OPC-28326, a selective peripheral vasodilator with angiogenic activity, mitigates postinfarction cardiac remodeling

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A LARGE MYOCARDIAL INFARCTION (MI) causes severe chronic heart failure with adverse remodeling of the left ventricle characterized by cavity dilatation and diminished cardiac performance (30). Two critical determinants of subsequent heart failure are the magnitude of the acute MI, which can be determined within several hours after an attack (32), and recanalization of the infarct-related artery, which, if performed early enough for myocardial salvage, reduces the size of the acute infarct, prevents subsequent heart failure, and improves prognosis (39). In addition, late reperfusion, beyond the window for myocardial salvage, also appears to reduce left ventricular (LV) remodeling and decrease mortality (16, 22). This finding serves as the basis for the “open artery hypothesis,” a proposal supported by much experimental and clinical evidence. A number of possible mechanisms by which an open infarct-related artery could confer benefit in ways other than by salvaging ischemic myocardium have been proposed, e.g., improving healing, scaffolding by blood flow or hemorrhage, awakening hibernating myocardium at the border zone, and improving collagen turnover and exertion antiapoptotic effects on salvaged cardiomyocytes (1, 11, 17, 29, 31, 36). In addition, we reported that in a rat MI model late reperfusion altered infarct tissue geometry, i.e., it made the infarcted wall thicker by promoting cell proliferation and reducing apoptosis among noncardiomyocytes in the infarct tissue, thereby reducing wall stress and mitigating LV remodeling and dysfunction (18, 24).

OPC-28326, 4-[(N-methyl-2-phenylethylamino)-1-(3,5-dimethyl-4-propionyl-amino)benzoyl] piperidine hydrochloride monohydrate, is a vasodilator that selectively increases peripheral blood flow in anesthetized open-chest dogs with minimal effect on other hemodynamic parameters (27). OPC-28326 acts as an α2-adrenergic antagonist, showing the highest affinity for the α2C-adrenoceptor subtype (28, 34). In addition, an earlier study reported that in an ischemic leg model OPC-28326 also exerted angiogenic effects via the Akt/endothelial nitric oxide synthase (eNOS) pathway (33). These properties of OPC-28326, vasodilatation and angiogenesis, are considered particularly effective for augmenting blood perfusion in ischemic tissue. We therefore hypothesized that treatment with OPC-28326 could bring about a late reperfusion-like effect in the infarcted heart, thereby mitigating postinfarction cardiac remodeling and dysfunction. To test that idea, in the present study we examined the effects of OPC-28326 on postinfarction survival and LV remodeling and function in a murine MI model, focusing on the architectural dynamics and blood perfusion of the infarct tissue.

MATERIALS AND METHODS

Animal experimental protocols. This study conforms to the Guide for the Care and Use of Laboratory Animals published by US National Institutes of Health (NIH Publication, 8th Edition, 2011) and was approved by the Institutional Animal Research Committee of Gifu University. Mice were initially anesthetized with 2% halothane in a mixture of N2O and O2 (0.5 l/min each) via a nasal mask and then
intubated with a 20-G intravenous catheter and ventilated with 0.5% halothane in a mixture of N2O (0.1 l/min) and O2 (0.5 l/min), using a rodent ventilator. MI was induced in 40 male C3H/He mice (8–10 wk of age; SLC Japan) by ligation of the left coronary artery as previously described (14, 21, 26). Not unlike humans, C3H/He mice showed a recovery of ischemic leg perfusion of ~50% after femoral artery ligation in the previous study (35). In contrast, the often used C57BL/6 mice are capable of entirely restoring blood flow in the ischemic limb between 2 and 4 wk after femoral artery ligation (6, 27), severely limiting the detection of long-term treatment effects of reagents. We thus used C3H/He mice showing a closer resemblance to humans in reaction against ischemic insult rather than C57BL/6 mice. Thirty-six mice were alive after surgery; the survival rate was 90%. After awakening from anesthesia, 36 mice were randomly assigned to the following 2 groups (n = 18 in each): a control group fed a normal diet and an OPC group fed a diet containing 0.05% OPC-28326. At this dose, according to our preliminary study, plasma concentrations were 20.24 ng/ml, respectively (4 each).

Physiological study. Echocardiography and cardiac catheterization were carried out before death, as described previously (15). Animals were anesthetized with an intraperitoneal injection of pentobarbital (30 mg/kg) once every examination. We assessed the adequacy of anesthesia by monitoring echocardiograms and respiratory rate. This relatively small dose of pentobarbital did not affect the level of blood pressure, heart rate, or the state of respiration in mice with any treatment. Echocardiograms were recorded using an echocardiographic system (Vevo770; Visualsonics) equipped with a 45-MHz imaging transducer. Following echocardiography, the right carotid artery was cannulated with a micromanometer-tipped catheter (SPR 671; Millar Instrument) that was advanced via the aorta into the left ventricle to record pressure and maximal and minimal dP/dt (±dP/dt), which are the most reliable parameters of LV systolic and diastolic function, respectively. LV diastolic average wall stress (LVWS) was calculated as described previously (4): LVWS = (LVEDP × Rm × 13.33)/(2 × LVWT), where LVEDP is the LV end-diastolic pressure (mmHg), LVWT = [(septum thickness + (3 × free wall thickness))/4, and Rm = chamber radius + (0.5 × LVWT).

Histological analysis. After the physiological studies, all surviving mice were euthanized by cervical dislocation following pentobarbital anesthesia (200 mg/kg ip) and their hearts were removed. The excised hearts were cut into two transverse slices at the midventricular level. The basal specimens were fixed in 10% buffered formalin and embedded in paraffin, after which 4-μm-thick sections were stained with hematoxylin-eosin and Masson’s trichrome or used for immunohistochemistry.

Immunohistochemistry. Deparaffinized 4-μm-thick sections were incubated with a primary antibody against Fk-1 (a vascular growth factor, 1:100 dilution; Santa Cruz Biotechnology), α-smooth muscle actin (α-SMA; a general cell proliferation marker, 1:100 dilution; Dako), or Ki-67 (a differentiation marker of smooth muscle cell, 1:25 dilution; Dako), after which they were immunostained using an ABC kit (Vector) with the chromogen diaminobenzidine HCI. Nuclei were stained with hematoxylin. Apoptosis was evaluated based on terminal deoxynucleotidyl transferase-mediated in situ nick-end labeling (TUNEL) using an ApopTag kit (Chemicon) according to the supplier’s instructions. Mouse mammary tissue served as a positive control.

To evaluate proliferation and apoptosis among nonmyocytes in the infarct tissue, we used double immunostaining in combination with TUNEL. Tissue sections were first incubated with anti-Ki-67 antibody followed by Alexa 488 or Fluorescein-FragEL (Oncogene) and then with anti-Fk-1 or anti-α-SMA antibody followed by Alexa 568. Nuclei were stained with Hoechst 33342. Immunofluorescent preparations were observed under a confocal microscope (LSM510; Zeiss).

Quantitative assessments of cardiomyocyte size (measured as the transverse diameter), fibrotic area, noncardiomyocyte population, vessel population (capillaries and small arteries), percent area of extravascular α-SMA-positive area, percent Ki-67-positive cells, and percent TUNEL-positive cells were performed on 20 randomly chosen high-power fields (>400) in each section using a multipurpose color image processor (BZ Analyzer; KEYENCE, Osaka, Japan). Noncardiomyocytes were defined as the cells other than cardiomyocytes in myocardium such as interstitial cells and vascular cells. The quantitative analysis was performed separately in infarcted area, noninfarcted area (salvaged myocardium), and border area, which contain both infarcted and noninfarcted areas in a high-power field under a microscope.

Western blotting. Proteins extracted from whole cardiac ventricles were subjected to 14% polyacrylamide gel electrophoresis and then transferred to polyvinylidene difluoride membranes. The membranes were then probed using primary antibodies against Akt, phosphorylated Akt (p-Akt), eNOS (Cell Signaling), p-eNOS (BD Pharmingen), and caspase-3 (Cell Signaling). Three to five hearts from each group were used for blotting. The blots were visualized using enhanced chemiluminescence (ECL; Amersham), and the signals were quantified by densitometry. α-Tubulin (Santa Cruz Biotechnology) served as the loading control.

Hypoxpyrobe staining. Tissue hypoxia was estimated using Hypoxyprobe (pimonidazole hydrochloride; Chemicon) as previously described (9). Hypoxyprobe uses an immunohistochemical reaction to detect local tissue hypoxia (2). In a low oxygen environment, the pimonidazole is reductively activated and binds to cysteine residues and thiols in hypoxic cells, forming protein adducts that can be detected using standard immunohistochemical methods.

Statistical analysis. Values are shown as means ± SE. Animal survival was analyzed using the Kaplan-Meier method with the log-rank Cox-Mantel test. The significance of differences in the findings was evaluated using t-tests or one-way ANOVA followed by Newman-Keul’s multiple comparisons test. Values of P < 0.05 were considered significant.

RESULTS

Survival and cardiac function and histology during the chronic stage of MI. Mice in the control and OPC groups (n = 18 in each) were followed up 4 wk post-MI. At that time, mice in the OPC group had a significantly greater rate of survival (83%) than those in the control group (44%; P < 0.05; Fig. 1). Echocardiography and cardiac catheterization carried out 4 wk post-MI revealed that, compared with the sham-operated mice, control MI mice had marked enlargement of the LV cavity and reduced cardiac function, as indicated by increased LV end-diastolic diameter, reduced LV percent fractional shortening, increased LV wall stress, and decreased ±dP/dt (Fig. 2). All of these structural and functional parameters were improved in the OPC group without lowering LV systolic pressure: LV end-diastolic diameter (control, 5.2 ± 0.25 mm vs. OPC, 4.7 ± 0.09 mm; P < 0.05); LV percent fractional shortening (control, 13 ± 1.5% vs. OPC, 23 ± 1.2%; P < 0.05); LV wall stress at diastole (control, 353 ± 53 dyne/mm² vs. OPC, 200 ± 25 dyne/mm²; P < 0.05).
that OPC-28326 mitigates post-MI remodeling and cardiac dysfunction.

Four weeks post-MI, heart weights were significantly lower than control in mice treated with OPC-28326 (control, 141 ± 6 mg vs. OPC, 117 ± 3 mg; P < 0.05), as were the heart-to-body weight ratios (control: 4.41 ± 0.29 mg/g vs. OPC, 3.74 ± 0.10 mg/g; P < 0.05). Hearts from control mice also showed marked LV dilatation with a thin infarct wall, while those from OPC-28326-treated mice had substantially smaller LV cavities and thicker infarct segments (control, 0.306 ± 0.019 mm vs. OPC, 0.440 ± 0.041 mm; P < 0.05) with shorter circumferential lengths (control, 9.1 ± 0.5 mm vs. OPC, 7.6 ± 0.5 mm; P < 0.05; Fig. 3). On the other hand, both the absolute area of the infarct and the percentage infarct area in a total LV area were comparable between the two groups (control, 2.5 ± 0.2 mm² vs. OPC, 2.7 ± 0.2 mm²; P = NS; and control, 39.7 ± 4.0% vs. OPC, 40.1 ± 4.1%; P = NS). Thus treatment with OPC-28326 resulted in a shorter and thicker infarct scar without changing the area of infarct. The thickness of the ventricu-

![Fig. 1. Kaplan-Meier analysis of postinfarction survival in mice (n = 6 for the sham operated group and n = 18 each for control group and OPC-28326 group at the start of the experiment).](image1.png)

![Fig. 2. Cardiac geometry and function during the chronic stage of myocardial infarction (4 wk post-MI). A: echocardiographic data: left ventricular diastolic diameter (LVDd); %fractional shortening (%FS); and heart rate. B: cardiac catheterization data: left ventricular peak systolic pressure (LVSP); left ventricular diastolic average wall stress (LVWS); and maximal and minimal first derivative of left ventricular pressure (+dP/dt and −dP/dt); n = 6 for the sham operated group, n = 8 for control group; n = 15 for OPC group. *P < 0.05 vs. sham; #P < 0.05 vs. control.](image2.png)
ular septum (noninfarcted myocardium) was significantly smaller in the OPC-treated group (control, 0.996 ± 0.022 mm vs. OPC, 0.893 ± 0.012 mm; \(P < 0.05\); Fig. 3), suggesting a less developed compensatory hypertrophy of the left ventricle in this group.

In addition, by 4 wk post-MI, the infarct area had been largely replaced by fibrous scar tissue in control mice (Fig. 4). In the OPC group, by contrast, abundant cellular components were present along with the collagen fibers. As a result, the population of interstitial noncardiomyocytes within the infarct area was significantly greater in the OPC group (Fig. 4). The numbers of Flk-1-positive blood vessels (mainly capillaries and \(\alpha\)-SMA-positive small arteries) as well as the percent area of extravascular \(\alpha\)-SMA-positive myofibroblasts were greater within infarct areas in the OPC group than in the control group (Fig. 4). The size of cardiomyocytes (assessed as the transverse diameter of cardiomyocytes) was significantly smaller in the OPC group than in the control group (Fig. 4 and Table 1), suggesting that compensatory hypertrophy of cardiomyocytes was less developed in the former. In addition, fibrosis was significantly less in the infarcted, border, and noninfarcted areas, of OPC-28326-treated hearts than the control hearts (Table 1). These findings support mitigation of pathological cardiac remodeling in OPC-28326-treated hearts.

**Subacute stage of MI.** As mentioned above, we observed greater numbers of myofibroblasts, capillaries, and small arteries within the infarct areas in OPC-28326-treated mice during the chronic stage of MI (4 wk post-MI). To clarify the mechanisms responsible for this difference in infarct...
tissue composition, we next evaluated cell proliferation and apoptosis among granulation tissue cells in hearts 5 days post-MI (subacute stage). Immunohistochemistry showed that the incidence of Ki-67 positivity was markedly greater in infarct tissue from OPC-28326-treated mice than in tissue from control mice (control, 9.6 ± 0.3% vs. OPC, 18 ± 0.5%; \( P < 0.05 \); Fig. 5A). Conversely, the prevalence of TUNEL positivity was significantly smaller in the OPC group (control, 3.0 ± 0.3% vs. OPC, 2.1 ± 0.2%; \( P < 0.05 \)), suggesting that OPC-28326 treatment reduced apoptosis among granulation tissue cells (Fig. 5B). Consistent with that idea, Western blot analysis showed that while little cleaved (active) caspase-3 was detected in sham-operated mouse hearts, markedly higher levels were detected in hearts with 1-wk-old MIs. On the other hand, the cleaved caspase-3 signal was significantly diminished in hearts treated with OPC-28326, compared with control (Fig. 5C). We checked cardiomyocyte proliferation by double immunofluorescence for myoglobin and Ki-67 (data not shown), but we could scarcely observe Ki-67-positive cardiomyocytes and the incidence was not different in control and OPC groups (0.0037 ± 0.0021 vs. 0.0042 ± 0.0027%; \( n = 6 \) each; \( P = \text{NS} \)).

It was previously reported that OPC-28326 dose dependently induces phosphorylation (activation) of Akt and eNOS in cultured endothelial cells (33). Western blot analysis of myocardial tissue 5 days post-MI revealed greater phosphorylation of both Akt and eNOS in control group than the sham-operated group (Fig. 6). Moreover, phosphorylation levels of both Akt and eNOS were even higher in the OPC group than the control group (Fig. 6).

**Hypoxic status of the infarct tissue.** To assess levels of tissue ischemia, we used Hypoxyprobe (pimonidazole hydrochloride) staining to examine the hypoxic status of the infarct tissue. As shown in Fig. 7, the staining was clearly weaker in infarct tissue from OPC-treated mice than from control mice. This suggests that hypoxia was substantially mitigated in the infarct tissue of OPC-treated mice.

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**Table 1. Morphometry of the hearts with 4-wk-old myocardial infarction treated with saline or OPC-28326**

<table>
<thead>
<tr>
<th></th>
<th>Infarct Area</th>
<th>Border Area</th>
<th>Non-infarct Area</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control ((n = 6))</td>
<td>OPC ((n = 6))</td>
<td>Control ((n = 6))</td>
</tr>
<tr>
<td>No. of nonmyocyte, per HPF</td>
<td>595 ± 14</td>
<td>667 ± 27*</td>
<td>625 ± 10</td>
</tr>
<tr>
<td>No. of Flk-1+ vessels, per HPF</td>
<td>5.1 ± 0.4</td>
<td>9.9 ± 0.7*</td>
<td>10.9 ± 0.5</td>
</tr>
<tr>
<td>No. of α-SMA+ vessels, per HPF</td>
<td>1.5 ± 0.3</td>
<td>2.9 ± 0.4*</td>
<td>1.6 ± 0.2</td>
</tr>
<tr>
<td>%Area of α-SMA+ myofibroblasts</td>
<td>5.0 ± 0.4</td>
<td>9.2 ± 0.5*</td>
<td>6.0 ± 0.2</td>
</tr>
<tr>
<td>Myocyte size, μm</td>
<td>NA</td>
<td>NA</td>
<td>20.2 ± 0.9</td>
</tr>
<tr>
<td>%Area of fibrosis</td>
<td>55.9 ± 3.4</td>
<td>45.2 ± 3.1*</td>
<td>32.3 ± 2.2</td>
</tr>
</tbody>
</table>

Values are means ± SE. Control, saline; OPC, OPC-28326; HPF, high-power field; α-SMA, α-smooth muscle actin; NA, not applicable. *\( P < 0.05 \), compared with control.
DISCUSSION

The results of the present study show that treatment with OPC-28326, started after the onset of MI, significantly improves survival and mitigates postinfarction cardiac remodeling and dysfunction during the chronic stage of MI in mice. OPC-28326 displayed angiogenic action probably via the Akt/eNOS pathway and mitigated ischemia of the infarcted tissue. This brought about an increase in nonmyocyte population of the infarcted area through increasing cell proliferation and reducing apoptosis (late reperfusion-like effect), thereby altering the infarct tissue dynamics (a shorter and thicker infarct wall) to result in reduction of wall stress, which might have attenuated progression of postinfarction cardiac remodeling and heart failure. Here we discuss those possible mechanisms underlying beneficial effects of OPC-28326 on the postinfarction heart in detail.

Pathophysiological mechanisms of the beneficial effects of OPC-28326 on postinfarction hearts. One remarkable finding of the present study is that OPC-28326 altered the geometry of the infarct scar without affecting its absolute area, i.e., the infarct segment was thicker and had a smaller circumferential length during the chronic stage in hearts from mice in the OPC group than control hearts. This is noteworthy because wall stress is directly proportional to cavity diameter and inversely proportional to wall thickness (Laplace’s law) and because wall stress and LV remodeling (dilatation) have a vicious relationship exacerbating one another (40). The wall stress in the OPC group was indeed smaller than in the control group. It is thus conceivable that the observed change in infarct geometry substantially improved the hemodynamic state of the heart.

Histologically, we observed greater numbers of cells, including abundant vascular cells and myofibroblasts, within the infarct scar in OPC-28326-treated hearts during the chronic stage of MI (5 days post-MI). These cells are destined to disappear via apoptosis during the natural course of healing after MI (6, 35), but we found that apoptosis was significantly inhibited in OPC-28326-treated hearts during the subacute stage (5 days post-MI). We previously reported that antiapoptotic treatment preserved the noncardiomyocyte population in the infarct area and significantly mitigated post-MI cardiac remodeling and dysfunction (12, 20). Moreover, OPC-28326 treatment significantly increased proliferation of both myofibroblasts and vascular components (including capillaries and small arteries) in the infarct area. These findings have two important implications. First, both diminished apoptosis and enhanced cell proliferation among granulation tissue cells during the subacute stage appear to contribute to the observed increase in the cell

Fig. 5. Cell proliferation and apoptosis in granulation tissue (infarcted area) in hearts during the subacute stage of MI (5 days post-MI). A: anti-Ki-67 immunostaining showing proliferating noncardiomyocytes (brown nuclei). The graph shows the prevalence of Ki-67-positive cells. B: terminal deoxynucleotidyl transferase-mediated in situ nick-end labeling (TUNEL) showing apoptotic noncardiomyocytes (brown nuclei). The graph shows the prevalence of apoptotic cells. C: Western blots showing myocardial expression of caspase-3. The graph shows the relative density of the cleaved caspase-3 blot. Scale bars = 20 μm; n = 6 each for control group and OPC group. *P < 0.05 vs. sham; #P < 0.05 vs. control.
population within scar tissue during the chronic stage, which likely preserves the infarct wall thickness. Second, myofibroblasts, which are known to play an important role in wound contraction during the healing process (9), could mediate contraction-induced reduction in the length of the infarct segment, thereby increasing infarct wall thickness. That in turn would alter the infarct tissue geometry, reducing wall stress and mitigating LV dilatation and dysfunction (20, 24, 37).

**OPC-28326 brings about a late reperfusion-like effect (pharmacological late reperfusion).** Late reperfusion, beyond the time window for myocardial salvage, also appears to reduce LV remodeling and decreases mortality (16, 22). This observation is the basis of the so-called open artery hypothesis, and a number of possible mechanisms by which an open infarct-related artery could confer benefit in ways other than salvaging ischemic myocardium have been proposed. Increased wall thickness without a reduction in infarct scar area is reportedly one of the morphological characteristics of postinfarction hearts receiving late reperfusion (10, 13, 16, 22). We previously reported that blood flow into the infarct area through late reperfusion 24 h after inducing MI in rats promoted cell proliferation and inhibited apoptosis among granulation tissue cells, which we suppose contributes significantly to the formation of a thick, cell-rich infarct scar during the chronic stage of MI (24). Of note is the striking similarity between the effects of late reperfusion and OPC-28326 treatment on infarct tissue dynamics, as shown in the present study. Moreover, when we assessed the ischemic status of the infarct tissue using Hypoxyprobe, we found that hypoxia was in fact reduced in the infarct tissue of OPC-28326-treated mice. Therefore, we propose that treatment with OPC-28326 after the onset of acute MI may induce a kind of pharmacological late reperfusion.

In the present study, we demonstrated an increased number of capillaries and small arteries in the OPC-treated postinfarction heart. We suppose such angiogenic action is one of the most important contributors to the beneficial effect of OPC-28326 on the postinfarction heart. Such action could relieve ischemia of the infarcted tissue through late reperfusion-like effect, decrease apoptosis while increase cell proliferation to alter geometry of the infarcted tissue, and finally mitigate adverse remodeling of the postinfarction heart. On the other hand, vasodilating action is unlikely to contribute to the benefits of OPC-28326 in the present setting because OPC-28326 does not increase coronary blood flow (27). Also unlikely is the possibility of afterload reduction due to peripheral vasodilating effect because systemic blood pressure was unchanged by the treatment with OPC-28326. We also noted that activation of Akt and eNOS in the postinfarction heart was augmented furthermore by the treatment with OPC-28326. Although it still remains unknown how the activation of eNOS promotes ischemia-induced angiogenesis, nitric oxide exerts several favorable effects on endothelial cells, including inhibition of apo-

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**Fig. 6.** Western blots show the relative levels of the indicated proteins in hearts during the subacute stage of MI (5 days post-MI). In the densitometric graphs, relative activation levels are indicated by the p-Akt-to-Akt and p-endothelial nitric oxide synthase (eNOS)-to-eNOS ratios; n = 3 each for control group and OPC group. *P < 0.05 vs. sham; #P < 0.05 vs. control.

**Fig. 7.** Tissue hypoxia assessed based on Hypoxyprobe staining of the hearts during the subacute stage of MI (5 days post-MI). Hypoxic tissue is stained intensely brown. A: control heart. B: OPC-28326-treated heart. Both preparations show cardiac tissues containing both infarcted area (I) and salvaged myocardium (S); n = 6 each for control group and OPC group. Scale bars = 50 μm.
ptosis (7) and promotion of migration (23). Sumi et al. (33) previously reported that OPC-28326 activated eNOS in cultured endothelial cells and that both blood flow recovering and angiogenic actions by OPC-26326 were abrogated in ischemic legs of eNOS-deficient mice. These results suggest that OPC-28326 promotes angiogenesis via direct activation of Akt-eNOS pathway in endothelial cells.

Limitations of the study. Although we showed the beneficial effect of OPC-28326 on postinfarction hearts, it should be cautioned that the benefit was evident in cases with large, transmural infarcts, which are relatively rare in humans, probably because of collateral development. Thus the outcome would be unknown if the therapy were applied to cases with subendocardial infarction.

In the present study, we used 8- to 10-wk-old mice, which are relatively young. In humans, however, MI is in general a disease that affects people with old age. Thus it might be warranted to confirm the efficacy of the reagents also in older mice.

Clinical implications. Rapid recanalization of the occluded coronary artery, which salvages ischemic myocardial cells, is at present the best clinical approach to treating acute MI. Unfortunately, most patients actually lose their chance for coronary reperfusion during the acute stage. In the present study, we found striking similarity between both the effects of OPC-28326 on postinfarction hearts, it should be remembered that the therapy were applied to cases with subendocardial infarction.


