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# Impact of different dosing regimens of clopidogrel on systemic oxidative stress in patients undergoing elective percutaneous coronary intervention

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## Letter to the editor

Percutaneous coronary intervention (PCI) directly injures coronary arterial wall, leading to an immediate increase of oxidative stress.[1] Antiplatelet therapy with clopidogrel is of paramount importance to prevent PCI-related thrombotic events. In the current study, we assessed the impact of different modalities of clopidogrel administration on systemic oxidative stress parameters in patients with stable coronary artery disease (CAD) undergoing elective PCI.

## Methods

We prospectively enrolled 28 consecutive patients at Campus Bio-Medico University (Rome) undergoing elective PCI with drug eluting stent implantation. All procedures were performed with informed consent and following all the guidelines for experimental investigation with human participants. All patients were receiving chronic low-dose aspirin (75–100 mg/day). In clopidogrel-naïve patients ($n = 19$), a 600-mg loading dose was administered immediately before the procedure (loading group). Patients on chronic therapy with clopidogrel 75 mg/day ($n = 9$) did not receive any further doses (maintenance group). Venous blood samples were collected at before (T0), and 6 (T1) and 24 (T2) h post-PCI. Oxidative stress was measured by a global evaluation of derivatives of reactive oxygen metabolites (DROM) and biological antioxidant test potential (BAP) using spectrophotometry-based FREE Carpe Diem assay (Diacron International, Grosseto, Italy). The DROM test detects the oxidization of *N*,*N*-diethyl-para-phenylenediamine as a chromogenic substrate by radicals converted from hydroperoxide, estimating the overall pro-oxidant component of oxidative stress, with results expressed as Carratelli unit (CARR U).[2] The BAP test examines the blood concentration of antioxidants as agents that can reduce iron from the ferric ($Fe^{3+}$) to ferrous ($Fe^{2+}$) form, indicating the physiological efficiency of the antioxidant defence systems.[3]

We used repeated measured analysis of variance (ANOVA) to evaluate the tendency of DROM and BAP test in the overall cohort and ANOVA test to assess the differences between the two groups at each time point. For all analyses, a $P$ less than 0.05 was considered statistically significant. All data were processed with SPSS 15.0 software (SPSS Inc., Chicago, Illinois).

## Results

In the overall cohort, DROM tended to increase from T0 ($280 \pm 57.4$ CARR U) to T1 ($301.4 \pm 61.5$ CARR U) and subsequently decrease at T2 ($289.2 \pm 60.1$ CARR U; ANOVA for repeated measures $P = 0.056$), whereas no significant differences in BAP levels were detected at the three time points (T0: $2038.3 \pm 402.9$ μmol/l; T1: $1958.2 \pm 294.1$ μmol/l; T2 $1985.8 \pm 313.2$ μmol/l; ANOVA for repeated measures $P = 0.607$). No significant differences were observed between the loading group and the maintenance group with respect to the clinical and procedural characteristics, including the medical treatment at baseline (Table 1). At T0, DROM values were similar in the two groups ($279.4 \pm 54.7$ in the maintenance group vs. $280.3 \pm 60.1$ CARR U in the loading group; $P = 0.972$). A significant increase in DROM was observed at T1 in the maintenance group ($317.0 \pm 59.6$ CARR U; $P = 0.039$ vs. T0), whereas this was blunted in the loading group ($294.0 \pm 62.6$ CARR U; $P = 0.534$ vs. T0). At T2, DROM values remained elevated also in the maintenance group ($316.8 \pm 68.1$ CARR U; $P = 0.041$ vs. T0), whereas in the loading group, DROM values returned to the baseline levels ($276.2 \pm 52.8$ CARR U; $P = 0.999$ vs. T0) (Fig. 1). Delta DROM, defined as the difference between DROM values at T2 and T0, was $-4.1 \pm 43.5$ CARR U in the loading group and $37.3 \pm 41.5$ CARR U in the maintenance

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**Table 1 Clinical and procedural characteristics**

| | Total group, $n=28$ | Clopidogrel loading dose 600 mg, $n=19$ | Clopidogrel maintenance dose 75 mg, $n=9$ | *P* value |
|---|---|---|---|---|
| Age | 68.7 ± 9.7 | 70.7 ± 8.4 | 64.4 ± 11.2 | 0.112 |
| Male sex | 21 (75) | 14 (74) | 7 (78) | 0.715 |
| BMI | 27.1 ± 3.8 | 27.5 ± 3.3 | 26.3 ± 4.8 | 0.443 |
| Diabetes mellitus type 2 | 13 (46) | 8 (42) | 5 (56) | 0.505 |
| Hypertension | 25 (89) | 18 (95) | 7 (78) | 0.175 |
| Smoke | 6 (21) | 4 (21) | 2 (22) | 0.944 |
| Hypercholesterolemia | 23 (82) | 15 (79) | 8 (89) | 0.521 |
| Family history of CAD | 12 (43) | 8 (42) | 4 (44) | 0.907 |
| Prior ACS | 8 (29) | 5 (26) | 3 (33) | 0.561 |
| Prior PCI | 9 (32) | 3 (16) | 6 (67) | 0.007 |
| Prior CABG | 1 (4) | 1 (5) | 0 (0) | 0.483 |
| Medical treatment at admission | | | | |
| Aspirin | 20 (71) | 14 (74) | 6 (67) | 0.701 |
| Nitroglycerin | 7 (25) | 3 (16) | 4 (44) | 0.102 |
| Beta blocker | 19 (68) | 14 (74) | 5 (56) | 0.337 |
| Calcium channel blockers | 6 (21) | 3 (16) | 3 (33) | 0.291 |
| Diuretic | 11 (39) | 8 (42) | 3 (33) | 0.657 |
| Sartan | 10 (36) | 8 (42) | 2 (22) | 0.305 |
| Ace-inhibitor | 12 (36) | 8 (42) | 4 (44) | 0.907 |
| Statin | 21 (75) | 13 (68) | 8 (89) | 0.243 |
| Multivassel PCI | 6 (21) | 4 (21) | 2 (22) | 0.994 |
| Stent number | 1.5 ± 1 | 1.63 ± 1.01 | 1.1 ± 0.78 | 0.186 |
| Total length of stents (mm) | 27 ± 17 | 30 ± 17 | 20 ± 15 | 0.148 |
| Contrast volume (ml) | 176 ± 56.2 | 180.6 ± 64.3 | 158.9 ± 37.1 | 0.357 |
| Contrast-induced acute kidney injury (AKI)[a] | 0 (0) | 0 (0) | 0 (0) | – |
| Periprocedural myocardial necrosis[b] | 11 (39) | 9 (47) | 2 (22) | 0.20 |
| Troponin I at T0 | 0.02 ± 0.01 | 0.02 ± 0.01 | 0.02 ± 0.01 | 0.86 |
| Troponin I at T2 | 0.66 ± 1.22 | 0.79 ± 0.22 | 0.22 ± 0.26 | 0.25 |
| Creatinine at T0 | 1.00 ± 0.26 | 1.02 ± 0.26 | 0.97 ± 0.26 | 0.63 |
| Creatinine at T2 | 1.01 ± 0.27 | 1 ± 0.25 | 1.06 ± 0.33 | 0.61 |

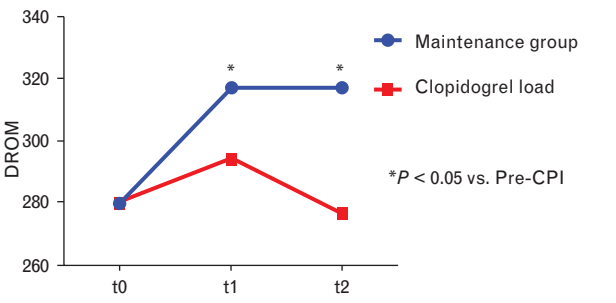
Values as expressed as *n* (%) or mean ± SD. ACS, acute coronary syndrome; AKI, acute kidney injury; CABG, coronary artery bypass grafting; CAD, coronary artery disease; PCI, percutaneous coronary intervention. [a] AKI was defined as at least 50% or at least 0.3 mg/dl increase of serum creatinine in 48 h. [b] Periprocedural myocardial necrosis was defined as Troponin I value more than 5 × 99th percentile.

group, respectively ($P=0.019$). BAP values were not significantly different between patients in the maintenance and loading groups at any study time point (T0: 2114.8 ± 581.7 vs. 2002.1 ± 298.1 μmol/l, $P=0.503$; T1: 2013.7 ± 276.2 vs. 1931.9 ± 305.9 μmol/l, $P=0.505$; T2: 1974.8 ± 289.8 vs. 1990.9 ± 331.3 μmol/l, $P=0.982$).

## Comment

Our data show that, unlike a chronic treatment, the administration of a loading dose of clopidogrel immediately before the procedure attenuates the increase of oxidative stress in patients with stable CAD undergoing PCI. Until now there are no randomized control trials investigating the use of the newer P2Y12 antagonists (prasugrel and ticagrelor) instead of clopidogrel in the setting of patients with stable CAD undergoing elective PCI, and, notwithstanding the more favourable pharmacological profile of prasugrel and ticagrelor, clopidogrel is still the most widely used P2Y12 antagonist. Furthermore, clopidogrel is the recommended P2Y12 antagonist in addition to aspirin in patients with an indication to oral anticoagulants undergoing elective PCI.[4] Clopidogrel may potentially prevent damage from oxidative stress through an inhibition of platelet activation that leads to reduced reactive oxygen species and proinflammatory cytokines production and increased nitric oxide bioavailability, and a production of antioxidant compounds as a result of a non-P2Y12 effect of clopidogrel metabolite which possesses a sulphhydryl (–SH) group that is able to react with sulphur-containing compounds.[5–7] By strengthening these mechanisms, our results suggest the potential benefit of a clopidogrel load and reload (in patients on chronic therapy) prior to PCI, to reduce oxidative stress parameters, although a previous study showed no overall clinical benefit using this strategy.[8] Of note, baseline DROM values were similar in the two groups, suggesting in line with previous evidence[7]

**Fig. 1**




Derivatives of reactive oxygen metabolites levels in the group of patients on chronic clopidogrel therapy and in the group of clopidogrel-naïve patients receiving preprocedural loading dose, at T0, T1 and T2.

that a chronic clopidogrel therapy with 75 mg does not significantly affect oxidative stress. Significantly, at 24 h, in patients who received clopidogrel loading dose, DROM levels return to the baseline differently from those who did not receive the loading dose in which DROM values remained higher. On the contrary, we did not assess oxidative stress beyond 24 h, but we would expect a slower reduction of DROM values to the baseline in patients not receiving clopidogrel loading dose and no significant variations of DROM values over time in patients treated with loading dose and shifted to 75 mg as suggested by the similar DROM values in the two groups at baseline.

In conclusion, PCI seems to induce a transient increase in DROM levels but has no significant effect on BAP. Compared with a chronic clopidogrel therapy, the administration of a 600-mg clopidogrel loading dose immediately before the procedure seems to attenuate the increase in DROM levels. Larger studies are warranted to clarify the effects of different clopidogrel strategies on the periprocedural oxidative balance in patients with stable CAD as well as with acute coronary syndrome undergoing PCI.

We believe that our study limitations were primarily the small sample size and lack of patient randomization, although the prospective design was important for avoiding selection bias. Although DROM and BAP test allow a global evaluation of oxidative stress balance, no other biological markers of oxidative stress were used, only systemic and not local oxidative stress was assessed, and we did not test the platelet reactivity and its possible correlation with the oxidative stress parameters. Finally, we did not evaluate oxidative stress trend beyond 24 h.

## Acknowledgements

### Conflicts of interest

There are no conflicts of interest.

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