Effects of transmural pressure on brachial artery mean blood velocity dynamics in humans

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Lott, Mary E. J., Michael D. Herr, and Lawrence I. Sinoway. Effects of transmural pressure on brachial artery mean blood velocity dynamics in humans. J Appl Physiol 93: 2137–2146, 2002. First published September 6, 2002; 10.1152/japplphysiol.00443.2002.—The effects of changes in transmural pressure on brachial artery mean blood velocity (MBV) were examined in humans. Transmural pressure was altered by using a specially designed pressure tank that raised or lowered forearm pressure by 50 mmHg within 0.2 s. Brachial MBV was measured with Doppler directly above the site of forearm pressure change. Pressure changes were evoked during resting conditions and after a 5-s handgrip contraction at 25% maximal voluntary contraction. The handgrip protocol selected was sufficiently vigorous to limit flow and sufficiently brief to prevent autonomic engagement. Changes in transmural pressure evoked directionally similar changes in MBV within 2 s. This was followed by large and rapid adjustments [−2.14 ± 0.24 cm/s (vasoconstriction) during negative pressure and +2.14 ± 0.45 cm/s (vasodilatation) during positive pressure]. These adjustments served to return MBV to resting levels. This regulatory influence remained operative after 5-s static handgrip contractions. Of note, changes in transmural pressure were capable of altering the timing of the peak MBV response (5 ± 0, 2 ± 0, 6 ± 1 s ambient, negative, and positive pressure, respectively) as well as the speed of MBV adjustment (−2.03 ± 0.18, −2.48 ± 0.15, −0.84 ± 0.19 cm·s⁻¹·s⁻¹·ambient, negative, and positive pressure, respectively) after handgrip contractions. Vascular responses, seen with changes in transmural pressure, provide evidence that the myogenic response is normally operative in the limb circulation of humans.

myogenic response; regional blood flow; exercise; Doppler velocity

THE MYOGENIC RESPONSE is an important contributor to blood flow autoregulation. It is the inherent ability of arterial blood vessels to respond to changes in transmural pressure (35). Animal studies (5, 9, 12, 21, 30, 35) have shown that blood vessels, particularly arteries and arterioles in the skeletal muscle and brain, exhibit a strong myogenic response that results in smooth muscle contraction as transmural pressure rises and in relaxation as transmural pressure falls (21). This pressure-sensitive mechanism has been shown to be independent of neural (10, 11) and endothelial influences (7, 8).

Compared with the vast number of animal studies performed (5, 21, 30, 35), there are fewer human studies that have examined the myogenic response. In humans, isolated coronary artery preparations have been shown to display a myogenic response to changes in pressure (31). Previous human studies investigating the myogenic response have used different models to alter transmural pressure, such as raising or lowering the limbs relative to the heart (20), head-up tilt (19) and dynamic handgrip exercises to evoke an increase followed by a rapid fall in blood pressure (36). However, these models are limited in that it is difficult to abruptly and precisely alter transmural pressure without also interfering with the ability to accurately measure rapid flow adjustments during the intervention. An alternative noninvasive method for altering local vascular transmural pressure without affecting the remainder of the systemic circulatory is to alter transmural limb pressure by means of a “limb pressure tank” (28, 29). With the use of this method to alter limb pressure, limb blood vessel movement can be minimized and the pressure throughout the exposed limb can be fully transmitted (29). Thus there are equal changes in arterial and venous pressure (VP). However, because of structural differences in these vessels (15), it is unclear whether increases in transmural pressure may alter the arterial-VP gradient. Early human studies (3, 14) that utilized the limb pressure tank showed forearm/leg vasoconstriction occurring 15–45 s after subatmospheric pressure (negative pressure) was withdrawn. However, the use of volume plethysmography to measure blood flow in these studies had two important limitations: 1) it is an indirect method that cannot be used to measure limb flow during the actual period of subatmospheric pressure; and 2) it has limited time resolutions and, therefore, lacks the ability to examine responses that occur immediately on return to atmospheric pressure.

Animal studies have shown that the myogenic response is still operative with skeletal muscle contrac-
tions (32, 39). During muscle contraction and relaxation, large changes in transmural pressure occur that make the study of this phenomenon important. On the basis of the results of these prior animal studies, we believe that further evaluation of the myogenic responses after human muscle contraction is warranted.

Accordingly, the purpose of this study was to examine the characteristics of both acute and sustained beat-by-beat changes at rest in brachial mean blood velocity (MBV) measured by Doppler as the transmural pressure was raised or lowered by using a pressurized tank device for human subjects. A brief static handgrip (SHG) stimulus was added to determine whether this regulatory system was operative in the presence of muscle contraction. We hypothesized that increases in transmural pressure would cause an initial increase in MBV followed by a rapid fall in limb MBV and that reductions in transmural pressure would lead to an initial fall in MBV that would be followed by a myogenic vasodilation. The results of these experiments demonstrate for the first time in humans that changes in transmural pressure evoke changes in limb flow dynamics at rest and after muscle contraction. These changes are entirely consistent with and best explained by the myogenic response.

METHODS

Subjects

Eighteen healthy (9 men and 9 women) subjects with a mean ± SE age of 24 ± 1 yr (range: 19–29 yr), height of 174 ± 2 cm (range: 156–188 cm), weight of 74 ± 3 kg (range: 50–90 kg), and a body mass index of 25 ± 1 kg/m² (range: 18–31 kg/m²) participated in this study. Subjects were active, normotensive, and nonsmokers. None of the subjects was receiving cardiovascular medications. Women were tested 20 ± 1 days into their menstrual cycle with four of nine women on oral contraceptives. Measurements were carried out in a quiet, dimly lit laboratory at a temperature between 21 and 24°C. Caffeine and exercise were avoided for 24 h before testing. The Institutional Review Board of The Milton S. Hershey Medical Center approved the experimental protocol. Subjects had the purposes and risks of the protocol explained to them, and written, informed consent was obtained from them before they entered the study.

Heart Rate, Blood Pressure, and Blood Velocity

Heart rate (HR) was monitored by standard electrocardiogram. Systolic and diastolic blood pressures were measured by using the volume clamp method (Finapres, Ohmeda, Madison, WI), and mean arterial blood pressure (MAP) was calculated from the Finapres waveform. An automated sphygmomanometer (Dinamap, Critikon, Tampa, FL) was used to confirm the Finapres pressure at the beginning of the study. Brachial artery MBV was measured on a beat-by-beat basis by using a 4-MHz pulsed-wave Doppler ultrasound probe (model 500M Multigun; Yonkers, NY) with Zero Crossing (Hokanson; Bellevue, WA) taped in a fixed position ~8 cm proximal to the antecubital fossa at a 45° angle of insonation. The Doppler probe was manually adjusted to obtain the maximal Doppler frequency shift. HR, MAP, and MBV were measured continuously and collected online at 100 Hz by using a MacLab system (AD Instruments, Castle Hill, Australia).

Muscle Voluntary Contraction

In this group of experiments, we tested the hypothesis that the ability of transmural pressure to alter flow dynamics in humans would be present even after muscle contraction. This hypothesis was based on prior animal studies that have shown that the myogenic was operative after contractions (32, 39). Before testing, three maximum voluntary handgrip efforts [maximal voluntary contraction (MVC)] were performed. The highest MVC value was determined, and 25% of MVC was used for the brief (5 s) SHG contractions.

Positive and Negative Pressure Forearm Tank

The lower aspect of the forearm was sealed at the elbow in an airtight tank. Two neoprene cuffs were fitted around the mid-aspect of the arm, which created a snug, nonconstricting seal. Negative and positive pressures were obtained from an external source and directed to a box containment. From this pressurized box, a manual switch opened the system to provide the appropriate pressure to the forearm tank within 0.2 s. Before the negative or positive conditions, a test trial was performed to ensure that the appropriate pressure (±50 mmHg) was present within the forearm tank.

Experimental Protocol

Subjects were placed supine with their nondominant forearm in the tank enclosure for ~20 min before any collection of data. The forearm was held at heart level. The Doppler probe on the upper forearm was taped into position, and electrocardiogram leads were attached. A Finapres device was attached to the subject’s opposite arm and positioned at heart level.

All subjects were exposed to three pressure conditions (ambient, negative, and positive) that were examined separately and randomly in two paradigms (resting and SHG). The negative and positive pressures used were −50 mmHg and +50 mmHg, respectively. After a 1-min baseline in the resting paradigm, the tank pressure was changed to ambient, negative, or positive pressure for 2 min followed by a 2 min recovery (i.e., release of pressure). Two to three trials were done for each pressure, and the responses were averaged. All trials were separated by a 5-min resting period.

SHG paradigm also utilized −50 mmHg, +50 mmHg, and ambient pressure. In these experiments, the subject squeezed a handgrip device (25% MVC) for 5 s at the same time the forearm tank pressure was raised, lowered, or kept the same. We postulated that 25% MVC for 5 s would limit muscle flow and evoke equivalent and “limited” levels of metabolic derangement across the three levels of transmural pressure chosen. Contraction was initiated at the time the pressure in the arm tank was changed to prevent any potential effects that a change in transmural pressure could have on flow to the contracting forearm. The period of contraction was kept short (5 s) to limit engagement of the autonomic nervous system.

A key component of this study was the use of MBV as an index of blood flow. This assumption is valid as long as vessel diameter remains relatively constant (flow = velocity × vessel area). Therefore, in a separate group of experiments (2 men and 2 women), brachial diameter was measured by ultrasonography with a linear array scanhead (range of 5–12 MHz; Advanced Technology Laboratories, model HDI 5000 CV, Bothell, WA). Diameter was measured during positive and negative tank pressure at rest and after static contraction. Multiple lumen-intima measurements (3–4) were performed and averaged during diastole for each study period.
Lastly, to examine the influences of increasing transmural pressure on the venous system, we measured VP changes during increases in transmural pressure (application of −50 mmHg and release of +50 mmHg) in a separate group of subjects (3 men and 1 woman). VP was measured by the placement of a 20-gauge catheter inserted retrograde into a deep forearm vein of the arm placed into the arm tank and connected to a pressure transducer (Abbott Laboratories: Chicago, IL). Perfusion pressure (PP) was calculated from VP (corrected to be additive) and MAP (i.e., PP = −VP + MAP). Forearm volume was also measured in two of the subjects by strain-gauge plethysmography (Hokanson EC-4 Plethysmograph, D. E. Hokanson). A mercury-ﬁlled strain gauge was placed around the forearm at the point of largest circumference and calibrated (38). MBV was measured during all of the protocols.

Data Analysis

In these studies HR, MAP, and MBV were measured on a beat-by-beat basis. Trials for each condition/paradigm were averaged for each individual. The transition heartbeat (HB) was identiﬁed as the HB between resting baseline and the ﬁrst HB at the designated pressure. On initial review of the data, steady states were reached within 20 HB (−20 s) after transmural pressure changes. Therefore, results from the ﬁrst 20–30 HB (20–30 s) after alterations in transmural pressure are presented in the text and diagrams.

Derivatives of the MBV tracings were estimated to examine the effects of transmural pressure on velocity transients. The time derivative of MBV was numerically approximated by using the three-point central difference approach (43). From these MBV derivative estimates, intervals of nearly constant slope were used to identify a rise-phase interval and two distinct phases during the “fall” interval.

To compare resting and SHG MBV responses, total area under the MBV curve, from the 1st to 30th HB on change of pressure/release of SHG, was calculated by the trapezoidal method with the baseline at zero levels Σ [1 HB(MBV1HB + MBVHB/2)]. One-way analysis of variance was used to examine the effects of transmural pressure on the various measured parameters. Data are presented as means ± SE, with P < 0.05 considered statistically signiﬁcant.

RESULTS

Diameter Responses to Changes in Transmural Pressure

There were no signiﬁcant differences between baseline, peak MBV (~2–3 HB), and 30th HB diameters after change in pressure/grip release (see Table 1). In addition, there was no signiﬁcant difference in diameters before the release of pressure and peak MBV and 30th HB (~30 s) after the release of pressure (see Table 1). Because diameter did not change, MBV was reﬂective of limb ﬂow. Thus MBV will be presented as an estimate for forearm blood ﬂow.

Resting Paradigm

Hemodynamics. As expected, during the ambient resting condition, HR and MAP remained relatively constant (58 ± 1 beat/min and 86 ± 1 mmHg, respectively). Negative and positive pressure had no effect on HR and MAP (Table 2). Forearm MBV under ambient conditions was 6.48 ± 0.10 cm/s (Fig. 1).

MBV during negative pressure. The change to negative pressure (i.e., increased transmural pressure) resulted in a rapid rise in MBV from 6.14 ± 0.26 to 17.50 ± 0.94 cm/s within 2 HB (~2 s; Fig. 1). The peak change in velocity was followed by a rapid fall in MBV. MBV at the 30th HB (~30 s) was actually less during sustained negative pressure than during the ambient condition (5.15 ± 1.20 vs. 6.22 ± 1.49 cm/s; P < 0.024).

Analysis of the rate of fall in velocity (after peak MBV was achieved) demonstrated two separate constrictor phases: an acute “rapid” constrictor phase (~2.16 ± 0.24 cm·s−1·s−1) and a “less rapid” second phase (~0.35 ± 0.05 cm·s−1·s−1; Fig. 2). Thus a rise in transmural pressure led to a powerful vasoconstrictor response that was initiated by the third HB. This effect of elevated transmural pressure led to sustained limb ﬂow responses that were below baseline.

MBV during positive pressure. The change to positive pressure (i.e., decrease in transmural pressure) resulted in an immediate drop in brachial MBV to 0.00 ± 0.02 cm/s (Fig. 1). Since MBV did not change, MBV was reﬂective of limb ﬂow. Thus MBV will be presented as an estimate for forearm blood flow.

Table 1. Brachial artery diameters (cm) with change and release of pressure

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Baseline</th>
<th>At Peak MBV</th>
<th>At 30th HB</th>
</tr>
</thead>
<tbody>
<tr>
<td>Change to pressure without and with SHG</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Negative pressure at rest</td>
<td>0.42 ± 0.03</td>
<td>0.43 ± 0.03</td>
<td>0.42 ± 0.03</td>
</tr>
<tr>
<td>Positive pressure at rest</td>
<td>0.43 ± 0.03</td>
<td>0.43 ± 0.03</td>
<td>0.44 ± 0.03</td>
</tr>
<tr>
<td>Negative pressure with SHG</td>
<td>0.43 ± 0.03</td>
<td>0.43 ± 0.03</td>
<td>0.43 ± 0.04</td>
</tr>
<tr>
<td>Positive pressure with SHG</td>
<td>0.43 ± 0.03</td>
<td>0.43 ± 0.03</td>
<td>0.43 ± 0.04</td>
</tr>
<tr>
<td>Release of pressure back to ambient pressure</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Negative pressure</td>
<td>0.43 ± 0.02</td>
<td>0.44 ± 0.02</td>
<td>0.44 ± 0.02</td>
</tr>
<tr>
<td>Positive pressure</td>
<td>0.45 ± 0.02</td>
<td>0.44 ± 0.02</td>
<td>0.44 ± 0.02</td>
</tr>
</tbody>
</table>

Values are means ± SE; n = 4 subjects. HB, heartbeat; MBV, mean blood velocity; SHG, static handgrip.

Table 2. Hemodynamics and MBV under different conditions and paradigms

<table>
<thead>
<tr>
<th>Condition</th>
<th>Ambient</th>
<th>Negative</th>
<th>Positive</th>
</tr>
</thead>
<tbody>
<tr>
<td>HR, beats/min</td>
<td>58 ± 1</td>
<td>59 ± 1</td>
<td>57 ± 1</td>
</tr>
<tr>
<td>SHG paradigm</td>
<td>60 ± 1</td>
<td>60 ± 1</td>
<td>60 ± 1</td>
</tr>
<tr>
<td>Recovery, return to ambient</td>
<td>59 ± 1</td>
<td>60 ± 1</td>
<td>59 ± 1</td>
</tr>
<tr>
<td>MAP, mmHg</td>
<td>86 ± 1</td>
<td>85 ± 1</td>
<td>85 ± 1</td>
</tr>
<tr>
<td>SHG paradigm</td>
<td>86 ± 1</td>
<td>86 ± 1</td>
<td>86 ± 1</td>
</tr>
<tr>
<td>Recovery, return to ambient</td>
<td>87 ± 1</td>
<td>87 ± 1</td>
<td>87 ± 1</td>
</tr>
<tr>
<td>Baseline MBV, cm/s</td>
<td>6.48 ± 0.10</td>
<td>6.14 ± 0.02</td>
<td>6.00 ± 0.48</td>
</tr>
<tr>
<td>SHG paradigm</td>
<td>6.44 ± 0.31</td>
<td>6.00 ± 0.22</td>
<td>6.04 ± 0.97</td>
</tr>
</tbody>
</table>

Values are means ± SE. MAP, mean arterial blood pressure.
0.65 cm/s (Fig. 1). This was followed by a subsequent rapid rise in MBV at a rate of $+2.14 \pm 0.45 \text{ cm} \cdot \text{s}^{-1} \cdot \text{HB}^{-1}$ reaching a steady state of $-4.56 \pm 0.33 \text{ cm/s}$ 3 HB (~3 s) after the nadir velocity was obtained. Thus a fall in transmural pressure evoked a rapid vasodilator response that was already evident by the third HB (~3 s) after pressure was changed.

**Handgrip Paradigm**

**MBV during ambient pressure.** Peak MBV ($26.80 \pm 1.43 \text{ cm/s}$) on handgrip release (i.e., transient increase in transmural pressure) occurred by the $5 \pm 0 \text{ HB}$ (Fig. 3). After peak MBV was achieved, the fall in velocity could again be described by two phases: a rapid early first phase ($-2.03 \pm 0.18 \text{ cm} \cdot \text{s}^{-1} \cdot \text{HB}^{-1}$) and a less rapid second phase ($-0.50 \pm 0.05 \text{ cm} \cdot \text{s}^{-1} \cdot \text{HB}^{-1}$). Compared with the resting condition, SHG more than doubled the “total” velocity area response observed over the 30 measured HBs (~30 s; $422.17 \pm 26.02$ vs. $183.65 \pm 9.48 \text{ cm} \cdot \text{s}^{-1} \cdot \text{HB}^{-1}$; $P < 0.001$; ambient SHG and rest, respectively).

**MBV during negative pressure.** Negative pressure (i.e., enhanced increased transmural pressure) led to a higher ($36.88 \pm 1.78 \text{ cm/s}$) and earlier peak MBV ($2 \pm 0 \text{ HB}$; both $P < 0.001$) than was seen during ambient handgrip (Fig. 3). With the higher sustained transmural pressure, the rate of decrease for the acute rapid and less rapid phase was greater than that seen during the ambient transient pressure condition (acute: $-2.48 \pm 0.15$ vs. $-2.03 \pm 0.18 \text{ cm} \cdot \text{s}^{-1} \cdot \text{HB}^{-1}$, $P < 0.039$; less rapid: $-0.85 \pm 0.08$ vs. $-0.50 \pm 0.05 \text{ cm} \cdot \text{s}^{-1} \cdot \text{HB}^{-1}$, $P = 0.004$). During the negative pressure condition (an increase in transmural pressure), SHG more than doubled the total MBV area response observed during the resting condition ($472.37 \pm 42.49$ vs. $218.96 \pm 13.66 \text{ cm} \cdot \text{s}^{-1} \cdot \text{HB}^{-1}$; $P < 0.001$).

**MBV during positive pressure.** With positive pressure (i.e., combination of reduced and increased trans-
mural pressures), the peak MBV after handgrip was less than that seen during the ambient condition (11.02 ± 0.87 vs. 26.80 ± 1.43 cm/s, P < 0.001) and nonsignificant later (6 ± 1 vs. 5 ± 0 HB) (Fig. 3). After MBV peaked, the rate of “acute rapid” fall in MBV was substantially less rapid than during the ambient condition (−0.84 ± 0.19 vs. −2.03 ± 0.18 cm·s⁻¹·s⁻¹, P < 0.001). There was no significant difference between the less rapid phases of positive and ambient SHG (0.22 ± 0.09 vs. −50 ± 0.05 cm·s⁻¹·s⁻¹). During positive pressure (sustained reduction in transmural pressure), SHG again more than doubled the total MBV area response seen during resting conditions. SHG had a 129% greater total MBV area under the curve compared with the positive resting paradigm (247.87 ± 15.87 vs. 108.17 ± 9.21 cm·s⁻¹·HB⁻¹, P < 0.001).

**Effects of the Return to Ambient Pressure on MBV**

After 2 min at a “pressurized condition” (±50 mmHg), pressure was rapidly (within 0.2 s) changed back to ambient pressure (observed transient changes in transmural pressure during recovery). MBV responses were not significantly different in the recoveries after the resting and SHG paradigms. Therefore, recovery MBV was pooled for each pressure condition.

**MBV on release of negative pressure.** This reduction in transmural pressure was associated with an immediate fall in MBV that was followed by a subsequent rise (vasodilatation) in MBV at a rate of +2.59 ± 0.17 cm·s⁻¹·s⁻¹ (Fig. 4). A peak velocity of 13.78 ± 0.86 cm/s was noted by the eighth HB (~8 s). This was followed by a fall in velocity, with MBV returning to ambient values by the 16th HB.

**MBV on release of positive pressure.** On release of positive pressure, the rise in transmural pressure was associated with a rapid rise in MBV that peaked at 26.09 ± 0.94 cm/s by the first HB (Fig. 4). This was followed by a fall in velocity that was −3.73 ± 0.23 cm·s⁻¹·s⁻¹ and −0.44 ± 0.04 cm·s⁻¹·s⁻¹ for the acute rapid and less rapid phases, respectively.

In Fig. 5, we compared resting velocity responses seen when we increased transmural pressure by either applying suction or withdrawing positive pressure. Peak MBV was higher (23.19 ± 2.29 vs. 17.50 ± 0.94 cm/s; P < 0.03), occurred earlier [1 ± 0 vs. 2 ± 0 HB (~1 ± 2 s]; P < 0.001], and exhibited a greater acute rapid phase after peak MBV (~3.31 ± 0.38 vs. −2.16 ± 0.24 cm·s⁻¹·s⁻¹, P < 0.014) for changes from positive to ambient pressure compared with changes from ambient to negative pressure, respectively (all P < 0.05).

In an analogous fashion, velocity responses during the two interventions that reduced transmural pressure are also presented (Fig. 5). The change from negative to ambient pressure evoked higher peak MBV (13.79 ± 1.59 vs. 4.56 ± 0.33 cm/s; P < 0.001) that occurred later [7 ± 1 vs. 4 ± 0 HB (~7 vs. 4 s); P < 0.001] than MBV obtained when the external pressure was changed from ambient to positive pressure. However, there was no significant difference in rates of rise (2.22 ± 0.29 vs. 2.14 ± 0.45 cm·s⁻¹·s⁻¹) between changes in pressure from negative to ambient and ambient to positive pressure, respectively.

**Effects of Increasing Transmural Pressure on the Venous System**

Venous system during increases in transmural pressure at rest. Resting VP was 7 ± 2 mmHg before negative pressure and 6 ± 2 mmHg before positive pressure. With the application of negative pressure, VP decreased (change of −16 ± 5 mmHg) by the second HB and was steadily maintained over the next four to six HB before slowly returning toward baseline values. However, during this initial time frame, the acute rapid phase of the fall in velocity (~1.78 ± 0.45 cm/s)
was completed. Thus a large arterial vasoconstriction effect was seen as PP remained relatively constant (Fig. 6A). Likewise, on release of positive pressure, there was a large vasoconstriction response with little change in PP (Fig. 6B). In addition, on the application of negative pressure, forearm volume increased steadily throughout the 2 min at −50 mmHg (change of 0.03 ± 0.20, 0.67 ± 0.21, and 2.97 ± 0.76% at the 2nd, 5th, and 30th HB, respectively). However, there were smaller changes in forearm volume with the release of positive pressure (change of −1.14 ± 1.16%, −0.51 ± 0.93, −0.01 ± 0.17% at the 2nd, 5th, and 30th HB, respectively). Again over the initial rapid phase of the fall in velocity, there were only small changes in venous volume.

VP during increases in transmural pressure and SHG. During negative pressure, on release of SHG, the quick reduction in VP (change of −5 ± 3 mmHg) by the first and second HB coincided with peak MBV (change of 23.99 ± 2.05 cm/s). VP then began to rise above baseline values over the next 7 s before returning toward baseline. Overall, there were small reductions in PP (change of −5 ± 3 mmHg) compared with the quick, rapid fall in velocity (change of −1.42 ± 0.11 cm/s) during the acute rapid vasoconstriction phase.

DISCUSSION

In these studies, we examined the effects of altering transmural pressure on brachial artery MBV. An external pressure device was utilized that altered external forearm pressure by ±50 mmHg within 0.2 s. We coupled this device with Doppler techniques that allowed for a beat-by-beat assessment of brachial artery MBV responses to these rapid changes in transmural pressures. These studies provide the most detailed examination to date of the effects of transmural pressure on peripheral blood velocity in humans. Moreover, these findings suggest that the myogenic response plays an important role in mediating the effects seen with changes in transmural pressure.

Previous studies have demonstrated that when external forearm pressure is altered, the changes in pressure are transmitted through the entire limb (28, 29). Accordingly, in the present report, it was assumed that changes in external pressure would be entirely transmitted to the vasculature of the forearm. Thus when tank pressure was lowered by 50 mmHg, transmural pressure would rise by 50 mmHg. Conversely, when tank pressure was raised by 50 mmHg, brachial artery transmural pressure would fall by 50 mmHg.

In this report, we increased transmural pressure under resting conditions two different ways: 1) by lowering tanking pressure to −50 mmHg, and 2) by returning tank pressure to ambient from +50 mmHg (see Fig. 5, top). We speculated that these two interventions would cause an initial increase in flow that was followed by a rapid fall in limb flow. We further hypothesized that, if the myogenic response was operative, then the rate of fall in flow (vasoconstriction) after peak flow would increase as a function of the transmural pressure. In an analogous fashion, we were able to reduce transmural pressure by both raising tank pressure to +50 mmHg and by withdrawing forearm suction (see Fig. 5, bottom). These interventions would be expected to lead to an initial fall in flow that should be followed by a myogenic vasodilation. The myogenic vasodilation response that occurred within 10 to 30 s has also been shown in animal studies (5, 39). With reductions in transmural pressure, animal studies have suggested that the arterial network vasodilates as a series-coupled (21, 23) or uniform vasodilation myogenic response (4). However, because intravascular pressure was not directly measured in our study during changes in arm tank pressure, we are unable to comment on any upstream flow regulation.
Study Findings

There were no significant changes in diameter as transmural pressure was changed. This lack of diameter change agrees with previous human studies examining brachial and femoral artery changes with exercise (34, 37). Therefore, changes in MBV in response to alterations in transmural pressure were used in examining the myogenic response. Strong linear relationships ($r^2 = 0.87–0.99$) between changes in Doppler MBV and changes in blood flow measured with strain gauge (41) and thermodilution (34) have been observed in other studies. Thus MBV was also employed as an index of flow in this study.

When transmural pressure was raised, we observed a rapid increase in MBV. This transient increase in flow has been previously described in animal experiments (2, 13, 39). The mechanisms responsible for this initial transient vasodilation are unclear, but it may reflect, in part, a suction effect of the veins and/or a passive stretch of the artery. Our findings suggest that it is likely that the more compliant veins lower their internal pressure immediately to create a suction effect and immediately increase arterial inflow. This effect is unlikely to represent a metabolic effect since it occurred so rapidly. Whatever the precise mechanism for this response, it was followed by a fall in MBV described by an early acute rapid phase and a less rapid second phase. Animal studies have also observed this rapid dynamic response occurring within 15 to 30 s on an increase in transmural pressure ($<1$ s) (32, 39). However, ramping applications of the pressure change (3 min) lead to a longer time course (3 min) for the completion of the vasoconstriction response (6). Thus vasoconstriction response is dependent on the stimulus's magnitude and speed of application. We used a relatively large pressure change that was delivered quite rapidly (0.2 s). To the best of our knowledge, the present report is the first in humans or animals to characterize blood velocity changes within a few seconds of a change in transmural pressure.

Infinite gain occurs when a control system is able to return a given variable to its equilibrium point (baseline) after perturbation of the system (16). In our study, an externally applied negative pressure produced a sudden and sustained increase in transmural pressure. As a result, MBV abruptly increased and...
then quickly decreased to a steady-state level below the ambient pressure baseline level. This steady-state response suggests that the myogenic response was continually engaged while negative pressure was maintained, an observation that is consistent with the concept of super regulation (22, 25). After an increase in transmural pressure induced by a rapid change from positive external pressure to ambient, the myogenic response did restore MBV back to baseline and thus approached an infinite gain response.

As mentioned earlier, the unique approach used in this report allowed us to raise transmural pressure by switching from ambient to negative pressure and by changing from positive to ambient pressure. After the initial rise in velocity, we noted an initial acute rapid rate of fall in blood velocity of $-2.16 \pm 0.24$ cm$^2$ s$^{-1}$. When we raised transmural pressure by the second method, i.e., by discontinuing positive pressure, we observed a rapid rise in MBV that was followed by an even more acute rapid rate of velocity reduction of $-3.31 \pm 0.038$ cm$^2$ s$^{-1}$ ($P < 0.014$). In addition, the quick reductions in VP associated with increases in transmural pressure suggest that a suction effect of the veins aided in the quick initial rise in MBV. These findings suggest that the initial level of transmural pressure may influence myogenic vasoconstriction; the lower the baseline transmural pressure and/or blood velocity, the more rapid the vasoconstrictor response. These detailed observations shown in humans are consistent with prior animal experiments (17, 26).

Parenthetically, these observations of a greater rate of fall in velocity for positive to ambient than for ambient to negative pressure argue against the possibility that our results can be explained by a venoarterial axon reflex (18). If this mechanism were the reason that an increased transmural pressure evoked vasoconstriction, then we would have expected the rate of fall in blood velocity to be greater when transmural pressure was raised by decreasing tank pressure than when by withdrawing positive pressure since the amount of venous distention would be greater during the former experimental condition.

MBV Responses to Muscle Contraction: Influence of Transmural Pressure

During start of the handgrip protocols, there was a brief 1- to 2-s quick reduction in MBV within 1–2 s to zero or negative velocities before MBV began to increase under the respective pressure conditions. Other studies (42) have shown this effect under ambient conditions, which is likely because of the contracting muscle compressing the blood vessels and creating high intramuscular pressures. This effect on muscle contraction leads to a work/flow mismatch and may lead to the release of metabolic productions that can cause vasodilation and make the evaluation of the myogenic response difficult. For this reason, a paradigm that would limit and control these variables was devised. We selected 25% MVC because this amount of contraction should limit flow to near zero under the three conditions (high, low, and normal transmural pressure). We felt that this would help ensure that the potentially confounding metabolic effects of contraction (2, 24) would be similar under the three experimental conditions. A very short single bout of contraction was selected. It was reasoned that one short contraction would limit the metabolic stimulus so that the potential effects of transmural pressure could still be evaluated. Five seconds of contraction was selected since it was possible that this exercise intervention would be of insufficient strength and duration to pre-

![Fig. 6. Perfusion pressure was calculated from venous pressure (light gray area; corrected to be additive) and mean arterial pressure (dark gray area). A: during changes to negative pressure (−50 mmHg), there were small changes in perfusion pressure with dramatic reductions in MBV (solid dark line). B: during the release of positive pressure (+50 mmHg) back to ambient pressure, there were small changes in perfusion pressure with quick reductions in MBV (solid dark line). Values are the group mean of $n = 4$ subjects in each pressure condition.](http://jap.physiology.org/)
vent autonomic engagement. Under these experimental conditions effects of differences in transmural pressure on flow velocity were still seen. Thus we conclude that in humans the myogenic response is operative after muscle contraction. These findings are consistent with prior animal studies (32, 39).

After handgrip, peak MBV rose above baseline values under all experimental conditions. On release of SHG, VP reductions corresponded to peak MBV in all conditions. After peak MBV was noted, the rate of fall in velocity toward baseline was inversely related to the transmural pressure. In this group of experiments, we found that there was a relationship between transmural pressure and when postcontraction peak MBV was noted. With −50 mmHg (increased transmural pressure), it occurred by the second HB (−2 s), with ambient pressure by the fifth HB (−5 s) and with positive pressure (decreased transmural pressure) by the sixth HB (−6 s). In addition, transmural pressure affected the rate of fall of blood velocity after peak MBV was achieved. Thus, with increased transmural pressure, the peak flow is higher, occurs earlier, and falls more rapidly. The physiological implications of these findings to flow regulation during rhythmic exercise needs to be further explored.

There have been a substantial number of animal studies examining the underlying mechanisms of the myogenic response (see reviews) (5, 21, 30, 35). However, there have been a limited number of human studies performed to examine this response, let alone its cellular mechanisms. Early human studies investigated the myogenic response indirectly by blood sampling (oxygen saturation) (1) and plethysmography after the release of negative pressure (transient transmural pressure reduction) (3, 14) or venous congestion (33). These reports suggested that the myogenic vasoconstriction occurred and was present even after transmural pressure was returned to baseline levels. Because of technical issues related to volume plethysmography, these studies could not evaluate flow for the first 8–10 s after negative pressure was withdrawn. In the present report, when negative pressure was withdrawn, we observed an initial fall in MBV within 1 s followed by a myogenic vasodilation period over the next 8 s and then a return to baseline MBV with 20 s. We did not observe the sustained flow reduction up to 1 min after release observed in prior reports (3, 14). It is unclear why our results are different from prior human reports. A recent study has suggested that arm elevation evokes transient and progressive vasodilation possibly related to venous emptying and myogenic mechanisms, respectively (40). Compared with the prior study, the myogenic vasodilator response presented in our report occurred earlier (3 vs. >10 s) and was of a different magnitude. These differences between the prior report and the present paper may be due to differences in study designs. Our results are consistent with rapid blood flow responses of vasoconstriction and vasodilation with negative and positive pressures, respectively, previously reported in animals (5, 21, 30, 35). These similarities include the effects of sustained positive and negative pressure on dynamic and static responses (5), the effects of baseline flow on the response, and the enhanced effectiveness of this response during muscle contractions (32, 39).

Myogenic vasoconstriction responses have been studied extensively in animals with clear changes in flow/velocity patterns with the identification of different phases (dynamic and static) (5). In our study, the dynamic (transient) myogenic response was able to be further delineated into two phases (rapid and less rapid) by derivative estimates. We believe that the present report is the first to characterize the myogenic response in such a fashion. We speculate that the phasic responses to increases in transmural pressure may reflect a more rate-sensitive response due to the influx of calcium into the smooth vascular muscle cell (27). This would suggest that a greater influx of calcium would be associated with a faster rapid vasoconstriction response. However, further studies are needed to examine this relationship. Myogenic vasodilation (reductions in transmural pressure) in animals has been less studied. Unlike vasoconstriction, vasodilation phases have not been identified. We speculate that reductions in intracellular calcium may be contributing to the vasodilation response (27). However, further studies are needed to determine whether changes in intracellular calcium are associated with myogenic vasodilation.

**Potential Limitations to the Study**

Alterations in PP may have occurred with the application of the neoprene arm cuffs due to venous congestion. To compensate for this possible effect, we used several different sized cuffs to securely, but not tightly, fit the subject’s forearm, and had all subjects rest ~20 min before testing after the arm cuffs were applied to establish a steady state in VP. Likewise, because of structural and response differences between the arteries and veins (15), external arm suction may have altered PP and contributed to the velocity changes observed. However, we observed rapid MBV vasoconstriction responses occurring within six HB with a relatively constant PP with the application of negative pressure. Therefore, another mechanism must be involved that leads to the reduction in MBV (vasoconstriction). We contend that this mechanism is the myogenic response. Lastly, we cannot totally exclude the possibility that metabolic factors were generated as blood velocity was reduced and that this velocity reduction evoked a metabolic vasodilation. Further studies will be necessary to better understand the relationship between metabolic and myogenic dilator systems.

This study represents the first time that flow responses to changes in transmural pressure were examined on a beat-to-beat basis by Doppler in humans at rest and after muscle contraction. The findings suggest that the myogenic response plays an important role in flow regulation in human subjects.

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