HEPARIN INDUCED THROMBOCYTOPENIA

Essay

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List of Abbreviations

Abb.	Full term
ACCP	the American College of Chest Physicians
ADP	Adenosine diphosphate
ATP	Adenosine triphosphate
COX	Cyclo-oxygenase enzyme
CPB	cardiopulmonary bypass
CVD	Cardio vascular disease
CVVH	continuous venovenous hemofiltration
CVVHD	continuous veno-veno hemodialysis
DIC	Disseminated intravascular coagulation
DTI	direct thrombin inhibitor
DVT	Deep venous thrombosis
ELISA	Enzyme-linked immunosorbent assay
HELLP	Hemolysis , Elevated Liver enzymes , Low Platelet count
HIPA	heparin-induced platelet aggregation assay
HIT	Heparin induced thrombocytopenia
HITT	Heparin induced thrombocytopenia and thrombosis
Ig	Immune globulin
IL	Inter leukin
INR	international normalized ratio
LMWH	Low molecular weight heparin
OCS	Open canalicular system
OD	optic density
PF4	Platelet factor 4
PSGL	P-selectin glycoprotein ligand
SRA	serotonin release assay
UFH	Unfractionated heparin
VWF	Von Willibrand factor

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Introduction

Heparin-induced thrombocytopenia (HIT) is not only a common but also a potentially serious drug adverse effect. Unlike other drug-induced thrombocytopenias, HIT does not usually cause bleeding, but instead causes thrombosis. Thrombosis in HIT can lead to limb gangrene (requiring leg amputation) or even death.

Heparin is widely used for thromboprophylaxis or treatment in many clinical situations, including cardiovascular and orthopaedic surgery and invasive procedures, acute coronary syndromes, venous thromboembolism, atrial fibrillation, peripheral occlusive disease, dialysis, and during extracorporeal circulation. ***(Chong , 2003)***

One third of hospitalised patients in the USA, or about 12 million a year, receive heparin**.** ***(Mahaffey and Lewis , 2000)***

Heparin-induced thrombocytopenia (HIT) is the most important and most frequent drug-induced type of thrombocytopenia. It is associated with significant morbidity and mortality if unrecognised.

Unfortunately, because thrombocytopenia is common in hospitalised patients and can be caused by a variety of factors, HIT often remains unrecognised and undiagnosed . ***(Wehler and Mehler , 2002)***

HIT may develop in two distinct forms: type I and type II. HIT type I (also known as heparin-associated thrombocytopenia) is a non-immunologic response to heparin treatment, mediated by a direct interaction between heparin and circulating platelets causing platelet clumping or sequestration. HIT type I affects up to 10% of patients, usually occurs within the first 48–72 h after initiation of heparin treatment, and is characterised by a mild and transient thrombocytopenia (rarely <100 000/mm^3), often returning to normal within 4 days once the heparin is withdrawn**.** ***(Franchini , 2005)***

No laboratory tests are required to diagnose HIT type I, and it is not associated with an increased risk of thrombosis, whereas HIT type II is immune-mediated and associated with a risk of thrombosis. It has recently been proposed that the term “HIT type I” be changed to “non-immune heparin associated thrombocytopenia” and that the term “HIT type II” be changed to “HIT” to avoid confusion between the two syndromes. ***(Rice , 2004)***

Physiology of platelets

Platelets were discovered by Giulio Bizzozero in 1882,but for many decades the dynamic and multifunctional nature of platelets remained a field of interest only for biologists. Anucleate, discoid platelets are the smallest blood particles which unveil their dynamicity through their morphology. ***(Ribatti and Crivellato,2007)***

Primarily they are associated with hemostasis, which is to initiate blood coagulation. Although very dynamic, they usually prefer to remain in inactive state and get activated only when a blood vessel is damaged. But hemostasis or blood coagulation is not the sole function of platelets; rather it is employed in several multifunctional attributes monitoring the homeostasis of the body. Its high sensitivity to different disease states eventually assigned it to be one of the most accessible markers. While keeping interactions with leukocytes and endothelial cells, it restores its behaviour as an important inflammatory marker ***(Cerletti et al.,2012)***.

Primarily, platelet activity is associated with the initiation of coagulation cascades. Damage in blood vessel makes the subendothelial surface the primary target site of

platelet action, where it establishes the hemostasis. Various pro-aggregatory stimuli also known as platelet agonists promote the action of platelet adhesion to the subendothelial surfaces. During this process, platelet changes its shape, releases its granule contents, and gradually forms aggregates by adhering with each other ***(Vinik et al.,2001)***. Thus its primary activity remains associated with minimizing blood loss. However, as discussed earlier platelets are not only confined in regulating hemostasis and thrombosis, but they also play many pivotal roles in disease pathophysiology. ***(Vinik et al.,2001)***

Platelet interaction and cardiovascular disease progression remain an unsolved riddle for many years. Platelet hyper-aggregation among the diabetic patients with CVD remains another striking area to explore. Platelet hyperactivity in various diseases provokes adverse effects in some cases, especially in coronary artery disease where hyperaggregation obstructs blood circulation.***(Sharma and Berger ,2011)***

Versatility of Platelets: Its Structural and Functional Aspects

Ultrastructure

Platelet ultrastructure reveals its behavioural peculiarities. Megakaryocytes of the bone marrow are site of platelet formation. Diameter of a mature platelet is 2-3 μm, which usually remains alive for 5–9 days. Approximately 2/3 of the platelets circulate in the blood and 1/3 is stored in the spleen. The normal platelet count is (150–400) × 10^3 per microliter of blood. Each megakaryocyte can produce 5000–10000 platelets. An average healthy adult can produce 10^{11} platelets per day; old platelets are destroyed by phagocytosis in the spleen and liver (Kupffer cells).

Platelets are unique in their structural assembly, though they are anucleate but have distinct mitochondria. Platelet plasma membrane, composed of phospholipid bilayer, is the site of expression of various surface receptors and lipid rafts which helps in signalling and intracellular trafficking. These markers include CD36, CD63, CD9, GPCR, IIbIIIa, and GLUT-3. These surface receptors also trigger the release of α granules which play a role in multiple functions, namely, coagulation, inflammation, atherosclerosis, antimicrobial host defense, angiogenesis, wound repair, and tumorigenesis ***(Blair and laumenhaft ,2009)***

Among these surface receptors, GPCR has been reported to play crucial role in ADP secretion from dense granules which is its major secretory product ***(Offermanns ,2006)***.

Asymmetrically arranged phospholipids (e.g., phosphatidylserine and phosphatidylinositol) present in the inner layer of the plasma membrane maintain the stability of its surface during nonprocoagulant state. During activation (Figure 1) platelet surface gradually exposes aminophospholipids by ATP-dependent floppases and scramblases to initiate coagulation cascades ***(Heemskerk et al.,2002)***. The open canalicular system (OCS) is the "tunnel" system present throughout the platelet cell and remains connected with the plasma membrane ***(Behnke and Forer,1998)***. The major role of OCS is to give entry of external elements into the platelets as well as to release its granule contents to the exterior. Other than being a major storage site for plasma membrane glycoproteins, it facilitates the formation of filopodia during platelet activation ***(Michelson ,1992)***.

Dense tubular system of platelets is a closed-channel network of residual endoplasmic reticulum and primarily involved in calcium sequestration with the help of cascades of reactions involving the activation of G protein-coupled receptor PAR-1 ***(Brass et al.,1993)***. The highly specialized

cytoskeleton of platelets maintains its discoid structures as well as protects the cell from getting sheared in bloodstream. It has three major components: (1) the spectrin-based membrane skeleton, (2) the actin cytoskeleton, and (3) the marginal microtubule coil.

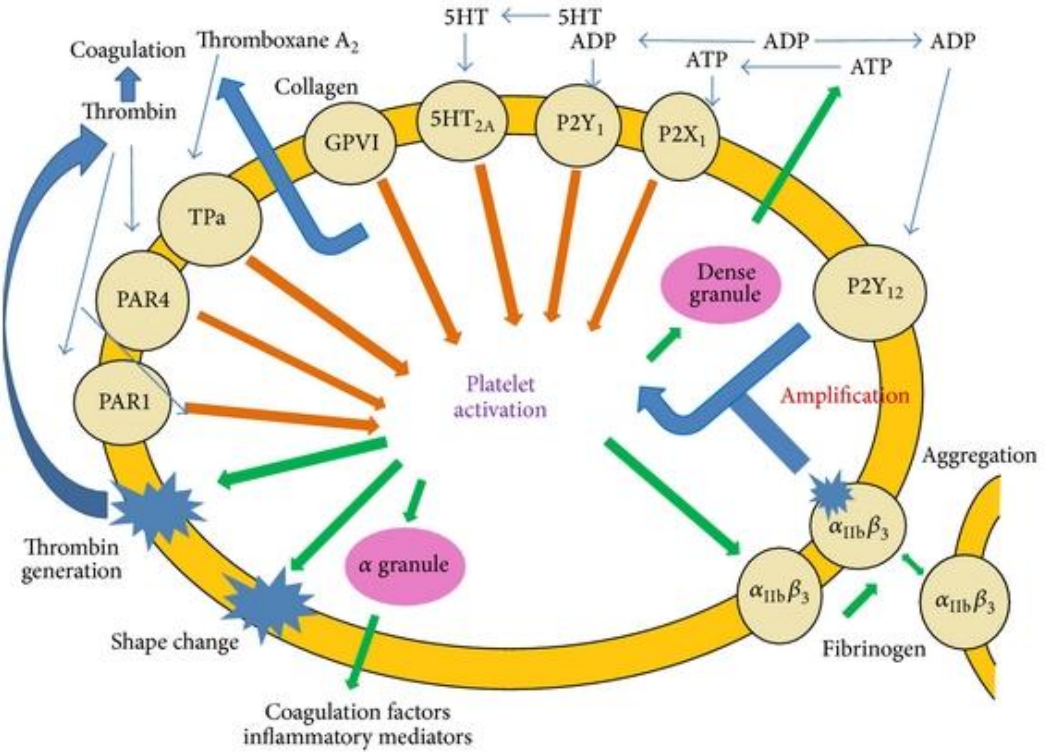

Figure 1: Platelet-activation mechanisms and role of the $P2Y_{12}$ receptor. Platelet activation leads to dense-granule secretion of ADP, which activates $P2Y_{12}$, inducing amplification of aggregation, procoagulant, and proinflammatory responses ***(Storey ,2008)***.

Platelets have two major storage granules, namely, α and dense granules, whose function is to store biologically active molecules precisely involved in initiation of coagulation and recruiting other cells during inflammation ***(Flaumenhaft ,2003)***.

The more prevalent α granule contains proteins (e.g., GPIIb/IIIa, fibrinogen, platelet factor 4 and Von Willibrand factor) which initiate the coagulation cascades. Numerous membrane proteins essential to platelet function are also packaged into α granule which includes GPIIbIIIa, P-selectin (CD62P), and CD36. α granules also have the bulk of cellular P-selectin in their membrane. P-selectin via P-selectin glycoprotein ligand (PSGL1) has been reported to recruit neutrophils ***(Folkman et al.,2001)***. Dense granules store a variety of hemostatically active molecules which are secreted during platelet activation; these include catecholamines, serotonin, calcium, adenosine 5′-diphosphate (ADP), and adenosine 5′-triphosphate (ATP). ADP is a weak platelet agonist, triggering platelet shape change, granule release, and aggregation ***(Thon and Italiano ,2012)***.