

Editorial Focus

Predicting thrombotic events: Creating a complex approach for a complex condition

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Effective antiplatelet therapy is one of the therapeutic cornerstones of cardiovascular medicine, with aspirin and clopidogrel being the most common antiplatelet drugs used in clinical practice. For example, the importance of aspirin therapy in cardiology patients is persuasively shown in a large number of trials. In the large meta-analysis by the Antithrombotic Trialists' Collaboration of 287 randomized trials of antiplatelet therapy of patients at high risk of occlusive vascular events, a 32% reduction of nonfatal myocardial infarction, non-fatal stroke and vascular death in patients treated with aspirin daily was demonstrated (1). Combination of aspirin and clopidogrel have been proven to be even more effective in certain conditions (for example, acute coronary syndromes and percutaneous coronary interventions [PCI]).

However, even despite the effectiveness of currently available antiplatelet agents, many patients fail to respond adequately to such therapy. This is evident by the occurrence of further clinical (thrombotic) events despite conventional antiplatelet treatment, which raises the following crucial questions: (i) how to increase the effectiveness of current antiplatelet therapies to prevent thrombotic events; and (ii) how to identify those under high risk of cardiovascular catastrophe? Thus, it is less of an issue of 'do we treat?' but rather a question of 'when to treat?' or 'how best to identify those patients to be treated?'

There are some similarities in relation to defining the problem of predicting of atherothrombotic events and so-called 'resistance' to aspirin and thienopyridines. Such 'aspirin resistance' has been defined in the laboratory setting by the failure of aspirin to sufficiently inhibit platelet aggregation (2). Indeed, laboratory-defined aspirin resistance can adversely affect the risk of cardiovascular complications. For example, amongst patients with prior ischemic attack or stroke, the incidence of aspirin resistance was significantly higher compared to asymptomatic patients with known cerebrovascular disease (3). At two-year follow-up, aspirin non-responders had a 10-fold increase in the risk of recurrent vascular events as compared to aspirin-sensitive patients (4). In patients with stable coronary artery disease undergoing PCI aspirin resistance was associated with a significant in-

crease in the risk of myocardial infarction, stroke, or death (hazard ratio 3.12) after 2.1 years follow-up.

A recent systematic review noted that patients who are resistant to aspirin are at a greater risk of clinically important cardiovascular morbidity long term than patients who are sensitive to aspirin, again emphasising the requirement of tests predicting adverse events (5). Of note, clopidogrel resistance, as defined by ADP-induced platelet aggregation, has also been reported to be associated with increases in thrombotic complications, again emphasising the need for reliable tests predicting the risk of adverse thrombotic events (6).

It is increasingly recognised that the antiplatelet effect of aspirin has some inter- and intra-patient variability (7). Indeed, thrombotic events still occur in patients who take their medication regularly, although it is recognised that some cases of aspirin resistance may in reality be due to poor drug compliance. Thus, the crucial unanswered question is 'how best to identify those patients at risk?'

A number of laboratory approaches have been developed to evaluate individual effectiveness of antiplatelet drugs (8). For example, light or optical aggregometry, which is generally considered the "gold standard" of platelet function assessment, is the most widely used technique for defining aspirin resistance. The main advantage of platelet aggregometry is that this technique estimates, although in an ex-vivo system, the most important function of platelets: their mutual aggregation within a glycoprotein IIb/IIIa-dependent manner.

Platelet aggregometry can predict major adverse cardiac events (9, 10). For example, patients receiving chronic clopidogrel therapy undergoing non-emergent PCI, who exhibit high on-treatment ADP-induced platelet aggregation, are at increased risk for post-procedural ischemic events suggesting potential utility of the approach to predict the risk of treatment failure (11).

Nonetheless, there are major disadvantages to platelet aggregometry as a clinical test of platelet function. For example, there is the necessity to run the assay rapidly (usually within 1–3 hours of blood collection) and with the necessary operator and inter-

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pre-er experience to guarantee sensitivity, specificity, and reproducibility.

Indeed, estimates of the prevalence of aspirin resistance vary widely (from 5.5% to 60%), reflecting the diversity of various laboratory assays and confounding from the broad range of disease states investigated (12). For this reason, most researchers are increasingly advocating the application of various laboratory tests with increasing number of novel approaches including the VerifyNow (Accumetrics, San Diego, CA, USA), Platelet Function Analyzer 100 (PFA-100; Dade Behring, Newark, DE, USA), and vasodilator-stimulated phosphoprotein (VASP) phosphorylation state (BioCytex, Marseilles, France) (13). Because of the variability among patients in the response of their platelets to antiplatelet therapy, there is increasing interest in the use of platelet function tests to monitor the effects of antiplatelet drugs, with the ultimate goal of guiding antiplatelet therapy to the optimal dose for prevention or treatment of thrombosis while minimizing side effects (14).

All of these methods have their own advantages, including the use in clinical settings, simplicity of use and a strict manufacturer developed procedure that may improve reproducibility of results. For example, the PFA-100 assay draws an anticoagulated blood sample under high-shear conditions through collagen-coated aperture in the presence of ADP or epinephrine providing advantages of simplicity, rapidity, low sample volume, physiologically relevant high shear. Aspirin non-responder status according to PFA-100 in patients with recurrent cerebral ischemic attacks has been reported to predict major adverse cardiac events (9).

However, hopes for overcoming the problem of reliable antiplatelet treatment monitoring using novel laboratory methods have not confirmed in all clinical studies. The prevalence of aspirin resistance – as defined by various tests – vary widely: the lowest figures are seen with optical aggregometry using arachidonic acid (6%) and the highest prevalence with the PFA-100 analyzer (26%) (15). Indeed, novel measures of platelet function (e.g. PFA-100) often lack of agreement with optical aggregometry in detection of aspirin resistance (16). Gum et al. reported a prospective study of aspirin resistance in 326 patients with stable cardiovascular disease and found the prevalence of aspirin resistance was 5.2% by platelet aggregation and 9.5% by PFA-100 (9). After 1.9 years follow-up, the major end-point of

death, myocardial infarction or stroke, occurred in 24% aspirin-resistant patients as determined by optical aggregation, and 10% patients were not aspirin-resistant. However, when they used PFA-100 to define aspirin resistance, there was no significant difference in end-points between those aspirin-resistant (15.1%) and those who were aspirin-sensitive (12.9%) (9). The incidence of aspirin non-responsiveness as 17% using the Ultegra-RPFA, 22% for PFA-100 compared with only 5% by optical aggregometry (17). Of note only 2% of patients were aspirin non-responders by all 3 tests (17).

But even leaving aside individual technical limitations of the available approaches, the limitation common for all tests is that platelet aggregation is evaluated separately using different agonists/stimulants. This limitation is based on the pathophysiology of platelet activation itself which in its very nature may be stimulated by multiple pathways. Platelet physiologists have admitted for a long time that it may not sufficient to block only one of such pathways (e.g. inactivation of cyclooxygenase by aspirin or blockade of platelet ADP receptors by thienopyridines), and this provides the basis for progressively more common combination antiplatelet therapy (18). Similarly, it might be difficult to expect that stimulation of any single pathway of platelet activation would provide comprehensive information on platelet aggregation status, and such information would probably be most accurate for the prediction of thrombotic events.

To overcome this limitation, the work by Gori et al. in the current issue of *Thrombosis and Haemostasis* proposes the use of a combination of platelet agonists for optical aggregometry, revealing an integrated platelet proaggregatory status in response to all major stimulation pathways together (19). Such an approach may indeed be more successful for predicting the risk of thrombotic events compared to separate use of the individual platelet agonists. It is especially important since the combined platelet stimulation performed by the authors includes pathways that currently serve as common targets for antiplatelet therapy in clinical practice (19). Their results may serve as a basis for further studies and intervention trials to validate this novel approach and furthermore, validate the concept that use of multiple agonists may become the method of choice for platelet treatment monitoring and – if validated – be predicting of adverse cardiovascular events.

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