Resistance to pressure-induced dilatation in femoral but not saphenous artery: physiological role of latch?

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Large elastic arteries contain considerable amounts of vascular smooth muscle (VSM), and reductions in elastic artery lumen diameters due to VSM contraction at physiological pressures are reported to range from 10% to over 50% (Refs. 3 and 7; reviewed by Refs. 8 and 14). In support of the contention that elastic arteries are normally partially constricted at physiological pressures are early reports that, by altering the contractile state of VSM, perturbations resulting in reductions in sympathetic tone can increase elastic artery diameters [e.g., abdominal aorta (11, 19) and femoral artery (FA) (10)], and perturbations that increase sympathetic tone can reduce elastic artery diameters (10, 11). More recent studies have revealed that inhibition of nitric oxide synthesis decreases elastic artery diameters (29), supporting the hypothesis that large elastic arteries are normally partially constricted. Moreover, as organ metabolic demands increase, regulation of organ blood flow shifts upstream from terminal arterioles to organ feed arteries (reviewed by Refs. 2 and 31), and early work on this topic shows that even the elastic FA can dilate to participate in regulating hindlimb blood flow (18). Importantly, during the extreme conditions of hemorrhage or hypoxia, contraction of elastic and muscular arteries can result in dramatic reductions in lumen diameters by as much as 90% (reviewed by Ref. 14). Thus a generally accepted view of elastic arteries is that they are neither maximally dilated nor maximally constricted at physiological pressures but likely reside at an intermediate state between these boundaries such that diameter changes appropriate to physiological demands can occur.

One clear mechanical distinction that has emerged when large-diameter elastic arteries are compared with smaller-diameter muscular distributing arteries is that muscular arteries can constrict nearly maximally, but elastic arteries cannot constrict to small diameters as pressure increases above the physiologically “resting” state. For example, using isolated, pressurized dog arteries contracted with a maximum concentration of norepinephrine, Cox (3) showed that the ability of FA to constrict began to decline when luminal pressures were increased above ~100 mmHg, whereas for saphenous artery (SA), a muscular distributing blood vessel and main branch of the FA, the “distending” pressure was ~200 mmHg. Thus, as blood pressure increases, as can occur during activation of the sympathetic nervous system, large elastic but not smaller muscular arteries appear to dilate (reviewed by Ref. 8). We recently found a second mechanical distinction between large-diameter elastic and smaller-diameter muscular arteries (16): isolated rings of rabbit SA do not maintain strong tonic isometric contractions on stimulation with KCl as do rings of FA. This presents an apparent paradox. That is, large elastic and smaller (but still large) muscular arteries, such as, respectively, the FA and SA, behave isometrically in a way that appears counter to their in situ behaviors. FA maintains strong isometric force indefinitely, whereas SA does not (16), but...
under isobaric conditions, SA can constrict at high blood pressures, whereas FA cannot (3).

A hallmark of large elastic arteries is their ability to maintain high levels of tonic contraction with a high energy economy, and this has been viewed as a consequence of the physiological requirement for the VSM of elastic arteries to remain contracted to withstand the continuous imposed load exerted on the muscle by blood pressure (reviewed by Refs. 21, 22). Physiological, molecular, and modeling studies provide compelling evidence that slow motor protein isoforms expressed in the tonic VSM of large elastic arteries permit the formation of slowly or noncycling cross bridges (latch bridges), resulting in sustained isometric contraction (Refs. 5, 12, 15, and 27; reviewed by Refs. 1, 22, 34). Phasic smooth muscles of visceral organs such as the urinary bladder (detrusor) express fast motor proteins, do not enter a “latch state,” and do not maintain strong contractions for long durations (reviewed by Ref. 32). Importantly, on the basis of kinetic modeling and empirical data revealing differences between FA and SA in motor protein expression, myosin light chain phosphatase activity, rate of force redevelopment, and temporal isometric contraction profile, we proposed that the reason for the ability of FA but not SA to maintain maximum stress levels is that FA but not SA has the capacity to form latch bridges (16). The goal of this study was to compare directly the mechanical behaviors of isolated, pressurized FA and SA. The present study, therefore, represents an attempt to understand the physiological relevance of latch-bridge formation by comparing the pressure-diameter relationships in two “tonic” arteries.

METHODS

Tissue preparation and artery diameter. Arteries from female New Zealand White rabbits (3–4 kg) were prepared as previously described (28) and stored in cold (4°C) physiological saline solution [PSS; in mM: 140 NaCl, 4.7 KCl, 1.2 MgSO4, 1.6 CaCl2, 1.2 NaHPO4, 2.0 3-(N-morpholino)propanesulfonic acid adjusted to pH 7.4, 0.02 Na2EDTA to chelate heavy metals, and 5.6 D-glucose made with high-purity (17 MΩ) deionized water]. Arteries were cleaned of adhering tissue by microdissection (Olympus SZX12). In arteries used for experiments shown in Fig. 2, the endothelium was removed by gently rubbing the intimal surface with a metal rod, and passive diameters (i.e., diameters measured when arteries were not stimulated to contract) were recorded in tissues incubated both in normal PSS containing CaCl2 (+Ca2+) and in a Ca2+-free PSS (−Ca2+) prepared by omitting CaCl2 and adding 1 mM EGTA to chelate contaminating CaCl2. For all other experiments, the endothelium remained intact, and passive diameters were measured by incubation of arteries in normal PSS (+Ca2+). Arteries were cut into 6- to 8-mm-wide tubes, secured at each end to a stainless steel cannula by using 4-0 silk suture, and immersed in PSS in a temperature-controlled pressure myograph (Danish Myo Technology) positioned on the stage of an inverted microscope (Nikon) housing a video camera connected to an online imaging system so that arterial outer wall diameter could be continuously recorded. The myograph was designed to permit no flow changes in arterial luminal pressures, and the measurement of artery diameters under isobaric conditions and during ramp increases in pressure. KCl (110 mM substituted isosmotically for NaCl) was used to maximally contract arteries and was added to the superfusate. In one set of experiments, the α-adrenergic receptor agonist phenylephrine (PE) was added to the superfusate to cause arterial constriction. After equilibration in PSS at 37°C for 1 h, each artery was pressurized to 40 mmHg and contracted with KCl for 10 min to test for viability, then washed to permit relaxation, and the pressure was returned to 0 mmHg. Pressures used in this study ranged from 0 mmHg to 200–240 mmHg. The average mean arterial pressure measured in vivo for normal New Zealand White rabbits derived from 16 published studies is 79 ± 0.6 mmHg. Three examples of studies from which blood pressure measurements were obtained are Refs. 20, 26, and 35.

Steady-state protocol. Outer wall diameters were recorded under passive (P, Fig. 1A) and active (stimulated with KCl) conditions at pressures ranging from 0 to 120 mmHg (for Fig. 2), to 200 and 240 mmHg (for Fig. 3), and to 200 mmHg (for Fig. 5). In this protocol, arteries were held at a constant pressure when stimulated to contract. Therefore, the recorded active diameters achieved by the artery in response to stimulation with KCl (and PE for Fig. 5) at a given luminal pressure were labeled isobaric active constrictions (IACs, see Fig. 1A). The temporal sequence for the protocol (Fig. 1A) was to

Fig. 1. Diagrams depicting steady-state passive and active [isobaric active constriction (IAC)] diameter protocol (A) and pressure-ramp protocol (B). See text for the actual final levels of pressure achieved. The x-axis is time. P, passive; 30°, 30 s; 10°, 10 min.
record the passive diameter at 0 mmHg and then contract with KCl and record IACs at 30 s (for Fig. 2) and 10 min (for Figs. 2–5). After KCl washout (Fig. 1A, Wash) to permit complete VSM relaxation and arterial dilatation (10 min), luminal pressure was step-increased by 10, 20, 40, or 50 mmHg (20-mmHg pressure steps are shown in Fig. 1A). After equilibration at the new pressure (10 min), passive diameter was recorded, the artery was contracted with KCl, and the IACs at 30 s and 10 min were recorded. This sequence was repeated until the final pressure was achieved.

**Ramp protocol.** After completion of the steady-state protocol, the VSM of each artery was relaxed so that full dilatation was achieved, depressurized to 0 mmHg, and then repressurized to 50 or 60 mmHg. After constriction by application of KCl for 10 min, arteries were subjected to a ramp increase in pressure at a rate of 0.5 mmHg/s (Fig. 1B). Diameter changes were recorded continuously, but only diameter responses corresponding to each 10-, 20-, or 25-mmHg incremental increase were used in data analyses.

**Wall stress calculation.** Given the assumptions that arterial density = 1.060 (mg/mm³), artery volume (V) = artery weight/artery density, steady-state diameter was achieved within 10 min, and every part of the artery tube of length L contracted to the same diameter, the inner diameter (a) could be accurately determined from the measured outer diameter (b) using the equation, V = Lπ(b² - d²), and wall stress (S) could be accurately calculated by multiplying luminal pressure (P) by the quotient of inner diameter (a) over twice the wall thickness (b - a), i.e., S = P(a/(b - a)) (4).

**Statistics.** ANOVA and the Student-Newman-Keuls test, or the Student’s t-test, were used where appropriate to determine significance, and the null hypothesis was rejected at P < 0.05. The population sample size (n value) refers to the number of rabbits, not the number of tissues. Statistical analyses and curve fitting were performed by using Prism 3.02 (GraphPad Software, San Diego, CA).

**RESULTS**

**Passive and active (IAC) pressure-diameter relationships.** For both SA (Fig. 2A) and FA (Fig. 2C), passive diameters measured in arteries incubated in Ca²⁺-containing (+Ca²⁺) record the passive diameter at 0 mmHg and then contract with KCl and record IACs at 30 s (for Fig. 2) and 10 min (for Figs. 2–5). After KCl washout (Fig. 1A, Wash) to permit complete VSM relaxation and arterial dilatation (10 min), luminal pressure was step-increased by 10, 20, 40, or 50 mmHg (20-mmHg pressure steps are shown in Fig. 1A). After equilibration at the new pressure (10 min), passive diameter was recorded, the artery was contracted with KCl, and the IACs at 30 s and 10 min were recorded. This sequence was repeated until the final pressure was achieved.

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**RESULTS**

**Passive and active (IAC) pressure-diameter relationships.** For both SA (Fig. 2A) and FA (Fig. 2C), passive diameters measured in arteries incubated in Ca²⁺-containing (+Ca²⁺)
and Ca²⁺-free (−Ca²⁺) solutions were identical (Fig. 2, A and C; compare solid symbols), indicating that these large distributing arteries did not develop myogenic tone. The steady-state (10 min) IAC produced by stimulation of SA with KCl was sufficient to cause significant reductions in diameters compared with passive diameters at all pressures, including 0 and 120 mmHg (Fig. 2A; compare open with closed symbols). The degree of steady-state constriction increased progressively from a small reduction in diameter of 0.3 mm produced at 0 degree of steady-state constriction increased progressively from a small reduction in diameter of 0.3 mm produced at 0 and at 120 mmHg (Fig. 2A). At the highest pressure measured, the absolute amount of constriction was reduced only slightly from that produced at 60 mmHg (Fig. 2A, double arrows next to the 60-mmHg data points), and at 40 mmHg, the reductions in diameter for FA at 30 s and 10 min were, respectively, 0.6 and 1.1 mm (Fig. 2C, double arrows next to the 60-mmHg data points). This temporal behavior in the degree of change in diameters (stronger early than late constriction in SA and stronger late than early constriction in FA) mimics the temporal behavior in isometric force produced in isolated artery rings (16).

Pressure-diameter and stress-diameter relationships in endothelium-intact tissues. A major distinction between FA and SA is that the degree of IAC was limited by pressure at relatively low pressures in FA compared with SA (compare Fig. 2, A and C). To determine whether this limitation in the ability of FA to constrict was due to disruption of internal

Also different when SA and FA were compared was the temporal response to KCl. In particular, at most pressures the degree of steady-state IAC in the SA was less (Fig. 2B), and in the FA was more (Fig. 2D), than that produced initially (30 s) on stimulation with KCl. That is, when stimulated with KCl, SA constricted to a greater degree early and then dilated, whereas FA constricted slowly to a maximally small diameter within 10 min. For example, at 60 mmHg, the reductions in diameter from the passive level to that induced by IAC for SA measured at 30 s and 10 min were, respectively, 0.7 and 0.6 mm (Fig. 2A, double arrows next to the 60-mmHg data points), and at 40 mmHg, the reductions in diameter for FA at 30 s and 10 min were, respectively, 0.6 and 1.1 mm (Fig. 2C, double arrows next to the 60-mmHg data points). This temporal behavior in the degree of change in diameters (stronger early than late constriction in SA and stronger late than early constriction in FA) mimics the temporal behavior in isometric force produced in isolated artery rings (16).

Fig. 4. A: IAC, B: FA, 60–120 mmHg. C: SA, 60–140 mmHg. D: summary of A and B. Active pressure-normalized diameter curves for femoral (FA) and saphenous (SA) arteries produced by the steady-state protocol where arteries were constricted isobarically for 10 min (IAC; open symbols in A–D) and produced by arterial constrictions at 60 mmHg for 10 min followed by ramp increases in pressure (P-Ramp) to 120 mmHg for FA (B, filled squares) and 140 mmHg for SA (C, filled circles). D summarizes results from A and B. Diameters were normalized so that the fully dilated and constricted diameters were, respectively, 1 and 0 (see A, dotted horizontal lines and labels). Double vertical dotted lines at ~80 mmHg indicate the average – SE and average + SE of mean arterial blood pressures published for New Zealand White rabbits. Data are means ± SE; n = 3–7. * P < 0.05 for FA vs. SA (A), IAC vs. P-Ramp (B). In B, 10° = 10 min.

Fig. 5. Active pressure-normalized diameter curves for FA (A) and SA (B) produced by the steady-state protocol where arteries were constricted isobarically for 10 min (IAC) with KCl (IAC, KCl) and phenylephrine (PE) at 1 µM (IAC, 1 PE) and 10 µM (IAC, 10 PE) and constricted for 10 min by 1 µM PE (P-Ramp, 1 PE) and 10 µM PE (P-Ramp, 10 PE) at 50 mmHg followed by ramp-increases in pressure to 150 mmHg for FA (A) and to 200 mmHg for SA (B). Diameters were normalized so that the fully dilated and constricted diameters were, respectively, 1 and 0, as in Fig. 4. Data are means ± SE; n = 3–6. * P < 0.05, IAC vs. P-Ramp responses when tissues were stimulated with 1 µM PE; IAC vs. P-Ramp responses when tissues were stimulated with 10 µM PE.
elastic lamina and damage to some medial smooth muscle located near the intima caused during the mechanical disruption of the endothelium or to a reduced ability of FA to generate stress, we subjected tissues that were not mechanically denuded of endothelium to a steady-state protocol, and pressures were increased to 200 mmHg for FA and 240 mmHg for SA. In addition, artery lengths and weights were recorded at the end of the experiment to permit calculation of wall stress. As reported in Fig. 2, on stimulation with KCl, FA failed to constrict significantly at pressures that did not limit SA constriction (Fig. 3A). The failure of FA to constrict at pressures that did not limit SA was not the result of a weaker ability of the VSM of FA to contract compared with SA, because the calculated maximum active stress induced by FA and SA were identical [-1 MPa, Fig. 3B, inset; compare peak values of SA and FA parabolic curves representing active stress calculated by subtracting passive from total stress values]. Moreover, the failure of FA to constrict compared with SA at high pressures was not due to a limitation in the ability of the FA to constrict because the absolute constriction was greater in FA compared with SA (Fig. 3B, inset; active stress curve for FA is broader than that for SA). Finally, the failure of FA to constrict at high pressures was not due to alterations resulting from endothelial denudation. However, the percent constriction in FA was shifted slightly to the left in endothelium-denuded compared with endothelium-intact arteries (compare Figs. 2C, open squares, and 3A, open squares), suggesting that agents released by endothelium, or mechanical denudation, may mildly alter the degree of IAC produced at high pressures. It was possible that release of relaxing agents from smooth muscle during the 10-min exposure to KCl limited the degree of constriction achieved by FA. However, in one FA tissue, no apparent differences in diameter responses were identified when the superfusate was washed and replaced every minute for each 10-min contraction with KCl during a full IAC protocol from 50 to 150 mmHg. These data suggest that if relaxing agents were released, the concentrations achieved within 10 min were insufficient to affect KCl-induced constriction.

The maximum active stress produced by FA occurred when arteries were stimulated with KCl at a relatively modest pressure of ~160 mmHg, and this corresponded to an artery diameter of 2.16 mm (Fig. 3A, top horizontal dashed line; Fig. 3B, right vertical dashed line). In SA, maximum active stress occurred at the considerably higher pressure of ~220 mmHg, which corresponded to an arterial diameter of 1.54 mm (Fig. 3A, bottom horizontal dashed line, and Fig. 3B, left vertical dashed line). Because Laplace’s law applied to arterial tubes (9) states that wall stress (S) is a function of the product of luminal pressure (P) and arterial diameter (d; i.e., $S \propto P \cdot d$), for a given stress, the larger diameter FA would support a lower pressure than the smaller diameter SA. This relationship was readily apparent when pressures and IAC diameters of SA and FA were compared at ~0.6 MPa. For example, the IAC diameter and pressure at ~0.6 MPa for SA were, respectively, ~1.35 mm and 160 mmHg, and for FA they were, respectively, ~1.9 mm and ~110 mmHg. Note that 1.35 × 160 = 1.9 × 110. These data support the hypothesis that the difference in absolute artery diameters is responsible for the difference in %IAC produced at high pressures when FA and SA are compared.

Diameter responses during steady-state IAC and during ramp increases in pressure after constriction at low pressure with KCl. Rings of FA can maintain high levels of isometric stress for a prolonged duration when stimulated to contract with KCl because of formation of latch bridges, whereas SA does not undergo latch and cannot maintain high levels of tonic isometric stress (16). However, at the average mean arterial pressure for rabbit of ~80 mmHg, FA does not constrict fully, whereas SA does (Fig. 4A, and see Figs. 2, A and C, and 3A). This appears paradoxical because the rationale for the necessity of VSM molecular motor proteins that permit formation of latch bridges and a strong and sustained isometric contraction does not appear to translate to the apparent function of VSM as the motors that alter both the caliber and compliance (14) of pressurized tubes. Thus this issue is one of translational interest because it is an attempt to understand in vivo arterial function on the basis of in vitro biomechanical and biochemical studies. That is, to fully understand the mechanisms regulating VSM contraction, the preparation of choice has usually been isolated rings or strips of VSM preloaded by stretching to its optimum length for muscle contraction ($L_o$) and stimulated to contract isometrically. This preparation permits highly controlled biomechanical and biochemical analyses to be performed simultaneously. However, at physiological pressures, the VSM of arteries are not maintained in a purely isometric state and, on stimulation, would likely actively constrict under pseudoisobaric conditions because of arterial VSM cell shortening. For example, when stimulated with KCl at arterial pressures of ~80 mmHg, fully relaxed FA would constrict from ~2.25 to ~1.35 mm (Fig. 3B, dotted line connecting passive- and active diameter-stress values at 80 mmHg), and because of Laplace’s law (9), wall stress would decrease from ~0.7 to ~0.2 MPa as the VSM cells shorten and the arterial tube diameter decreases. A purely isometric VSM contraction when cells are stretched to $L_o$ may be a very rare event in a large artery under physiological conditions. What, then, is the role of latch-bridge formation in large elastic arteries? To answer this question, passive arteries maintained under an isobaric condition of 60 mmHg (approximately normal physiological pressures) were constricted with KCl for 10 min and then subjected to a ramp increase in pressure. The question addressed was, do constricted FA and SA respond to ramp increases in pressure by immediately dilating to the diameter achieved during a steady-state IAC, or can one or both of these arteries resist the pressure increase by remaining constricted?

Steady-state pressure-active diameter curves for FA and SA, normalized to fully dilated (passive) and fully constricted (IAC at 40 mmHg for FA and 60 mmHg for SA) diameters were sigmoidal, and because FA constricted to a lesser degree than SA for a given pressure above 80 mmHg, the SA curve was rightward-shifted compared with the FA curve (Fig. 4A). Although the FA was more dilated than SA at pressures ranging from 80 to 120 mmHg during steady-state IAC, a fully constricted FA remained constricted when subjected to a ramp increase in pressure from 60 to 120 mmHg (Fig. 4B). The level of constriction held during the ramp increase in pressure was significantly greater than the IAC diameter that the artery could produce (Fig. 4B). That is, FA could bear higher pressures when the VSM was contracted and the artery fully constricted at lower pressures and ramped to higher pressures than when the VSM was contracted and the artery made to contract.
isobarically at the higher pressures. This was not true for SA, where both IAC and pressure-ramped diameters were identical (Fig. 4C). The level of constriction measured during a ramp increase in pressure was retained by FA for at least 90 s (the approximate duration of the pressure ramp). Moreover, FA appeared to be very slow to dilate toward the IAC values because arteries subjected to a ramp pressure increase from 60 to 120 mmHg “slipped” only by ~36% when held for 10 min at 120 mmHg after the pressure ramp (Fig. 4B, arrow from solid square to gray diamond, n = 2).

These data suggest that one role of latch-bridge formation in FA was to resist dilatation on increases in pressure such that FA could remain for many minutes in a constricted state on transient increases in pressure of at least 40 mmHg above the normal physiological level of ~80 mmHg. Moreover, these data support the contention that SA may not have a physiological requirement for latch-bridge formation because SA, unlike FA, could constrict between 100% and ~75% at pressures between, respectively, 80 and 120 mmHg (Fig. 4C), presumably due to their smaller diameters (see Fig. 3). In summary, FA appears to possess at least two pressure-active diameter curves (Fig. 4D, open and solid squares) and likely can be made to constrict anywhere between the two curves, depending on the sequence of events (constriction followed by pressure increase, or pressure increase followed by constriction), whereas SA has a unique pressure-active diameter relationship not dependent on event sequence (Fig. 4D, open circles).

Diameter responses during steady-state IAC and during ramp increases in pressure after constriction at low pressure with the α-adrenergic receptor agonist PE. KCl bypasses receptors to contract VSM by causing membrane depolarization. To determine whether activation of a G protein-coupled receptor agonist produces similar results to KCl, FA and SA were stimulated with PE, and diameters were measured at steady-state IAC and after ramp increases in pressure. In FA, 1 μM PE produced responses much like KCl, and 10 μM PE shifted the steady-state IAC curve to the right but still could not cause FA to constrict when pressure was 150 mmHg (Fig. 5A). However, when subjected to a ramp increase in pressure from 50 to 150 mmHg while constricted by 1 and 10 μM PE, FA maintained the constricted state (Fig. 5A, P-Ramp, 10 and 1 PE), as seen also when constricted with KCl (see Fig. 4B). That is, FA could withstand high distending pressures when first constricted with KCl or PE. PE produced a response that was also similar to KCl in SA, although the effects were somewhat more complex (Fig. 5B). In particular, some, but not all, SAs tested could maintain a constricted state when stimulated with 10 μM PE and subjected to a ramp increase in pressure from 50 to 200 mmHg. Because of the high variability in the response, the IAC and pressure-ramped final diameters at 150 and 200 mmHg were not significantly different.

**DISCUSSION**

Our recent study shows that the large elastic FA fits the classification of a tonic smooth muscle that depends on latch-bridge formation for the maintenance of strong isometric stress and that the tonic phase of contraction in the smaller more muscular SA is weaker than the early phasic phase by approximately one-half because of a lack of latch-bridge formation (16). The present study extends our previous work by exploring the functional consequences of differential motor protein expression and muscle contractile behavior by comparing diameter responses in pressurized FA and SA. The results support the hypothesis that latch-bridge formation by FA permits maintenance of a constricted artery in the face of pressure increases above the normal mean arterial pressure. Perhaps the most novel aspect of this work is that it expands the proposed role of latch-bridge function. The original concept is that latch-bridge function is solely to maintain strong isometric force at Lₐ (reviewed by Ref. 22), a mechanical situation we believe is unlikely to occur routinely in an artery tube in vivo. In our working hypothesis, the latch bridge is envisioned to prevent motor protein “slippage” at short muscle lengths so that the arterial tube can remain constricted for some finite time under conditions that cause increased blood pressures above the resting levels.

When employing a protocol whereby luminal pressure was increased stepwise in relaxed arteries and the VSM was permitted to contract to KCl under an isobaric condition (IAC), SA was found to constrict to a greater degree than FA at all pressures above the resting physiological range up to a pressure of ~200 mmHg. This behavior in rabbit FA and SA was qualitatively identical to and quantitatively similar to that...
measured in dog FA and SA (3). As in dog, the maximum stress produced at the optimum diameter for muscle contraction was not different when FA and SA were compared, indicating that the greater ability of SA to constrict at pressures between 100 and 200 mmHg resided in the arterial tube geometry (3). That is, because the passive tube diameter of SA is smaller than FA at each pressure (see Fig. 3A) and the law of Laplace states that wall stress is proportional to the product of pressure and diameter (9), smaller diameter arteries will be subjected to lower wall stresses for a given luminal pressure. Thus maximum stress generated by the VSM of SA occurred at a diameter of ~1.54 mm and pressure of ~220 mmHg (see Fig. 3B). However, at this pressure, activation of VSM could not cause SA to constrict (see Fig. 3A) but would have shifted some of the load from parallel elastic elements to the contractile proteins and elements in series with them (see Ref. 8 for review). Perhaps more importantly, at the normal resting mean arterial pressure for rabbit of ~80 mmHg, SA could constrict nearly maximally, and even at 120 mmHg, SA could constrict ~80% from a fully dilated diameter. The VSM of SA, therefore, does not appear to have a physiological requirement for sustaining a strong, isometric contraction. Rather, when considering pressures between 60 and 120 mmHg, the VSM of SA appears to operate within a broad isotonic but narrow isometric window (Fig. 6A, hatched area). Rapid movement between these states may have more profound physiological relevance than prolonged maintenance of high isometric stresses. Such behavior does not support a requirement for lattice bridges but does appear to support a requirement for “fast” motor proteins.

Qualitatively, SA and FA behave similarly. However, at pressures above 80 mmHg, FA cannot generate the same degree of constriction as SA. This limitation is not due to an inability of FA to generate high stresses because the maximum stress produced by FA and SA was equivalent. Moreover, FA did not appear to be limited by a reduced ability of VSM to shorten because the absolute amount of constriction produced by FA was greater than that produced by SA, and this was evident not only when absolute diameter curves were compared (see Fig. 2, A and C) but also when the widths of the active stress-diameter curves were compared (see Fig. 3B, inset). The reason that FA could not generate the same degree of constriction as SA at pressures above 80 mmHg appears to reside in the geometric constraint of a tube and the constraints imposed by the ascending limb of the length-tension curve of VSM, which limits the degree of stress that can be produced at short lengths (13, 17). Such a relationship agrees well with data presented in the late 1970s by Cox (3), who compared pressure-diameter curves in dog iliac artery, FA and SA, and proposed that the differential ability of FA and SA to constrict at high pressures resided, in part, in the differences in wall tensions inherent in these two arteries that display large differences in lumen diameters. In short, because the FA is a larger-diameter, thinner-walled tube than the SA, for a given wall stress, the luminal pressure in the SA will be greater than that in the FA. At each comparable relative diameter, pressures on the ascending limb of the active stress-diameter curve, a surrogate for the VSM length-active stress curve (7), will therefore be higher for SA than FA. Thus, for comparable pressures above 80 mmHg (e.g., 120 mmHg, Fig. 6), constriction from the fully dilated, passive state to the fully constricted active state will proceed further down the stress-diameter curve in SA than in FA (Fig. 6, compare the lengths of the dashed arrows connecting iso-baric diameters at 120 mmHg for SA in A with FA in B). However, we propose that one previously unrecognized function of the latch bridge is to extend the active stress-diameter curve (Fig. 6B, dotted line labeled “Proposed Extension”) such that FA could remain at small diameters when first constricted to that diameter at physiological resting arterial pressures before the loading that occurred when subjected to higher pressures. That is, we propose that the dramatic increase in the absolute amount of constriction that can be maintained at high pressures in FA subjected to a pressure-ramp protocol is due to latch-bridge formation.

It is noteworthy that the extent of constriction induced by KCl in FA was greater at 10 min than 30 s and that the opposite was true for SA because our previous study (16) shows that the degree of isometric contraction at L0 in FA is also greater at 10 min than 30 s, and the opposite is true for SA. The level of stress produced under isometric contraction is dependent on the number of attached cross bridges (5), but under isobaric shortening, the number of attached cross bridges likely declines as the muscle cells shorten and move down their length-tension curves. Thus the slower development of both maximum isometric contraction and isobaric constriction by FA compared with SA may reflect the slower cross-bridge cycling rate in FA caused by slow motor protein isoform expression and formation of latch bridges (16) that impose an internal load against which cycling cross bridges must act (6).

Perspectives. It is now well established that elevated large-artery stiffness is associated with morbidity and fatal cardiovascular events, including cardiac failure and microvascular disease (reviewed by Refs. 24, 25, 30, 33). A proposed link between increased elastic artery stiffness and cardiovascular events involves augmentation of arterial pressure waves because of increased pulse wave velocity and earlier pulse wave reflection from the periphery (reviewed by Ref. 23). A reduced ability of large elastic arteries to constrict would lead to dilated and stiffened arteries, especially at the higher blood pressures witnessed in hypertension and aging. The present study proposes that appropriate VSM contraction and latch-bridge formation may enhance the ability of elastic arteries to remain constricted and resist pressure increases and, thus, remain compliant. Additional experiments directed toward understanding the relationships between VSM molecular motor regulatory mechanisms and in vivo arterial function (i.e., translational studies) will likely provide novel insights into mechanisms causing vascular dysfunction related to cardiovascular morbidity.

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