Exercise-induced and nitroglycerin-induced myocardial preconditioning improves hemodynamics in patients with angina

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Crisafulli, Antonio, Franco Melis, Filippo Tocco, Uberto M. Santoboni, Carlo Lai, Giovanni Angioy, Luigi Lorrai, Gianluigi Pittau, Alberto Concu, and Pasquale Pagliaro. Exercise-induced and nitroglycerin-induced myocardial preconditioning improves hemodynamics in patients with angina. Am J Physiol Heart Circ Physiol 287: H235–H242, 2004. First published March 11, 2004; 10.1152/ajpheart.00989.2003.—In humans, regional myocardial dysfunction during ischemia may be improved by ischemic and pharmacological preconditioning. We assessed the possibility that exercise- and nitroglycerin-induced myocardial preconditioning may improve global cardiac performance during subsequent efforts in patients with angina. Ten patients suffering from chronic stable angina and ten healthy volunteers were studied. Through impedance cardiography we assessed hemodynamics during a maximal exercise test, which was used as a baseline (Bas test) and considered as a preconditioning exercise. The Bas test was followed by a sequence of maximal efforts performed during the first (FWOP; 30 min after the Bas test) and second (SWOP; 48 h after the Bas test) windows of protection conferred by ischemic preconditioning. Hemodynamics was further evaluated during maximal exercise performed 48 h later with pharmacologically induced SWOP (PI-SWOP) obtained by transdermal administration of 10 mg of nitroglycerin. In the angina patients, FWOP, SWOP, and PI-SWOP delayed the time to ischemia and allowed them to achieve higher workloads compared with the Bas test. Furthermore, heart rate and cardiac output at peak exercise were enhanced during all the preconditioning phases with respect to the Bas test. However, only SWOP and PI-SWOP increased myocardial contractility and stroke volume. No changes in hemodynamics were detectable in the control subjects. This study demonstrates that in patients with stable angina, although hemodynamics during exercise can be positively improved during both FWOP and SWOP, differences exist between these two phases. Furthermore, the mimicking of exercise-induced SWOP by PI-SWOP with transdermal nitroglycerin may represent an important clinical aspect.

Effect is controversial in FWOP (5, 8, 34). The increased synthesis and activity of several enzymes is considered one of the major mechanisms that contribute to the mediation of late preconditioning (5, 8, 34, 47). The expression of many of these enzymes may also be increased by exercise (16) and NO donor pretreatment (16, 21).

Evidence for preconditioning in humans derives from in vitro studies (39) and studies performed during coronary angioplasty, surgery, or exercise (14, 24, 46). In particular, the warm-up phenomenon (i.e., enhanced resistance to further ischemia in patients suffering from angina a few minutes after a previous effort) has also been attributed to a preconditioning-like effect, mainly because the time course of the warm-up is consistent with the protection afforded by FWOP (42). In addition, it was recently demonstrated in patients with stable angina that exercise induces an increase in time to ST segment depression during a subsequent effort in the FWOP and SWOP periods after exercise-induced ischemia (23, 28). Studies have also demonstrated that intravenous or transdermal nitroglycerin may induce SWOP-like cardioprotective effects in humans and animals (1, 21, 25). In particular, it appears to be certain that regional myocardial dysfunction occurring during ischemia is reduced during both FWOP (i.e., a few minutes after a previous angioplasty; Ref. 26), and pharmacological SWOP (i.e., 48 h after intravenous nitroglycerin; Ref. 25). Whether or not this improvement of regional function results in an improvement of global cardiac performance remains to be elucidated.

If the protection observed in the ischemic myocardium afforded by IP and pharmacological preconditioning were also applicable to exercise-induced preconditioning, an improvement in global left ventricular function during exercise in FWOP and SWOP would be predicted. Alternatively, because of the mismatch between oxygen supply and demand occurring during exercise resembling the stunning condition (i.e., oxygenated blood is supplied to an ischemic myocardium), it may be hypothesized that during exercise-induced ischemia the improvement in global cardiac performance may occur to an important extent only during SWOP, as is the case in real myocardial stunning (5, 8, 34).

Therefore, this study was designed to examine in patients with stable angina whether exercise-induced preconditioning improves global cardiac performance in a similar way during exercise performed in the FWOP and SWOP periods. Moreover, we aimed to determined whether nitroglycerin-induced

ischemia; cardiac output; myocardial contractility; nitroglycerin

SHORT EPISODES of nonlethal ischemia render the myocardium more resistant to subsequent ischemic insults. This phenomenon, known as ischemic preconditioning (IP) (31), is characterized by a biphasic pattern: a first window of protection (FWOP), which is active immediately and lasts for 2–3 h, and a second window of protection (SWOP), which starts 12–24 h later and lasts until 72–90 h after the initial ischemia (delayed preconditioning). FWOP, also called early preconditioning, is more potent than SWOP in reducing infarct size. However, although SWOP always attenuates myocardial stunning, this

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SWOP may mimic exercise-induced SWOP. For comparative purposes we also evaluated cardiac performance in normal sedentary subjects before and after exercise- and nitroglycerin-induced preconditioning.

METHODS

Study Population

Two groups of subjects were studied. The angina subject group (Ang group) consisted of 10 subjects with chronic stable angina diagnosed on the basis of symptoms and ST segment depression >0.1 mV during a previous exercise test and with isolated left anterior descending coronary insufficiency based on >70% diameter stenosis on coronary angiography. We selected patients with a single location of ischemia to limit the influences on global function related to the site of ischemia. Exclusion criteria were occurrence of significant stenosis on the right and/or left circumflex coronary arteries, involvement in exercise-training programs, acute myocardial infarction or unstable angina during the preceding 3 mo, aortic stenosis, heart failure, severe hypertension, valvular insufficiency, diabetes mellitus, and resting ST segment changes on the ECG. All subjects were male, and six had a history of myocardial infarction. No one was physically active. The demographic characteristics of the Ang group are shown in Table 1.

Although it is unlikely that one or two exercise tests induce a generic training effect in patients with angina, to exclude this effect and to verify whether exercise-induced ischemia (ST depression) is necessary to obtain beneficial preconditioning effects, we chose as the control group (Con group) 10 healthy male subjects with a sedentary lifestyle and not involved in any physical training program. All Con group subjects were free from cardiac disease as determined by anamnesis, physical examination, and normal resting ECG. The demographic characteristics are shown in Table 1.

Each subject gave written informed consent to take part in the study, which was approved by the local ethical committee and conforms to the principles of the Declaration of Helsinki.

Experimental Protocol

All experiments were carried out in a temperature-controlled air-conditioned room. Each subject underwent the following study protocol.

Exercise-induced preconditioning. All subjects underwent a maximal exercise test performed with an electromagnetically braked cycle ergometer (Tunturi EL 400). The test consisted of a 10 W/min incremental exercise starting from 10 W at a pedaling frequency of 60 rpm. Criteria for terminating the test were physical exhaustion, attainment of maximal age-related heart rate (HR), electrocardiogram ST segment depression >2.0 mV, severe chest pain, and severe dysrhythmias for the Ang group and physical exhaustion and/or attainment of maximal age-related HR for the Con group. This test was considered as the preconditioning episode and as the baseline test (Bas test). Both groups were then involved in a second maximal effort 30 min later (FWOP test). A third maximal exercise test (SWOP test) was performed 48 h after the Bas test.

Pharmacologically induced SWOP. Forty-eight hours after an overnight (8 h) transdermal administration of 10 mg of nitroglycerin, all subjects performed a maximal cycle-ergometer test identical in end point criteria to the Bas test. All exercise tests were identical in end point criteria to the Bas test. Exercise-induced preconditioning tests and the pharmacologically induced SWOP (PI-SWOP) test were assigned in a random order and were separated from each other by 1 wk.

Patients were requested to avoid heavy efforts that could induce angina or require the administration of nitrates. All medications were continued except for nitrates, which were discontinued 1 wk before the subject entered the study protocol.

The level of ST segment depression was measured 0.08 s after the J point by means of an ECG computer with signal-averaging capacity (Marquette Cardiosoft 4.1, Freiburg, Germany). The lead with the greatest ST segment depression was used for subsequent analysis. ECG traces were checked in a blinded fashion by an investigator to ensure that the computer was not influenced by wandering baseline, development of bundle branch block, or ectopic beats.

Hemodynamics Assessment

Hemodynamics was assessed with an impedance cardiograph (NCCOM 3; BoMed, Irvine, CA) by an experienced operator. The impedance method is commonly utilized to measure cardiodynamics during exercise either in normal subjects or in patients (11, 12, 19) as well as in ischemic disease (2, 32, 41). This technique assumes that when an electrical current circulates through the thorax, the pulsatile aortic blood flow causes a proportional fluctuation in electrical conductivity. As a consequence, changes in thoracic electrical impedance during systole are representative of stroke volume (SV) (44). Several formulas have been developed for the electrical estimation of SV. All these formulas are based on the measurement of several reference points on the impedance waveforms (3, 11, 44). Therefore, through impedance and ECG traces we assessed the following parameters: SV, which was calculated with the Sramek-Bernstein equation (3); HR calculated as the reciprocal of the R-R interval; cardiac output (CO), obtained by multiplying SV × HR; and myocardial contractility, considered to be inversely related to the prejunction period-to-ventricular ejection time ratio (PEP/VET). This ratio correlates with the angiographic ejection fraction very well and represents an inverse index of global ventricular performance (17, 27). Subjects were also connected to a manual spaghmomanometer for systolic (SBP) and diastolic (DBP) blood pressure assessment. To calculate mean blood pressure (MBP) we used the method previously described by Moran and coworkers (30), which allows calculation of MBP by taking into account changes in the diastolic and systolic periods caused by exercise tachycardia. Systemic vascular resistances (SVR) were obtained by multiplying the MBP-to-CO ratio by 80, where 80 is a conversion factor to change units to standard resistance units. Double product (DP) was calculated as HR × SBP.

Statistical Analysis

Statistical analysis was performed with commercially available software (SigmaStat 2.03). Data were averaged for 1 min. Two sets of comparisons were made, one between resting data and the other between peak exercise changes (i.e., values of hemodynamic variables reached during the last minute of exercises) in response to preconditioning maneuvers. To compare cardiodynamic responses we reported data as mean ± SE percent changes from rest and peak exercise values of the Bas test. Comparisons were performed with two-way
ANCOVA for repeated measures (factors: group and condition). Post hoc comparison between groups at an individual time point was performed with Student’s $t$-test for unpaired data. Contrasts between conditions in the same group were analyzed with the Tukey post hoc test. Statistical significance was set at a $P$ value of $<0.05$ in all cases.

**RESULTS**

Table 1 shows that the groups enrolled in the study were comparable in age, weight, and height. Table 2 summarizes absolute values of hemodynamic data at rest and peak exercise during the Bas test.

All resting data were collected just before the various exercise tests and are shown in Table 3 as percent changes from resting data of the Bas test, which were considered as 100%. Because no differences were found between groups or conditions, it can be inferred that both exercise-induced and pharmacologically induced preconditioning did not affect hemodynamics at rest.

Figure 1A shows ST segment changes at peak exercise. As expected, there was a marked difference in ST behavior between groups. Specifically, there was no ST depression in the Con group. In contrast, exercise produced a significant depression in the ST segment in the Ang group subjects. In particular,

Table 2. Absolute values of hemodynamic data at rest and at peak exercise during the baseline test

<table>
<thead>
<tr>
<th></th>
<th>Rest</th>
<th>Peak Exercise</th>
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<tbody>
<tr>
<td><strong>HR, beats/min</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ang</td>
<td>76.1±3.7</td>
<td>117.5±4.6</td>
</tr>
<tr>
<td>Con</td>
<td>83±3.8</td>
<td>143±7.2</td>
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<tr>
<td><strong>SV, ml</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ang</td>
<td>59.7±6.7</td>
<td>82.7±7.3</td>
</tr>
<tr>
<td>Con</td>
<td>58.8±2.3</td>
<td>99.1±6.4</td>
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<tr>
<td><strong>CO, l/min</strong></td>
<td></td>
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<tr>
<td>Ang</td>
<td>4.5±0.5</td>
<td>9.6±0.8</td>
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<tr>
<td>Con</td>
<td>4.9±0.3</td>
<td>14.4±1.4</td>
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<tr>
<td><strong>PEP/VET</strong></td>
<td></td>
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<tr>
<td>Ang</td>
<td>0.52±0.03</td>
<td>0.44±0.03</td>
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<tr>
<td>Con</td>
<td>0.54±0.02</td>
<td>0.35±0.03</td>
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<tr>
<td><strong>SBP, mmHg</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ang</td>
<td>123.9±3.1</td>
<td>170±5.4</td>
</tr>
<tr>
<td>Con</td>
<td>118.5±5.1</td>
<td>191±8.3</td>
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<tr>
<td><strong>DBP, mmHg</strong></td>
<td></td>
<td></td>
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<tr>
<td>Ang</td>
<td>77.2±3.8</td>
<td>84.4±5</td>
</tr>
<tr>
<td>Con</td>
<td>72.5±2.8</td>
<td>84±5.2</td>
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<tr>
<td><strong>MBP, mmHg</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ang</td>
<td>91.2±2.7</td>
<td>116±4.5</td>
</tr>
<tr>
<td>Con</td>
<td>87±2.9</td>
<td>131±3.5</td>
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<tr>
<td><strong>SVR, dyn-s·cm$^{-5}$</strong></td>
<td></td>
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</tr>
<tr>
<td>Ang</td>
<td>1,861.7±245.7</td>
<td>1,037.2±103.5</td>
</tr>
<tr>
<td>Con</td>
<td>1,476.2±117.6</td>
<td>791±77.8</td>
</tr>
<tr>
<td><strong>DP, mmHg·beat·min$^{-1}$</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ang</td>
<td>9,376.2±377.6</td>
<td>2,008.2±1068.3</td>
</tr>
<tr>
<td>Con</td>
<td>9,867.2±543.8</td>
<td>27,434.6±1813.2</td>
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<tr>
<td><strong>Workload, W</strong></td>
<td></td>
<td></td>
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<tr>
<td>Ang</td>
<td>116.5±5.9</td>
<td>148±9</td>
</tr>
<tr>
<td>Con</td>
<td>116.5±5.9</td>
<td>148±9</td>
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<tr>
<td><strong>Total exercise duration, min</strong></td>
<td></td>
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</tr>
<tr>
<td>Ang</td>
<td>11.6±0.59</td>
<td>14.8±0.9</td>
</tr>
<tr>
<td>Con</td>
<td>11.6±0.59</td>
<td>14.8±0.9</td>
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</tbody>
</table>

Values are means ± SE. HR, heart rate; SV, stroke volume; CO, cardiac output; PEP/VET, prejection period-to-ventricular ejection time ratio; SBP, systolic blood pressure; DBP, diastolic blood pressure; MBP, mean blood pressure; SVR, systemic vascular resistance; DP, double product.

all patients reached at least 0.15 mV of depression at peak exercise in all protocol tests. There were no differences in the level of ST segment depression in protocol sessions for the Ang group. However, as can be seen in Fig. 1B (time course of ST segment depression as a function of exercise time in the Ang group), FWOP, SWOP, and PI-SWOP delayed the time to ischemia with respect to the Bas test. This allowed Ang group subjects to exercise longer (Fig. 1C) before experiencing angina and to achieve higher workload values at peak exercise (Wpeak; Fig. 1D) after the preconditioning maneuvers compared with baseline.

**Hemodynamics**

Figures 2–4 exhibit percent changes in hemodynamic parameters reached at peak exercise during FWOP, SWOP, and PI-SWOP tests with respect to the peak exercise of the Bas test (100%).

**Con group.** After all preconditioning maneuvers, the Con group showed values not different from the Bas test for all hemodynamic variables considered (Figs. 2–4).

**Ang group.** In this group, after all preconditioning maneuvers HR at peak exercise (HRpeak; Fig. 2A) was similarly and significantly enhanced, being +8–9% of the Bas test values.

Exercise-induced and pharmacologically induced delayed preconditioning caused an increase in SV at peak exercise (SVpeak) of 25 ± 6% and 24 ± 5%, respectively, with respect to the Bas test (Fig. 2B). However, the increase in SVpeak (15 ± 4%) was not statistically significant during FWOP.
As a consequence of HR and SV behavior in this group, CO at peak exercise (CO \(_{\text{peak}}\); Fig. 2C) was enhanced during FWOP (+24 \( \pm \) 5%), SWOP (+34 \( \pm \) 6%), and PI-SWOP (+33 \( \pm \) 6%) compared with the Bas test. It is noteworthy that during SWOP the increase in CO was due to a significant increase in HR and SV, whereas during FWOP the increase in SV did not reach statistical significance.

Moreover, during SWOP, statistical analysis revealed significant differences in terms of PEP/VET (Fig. 2D) at peak exercise (PEP/VET \(_{\text{peak}}\)). In fact, PEP/VET \(_{\text{peak}}\) was reduced by 11 \( \pm \) 2% and 13 \( \pm \) 2% during SWOP and PI-SWOP, respectively, with respect to the Bas test. Because this parameter is inversely related to myocardial contractility, this reduction meant that contractility was enhanced at these settings. On the contrary, the modest reduction (7 \( \pm \) 4%) in the inverse index of contractility, PEP/VET, was not statistically significant during FWOP.

SBP (SBP \(_{\text{peak}}\)), DBP (DBP \(_{\text{peak}}\)), and MBP (MBP \(_{\text{peak}}\)) at peak exercise did not show any variation in response to preconditioning maneuvers compared with the Bas test (Fig. 3, A–C, respectively). Figure 3D shows that SVR at peak exercise (SVR \(_{\text{peak}}\)) were reduced during FWOP (−15 \( \pm \) 3), SWOP (−20 \( \pm \) 4), and PI-SWOP (−21 \( \pm \) 4) with respect to the Bas test. Finally, DP at peak exercise (DP \(_{\text{peak}}\)) was not influenced by preconditioning maneuvers (Fig. 4).
DISCUSSION

This is the first clinical study that demonstrates that exercise-induced and pharmacologically induced preconditioning can positively affect hemodynamics during subsequent exercise in patients suffering from stable angina. In fact, we found that HR and CO at peak exercise were improved during the FWOP and SWOP periods after exercise-induced ischemia, without any significant changes in blood pressure and DP. Importantly, SWOP also induced a significant improvement in SV and myocardial contractility. Similarly, the transdermal administration of nitroglycerin was effective in mimicking the positive effects of exercise-induced delayed protection. Because this global improvement in hemodynamics was not present in the Con group, it was not due to a generic training effect.

During FWOP the increase in HR is likely due to a difference in exercise duration, i.e., the possibility of exercising longer without ischemic symptoms. In fact, together with the improvement in hemodynamic parameters, we also found in the Ang group blunted signs of myocardial ischemia (delay in ST segment depression) as well as higher exercise capacity (increased W_peak). These observations are consistent with the warm-up concept in angina (42).

During SWOP we found a similar improvement in these parameters (HR, ST, and W_peak), independent of whether SWOP was induced by exercise or nitroglycerin. This is in accordance with the recent findings of Lambiase and coworkers (23), who demonstrated an increase in time to ST depression in exercise-induced SWOP in humans. The major difference between our study and that of Lambiase et al. (23) is that we analyzed global performance in the same subjects in which we measured ECG. To the best of our knowledge, our study is the first to document an improvement in both ischemic signs and hemodynamics during both exercise- and nitroglycerin-induced preconditioning.

Importantly, the data reported here represent the first evidence that hemodynamic responses during exercise are different in FWOP and SWOP. In fact, a significant improvement in global myocardial contractility (indexed by PEP/VET) and SV were observed in SWOP but not in FWOP. It is therefore likely that during exercise in the Ang group the ischemia-induced regional dysfunction was lowered importantly only in the SWOP period. This difference is at variance with previous studies in which coronary angioplasty was used as the setting for studying preconditioning. In these studies, regional dysfunction during ischemia was reduced during both FWOP and SWOP (25, 26). We can argue that during FWOP the regional improvement is not strong enough to result in a significant improvement in global function. An alternative explanation for these differences may reside in the protocol used to induce preconditioning and to detect protection (angioplasty vs. exercise-induced ischemia). It is noteworthy that the ischemic dysfunction observed during a mismatch between oxygen supply and demand (e.g., during exercise) is quite different from that observed during coronary occlusion (e.g., during angioplasty). In fact, during exercise, oxygen supply, although insufficient, increases and reactive oxide species (ROS) formation may increase (22); differently, ROS do not increase during coronary occlusion. On the basis of evidence that during exercise an improved contractility occurs during SWOP but not during FWOP (similar to what is seen in animal studies when stunning is considered as the end point) (6, 13, 21, 37), we can

Fig. 3. Percent changes with respect to Bas test of hemodynamic data at peak exercise. A: systolic blood pressure (SBP). B: diastolic blood pressure (DBP). C: mean blood pressure (MBP). D: systemic vascular resistance (SVR). Values are means ± SE. *P < 0.05 vs. Con group; †P < 0.05 vs. Bas test.

Fig. 4. Percent changes with respect to Bas test of double product (DP) at peak exercise. Values are means ± SE.
speculate that our findings of improved contractility during exercise may be correlated with the mechanisms involved in protection against stunning (i.e., reduced formation of ROS and/or increased endogenous antioxidant defense). This hypothesis is supported by the recent findings of Michaelides and coworkers (28) showing that the beneficial effect (delayed time to onset of angina and ST depression) on exercise-induced ischemia observed the day after a first effort can be attributed to increased extracellular superoxide dismutase activity. It is noteworthy that an upregulation of this enzyme can be triggered by IP (5, 8, 34, 47), exercise (16, 28), and NO donor pretreatment (16, 21).

The findings that pharmacologically induced and exercise-induced SWOP similarly improve contractility support each other and are in line with the idea that solely in this phase may an improved cardiac performance be importantly appreciated during exercise, as is the case in myocardial stunning (5, 8, 34). Moreover, the fact that in experimental animals NO plays a pivotal role in both the triggering and mediation phases of delayed protection (1, 5) is in agreement with our findings that nitrates are able to mimic the effects of exercise-induced delayed protection in terms of tolerance to ischemia and cardiac performance in subjects with angina.

Our study does not reveal any effect of exercise-induced and pharmacologically induced preconditioning on blood pressure parameters, which did not show any difference in behavior during all the protocol sessions. This result may find an explanation in the fact that, although the CO\textsubscript{peak} increased, the SVR\textsubscript{peak} reduced in all protocol sessions in the Ang group. This observation of SVR reduction is intriguing and deserves further study to ascertain whether it may be attributable to the higher workloads achieved, to the consequent accumulation of end products of muscle metabolism, and/or to an enhanced vasodilation achieved via an improved endothelial function. Indeed, endothelium-dependent vasodilatation can be improved by the early and delayed phases of IP (6, 18, 21, 34) as well as by NO donor pretreatment (16, 21).

Finally, the DP\textsubscript{peak} was unaffected by the preconditioning protocol. This result suggests that myocardial oxygen consumption was not increased by FWOP, SWOP, or PI-SWOP. This finding is consistent with the observations of Okazaky and coworkers (33), who demonstrated reduced myocardial oxygen consumption (i.e., enhanced heart efficiency) during warm-up.

**Methodological Considerations**

Studies performed in normal rats (45) and dogs (15) show that exercise induces early and late myocardial preconditioning against infarction and that ischemia is not required to trigger protection. The fact that healthy subjects received no benefit from nitroglycerin pretreatment and from exercise-induced FWOP and SWOP does not exclude their hearts being preconditioned but supports the concept that ischemia must occur in the protection periods for any beneficial effect of preconditioning to be detected. Thus it is likely that the differences observed in clinical and experimental studies are attributable to the different end points used during the protection phase. In fact, the studies on animals use infarct size as the end point to demonstrate the occurrence of protection. For ethical reasons, no clinical study can meet this condition. Hence, end points appropriate to human studies are used, such as ST ischemic changes and chest pain (23, 25, 28) as well as myocardial ischemic dysfunction (25, 26) and hemodynamics (present study). These end points indirectly reflect the degree of ischemia, and this may be a limitation on studies dealing with preconditioning in humans. However, it is our opinion, as well as that of other authors (20, 25, 26), that in the clinical scenario of ischemic disease use of multiple surrogate end points is the best approach. In fact, the sole changes in ST segment shift are not predictive of changes in tolerance to ischemia and in heart performance (26). Hence, in our study, the contemporary findings of delayed ischemic signs and of improved cardiac performance support each other in demonstrating the possibility of cardiac preconditioning in angina patients.

The Fick and dye-dilution methods are considered the “gold standard” for hemodynamic assessment (43). Inasmuch as these methods are invasive, their use is not permitted in studies involving healthy subjects or patients who do not require invasive procedures for diagnostic or therapeutic purposes. Among noninvasive methods the choice is restricted to rebreathing, Doppler echocardiography, and impedance cardiography. However, none of these methods has yet been unani-

**Clinical Implications**

Our study extends the clinical usefulness of nitrate administration by including the possibility that this drug, which is one of the most widely utilized therapeutic tools, also elicits late preconditioning against myocardial dysfunction, thus improving cardiac performance during exercise in ischemic patients. This study provides the first evidence that transdermal nitroglycerin can put the human heart in a preconditioned state. Transdermal application of this drug also elicited late preconditioning-like protection in rabbits (21). Differently from most agents that elicit late preconditioning in humans and animals, this does not require intravenous infusion, it is not toxic, and it is easily applicable. Such a property may be as important as, or possibly even more important than, the reported preconditioning action of intravenous nitroglycerin (25) or adenosine (26) infusion. Finally, we stress once again that nitroglycerin is a multifaceted drug and that care must be taken when it is used in a clinical scenario. Moreover, although the present study and other studies (see, e.g., Ref. 25) suggest the potential useful-
ness of the application of acute nitrates in inducing protection, it has not been demonstrated that the prolonged administration of such drugs can afford similar protection.

In conclusion, the results of the present study show that hemodynamics during exercise can be positively affected by early and delayed phases of exercise-induced IP in patients with stable angina. In fact, at peak exercise during both protecting phases, HR and CO were significantly enhanced with respect to nonpreconditioned effort. However, myocardial contractility and SV increased only during delayed preconditioning, thus suggesting a substantial anti-ischemic effect in this phase. Furthermore, the pharmacological preconditioning afforded by transdermal administration of nitrates in patients with stable angina effectively mimicked the hemodynamic effects of exercise-induced delayed preconditioning.

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