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Microcirculatory Basis for Nonuniform Flow Delivery with Intravenous Nitroprusside

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Background: **The purpose of this study was to determine the effects of systemic infusions of nitroglycerin and sodium nitroprusside on flow distribution and wall shear stress in the microcirculation.**

Methods: **With university approval, the cremaster muscle of 28 anesthetized (70 mg/kg pentobarbital given intraperitoneally) hamsters (Harlan Sprague Dawley: Syrian; weight, 121 ± 11 g [mean ± SD]) was observed using *in vivo* fluorescence microscopy. Arteriolar diameter, erythrocyte flux, and velocity were measured for a feed arteriole and its sequential branches. Observations were made during control (mean arterial pressure, 88 ± 4 mmHg) and after 30 min of intravenous delivery of sodium nitroprusside or nitroglycerin, titrated to decrease mean arterial pressure by 20 mmHg.**

Results: **Sodium nitroprusside significantly dilated select upstream portions of the network (23 ± 2.6 to 29 ± 2.6 μm); no arterioles were dilated with nitroglycerin. Erythrocyte flux into the feed (*i.e.*, inflow into the arteriolar network) and into the sequential branches (*i.e.*, distribution within the network) were evaluated. With nitroglycerin, inflow decreased significantly from 1,560 ± 335 to 855 ± 171 cells/s, and flux into the branches decreased evenly. With sodium nitroprusside, inflow increased significantly to 2,600 ± 918 cells/s, yet cells were "stolen" from upstream branches (a decrease from 425 ± 67 to 309 ± 87 cells/s in the first branch). Excess flow passed into a downstream "thorough-fare channel," significantly increasing flux from 347 ± 111 to 761 ± 246 cells/s. Wall shear stress decreased uniformly with nitroglycerin infusion, with a decrease in the feed from 8.8 ± 2.5 to 6 ± 1.7 dyn/cm^2. With sodium nitroprusside, variable changes occurred that were location specific within the network. For instance, at the inflow point to the network, wall shear stress changed from 8.3 ± 2.5 to 4.2 ± 3.3 dyn/cm^2.**

Conclusions: **Nitroglycerin infusion promoted homogeneity of flow. Sodium nitroprusside significantly increased the heterogeneity of flow within this arteriolar network; an anatomic location for steal induced by sodium nitroprusside is identified. (Key words: Arteriole; autoregulation; steal phenomenon; wall shear stress.)**

NITROGLYCERIN and sodium nitroprusside have been used commonly as vasodilators for decades. These nitric oxide donors provide an undeniable benefit for a range of cardiovascular disease states, such as congestive heart failure, and are used to control blood pressure in hypertensive patients and to induce hypotension during surgical procedures.[1–5] The systemic effects of these nitrovasodilators are well documented to include decreased preload with nitroglycerin and decreased afterload with nitroprusside. Although these systemic effects are well known and accepted in the clinical community, knowledge of the effect on microcirculatory flow and resistance is relatively sparse (for example, see Enrich *et al.*,[6] Longnecker *et al.*,[7] Auer,[8] and Endrich *et al.*[9]), or inconsistent (for example, see Ovadia-Tirosh *et al.* [10] and Hauss *et al.*[11]). In general, with nitroglycerin, organ flow is preserved, whereas with nitroprusside there is increased heterogeneity of flow ("steal"). This may be the result of differing systemic mechanisms by which these agents decrease mean arterial pressure or affect baroreflexes.[5] However, direct evidence exists that microvessels of different sizes respond differently to directly applied nitroglycerin, with those smaller than 50 μm dilating only transiently and then becoming overridden by autoregulation[12,13] or not dilating at all[14] (see also Harrison and Bates[4]). Furthermore, the steal phenomenon with nitroprusside is very likely a microcirculatory effect.[6,15]

However, despite decreased tissue oxygenation with nitroprusside,[6,10,11] there is an equivalent dilation for large

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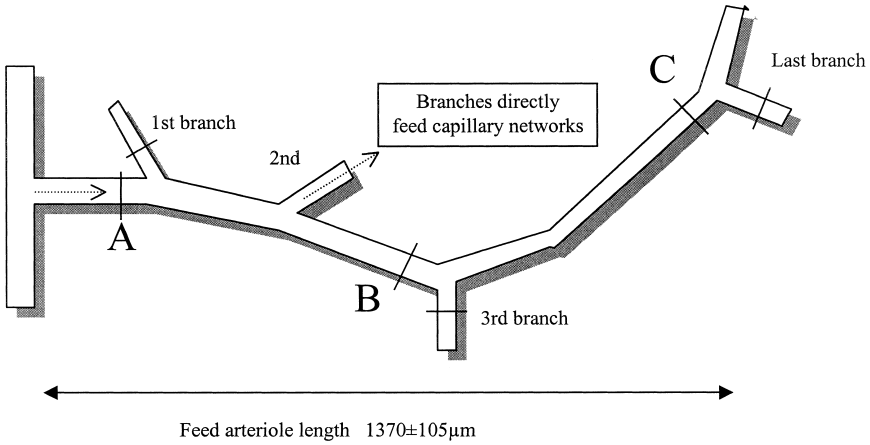

Fig. 1. The experimental test site, which is a third-order transverse arteriole and its sequential fourth-order branches. Observations were made at the feed (points A, B, and C) and at the sequential branches (first, third, and last). Each branch arteriole regulates the inflow to a separate capillary network; flow into the branch arterioles is organized and can be regulated by physiologic stimuli. The hemodynamic characteristics of this site are well described (see text). The total feed vessel length is given as the mean ± SEM (n = 16 animals).

and small microvessels.[14] The cause of the steal phenomenon and its extent into the peripheral bed are unknown. The current microcirculatory data show that changes in vessel diameter do not by themselves tell how the flow is altered in intact vascular networks.[16–19] A study examining both diameter and flow changes is required.

Extensive work has identified and characterized the flow within a specific arteriolar network in the microvasculature of the hamster cremaster striated muscle preparation.[20–25] It is clear that the behavior of the downstream arteriolar branches is highly coordinated with the actions of the upstream branches, with distinct differences in arteriolar network behavior for metabolic versus adrenergic stimuli.[26] In addition, there is no coordinated behavior when the endothelial-dependent dilatory pathways (*i.e.*, nitric oxide) are blocked. Thus, endogenous nitric oxide plays a role in coordinating the arteriolar network behavior. Furthermore, flow-dependent responses in the microcirculation tightly link endogenous nitric oxide to the prevailing wall shear stress (for example, see Koller and Kaley[27]). Our question is whether intravenous nitric oxide donors will induce coordinated microvascular responses. We have evaluated the effect of intravenous administration of these nitrovasodilators on microcirculatory diameter, flow, and wall shear stress within an intact arteriolar network.

Materials and Methods

Preparation

Sixteen adult male golden hamsters (Harlan Sprague Dawley: Syrian; age, 80 ± 5 days old; weight, 121 ± 11 g [mean ± SD]) were anesthetized with 70 mg/kg pentobarbital sodium given intraperitoneally, had tracheostomies, and were maintained with a constant infusion of 10 mg/ml pentobarbital sodium at a rate of 0.56 ml/h to replace respiratory fluid losses. The systemic hematocrit was not different before (56 ± 1%) or after (55 ± 1%) the preparation. Deep-body temperature was monitored throughout the experiment and maintained between 37°C and 38°C. Mean arterial pressure (femoral) was monitored throughout the experiment. The right cremaster muscle was prepared for *in vivo* microcirculatory observations.[23,26] The preparation was superfused continuously with bicarbonate-buffered physiologic salt solution (control superfusate) containing 132 mM NaCl, 4.7 mM KCl, 2 mM $CaCl_2$, 1.2 mM $MgSO_4$, 20 mM $NaHCO_3$ (equilibrated with 5% carbon dioxide$_{(gas)}$, 95% $N_{2(gas)}$; *p*H 7.4 ± 0.5 at 34°C). Erythrocytes from age- and weight-matched animals (82 ± 6 days old, 124 ± 11 g, n = 12) were labeled with substituted tetramethyl rhodamine isothiocyanate (XRITC cells; Molecular Probes, Eugene, OR) according to an established protocol.[28,29] These were used to measure blood flow parameters.

Observation Site

Observations were made along a third-order transverse (feed) arteriole and its fourth-order branch arterioles (fig. 1). The site was located in each preparation as described previously.[18,30] This site has a well-described architecture and flow behavior.[26,31] Each branch controls flow into adjacent capillary networks in this muscle.[21,22,32] Furthermore, the organization and behavior of this site suggests that it is a functional unit, which is repeated across this muscle tissue. Thus, by studying this group of arterioles, we describe flow characteristics of the tissue.

Experimental Protocol

During the 60-min stabilization period, the fluorescently labeled erythrocytes were injected into the hamster, and the presence of vasoactive tone was confirmed by dilation to topical application of 10^{-4} M adenosine and constriction to 5% oxygen bubbled into the suffusate solution. The test site was videotaped after stabilization (control), and then drug infusion was begun immediately. The site was videotaped again at the end of the 30-min infusion of nitroglycerin or sodium nitroprusside. The drugs were titrated to decrease the hamsters' mean arterial pressure by 20 mmHg; the infusion rates are given in Results. So that our results were not biased by time-dependent changes, videotaping periods were kept short (approximately 3 min), and for alternate experiments we began videotaping at the first or last branch point. One drug was tested per animal; drugs were assigned randomly to the animals.

Measurements

The diameter (in micrometers), fluorescent erythrocyte flux (cells/s), and velocity (μm/s) were measured off-line from the videotaped image using a calibrated video-caliper system and a software computer program developed specifically for this application (in the Department of Anesthesiology of our institution). These parameters were measured for the feed vessel and the first, third, and last branches, as indicated in figure 1, to examine hemodynamic changes into and within the arteriolar network. Hemodynamic parameters were calculated as described previously.[18] Briefly, cell flux, F cells/s, is calculated according to the formula $F = (m_t/p)/t$, where m_t is the number of fluorescent cells crossing a specified vessel plane in time, t, and p is the fraction of fluorescent cells in the total erythrocyte population. Fluorescent cells were considered to represent the total population.[28,29] Individual velocities, expressed in micrometers per second, were measured as the distance traveled in one video field (1/60th s) for all fluorescent cells crossing the specified sampling plane during a 30-s time interval. The mean axial cell velocity, v_c, was calculated from their harmonic mean, and v_c was used to approximate the average velocity of blood in the vessel.[33] Hematocrit, the time-averaged volume fraction of cells in the vessel, was calculated as $H = F \cdot V_c/v_c \cdot \pi r^2)$, where r is the vessel radius and V_c is the mean corpuscular volume (58×10^{-12} ml). The apparent viscosity, η_{app}, was calculated from the relation between vessel hematocrit, vessel diameter, and relative viscosity.[34] The shear rate, γs^{-1}, was calculated as $\gamma = 8 \cdot v_c/D$. Wall shear stress, $T\omega$ dynes/cm^2, was calculated as $T\omega = \eta_{app} \cdot \gamma$.

Statistical Analyses

The calculated values were pooled by feed segment or branch location and test condition to determine the population means and standard errors. Group comparisons were made between control and test conditions for the feed arteriole segments by analysis of variance, and differences were evaluated with paired Student *t* tests. There were no differences between the control measurements for the two groups. For the branch arterioles, changes from control were calculated as test − control. For all analyses, differences were considered significant when $P \leq 0.05$; $0.05 \leq P \leq 0.1$ are given in the text or legends. Changes in variability within populations were evaluated using the coefficient of variation (CV = SD/mean)[35] and the Moses test for dispersion.[36]

Results

Mean arterial pressure was 88 ± 4 mmHg (mean ± SD) during control conditions. Infusing sodium nitroprusside (20 ± 4 $\mu g \cdot kg^{-1} \cdot min^{-1}$) significantly decreased the mean arterial pressure to 64 ± 5 mmHg, a decrease of 27%. With nitroglycerin (7 ± 3 $\mu g \cdot kg^{-1} \cdot min^{-1}$), mean arterial pressure decreased significantly to 67 ± 3 mmHg, a decrease of 23%.

Arteriolar Diameter

Figure 1 illustrates the arteriolar network. During control conditions, arteriolar diameter was greater upstream at the entrance of the arteriolar network (feed segment before the first branch) compared with the downstream regions of the same feed, as previously.[18] Along the length of the feed vessel, the feed decreased in diameter from 23.3 ± 2.6 μm at point A of figure 1, to 17.8 ± 1.6 μm immediately proximal to the last branch point (point C, figure 1, with data in figure 2). Similarly, the diameters for the branches ranged from 19.3 ± 2 μm for the first branch to 16.2 ± 1.3 μm for the last branch. Nitroglycerin had no effect on the arteriolar diameters throughout the arteriolar network, for the feed (fig. 2A), or for the branches (fig. 2B). In contrast, nitroprusside significantly dilated the feed vessel at the entrance of the network to 29.3 ± 2.6 μm, down to the level of the third branch (fig. 2A). Only the first branch arteriole dilated significantly with nitroprusside (third branch, $P = 0.1$). Thus, the response to the systemic infusion of nitroprusside

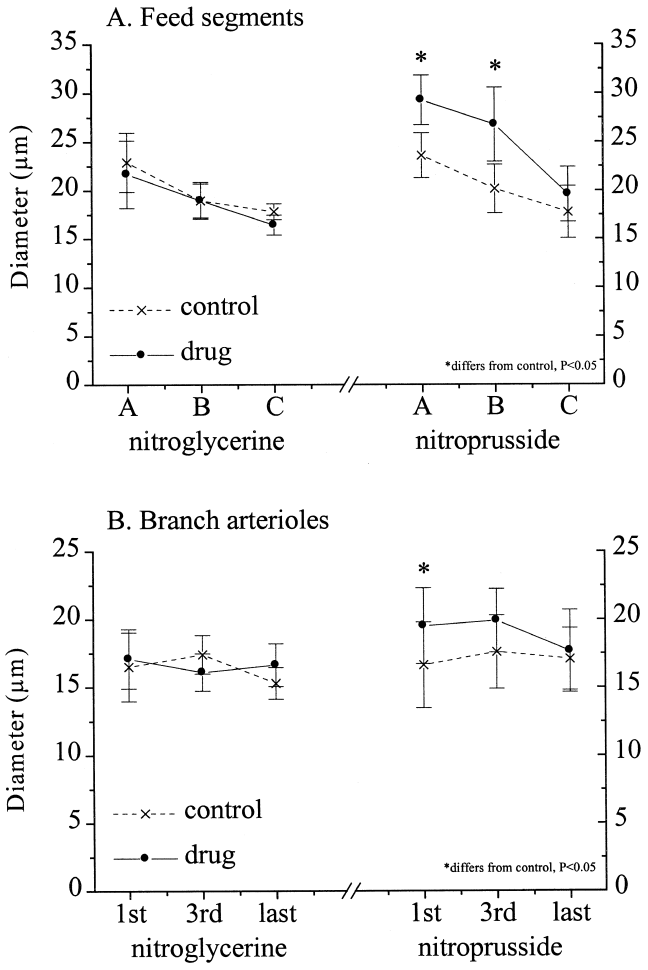

Fig. 2. The baseline control (x) diameters (μm), and diameters after the 30-min test infusion (●) of nitroprusside (n = 8) or nitroglycerin (n = 8). Diameters are shown for (*A*) the feed and (*B*) branch arterioles at the first, third, and last branch points, as shown in figure 1. *Differs from control.

was noticeably heterogeneous, with pronounced dilation only occurring upstream.

Erythrocyte Flux

Erythrocyte flux was used to evaluate changes in inflow supply to this arteriolar network (*e.g.*, inflow to the feed vessel, marked as point A in figure 1) and changes in flow distribution within this network (*e.g.*, cell flux to each sequential branch). In controls, the total inflow into the feed vessel was 1,560 ± 335 cells/s. Nitroglycerin infusion significantly decreased the cell flux into the network by half to 855 ± 171 cells/s (fig. 3A). With nitroprusside infusion, cell flux into the network was 2,600 ± 918 cells/s. Because of a corresponding increase in variability of cell flux with nitroprusside, this was not a significant increase ($P = 0.07$). In fact, the coefficient of variation for cell flux increased from the control value of 0.67 (all control) to 0.93 with nitroprusside, and it decreased to 0.49 with nitroglycerin. Using a nonparametric statistical test for variation, also called dispersion, we found that the amount of dispersion in cell flux was significantly greater with nitroprusside than with nitroglycerin, or during control (by the Moses test, $P < 0.01$; see Daniel[36]). This means that the cell inflow to the feed actually became less predictable across animals with nitroprusside and more predictable with nitroglycerin. Thus, the inflow supply capacity decreased and became more uniform with nitroglycerin, and in contrast increased yet became more variable with nitroprusside.

Figure 3A shows that the cell flux remained suppressed along the length of the feed with nitroglycerin

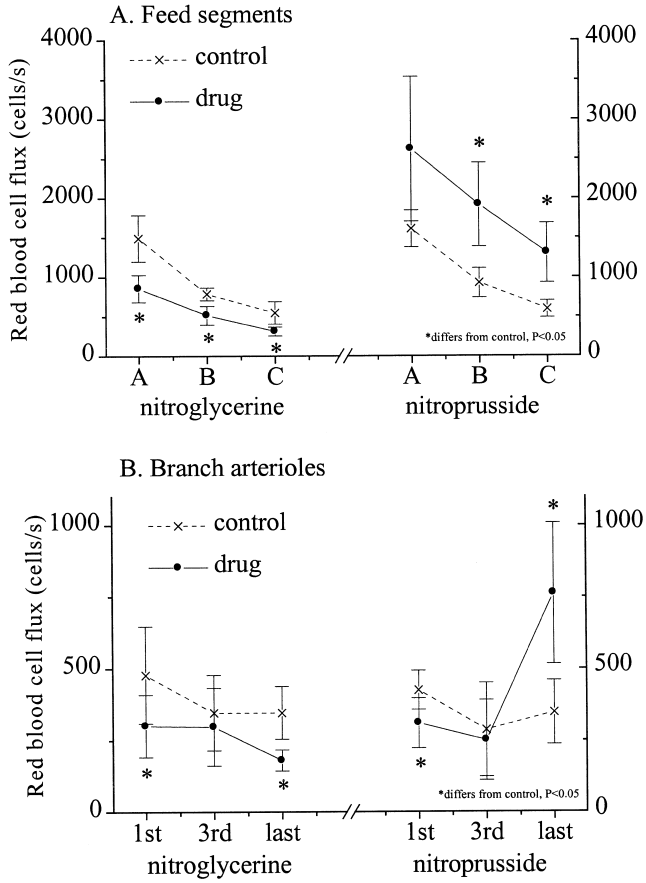

Fig. 3. The baseline control (x) erythrocyte flux (cells/s) values and flux after the 30-min test infusion (●) of nitroprusside (n = 8) or nitroglycerin (n = 8). Flux values are shown for (*A*) the feed and (*B*) branch arterioles at the first, third, and last branch points, as shown in figure 1. *Differs from control.

and remained elevated along the feed with nitroprusside. Within the arteriolar network, cell flux uniformly decreased into each branch along the feed with nitroglycerin (fig. 3B). With nitroprusside, cell flux changes were very consistent between animals and showed a decrease to the upstream branches (significant at the first branch). All of the additional inflow was channeled to the last branches of this network. As we have shown for this network in other studies,[18,37] the last branch point exhibits a characteristic flow split of 60% to the anatomically larger branch. Data for the larger "last" branch are contained in figure 3B, which shows a significant increase in cell flux with nitroprusside and a significant decrease with nitroglycerin. The smaller of the last branches always received 39 ± 3% of the flow presented to the last feed during either control, or with nitroglycerin or nitroprusside. The cell flux into the smaller branch increased from 246 ± 115 to 543 ± 153 cells/s with nitroprusside, and significantly decreased from 212 ± 40 to 131 ± 24 cells/s with nitroglycerin. Thus, the terminal bifurcation of this network provides a "run-off" for the excess flow with nitroprusside. With nitroglycerin, there is decreased cell flux to all branches, independent of location within the network.

Wall Shear Stress

Normally, this arteriolar network has a defined and predictable wall shear stress distribution, with a constant wall shear stress along the feed vessel and a relatively higher wall shear stress in the first and second upstream branches. Similarly, in the current study, during control conditions the wall shear stress values were constant along the feed vessel (fig. 4A) and significantly greater in the first compared with the other branches (fig. 4B). With nitroglycerin, wall shear stress decreased at each position along the feed, with significant reductions at the first and last feed segments (fig. 4A). This was due to a uniform decrease in cell velocity (first feed: from 905 ± 175 to 580 ± 160 μm/s) along the length of the feed (last feed segment: from 868 ± 191 to 582 ± 140 μm/s), without diameter changes (fig. 2). With nitroprusside, wall shear stress did not change uniformly along the feed vessel but rather decreased at the first bifurcation and increased at the last bifurcation. This was a result of increased diameter (fig. 2) without a change in cell velocity at the first feed (from 840 ± 166 to 824 ± 165 μm/s), and a result of only increased velocity at the last feed segment (from 932 ± 162 to 1,320 ± 310 μm/s). The calculated apparent viscosity did not change significantly from control (2.0 ± 0.1 cP) to with nitroprusside (2.2 ± 0.3 cP) or with nitroglycerin (2.2 ± 0.4 cP).

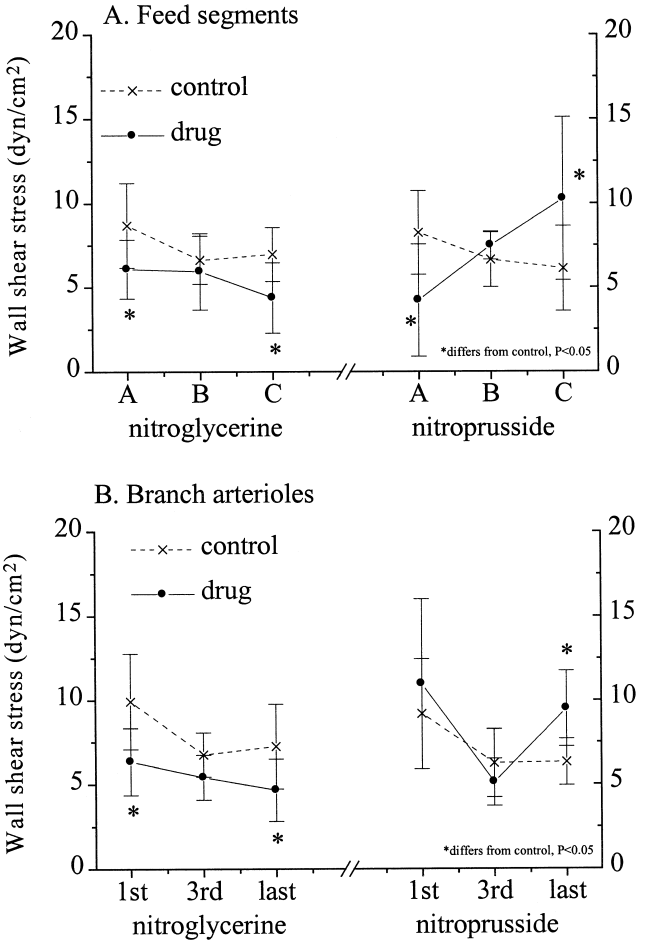

Fig. 4. The baseline control (x) wall shear stress (dyn/cm^2) values and wall shear stress after the 30-min test infusion (●) of nitroprusside (n = 8) or nitroglycerin (n = 8). Wall shear stress values are shown for (*A*) the feed and (*B*) branch arterioles at the first, third, and last branch points, as shown in figure 1. *Differs from control.

In the branch arterioles, the wall shear stress decreased significantly with nitroglycerin (at the first and last branch points), as had the cell flux (compare figures 3B with 4B). Thus the flux and wall shear stress were linked and the effect of nitroglycerin was uniform. With nitroprusside, however, significant changes in wall shear stress were not mirrored by corresponding changes in cell flux, except at the last branch. The wall shear stress was thus suppressed and made uniform throughout the arteriolar network with nitroglycerin, whereas it became disorganized and more variable with nitroprusside.

Discussion

This study shows that systemic infusion of two nitrovasodilators, nitroglycerin and sodium nitroprusside, each affect flow in an arteriolar network deep within the tissue in specific and quantifiably different ways. Our primary finding is that nitroprusside infusion increases the heterogeneity of flow, whereas nitroglycerin promotes a homogeneous distribution of flow.

It is well documented that nitroglycerin and nitroprusside have different effects within the systemic circulation.[2,5] Nitroprusside decreases preload and afterload, with the major dilatory effect on the systemic resistance vessels (afterload). Nitroglycerin has its greatest dilatory effects on the venous capacitance vessels, leading to a subsequent decrease in preload. One of the most important clinical aspects related to the perfusion abilities of these two drugs is the concept of coronary steal. Nitroprusside infusion has been shown, in fact, to increase ischemia in patients while trying to treat it.[2] It is postulated, but not yet shown, that the inappropriate dilation of small coronary arteries is the cause of this phenomenon. Nitroglycerin, in contrast, displays an extreme selectivity for the vasculature it dilates, acting only on larger conductance vessels that increase collateral blood flow, serving to redirect blood to ischemic areas.[11,12,14] The current study addresses whether these differences seen at the macroscopic level can be measured at the microvascular level.

Microvascular flow distribution is highly organized and tightly regulated. Within the arteriolar network we studied in the current investigation, we showed previously that different patterns of flow distribution can be triggered with metabolic or with adrenergic stimuli. With an equivalent constriction to norepinephrine (applied to the tissue) or to oxygen (to the tissue), we can repeatedly induce distinctly agonist-dependent and highly organized changes for cell flux and wall shear stress.[18,26] Dilation to tissue-applied adenosine also induces characteristic changes in this arteriolar network.[25,31,38] In the current study, we draw conclusions about heterogeneity of flow within the tissue. We can only make those conclusions because we have characterized extensively the behavior of the "generic" arteriolar network in this tissue. Changes within this network (*e.g.*, into the branches) describe heterogeneity of flow into and among separate capillary networks. Another key concept is that we know this arteriolar network is repeated across the tissue. Changes into the network itself will tell us how flow is distributed into other similar units across the tissue, and thus will tell us about regulation of flow across the tissue.

Microcirculatory Evidence of Systemic Effects

With nitroglycerin treatment, the arteriolar network experienced virtually no change in arteriolar diameters, as expected and consistent with the primary systemic effect of nitroglycerin to dilate the large veins, and thus to decrease preload.[12,14,39] The resulting decrease in arterial pressure may also decrease total flow into individual organs, including skeletal muscle.[3,6,40,41] Our observed decrease in the flow to the arteriolar network deep within the tissue is consistent with the studies that report a decrease in flow to the organ. Our study describes flow within the skeletal muscle. We observe an arteriolar network that is a repeating architectural unit across the tissue and that functionally behaves in a coordinated manner. Because the flow decreased uniformly both into and within this arteriolar network, our study predicts that nitroglycerin infusion has maintained a uniform distribution of the available flow across the entire tissue. Other studies have suggested that tissue autoregulation is retained with nitroglycerin exposure,[12,40] and our uniform change in flow is further proof of intact autoregulation mechanisms. Furthermore, nitroglycerin is also known to retain or improve tissue oxygenation.[6,10,11] This is consistent with our observed decrease in total flow, allowing more time for oxygen extraction, and our observed uniformity of flow, allowing uniform distribution of the oxygen to the capillaries. Our study provides direct evidence that nitroglycerin infusion uniformly changes flow within skeletal muscle tissue, thus retaining or improving homogeneity of flow within units defined by this small arteriolar network.

Nitroprusside, in contrast, did not have a uniform effect. Select upstream portions of the arteriolar network became dilated. The group of arterioles that we studied controls flow distribution into adjacent capillary networks, and thus controls oxygen supply capacity to the tissue.[22,25] It is interesting to note that the dilation to nitroprusside quite specifically extended only through one half of the feed vessel (*i.e.*, through one half of this arteriolar network). Arteriolar dilation alone is consistent with the documented afterload effects of nitroprusside.[5] However, directly induced dilation by nitroprusside cannot explain the pattern of dilation seen within this arteriolar network. These data suggest that the direct effects of nitroprusside

extend well into the microcirculation in this skeletal muscle and yet halt within the very arteriolar network that controls tissue perfusion. This is consistent with previous studies documenting size and location specificity for arteriolar dilation to intravenous nitroprusside, with variation in the amount of dilation in smaller arterioles.[7,8] Concurrently, we see a doubling of the inflow into this arteriolar network; skeletal muscle flow is reported to be elevated during intravenous nitroprusside in many[3,41,42] but not all studies.[6,40] Nitroprusside, acting in an opposite manner compared with nitroglycerin, causes a loss of organ autoregulation of flow in many tissues,[2,6,40,42,43] with concurrent decreased peripheral oxygenation.[10,11] Decreased oxygenation would be consistent in a scenario with elevated flow and nonuniform flow delivery. Perhaps the loss of autoregulatory ability is the reason that microvascular steal occurs. Our data provide direct evidence that the steal phenomenon occurs in this group of arterioles. Specifically, we have identified that the terminal arteriolar bifurcation of this network serves as a conduit for the excess flow. This finding has enormous implications for the control of microcirculatory flow and suggests that the terminal branches represent the entrance to a thorough-fare channel through the capillary beds. Furthermore, erythrocytes were stolen from the upstream branches, even though the feed inflow doubled. The curious part is that there was a uniform decrease in cell flux to the first branch, despite an increased variability elsewhere in this same network. We interpret this steal behavior as an organized response for this group of vessels, because it occurred at the same vascular branches in each animal studied. Thus, these data show that microvascular steal occurs in a prescribed and organized manner with nitroprusside. Thus, the flow within this group of arterioles is predictable by branch location (first, second, and so forth), albeit is highly heterogeneous between branches. Separately, between animals, the effect of nitroprusside on flow into this network was not uniform, as seen by the huge increase in variability between animals. This means that across the tissue, flow was heterogeneous and not predictable into the groups of feed/branch arterioles. We conclude that nitroprusside infusion increases the heterogeneity of flow within skeletal muscle, perhaps in a prescribed or orderly way, but in a pattern that we have not yet deciphered.

Direct versus *Indirect Effects*

Previously, we showed that changes in erythrocyte flux and in wall shear stress are tightly coupled when endogenous nitric oxide pathways are intact. In the current study, we found that with exogenous intravascular nitroglycerin, this link between cell flux and wall shear stress is maintained, suggesting that the arteriolar network retains its intrinsic control capability. Importantly, we did not observe direct dilation to nitroglycerin at this level. The lack of dilation to nitroglycerin in our study is likely a result of the finding that nitroglycerin must undergo a complex bioconversion to release nitric oxide, a process that only larger vessels can complete.[14] Coupled with the short half-life of nitric oxide, it is likely that our observations did not result from a direct effect of nitric oxide on the microcirculation. We conclude that the microcirculatory effects of nitroglycerin are therefore primarily the result of its systemic effects to decrease preload, and thus mean arterial pressure. Our observations reflect the intrinsic microcirculatory reflex response (*e.g.*, the complex phenomenon called autoregulation). What the current study does not address is the additional contribution of flow control at the venular end. That remains for future studies.

The mechanism of action of nitroprusside is complex and involves spontaneous release of nitric oxide and of cyanide. Cyanide toxicity is a serious consideration for patients receiving prolonged nitroprusside therapy.[44] The body can buffer up to 175 μg/kg cyanide per kilogram of body weight; this corresponds to 450–500 μg/kg total infused nitroprusside. During the 30-min infusion, animals in our study would have received only 6.6 μg/kg total nitroprusside, which is significantly less than the buffering capacity suggested for the resulting cyanide. We conclude that the microcirculatory effects resulting from nitroprusside infusion are not related to cyanide or cyanide toxicity. What remains is a complex interaction between the direct systemic effect to decrease MAP by decreasing afterload, the direct effect of the spontaneously generated nitric oxide to dilate blood vessels, and the microcirculatory reflex responses to these direct effects. We know that spontaneous nitric oxide donors exert a direct effect on these microvessels.[12,23,37,45–47] We also have found that nitroprusside and SIN-1 (3-morpholinosydnonimine, another spontaneous nitric oxide donor) each trigger vascular communicating responses.[37] Thus, the response to sodium nitroprusside involves many components, and our findings in

the current study may be the result of a direct effect of nitric oxide to dilate and a complex indirect response.

In conclusion, this study evaluated the microcirculatory effects of decreased mean arterial pressure by two nitrovasodilators. The data show direct evidence of a microcirculatory basis for nitroprusside-induced vascular steal and decreased afterload, extending well into the microcirculation of this skeletal muscle model. The data with nitroglycerin, in contrast, show no evidence of a direct effect on the microcirculation, and instead may illustrate the consequent autoregulatory effects in skeletal muscle of systemic nitroglycerin (or hypotension).

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