Bionic Baroreceptor Corrects Postural Hypotension in Rats With Impaired Baroreceptor

Kazuya Hosokawa, MD; Tomomi Ide, MD, PhD; Tomoyuki Tobushi, MD; Kazuo Sakamoto, MD; Ken Onitsuka, MD; Takaumi Sakamoto, MD; Takeo Fujino, MD; Keita Saku, MD; Kenji Sunagawa, MD, PhD

Background—Impairment of the arterial baroreflex causes orthostatic hypotension. Arterial baroreceptor sensitivity degrades with age. Thus, an impaired baroreceptor plays a pivotal role in orthostatic hypotension in most elderly patients. There is no effective treatment for orthostatic hypotension. The aims of this investigation were to develop a bionic baroreceptor (BBR) and to verify whether it corrects postural hypotension.

Methods and Results—The BBR consists of a pressure sensor, a regulator, and a neurostimulator. In 35 Sprague-Dawley rats, we vascularly and neurally isolated the baroreceptor regions and attached electrodes to the aortic depressor nerve for stimulation. To mimic impaired baroreceptors, we maintained intracarotid sinus pressure at 60 mm Hg during activation of the BBR. Native baroreflex was reproduced by matching intracarotid sinus pressure to the instantaneous pulsatile aortic pressure. The encoding rule for translating intracarotid sinus pressure into stimulation of the aortic depressor nerve was identified by a white noise technique and applied to the regulator. The open-loop arterial pressure response to intracarotid sinus pressure (n = 7) and upright tilt–induced changes in arterial pressure (n = 7) were compared between native baroreceptor and BBR conditions. The intracarotid sinus pressure–arterial pressure relationships were comparable. Compared with the absence of baroreflex, the BBR corrected tilt-induced hypotension as effectively as under native baroreceptor conditions (native, −39±5 mm Hg; BBR, −41±5 mm Hg; absence, −63±5 mm Hg; P < 0.05).

Conclusions—The BBR restores the pressure buffering function. Although this research demonstrated feasibility of the BBR, further research is needed to verify its long-term effect and safety in larger animal models and humans. (Circulation. 2012;126:1278-1285.)

Key Words: baroreceptors ■ blood pressure ■ electric stimulation ■ hypotension (low blood pressure) ■ nervous system, autonomic

The prevalence of orthostatic hypotension (OH) increases with age and reaches 20% in those older than 65 years of age.1,2 Various characteristic symptoms of OH, including dizziness, lightheadedness, visual blurring, chest discomfort, and syncope, deteriorate the quality of life. Recurrent falls, often experienced by OH patients, may cause hip fracture and head trauma. Meanwhile, 10% to 31% of patients presenting to emergency departments with syncope are diagnosed with OH-related syncope,3–5 and OH accounts for 0.4% of all hospital admissions.6 Although OH imposes a heavy burden in terms of medical costs, treatments for OH such as pharmacological pressor agents, elastic lower-limb stockings, and avoidance of rapid postural change remain unsatisfactory.

Clinical Perspective on p 1285 Arterial baroreflex dysfunction is well recognized as one of the most important causes of OH. Regardless of the disease (spinal cord injury, primary neurogenic diseases, peripheral neuropathies, or baroreceptor insensitivities), wherever the baroreflex is impaired, orthostatic dysregulation ensues. Previously, Sato et al7 reported that a bionic baroreflex system that electrically stimulates the efferent sympathetic nerve trunk as required is capable of preventing postural hypotension. On the other hand, OH is more likely to be observed in hypertensive patients.8,9 The comorbid condition of hypertension complicates the therapeutic strategy for maintaining optimal blood pressure. The previous bionic baroreflex system has several limitations. First, although the system has a pressor action, it is not capable of inducing a depressor effect. Second, the system stimulates the efferent sympathetic nerve, which may affect not only blood pressure but also other abdominal organs, and hence, it carries a risk of adverse effects. Therefore, there is still room for improvement.

Baroreceptor sensitivity is known to degrade physiologically with advancing age, and arteriosclerosis in major arteries also causes baroreceptor impairment. Several studies...
demonstrated that the arterial baroreceptor afferent root plays an important role in long-term blood pressure regulation.\textsuperscript{10–13} Recent clinical trials of baroreceptor activation therapy for resistant hypertension have shown that the therapy confers not only long-term blood pressure–lowering effects\textsuperscript{14,15} but also an immediate pressor response when the baroreceptor stimulation is deactivated. These facts and results suggest that an impaired baroreceptor plays a pivotal role in elderly OH patients who have comorbid hypertension. Therefore, an alternative bionic baroreflex system designed to substitute for impaired baroreceptors may restore bidirectional arterial pressure (AP) regulation, thereby preventing both OH and comorbid hypertension in those patients. To the best of our knowledge, there is no effective treatment for baroreceptor impairment. The purposes of the present investigation were to develop an alternative bionic baroreflex system that substitutes for the native baroreceptor and to verify its performance in AP regulation against postural hypotension.

Methods

Animals and Surgical Procedures

Experiments and animal care were approved by the Committee on Ethics of Animal Experiment, Kyushu University Graduate School of Medical Sciences, and performed in strict accordance with the Guide for the Care and Use of Laboratory Animals published by the US National Institutes of Health. Thirty-five Sprague Dawley rats weighing 620±15 g were anesthetized by intraperitoneal injection (2\textsuperscript{mg}/kg) of a mixture of Medetomidine (H11006) 0.15 g and Ketamine (H9004) 15 g were anesthetized by intraperitoneal injection (2\textsuperscript{mg}/kg) of a mixture of Medetomidine (H11006) 0.15 g and Ketamine (H9004) 15 g. An appropriate level of anesthesia was maintained via oxygen-enriched air spontaneously. At the end of the experiments, we confirmed that the arterial blood pH, PCO\textsubscript{2}, and bicarbonate were not only long-term blood pressure–lowering effects\textsuperscript{14,15} but also an immediate pressor response when the baroreceptor stimulation is deactivated. These facts and results suggest that an impaired baroreceptor plays a pivotal role in elderly OH patients who have comorbid hypertension. Therefore, an alternative bionic baroreflex system designed to substitute for impaired baroreceptors may restore bidirectional arterial pressure (AP) regulation, thereby preventing both OH and comorbid hypertension in those patients. To the best of our knowledge, there is no effective treatment for baroreceptor impairment. The purposes of the present investigation were to develop an alternative bionic baroreflex system that substitutes for the native baroreceptor and to verify its performance in AP regulation against postural hypotension.

Figure 1. The native baroreflex feedback loop and framework of the bionic baroreceptor. The native baroreflex consists of baroreceptors, the vasomotor center, and the cardiovascular actuators. The baroreceptors translate arterial pressure (AP) to the carotid sinus nerve or the aortic depressor nerve firing. In the bionic baroreceptor, a pressure sensor senses AP and a regulator translates AP into neurostimulation signals, and then a neurostimulator generates electric pulse trains. The regulator changes the frequency of the electric pulse train.

Bionic Baroreflex System

Feedback loops of the arterial baroreflex are shown in Figure 1. The native arterial baroreceptor senses a change in AP and transmits the message to the vasomotor center via afferent nerves (aortic depressor nerves and carotid sinus nerves). The vasomotor center modulates sympathetic outflow depending on inputs from the baroreceptors. The efferent sympathetic nerve firing facilitates the cardiovascular actuators and induces the resultant AP change. This negative feedback loop is the biological mechanism that stabilizes blood pressure. The bionic baroreceptor consists of a pressure sensor (DX-360, MEG-5200; Nihonkohden), a neurostimulator (SEN-3401, Nihonkohden; AD202JN, Analog Devices), and a regulator (Studio 1410; Dell). The bionic pressure sensor senses AP, and the regulator translates AP into neurostimulation (STM stimulation of the aortic depressor nerve), thus mimicking the native baroreceptor. Therefore, the bionic baroreceptor is expected to substitute for impaired baroreceptors. The neurostimulator generates an electric rectangular pulse train (5-V intensity, 0.2-ms pulse width) according to the command signal from the regulator. The regulator updates the frequency of the pulse train every 10 ms.

Protocol 1: Development of Regulator in Bionic Baroreflex System

To eliminate the native baroreceptor afferent signals and to mimic an impaired baroreceptor, we maintained CSP at 60 mm Hg during the bionic baroreflex condition. Because lowering CSP induces baroreflex-mediated hypertension, we stimulated the aortic depressor nerve at a constant frequency and titrated upward until the mean AP was maintained at the same level as in the closed-loop baroreflex condition (Figure 2). The stimulation frequency required to maintain the mean AP was defined as the bias frequency. Then we identified the operating rule required for the bionic regulator to translate ACSP into ΔSTM. Details of identification of the operating rule are illustrated in Figure 3. The total transfer function from CSP to AP (H\textsubscript{CSP-AP}) can be divided into 2 components: The transfer function translating CSP into STM by the bionic regulator (H\textsubscript{Bionic}) and the transfer function from STM to AP (H\textsubscript{STM-AP}). H\textsubscript{Bionic} is obtained from H\textsubscript{CSP-AP}/H\textsubscript{STM-AP} and H\textsubscript{CSP-AP} and H\textsubscript{STM-AP} can be measured experimentally in individual rats (n=35; protocol 1). We randomly changed CSP in binary values (mean AP±20 mm Hg) every 500 ms and obtained CSP-AP data sets. To obtain STM-AP data sets, we randomly altered the frequencies of STM in binary values (bias frequency±80% of bias frequency) every 500 ms. These random perturbations were performed for 10 to 20 minutes, and stable 5-minute data sets were used for the following analysis. Experimental data pairs were recorded at a 200-Hz sampling interval. The input-output data pairs were divided into 5 segments and processed with 50% overlapping bins of 20,000 points each by a fast Fourier transform algorithm to identify H\textsubscript{CSP-AP} and H\textsubscript{STM-AP}. H\textsubscript{Bionic} was calculated as the ratio of H\textsubscript{CSP-AP} to H\textsubscript{STM-AP}. The H\textsubscript{Bionic} obtained was converted to impulse response (h\textsubscript{Bionic}), which is the operating rule, by an inverse fast Fourier transform algorithm. Gains and phase lags >0.5 Hz (poor coherence spectrum caused by spontaneous breathing and low gain input) were replaced with the average gain.
and a phase lag <0.5 Hz. In the following experiments, we applied the individual operating rule to the regulator for each rat.

**Protocol 2: Validation of Bionic Pressure Regulation Under Open-Loop Conditions**

To examine the performance of bionic AP regulation under open-loop conditions, the AP responses to stepwise CSP increases were examined (n=7). To mimic the native baroreflex condition, we deactivated the bionic baroreceptor and increased the native CSP stepwise every 20 seconds: 60, 80, 100, 115, 130, 150, and 170 mm Hg (native). To obtain the bionic baroreflex condition, we reactivated the bionic system and imposed the same stepwise pressure change on the bionic pressure sensor while keeping the native CSP at 60 mm Hg (bionic). We conducted the stepwise cycle...
under native and bionic conditions alternately for a total of 6 times in each rat. The average AP for the last 5 seconds of each step was plotted on a CSP-AP diagram. The CSP-AP data sets for the corresponding 3 cycles were averaged as native or bionic conditions, respectively. To compare the baroreflex function between native and bionic conditions, the CSP-AP diagrams were fitted to a 4-parameter logistic function and the coefficients compared.17

Protocol 3: Validation of Bionic Pressure Buffering Effect Against Head-Up Tilt Under Closed-Loop Conditions

To examine whether the bionic baroreceptor corrects postural hypotension, we compared AP changes between native and bionic conditions in rats placed on a tilt table (80°, 30 seconds; n=7). To attempt to reproduce the closed-loop native baroreflex condition, the native CSP was matched to the instantaneous pulsatile aortic pressure (native). Conversely, the closed-loop bionic baroreflex condition was obtained by inputting the instantaneous pulsatile aortic pressure into the bionic sensor (bionic). CSP was maintained at 60 mm Hg during the bionic condition. To compare the 2 conditions with the absence of baroreflex feedback, we repeated head-up tilt tests while keeping CSP at baseline supine AP (open-loop baroreflex condition [open]). The 3 conditions (native, bionic, and open) were performed in random sequence in the same animals. We compared AP responses at 5, 15, and 25 seconds after upright tilting (ΔAPᵢ, ΔAP₁₅, and ΔAP₂₅).

Statistical Analysis

All data are presented as mean±SE. The gains at 0.01, 0.05, and 0.5 Hz (G₀.₀₁, G₀.₀₅, and G₀.₅), and the slope of dynamic gain (G_slope) between 0.05 and 0.5 Hz were derived from the transfer functions. In protocol 2, the CSP-AP relationships were fitted to the 4-parameter logistic function: AP=P₁/(1+e^(-P₂·CSP-P₃))+P₄, where P₁, P₂, P₃, and P₄ represent response ranges of AP, coefficient of gain, CSP corresponding to the midpoint operation, and minimum AP in operation, respectively. The nonlinear least-squares method with a generalized reduced-gradient algorithm was used for curve fitting. Student paired t tests and repeated-measures ANOVA with post hoc Tukey’s honestly significant difference tests were used to detect statistical significance. Statistical analyses were performed with JMP release 8.0 (SAS Institute Inc) and Microsoft Office Excel 2010 (Microsoft Corp). P<0.05 was considered to indicate a significant difference.

Results

Protocol 1: Development of Regulator in Bionic Baroreflex System

The mean AP under the native closed-loop baroreflex condition was 99±2 mm Hg, and the bias frequency was 13.9±1.4 Hz. The characteristics of H_CSP-AP approximated a second-order low-pass filter (Figure 4A). The gains (mm Hg/mm Hg)
were as follows: $G_{0.01}$, 0.71±0.04; $G_{0.05}$, 0.69±0.04; and $G_{0.5}$, 0.08±0.01. $G_{\text{slope}}$ was $-9.7\pm0.5$ dB/decade. The phase lag was close to $\pi$ radians at 0.01 Hz and out of phase up to 0.08 Hz, which reflects the negative feedback operation. The characteristics of $H_{\text{STM-AP}}$ were similar to $H_{\text{CSP-AP}}$ (Figure 4A). The gains (mm Hg/mm Hg) were as follows: $G_{0.01}$, 2.25±0.21; $G_{0.05}$, 1.94±0.20; and $G_{0.5}$, 0.14±0.02. $G_{\text{slope}}$ was $-10.8\pm0.1$ dB/decade. The phase lag was close to $\pi$ radians at 0.01 Hz and out of phase up to 0.07 Hz, similar to $H_{\text{CSP-AP}}$. The coherence of $H_{\text{CSP-AP}}$ and $H_{\text{STM-AP}}$ below 0.6 Hz exceeded 0.6. $H_{\text{Bionic}}$ is shown in Figure 4B. The gain was 0.6 Hz/mm Hg at 0.01 Hz, remained almost flat up to 0.1 Hz, and then increased slightly with an increase in frequency. The phase lags were close to zero up to 0.3 Hz. Because the impulse response ($h_{\text{Bionic}}$) approximated the delta function (Figure 4C), a simple gain scalar that was the time integral of $h_{\text{Bionic}}$ was applied to the regulator to avoid computational burden.

Protocol 2: Validation of Bionic Pressure Regulation Under Open-Loop Conditions

The representative time-tracking data are shown in Figure 5A. In the CSP-AP diagram (Figure 5B; pooled data), every AP response to CSPs through the bionic baroreceptor was similar to the native baroreflex. The coefficients that corresponded to the fitted 4-parameter logistic function curve and the maximal gain revealed nonsignificant differences between them (Table). The result ensured that the bionic baroreflex reasonably replicated the native baroreflex in terms of AP regulation.

Discussion

This report is the first to describe a novel bionic baroreceptor that implements baroreceptor transduction as the native baroreceptor response to ameliorate upright tilt–induced hypotension in rats with impaired baroreceptors.

A new concept of the bionic baroreceptor is that it theoretically permits bidirectional AP regulation to prevent both postural hypotension and hypertension in patients with OH caused by an impaired baroreceptor. Because the bionic baroreceptor controls neurostimulation in response to AP by a bionic closed-loop feedback in a manner as effective as the native baroreflex, bionic baroreceptor therapy can avoid both excessive AP fall and rise, which are often observed with conventional therapies. We demonstrated herein a bionic pressure buffering effect only for a short period; however, the existing technology using electric field stimulation, which has already been used in clinical trials, is theoretically applicable for blood pressure regulation in the long term.

Table. Parameters of the Sigmoidal Logistic Function

<table>
<thead>
<tr>
<th></th>
<th>Native</th>
<th>Bionic</th>
<th>$P$ Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>$P_1$: Range, mm Hg</td>
<td>84±8</td>
<td>80±9</td>
<td>0.39</td>
</tr>
<tr>
<td>$P_2$: Coefficient of gain, mm Hg$^{-1}$</td>
<td>0.11±0.02</td>
<td>0.12±0.03</td>
<td>0.30</td>
</tr>
<tr>
<td>$P_3$: Midpoint, mm Hg</td>
<td>111±5</td>
<td>110±6</td>
<td>0.42</td>
</tr>
<tr>
<td>$P_4$: Min, mm Hg</td>
<td>65±5</td>
<td>68±6</td>
<td>0.42</td>
</tr>
<tr>
<td>Gain max, unit less</td>
<td>$-2.27\pm0.33$</td>
<td>$-2.07\pm0.43$</td>
<td>0.46</td>
</tr>
</tbody>
</table>

Data are expressed as mean±SE. The curves of intracarotid sinus pressure (CSP)–arterial pressure (AP) relationships (in protocol 2) were approximated by the 4-parameter logistic function: $AP=\frac{P_1}{1+e^{P_2(CSP-P_3)}}+P_4$, where $P_1$, $P_2$, $P_3$, and $P_4$ represent response ranges of arterial pressure, coefficient of gain, carotid sinus pressure corresponding to the midpoint operation, and minimum arterial pressure in operation, respectively. Gain max equals $\frac{P_1P_4}{P_3}$ at $CSP=P_3$. Student paired t tests were used for statistical analyses.
Several epidemiological studies have reported that systolic hypertension, 2, 8 carotid wall thickening, 2 and high pulse-wave velocity 18 are significantly associated with OH. Moreover, the fact that OH is an independent risk factor for cardiovascular events and mortality even in middle-aged adults suggests a close association between arteriosclerosis and OH. 19–22 It is therefore reasonable to consider that arteriosclerosis in the baroreceptor regions induces arterial baroreceptor insensitivity and thereby plays a major pathogenetic role in most OH patients. A bionic baroreceptor would benefit many OH patients.

**Simplified Operating Rule Promised Physiological AP Regulation**

Even though we applied a simple gain scalar to the regulator, the bionic baroreceptor successfully reproduced physiological pressure regulation. To examine the adequacy of the simplification, we simulated AP responses to a depressor perturbation, comparing the raw impulse response with the simple scalar (Figure 7). The simulation showed that AP recoveries after depressor disturbance were comparable between the raw impulse response and the simple gain scalar. The characteristics of H_bionic observed in the present study were consistent with those of our previous studies in rabbits 23, 24 and canines, 25 which suggests that the transduction from CSP to afferent nerve firing or to STM is conserved across species.

### Bionic Pressure Regulation Under Open-Loop Conditions

The regulator linearly changed the frequency of neurostimulation in response to CSP. Nevertheless, the bionic baroreflex control of AP is in the form of a sigmoidal response to CSP, similar to the native baroreflex. Afferent nervous firing in response to CSP is characterized by a sigmoidal relation and is saturated beyond the physiological range. 16, 23, 26, 27 Similarly, high-frequency STM also becomes saturated in its depressor effect. 28, 29 The sigmoidal CSP-AP relationship of the native baroreflex reflects either or both saturations, whereas that of the bionic baroreflex reflects STM saturation.

### Bionic Pressure Buffering Effect Against Head-Up Tilt Under Closed-Loop Conditions

In protocol 3, we demonstrated that the bionic baroreceptor ameliorated head-up tilt–induced hypotension under the bionic closed-loop baroreflex condition. The changes in AP were identical in both the bionic and native conditions, which indicates that the bionic baroreceptor completely substituted for the native baroreceptors. In addition, the bionic baroreceptor suppressed the baseline supine hypertension. If the bias frequency were underestimated, the supine AP would increase up to 142±4 mm Hg according to the value of AP at CSP=60 mm Hg (Figure 5B). Therefore, this result verifies that the bionic baroreceptor reproduces bidirectional AP regulation.

The pressor effect of the bionic baroreceptor is attributed to subtraction of the corresponding frequency from the bias frequency according to the fall in AP. Because the native baroreceptor reduces afferent nerve firing when the AP drops, the bionic pressor mechanism appears to be reasonable and physiological. Moreover, the bias frequency compensates for the basal afferent signal from the baroreceptors and would be a therapeutic tool for comorbid supine hypertension, as in baroreflex activation therapy. 10–13

Postural hypotension was not completely prevented even in the native baroreflex condition (Figure 6A). It was thought that this was because the native baroreceptor itself was damaged by the surgical preparation. Thus, the bionic baroreceptor also could not produce a sufficiently antihypotensive effect.

### Limitations and Considerations for Clinical Application

First, the present study is preliminary and demonstrated only the very short-term effect and feasibility of this approach.
Further research is needed to verify the long-term effect and safety in larger animal models and eventually in humans. Second, for further development to practical use, it is essential to develop a safe, durable, and reliable implantable pressure sensor. Although no implantable pressure sensor is commercially available, several devices, such as the CardioMEMS sensor (CardioMEMS, Inc)\textsuperscript{30} for pulmonary arterial pressure, Chronicle (Medtronic, Inc)\textsuperscript{31} for right ventricular pressure, and HeartPOD (St Jude Medical, Inc)\textsuperscript{32} for left atrial pressure, have already been used in patients with heart failure, and the safety and feasibility of these devices have been reported. A long-term implantable AP sensor will become available in the near future.

Clinical Significances and Perspectives
A novel bionic baroreceptor system would be an attractive therapy for OH with comorbid hypertension caused by impaired baroreceptor function. Because the bionic baroreceptor substitutes for physiological baroreceptor functions, the device should be effective in any patient with impaired baroreceptor function, regardless of the underlying disease. It may also provide safer and more adaptive blood pressure–lowering therapy for most patients with hypertension if used in combination with baroreflex activation therapy. Additionally, blood pressure variability was considered as an independent risk of developing cardiovascular disease.\textsuperscript{33} Because the bionic baroreceptor is theoretically able to suppress blood pressure variability independent of the mean level of blood pressure by adjusting the gain scalar, the system may help to elucidate the causal association between blood pressure variance and cardiovascular events and mortality.

Acknowledgments
The authors thank Toru Kawada, MD, PhD, and Yoshinori Murayama, MS, for technical advice.

Sources of Funding
This study was supported in part by grants from the Ministry of Health Labour and Welfare of Japan (Health and Labour Sciences research grant for clinical research and Health and Labour Sciences research grant for research on medical devices for improving impaired quality of life) and the Japan Society for the Promotion of Science (grant-in-aid for scientific research, No. 18100006).

Disclosures
None.

References
Orthostatic hypotension is one of the most common medical problems. The prevalence of orthostatic hypotension increases with age and reaches 20% in those older than 65 years. Similarly, baroreceptor sensitivity degrades with aging and arteriosclerosis. Because arterial baroreflex dysfunction encompasses orthostatic intolerance, it is reasonable to consider that an impaired baroreceptor plays a pivotal role in orthostatic hypotension in most elderly patients. However, there is no available treatment for baroreceptor impairment. In the present study, we developed a bionic baroreceptor that consists of a pressure sensor, a regulator, and a neurostimulator. The bionic regulator translates the aortic pressure into electric stimulation of the aortic depressor nerve in real time as if it were the native baroreceptor. We demonstrated that a bionic baroreceptor system restores the pressure buffering function against head-up tilt-induced hypotension in rats without baroreflex function as well as the native baroreflex. This study proposes that application of a bionic baroreceptor could be a novel therapeutic tool for patients with orthostatic hypotension caused by baroreceptor impairment. In addition, our bionic baroreceptor theoretically enables bidirectional arterial pressure regulation. Therefore, this system will be specifically beneficial for patients with coexistent supine hypertension and orthostatic hypotension, which often causes a particularly difficult therapeutic dilemma.
Bionic Baroreceptor Corrects Postural Hypotension in Rats With Impaired Baroreceptor
Kazuya Hosokawa, Tomomi Ide, Tomoyuki Tobushi, Kazuo Sakamoto, Ken Onitsuka,
Takafumi Sakamoto, Takeo Fujino, Keita Saku and Kenji Sunagawa

Circulation. 2012;126:1278-1285; originally published online July 31, 2012;
doi: 10.1161/CIRCULATIONAHA.112.108357

Circulation is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2012 American Heart Association, Inc. All rights reserved.
Print ISSN: 0009-7322. Online ISSN: 1524-4539

The online version of this article, along with updated information and services, is located on the
World Wide Web at:
http://circ.ahajournals.org/content/126/10/1278

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published
in Circulation can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial
Office. Once the online version of the published article for which permission is being requested is located,
click Request Permissions in the middle column of the Web page under Services. Further information about
this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to Circulation is online at:
http://circ.ahajournals.org//subscriptions/