Effective platelet inhibition has become a cornerstone in the management of patients with acute coronary syndromes (ACSs). The addition of clopidogrel, a blocker of the ADP platelet receptor, to aspirin therapy significantly reduces major cardiovascular events in ACS patients and reduces the frequency of percutaneous coronary intervention (PCI) with stent implantation. As PCI became the dominant approach for myocardial revascularization, clopidogrel use grew steadily, and clopidogrel is now the second most highly prescribed medicine in the US. Even though clopidogrel was approved for use in 1997, its mechanism of action continues to be progressively understood.

Evidence for the Involvement of CYP2C19

The response to clopidogrel is highly variable among patients. Patients with low or incomplete platelet inhibition (so-called high residual on-clopidogrel platelet reactivity) are at a greater risk of cardiovascular events (1). Multiple mechanisms have been proposed for the variable response to clopidogrel. Pharmacokinetic and pharmacodynamic analyses have established that a substantial portion of the variation in platelet response is explained by the variability in plasma concentrations of the active metabolite of clopidogrel (2–4). Clopidogrel is a prodrug that requires bioactivation to achieve its antiplatelet efficacy. Only 15% of the produg is available for transformation to the active agent; the remaining 85% is hydrolyzed by esterases into inactive forms. The cytochrome P450 enzyme CYP2C19 is involved in the metabolism of clopidogrel (5). Like many other members of the cytochrome P450 gene superfamily, the CYP2C19 (cytochrome P450, family 2, subfamily C, polypeptide 19) gene is highly polymorphic, with >25 variant alleles known. The most common CYP2C19 loss-of-function allele is *2 (c.681G>A; rs4244285), with allele frequencies of approximately 15% in Caucasians and Africans, and 29%–35% in Asians. Carriers of two *2 alleles are defined as poor metabolizers, whereas carriers of only one *2 allele are defined as intermediate metabolizers and noncarriers are defined as extensive metabolizers. On the other hand, the CYP2C19*17 allele (c.-806C>T; rs12248560) leads to increased CYP2C19 activity, owing to enhanced transcription. The allelic frequency ranges from 3% to 21%, depending on ethnicity (6).

In 2006, we documented a >25% reduction in platelet responsiveness to clopidogrel in healthy volunteer carriers of the loss-of-function CYP2C19*2 allele (7). Since then, many pharmacologic studies have corroborated this result and further showed that CYP2C19*2 carriers have reduced active clopidogrel metabolites (2, 3). Subsequently, substantial evidence has linked the CYP2C19 genotype with the clinical response among clopidogrel-treated patients with ACS (8, 9). The growing body of literature implicating CYP2C19*2 in adverse clopidogrel responses prompted the US Food and Drug Administration to implement a boxed warning on the clopidogrel label describing the relationship between CYP2C19 pharmacogenetics and drug response and particularly noting the diminished effectiveness in poor metabolizers (10). Recent evidence also suggests that clopidogrel response is improved in carriers of the CYP2C19*17 allele (11, 12).

Given these data, the use of CYP2C19 genetic testing to guide antiplatelet therapy in patients with coronary artery disease appears to be an appealing strategy. CYP2C19 genetic testing is already available through academic and commercial clinical laboratories. The enthusiasm for CYP2C19 genetic testing, however, has been tempered by 4 issues that have not been properly addressed or clearly understood.

1. Whom to Test?
A crucial challenge is clearly defining the target population for clopidogrel genetic testing. The aim is to use a test that identifies the right drug at the right dose for the right patient; however, this aim would be achieved...
only by offering the right test at the right time under the right clinical conditions. Importantly, the most definitive studies that have showed a relationship between CYP2C19 genotype and clopidogrel response have been conducted in ACS patients (8). Thus, there is currently no evidence for applying such findings to other indications for clopidogrel, including atrial fibrillation, stroke, peripheral artery diseases, and chronic stable angina, and there is no evidence supporting the systematic use of CYP2C19 genetic testing in all clopidogrel-treated patients (6).

In addition, CYP2C19 genetic testing would not apply to all ACS patients. The strongest association between CYP2C19 genotype and adverse cardiovascular outcomes appears to be reserved for those patients undergoing PCI with stenting. The genetic substudy of the large-scale PLATO (Platelet Inhibition and Patient Outcomes) trial investigating clopidogrel vs ticagrelor in ACS found a significant effect of the CYP2C19*2 allele in patients undergoing PCI (odds ratio for *2 carriers vs noncarriers, 1.33; 95% CI, 1.03–1.71) but no such effect in medically managed patients (odds ratio for *2 carriers vs noncarriers, 0.93; 95% CI, 0.66–1.32) (13, 14). Similarly, CYP2C19 genotype had no significant effect in the CURE (Clopidogrel in Unstable Angina to Prevent Recurrent Events) trial, in which <15% of the ACS patients underwent PCI (15), but it did have a highly significant effect in the TRITON (Trial to Assess Improvement in Therapeutic Outcomes by Optimizing Platelet Inhibition with Prasugrel) trial, in which all of the patients were treated with PCI (3). Finally, most of the evidence supports a greater influence of CYP2C19 genotype on the events occurring within the first postprocedure month, particularly stent thrombosis, a procedural complication that can be prevented with effective platelet inhibition (8, 9).

2. HOW TO TEST?
CYP2C19 genotyping is currently available through a number of laboratories; however, the turnaround time is often several days, which is too long to efficiently translate test results into effective therapy for ACS patients with a PCI. A large number of events happen within the first hours after a PCI, and any adaptation of treatment should be proposed early in the patient’s management. During the last decade, interventional cardiologists have been very favorably disposed toward adjusting antiplatelet therapy, especially with clopidogrel. Numerous teams have evaluated higher clopidogrel loading doses at the time of PCI and found that a higher dose was associated with a decrease in postprocedural atherothrombotic complications (16). Interventional cardiologists have also tended to use point-of-care assays to monitor platelet function, even with patients with high platelet reactivity on treatment, and have adapted antiplatelet therapy for these patients (1, 16). The key factor in both situations was a more rapid access to the results and to the intervention. A rapid and low-cost bedside test will thus be required to allow interventional cardiologists to obtain a rapid genetic profile of the patient under treatment and to integrate such information into a viable therapeutic algorithm. Another option is to perform broad genotyping (beyond CYP2C19) before any particular clinical disease arises so that genotyping information will be available immediately when an event occurs. This more general approach will require extensive developments in bioinformatics so that each patient’s profiling and relevant genetic information is efficiently picked up and used at the time of an event. It is noteworthy that combining genetic testing with clinical variables and/or with a platelet function assay may provide valuable information to assist the physician in further adapting treatment.

3. HOW TO INTERPRET RESULTS?
One of the major objections to the use of CYP2C19 genetic testing is that CYP2C19 explains only a limited portion of the variable response to clopidogrel (estimated at “only” 12% in a genomewide association study) (17). The misleading assumption here is that simply assessing a genotype would allow one to distinguish clopidogrel responders from nonresponders directly and, ultimately, the patients who would present clinical outcomes linked to an inappropriate response to clopidogrel. Such an expectation is not a realistic one because the response to clopidogrel is not monogenic (in contrast to a genetic disease) but rather is influenced by multiple factors and multiple genes. The current distinction between responders and nonresponders is defined in most cases by the results of point-of-care platelet assays that may not correlate strongly with clinical events. Thus, the CYP2C19 genetic profile must be seen as a risk factor for an impaired response to clopidogrel and to subsequent recurrent cardiovascular events. Two metaanalyses (8, 9) have shown that CYP2C19*2 carriers treated with clopidogrel are at an increased risk for cardiovascular events, with the risk showing a gene dosage effect. Compared with noncarriers, the risk for developing major adverse cardiovascular events is 1.55 (95% CI, 1.11–2.17) for heterozygotes (intermediate metabolizers) and 1.76 (95% CI, 1.24–2.50) for homozygotes (poor metabolizers). The increased risk of stent thrombosis is 2.67 (95% CI, 1.69–4.22) for heterozygotes and 3.97 (95% CI, 1.75–9.02) for homozygotes.

Actually, clopidogrel prescribers are well aware of this concept, because the underlying conditions they are treating are also influenced by multiple risk factors. For instance, patients with arterial hyperten-
or high LDL cholesterol concentrations have a higher risk for developing atherosclerosis and consequent cardiovascular outcomes, but the predictive performance is not absolute. The integration of the CYP2C19 genetic profile into a global clinico-genetic model would help in interpreting the results. Similarly, other genetic variants are likely to influence clopidogrel response, and such effects may be additive to those of CYP2C19 genotype. Some gene candidates have recently been proposed, but they require more validation.

4. HOW TO RESPOND TO CYP2C19 GENOTYPE RESULTS
Two options might ameliorate the risk in patients with an impaired response to clopidogrel: switching to a novel P2Y12 receptor inhibitor or increasing the clopidogrel dose. Prasugrel and ticagrelor are new antiplatelet agents that, like clopidogrel, act on the P2Y12 platelet receptor, but their antiplatelet activity does not depend on CYP2C19; however, prasugrel and ticagrelor may not be substitutes for clopidogrel in all patients. The use of prasugrel is associated with an increased risk of bleeding (including fatal bleeding), is contraindicated in some patients, and represents a higher expense as clopidogrel progressively comes off patent worldwide. Ticagrelor has just been approved in the US and has an efficacy and safety profile similar to that of prasugrel (18), according to indirect-comparison data. On the other hand, the systematic use of 150 mg clopidogrel per day in PCI patients with a high on-clopidogrel platelet reactivity in the GRAVITAS (Gauging Responsiveness with a VerifyNow Assay—Impact on Thrombosis and Safety) trial did not significantly reduce the rate of cardiovascular events compared with 75 mg clopidogrel per day (19). In a recent pharmacokinetic/pharmacodynamic study, we found that clopidogrel resistance could be overcome by increasing the loading dose to 900 mg in *1/*2 heterozygotes but not in *2/*2 homozygotes (2).

The current literature supports the use of new thienopyridine agents in CYP2C19 *2/*2 homozygotes, provided it is not contraindicated clinically. Patients with CYP2C19 extensive metabolizer or rapid metabolizer phenotypes (i.e., *1/*1, *1/*17, and *17/*17) might receive standard clopidogrel dosing as recommended in the new product insert (see Fig. 1). Finally, the most challenging patient population is the CYP2C19 intermediate-metabolizer phenotype (*1/*2), because some studies, but not all, have suggested that high residual platelet reactivity might be improved by increasing the clopidogrel dosage in these patients. Given the negative results from the GRAVITAS trial (19), the switch to new P2Y12 receptor inhibitors appears to be a preferable option, but more data are clearly needed. Overall, the options exist, but the optimal therapeutic strategy remains to be clearly defined.

Rapid and easy access to CYP2C19 genetic testing will be an incentive to integrate this marker into the optimal therapeutic algorithm (e.g., Fig. 1).

Conclusions

CYP2C19 genetic testing is able to identify subgroups of patients who will not benefit from the standard clopidogrel therapy. The future relies on a rapid, low-cost genetic test that will optimally integrate the CYP2C19 genotype into accurate predictive algorithms and thus allow efficient genotype-guided antiplatelet therapy.

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References


