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REGULAR ARTICLE

Relationship between changes in platelet reactivity and changes in platelet receptor expression induced by physical exercise

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Abstract

Introduction: In previous studies we have consistently shown a significant increase of platelet reactivity after exercise in patients with obstructive coronary artery disease (CAD). We also observed a significant individual variability in the response to exercise of platelet reactivity in these patients. Whether exercise-induced changes in platelet reactivity correlate with changes in platelet membrane receptors in patients with CAD is unknown.
Methods: We studied 26 patients with stable CAD and 10 matched healthy controls who underwent a symptom-limited treadmill exercise stress test. Venous blood samples were collected at rest and within 5 min of peak exercise. Platelet reactivity was measured by the PFA-100 method as time to occlude (closure time, CT) a ring coated with collagen/adenosine diphosphate (C/ADP). Platelet expression of glycoprotein (GP) IIb/IIIa, in both global (CD41) and active form (PAC-1), and P-selectin (CD62P) and formation of leukocyte–platelet aggregates were assessed by flow cytometry.
Results: After exercise CT did not change in controls (85.4±12 to 84.0±9 s, $p=0.37$), whereas it decreased in CAD patients (98.8±24 to 91.4±25 s, $p<0.001$). After

Abbreviations: CAD, coronary artery disease; PFA, platelet function analyzer; C/ADP, collagen/adenosine diphosphate; CT, closure time; ECG, electrocardiogram; PMN, polymorphonuclear cells; MONO, monocyte; GP, glycoprotein.

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exercise, CD41 and PAC-1 platelet expression increased significantly in CAD patients ($p=0.04$ for both), but not in controls ($p=0.39$ and $p=0.98$, respectively). To evaluate the relationship between the response to exercise of platelet reactivity and of platelet receptor expression, CAD patients were divided into two groups: CAD group 1 (16 patients, decrease in CT >5 s after exercise) and CAD group 2 (10 patients no increase in platelet reactivity after exercise). CD41 and PAC-1 expression increased in CAD group 1 ($p=0.008$ and $p=0.026$, respectively) but not in CAD group 2 ($p=0.39$ and $p=0.50$, respectively). No significant differences were observed between the 2 groups for changes in CD62P and leukocyte–platelet aggregates.
Conclusions: Our data show that, in patients with stable CAD, an increased platelet reactivity to C/ADP stimulation after exercise, as assessed by the PFA-100 method, is specifically associated with an increased expression of platelet GP IIb/IIIa receptor.

Introduction

Platelets are involved both in the mechanisms of atherogenesis [1] and in the thrombotic complications of atherosclerosis [2]. Several previous studies assessed the effects of lifestyle factors on platelet function [3–5]. The effects of exercise, in particular, have been extensively studied, but the results are contentious [4,6–11]. The reasons for that are likely multiple and include differences in patient selection, drug therapy and methods employed to assess study platelet function. Furthermore, techniques used to assess platelet reactivity, either in vitro or in vivo, are often associated with considerable methodologic difficulties, which might also account, at least in part, for the discrepancies reported in previous studies [12].

Recently, a new simple method has been proposed to measure platelet reactivity, the platelet function analyser (PFA)-100. By this method, platelet reactivity is measured as the time needed to occlude a ring in a cartridge coated with either collagen and adenosine diphosphate (C/ADP) or collagen and norepinephrine [13,14].

Using the PFA-100 method, in previous studies we have consistently shown a significant increase of platelet reactivity in response to C/ADP after exercise in patients with obstructive coronary artery disease (CAD) [9–11]. However, we also observed a significant individual variability in the response to exercise of platelet reactivity in these patients [10]. It is not known at present whether these individual differences are or not related to differences in changes of membrane platelet receptors in response to exercise.

To address this issue, in this study we compared the exercise-induced changes of platelet reactivity (as assessed by the PFA-100 method) with the changes of platelet surface receptors and leukocyte–platelet aggregates, as assessed by flow cytometry, in a group of CAD patients.

Methods

Study groups

We studied 26 patients (65±9 years, 20 men) with clinically stable obstructive CAD, documented at coronary angiography (>50% stenosis in ≥1 major epicardial coronary artery). All patients were studied while taking their usual medications. Patients receiving anticoagulant drugs or antiplatelet agents other than aspirin were excluded from the study. Patients taking aspirin were not excluded as previous reports showed that the platelet response to exercise, as assessed by the methods used in the present study (see below), is not significantly influenced by aspirin treatment [15,16].

Ten subjects without any evidence of CAD (60±3 years, 4 men) were studied as a control group. These subjects were enrolled from the non-medical staff of our hospital and were selected to be comparable to CAD patients as to age and gender. Their clinical history excluded any potential cardiac symptom and all had normal standard 12 leads electrocardiogram (ECG), exercise stress test and two-dimensional and Doppler echocardiography. The study complies with the Declaration of Helsinki and was approved by our institutional review board; all subjects gave their written informed consent to participate in the study.

Exercise stress test

After 30 min of rest in a sitting position, a symptom/sign-limited exercise stress test was performed according to a standard Bruce protocol. Leads II, V_2 and V_5 were monitored continuously. A 12-lead ECG was printed at the end of each stage or when clinically indicated, and at 1-minute intervals in the recovery phase. Blood pressure was measured at