Regulation of Vascular Tone and Pulse Wave Velocity in Human Muscular Conduit Arteries

Selective Effects of Nitric Oxide Donors to Dilate Muscular Arteries Relative to Resistance Vessels

Henry Fok, Benyu Jiang, Brian Clapp, Phil Chowienczyk

Abstract—Arterial tone in muscular conduit arteries may influence pressure wave reflection through changes in diameter and pulse wave velocity. We examined the relative specificity of vasodilator drugs for radial artery and forearm resistance vessels during intrabrachial arterial infusion. The nitric oxide (NO) donors, nitroglycerine and nitroprusside, and brain natriuretic peptide were compared with the α-adrenergic antagonist phentolamine, calcium-channel antagonist verapamil, and hydralazine. Radial artery diameter was measured by high resolution ultrasound, forearm blood flow by strain gauge plethysmography, and pulse wave velocity by pressure recording cuffs placed over the distal brachial and radial arteries. Norepinephrine was used to constrict the radial artery to generate a greater range of vasodilator tone when examining pulse wave velocity. Despite dilating resistance vasculature, phentolamine and verapamil had little effect on radial artery diameter (mean dilation <9%). By contrast, for comparable actions on resistance vessels, nitroglycerine and nitroprusside but not brain natriuretic peptide had powerful actions to dilate the radial artery (dilations of 31.3±3.6%, 23.6±3.1%, and 9.8±2.0% for nitroglycerine, nitroprusside, and brain natriuretic peptide, respectively). Changes in pulse wave velocity followed those in arterial diameter irrespective of the signaling pathway used to modulate arterial tone (R=−0.89, P<0.05). Basal tone in human muscular arteries is relatively unaffected by α-adrenergic or calcium-channel blockade, but is functionally or directly antagonized by NO donors. The differential response to NO donors suggests that there is potential to manipulate the downstream pathway to confer greater specificity for large arteries with a resultant decrease in pressure wave reflection and systolic blood pressure. (Hypertension. 2012;60:1220-1225.)

Key Words: α-adrenergic antagonists ■ brain natriuretic peptide ■ calcium-channel antagonists ■ conduit arteries ■ cyclic guanosine monophosphate ■ nitric oxide ■ nitroglycerin

Human pharmacology of vasodilator drugs has focused almost exclusively on their actions on vascular smooth muscle in resistance vessels composed of the arterioles and microvessels <100 μm in diameter that determine peripheral vascular resistance and, hence, mean arterial blood pressure. The pulsatile component of blood pressure, which is of more prognostic importance than mean arterial pressure, especially in older subjects, is, however, dependent on characteristics of large elastic and muscular conduit arteries. Vasodilator drugs have little effect on large elastic arteries (other than the passive effect because of a change in blood pressure), presumably because these have little smooth muscle. By contrast, conduit arteries are composed mainly of vascular smooth muscle and, dependent on basal vascular tone, might be expected to be responsive to vasodilators. Conduit arteries influence the pulsatile component of blood pressure through their contribution to total arterial functional compliance or reservoir property and to pressure wave reflections. Pressure wave reflections may be important over and above their contribution to blood pressure, because they are predictive of cardiovascular mortality independent of conventional blood pressure components. The effects of wave reflections may be influenced both by artery diameter and local arterial pulse wave velocity (PWV) within conduit arteries, because this PWV influences the timing and functional consequence of reflections in terms of ventricular loading. The aim of the present study was to first compare the effects of different classes of vasodilator drug on arterial tone and vasodilation of the radial artery, as an example of a muscular conduit artery, and on forearm resistance vessels representative of those contributing to total peripheral vascular resistance. A second objective was to examine the relationship of local PWV within the radial artery to vascular tone and diameter. By infusing drugs locally into the brachial artery we were able to study these effects independent of any change in blood pressure or activation of neurohormonal reflexes. We examined drugs acting through the cyclic guanosine monophosphate (cGMP) pathway, the NO donors nitroglycerin (NTG) and nitroprusside (NP) acting on soluble...
guanylyl cyclase (sGC), and brain natriuretic peptide (BNP) acting on particulate guanylyl cyclase (pGC), because there is indirect evidence that these may exhibit relative specificity for muscular arteries.\(^7\) We compared these cGMP agents to the \(\alpha\)-adrenergic antagonist phentolamine (PHT) that would oppose sympathetically induced tone, the calcium-channel antagonist verapamil (VER) that may antagonize myogenic tone, and hydralazine (HYD), a vasodilator acting through multiple pathways.

**Methods**

**Participants**

Participants were normotensive healthy men aged 19 to 53 years (mean±SD, 27.6±8.4 years). All were asymptomatic with no history of or risk factors for cardiovascular disease (mean serum total cholesterol, 4.3±1.1 mmol/L, and blood pressure 126±10/73±9 mm Hg) and were on no regular medication. As a result of the large number of drugs examined and constraints on the duration of each intra-arterial study and number of arterial cannulations performed on each subject, not all drugs could be administered to all subjects. However, as far as possible, the allocation of study drug to subjects was randomized to avoid bias. Forty subjects were studied in total with effects of each drug being studied in a minimum of 8 subjects (except as noted below for norepinephrine [NE], where \(n=7\)) and an average number of 2 drugs studied per subject. There were no significant differences in subject characteristics (age, blood pressure, or serum lipids) between subjects receiving the various drugs. The study was approved by the London Westminster Research Ethics Committee, and written informed consent was obtained from all participants.

**Study 1: Effects of Vasodilators on Muscular Conduit Arteries and Resistant Arteries**

Studies were performed in a quiet temperature-controlled room (24–26°C). Subjects were studied at approximately the same time of day, after a standardized light meal, and were asked to avoid caffeine and alcohol on the day of the study. Subjects lay supine for \(\geq 30\) minutes before measurements were initiated. Brachial blood pressure was monitored noninvasively over the left arm throughout the study (Intellieve MP30, Philips). A 27-gauge unmounted steel needle (Cooper Needle Works) sealed with dental wax to an epidural catheter (Portex, Smiths Medical, United Kingdom) was inserted into the right brachial artery using \(\leq 1\) mL of 1% lidocaine hydrochloride (Braun Pharmaceuticals) to provide local anesthesia. Either 0.9% saline or drugs dissolved in 0.9% saline vehicle were infused at 1 mL/min using a syringe driver (Injectomat, Agilia, Fresenius Kabi, Germany). Cumulative doses of comparator drugs NP (0.3, 1.0, 3.0 \(\mu\)g/min, \(n=11\)), NTG (0.03, 0.10, 0.30, 1.00 \(\mu\)g/min, \(n=8\)), nesiritide (BNP, 0.1, 0.3, 1.0, 3.0 \(\mu\)g/min, \(n=8\)), PHT (10, 30, 100 \(\mu\)g/min, \(n=9\)), VER (10, 30, 60 \(\mu\)g/min, \(n=8\)), and HYD (10, 30, 100 \(\mu\)g/min, \(n=8\)) were infused for 12 minutes (at each dose). With the exception of NP (the infusion of which was followed by a washout period and then infusion of a second drug), the duration of action of drugs was too long to allow for \(\geq 1\) drug to be infused on 1 visit, and, therefore, each drug was given on separate occasions.

A 10-MHz ultrasound probe (Aspen, Siemens, Germany) was placed 3 to 5 cm distal to the point of arterial cannulation to image the right radial artery. Once a satisfactory 2-dimensional radial artery image was acquired, the ultrasound probe was placed in a stereotactic holder with micrometer adjustment to allow fine adjustment to maintain image quality throughout the study. End-diastolic (electrocardiogram referenced) radial artery diameter (RAD) was obtained from images acquired every 3 seconds over 2 minutes, after 7-minute infusion when steady state had been achieved (before measurement of forearm blood flow [FBF] as described below to prevent cuff inflation from interfering with ultrasound images). Images were analyzed off line using automated software (brachial analyser, Medical Imaging Applications), which provided a mean diameter over a length of artery of \(\approx 10\) mm. The coefficient of variation of arterial diameter measurements repeated at baseline and after drug washout was 4%.

After acquisition of ultrasound images, FBF measurements were taken in both arms simultaneously by venous occlusion strain gauge plethysmography (Hokanson) during the final 9 to 12 minutes of drug infusion. Wrist cuffs were inflated to 180 mm Hg. Upper-arm cuffs were inflated to 40 mm Hg intermittently and 5 measurements used to calculate mean FBF.

**Study 2: Relationship Between PWV and Muscular Conduit Artery Diameter**

In this study the aim was to examine the relation between RAD and PWV irrespective of FBF. To modulate arterial tone over a wider range, we used NE (0.01, 0.02, 0.04 \(\mu\)g/min, \(n=7\)) infused into the brachial artery alone and coinfused with PHT to generate \(\beta\)-adrenergic–mediated vasodilation (\(\beta\)-adrenergic vasodilation being opposed by \(\alpha\)-adrenergic mediated vasoconstriction). NTG (0.03, 0.10, 0.30, 1.00 \(\mu\)g/min, \(n=8\)) was used to generate an increase in RAD. PWV was measured over the brachial to radial path from simultaneous pressure cuff recordings at each site using the Vicorder device (Skidmore Medical). The path distance was taken from the proximal edge of the upper arm cuff to that of the wrist cuff. RAD and PWV were measured at baseline and after 8 minutes of infusion of each dose of drug. The Vicorder device uses a modified cross-correlation algorithm for measuring transit times with a resolution of 1.8 ms, and the coefficient of variation of readings repeated during the course of 1 study was 3%. Characteristic impedance of the forearm vascular bed (\(Z_c\)) was calculated from PWV and RAD using the relation:

\[
Z_c = \frac{4\rho \cdot PWV}{\pi \cdot RAD^2}
\]

derived from the water hammer equation,\(^4\) where \(\rho\) is blood density. Changes in PWV were partitioned into those attributed to a change in the intrinsic elasticity of the arterial wall as represented by Young’s incremental elastic modulus (Einc), thickness of the arterial wall (\(h\)), and RAD using the Moens-Korteweg equation:

\[
PWV = \sqrt{\frac{Einc \cdot h}{\rho \cdot RAD}}
\]

**Drugs**

Drugs were obtained from Baxter Healthcare (saline), Hospira Incorporation (NP and NTG), Alliance Pharmaceuticals (PHT), Sovereign Medical (HYD), Abbott Laboratories (VER), Scios Incorporation (BNP/Nesiritide), and Aguettant Ltd (NE).

**Statistical Analysis**

Results are summarized as mean±SEM. To compare the effects of drugs on RAD for the same degree of action on resistance vessels, dose-response curves were extrapolated, for each subject, to obtain a RAD representative of fixed absolute increments in FBF of 1, 2, and 3 mL/min per 100 mL. Differences between means were evaluated for statistical significance by ANOVA (for repeated measures whereas appropriate). SPSS version 16 was used for all statistical testing, and statistical significance was considered when \(P<0.05\).

**Results**

**Study 1**

Local intra-arterial infusion of the vasodilator drugs produced a dose-dependent increase in FBF in the infused arm with an \(\approx 2\)- 3-fold increase in FBF at the highest dose (Table). There were minor or nonsignificant changes in FBF in the noninfused control arm (Table) indicating that local effects in the infused arm were unlikely to be influenced by changes in systemic hemodynamics. With the exception...
of PHT, all vasodilators produced a significant increase in RAD in the infused arm. However, effects on RAD differed between the drugs. This was more marked when effects on RAD were compared for a given increase in FBF (Figure 1). PHT, VER, and BNP produced relatively little dilation (mean dilation <9%). NTG and to a lesser extent NP produced greater dilation (Figure 1, P<0.01 and P<0.05 compared with PHT). For the lowest increase in FBF of 1 mL/min per 100 mL, dilation was 1.5±1.0%, 7.8±1.6%, and 15.8±3.2% for PHT, NP, and NTG, respectively (P<0.01 for NTG versus PHT) and at the highest increase in FBF of 3 mL/min/100 mL, 3.4±1.4%, 16.8±2.6%, and 26.0±3.5% for PHT, NP, and NTG, respectively (P<0.0001 for NTG versus PHT and P<0.001 for NP versus PHT). HYD produced an intermediate degree of dilation and the order of efficacy to dilate the radial artery was NTG>NP>HYD>BNP>VER>PHT, irrespective of degree of dilation and the order of efficacy to dilate the radial artery was NTG>NP>HYD>BNP>VER>PHT, irrespective of degree of forearm resistance vessel dilation, the vasodilator drugs studied differed widely in their effects on RAD. The NO donor effects on the muscular artery, with NTG significantly more potent than the other drugs that produce the same increase in forearm blood flow (FBF). **P<0.01 vs phentolamine, verapamil, and brain natriuretic peptide (BNP). *P<0.05 vs phentolamine. Bars represent mean values; error bars are SEM, n=9, 8, 8, 8, 11, and 8 for phentolamine, verapamil, BNP, hydralazine, nitroprusside, and nitroglycerin, respectively.

**Discussion**

To our knowledge, this is the first study to systematically examine the effects of different classes of vasodilator drugs on both muscular arteries and resistance vessels. For the same degree of forearm resistance vessel dilation, the vasodilator drugs studied differed widely in their effects on RAD. The NO donors NTG and NP acting through sGC had marked vasodilator effects on the muscular artery, with NTG significantly more effective than NP. NTG produced a >20% increase in RAD. The functional consequences of such an increase in diameter result from the change in local Zc influencing the local dynamic pressure/flow relation with greater initial flow and hence shear stress for a given pressure and a reduction in pressure wave reflection. Assuming no change in intrinsic arterial elasticity, Zc is proportional to RAD and, therefore, a 20% increase in RAD will produce an ≈50% reduction in Zc. By
contrast to NTG and NP, BNP, acting through pGC rather than sGC, had less effect than either NTG or NP. Differential effects of the NO donors could relate to variation in the NO species released (eg, different redox states or thiol intermediates) with potential actions on potassium channels and mechanism other than cGMP. Differences between the sGC and pGC activators could be attributed to spatial compartmentalization of cGMP production, whereby a higher concentration of cGMP accumulates close to the sarcolemma when activated by sGC in the cytosol as opposed to membrane-bound isoform pGC.9,10

The marked vasodilator effects of NTG demonstrate that there is considerable resting tone within muscular arteries. This could be sympathetically mediated (as in resistance vessels) or because of an intrinsic myogenic response (not necessarily causing vasoconstriction in response to an increase in transmural pressure but an increase in tone) or another mechanism. Despite producing similar effects on resistance vasculature to NTG and reversing constriction produced by the α-adrenergic agonist NE, the α-adrenergic antagonist PHT had relatively little effect on muscular artery diameter. This suggests that there is little sympathetically mediated resting tone within these muscular arteries. The myogenic response is thought to be mediated in part through calcium channels and, in some vessels, is inhibited by the l-type calcium-channel blocker VER.11 In the present study, however, we found that, whereas VER dilated resistance vessels with similar efficacy to PHT, it had only minor effects on the muscular artery. Thus, if a myogenic response is implicated in resting tone within muscular arteries it is likely that this is mediated through a mechanism other than l-type calcium channels but which is functionally or directly antagonized by an NO donor. In this regard it is notable that NO donors reduce myogenic tone in rat arteries by a mechanism thought to involve activation of calcium-dependent potassium channels.12

A second objective of our study was to examine the relationship between arterial vasodilator tone and local PWV in the radial artery. Previous studies have demonstrated that PWV is influenced by NO, with PWV increased or reduced by inhibition or stimulation of NO synthase, respectively,13 and decreased by administration of an exogenous NO donor.14 However, it is unlikely that these findings relate to a specific effect of NO on arterial elasticity, and PWV may simply relate to arterial smooth muscle tone irrespective of the signaling pathway through which this is modulated. In the present study we altered arterial vasodilator tone over a wide range, using NE to constrict the artery, PHT to antagonize the actions of NE on α-adrenergic receptors and thus dilate the artery through its unopposed vasodilator actions on β-adrenergic receptors, and NTG as an NO donor and dilator. Changes in PWV were closely related to those in arterial diameter, irrespective of the vasoactive drug used to modulate arterial tone. This suggests that, under physiological conditions, PWV in muscular arteries is determined by smooth muscle tone rather than being influenced by a specific signaling pathway. Because PWV is known to be dependent on transmural pressure,14 this finding is consistent with arterial tone in muscular arteries being generated mainly as an intrinsic myogenic response to transmural pressure. Although a measure of arterial stiffness, PWV may be influenced both by arterial diameter and the intrinsic elasticity of the arterial wall. The Moens-Korteweg equation allows change in PWV to be partitioned into that attributed to the change in Einc.h and RAD, with the percentage of change in PWV predicted to be 50% of that arising from change in RAD if Einc.h remains constant. Figure 2 shows that the percentage of change in PWV was ≈25% of that in RAD, suggesting that Einc.h increases. Because an increase in RAD necessarily implies a reduction in h as the wall is stretched, our results suggest a paradoxical increase in Einc. This behavior exemplifies the complex relations among local geometry, wall stiffness, and PWV.14,15

The clinical relevance of our findings relates to the potential to selectively reduce tone of muscular arteries, with the resultant vasodilatation and decrease in PWV reducing pressure wave reflection and delaying the time of its arrival in systole, actions that will reduce systolic blood pressure and pulse pressure.16

Figure 2. A, Relation between changes in radial artery pulse wave velocity (PWV) and diameter during modulation of arterial tone with nitroglycerin (NTG, n=8), phenolamine (PHT, n=8), and norepinephrine (NE, n=7) alone and with PHT (n=7). Each symbol represents mean values for each dose of drug. The solid line is the line of best fit by linear regression; Pearson correlation coefficient, R=−0.89, P<0.05. B, Relation between change in local characteristic impedance (Zc) and radial artery diameter during modulation of arterial tone as in A. R=−0.99, P<0.001.
Selective vasodilatation of muscular as opposed to resistance vessels means that diastolic pressure will be relatively unaffected. Such hemodynamic effects are likely to be of particular benefit in subjects with isolated systolic hypertension in whom myocardial wall stress and oxygen consumption are elevated because of raised systolic pressure but in whom a reduction of diastolic pressure in parallel with systolic pressure may compromise myocardial perfusion during diastole. It is notable that NTG is remarkably effective at reducing systolic pressure (particularly central systolic pressure) with little effect on diastolic pressure. The present results suggest that, of currently available vasodilator agents, NO donors show the greatest selectivity for muscular arteries. Furthermore, the heterogeneity between NO donors suggests that this is dependent on the exact NO signaling pathway and that there may be potential to exploit this to enhance the selectivity of these agents to dilate muscular arteries. In this regard, it is notable that the NO donor sinitrodil can increase compliance of the brachial artery without effects on systemic vascular resistance. It should be noted that the present study provides insight into the potential to exploit this to enhance the selectivity of these agents to dilate muscular arteries. In conclusion, the smooth muscle tone of conduit arteries that determines arterial diameter and PWV is likely to be generated by an intrinsic myogenic response to transmural pressure and, for an equivalent action on resistance vessels, is functionally or directly antagonized by NO donors more effectively than by α-adrenergic or calcium-channel blockade.

Perspectives
Although the regulation of vascular tone in resistance vasculature has been studied extensively, relatively little is known regarding that in muscular conduit arteries. Such tone in muscular arteries is likely to be important because it influences systemic hemodynamics through effects on the functional compliance of the arterial tree and on pressure wave reflection. The present study suggests that, under resting conditions, the tone of muscular arteries is largely independent of sympathetic outflow acting on α-adrenergic receptors but is generated as a myogenic response to transmural pressure. NO donors seem highly effective at antagonizing such resting tone with greater action on muscular arteries relative to resistance vessels than calcium-channel antagonists. However, there is considerable heterogeneity between drugs acting through the NO-cGMP pathway in their selectivity for muscular arteries. This suggests that there is the potential to design drugs acting through this pathway that dilate muscular arteries but have minimal effects on resistance vasculature and that would be useful in treating systolic hypertension.

In conclusion, the smooth muscle tone of conduit arteries that determines arterial diameter and PWV is likely to be generated by an intrinsic myogenic response to transmural pressure and, for an equivalent action on resistance vessels, is functionally or directly antagonized by NO donors more effectively than by α-adrenergic or calcium-channel blockade.

Sources of Funding
This work was supported by a British Heart Foundation Clinical Research Training Fellowship FS/11/11/28634 to Dr Fok. We also acknowledge financial support from the Department of Health via the National Institute for Health Research comprehensive Biomedical Research Centre award to Guy’s and St Thomas’ National Health Service Foundation Trust in partnership with King’s College London and King’s College Hospital National Health Service Foundation Trust.

Disclosures
Brian Clapp and Phil Chowienczyk have an interest in Centron Diagnostics, a King’s College London “spin-out” company developing technology for central blood pressure measurement.

References
16. Pauca AL, Kon ND, O’Rourke MF. Benefit of glyceryl trinitrate on arterial stiffness is directly due to effects on peripheral arteries. Heart. 2005;91:1428–1432.

**Novelty and Significance**

**What Is New?**
- In comparison to resistance vessels, tone of conduit arteries is insensitive to α-adrenergic blockade, calcium-channel blockade, or brain natriuretic peptide.
- It is sensitive to the nitrovasodilators nitroglycerin and nitroprusside.
- Pulse wave velocity in conduit arteries is dependent on arterial tone.

**What Is Relevant?**
- Arterial tone in conduit arteries may be mediated by a myogenic mechanism antagonized by nitric oxide.

- Arterial tone and pulse wave velocity in conduit arteries influence pulse pressure augmentation.
- Nitrovasodilators may have relatively selective actions on conduit arteries.
- Differences between vasodilators acting through cyclic guanosine monophosphate suggest that there is potential to design drugs with greater specificity for conduit arteries.

Regulation of Vascular Tone and Pulse Wave Velocity in Human Muscular Conduit Arteries: Selective Effects of Nitric Oxide Donors to Dilate Muscular Arteries Relative to Resistance Vessels

Henry Fok, Benyu Jiang, Brian Clapp and Phil Chowienczyk

Hypertension. 2012;60:1220-1225; originally published online October 8, 2012; doi: 10.1161/HYPERTENSIONAHA.112.198788

Hypertension is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2012 American Heart Association, Inc. All rights reserved.
Print ISSN: 0194-911X. Online ISSN: 1524-4563

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://hyper.ahajournals.org/content/60/5/1220

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