Pharmacodynamic Comparison of Pitavastatin Versus Atorvastatin on Platelet Reactivity in Patients With Coronary Artery Disease Treated With Dual Antiplatelet Therapy – The PORTO Trial –
Francesco Pelliccia, MD, PhD; Giuseppe Rosano, MD, PhD; Giuseppe Marazzi, MD, PhD; Cristiana Vitale, MD, PhD; Ilaria Spoletini, MD; Ferdinando Franzoni, MD; Giuseppe Speziale, MD; Marina Polacco, MD; Cesare Greco, MD; Carlo Gaudio, MD

Background: Levels of platelet reactivity in patients on dual antiplatelet therapy (DAPT) can be influenced by concomitant treatment with statins. We verified if the pharmacodynamic effects of CYP3A4-metabolized statins (atorvastatin) and non-CYP3A4-metabolized statins (pitavastatin) differ in patients with coronary artery disease (CAD) treated with DAPT.

Methods and Results: A total of 155 CAD patients receiving DAPT (clopidogrel 75 mg plus aspirin 100 mg) entered the PORTO trial. Patients were randomly assigned to atorvastatin (20 mg day) or pitavastatin (4 mg day) for 30 days, and then switched to the other drug for 30 days. Platelet reactivity was expressed as VerifyNow P2Y12 platelet response units (PRU) before and after each 30-day treatment period. High platelet reactivity was defined as PRU >208. As compared with pretreatment (192±49), PRU was significantly higher after 30-day atorvastatin (210±56; P=0.003), but was unchanged after 30-day pitavastatin (199±47 PRU, NS). In the 48 patients with PRU >208 at baseline (232±44), PRU increased significantly after 30-day atorvastatin (258±41; P=0.004), but not after 30-day pitavastatin (237±43, NS). In the 107 patients with PRU <208 at baseline (174±52), PRU did not change significantly with respect to baseline either after 30-day atorvastatin (188±61, NS) or after 30-day pitavastatin (181±59, NS).

Conclusions: Pitavastatin, a non-CYP3A4-metabolized statin, does not affect clopidogrel’s response as compared with atorvastatin in patients who are borderline or poor responders to DAPT. (Circ J 2014; 78: 679–684)

Key Words: Clopidogrel; Coronary artery disease; Percutaneous coronary intervention; Platelet reactivity

High platelet reactivity in patients on dual antiplatelet therapy (DAPT) has been shown to be an independent risk factor for recurrent ischemic events.1,2 Levels of platelet reactivity can be influenced by concomitant treatment with medications (ie, statins) that might inhibit the CYP4504 system involved in the activation of clopidogrel.3,4 Although all statins share a common mechanism of action, they differ in terms of their chemical structures, pharmacokinetic profiles, and lipid-modifying efficacy.5-8 The chemical structures of statins govern their water solubility, which in turn influences their absorption, distribution, metabolism and excretion.9 Atorvastatin, simvastatin, lovastatin, fluvastatin, and pitavastatin are relatively lipophilic compounds, but pravastatin and rosuvastatin are relatively hydrophilic and not significantly metabolized by cytochrome P(450) enzymes.10 Lipophilic statins are more susceptible to metabolism by the cytochrome P(450) system, except for pitavastatin, which is mostly excreted unchanged in bile and undergoes minimal biotransformation through the cytochrome P450 system.11-16

Editorial p 592

We reasoned that consideration of the differences among the lipophilic statins helps to provide a rational basis for their...
use in clinical practice, especially in patients who are receiving DAPT. Accordingly, the primary objective of this study was to compare the pharmacodynamic effects of a CYP3A4-metabolized statin (atorvastatin) with those of a non-CYP3A4-metabolized statin (pitavastatin) in patients with coronary artery disease (CAD) treated with DAPT.

Methods
Study Population
All stable patients with angiographically documented CAD who underwent percutaneous coronary intervention with 1 or more drug-eluting stents between January 2011 and December 2012 were screened for the PORTO trial. By protocol, only patients on chronic (>10 days) therapy with clopidogrel (75 mg/day) were considered for the study. Patients were not considered eligible if they had contraindications to statin treatment, platelet function disorder or platelet count <150,000/ml, hemoglobin<10 g/dl, need for warfarin treatment, active liver disease or liver cirrhosis, unexplained transaminase increase >2-fold the upper limit of normal, peripheral muscle disease or creatine kinase >2.5-fold the upper limit of normal, current treatment with proton-pump inhibitors and/or omega-3, acute renal failure or endstage renal failure requiring dialysis, active bleeding or recent bleeding diathesis (within the past month), previous hemorrhagic stroke, malignancy, and refusal of consent. The study was conducted in accordance with the Declaration of Helsinki, and was approved by the Institutional Board Review Committee. All eligible patients gave informed, written consent. The PORTO trial is registered at ClinicalTrials.gov (Identifier: NCT01648829).

Study Design
The PORTO trial was a prospective, randomized, 2-period, cross-over trial. The study design is illustrated in Figure 1. Patients were screened at the time of 1-month post-angioplasty follow-up visit and only those who were deemed to be eligible for the study were asked to participate in the PORTO trial. A total of 155 patients agreed, entered the initial wash-out period from any concomitant statin therapy and 1 week later, all patients had a VerifyNow P2Y12 assay (Accumetrics, San Diego, CA, USA) to assess their baseline platelet function. Enrolled patients were then randomly assigned in a 1:1 fashion to a regimen of atorvastatin (20 mg day) or pitavastatin (4 mg day) for 30 days. After another 1-week wash-out period to avoid any carryover effect, cross-over was performed and patients switched to the other statin, which was continued for 30 days (Figure 1). Patients were advised to take clopidogrel at 06:00 hours and the statin dose at 22:00 hours throughout the study period. Platelet reactivity was measured at baseline after the 1-week statin wash-out period and at the end of each 30-day treatment period. Compliance and adverse events were assessed by a single investigator (M.P.) based on interview, pill counting, and a questionnaire.

The primary endpoint of the trial was the change in P2Y12 reaction units (PRU) as compared with baseline values after each 30-day treatment period.

Platelet Reactivity
Blood samples for platelet function testing were collected 2 h after the ingestion of the last clopidogrel dose (06:00 hours) and 10 h after the ingestion of the last statin dose (22:00 hours). Platelet reactivity was evaluated by the VerifyNow P2Y12 assay, which is a whole blood, point-of-care turbidimetric assay that measures responsiveness to P2Y12 antagonists. The test measures ADP-induced platelet agglutination as an increase in light transmittance and uses a proprietary algorithm to report values in PRU, which measures platelet aggregation in separate channels in response to ADP. Also, the device estimates the maximal platelet function independent of P2Y12 receptor blockade (BASE), which represents total platelet function in response to thrombin receptor-activating peptide. The instrument provides the percent platelet P2Y12 inhibition (IPA), calculated by comparing the PRU from the ADP channel to the PRU from the thrombin receptor-activating peptide channel. High platelet reactivity was defined as PRU >208, a cut-off value previously proposed. The PRU cut-off value of 235 proposed by the GRAVITAS investigators was also considered.

Coronary Angiography and Intervention
All interventions were performed according to standard techniques. Analysis of coronary angiograms obtained before and
after angioplasty was independently performed by the core laboratory ROMA (Ricerche Orientate alla Malattia Aterosclerotica). Percutaneous coronary intervention was performed according to current standard guidelines, with the type of stent implanted left to the discretion of the operator.

**Adjunct Drugs**

According to our institutional protocol, all stable patients scheduled to undergo elective angiography are prescribed aspirin (100 mg/day) and clopidogrel (300 mg loading dose followed by a maintenance dose of 75 mg/day) for at least 7 days. During angioplasty, patients received procedural anticoagulation with weight-adjusted doses (100 U/kg) of unfractionated heparin and additional doses of unfractionated heparin given to achieve and maintain activated clotting time >300 s. After angioplasty, the protocol-mediated antiplatelet therapy consisted of aspirin 100 mg/day indefinitely and clopidogrel 75 mg/day for 12 months. Other medications such as β-blockers, angiotensin-converting enzyme inhibitors, and angiotensin-receptor blockers were given as appropriate. Other lipid-lowering treatment or medications that affect CYP3A4–mediated drug metabolism were not permitted during the study, including erythromycin, antimycotic agents, and cyclosporine. Any change in medications was not permitted during the study period.

**Sample Size Estimation**

The sample size calculation was performed by assuming mean ± standard deviation (SD) of PRU under atorvastatin treatment of 180 ± 50. The null hypothesis was that there was no difference in clopidogrel’s effect between atorvastatin and pitavastatin groups, and the study was powered to reject this hypothesis. We calculated that we needed to include 146 patients to be able to detect a 15% difference in mean PRU values with a power of 95% and a 2-sided α value of 0.05.

**Statistical Analysis**

Data are presented as mean ± SD for continuous variables or frequency percentages for categorical variables. Kolmogorov-Smirnov testing was applied to assess normality of distribution for continuous variables. Chi-square test, or Fisher’s exact tests, when appropriate, were used to compare differences between categorical variables. Non-normally distributed continuous variables were compared by Kruskal-Wallis test and Mann-Whitney U test. The repeated-measure analysis of variance was used to evaluate platelet reactivity changes over time. All analyses were performed with the S-Plus statistical package (Mathsoft Inc, Seattle, WA, USA). A 2-sided P value < 0.05 was considered statistically significant.

**Results**

**Patients Characteristics**

Study patients demographics and baseline characteristics are shown in **Table 1**. The mean age for the entire study cohort was 65 ± 16 years; 22% of patients had diabetes mellitus, 58% hypertension and 76% dyslipidemia. A significant proportion of patients had had myocardial infarction and a previous revascularization procedure, and 59% showed multivessel CAD.

**Study Treatment**

All patients enrolled in the study underwent all the platelet reactivity testing as scheduled, and all were compliant with the study protocol. As regards side effects, 4 patients (2 during atorvastatin and 2 during pitavastatin treatment) had a transaminase increase >2-fold the upper limit of normal. Withdrawal atorvastatin and 2 during pitavastatin treatment) had a transaminase increase >2-fold the upper limit of normal. Withdrawal from the study drug for non-medical reasons occurred in 1 case during the 30-day atorvastatin treatment and in 1 during pitavastatin treatment.

**Overall Platelet Reactivity**

Results of platelet reactivity at each time point for both groups are presented in **Table 2**. Statistical analysis showed that the results of platelet reactivity assessment were normally distributed. As compared with the pretreatment value (192 ± 49), PRU was significantly higher after the 30-day treatment period with atorvastatin (210 ± 56; P = 0.003 vs. baseline), but was unchanged after the 30-day treatment period with pitavastatin (199 ± 47, NS vs. baseline) (**Figure 2**). BASE values did not significantly change as compared with pretreatment evaluation either after the 30-day treatment with atorvastatin or after 30-day treatment with pitavastatin. Similarly, the mean IPA did not significantly change as compared with pretreatment evaluation either after 30-day treatment with atorvastatin or after 30-day treatment with pitavastatin (**Figure 2**).

**Platelet Reactivity in Patients With Baseline Normal or High PRU**

Differences in platelet reactivity with atorvastatin were mainly related to individual baseline values of PRU (**Table 3**). In the 48 patients with a PRU > 208 at baseline (mean ± SD

---

**Table 1. Baseline Clinical and Angiographic Characteristics of the Study Patients**

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Overall study population (n=155)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age (years)</strong></td>
<td>65 ± 16</td>
</tr>
<tr>
<td><strong>Male sex</strong></td>
<td>110 (71%)</td>
</tr>
<tr>
<td><strong>Medical history</strong></td>
<td></td>
</tr>
<tr>
<td>Current smoking</td>
<td>74 (48%)</td>
</tr>
<tr>
<td>Hypertension</td>
<td>90 (58%)</td>
</tr>
<tr>
<td>Total cholesterol &gt;200 mg/dl</td>
<td>118 (76%)</td>
</tr>
<tr>
<td>Diabetes mellitus</td>
<td>31 (20%)</td>
</tr>
<tr>
<td>History of angina</td>
<td>60 (39%)</td>
</tr>
<tr>
<td>Previous myocardial infarction</td>
<td>46 (30%)</td>
</tr>
<tr>
<td>Previous PCI</td>
<td>38 (25%)</td>
</tr>
<tr>
<td>Previous CABG</td>
<td>21 (14%)</td>
</tr>
<tr>
<td><strong>Laboratory data</strong></td>
<td></td>
</tr>
<tr>
<td>Angiographic features</td>
<td></td>
</tr>
<tr>
<td>1-vessel</td>
<td>63 (41%)</td>
</tr>
<tr>
<td>2- or 3-vessel</td>
<td>91 (59%)</td>
</tr>
<tr>
<td>LV ejection fraction</td>
<td>58 ± 13</td>
</tr>
<tr>
<td>Blood creatinine (mg/dl)</td>
<td>1.21 ± 0.52</td>
</tr>
<tr>
<td>Total cholesterol (mg/dl)</td>
<td>221 ± 61</td>
</tr>
<tr>
<td>Triglyceride (mg/dl)</td>
<td>145 ± 69</td>
</tr>
<tr>
<td>HDL cholesterol (mg/dl)</td>
<td>39 ± 20</td>
</tr>
<tr>
<td>LDL cholesterol (mg/dl)</td>
<td>132 ± 31</td>
</tr>
<tr>
<td><strong>Concomitant medications</strong></td>
<td></td>
</tr>
<tr>
<td>β-blockers</td>
<td>138 (89%)</td>
</tr>
<tr>
<td>Calcium antagonist</td>
<td>37 (29%)</td>
</tr>
<tr>
<td>ACEI</td>
<td>119 (77%)</td>
</tr>
<tr>
<td>Angiotensin-receptor blockers</td>
<td>39 (25%)</td>
</tr>
<tr>
<td>Nitrates</td>
<td>68 (44%)</td>
</tr>
</tbody>
</table>

Data are n (%) or mean ± SD. ACEI, angiotensin-converting enzyme inhibitor; CABG, coronary artery bypass grafting; LV, left ventricular; PCI, percutaneous coronary intervention.
30 days is associated with improved rates of optimal clopidogrel response as compared with atorvastatin. Noteworthy, the favorable effects of pitavastatin mainly occurred in patients showing high platelet reactivity while on DAPT.

Clopidogrel is a prodrug metabolized in the liver via the cytochrome P450 (CYP) 3A4 system to the active compound that inhibits the P2Y(12) ADP platelet receptor. Some studies suggested that lipophilic statins (ie, atorvastatin, lovastatin and simvastatin) may competitively inhibit CYP3A4 and decrease the generation of clopidogrel’s active metabolite, thereby reducing its antiplatelet effects.

Lau et al originally found that atorvastatin but not pravastatin treatment was associated with a dose-dependent reduction of the antiplatelet activity of clopidogrel 6–8 days after clopidogrel initiation. Their findings were not confirmed subsequently by other investigators who did not observe any drug-drug interaction between clopidogrel and lipophilic statins. However, the recent ACCEL-STATIN study showed that in clopidogrel-treated patients with high platelet reactivity during chronic coadministration of atorvastatin, switching to a non-CYP3A-metabolized statin resulted in a significant decrease in platelet reactivity and the prevalence of high platelet reactivity. The investigators concluded that their study supported the beneficial effect of replacing atorvastatin therapy with a non-CYP3A-metabolized statin with regard to reducing platelet reactivity in clopidogrel-treated patients with high platelet reactivity. The authors stated that the ACCEL-

<table>
<thead>
<tr>
<th>Table 2. Distribution of Platelet Function Parameters Over Study Time Course</th>
</tr>
</thead>
<tbody>
<tr>
<td>PRU</td>
</tr>
<tr>
<td>PRU</td>
</tr>
<tr>
<td>Absolute PRU difference from baseline</td>
</tr>
<tr>
<td>Relative PRU difference from baseline (%)</td>
</tr>
<tr>
<td>BASE</td>
</tr>
<tr>
<td>Absolute PRU difference from baseline</td>
</tr>
<tr>
<td>Relative PRU difference from baseline (%)</td>
</tr>
<tr>
<td>IPA, %</td>
</tr>
</tbody>
</table>

Data are n (%) or mean ± SD. *P=0.003 as compared with corresponding value at baseline.

BASE, P2Y12 receptor blockade; IPA, Platelet P2Y12 inhibition; PRU, P2Y12 reaction units.

Discussion

This is the first prospective, randomized, cross-over study powered to detect differences between atorvastatin and pitavastatin regarding possible interferences with the antiplatelet efficacy of clopidogrel maintenance treatment in patients with CAD. Results of the PORTO trial show that the use of pitavastatin for 30 days is associated with improved rates of optimal clopidogrel response as compared with atorvastatin. Noteworthy, the favorable effects of pitavastatin mainly occurred in patients showing high platelet reactivity while on DAPT.

Clopidogrel is a prodrug metabolized in the liver via the cytochrome P450 (CYP) 3A4 system to the active compound that inhibits the P2Y(12) ADP platelet receptor. Some studies suggested that lipophilic statins (ie, atorvastatin, lovastatin and simvastatin) may competitively inhibit CYP3A4 and decrease the generation of clopidogrel’s active metabolite, thereby reducing its antiplatelet effects. Lau et al originally found that atorvastatin but not pravastatin treatment was associated with a dose-dependent reduction of the antiplatelet activity of clopidogrel 6–8 days after clopidogrel initiation. Their findings were not confirmed subsequently by other investigators who did not observe any drug-drug interaction between clopidogrel and lipophilic statins. However, the recent ACCEL-STATIN study showed that in clopidogrel-treated patients with high platelet reactivity during chronic coadministration of atorvastatin, switching to a non-CYP3A-metabolized statin resulted in a significant decrease in platelet reactivity and the prevalence of high platelet reactivity. The investigators concluded that their study supported the beneficial effect of replacing atorvastatin therapy with a non-CYP3A-metabolized statin with regard to reducing platelet reactivity in clopidogrel-treated patients with high platelet reactivity. The authors stated that the ACCEL-

![Figure 2. P2Y12 reaction units (Left) and percent inhibition (Right) at the end of the 30-day atorvastatin vs. pitavastatin therapy. *P=0.003 as compared with corresponding value at baseline.](image-url)
Study Limitations

Only the VerifyNow P2Y12 assay was used to evaluate platelet function. Also, we did not look for CYP2C19*2 polymorphism or other polymorphisms (eg, CYP3A4/5, ABCB1) that might be associated with clopidogrel response. Given the pilot nature of this study, it was not designed to evaluate clinical outcomes, which would require larger populations. We did not measure clopidogrel and its active metabolite, similarly to most other studies. The levels of clopidogrel and its active metabolite are, however, likely to be similar either on atorvastatin or pitavastatin as the platelet inhibitory effects were similar. In addition, one should take into account the possibility that our results might have been affected in part by the evolving pattern over time of on-clopidogrel platelet reactivity.

A further limitation is that PRU was measured during chronic DAPT. However, PRU following the loading dose of clopidogrel is more strongly correlated with thrombotic events than during the maintenance dose.

Finally, confounders could interfere with alternative metabolic pathways of pitavastatin, but possible effects of differences in ongoing medications (nitrates and angiotensin-inhibiting drugs) on clopidogrel efficacy are unlikely, as med-

STATIN study results suggested that switching from a CYP3A4-metabolized statin to a non-CYP3A4-metabolized statin enhanced the antiplatelet effect of clopidogrel and overcame high platelet reactivity observed during atorvastatin therapy in some patients. The results of the PORTO trial confirm the previous observations and provide the novel information that pitavastatin constitutes a further option for poor responders to clopidogrel when atorvastatin must be switched.

The bioavailability of lipophilic statins is largely dependent on CYP3A-mediated first-pass metabolism in the intestine and liver. Lipophilic statins, such as atorvastatin, are inhibitors of CYP3A4 and ABCB1 in the intestine and liver. The most important differences between pitavastatin and other statins are related to metabolism. As a result, most of the bioavailable fraction of an oral dose of pitavastatin is excreted unchanged in the bile and is then ready for enterohepatic circulation by reabsorption in the small bowel. Less than 5% of a dose of pitavastatin is excreted in the urine. Thus, pitavastatin has a unique metabolic profile compared with other statins, which contributes to increased bioavailability, longer duration of action and lower probability of drug-food or drug-drug interactions.
The results of the PORTO trial have important clinical implications in real-world practice. The use of pitavastatin, which is associated with more favorable rates of optimal clopidogrel response as compared with atorvastatin, appears to be mandatory for patients showing high platelet reactivity while on DAPT. In addition, pitavastatin may be considered in patients with borderline platelet inhibition with DAPT, in whom the use of CYP3A4 metabolized statin coadministration may further decrease the antiplatelet effect of clopidogrel.

Acknowledgments
The PORTO trial was supported by the Sapienza University of Rome.

Disclosures
There were no external sources of support for this research. No author has any conflict of interest to disclose.

References