Antiplatelet Therapy –Recent Advances

*Rahul Saini, Sunaina

1. Department of pharmacology, PGIMS, Rohtak, Haryana, India.
2. Department of Radiology, PGIMS, Rohtak, Haryana, India.

ABSTRACT

Haemostatic function of platelets is vital & their pathological role is potentially lethal. In pathological states there is over activation and aggregation of blood platelets resulting in atherothrombosis causes various states from angina to acute coronary syndromes, stroke and peripheral artery diseases. Platelets are anuclear subcellular fragments derived from megakaryocytes that circulate in blood as small anucleate ovoid discs. Under normal conditions they do not interact with the intact endothelial cell lining throughout the vasculature. Disruption of this well-regulated balance leads to pathologic conditions, such as thrombosis and bleeding. Although controlled plug formation is desired for the prevention of excessive blood loss and for promoting wound healing, several pathological conditions may result in the formation of occlusive thrombi leading to severe clinical complications, including myocardial infarction and ischemic stroke. The formation of a stable platelet plug occurs in three different stages: platelet adhesion, amplification of platelet activation and platelet aggregation. Many strategies have been pursued to lower the risk of pathological thrombus formation by interfering either with platelet adhesion, activation or aggregation. We here review clinically established antiplatelet targets and promising new antiplatelet strategies that are under investigation.

*Author for Correspondence:
Rahul Saini
P-40A, indraprastha, Rohtak, Haryana, India.
E-mail:drrahulnanu@gmail.com

INTRODUCTION

Haemostasis is maintain in healthy vasculature by various processes that include the platelet system, coagulants, anticoagulants and fibrinolytic pathways [1]. Such processes evolved to maintain the blood in a fluid state under physiological conditions and to arrest bleeding after vascular injury [2]. The ideal antiplatelet agent may depend on the clinical indication. In the acute setting of thrombosis, a powerful agent that has rapid onset of action and rapid offset of action may be most advantageous. To achieve these objectives, parenteral agents are most commonly used. Long-term therapy with oral agents means that a long half-life is required, in order that once- or twice-daily dosing is possible. A shorter half-life may be advantageous to allow earlier return of platelet function when hemostasis is necessary, but noncompliance with a short half-life agent could be dangerous, resulting in acute complications such as stent thrombosis [3]. The ultimate goal remains to prevent thrombosis while preserving hemostasis. Ongoing clinical trials will determine whether any agent currently being tested can achieve this goal. Currently available oral antiplatelet agents include aspirin, an irreversible inhibitor of cyclooxygenase1–mediated TXA2 synthesis, and the $P2Y_{12}$ antagonist's clopidogrel and prasugrel which selectively and irreversibly bind to the $P2Y_{12}$ ADP receptor. Both aspirin and $P2Y_{12}$ antagonists have demonstrated ischemic benefits in patients with atherothrombotic disease [4]. But as they inhibit only one agonist pathway so limited efficacy. Specific inhibition of single agonistic pathway leaves alternative routes

to platelet activation unaffected, this further limit their effectiveness. Treatment with the combination of aspirin plus a $P2Y_{12}$ antagonist (dual antiplatelet therapy) has demonstrated greater efficacy versus monotherapy with either class of,but there is increase risk of bleeding [5]. And also, patients receiving dual antiplatelet therapy remain at substantial risk of ischemic events [6]. There is need for better side effect profile drugs with good efficacy.

Current oral antiplatelet agents: benefits and risks

Aspirin

It is currently the most famous & most widely used agent in antiplatelet therapy. The antiplatelet action of aspirin consists of blocking the TXA2 synthesis by the irreversible acetylation of Ser-529 and Ser-516 in human arachidonate cyclooxygenase (COX)1 and COX2 respectively. Human platelets only express COX1 although residual amounts of COX2 can be detected in newly formed platelets [7]. The efficacy of aspirin in the secondary prevention of ischemic stroke, MI, and vascular death in patients with known atherosclerosis is evident in the large meta-analysis by the Antithrombotic Trialists Collaboration (ATC) which incorporated over 100 trials with aspirin.[8] They showed that 25%reduction in all vascular events, a 30% reduction in MI, and a 15% reduction in death were reported in high-risk patients treated with aspirin compared with placebo. The benefit across the full spectrum of events outweighed the bleeding risk substantially [8]. The optimal dose of aspirin for patients undergoing revascularization, or after an ACS event, remains debatable. Although observational studies and meta-analyses of previous trials have shown no incremental benefit of aspirin doses greater than 75 mg for secondary prevention and following ACS or PCI [9,10]. In the Clopidogrel and Aspirin Optimal Dose Usage to Reduce Recurrent Events-Seventh Organization to Assess Strategies in Ischemic Syndromes (CURRENT-OASIS 7) trial, 25,086 patients with ACS were randomly assigned to receive low-dose (75–100 mg per day) or high-dose (300–325 mg per day) aspirin for maintenance therapy after an initial loading dose. The data from this trial support no difference in the primary end point of cardiovascular death, MI, or stroke at 30 days between the two doses in either the medically managed or invasively managed (PCI) groups [11]. Although no major differences in efficacy and safety were reported between the two doses, observational analyses from previous trials have suggested an increase in major bleeding with chronic high-dose aspirin therapy [12].

GP IIb-IIIa inhibitors

The glycoprotein IIb/IIIa receptor is the most abundant receptor on the platelet surface [13]. Three parenteral inhibitors of this receptor are abciximab, tirofiban, and eptifibatide. They prevent the formation of interplatelet bridges and aggregates. Oral administration of GIIb/IIIa inhibitors has failed to demonstrate any benefit [14]. Therefore, these drugs are only administered within the hospital setting and are indicated for use in high-risk ACS patients undergoing PCI [15]. Eptifibatide and tirofiban reversibly and competitively bind to the GIIb/IIIa receptor, whereas abciximab irreversibly binds to the GIIb/IIIa receptor and blocks the binding of fibrinogen and other adhesion molecules [16]. Although these agents have a role in certain clinical settings, their use has decreased with the advent and routine use of thienopyridines and newer anti-coagulants. Broad use of glycol-protein IIb/IIIa inhibitors (GPIs) occurred in some practices before the administration of clopidogrel prior to PCI became common. Many evidences supported the use of intravenous GPIs in the setting of PCI for both stable and unstable coronary artery disease for the reduction of periprocedural MI [17]. A large meta-analysis of over 30,000 patients who received GPIs for the medical management of ACS demonstrated a relative risk reduction (RRR) of 9% (P = 0.015) for mortality and MI at 30 days compared with placebo, at the expense of a 1% absolute increase in bleeding risk (2.4% vs1.4%, P <0.0001) [18]. Three sequential trials of GPIs in which 600 mg of clopidogrel was also administered at least 2 h before PCI in low-risk, moderate-risk, and high-risk patients, to reflect the current state of

practice. In low-risk, stable patients undergoing elective PCI, the Intracoronary Stenting and Antithrombotic Regimen-Rapid Early Action for Coronary Treatment (ISAR-REACT) trial demonstrated no additional benefit of abciximab over placebo in the reduction of ischemic complications or mortality [19]. Similar results were found in moderate-risk, stable patients with diabetes who were undergoing elective PCI, in the ISAR-Is Abciximab a Superior Way to Eliminate Elevated Thrombotic Risk in Diabetics? (ISAR-SWEET) trial [20]. Although ADP antagonism has remarkably influenced periprocedural ischemic outcomes; however, intravenous GPIs might still have an incremental benefit on a background of DAPT and heparin, particularly in high-risk patients with ACS. The incremental value of additional platelet inhibition through the use of GPIs will be further challenged in the era of more-potent ADP blockade with newer agents. Additionally, intracoronary administration of GPIs has demonstrated greater platelet inhibition with improved coronary flow and microvascular perfusion compared with the parenteral counterpart, and is a subject of current investigation [21]. In addition to efficacy, the appropriate timing of adjunctive intravenous GPI therapy in the setting of PCI in patients with non-ST-segment elevation (NSTE)-ACS has long been debated. Studies demonstrated benefit when these drugs were used up to 24 h before PCI immediately before revascularization, and even with thienopyridine loading [22,23]. In the Early Glycoprotein IIb/IIIa Inhibition in Non-ST-Segment Elevation Acute Coronary Syndrome (EARLYACS) trial, 9,492 patients with high risk NSTE-ACS were randomly assigned to receive routine early eptifibatide (12–96 h) before angiography or to placebo with a delayed provisional use of eptifibatide after angiography, but before intervention, for high-risk features or 'bailout' therapy for a thrombotic complication. No differences in mortality, MI, recurrent ischemic events, or thrombotic bailout at 96 h were seen between the two groups (9.3% versus 10.0%,P = 0.23). Bleeding, however, was increased in the group that received routine early use of eptifibatide [24]. The prior Acute Catheterization and Urgent Intervention Triage (ACUITY) timing study had broadly similar findings [25]. In the modern era of thienopyridine loading, routine GPI use before PCI in the setting of NSTE-ACS continues to decline. Moreover,this treatment accentuates bleeding risk.

Phosphodiesterase inhibitors
Elevation of the levels of cyclic nucleotides cAMP and cGMP in the platelet cytosol stimulates signalling pathways that inhibit platelet activation. Inhibitory effects of endothelium-derived nitric oxide and PGI2 on platelet function are mediated through stimulation of cGMP- and cAMP-dependent signalling mechanisms, respectively. The inhibition of phosphodiesterase enzymes, which metabolize these second messengers, therefore suppresses platelet function [26].

Cilostazol
It is a selective PDE type III (PDE III) inhibitor, increases cAMP levels in platelets, endothelial and smooth muscle cells, having vasodilatory andantiplatelet properties [27]. It was approved by the FDA in 1998 for the treatment of symptoms of intermittent claudication. Pharmacodynamic studies showed that adjunctive cilostazol therapy resulted in greater platelet inhibition than dual antiplatelet therapy [28]. A meta-analysis of 10 trials demonstrated reduction in angiographic restenosis associated with triple antiplatelet therapy in patients undergoing PCI, but no difference in stent thrombosis [29]. In a study of 960 patients who underwent drug-eluting stent implantation, triple antiplatelet therapy with cilostazol, aspirin, and clopidogrel reduced platelet reactivity when compared with standard DAPT; however, this reduction did not translate into improved clinical outcomes [30]. It has predominantly been studied in Koreans, a population with a high prevalence of clopidogrel loss-of-function alleles, which is likely to preclude the broad extrapolation of its benefits. Moreover, recent studies have shown that adjunctive cilostazol to dual antiplatelet therapy after PCI can achieve greater platelet inhibition than high maintenance-dose clopidogrel of 150 mg daily [31] even in patients with

theCYP2C19 mutant allele [28]. It also has been shown to prevent the recurrence of cerebral infarction and the progression of symptomatic intracranial arterial stenosis [32]. It has been shown to be particularly effective in diabetic patients [33]. Clinical trials in large and diverse populations are necessary to validate the potential benefits of it in patients with CAD or cerebrovascular disease. Its most common sideeffects include headache, tachycardia, palpitations, soft stools and diarrhea. It should be avoided in patients with congestive heart failure of any severity because of increased mortality risk [34].

Dipyridamole

It is a pyrimidopyrimidine derivative with antiplatelet and vasodilator properties. The antiplatelet effects of dipyridamole have been reported to be due to several mechanisms, including inhibition of the cyclic guanosine monophosphate (cGMP) phosphodiesterase (PDE) type V enzyme [35]. A study, ESPS-2 (European Stroke Prevention Study), showed that aspirin plus dipyridamole was significantly more effective than aspirin alone in secondary prevention of stroke (relative risk reduction 23.1%; p= 0.006) and conveyed a similarly low risk of severe bleeding (1.6% vs 1.2%) [36]. Another study ESPRIT (European/Australasian Stroke Prevention in Reversible Ischemia Trial) demonstrated that the incidence rate of the composite primary outcome (nonfatal MI, nonfatal stroke, vascular death, or major bleeding complication) was significantly lower in patients receiving aspirin plus dipyridamole than in those using aspirin alone (12.7% vs 15.7%) [37]. However, aspirin plus dipyridamole was not superior to clopidogrel in the treatment of recurrent stroke in the recently completed PROFESS (Prevention Regimen for Effectively Avoiding Second Strokes) trial [38].

$P2Y_{12}$ADP receptor antagonists

ADP is a secondary messenger molecule released in response to platelet adhesion to the endothelium that occurs during vascular injury or plaque rupture. This molecule serves a pivotal role in the activation of platelets and in the amplification of this response by promoting the release of prothrombotic factors. These agents cause Inhibition of ADP at the $P2Y_{12}$ receptor. $P2Y_{12}$receptor antagonists include ticlopidine, clopidogrel, prasugrel, and ticagrelor and other compounds under late-stage development (cangrelor, elinogrel). Ticlopidine, clopidogrel, and prasugrel represent 3 generations of thienopyridines that selectively and irreversibly inhibit the $P2Y_{12}$ receptor.

Ticlopidine

Ticlopidine was the first thienopyridine to enter clinical usage in the early 1990s and demonstrated significant benefit over placebo and equal efficacy to aspirin,for the prevention of secondary cardiovascular events after ACS [39]. Thienopyridines are prodrugs that require metabolism to generate the active compound that selectively and irreversibly inhibits the$P2Y_{12}$ receptor [40]. Clinical trials demonstrated that, in patients undergoing coronary stenting, better clinical outcomes were achieved with the combined use of aspirin and ticlopidine than aspirin alone or aspirin plus warfarin [41]. In the TASS (Ticlopidine Aspirin Stroke Study), ticlopidine was superior to aspirin for prevention of the primary endpoint, nonfatal stroke or death. Two limitations with the use of ticlopidine are: its safety profile, such as neutropenia, thrombotic thrombocytopenic purpura, rash and its inability to induce platelet inhibition rapidly [42]. It has been largely replaced in clinical practice by clopidogrel, which has the same benefitcial properties as ticlopidine but without its limitations.

Clopidogrel

Clopidogrel is a second-generation thienopyridine & binds irreversibly to $P2Y_{12}$receptor. Irreversible binding to the$P2Y_{12}$ receptor means that platelet turnover is required to restore platelet function. Complete platelet turnover requires 7–10 days. It also has advantage of being administered orally. When taken at 75 mg day per day, it is able to reduce ADP-induced aggregation by 40–60%. It is reported to be more effective than aspirin (325 mg days per day) in preventing vascular death, MI or ischemic stroke but with reduced gastric irritation. When used at higher doses, however, it increases bleeding time [43]. It has been extensively

studied and variable pharmacodynamic effects have been associated with variation in absorption, variation in metabolism and genetic variation in the $P2Y_{12}$ receptor [44]. It is converted to its active metabolite by the cytochrome P450 system. Only 15% of it becomes an active metabolite. It has a slow onset of action unless loading doses are used. Without a loading dose, steady state effects are not apparent until approximately 5 days. By contrast, steady state pharmacodynamic effects are apparent 18–24 h after a 300-mg loading dose and 2 h after a 600-mg loading dose [45]. Side effects such as cardiac events, gastrointestinal problems, neutropenia and thrombotic thrombocytopenic purpure have been reported [46].

Emerging antiplatelet agents

Novel $P2Y_{12}$ inhibitors

Newer $P2Y_{12}$ inhibiting antiplatelet therapies have a more favorable pharmacokinetic and pharmacodynamic profile than clopidogrel and do not appear to interact with the genetic polymorphisms

Prasugrel

It is a third-generation oral thienopyridine which is chemically different from clopidogrel. It is a prodrug that irreversibly binds to the platelet $P2Y_{12}$ receptor. It is converted to its active metabolite more effectively than clopidogrel, allowing for greater bioavailability and faster onset of action [47]. It is more powerful antiplatelet agent with less interindividual variability and a more consistent antiplatelet effect [48]. After a loading dose of 60 mg maximal plasma concentration is achieved within approximately 30 min [47]. It was approved in 2009 in both the United States and Europe. It causes more potent inhibition of platelet aggregation and a more consistent platelet response than standard- and high-dose clopidogrel [49]. TRITON (Trial to Assess Improvement in Therapeutic Outcomes by Optimizing Platelet Inhibition with Prasugrel) showed prasugrel to be superior to standard-dose clopidogrel in reducing ischemic events in patients with ACS scheduled for PCI, although prasugrel was associated with a significantly higher risk of major bleeding events [50]. However,patients with stroke or TIA had net clinical harm from prasugrel and those =75 years old or who weighed<60 kg had no net benefit. It demonstrated the greatest benefit among patients with DM and those presenting with STEMI undergoing primary PCI in whom there were no differences in major bleeding complications [51]. In patients who are at high risk of bleeding, particularly the elderly (age>75 years) or underweight (<60 kg), the risks and benefits of prasugrel use must be carefully weighed. Its use is contraindicated in patients who have had a prior stroke or transient ischemic attack. In spite of these limitations, the efficacy of prasugrel is not affected by concurrent PPI administration or genetic polymorphisms. It is only approved for use in patients with ACS undergoing PCI, switching patients from clopidogrel to prasugrel after ACS has demonstrated incremental platelet inhibition [52]. It appears to be better suited for selected patients such as those with diabetes mellitus and STEMI. The more powerful antiplatelet effect and irreversible binding poses additional risk for patients undergoing surgical procedures.

Ticagrelor

It is a cyclopentyltriazoloprimidine and a direct and reversible $P2Y_{12}$ antagonist, has recently approved in the United States and Europe. Like prasugrel, ticagrelor acts more rapidly and is a more potent inhibitor of platelets than clopidogrel and did not significantly increase major bleeding compared with clopidogrel. However, the occurrence of dyspnea and ventricular pause were greater, in an apparently dose dependent manner, in patients receiving ticagrelor than in patients receiving clopidogrel [53]. The PLATO (Platelet Inhibition and Patient Outcomes) trial randomized 18,624 ACS patients to receive ticagrelor or clopidogrel. The overall trial demonstrated a significant reduction in the primary endpoint a composite of death from vascular causes, MI, or stroke at 12 months with ticagrelor, without an increase in overall major bleeding although non-surgical bleeding was significantly increased to a similar extent than that observed with prasugrel and there was a higher rate of drug discontinuation due to dyspnea [54]. Given its reversible affinity towards $P2Y_{12}$, ticagrelor might redistribute

itself to new platelets that enter the circulation before the administration of the next dose, which could account for the mortality benefit. Additionally, a potential off-target benefit of ticagrelor is its inhibition of adenosine uptake into red blood cells, which results in improved myocardial blood flow. Currently known genetic polymorphisms do not alter the efficacy of ticagrelor [55].

Cangrelor

It is a chemical modification of ticagrelor yielded, a reversible intravenous$P2Y_{12}$ receptor antagonist with a half-life of 5–9 min. It achieves steady state concentration within minutes and platelet function recovers within 2 h after the infusion is discontinued. In CHAMPION PCI (Comparison of IV Cangrelor to Clopidogrel in coronary stenting), a phase III trial (8,820 pts) are randomized to cangrelor 30 mcg per kg IV & clopidogrel 600 mg orally followed by infusion administered before PCI [56]. Another CHAMPION PLATFORM trial, a double-blind, placebo-controlled trial that randomized more than 5000 patients with ACS undergoing PCI to either placebo or cangrelor given as an intravenous bolus of 30 mcg/kg followed by infusion administered before PCI [57]. Neither trial demonstrated a significant difference in the primary end point of death, MI, or revascularization at 48 h. Stent thrombosis and mortality, however, were reduced with cangrelor at 48 h in the CHAMPION PLATFORM study.

Elinogrel

It is a potent oral and intravenous (IV) third-generation$P2Y_{12}$ antagonist that has potential advantages over clopidogrel and prasugrel. One of these is a direct mode of action that does not require conversion into an active drug; this direct action can result in reduced variability in patient response. The reversible and competitive nature of $P2Y_{12}$ receptor binding with it could result in its displacement by ADP at sites of bleeding, which are characterized by low flow, low shear rate, and higher ADP concentrations. For this reason, it may provide a more favorable safety profile as compared with irreversibly acting agents such as prasugrel and clopidogrel [58].

The INNOVATE-PCI study was a phase 2b study with 652 patients undergoing elective PCI randomized to clopidogrel or to IV elinogrel followed by oral elinogrel. Although treatment with elinogrel was associated with a more rapid and more potent inhibition of ex vivo ADP-induced platelet activation, there were no significant differences in the rates of ischemic events between the clopidogrel and elinogrel treatment arms at 24 hours or 120 days [58].

PAR-1 Inhibitors

Selective inhibition of the principal protease activated receptor (PAR)-1 for thrombin, the most potent platelet activator, represents a promising novel strategy to reduce ischemic events without increasing the risk of bleeding. Two PAR-1 receptor antagonists are currently being tested in clinical trials, vorapaxar (SCH 530348) and atopaxar (E5555).

Vorapaxar

It is an orally active PAR-1 antagonist that blocks thrombin-mediated platelet activation without interfering with the thrombin mediated cleavage of fibrinogen [59]. A single dose of vorapaxar inhibits platelet function within 1 h after administration and an effect was still evident after 72 h. The drug has a rather long half-life of 126–296 h. [60]. Platelet function returns to normal within 2–3 weeks after cessation of the drug. The safety and efficacy of vorapaxar have already been tested in various Phase II trials, including the TRAPCI (Thrombin Receptor Antagonist for Cardiovascular Event Reduction in Percutaneous Coronary Interventions) in which vorapaxar when used in combination with the standard of care therapy in patients scheduled to undergo non-urgent PCI. In it patients (n = 1030) were randomized to oral vorapaxar or placebo in a 3:1 ratio on top of standard antithrombotic therapy, including aspirin, clopidogrel, and the anticoagulant of choice. No significant increase was observed in TIMI major and minor bleeding (primary endpoint) with vorapaxar vs standard of care (2.8% vs 3.3%; p= 0.58). There was a trend toward a lower rate of death and major adverse cardiac events (MACE), including MI in the vorapaxar treated group

compared with the placebo-treated group [61].

Atopaxar

It is another member of the thrombin receptor antagonist group & was evaluated in two phase II trials-LANCELOT (Japanese lessons from antagonizing the Cellular Effects of Thrombin) and LANCELOT. These trials were designed to evaluate the safety and tolerability of atopaxar in ACS and CAD patients as their primary objective. The results of these trials have been recently reported and overall show that atopaxar was not associated with any increase in serious bleeding events in both ACS and CAD patients, although there was a significant dose-dependent increase in liver function abnormalities [62].

CONCLUSION

Atherothrombotic disease is a major health burden with significant economic impact. Patients with prior ACS or stroke are at significant risk of experiencing a recurrent event or an atherothrombotic event in another vascular bed. Patients with PAD are also at increased risk of acute atherothrombotic events. Antiplatelet therapy with aspirin, clopidogrel, the combination of aspirin plus clopidogrel, and the combination of aspirin plus ER dipyridamole have been shown in numerous clinical trials to be both effective and safe in the secondary prevention. However, recurrent event rates remain considerably high with currently available antiplatelet treatment regimens. With a strong clinical need to improve the efficacy and safety of current anti-platelet therapies in various clinical scenarios, a small number of characterized platelet receptors, which include proven anti-platelet drug targets, are currently the focus of drug discovery and clinical trials for the prevention and treatment of thrombosis. These include ADP receptor ($P2Y_{12}$) antagonists and molecules that inhibit activation of PARs for thrombin. A more understanding of the platelet regulatory systems is, however, likely to result in the development of more refined, safer and more efficacious approaches to prevent thrombosis. So no doubt there is always need of better antiplatelet drugs with balance of safety, efficacy & cost.

REFERENCES

1. Davi G, Patrono C. Platelet activation and atherothrombosis.N Engl J Med 2007 Dec 13;357 (24):2482-94.
2. Rosenberg RD, Aird WC. Vascular bed–specific hemostasis and hypercoagulable states.N Engl J Med 1999 May 20;340(20):1555-64.
3. Lischke S, Schneider DJ. Recent developments in the use of antiplatelet agents to prevent cardiovascular events.Future Cardiol 2011 May;7(3):403-13.
4. Wiviott SD, Braunwald E, McCabe CH, Montalescot G, Ruzyllo W, Gottlieb S et al. Prasugrel versus clopidogrel in patients with acute coronary syndromes.N Engl J Med 2007 Nov 15;357(20):2001-15.
5. Jennings LK. Mechanisms of platelet activation: need for new strategies to protect against platelet-mediated atherothrombosis. Thromb Haemost 2009 Aug;102(2):248-57.
6. Wallentin L, Becker RC, Budaj A, Cannon CP, Emanuelsson H, Held C et al. Ticagrelor versus clopidogrel in patients with acute coronary syndromes.N Engl J Med 2009 Sep 10;361(11):1045-57.
7. Rocca B, Secchiero P, Ciabattoni G, Ranelletti FO, Catani L, Guidotti L et al.Cyclooxygenase-2 expression is induced during human megakaryopoiesis and characterizes newly formed platelets. Proc Natl Acad Sci USA 2002 May 28;99(11):7634-9.
8. Antithrombotic Trialists' Collaboration. Collaborative meta-analysis of randomised trials of antiplatelet therapy for prevention of death, myocardial infarction, and stroke in high risk patients.BMJ 2002 Jan 12;324(7329):71-86.
9. Berger JS, Brown DL, Burke GL, Oberman A, Kostis JB, Langer RD et al. Aspirin use, dose, and clinical outcomes in postmenopausal women with stable cardiovascular disease: the Women's Health Initiative Observational Study. Circ Cardiovasc Qual Outcomes 2009 Mar;2(2):78-87.
10. Jolly SS, Pogue J, Haladyn K, Peters RJ, Fox KA, AvezumA et al. Effects of aspirin dose on ischemic events and bleeding after percutaneous coronary intervention: insights from the PCI-CURE study. Eur Heart J 2009 Apr;30(8):900-7.
11. Mehta SR, Tanguay JF, Eikelboom JW, Jolly SS, Joyner CD, Granger CB et al. Double-dose versus standard dose clopidogrel and high-dose versus low-dose aspirin in individuals undergoing percutaneous coronary intervention for acute coronary syndromes (CURRENT-OASIS 7): a randomized factorial trial. Lancet 2010 Oct 9;376(9748):1233-43.

12. Peters RJ, Mehta SR, Fox KA, Zhao F, Lewis BS, Kopecky SL et al. Effects of aspirin dose when used alone or in combination with clopidogrel in patients with acute coronary syndromes: observations from the Clopidogrel in Unstable angina to prevent Recurrent Events (CURE) study.Circulation 2003 Oct 7;108(14):1682-7.

13. French DL, Seligsohn U. Platelet Glycoprotein IIb/IIIa Receptors and Glanzmann's Thrombasthenia. Arterioscler Thromb Vasc Biol 2000 Mar;20(3):607-10.

14. Chew DP, Bhatt DL, Sapp S, Topol EJ. Increased mortality with oral platelet glycoprotein IIb/IIIa antagonists: a meta-analysis of phase III multicenter randomized trials.Circulation 2001 Jan 16;103(2):201-6.

15. Kastrati A, Mehilli J, Neumann FJ, Dotzer F, ten Berg J, Bollwein H. Abciximab in patients with acute coronary syndromes undergoing percutaneous coronary intervention after clopidogrel pretreatment: the ISAR-REACT2 randomized trial. JAMA 2006 Apr 5;295(13):1531-8.

16. Kleiman NS. Pharmacokinetics and pharmacodynamics of glycoprotein IIb/IIIa inhibitors. Am Heart J 1999 Oct;138(4 Pt 2):263-75.

17. Bhatt DL. Glycoprotein IIb/IIIainhibitors : do they still have a role? J Am CollCardiol 2011 Mar 8;57(10):1200-1.

18. Boersma E, Harrington RA, Moliterno DJ, White H, Théroux P, Van de Werf F et al. Platelet glycoprotein IIb/IIIa inhibitors in acute coronary syndromes: a metaanalysis of all major randomised clinical trials.Lancet 2002 Jan 19;359(9302):189-98.

19. Kastrati A, Mehilli J, Schühlen H, Dirschinger J, Dotzer F, ten Berg JMet al. A clinical trial of abciximab in electivepercutaneous coronary intervention after pretreatment with clopidogrel.N Engl J Med 2004 Jan 15;350(3):232-8.

20. Mehilli J, Kastrati A, Schühlen H, Dibra A, Dotzer F, von Beckerath N et al. Randomized clinical trial of abciximab in diabetic patients undergoing elective percutaneous coronary interventions after treatment with a high loading dose of clopidogrel. Circulation 2004 Dec 14;110(24):3627-35.

21. Deibele AJ, Jennings LK, Tcheng J, Neva C, Earhart AD, Gibson CM. Intracoronary eptifibatide bolus administration during percutaneous coronary revascularization for acute coronary syndromes with evaluation of platelet glycoprotein IIb/IIIa receptor occupancy and platelet function: the Intracoronary Eptifibatide (ICE) Trial. Circulation 2010 Feb 16;121(6):784-91.

22. Boersma E, Akkerhuis KM, Theroux P, Califf RM, Topol EJ, Simoons ML. Platelet glycoprotein IIb/receptor inhibition in non-ST-elevation acute coronary syndromes: early benefit during medical treatment only, with additional protection during percutaneous coronary intervention. Circulation 1999 Nov 16;100(20):2045-8.

23. Kastrati A, Mehilli J, Neumann FJ, Dotzer F, ten Berg J, Bollwein H et al. Abciximab in patients with acute coronary syndromes undergoing percutaneous coronary intervention after clopidogrel pretreatment: the ISAR-REACT 2 randomized trial. JAMA 2006 Apr 5;295(13):1531-8.

24. Giugliano RP, White JA, Bode C, Armstrong PW, Montalescot G, Lewis BSet al. Early versus delayed, provisional eptifibatide in acute coronary syndromes. N Engl J Med 2009 May 21;360(21):2176-90.

25. Stone GW, Bertrand ME, Moses JW, Ohman EM, Lincoff AM, Ware JHet al. Routine upstream initiation vs deferred selective use of glycoprotein IIb/IIIa inhibitors in acute coronary syndromes: the ACUITY Timing trial. JAMA 2007 Feb 14;297(6):591-602.

26. Manns JM, Brenna KJ, Colman RW, Sheth SB . Differential regulation of human platelet responses by cGMP inhibited and stimulated cAMP phosphodiesterases. Thromb Haemost. 2002 May;87(5):873-9.

27. Goto S. Cilostazol: potential mechanism of action for antithrombotic effects accompanied by a low rate of bleeding. Atheroscler Suppl 2005 Dec 15;6(4):3-11.

28. Hwang SJ, Jeong YH, Kim IS, Park KS, Kang MK, Koh JS et al. Cytochrome 2C19 Polymorphism and Response to Adjunctive Cilostazol Versus High Maintenance-Dose Clopidogrel in Patients Undergoing Percutaneous Coronary Intervention. Circ Cardiovasc Interv 2010 Oct;3(5):450-9.

29. Tamhane U, Meier P, Chetcuti S, Chen KY, Rha SW, Grossman MPet al. Efficacy of cilostazol in reducing restenosis in patients undergoing contemporary stent based PCI: a meta-analysis of randomised controlled trials. Euro Intervention 2009 Aug;5(3):384-93.

30. Suh JW, Lee SP, Park KW, Lee HY, Kang HJ, Koo BKet al. Multicenter randomized trial evaluating the efficacy of cilostazol on ischemic vascular complications after drug-eluting stent implantation for coronary heart disease: results of the CILON-T (influence of CILostazol-based triple antiplatelet therapy ON ischemic complication after drug-eluting

stenT implantation) trial. J Am Coll Cardiol 2011 Jan 18;57(3):280-9.

31. Jeong YH, Lee SW, Choi BR, Kim IS, Seo MK, Kwak CH et al. Randomized comparison of adjunctive cilostazol versus high maintenance dose clopidogrel in patients with high post-treatment platelet reactivity: results of the ACCEL-RESISTANCE (Adjunctive Cilostazol Versus High Maintenance Dose Clopidogrel in Patients With Clopidogrel Resistance) randomized study. J Am Coll Cardiol 2009 Mar 31;53(13):1101-9.

32. Shinohara Y, Katayama Y, Uchiyama S, Yamaguchi T, Handa S, Matsuoka K et al.Cilostazol for prevention of secondary stroke (CSPS 2): an aspirin-controlled, double-blind, randomised non-inferiority trial. Lancet Neurol 2010 Oct;9(10):959-68.

33. Angiolillo DJ, Capranzano P, Goto S, Aslam M, Desai B, Charlton RK et al.A randomized study assessing the impact of cilostazol on platelet function profiles in patients with diabetes mellitus and coronary artery disease on dual antiplatelet therapy: results of the OPTIMUS-2 study.Eur Heart J 2008 Sep;29(18):2202-11.

34. da Rosa MP, Baroni GV, Portal VL. Cilostazol, a phosphodiesterase inhibitor: future perspectives in atherosclerosis. Arq Bras Cardiol 2006 Nov;87(5):e222-6.

35. Aktas B, Utz A, Hoenig-Liedl P, Walter U, Geiger J. Dipyridamole enhances NO/cGMP-mediated vasodilator-stimulated phosphoprotein phosphorylation and signaling in human platelets: in vitro and in vivo/ex vivo studies. Stroke 2003 Mar;34(3):764-9.

36. Diener HC, Cunha L, Forbes C, Sivenius J, Smets P, Lowenthal A. European Stroke Prevention Study 2.Dipyridamole and acetylsalicylic acid in the secondary prevention of stroke.J Neurol Sci 1996 Nov;143(1-2):1-13.

37. Halkes PH, van Gijn J, Kappelle LJ, Koudstaal PJ, Algra A:Aspirin plus dipyridamole versus aspirin alone after cerebral ischaemia of arterial origin (ESPRIT): randomised controlled trial. Lancet 2006 May 20;367(9523):1665-73.

38. Sacco RL, Diener HC, Yusuf S, Cotton D, Ounpuu S, Lawton WA et al. Aspirin and extended-release dipyridamole versus clopidogrel for recurrent stroke. N Engl J Med 2008 Sep 18;359(12):1238-51.

39. Scrutinio D, Cimminiello C, Marubini E, Pitzalis MV, Di Biase M, Rizzon P.Ticlopidine versus aspirin after myocardial infarction (STAMI) trial. J Am CollCardiol 2001 Apr;37(5):1259-65.

40. Brandt JT, Payne CD, Wiviott SD, Weerakkody G, Farid NA, Small DSet al. A comparison of prasugrel and clopidogrel loading doses on platelet function:magnitude of platelet inhibition is related to active metabolite formation. Am Heart J 2007 Jan;153(1):66.e9-16.

41. Schömig A, Neumann FJ, Kastrati A, Schühlen H, Blasini R, Hadamitzky M et al. A randomized comparison of antiplatelet and anticoagulant therapy after the placement of coronary-artery stents. N Engl J Med 1996 Apr 25;334(17):1084-9.

42. Bertrand ME, Rupprecht HJ, Urban P, Gershlick AH. Double-blind study of the safety of clopidogrel with and without a loading dose in combination with aspirin compared with ticlopidine in combination with aspirin after coronary stenting: the Clopidogrel Aspirin Stent International Cooperative Study (CLASSICS). Circulation 2000 Aug 8;102(6):624-9.

43. Thebault JJ, Kieffer G, Cariou R. Single-dose pharmacodynamics of clopidogrel. Semin Thromb Hemost 1999;25Suppl 2:3-8.

44. Momary KM, Dorsch MP. Factors associated with clopidogrel non responsiveness. Future Cardiol 2010 Mar;6(2):195-210.

45. Von BN, Taubert D, Pogatsa MG, Schömig E, Kastrati A, Schömig A. Absorption, metabolization, andantiplatelet effects of 300-, 600-, and 900-mg loading doses of clopidogrel: results of the ISAR-CHOICE (Intracoronary Stenting and Antithrombotic Regimen: Choose Between High Oral Doses for Immediate Clopidogrel Effect) trial. Circulation 2005 Nov 8;112(19):2946-50.

46. Bennett CL, Connors JM, Carwile JM, Moake JL, Bell WR, Tarantolo SR et al. Thrombotic thrombocytopenic purpura associated with clopidogrel. N Engl J Med 2000 Jun 15;342(24):1773-7.

47. Farid NA, Smith RL, Gillespie TA, Rash TJ, Blair PE, Kurihara A et al.The disposition of prasugrel, a novel thienopyridine, in humans. Drug Metab Dispos 2007 Jul;35(7):1096-104.

48. Wallentin L, Varenhorst C, James S, Erlinge D, Braun OO, Jakubowski JA et al.:Prasugrel achieves greater and faster P2Y12 receptor mediated platelet inhibition thanclopidogrel due to more efficient generation of its active metabolite in aspirin-treated patients with coronary artery disease. Eur Heart J 2008 Jan;29(1):21-30.

49. Payne CD, Li YG, Small DS, Ernest CS 2nd, Farid NA, Jakubowski JA et al. Increased

active metabolite formation explains the greater platelet inhibition with prasugrel compared to high-dose clopidogrel. J Cardiovasc Pharmacol 2007 Nov;50(5):555-62.

50. Wiviott SD, Braunwald E, McCabe CH, Montalescot G, Ruzyllo W, Gottlieb S et al.Prasugrel versus clopidogrel in patients with acute coronary syndromes.N Engl J Med 2007 Nov 15;357(20):2001-15.

51. Wiviott SD, Braunwald E, Angiolillo DJ, Meisel S, Dalby AJ, Verheugt FW et al. Greater clinical benefit of more intensive oral antiplatelet therapy with prasugrel in patients with diabetes mellitus in the trial to assess improvement in therapeutic outcomes by optimizing platelet inhibition with prasugrel-Thrombolysis in Myocardial Infarction 38.Circulation 2008 Oct 14;118(16):1626-36.

52. Angiolillo DJ, Saucedo JF, Deraad R, Frelinger AL, Gurbel PA, Costigan TMet al. Increased platelet inhibition after switching from maintenance clopidogrel to prasugrel in patients with acute coronary syndromes: results of the SWAP (SWitching Anti Platelet) study. J Am CollCardiol 2010 Sep 21;56(13):1017-23.

53. Husted S, Emanuelsson H, Heptinstall S, Sandset PM, Wickens M, Peters G. Pharmaco- dynamics, pharmacokinetics, and safety of the oral reversible P2Y12 antagonist AZD6140 with aspirin in patients with atherosclerosis: double-blind comparison to clopidogrel with aspirin. Eur Heart J 2006 May;27(9):1038-47.

54. Wallentin L, Becker RC, Budaj A, Cannon CP, Emanuelsson H, Held C et al. Ticagrelor versus clopidogrel in patients with acute coronary syndromes.N Engl J Med 2009 Sep 10;361(11):1045-57.

55. Tantry US, Bliden KP, Wei C, Storey RF, Armstrong M, Butler Ket al. First analysis of the relation between CYP2C19 genotype and pharmacodynamics in patients treated with ticagrelor versus clopidogrel: the ONSET/OFFSET and RESPOND genotype studies. Circ Cardiovasc Genet 2010 Dec;3(6):556-66.

56. Harrington RA, Stone GW, McNulty S, White HD, Lincoff AM, Gibson CMet al. Platelet inhibition with cangrelor in patients undergoing PCI.N Engl J Med 2009 Dec 10;361(24):2318-29.

57. Bhatt DL, Lincoff AM, Gibson CM, Stone GW, McNulty S, Montalescot Get al. Intravenous platelet blockade with cangrelor during PCI. N Engl J Med 2009 Dec 10;361(24):2330-41.

58. Leonardi S, Rao SV, Harrington RA, Bhatt DL, Gibson CM, Roe MTet al. Rationale and design of the randomized, double-blind trial testing INtraveNous and Oral administration of elinogrel, a selective and reversible P2Y(12)-receptor inhibitor, versus clopidogrel to eVAluate Tolerability and Efficacy in non urgent Percutaneous Coronary Interventions patients(INNOVATE-PCI).Am Heart J 2010 Jul;160(1):65-72.

59. Chintala M, Shimizu K, Ogawa M, Yamaguchi H, Doi M, Jensen P. Basic and translational research on proteinase activated receptors: antagonism of the proteinase-activated receptor 1 for thrombin, a novel approach to antiplatelet therapy for atherothrombotic disease. J Pharmacol Sci 2008 Dec;108(4):433-8.

60. T. Kosoglou, L. Reyderman, C. Kasserra, S. Young, J. Pei, S. E. Maxwell ,et al. Optimizing dose of the novel thrombin receptor antagonist SCH 530348 based on pharmacodynamics and pharmacokinetics in healthy subjects. Clin.Pharmacol.Ther 2008 Mar;83(Suppl. 1):S-55.

61. Becker RC, Moliterno DJ, Jennings LK, Pieper KS, Pei J, Niederman A et al.Safety and tolerability of SCH530348 in patients undergoing non-urgent percutaneous coronary intervention: a randomised, double-blind, placebo-controlled phase II study. Lancet 2009 Mar 14;373(9667):919-28.

62. Goto S, Ogawa H, Takeuchi M, Flather MD, Bhatt DL. Double-blind, placebo-controlled Phase II studies of the protease-activated receptor 1 antagonist E5555 (atopaxar) in Japanese patients with acute coronary syndrome or high-risk coronary artery disease. Eur Heart J 2010 Nov;31(21):2601-13.