Fluid Restitution and Blood Volume Redistribution in Anesthetized Rabbits in Response to Vasoactive Drugs

Susan M. Anderson, BM, FCAnaesa; George F. Rich, MD, PhD;
Christine Roos, MS; L.P. Lee, PhD; J.S. Lee, PhD

**Background** Vasoactive drugs could alter the fluid restitution from the tissue and redistribute blood volume between the macrocirculation and microcirculation.

**Methods and Results** With bolus injections of vasoactive drugs in anesthetized rabbits, we measured the changes in blood and plasma density for the determination of the volume of restitution and redistribution. Epinephrine 3.5 µg/kg caused a fluid loss to the tissue, leading to a transient decrease in total blood volume by 2.30 mL/kg. Because of blood volume redistribution, the peak volume reduction was accompanied by a volume reduction of 0.81 mL/kg from the macrocirculation and 1.49 mL/kg from the microcirculation. Phenylephrine 70 µg/kg caused a peak reduction in total blood volume of 1.40 mL/kg (with 0.41 mL/kg from macrocirculation and 0.99 mL/kg from microcirculation). Nitroprusside 7 µg/kg increased the blood volume by 1.44 mL/kg (0.83 mL/kg macro and 0.61 mL/kg micro), nitroglycerin 7 µg/kg by 1.48 mL/kg (0.97 mL/kg macro and 0.51 mL/kg micro), and isoproterenol 7 µg/kg by 2.07 mL/kg (0.68 mL/kg macro and 1.39 mL/kg micro). All plasma (or blood) density changes measured for the five drugs (with epinephrine, phenylephrine, and nitroprusside done over a wide dosage range) correlated linearly with the drug-induced changes in arterial pressures.

**Conclusions** These results indicate that vasoactive drugs alter total blood volume and the volume of microcirculation and macrocirculation. (Circulation. 1994;90:509-514.)

**Key Words** • blood volume • microcirculation • nitroglycerin • vasoconstriction • vasodilation

Vasoactive drugs constrict or dilate blood vessels. The change in the microvascular pressure could alter the fluid being restored or filtered between the blood and tissue compartments and therefore change the total blood volume. Fluid restitution in response to vasoactive drugs has been assessed by measurement of the slow changes in organ weight or altered radionuclide counts of specific organs. We sought to quantify the total contribution in fluid restitution from all organs using a density-measuring technique. The overall pressor effect of drugs on total blood volume has been studied by measurement of the blood volume transferred from the circulation to an extracorporeal reservoir. However, because fluid restitution is part of the blood volume transfer to the reservoir, restitution needs to be subtracted to quantify the actual blood volume shift for a more accurate assessment of the effect of drugs on the circulation.

The microcirculation is defined as the part of the circulation composed of microvessels with a diameter <200 µm because of the presence of the Fahraeus effect. By this definition, the microvascular volume is comparable to the macrovascular volume. Recent microvascular measurements indicate that a significant shift of blood volume out of the microcirculation is produced by hemorrhage in the rat spinotrapezius muscle or is activated by bilateral carotid sinus occlusion in the rat intestinal microvascular bed. Nitroprusside and nitroglycerin dilate the microcirculation of striated muscle in dorsal skin folds of hamsters during drug-induced hypotension. By measuring the changes in plasma and blood density with consideration of the Fahraeus effect, LaForte et al estimated, for anesthetized rabbits subjected to a 10% hemorrhage (or 7.1 mL/kg), that 5% of the blood volume (or 3.5 mL/kg) is shifted from microcirculation to macrocirculation, while 0.5% of the volume (or 0.4 mL/kg) is restored from the tissue. This indicates the importance of microvascular volume shift in minimizing the effect of hemorrhage on macrocirculation, since its volume is reduced by 3.2 mL/kg instead of 7.1 mL/kg. In this investigation, we used this density technique to quantify the volume of fluid restitution and blood volume redistribution produced by the intravenous bolus injections of epinephrine, phenylephrine, nitroprusside, nitroglycerin, and isoproterenol in anesthetized rabbits. Our objective is to assess the pressor effect of these drugs on the total blood volume, microvascular volume, and macrovascular volume for a better understanding of their roles in blood volume regulation.

**Surgery**

The surgery and protocol, which followed the National Institutes of Health guidelines on animal experimentation, were approved by the Animal Research Committee at the University of Virginia. New Zealand White male rabbits weighing 2.7 to 3.4 kg were sedated with chlorpromazine (6.7 mg/kg IM) and anesthetized with sodium pentobarbital (20 mg/kg IV). Anesthesia was maintained with incremental sodium pentobarbital (1.5 mg/kg) every hour. After subcutane-
ous infiltration with lidocaine 1%, the trachea, right common carotid, left femoral artery, and left jugular vein were isolated. A cannula was inserted through a tracheostomy, and the rabbit was allowed to ventilate spontaneously. A polyethylene cannula was inserted 5 cm into the carotid artery, and heparin (5000 U) was injected. The remaining isolated vessels were cannulated in a similar manner. The blood was withdrawn via the right common carotid artery by a roller pump (Masterflex) at a rate of 20 mL/min, passed through a density meter (DMA 620W, Parr), and returned to the rabbit via the left jugular cannula. Another jugular cannula was connected to a Statham pressure transducer and a femoral artery cannula to another transducer. The blood density, arterial pressure, and venous pressure were continuously recorded on a Hewlett Packard 7700 series monitor. From the latter two measurements, the mean arterial pressure (MAP) and mean venous pressure were calculated. The former was about 75 to 80 mm Hg, which, because of anesthesia, is lower than that of conscious rabbits. With a similar density meter, plasma density was measured discretely from centrifuged blood samples (1 mL) taken before the injection of the drug, during the peak blood density, and 3 minutes after.

Protocol

After a period of stabilization from instrumentation (approximately half an hour), we initiated the study on the effects of a bolus of a vasoactive drug on arterial pressure, blood density, and plasma density. A series of syrings was prepared, each containing 0.1 to 0.2 mL of saline and having one of the following dosages: epinephrine 3.5 μg/kg, nitroglycerin 7 μg/kg, phenylephrine 70 μg/kg, sodium nitroprusside 7 μg/kg, or isoproterenol 7 μg/kg. The drug was injected via a needle inserted through a rubber cap to the midstream of the jugular venous cannula. In a random series, each injection was separated by 15 minutes to allow the blood density and arterial pressure to return to baseline. For a total blood volume of 200 mL, the lower density of injected saline decreased the blood density by 0.04 g/L and plasma density by 0.02 g/L. Both changes are negligible in comparison with the observed peak changes.

Dose-response curves for epinephrine (0.5 to 25 μg/kg) and nitroprusside (0.7 to 30 μg/kg) were obtained from four rabbits. For experiments with the injection dose of 3.5 μg/kg (epinephrine) or 7 μg/kg (nitroprusside), a bolus of isotonic saline was also injected into the jugular catheter at a time preceding the drug injection and then around the time of peak MAP change. The resulting transient density decrease in arterial blood was used to calculate cardiac output. With the change in MAP and mean central venous pressure, the total peripheral resistances preinjection and postinjection were estimated. Two of these rabbits were also splenectomized, followed by the measurement of density changes due to bolus injections of epinephrine (3.5 μg/kg) and nitroprusside (7 μg/kg). For four other rabbits, phenylephrine (8 to 320 μg/kg) was injected to obtain its dose-response curve.

Data Analysis

Two-sided Student's t test was used to determine statistical significance for density and pressure changes. The mean change in plasma density for each drug was used to calculate the change in the total blood volume (∆Vₖ) induced by fluid restitution from the preinjection time to the time around the peak MAP and blood density. The fluid restitution and a shift of blood volume from the low-hematocrit microcirculation can alter the arterial hematocrit. Once the change in arterial hematocrit is calculated from the mean changes in plasma and blood density, we then determine the change in microvascular volume ∆Vₘₑ. In the “Appendix,” we summarize the equations used for the calculation. More details are given in the paper by LaForté et al12 and the supplement filed with National Auxiliary Publication Service.13 The blood volume redistributions were computed from a total blood volume of 71 mL/kg.14 Finally, the concurrent change in the macrovascular volume, ∆Vₘₑ, is identified as ∆Vₖ – ∆Vₘₑ.

Results

Continuous tracings of blood density and arterial pressure changes after a 10-μg bolus of epinephrine are shown in Fig 1. The arterial pressure increased over a period of 40 seconds to a peak value and then returned to the baseline value. There was initially a transient density decrease caused by the injection of the lower-density saline. This was followed by a maximum density peak reflecting the summation of fluid restitution and microvascular volume shift of the intact animal to epinephrine. The discrete measurements of plasma density had transient changes similar to those of blood density.

The peak changes in MAP, blood density, and plasma density resulting from injection of each vasoactive drug are shown in Table 1. Epinephrine 3.5 μg/kg (n=19) significantly increased MAP (P<.01), blood density (P<.01), and plasma density (P<.01). The change in arterial hematocrit calculated from these two density changes and Equation 3 showed an increase of 1.1% over the preinjection arterial hematocrit of 36.3%. Phenylephrine 70 μg/kg (n=13) produced similar significant changes (P<.01 for all variables). Sodium nitroprusside 7 μg/kg bolus (n=18) significantly decreased arterial pressure, blood density, and plasma density (P<.01 for all variables). The two density decreases corresponded to a reduction of 0.8% in the arterial hematocrit. Likewise, 7 μg/kg nitroglycerin (n=10) or isoproterenol (n=6) showed similar changes and significance. The changes in MAP and blood density for epinephrine and phenylephrine were significantly dif-
isoproterenol decrease the blood volume; and isoproterenol, change in plasma density. Values are mean±SEM.

*The drug dosage was 3.5 \( \mu \)g/kg for epinephrine, 70 \( \mu \)g/kg for phenylephrine, and 7 \( \mu \)g/kg for others. A positive value in \( \Delta P \), for example, corresponds to a decrease in pressure (the preinjection time) to the time of peak pressure. The changes reported here indicate that these dosages significantly alter the mean arterial pressure and blood and plasma densities from control, with \( P<.01 \).

Different from each other and from the other drugs (\( P<.01 \)), MAP and blood density changes were not significantly different between nitroprusside, nitroglycerin, and isoproterenol. Plasma density changes were different between epinephrine and phenylephrine (\( P<.05 \)) and the other drugs (\( P<.01 \)). The plasma density changes with nitroprusside, nitroglycerin, and isoproterenol were not significantly different.

The peak changes in the total blood volume and macrovascular and microvascular volumes calculated from the peak changes in blood and plasma densities for the five vasoactive drugs at a given dose are presented in Table 2. An intravenous bolus of epinephrine caused a loss of fluid from the blood compartment with a reduction in macrovascular and microvascular volumes. Concurrent with the volumetric redistribution, the cardiac output increased from the preinjection level by 52±8\% during peak pressure change (n=4, \( P<.01 \)), and the total peripheral resistance decreased by 6±3\% (insignificant). Phenylephrine also caused fluid movement from the blood compartment, but to a lesser degree.

Sodium nitroprusside caused fluid movement from the tissue to the blood compartment. The total blood volume increased, as did the macrovascular and microvascular volumes. The cardiac output during peak MAP was 25±10\% (n=4 rabbits, \( P<.05 \)) lower than the preinjection value. Because of the larger decrease in MAP, the total peripheral resistance was decreased by 16±6\% (\( P<.05 \)). Intravenous nitroglycerin injection also caused an increase in total blood volume, although mostly in the macrocirculation, with smaller effects on the microcirculation. Isoproterenol caused an increase in total blood volume with expansion for both macrocirculation and microcirculation.

The changes in blood and plasma density with all vasoactive drugs and dosages are plotted against the change in MAP in Fig 2. The regression coefficient (\( r=.873 \)) for plasma density changes with respect to pressure changes is linear. Likewise, there was a strong linear relation (\( r=.938 \)) between blood density and MAP. The dose-response curves for blood and plasma density for epinephrine, phenylephrine, and nitroprusside are presented in Fig 3.

**Discussion**

Epinephrine and phenylephrine are vasconstrictors that increase MAP. Our results indicate that these drugs produce a transient increase in blood density, plasma density, and hematocrit. As the drug effect subsides, these quantities return to their preinjection values, indicating recovery to the original blood volume. The time course of hematocrit change after a bolus injection of epinephrine is comparable to that obtained by Hannon et al\textsuperscript{15} in splenectomized pigs. When a
higher dose of epinephrine is injected, we observe that the time for the density to decay back to its control value is longer. For epinephrine and phenylephrine, the transient increase in plasma density is caused by fluid filtration to the tissue, which decreases the total blood volume. The volume decrease is in agreement with the observation that epinephrine injection initiates a water efflux in the dog forelimb. Our analysis of the changes in blood and plasma densities indicates that about one third of the reduction in total blood volume comes from the macrocirculation and two thirds from the microcirculation.

Intravenous bolus injection of sodium nitroprusside, nitroglycerin, or isoproterenol produced a transient decrease in MAP, blood density, plasma density, and arterial hematocrit. Our analysis of plasma density change indicates that total blood volume increases as do the corresponding macrovascular and microvascular volumes. The dilation of microcirculation is comparable to the observed dilation of arterioles, precapillaries, and venules of skin fold during the infusion of nitroprusside and nitroglycerin in hamsters. Because of the microvascular dilation, the volume change of the microcirculation is less than the volume change of the entire circulation. In contrast, the pulmonary intravascular volume in dogs has been found to decrease after a low-dose infusion of nitroglycerin. Our results on the changes in the total blood volume suggest that if pulmonary blood volume is decreased, it is overwhelmed by the volume expansion in other organs such that the overall effect of nitroglycerin is an increase in total blood volume.

Our results indicate that epinephrine increases the peak MAP and peak cardiac output without significant change in the total peripheral resistance and venous pressure. The plasma density change relates almost linearly with the change in total blood volume (Equation 3). If the blood volume change is due to a change in transcapillary fluid flux but not an alteration in hydraulic conductance, then the Starling hypothesis and the correlation between the plasma density change and peak change in MAP indicate that the microcirculatory pressure is increased by epinephrine injection. Although epinephrine could induce different response in the preresistance and postresistance, the increase in MAP induced by epinephrine may be the primary factor producing the predicted overall increase in microcirculatory pressure. A similar implication applies to the vasoconstrictor phenylephrine. For the vasodilators nitroprusside, nitroglycerin, and isoproterenol, the decrease in MAP may be the factor producing a decrease in microcirculatory pressure and subsequently a reduction in fluid filtration to the tissue. If there is no change in the lymphatic flux, the total blood volume experiences an increase.

The plasma density change at about 1.5 minutes after the injection of epinephrine (3.5 μg/kg) indicates a decrease in total blood volume by 2.30 mL/kg. A constant transcapillary flux of about 1.5 to 3 mL/
(min · kg) during this period could produce the blood volume change. This large flux can be produced in view of the observations that (1) the normal lymphatic flow is about 0.05 to 0.1 mL/(min · kg), (2) the interstitial fluid pressure and lymphatic activity can increase the lymphatic flux by 10 to 50 times, (3) the transcapillary fluid flux at the arterial ends of the capillaries can be 10 times higher than the lymphatic flux, (4) the hydraulic conductances of various organs range from 0.1 to 1 mL/(min · mm Hg · kg), and (5) the peak MAP (113 mm Hg) is 50% higher than the preinjection MAP (75 mm Hg). Similar consistency is obtained for the blood volume changes estimated for other vasoactive drugs.

For some organs, vasoconstrictors such as epinephrine produce vasodilation. However, our result of a net decrease in total microvascular volume suggests that vasoconstriction induced directly by epinephrine overwhelms these two vasodilation effects. The finding of a net increase in microvascular volume for nitroprusside, nitroglycerin, and isoproterenol may also imply that their vasodilation effect predominates.

The change in plasma density after the injection of epinephrine (Table 1) yields a 1% increase in the whole-body hematocrit (from 29.8% to 30.8% as calculated from Equation 1). Since the shift of blood volume from microcirculation to macrocirculation does not change the total blood volume and total red blood cell volume, the change in microvascular volume will not alter the whole-body hematocrit. The calculation from Equation 2 shows that the postinjection $F_{\text{cell}}$ is 0.822, a value close to the preinjection value of 0.82. If there is no change in microvascular volume, the $F_{\text{cell}}$ ratio is calculated as 0.814. For phenylephrine injection (70 µg/kg), the whole-body hematocrit increased by 0.6% and the $F_{\text{cell}}$ ratio by 0.002. For the vasodilator results listed in Table 1, the whole-body hematocrit decreased by 0.6% to 0.9%, and the postinjection $F_{\text{cell}}$ ratio differs from the preinjection ratio by a value <0.002.

The vasoactive drugs were injected randomly and after the pressure and blood density had returned to their control values to ensure that the measurements were independent. The independence is supported by the linear relation found between the pressure and density changes for all drugs combined and the consistency of the dose-response curves. The calculation of fluid restitution and microvascular volume shift from the density changes is based on three assumptions: (1) the density of restored fluid takes the density of lymphatic fluid from muscle bed, (2) the microvascular hematocrit changes in proportion to arterial hematocrit change, and (3) the contribution from the small rabbit spleen is negligible. The discussion given in Appendix A of Reference 13 supports the use of the equations deduced from these assumptions to calculate the volumetric changes in the rabbit or splenectomized animals.

When vasoactive drugs are injected, the change in microcirculatory pressure may alter the protein content of the filtrate, eg, by an increase of 20%. This is equivalent to a change of the filtrate density from 1005 to 1006 g/L. For this higher density, the new estimate of $\Delta V_i$ is 8% lower than the previous estimate, a negligible error. It is known that epinephrine and isoproterenol cause contraction of the spleen. If this significantly changed the hematocrit of rabbits, we would see an increase in hematocrit for both drugs. Instead, our calculations indicate that epinephrine increased the hematocrit, whereas isoproterenol decreased hematocrit. In two splenectomized rabbits, the peak variations in plasma and blood density after epinephrine or nitroprusside injection were not different from the control values. We conclude that the contribution of the rabbit spleen is negligible.

We have demonstrated that the correlation between the microvascular volume shift and the hemorrhaged volume is similar for anesthetized and conscious rabbits. Since the vasoactive drugs will have similar effects on blood pressure for anesthetized and conscious rabbits, it is likely that the effects of these drugs on the blood volume redistribution found here may be similar for the conscious rabbits. The present density protocol has the potential to study the response of vasoactive drugs in conscious rabbits.

Ogilvie and Zborowska-Bliski assessed the vascular capacitance of splenectomized dogs and observed that nitroglycerin and nitroprusside increase the “unstressed” vascular volume. The present study suggests that their baseline measurement corresponds to a volume expanded by the fluid restitution, which could alter their estimate of the unstressed vascular blood volume. The measurement of the transfer of blood volume out of the circulation caused by the infusion of epinephrine at 1.2 µg/(kg · min) indicates that the unstressed vascular volume is changed by 8.02 mL/kg. For a bolus injection of epinephrine at 3.5 µg/kg, we found that the peak reduction in total blood volume, which resulted from a negative fluid restitution, was 2.3 mL/kg. Although the experimental protocols to obtain these two volume changes are different, their comparable values indicate that fluid restitution needs to be measured and corrected for accurate assessment of vascular capacitance by the blood volume transfer technique.

The microvascular volume shift produced by the drug injections ranged from 30% to 70% of the peak changes in total blood volume (Table 2). These results indicate that the microvascular volume shift is a significant part of the blood volume transferred from the circulation to the extracorporeal reservoir and that the microvascular compliance is an important component of the total vascular compliance. Assessment of the diameter changes of microvessels in the rat spinotrapezius muscle and intestine indicates that 57% to 80% of the microvascular volume shift originates from the venules. Accordingly, the venules could be the major contributor of the microvascular volume shift found by the present study.

In summary, blood and plasma density changes occur after intravenous administration of vasoactive drugs in anesthetized rabbits. Calculations from the blood and plasma density changes indicate that epinephrine and phenylephrine cause a transient loss of fluid from the blood compartment, leading to a reduction in total blood volume. The reduction occurs concurrently with a reduction in the macrovascular and microvascular volumes. In contrast, nitroprusside, nitroglycerin, and isoproterenol induce a fluid restitution and microvascular volume shift to produce a transient increase in total blood volume and microvascular and macrovascular volumes. These results indicate that the volumetric changes are an important pressor effect of these vasoactive drugs on the cardiovascular system.
Appendix

Equations to Calculate Changes in Total Blood Volume and Microvascular and Macrovacular Blood Volumes

The equations used for the calculation and the rationale for their derivation are summarized here. Let the preinjection blood volume of all microvessels having a diameter <200 μm and flowing with blood be $V_{\text{mic}}$. The tube hematocrit for vessels of this size is smaller than the arterial hematocrit, $H_a$. With the volume average of the tube hematocrits of these microvessels identified as $H_{\text{mic}}$, the total red blood cell (RBC) volume is expressed as $H_{\text{mic}} \cdot V_{\text{mic}}$. Let the preinjection total blood volume be $V_b$. The macrovascular volume $V_{\text{mac}}$ is taken as $V_b - V_{\text{mic}}$. The blood flowing in the large vessels in the macrocirculation has the hematocrit $H_a$. According to the definition of the whole-body hematocrit $H_w$ and the $F_{\text{cell}}$ ratio ($=H_w/H_a$), one can derive, with $\alpha$ as $H_{\text{mic}}/H_a$, these two relations:

$$H_w = H_a - (H_a - H_{\text{mic}})V_{\text{mic}}/V_b$$

and

$$F_{\text{cell}} = 1 - (1 - \alpha)V_{\text{mic}}/V_b$$

Fluid is restored from the tissue because of drug injections. Let the volume expansion to the time of peak blood density be $\Delta V_s$ and the fluid density be $\rho_f$. If the density is different from the preinjection plasma density ($\rho_p$), the fluid restitution changes it to $\rho_p'$, with the prime representing the time of peak blood density. The consideration of mass balance for this mixing process yields the following equation to calculate $\Delta V_b$ from the change in plasma density:

$$\Delta V_b/V_b = (1 - H_a)(\rho_p - \rho_p')/(\rho_p' - \rho_f)$$

The drug injection does not change the osmotic pressure of plasma. Thus, it is reasonable to regard the RBC density $\rho_r$ as constant. Through the use of the dependence of $\rho_r$ on $\rho_p$, $\rho_f$, and $H_a$, one can calculate the change in the arterial hematocrit imposed by the drug injection as

$$\Delta H_a = H_a[(\rho_r - \rho_p') - (\rho_r - \rho_f')(1 - H_a)]/[(\rho_r - \rho_f) + (\rho_r - \rho_p')(1 - H_a)]$$

Both the fluid restitution (Equation 3) and the shift of blood from microcirculation to macrocirculation contribute to the arterial hematocrit change in Equation 4. The consideration of no change in the total circulating RBC volume leads to the following equation to calculate the microvascular volume change:

$$\Delta V_{\text{mic}}/V_b = -(\Delta H_a/H_a)F_{\text{cell}} + \Delta V_s/V_b)/(1 - \alpha)$$

These five equations are Equations 2, 3, 21, 23, and 26 in the supplement (see Reference 13) once $\Delta V_s$ is identified as $\Delta V_s$, $\Delta V_{\text{mic}}$, and $\Delta V_{\text{mac}}$, and the shed blood volume $\Delta V$ as 0.

To carry out the calculation, we take $F_{\text{cell}}$ of the rabbit as 0.82, $\alpha$ as 0.64, and $\rho_f$ as 1005 g/L. The former two values correspond to a microvascular volume 50% of the total blood volume. A discussion on the following four points is detailed in References 12 and 14 and the supplement. First are the morphometric data and direct microvascular evidence supporting the use of these values. Then, because of the short recirculation time for the rabbit, we show that Equations 3, 4, and 5 are applicable to the present quasi-steady experiments. Third, a sensitivity analysis is made using other likely values for $F_{\text{cell}}$, $\alpha$, and $\rho_f$ to demonstrate minor corrections in volume calculation. Fourth, the role of splanchic beds, the negligible contribution from the small rabbit spleen, and the implication of tissue hematocrit measurement on the interrelation between microvascular volume shift and arterial hematocrit change are examined.

Acknowledgment

This study is supported in part by grant HL-40893 from the National Heart, Lung, and Blood Institute.

References

Fluid restitution and blood volume redistribution in anesthetized rabbits in response to vasoactive drugs.
S M Anderson, G F Rich, C Roos, L P Lee and J S Lee

Circulation. 1994;90:509-514
doi: 10.1161/01.CIR.90.1.509

Circulation is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1994 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/90/1/509

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/