Cardiovascular Response to Static Exercise during Selective Autonomic Blockade in the Conscious Cat

GEORGE DIEPSTRA, WILLIAM GONYEA, AND JERE H. MITCHELL

SUMMARY Selective autonomic blockade with propranolol, atropine, and combined atropine and propranolol was used to elucidate the role of the autonomic nervous system in the cardiovascular responses that occur during voluntary static exercise in conscious cats. Seven animals were operantly conditioned to hold a bar against a fixed resistance for a constant time of 15 seconds and were then placed on an exercise regimen which consisted of small weekly increments in resistance. With a resistance of 100 g, heart rate (HR) increased by 7%. With exercise at a resistance of 200 g, HR (10%), left ventricular systolic pressure (LVSP, 16%), and LV max dp/dt (18%) increased, and significant changes in these parameters persisted throughout the remainder of the training period. β-Adrenergic receptor blockade with propranolol abolished the increase in LV max dp/dt, whereas HR and LVSP increased. After atropine, the increase in HR was abolished at the early training stages, whereas LVSP and LV max dp/dt increased. Administration of atropine and propranolol blocked the increase in HR and LV max dp/dt responses, whereas LVSP increased. In this study, the increased HR in response to exercise was mediated primarily by the parasympathetic nervous system, whereas the increases in LV max dp/dt were mediated by the sympathetic nervous system. Furthermore, the bradycardia accompanying chronic performance of isometric exercise resulted from both an increase in vagal tone and a decrease in sympathetic tone. Circ Res 47: 530–535, 1980

ISOMETRIC exercise has long been known to cause dramatic increases in arterial blood pressure and more moderate increases in heart rate (Alam and Smirk, 1937, 1938; Lind et al., 1964). Systolic, diastolic, and mean blood pressures all increase in response to this type of effort (Humphreys and Lind, 1963; Lind et al., 1964). More recently, static efforts also have been shown to elicit an increase in left ventricular contractile force (Fisher and Nutter, 1974; Mitchell et al., 1977; Mullins et al., 1970).

The rapid onset of the cardiovascular changes to isometric exercise strongly suggests a neurogenic mechanism (Mitchell and Wildenthal, 1974; Petro et al., 1970). The efferent mechanisms responsible for these circulatory changes have been the focus of several studies. The initial HR response during isometric exercise has been shown to be mediated by the parasympathetic nervous system in human subjects (Freyschuss, 1970; Martin et al., 1974), whereas this response is mediated by the sympathetic nervous system in anesthetized dogs and cats (Crayton et al., 1979; Mitchell et al., 1977). The pressor response accompanying static efforts persists after beta-adrenergic blockade in humans (MacDonald et al., 1966; Martin et al., 1974), whereas it is abolished by this procedure in anesthetized dogs (Crayton et al., 1979). Although these discrepant findings may be due to species differences, it is becoming increasingly apparent that data collected from anesthetized experimental animals are not necessarily descriptive of the spontaneous responses in normal conscious subjects. A number of investigators have noted that the commonly used anesthetic agents have marked effects on the cardiovascular system (Brown and Hilton, 1956; Peiss and Manning, 1964; Price et al., 1965; Van Citters et al., 1964). These findings may relate to the discrepancies that exist between studies on animals and humans conducted to elucidate the efferent mechanisms responsible for the cardiovascular changes accompanying isometric exercise.

The present study was undertaken to describe the efferent mechanisms responsible for the cardiovascular responses during static efforts in the conscious animal preparation developed by Gonyea and Ericson (1976). A preliminary report of this work has been published elsewhere (Diepstra et al., 1977).

Methods

Seven adult cats were operantly conditioned to perform isometric exercise at increasing weight loads and to hold the weight for 15 seconds. The training procedure and equipment developed for this study were an extensive modification of that used by Gonyea and Ericson (1976) in which cats were trained to perform weight-lifting exercise. The
The general design of the exercise apparatus is illustrated in Figure 1. A clear plastic enclosure with a tunnel 1.5 inches deep at one end was used in the training program. The training procedure consisted of conditioning 23-hour food-deprived cats to extend their right forelimb through the tunnel and grasp a hinged bar and to hold it against a microswitch for 15 seconds. The conditioning was done in several stages to shape the desired response. Weights attached to the hinged bar via a pulley provided the load that the cats must hold. The hinged bar was set between two microswitches that provided an upper and lower threshold. An external clock, which could be preset, provided the holding interval for the exercise event. When the hinged bar was held against the upper threshold microswitch, the preset clock was activated, along with a buzzer located inside the box. The buzzer provided audio feedback as long as the microswitch was kept closed. When the cat maintained the proper tension for a predetermined holding interval (15 seconds), a feeding apparatus dispensed a food reward. This procedure activated an external counter that registered a single event.

Following this initial behavioral conditioning, the cats were placed on a static exercise training regimen which consisted of 5 consecutive days of training and then 2 days of rest. The training regimen lasted approximately 4 months, during which the initial week of training commenced with all cats holding 100 g for 15 seconds. The weight that the cats held was then increased weekly by 50 g until the animals experienced difficulty in maintaining the holding interval, at which time increments of 10 to 30 g were instituted. The cats were housed in an isolated colony where water was given ad libitum, and received food only for successful exercise events in the training enclosure and on the first day of rest in the isolated colony.

After the cats had been conditioned to perform the exercise, but before being placed on the training regimen, they were anesthetized with sodium pentobarbital (35 mg/kg), given intraperitoneally, and a left thoracotomy was performed at the level of the 6th intercostal space. A calibrated Konigsberg P-3.5 solid state pressure transducer was inserted into the left ventricular chamber via the apex and a catheter filled with heparinized saline was implanted in the left atrium. Both were exteriorized between the scapulae and placed in pouches located on a modified small animal harness. After a 1- to 2-week recuperative period, the instrumented cats were returned to the training regimen. Left ventricular systolic pressure (LVSP), rate of left ventricular pressure development (LV dp/dt), and heart rate (HR) were recorded on an Electronics for Medicine recorder. Left ventricular dp/dt was measured from the left ventricular pressure signal with an R/C differentiator and an ECG preamplifier. Heart rate was recorded through a calibrated tachographic preamplifier triggered from the pulsatile left ventricular pressure signal.

The cardiovascular parameters were recorded continuously while each cat was in the training enclosure at each 100 g increment in the training regimen. Body weight measurements were obtained daily, and a record was kept of the daily number of events at each weight load throughout the training regimen.

β-Adrenergic receptor blockade, vagal blockade, and combined β-adrenergic receptor blockade and vagal blockade were effected at each 100 g increment in the training regimen for the seven cats. Propranolol (1 mg/kg) was administered to produce β-adrenergic receptor blockade, atropine (1 mg/cat) was administered to produce vagal blockade, and combined blockade was effected with propranolol plus atropine given via the left atrial catheter. Selective autonomic blockade was instituted on separate days so that the blockade procedure lasted 3 consecutive days at each designated training level for each cat. The sequence of the drug administration was randomized.

Two additional exercise control cats were placed on a 3-month training regimen that consisted of holding the same weight (20 g) for 15 seconds. The
exercise intensity for these animals was maintained far below that necessary to induce an increase in LVSP pressure. In addition, three control cats were maintained in the isolated colony as cage-confined controls for 3 months. In all animals LVSP, LV dp/dt, and HR were measured.

Statistical significance between two samples was determined using the Student’s paired t-test. The Student-Newman-Keuls Multiple Range Test with single factor analysis of variance (Zar, 1974) was used for comparisons of three or more samples. Differences were considered statistically significant if the 0.05 probability level was attained.

Results

Comparison of the average pre-exercise and peak response values for HR, LVSP, and LV max dp/dt during isometric exercise are presented in Table 1 for normal (N), propranolol-treated (P), atropine-treated (A), and propranolol plus atropine-treated (P+A) conditions during increasing exercise resistance.

Responses to Isometric Exercise before Drug Intervention

The exercise subjects’ mean body weight did not change significantly during the training period, indicating that the diet was sufficient to prevent weight loss during the training regimen. Significant increases in HR from 218 ± 3 beats/min to 234 ± 3 beats/min were observed at the outset of the training program when the cats were holding 100 g for 15 seconds. HR increased significantly throughout the remainder of the training period, reaching a maximal response (+33 ± 4 beats/min) at the 400 g training stage. The rise in HR began before the onset of exercise, reached a peak in the first 5 seconds of exercise, and then decreased to the pre-exercise level. LVSP (+23 ± 3 mm Hg) increased significantly in response to isometric exercise when the holding tension was 200 g. Accompanying the rise in LVSP, LV max dp/dt increased from 3495 ± 146 mm Hg/sec to 4236 ± 178 mm Hg/sec. Significant increases in these two parameters persisted throughout the remainder of the training period.

During the time course of the training program, significant reductions in pre-exercise HR from 218 ± 3 to 142 ± 2 beats/min and peak exercise HR from 234 ± 3 to 165 ± 3 beats/min were observed during the training regimen (Table 1). Cats trained for 3 months at a constant holding tension (20 g) also exhibited a similar reduction in pre-exercise HR after training. No reductions in HR were observed in the cage confined control cats.

Responses to Isometric Exercise after Propranolol

Administration of propranolol resulted in reductions in pre-exercise values for HR and LV max dp/dt and more moderate reductions in pre-exercise

Table 1 Cardiovascular Responses to Isometric Exercise before and after Autonomic Blockade

<table>
<thead>
<tr>
<th>Holding tension (g)</th>
<th>Pre-exercise HR (beats/min)</th>
<th>Peak exercise HR (beats/min)</th>
<th>Pre-exercise LVSP (mm Hg)</th>
<th>Peak exercise LVSP (mm Hg)</th>
<th>Pre-exercise LVmax dp/dt (mm Hg/sec)</th>
<th>Peak exercise LVmax dp/dt (mm Hg/sec)</th>
</tr>
</thead>
<tbody>
<tr>
<td>100 N</td>
<td>218 ± 3</td>
<td>234 ± 3</td>
<td>139 ± 3</td>
<td>145 ± 4</td>
<td>3739 ± 228</td>
<td>3824 ± 232</td>
</tr>
<tr>
<td>P</td>
<td>134 ± 4</td>
<td>145 ± 2</td>
<td>128 ± 4</td>
<td>136 ± 5</td>
<td>2788 ± 166</td>
<td>2795 ± 104</td>
</tr>
<tr>
<td>A</td>
<td>275 ± 3</td>
<td>279 ± 4</td>
<td>151 ± 5</td>
<td>154 ± 3</td>
<td>4892 ± 542</td>
<td>5085 ± 337</td>
</tr>
<tr>
<td>P+A</td>
<td>158 ± 4</td>
<td>159 ± 3</td>
<td>140 ± 5</td>
<td>148 ± 4</td>
<td>2814 ± 86</td>
<td>2834 ± 52</td>
</tr>
<tr>
<td>200 N</td>
<td>175 ± 5*</td>
<td>190 ± 4*</td>
<td>145 ± 5</td>
<td>168 ± 5**</td>
<td>3594 ± 146</td>
<td>4236 ± 178*</td>
</tr>
<tr>
<td>P</td>
<td>137 ± 2</td>
<td>151 ± 4</td>
<td>134 ± 6</td>
<td>152 ± 5**</td>
<td>2898 ± 109</td>
<td>2930 ± 98</td>
</tr>
<tr>
<td>A</td>
<td>232 ± 8*</td>
<td>241 ± 4</td>
<td>149 ± 7</td>
<td>177 ± 6**</td>
<td>4219 ± 191</td>
<td>5345 ± 214*</td>
</tr>
<tr>
<td>P+A</td>
<td>159 ± 4</td>
<td>160 ± 2</td>
<td>142 ± 3</td>
<td>160 ± 2</td>
<td>2809 ± 113</td>
<td>2654 ± 93</td>
</tr>
<tr>
<td>300 N</td>
<td>171 ± 4*</td>
<td>194 ± 3*</td>
<td>134 ± 4</td>
<td>165 ± 3**</td>
<td>3743 ± 141</td>
<td>4377 ± 167*</td>
</tr>
<tr>
<td>P</td>
<td>133 ± 4</td>
<td>148 ± 3</td>
<td>133 ± 4</td>
<td>150 ± 3</td>
<td>2737 ± 182</td>
<td>2878 ± 177</td>
</tr>
<tr>
<td>A</td>
<td>241 ± 7*</td>
<td>248 ± 6</td>
<td>143 ± 5</td>
<td>164 ± 4*</td>
<td>4057 ± 220</td>
<td>4830 ± 202*</td>
</tr>
<tr>
<td>P+A</td>
<td>149 ± 5</td>
<td>155 ± 3</td>
<td>133 ± 4</td>
<td>153 ± 5*</td>
<td>2983 ± 241</td>
<td>3066 ± 144</td>
</tr>
<tr>
<td>400 N</td>
<td>147 ± 4**</td>
<td>180 ± 5**</td>
<td>140 ± 4</td>
<td>166 ± 2**</td>
<td>3319 ± 201</td>
<td>4167 ± 228*</td>
</tr>
<tr>
<td>P</td>
<td>115 ± 5**</td>
<td>125 ± 4**</td>
<td>127 ± 3</td>
<td>141 ± 2</td>
<td>2692 ± 127</td>
<td>2615 ± 159</td>
</tr>
<tr>
<td>A</td>
<td>222 ± 7**</td>
<td>240 ± 2**</td>
<td>147 ± 1</td>
<td>174 ± 2**</td>
<td>4508 ± 241</td>
<td>5381 ± 144*</td>
</tr>
<tr>
<td>P+A</td>
<td>150 ± 3</td>
<td>152 ± 2</td>
<td>135 ± 2</td>
<td>152 ± 3*</td>
<td>2788 ± 115</td>
<td>2809 ± 90</td>
</tr>
<tr>
<td>500 N</td>
<td>142 ± 2**</td>
<td>165 ± 3**</td>
<td>135 ± 2</td>
<td>159 ± 2**</td>
<td>3013 ± 147*</td>
<td>3945 ± 184*</td>
</tr>
<tr>
<td>P</td>
<td>119 ± 2*</td>
<td>138 ± 3</td>
<td>126 ± 1</td>
<td>143 ± 2*</td>
<td>2562 ± 98</td>
<td>2596 ± 102</td>
</tr>
<tr>
<td>A</td>
<td>210 ± 4**</td>
<td>224 ± 2**</td>
<td>148 ± 2</td>
<td>167 ± 3*</td>
<td>3710 ± 158*</td>
<td>4613 ± 196*</td>
</tr>
<tr>
<td>P+A</td>
<td>153 ± 3</td>
<td>154 ± 1</td>
<td>144 ± 3</td>
<td>164 ± 3*</td>
<td>2515 ± 112</td>
<td>2544 ± 126</td>
</tr>
</tbody>
</table>

Results are expressed as mean ± SE. N = normal, P = propranolol, A = atropine, P + A = propranolol + atropine.

* Response to exercise significant from pre-exercise values at P < 0.05. Multiple comparisons were made between 100 and 500 g training stages for each parameter. a Difference from 100 g training stage found to be significant (P < 0.001). b Difference from 300 g training stage found to be significant (P < 0.05). c Difference from 100 g training stage found to be significant (P < 0.05). d Difference from 400 g training stage found to be significant (P < 0.05).
LVSP throughout the training regimen when compared with respective pre-exercise values prior to drug intervention. After propranolol at the 100 g training stage, HR increased from 134 ± 4 to 145 ± 2 beats/min. A significant rise in HR was observed throughout the remainder of the training period, with the time course of the HR changes being similar to that observed before propranolol. LVSP exhibited a significant rise at the 200 g training stage from 134 ± 6 to 152 ± 5 mm Hg. The rise in LVSP in response to static exercise was maintained for the remainder of the training regimen. No change in LV max dp/dt was recorded at any of the training stages. Significant reductions in pre-exercise HR from 133 ± 4 to 113 ± 5 beats/min and peak exercise HR from 148 ± 3 to 125 ± 4 beats/min were observed after 11.3 ± 0.5 weeks of training when the cats were holding 400 g (Table 1; Fig. 2).

### Responses to Isometric Exercise after Atropine

Administration of atropine resulted in increases in pre-exercise values for HR and LV max dp/dt at each holding tension (Table 1). No significant change in any parameter was observed at the 100 g training stage after atropine. At the 200 g training stage, increases in both LVSP from 149 ± 7 mm Hg to 177 ± 6 mm Hg and LV max dp/dt from 4219 ± 191 mm Hg/sec to 5345 ± 214 mm Hg/sec were recorded. LVSP and LV max dp/dt continued to increase in response to isometric exercise at subsequent training levels. No change in HR was observed until the exercise subjects were holding 400 g for 15 seconds. HR increased from 222 ± 7 to 240 ± 2 beats/min. Peak HR was reached in the first 10 seconds of exercise and remained elevated throughout the exercise event. HR responses at the 500 g training level were similar to those obtained at the 400 g training level.

### Responses to Isometric Exercise after Atropine plus Propranolol

Throughout the training regimen after atropine and propranolol, no significant change was observed in HR and LV max dp/dt during isometric exercise (Table 1). No significant change was seen in LVSP at the 100 g training stage during exercise. At the 200 g training level, a significant increase in LVSP (+ 13%) was observed during isometric exercise. Similar increases in LVSP were recorded during exercise throughout the remainder of the training regimen. After combined blockade, the values of pre-exercise HR and peak exercise HR did not change from their initial values during the time course of the training regimen (Fig. 4).

### Discussion

During this study, increases in left ventricular systolic pressure in response to isometric exercise were first observed after approximately 2 weeks of training and were maintained throughout the remainder of the training period. The pressor response was found to persist after β-adrenergic receptor blockade with propranolol, although the magnitude of the change was reduced. The rise in pressure could be the result of increased cardiac...
output, increased peripheral resistance, or both. However, in most human subjects performing isometric exercise, the cardiac output response seems to predominate (Donald et al., 1967).

Following β-adrenergic receptor blockade with propranolol, several investigators (MacDonald et al., 1966; Martin et al., 1974) have observed that the arterial blood pressure still increases during isometric exercise despite pharmacological blockade of the cardiac output response. Martin et al. (1974) concluded that the increase in arterial pressure after propranolol was due to a compensatory increase in systemic vascular resistance, since no increase in cardiac output was observed during exercise after propranolol. In the anesthetized cat, increases in arterial pressure also have been seen during induced isometric exercise after propranolol, although they were reduced in magnitude when compared to exercise before propranolol (Mitchell et al., 1977). Crayton et al. (1979) found no change in arterial pressure during sustained contractions after propranolol in anesthetized dogs; however, he observed that, following propranolol administration, baseline total peripheral resistance increased dramatically. Furthermore, α-adrenergic receptor blockade with phenoxybenzamine or phentolamine attenuates the rise in arterial blood pressure in response to a static effort indicating partial mediation of the response by α-adrenergic receptors (Crayton et al., 1979; Freyschuss, 1970). It appears that an increase in systemic vascular resistance resulting in an elevation in blood pressure occurs even when the cardiac output response to isometric exercise is prevented pharmacologically. Attempts to effect an α-adrenergic receptor blockade in the conscious cat were unsuccessful.

Increases in LVSP during isometric exercise were accompanied by a significant rise in LV max dp/dt in the conscious cat. In several cats, mean left atrial pressure was measured periodically as an index of left ventricular end-diastolic pressure. Isometric exercise resulted in no significant change in mean left atrial pressure, while dp/dt increased. Other investigators (Fisher and Nutter, 1974; Grossman et al., 1973; Mitchell et al., 1977) have shown similar responses in humans, cats, and dogs, indicating that isometric exercise causes an increase in the contractile state of the left ventricle.

β-Adrenergic receptor blockade abolished the increase in LV max dp/dt in the conscious cat. Mitchell et al. (1977) also established that propranolol abolished the rise in LV max dp/dt during static contractions in the anesthetized cat, indicating mediation of this response by the sympathetic nervous system.

A characteristic feature of the responses to isometric exercise in the present study was a rapid increase in HR. The maximum HR response was reached in the initial 5 seconds of exercise, and then decreased to the pre-exercise level, whereas LVSP continued to rise. It was hypothesized that a major aspect of this HR response may be an anticipatory "overshoot" in expectation of food or exercise which elicits homeostatic mechanisms during exercise to return the HR to near pre-exercise levels (Diepstra et al., 1977).

β-Adrenergic receptor blockade with propranolol had little effect on the HR response curve, while vagal blockade with atropine abolished the HR response to isometric exercise at the initial training levels. This agrees with previous findings for human subjects in which the initial tachycardia during sustained handgrip was found to be mediated by withdrawal of vagal dominance (Borst et al., 1972; Freyschuss, 1970; Martin et al., 1974; Paulev, 1971). Experiments on anesthetized animals have demonstrated that the chronotropic response to isometric exercise can be abolished following propranolol administration (Mitchell et al., 1977; Crayton et al., 1979); however, it must be noted that anesthetic agents can markedly affect the cardiovascular system (Peiss and Manning, 1964; Price et al., 1965). Crayton et al. (1979) demonstrated a minimal effect of atropine administration on resting heart rate in his preparation. Thus, these anesthetized animal preparations may be characterized by a lack of significant vagal cardio-inhibitory activity so that the only mechanism available to increase HR in these animals is the sympathetic nervous system.

Martin et al. (1974) found that sympathetic stimulation was a secondary mechanism for increasing the HR in human subjects performing sustained handgrip, becoming operative only after the first mechanism of vagal withdrawal had been used. They observed that atropine was ineffective in blocking the HR response during the last 2.5 minutes of a 3-minute contraction, but combined atropine-propranolol blockade was effective in suppressing this portion of the response. In the present study, the high HR's following atropine administration would tend to mask any influence by the sym-

![Figure 4 Relationship between HR and holding tension after atropine plus propranolol.](http://circres.ahajournals.org/)
pathetic nervous system. However, at the latter training stages of the exercise program (400 and 500 g training stage), pre-exercise HR after atropine was reduced significantly from the previous training stages, and a small rise in HR was observed during exercise at these levels of training. The increase in HR after atropine was slower in onset when compared to the normal response and the response after propranolol. This correlates with the observation that there is a longer latency period (3-6 seconds) between sympathetic stimulation and cardiac acceleration than between parasympathetic withdrawal and tachycardia (Petro et al., 1970). Also, HR remained elevated and did not tend to return to pre-exercise levels during exercise, indicating mediation by the parasympathetic nervous system to correct for the “overshoot.” Combined autonomic blockade completely abolished the changes in HR.

The exercised cats on the progressive holding tension schedule exhibited significant reductions in pre-exercise HR and peak exercise HR after 2 weeks of training, prior to the development of significant pressor responses. Comparable changes were observed during the initial weeks of training in subjects on a regimen in which the holding tension was kept constant (20 g) throughout the exercise program. This initial reduction in HR probably represents a period of behavioral adaptation in which the animals adjust to the environment of the experimental apparatus and the daily routine of the training procedure. Autonomic blockade showed that this alteration was mediated by the sympathetic nervous system. A small but significant secondary reduction in preload HR and peak exercise HR was observed after approximately 11 to 12 weeks of training. This bradycardia was significant when compared to the HR’s of cats exercised at a constant load of only 20 g (Diepstra et al., 1977). To unmask the intrinsic HR, simultaneous sympathetic and parasympathetic blockade was effected with propranolol and atropine. No change in intrinsic F HR was observed in the animals during the training period, indicating a substantial reduction in the HR was entirely mediated by neural mechanisms.

Thus, the discrepancies that exist in the literature concerning the effector mechanisms responsible for the cardiovascular changes during isometric exercise seem to be related to the differing preparations, anesthetized vs. conscious. The autonomic mechanisms functioning to induce the cardiovascular changes accompanying isometric exercise in the conscious cat more closely resemble those elicited during static exercise in humans and, in this regard, the experimental model presently employed will be useful for future comparative studies.

Acknowledgments

We would like to extend our appreciation to Francine Gonyea and Mark Young for their technical assistance and to Glenda Graham, Roselyn Powell, Dianne Ralston, and Billie Price for their help in preparing the manuscript.

References

Alam M, Smirk FH (1957) Observations in man upon a blood pressure raising reflex arising from the voluntary muscles. J Physiol (Lond) 139: 372-383
Alam M, Smirk FH (1938) Observations in man on a pulse-reflex from the voluntary muscles of the legs. J Physiol (Lond) 92: 167-177
Humphreys PW, Lind AR (1963) The blood flow through active and inactive muscles of the forearm during sustained handgrip contractions. J Physiol (Lond) 160: 120-131
Cardiovascular response to static exercise during selective autonomic blockade in the conscious cat.

G Diepstra, W Gonyea and J H Mitchell

*Circ Res.* 1980;47:530-535
doi: 10.1161/01.RES.47.4.530

*Circulation Research* is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1980 American Heart Association, Inc. All rights reserved.
Print ISSN: 0009-7330. Online ISSN: 1524-4571

The online version of this article, along with updated information and services, is located on the World Wide Web at:

http://circres.ahajournals.org/content/47/4/530

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in *Circulation Research* 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 Research* is online at:
http://circres.ahajournals.org/subscriptions/