Antiplatelet drugs in patients with enhanced platelet turnover: biomarkers versus platelet function testing

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Summary
Platelets are key players in atherothrombosis. Antiplatelet therapy comprising aspirin alone or with P2Y₁₂-inhibitors are effective for prevention of atherothrombotic complications. However, there is interindividual variability in the response to antiplatelet drugs, leaving some patients at increased risk of recurrent atherothrombotic events. Several risk factors associated with high on-treatment platelet reactivity (HTPR), including elevated platelet turnover, have been identified. Platelet turnover is adequately estimated from the fraction of reticulated platelets. Reticulated platelets are young platelets, characterised by residual messenger RNA. They are larger, haemostatically more active and there is evidence that platelet turnover is a causal and prognostic factor in atherothrombotic disease. Whether platelet turnover per se represents a key factor in pathogenesis, progression and prognosis of atherothrombotic diseases (with focus on acute coronary syndromes) or whether it merely facilitates insufficient platelet inhibition will be discussed in this state-of-the-art review.

Keywords
Platelet turnover, reticulated platelets, mean platelet volume, P2Y₁₂ inhibitors, aspirin

Introduction
Platelets are important to repair continuously occurring endothelial lesions, initiate thrombus formation after vascular damage and modulate wound healing, inflammation, as well as angiogenesis. On the other hand, they are key factors in progression of atherosclerosis and atherothrombosis (1, 2). Atherosclerosis is the underlying pathophysiological process of a variety of cerebro- and cardiovascular diseases (2). As described by Virchow (3), factors from the vessel wall, changes in blood flow and the blood itself (platelets and coagulation factors) might also contribute to the transition from stable coronary artery disease (CAD) into acute coronary syndrome (ACS) (2). Thrombus formation over plaques is initiated either due to erosion of endothelial cells with exposure of subendothelial structures or due to plaque disruption with intraplaque thrombus formation and sudden increase in plaque volume or rupture, with exposure of the highly thrombogenic core to the blood (4, 5). Blood flow velocity decreases from the centre of the vessel to layers close to the vessel wall with maximal shear forces. The combination of shear acceleration, peak shear stress and rapid shear deceleration at vascular bifurcation, or even more pronounced at ruptured plaques or preexisting thrombus, produce substantial platelet aggregates, independently of platelet activators (6). Local platelet activators, including adenosine diphosphate (ADP), thromboxane A₂ (TXA₂) and thrombin further activate the loosely attached platelets and stabilise their position by aggregation (2). Evidence that platelets are key elements in ACS derives from the observation that they are the main component of coronary artery thrombi in virtually all patients with ACS (7). Levels of TXA₂ and prostacyclin metabolites parallel undulant ischaemic episodes and associate with plasma creatinine-kinase levels (8). Moreover, an increase in platelet count and size have also been linked to ACS, cardiovascular death and reinfarction. Larger and denser platelets are more reactive since they are able to produce and release more thrombogenic substances and to express more specific surface receptors (9). Finally, the proof of concept that platelets are essentially involved in atherothrombosis derives from broad clinical evidence that antiplatelet agents reduce cardiovascular risk (10). Aspirin alone or in combination with P2Y₁₂ inhibitors constitute the cornerstones in treatment and secondary prevention of ACS (11–14).

An insufficient pharmacological platelet inhibition is most often defined as high on-treatment platelet reactivity (HTPR) according to cut-offs from various ex vivo tests (15–18) and is considered a risk factor for recurrent ischaemic events (19). HTPR is multifactorial (2, 15) and recent studies have raised the question whether enhanced platelet turnover as seen in ACS or diabetes interferes with the anti-aggregatory effects of antiplatelet agents

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Platelets are approximately 2 µm large, discoid and anucleate cellular fragments with residual messenger RNA (mRNA) and preserved ability to synthesise various proteins (24). After 8–10 days, platelets disappear from circulation. To maintain a stable normal platelet count in adults (150,000–350,000 per mm$^3$) $10^{11}$ new platelets have to be released every day ranging to a 20-fold increase in times of greater demand (25).

Platelet production starts with differentiation of a haematopoietic stem cell and is completed when mature megakaryocytes release fragments into the vasculature, further splitting up into platelets. The exact description of thrombopoiesis is outside the scope of this review, and new insights have recently been reviewed elsewhere (23, 26). Briefly, the process that megakaryocytes undergo 3–5, so-called endomitoses, thus generating polyplloid megakaryocytes, is unique among mammalian cells. In doing so, the volume of megakaryocytes doubles with each nuclear endomitosis (27). Number and size of megakaryocytes define the quantity (and quality) of cytoplasm for platelet production. Megakaryopoiesis is regulated by a plethora of growth factors (28), including interleukin (IL)-1beta, IL-3, IL-6, IL-11, stem cell factor, and erythropoietin, respectively, while thrombopoietin is the primary regulator of thrombopoiesis, acting via the c-Mpl-receptor (29). It acts at each stage of megakaryopoiesis, resulting in increased megakaryocyte size, polyploidisation and, finally, increased platelet production (29).

Greater megakaryocyte ploidy and changes in megakaryocyte DNA positively correlate with expression of megakaryocyte mRNA for known procoagulatory mediators such as fibrinogen (30), platelet-derived growth factor B–chain (31), or glycoprotein (GP) IIb (32). Hence, the aforementioned increase in megakaryocyte ploidy not only helps to produce more platelets (quantity), but also more active platelets (quality) to fulfill increased haemostatic needs.

Several models describe the mechanism of ultimate platelet release from the bone-marrow or from megakaryocytes into circulation. Of these, the most preferred one is the proplatelet-flow-model: beginning at a yet unidentified signal, mature megakaryocytes restructure their cytoplasm and build elongated pseudopodia, the so-called proplatelets (33). Newly formed platelets or proplatelets must enter the capillaries without local clotting when they pass the subendothelial space, and it has been suggested that the problem of untimely platelet activation is circumvented as megakaryocytes extend their pseudopodia into the lumen of local sinuoidal vessels (34). These processes are sheared off by local blood flow (34). Other models suggest megakaryocyte fragmentation in the peripheral or pulmonary circulation. The signals defining the size (and quality) of a mature platelet should be further studied.

Platelets newly released from megakaryocytes are – in analogy to reticulated red blood cells – known as reticulated platelets (35). The identification of reticulated platelets can be identified in flow cytometers based on a combination of light scatter (cell size) and staining of residual mRNA, which is usually degraded after 24 hours (h) in circulation. The fraction of reticulated platelets is typically increased in situations with enhanced thrombopoiesis, platelet turnover and higher megakaryocyte activity (36).

### Platelet indices

Whereas platelet counting combined with analysis of platelet size is routinely performed in most laboratories, the evaluation of platelet kinetics is rather uncommon, as information about platelet production, platelet life span and degradation could only be collected by means of radio-labelling of platelets (37).

### Platelet count and platelet distribution

Platelet counting is an important screening measure to identify platelet diseases. Over the last decades, development from manual counting techniques over automated impedance techniques to optical scatter techniques or even a combination of optical with flow cytometric information has led to better results as small, non-platelet particles as well as very large platelets can now be correctly assigned (38).

In patients with atherosclerosis and its complications, measuring platelet count might provide additional prognostic insight. A reduced platelet count (< 150,000 per mm$^3$) before percutaneous coronary intervention (PCI) is linked to an increase in major bleedings and gastrointestinal bleedings and consequently to increased in-hospital mortality (39). Yet, with thrombocytopenia following PCI, the situation becomes more complex, since it results in haemorrhagic complications with excess mortality (40) and also more ischaemic complications (41). Vice versa, higher platelet count at baseline has been linked to ischaemic complications (42–44). Obviously, a stable platelet count is important to maintain vascular integrity, to meet haemostatic needs to prevent bleeding and to avoid thrombotic events on the other hand. Yet, data are not easily interpreted and, therefore, "newer" platelet indices might facilitate a better estimation of the patients’ (platelet) haemostatic balance. These newer platelet indices provide additional information regarding platelet size, granularity, age, or turnover.

The automated blood and platelet counters also display the distribution curve of platelets based on platelet volume. Unlike all other mammalian cells following normal distribution, the distribution curve is log normal, suggesting a high variability in...
platelet size, which seems to be the result of the unique endomi-
totic production of platelets from megakaryocytes (23). During
stable platelet production, there is a negative correlation of platelet
count with mean platelet volume (MPV), in order to maintain a
constant circulating mass of platelets (23). When steady-state is
disturbed, the initial response is rapid formation of larger, denser
and more reactive platelets, as after acute blood loss: The size of re-
ticulated platelets increase (45). There is a positive correlation of
platelet size with younger age, function, density (granules, mito-
chondria and functional organelles), synthesis of TXA₂, trans-
forming growth factor beta and with the density of expression of
GPIIb/IIIa per defined unit of plasma membrane (23).

Similarly to platelet size, after a decrease in circulating platelet
mass, megakaryocyte ploidy and volume is upregulated (27, 46–48). Megakaryocyte ploidy (and volume (49)) has been linked to
larger and more-reactive platelets in steady state as well as in
times of increased platelet formation (50). There is evidence that
megakaryocyte ploidy and volume is associated with increase in
MPV and platelet function (27, 46). How these obviously impor-
tant processes are regulated remains to be elucidated.

Since MPV is easy to measure, it has often been used as a sur-
rrogate marker to estimate platelet turnover. It is the most widely
investigated physical platelet variable and there is evidence in ACS,
that platelet volume might serve as a causal and prognostic factor
(23): Platelets of patients with ACS are denser, express more
GPIIb/IIa-receptors (51) and have increased volumes (52, 53);
there is a correlation of MPV with severity / acuity of CAD (54)
and it has been shown that with larger platelets, bleeding time is
reduced (55). Interestingly, these clinical observations seem to
hold true also in the current era of combined antiplatelet therapies
(56). Several studies have meanwhile shown that MPV – as sur-
rrogate marker for platelet turnover – might increase on-treatment
platelet function (►Table 1). Data on the direct comparison of
platelet function tests versus MPV in regard to cardiovascular out-
comes are scarce. One group has shown that MPV might be re-
lated to cardiovascular death following PCI, whereas platelet func-
tion according to the Verify Now assay might not be predictive for
cardiovascular death in this rather small cohort (57). The lifespan
of a platelet is 8–10 days, and about 90 % of platelets measured
within hours after the ACS would thus have circulated already be-
fore the ischaemic event. A causal relationship that the presence of
larger, more reactive platelets could trigger the development of ACS
seems plausible. However, a recent investigation has shown,
that MPV does not aid in diagnosing ACS in patients presenting at
emergency departments with chest pain (58), and another investi-
gation has suggested that MPV might not be related to the extent
of CAD (59). In contrast, decades ago, studies began suggesting a
causative role of MPV in ACS / progression of CAD (52), which
was confirmed in a large epidemiological study in the general
population, including approximately 40,000 persons (60). Of
these, 1,300 persons developed ACS over a four-year observation
period. MPV was found to best predict ACS, compared to estab-
lished cardiovascular risk factors (60). Meanwhile, profound evi-
dence has been generated which relates MPV independently with
cardiovascular outcomes. This was successfully summarised in a
recent meta-analysis confirming that MPV independently predicts
myocardial infarction, death after myocardial infarction or reste-
nosis after PCI (56).

A further confirmation of the causative role of platelet produc-
tion / quality of platelets with regard to development of athero-
sclerosis and its complications derives from previous studies on
bone marrow biopsies. Patients with sudden cardiac death had sig-
ificantly higher volumes of megakaryocytes compared to matched
persons who died from traumatic death (61). Fur-
thermore, in the same study, there was good correlation of MPV
with megakaryocyte size in the patients with sudden cardiac death
(61).

Platelet kinetics and platelet turnover

The gold standard for measuring platelet kinetics (including mean
platelet lifespan, platelet production rate) is radioactive labelling of
platelets, with disadvantages for patients and personnel inherent in
studies using radioactive tracers. Further problems are extracorpo-
real platelet manipulation with potential activation and loss of cer-
tain platelet populations as well as the duration of such tests (62).
In search for a new technique allowing fast information about the
thrombopoietic state, a flow cytometric method estimating platelet
turnover from peripheral blood offers a new and attractive answer
to this problem (63).

The principle of determination of platelet turnover from pe-
 peripheral blood affords a specific marker that identifies old versus
new platelets. In the late 1960s, it was described that newly re-
leased platelets contain residual mRNA and rough endoplasmatic
reticulum, which could be stained, i.e. the basis for the estimation
of platelet turnover from peripheral blood (45). In analogy to reti-
culocytes, young platelets were termed reticulated platelets. Sev-
eral dyes had been used for staining of platelets, of which thiazo-
le orange was the most popular (64). There is no doubt that reticu-
lated platelets represent the youngest platelets in circulation (35,
36), but drawbacks in (unspecific) staining with thiazol orange as
well as difficulties in standardising the technique has led to devel-
 opment of automated blood counters with the ability to determine
the reticulated platelet fraction (RPF) or reticulated platelet count
(RPC) from whole blood (65). Synonyms used for the results from
automated counters are „immature platelet fraction“ and „imma-
ture platelet count“. However, in our opinion these terms are mis-
leading, since the term „immature“ implies that these platelets
must undergo a process of maturation. Among the automated ana-
lysers, the most widely used is the Sysmex XE-2100 (65). Platelet
RNA is stained with two fluorescent dyes, polymethine and ox-
azine. The forward scatter light (cell volume) and fluorescence in-
tensity (RNA content) are measured and older and younger pla-
telets are identified (66). Compared to conventional thiazol orange
staining, there is moderate correlation, at least in patients with
thrombocytopenia with peripheral platelet destruction (67). In the
general population, RPF was between 1.7–3.4 %, which might in-
crease to 17 % in patients with peripheral platelet consumption or
destruction (67).
Referring to above, reticulated platelets increase their size in haemostatic needs (45). Furthermore, platelet size and age seem to be closely related (overlapping) as the RPF is much greater in a pool of larger platelets compared to a pool of smaller platelets (21) and RPF is highly intercorrelated with MPV suggesting that younger platelets are larger (21, 66). The (young) reticulated platelets differ from their older counterparts, as they are more reactive compared to older (and smaller) platelets (21, 68, 69).

A higher number of reticulated platelets (and elevated MPV) has been found in patients with stable CAD compared to controls (21, 70). Indeed, elevated consumption of platelets might be due to the interaction of platelets with the atherosclerotic vessel wall (71). In ACS, platelet kinetics are similarly altered: Several decades ago, it was shown that patients with myocardial infarction had a shorter mean platelet lifespan and an elevated platelet production rate compared to controls (72, 73) and a causal role of the megakaryocyte-platelet system with development or progression of atherosclerosis was discussed. Recent investigations could confirm the association of reticulated platelets with ACS. The RPF was significantly different, with a stepwise increase between healthy controls, patients with stable CAD, patients with non ST-elevation myocardial infarction and ST-elevation myocardial infarction (74). Similar results have been reported in studies comparing patients with or without ACS (75, 76), and an interesting confirmation of the findings derives from a study demonstrating increased platelet turnover in patients with previous definite stent thrombosis (77). We recently have shown that in patients with ACS there is a significant decline of RPF following successful treatment of coronary artery stenoses or occlusions (66). Otherwise, one group has shown that reticulated platelets might not assist in diagnosing ACS in patients presenting at emergency departments (58). Correlations between RPF and MPV as well as with platelet function tests encourage the hypothesis that in ACS platelets are not only larger, but younger and haemostatically more active (21, 69, 78) (Table 1). And it could be confirmed that reticulated platelets are more eager to participate in thrombus formation under shear conditions of coronary artery stenosis compared to older platelets (79). Interestingly, over 90% of the platelets measured at the time of an ischaemic event would have been in circulation before the event – an image of megakaryocyte activity one or two days before the ACS. Platelet consumption in chronic atherosclerosis (or even earlier in patients with endothelial dysfunction / activation) might alter megakaryocyte activity with presence of more active platelets that perpetuate atherosclerosis and / or changes in megakaryocyte activity, and presence of more active platelets might trigger the transition to ACS (Figure 1).

However, outcome data with regard to RPF are scarce: In one study of patients with ACS, RPF was independently associated with mortality (80). Results from a very small investigation also pointed in the same direction (81). Similarly, we recently have shown that RPF was independently predictive for major adverse cardiac events in patients undergoing coronary intervention with stenting, which was mainly driven by re-infarctions (66). Interestingly, vasodilator stimulated phosphoprotein-phosphorylation assay and multiple electrode aggregometry were not independently predictive with regard to the combined endpoint (66).

Antiplatelet drugs and enhanced platelet turnover

Antiplatelet drugs are an essential preventive measure in patients with CAD (82). According to recent guidelines, aspirin 75–100 mg daily is recommended indefinitely in all CAD patients, and the combination with a P2Y12-inhibitor (clopidogrel 75 mg daily, prasugrel 10 mg daily or ticagrelor 90 mg twice-daily) is recommended for patients with ACS or after coronary intervention for a certain time (83).

However, numerous studies have shown inter-individual variability in the (platelet) response to aspirin therapy. HTPR during treatment with aspirin is also linked with cardiovascular events (84). It seems that once-daily dosing of aspirin is inadequate for some patients (85), since platelet function and synthesis of platelet TXA2 recovers during a 24-h dosing interval (85–87). High-risk patients, i.e. diabetics, patients with severe atherosclerosis and / or with increased platelet turnover might benefit from different antiplatelet strategies (69, 88–90).

Furthermore, clopidogrel might not be effective in some patients due to various reasons, including genetic, metabolic, and others such as non-compliance, drug interactions, as well as several comorbidities (2, 15). Increased platelet turnover was identified as one factor associated with reduced antiplatelet effect of clopidogrel (21, 70, 78) (Table 1), being more important in the acute phase of ACS according to multivariable modelling (78). Numerous platelet function tests are predictive with regard to cardiovascular outcomes (18, 19, 91–95) and there seems to be a target zone of optimal platelet inhibition to minimise bleeding and ischaemic complications (19). A comprehensive review of recent advantages of platelet function testing in the cardiovascular arena is outside the scope of this review and has been elegantly reviewed elsewhere (15, 96).

It is not surprising that the ischaemic risk in patients with HTPR during treatment with clopidogrel compared to patients without HTPR is similar to the risk of patients on aspirin monotherapy, compared to patients with dual antiplatelet therapy (18, 95). Yet, after initial promising results, the transformation of this knowledge into safer and improved tailored antiplatelet regimens has failed so far (97–101). In light of the successful arrival of more potent P2Y12-receptor blocking agents, prasugrel and ticagrelor, in the ACS arena the neutral studies with regard to tailored antiplatelet therapy seem to have limited impact (13, 14, 102, 103). However, there have recently been reports that even during treatment with prasugrel, HTPR might be an issue (104), and enhanced platelet turnover could be defined as a factor contributing to HTPR (22) (Table 1).
Table 1: Antiplatelet drugs in patients with enhanced platelet turnover: MPV or reticulated platelets as estimators of platelet turnover and on-treatment platelet function. Abbreviations: MPV = mean platelet volume; LTA = light transmission aggregometry; WBA = whole blood aggregometry; VASP-P = vasodilator-stimulated phosphoprotein phosphorylation; CAD = coronary artery disease; STEMI = ST-elevation myocardial infarction; NSTEMI = Non ST-elevation myocardial infarction; PCI = percutaneous coronary intervention; ACS = acute coronary syndrome.

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<td>MPV PFA 100 Multiplate</td>
<td>211</td>
<td>CAD</td>
<td>Not increased</td>
<td></td>
</tr>
<tr>
<td>Durmaz et al. (140)</td>
<td>Observational</td>
<td>Aspirin</td>
<td>MPV PFA 100</td>
<td>69</td>
<td>CAD</td>
<td>Increased</td>
<td></td>
</tr>
</tbody>
</table>
Aspirin

TXA$_2$ acts as a potent feedback mediator and activates platelets via the platelet TXA$_2$ receptors (2). Aspirin mediates its antithrombotic effects via irreversible acetylation of platelet cyclooxygenase (COX) with 97–99% inhibition of platelet TXA$_2$ formation for the whole lifespan of a platelet (105, 106). The antithrombotic effects of blocking COX derive from shifting the balance between TXA$_2$/prostacyclin production towards prostacyclin (2).

Consequently, recovery of platelet TXA$_2$ synthesis and platelet function after administration of aspirin would theoretically require re-synthesis of platelet COX or the release of new platelets into the blood. There is considerable evidence that once-daily dosing of aspirin is not effective enough in high-risk patients like diabetics, or patients with severe CAD. Selected patients might thus benefit from individualised or intensified treatment strategies (85, 89, 90, 107). Of note, very high doses of aspirin might have inverse effects, since an increase in bleeding time is observed only after several hours when endothelial COX has recovered (108). Indeed, there is a relevant recovery of platelet function at the end of a 24-h dosing interval in patients as well as in healthy controls treated with once-daily aspirin (85, 86, 109, 110). Dating back to the 1980s, it has been shown that diabetics with enhanced platelet turnover recover platelet aggregation and TXA$_2$ synthesis significantly faster than healthy controls (111). More recent investigations have described HTPR during aspirin treatment in association with elevated levels of reticulated platelets (Table 1). In an elegant study, 60 volunteers received a single dose of 325 mg aspirin. Light transmission aggregometry was measured before and 24 hours after administration of aspirin. Participants were stratified according to tertiles of RPF: Aggregation, before and after aspirin was higher in the upper tertile, as was platelet TXA$_2$ synthesis recovery (112). Explanations might include that newly formed platelets contain mRNA to produce GPIIb/IIIa receptors, COX-2 and procoagulant proteins of the alpha-granules including fibrinogen and von Willebrand factor (112–114). The expression of COX-2 might be relevant, since COX-2 is not inhibited by low-dose aspirin, which gives rise to an increased synthesis of TXA$_2$ (5, 15).

Moreover, the elevated platelet turnover per se might be important: Non-acetylated platelets are released into the blood and, depending on the turnover rate, might cause insufficient overall platelet inhibition, especially at the end of the usual 24-h dosing interval. A recent investigation has questioned this hypothesis: In a randomised cross-over trial in diabetic patients, twice daily dosing of aspirin was associated with decreased platelet function, but markers of platelet turnover did not help to identify patients who benefited from intensified aspirin therapy (115). However, in essential thrombocythaemia – a natural model of increased platelet turnover – platelet TXA$_2$-recovery was faster compared to healthy controls receiving the same dose of aspirin (110, 116). In order to rule out that platelets in essential thrombocythaemia are "resistant" to aspirin, the addition of aspirin "in vitro" to these platelets led to complete abolishment of platelet TXA$_2$ synthesis (110). In summary, these findings suggest that elevated platelet turnover actually determines a less and shorter than expected antiplatelet effect of aspirin, leaving patients unprotected for several hours every day.

Clopidogrel and newer P2Y$_{12}$-inhibitors

Clopidogrel mediates its antithrombotic effect through irreversible inhibition of the platelet ADP P2Y$_{12}$-receptor (2). ADP derives primarily from dense granules of platelets and acts on platelets via the P2Y$_1$ and P2Y$_{12}$-receptors (117). ADP signaling via the specific platelet ADP receptors ultimately leads to platelet activation and aggregation via binding of fibrinogen to expressed GPIIb/IIIa receptors (2).
There is no doubt that clopidogrel has improved outcome of patients with CAD undergoing PCI and after ACS (82). Yet, in a certain proportion of patients clopidogrel lacks efficacy (18, 19, 91–95). Comparable to aspirin, since clopidogrel irreversibly blocks the platelet P2Y₁₂-receptor, full regeneration of platelet function requires exchange of the platelet pool by new, non-inhibited platelets. Although the plasma half-life of the active metabolite of clopidogrel (8 h) is markedly longer compared to plasma half-life of enteric-coated aspirin (4 h), it is an interesting hypothesis, that elevated platelet turnover might also partly explain HTPR during clopidogrel therapy (20). Several studies have meanwhile confirmed an independent positive association of platelet turnover with HTPR under clopidogrel, as assessed with different platelet function test systems (21, 70, 78) (Table 1). These data suggest that elevated platelet turnover might also suffice to overcome the platelet inhibitory effects of clopidogrel. With regard to the receptor specific VASP-assay, blockade of 90% of P2Y₁₂-receptors is translated into a platelet reactivity index of 50% (118), which is the recommended clinical cut-off for HTPR in this assay. Indeed the VASP assay is not very sensitive to low levels of platelet inhibition, but on the other side this seems to be the clinically relevant cut-off. If slightly more than 10% uninhibited P2Y₁₂-receptors suffice to cause ischaemic events, it is plausible that more than 10% of uninhibited newly released platelets might cause ischaemic events. However, in case the abovementioned findings are true, a diurnal variation in the antiplatelet effect of once-daily clopidogrel may be postulated.

Furthermore, we have recently speculated, whether this phenomenon might also hold true for prasugrel, as illustrated in a case-report of a 76-year-old women suffering two definite stent thromboses during dual antiplatelet therapy with clopidogrel and with prasugrel with HTPR under both substances. Indeed, the most interesting fact about this case was the distinct elevation of platelet turnover preceeding each stent thrombosis (20). Meanwhile, this hypothesis has been confirmed: In fact, elevated platelet turnover could be related to HTPR under prasugrel (22).

Conclusions and outlook

Patients with enhanced platelet turnover are at increased risk of developing atherothrombotic cardiovascular events such as ACS. Following ACS, the risk for further atherothrombotic complications is increased. Whether the shift of the megakaryocyte-platelet system towards a more aggressive platelet pool is cause or condition of atherothrombosis still remains elusive.

The most important question is, which signals regulate platelet turnover, size and activity. Solving this issue would allow us to enlarge our specific anti-atherothrombotic armamentarium:

With currently available drugs, only changes in dosing intervals may improve outcomes. A future perspective to reduce progression of atherothrombosis and platelet turnover lies in the modulation of adhesion of platelets to activated endothelium, i.e. blockade of GPIb (vWF, ADAMTS-13), P-selectin, GPVI or C-type lectin receptor-2 (2). Already, some promising results derive from a study selectively inhibiting thrombopoietin-dependent platelet formation (in baboons). Reduced platelet production and count has led to a significant reduction in arterial thrombosis without a major increase in bleeding complications (119).

Platelet turnover is a key factor for insufficient platelet inhibition in response to aspirin and clopidogrel treatment. Except for our own preliminary data, we are only aware of one study comparing specific agonist-dependent platelet function tests with platelet turnover in patients treated with antiplatelet drugs with regard to outcome (57, 66). It was not surprising that platelet function tests are of less predictive value compared to platelet turnover. Platelet turnover might represent a major prognostic factor, whereas specific platelet function tests in principle only allow an estimation of atherothrombotic risk in certain situations (if an otherwise helpful drug does not work, the risk is increased). Thus, direct comparisons do not seem relevant.

Conflicts of interest

None declared.

References