

# Tests of hemostasis

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## Hemostasis

Haemostasis means prevention of blood loss from blood vessels.

Bleeding is stopped by several mechanisms, which are:

- 1. Local vasoconstriction**
- 2. Formation of platelet plug**
- 3. Blood coagulation**

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## 1. Local vasoconstriction

- Immediately after injury, constriction of an injured arteriole or small artery may be so marked that its lumen is obliterated. The vasoconstriction is probably due to:
  - Liberation of serotonin and other vasoconstrictors from platelets.
  - Local myogenic contraction of the blood vessel.

## 2. Formation of platelet plug

Platelets adhere to damaged endothelium to form a platelet plug (***primary hemostasis***). Platelets adhere to the collagen fibers of a wound, then release chemical messengers such as adenosine diphosphate (ADP), serotonin and thromboxane  $A_2$ , causing more platelets to stick to the area, release their contents, and enhance vascular spasms. As more chemicals are released more platelets stick and release their chemicals; creating a platelet plug.

Hemostasis

### 3. Blood coagulation

Blood coagulation is dependent upon a number of factors, which interact to produce the prothrombin conversion factor, which then convert prothrombin to thrombin and the later convert the fibrinogen to fibrin, which appears to be important in stabilizing the platelet thrombus. Fibrin mesh helps hold the plug in place. Red and white blood cells become caught up in the fibrin mesh which causes the clot to become even stronger. This step of coagulation is referred to as **secondary hemostasis**

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Hemostasis

### Why is platelet adhesion clinically important?

Platelet adhesion is required for primary hemostasis. Platelets do not adhere to healthy endothelium. Intact endothelial cells secrete antithrombotic substances such as *prostacyclin (PGI)*, a prostaglandin and platelet inhibitor. Platelets are also repelled by the negatively charged surface of intact endothelium.

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## Tests of hemostasis

1. Whole blood coagulation time (Clotting time).
2. Bleeding time.
3. Prothrombin time (PT).
4. Thrombin time (TT).
5. Platelet count (Thrombocytes).

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### Tests of hemostasis

#### 1. Whole blood coagulation time (Clotting time)

It is the time that elapses from the start of bleeding till the formation of clot.



#### 2. Bleeding time

It is the time needed for bleeding to stop when small injury of skin occurs.

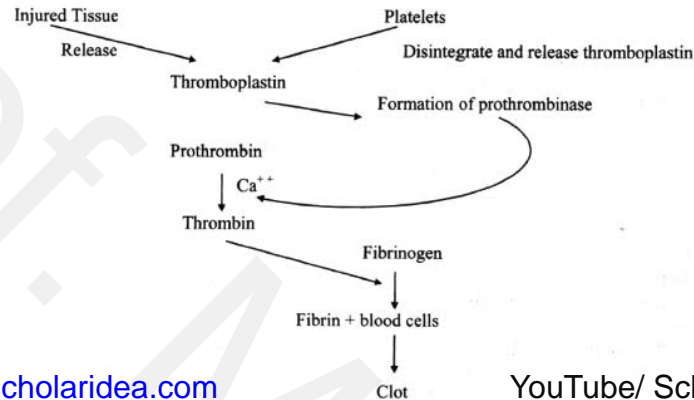
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**3. Prothrombin time (PT)**

It is the time required for clotting to take place in citrated plasma to which optimum amount of thromboplastin and calcium ions have been added. It is prolonged with severe liver disease or vitamin K deficiency.



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**4. Thrombin time (TT)**

This is the time taken for fibrin to be formed after the addition of thrombin to citrated plasma sample. It is a measure of the amount of fibrinogen present and the ability to convert it to fibrin. A prolonged thrombin time is found with a deficiency of fibrinogen or with difficulty in the production of fibrinogen.

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## 5. Platelet count (Thrombocytes count)

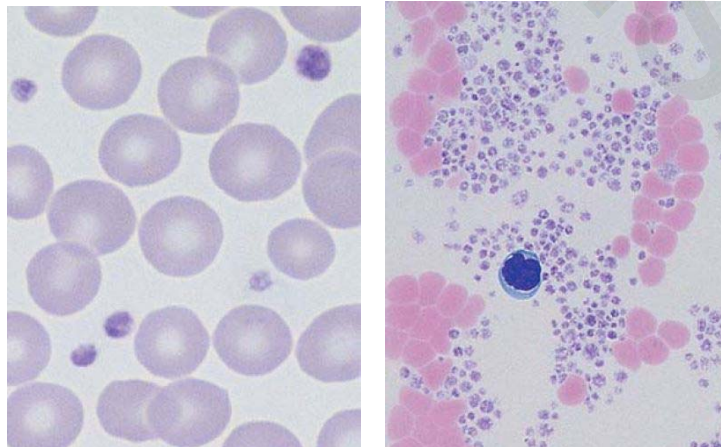
Platelets in mammals are fragments that contain small pink-red granules. Shed into the blood from megakaryocytes in bone marrow

Like all circulating blood cells, platelets are bone marrow derived. The first recognizable platelet precursor is the megakaryoblast, which undergo endomitosis (nuclear division without cytoplasmic division) to form megakaryocytes.

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On routine blood film, platelets are recognized as small anucleate discoid cytoplasmic fragments containing variable numbers of purple granules.



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## Platelet Lifespan

As with all other circulating cells, the platelet has a finite circulating lifespan. Dog platelets circulate for approximately 5-7 days. While cat platelets survive only a little more than a day. Cells of the monocytes and macrophages are responsible for the removal of effete platelets. Nearly half are removed by splenic macrophages and a third by macrophages of the liver.

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### 5. Platelet count (Thrombocytes)

#### Methods of counting

##### 1. Direct method

- a. Haemocytometer.
- b. Automatic blood cell counter.

##### 2. Indirect method: blood film.

The platelets per oil immersion field on a stained blood smear are counted and compared with the number of red or white cells. For example, the number of platelets per 100 white blood cells multiplied by the total white count is an estimate of the platelet count. Another method is to simply count the number of platelets per oil immersion field where one /oil is equivalent to 15,000/ul.

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## **HEMORRHAGIC DISORDERS**

### **I. Abnormalities of blood platelets**

### **II. Hemorrhagic disorders due to defect in the clotting mechanisms.**

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## **HEMORRHAGIC DISORDERS**

### **I. Abnormalities of blood platelets**

#### **A. Quantitative Abnormalities**

- a. Thrombocytopenia.**
- b. Thrombocytosis.**
- c. Thrombocythemia.**

#### **B. Qualitative Abnormalities**

- a. Thrombocytopathia**

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### a. Thrombocytopenia

Thrombocytopenia, or reduction in the number of circulating platelets, is caused by one of four mechanisms:

1. Increased peripheral utilization of platelets.
2. Increased destruction of platelets.
3. Increased sequestration of platelets.
4. Reduced platelet production in the bone marrow.

### 1. Increased peripheral utilization of platelets

Increased peripheral utilization results when there is increased systemic demand for platelets.

**This occurs in two conditions:**

- Disseminated intravascular coagulopathy (DIC)
- Blood loss.

### **Disseminated intravascular coagulopathy (DIC)**

DIC is a secondary syndrome associated with underlying severe disease. In most cases, the underlying process is inflammatory. but DIC also occurs in some cases of neoplasia, marked tissue necrosis and shock. Regardless of the inciting cause, DIC is a syndrome where excessive stimulation of the coagulation cascade leads to the peripheral consumption of both coagulation factors and platelets.

#### **Laboratory findings:**

- Thrombocytopenia.
- Prolonged prothrombin time
- Decreased fibrinogen levels.

### **2. Increased destruction of platelets**

In case of immune-mediated thrombocytopenia may be caused by circulating antiplatelet antibodies

### **3. Increased sequestration of platelets**

Thrombocytopenia can occur in cases of hepatomegaly or splenomegaly as a result of sequestration of platelets in the enlarged organs. This condition is much more common in humans than in animals, and rare in dogs and cats. Hypothermia has been demonstrated to cause platelet sequestration in the liver. Thrombocytopenia in endotoxemia is believed to be at least partially the result of sequestration in the lung.

#### 4. Reduced platelet production in the bone marrow

Hypoproliferative thrombocytopenia is the direct result of reduced megakaryocytopoiesis.

In most cases, there is reduced production of at least one other cell line; hemograms generally reflect anemia and/or leukopenia in addition to thrombocytopenia.

#### HEMORRHAGIC DISORDERS

##### b. Thrombocytosis

Increase the number of circulating blood platelets.

Most cases of thrombocytosis are secondary or reactive. Thrombocytosis can be seen - secondary to splenic contraction (eg, with excitement or exercise), elevated circulating glucocorticoids, splenectomy. For the most part, reactive thrombocytosis is clinically insignificant.

### c. Thrombocythaemia

Means persistent increase in circulating blood platelets.

**Causes:**

- Megakaryoblastic tumors.
- Over production of thrombopoietin from the kidney.
- Polycythemia Vera.

### d. Thrombocytopathia

Platelet normal in number but abnormal in function.

These platelets may have a normal or abnormal appearance, they may be:

- Defective in adhesiveness.
- Defective in aggregation.

**Laboratory findings:**

- Hemorrhagic diathesis.
- Normal levels of blood coagulation factors.
- Normal platelet count.
- Prolonged bleeding time.

## II. Haemorrhagic disorders due to defect in the clotting mechanisms

- An increase in the clotting time, with normal values for the bleeding time and platelet count, indicates existence of haemophilia i. e. a deficiency of one or more of the factors necessary for normal coagulation.

- An increase in both the clotting time and bleeding time (usually associated with normal level of platelets) most probably indicates a deficiency of prothrombin (Hypoprothrombinemia). This is usually due to either:
  - Liver disease as hepatic toxins, hepatitis or obstructive jaundice.
  - Warfarin poisoning.
  - **Vitamin K deficiency.**
    - a. Inadequate diet from excessive antibiotics either as medicaments or in feed, which interfere with bacterial synthesis of vitamin K.
    - b. Deficiency of bile, which interfere with absorption of vitamin K.

**Summary of changes in some  
diseases causing increased bleeding  
tendency**

Test	Hypoprothrom- -binaemia	Thromb- cytopenia	Haemophilia	Traumatic
Clotting time	Increased	Usually normal	Increased	Normal
Bleeding time	Increased	Increased	Normal	Normal
Thrombocyte count	Normal	Decreased	Normal	Increased
Prothrombin time	Increased	Normal	Usually normal	Normal

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