa (formed from FXIII by FIIa), is converted to **Insoluble crosslinked fibrin** by Ca²⁺.
 - Thrombin (FIIa) also activates FVIII to FVIIIa and FXII to FXIIa, feeding back into the initiation phase.

Principle:

When reagent thromboplastin--to which calcium has been added--is mixed with plasma (derived from sodium citrated whole blood), the time (in seconds) it takes for the formation of a clot is reported as the Prothrombin time (PT).

Calcium is necessary for the correct orientation and binding of a number of complexes including : tissue factor-VIIa, IXa-VIIIa, and Va-Xa.

**SPECIMEN:**

Citrated plasma: 1 part of sodium citrate solution (0.11 mol/ L) to 9 part of venous blood, avoiding the formation of foam.

Control: normal plasma (Commercial, Pooled Plasma).

EQUIPMENTS:

- 100 μ L micropipettes (0.10 mL)

- Stop Watch.
- Reagent
- Test tubes
- Water bath (37°C)

Procedure:

1. Bring all reagents, controls and sample to room temperature 15 minutes prior to testing.
2. Pre-warm PT reagent at 37°C for 5 minutes.
3. Pipette 100µl of PT reagent to each tube.
4. Add 50 µl of sample, controls to the tubes prepared in step 3, start stop watch , mix in a water bath (37°C) for 8 seconds , then record the time required for clot formation .

RESULTS:

Prothrombin Time Ratio (PTR) = Clot time of the test plasma / Clot time of the control plasma

Reference ranges:

- ❑ PT: 11.0 – 13.0 seconds.
- ❑ Normal control sample: (11-16 seconds)
- ❑ PTR: 1.0 ± 0.15

When is it ordered?

- ↻ Used to monitor oral anticoagulant therapy (Warfarin / Coumadin).
- ↻ When a patient who is not taking anti-coagulant drugs has signs or symptoms of a bleeding disorder
- ↻ When a patient is to undergo an invasive medical procedure, such as surgery, to ensure normal clotting ability.

An elevated Prothrombin time may indicate the presence of

1. Vitamin K deficiency
2. DIC
3. Liver disease
4. Presence of FSP's
5. A deficiency in one or more of the Concerning factors: Factor I (Fibrinogen), Factor II (Prothrombin), Factor V (Proaccelerin, Labile Factor), Factor VII (Proconvertin, Stable Factor, Factor X (Stuart-Prower Factor, Factor XIII (Fibrin Stabilizing Factor)

In addition, inhibitors can cause prolonged PT's

Interpretation of Result:

- ☐ A Normal Plasma is used to evaluate routine result.
- ☐ Patients with lupus anticoagulants are not be requested for PT as they have antiphospholipid

Interfering Factors:

- ☐ Diet: ingestion of excessive green, leafy vegetables will increase the absorption of vit-K, which promotes blood clots.
- ☐ Alcoholism, Prolonged PT levels
- ☐ Diarrhea and vomiting decrease PT because of dehydration.
- ☐ Quality of Vein puncture.
- ☐ Medication : Antibiotics , Aspirin, Cimethidine....
- ☐ Prolonged Storage of plasma at 4° C.