Biomechanical response of femoral vein to chronic
elevation of blood pressure in rabbits

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noral deep vein thrombosis and valvular dysfunction induce
venous hypertension. To know the effects of the hypertension
on venous mechanics, blood pressure in the left femoral
vein in the rabbit was chronically elevated by the constriction
of the left external iliac vein. Wall dimensions and biomechani-
cal properties of the femoral vein were studied in vitro at 1,
2, or 4 wk after surgery. Blood pressure measured immedi-
ately before the animal was killed was significantly higher in
the left femoral vein than in the sham-operated, contralat-
eral vein. Wall thickness was increased by blood pressure
ereation even at 1 wk, which restored circumferential wall
stress to a control level. The stress was kept at normal up to
4 wk. Vascular tone and vascular contractility were in-
creased by the elevation of blood pressure; however, wall
elasticity and compliance were kept at a normal level. These
results are very similar to those observed in hypertensive
arteries, indicating that not only arteries but veins optimally
operate against blood pressure elevation.

hypertrophy; remodeling; wall stress

MANY EXPERIMENTAL STUDIES and clinical observations
have shown that arteries change their dimensions and
properties in response to the chronic elevation of blood
pressure (10, 14, 16, 17, 19, 41). One of the specific
biomechanical phenomena appearing in response to
arterial hypertension is wall thickening, which re-
stores wall hoop stress to a normal, control level. In
addition, vascular tone is increased, although arterial
elasticity at in vivo working pressure changes to an
optimal level. For example, Matsumoto and Hayashi
(26, 27) demonstrated in the rat with induced Gold-
blatt hypertension that the aortic hoop stress at in vivo
systolic pressure, which should have been high soon
after the induction of hypertension, was rapidly (with-
in 2 wk) restored to a physiologically normal level
because of wall hypertrophy and remained at the nor-
mal level thereafter. On one hand, the elastic modulus
of the wall, which was also high soon after the induc-
tion of hypertension, became normal after a relatively
long period of time, i.e., at 16 wk. In addition, Fridetz
et al. (8, 9) have shown that normal vascular tone related
to the smooth muscle cell, which is the sensing and
effecting element of the adaptation process, was in-
creased during the early phase of arterial adaptation to
hypertension.

As far as we know only Monos et al. (30, 32) directly
studied the biomechanical effects of the chronic eleva-
tion of blood pressure in the vein, and much less is
known about the remodeling of the venous wall com-
pared with arteries. They chronically increased blood
pressure in the vein of the lower extremity by exposing
rats to head-up tilt for 2 wk, and they studied the
pressure-diameter relations and wall thickness of the
saphenous vein excised from the animals. When com-
pared at the same pressures, the external diameter
was larger and the wall distensibility was smaller in
the tilt animals than in the age-matched, nontilt ani-
imals, although there was no difference in the wall
thickness between them. Clinically, chronic venous
diseases like iliopheimal deep vein thrombosis and val-
vular dysfunction, which are accompanied by varicos-
ties, edemas, pigmentation, and so on, induce venous
hypertension (1, 23, 36, 39, 52). Applying an indirect
method to the patients having such chronic venous
insufficiency as a combination of reflux and obstruc-
tion, Neglen and Raju (35) observed that the femoral
and the popliteal veins, which were most likely exposed
to high blood pressure, were less compliant than those
in normal, healthy patients. However, there is a great
paucity of information related to the effects of blood
pressure elevation on venous mechanics.

We hypothesized that the vein exhibits biomechani-
cal responses to the chronic increase of blood pressure
similar to the artery, because like arteries, veins also
consist of three layers each of which contains many of
the layer-specific constituents (elastin, collagen, and
smooth muscle cell) found in the arterial wall, although
the content and distribution of these constituents differ
from those in arteries. The purpose of the present
study was to know the detailed biomechanics of the
vein chronically exposed to high blood pressure and to

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compare the results with those observed in hypertensive arteries.

MATERIALS AND METHODS

In vivo experimental procedures and materials. Eighteen female Japanese white rabbits weighing 3.0 ± 0.1 (means ± SD) kg were used for the experiments. After each animal was anesthetized by the injection of pentobarbital sodium via the auricular vein, the external iliac veins at both sides were exposed. At a position in the left external iliac vein ~5 mm proximal from the deep femoral vein, a syringe needle having the external diameter of 0.55 mm was placed beside the external iliac vein in parallel, and the vein and the needle were tied together with 4-0 braided silk suture from the outside. The needle was then gently pulled out. With this procedure, almost the same degree of constriction was made in the left external iliac vein (Fig. 1). This treatment was not applied to the right external iliac vein. As will be mentioned below (see RESULTS), the blood pressure in the left femoral vein was significantly increased (HP group), whereas the blood pressure in the contralateral femoral vein was maintained at normal level (Sham group).

At 1, 2, or 4 wk after the operation (6 animals for each period), each animal was anesthetized with pentobarbital sodium, and both femoral veins were exposed in a supine position. Blood pressure in each of the veins was measured using a plastic water column via an indwelling needle (0.55 mm in diameter) inserted through the deep femoral vein. The surface of each femoral vein was then marked with saturated gentian violet solution at the interval of 3 mm to identify the in vivo extension ratio (Δi), which is the ratio of in vivo vascular length to the length measured after excision. An ~10-mm-long tubular segment was excised and kept in Krebs-Ringer solution at 4°C until biomechanical tests were performed within 24 h after excision.

These animal experiments were done under the “Guidelines for Animal Experiments,” Graduate School of Engineering Science, Osaka University.

Biomechanical tests. The experimental system, shown in Fig. 2, was used for the measurements of intraluminal pressure-external diameter (P–D) data. Although axial force was also measured using a load cell incorporated in the system, the data were not used for the present study. A tubular specimen was mounted in the bath filled with Krebs-Ringer solution, which was kept at 37°C and aerated with a 95% O2-5% CO2 gas mixture. It was then extended to the in vivo length in reference to the above-mentioned markers dotted on the specimen surface. A diaphragm-type actuator, which was controlled with a sequencer, was used to apply internal pressure to the specimen. The vessel was inflated and deflated at a rate of ~1.3 mmHg/s. The pressure was measured with a fluid-filled pressure transducer (MPU-0.5-290-0-3, Orientec; Tokyo, Japan), while the external diameter of the specimen was determined with a video dimension analyzer (C3161, Hamamatsu Photonics; Hamamatsu, Japan) combined with a CCD camera (WV-BD400, Panasonic; Osaka, Japan). The accuracies of the measurements of pressure and diameter were ±0.5 mmHg and ±5 μm, respectively.

First, each vein was preconditioned by means of several inflation-deflation loops between the internal pressures of 0 and 30 mmHg until the P–D curve became reproducible. The ascending limb of the last stable loop was used for data analysis under normal conditions (normal vascular smooth muscle tone). Internal pressure was then maintained at 10 mmHg, and norepinephrine (NE) was gradually added to the bath. When vascular contraction became maximal, pressure was lowered to 0 mmHg and subsequently raised to 30 mmHg, yielding the P–D0 curve under maximal contraction. After the vessel was rinsed with Krebs-Ringer solution, internal pressure was set back to 10 mmHg, and papaverine (10^-4 mol/l) was added to the bath. Femoral veins in the rabbit are completely relaxed with this concentration of papaverine. When dilatation reached its maximum, several inflation-deflation loops were performed between 0 and 30 mmHg until the P–D0 curve became reproducible. The ascending limb of the last stable curve was used for data analysis as a representative of vascular response under total relaxation. For simplicity, we shall henceforth refer to normal resting (Krebs-Ringer solution), maximally contracted (NE), and totally dilated (papaverine) conditions as the “normal,” “active,” and “passive” conditions, respectively.

After pressure-diameter tests, a 0.5-mm-thick ring was cut out from the middle part of each vascular segment. The ring was placed in a physiological saline solution bath, which was kept at 37°C, and its cross-sectional image was taken into an image analyzer (PIAS-III, PIAS; Osaka, Japan) via a stereoscope (SMZ-10, Nikon) and a CCD camera (WV-BD400, Panasonic; Osaka, Japan). The external and internal diameters (di and do, respectively) of the ring specimen were measured by using the analyzer from four directions at the angular
interval of 45°. Results were based on the average of the measurements from these directions.

Data analysis. Considering that the venous wall is thin, wall stress in the circumferential direction (i.e., circumferential wall stress or wall hoop stress) was calculated from the following Laplacian equation (19)

\[
\sigma_s = \frac{P_i D_i}{2T}
\]

where \(D_i\) and \(T = (D_o - D_i)/2\) are internal diameter and wall thickness at each pressure, respectively. \(D_i\) was calculated from \(d_o, D_o, D_i\), and \(\lambda_s\), assuming the incompressibility of wall material (3, 4)

\[
D_i = \sqrt{D_o^2 - d_o^2}/\lambda_s
\]

The vascular compliance, i.e., the distensibility of wall, was calculated from (12, 16, 17)

\[
C_v = \frac{\Delta V}{\Delta P} = \frac{\Delta P (2D_o + \Delta D_i)}{\Delta P D_i^2}
\]

where \(\Delta P_i\) is the increment of pressure at \(P_i\) (2.5 mmHg), and \(\Delta D_i\) is the increment of internal diameter developed by \(\Delta P_i\).

The incremental elastic modulus of wall was calculated from (16–18)

\[
H_\text{wall} = \frac{2D_o}{D_o^2 - D_i^2} \left( \frac{\Delta P D_i^2}{\Delta D_o} + P_i D_i \right)
\]

To express the magnitude of vascular contraction, we used diameter response given by (5)

\[
\frac{\delta D_o}{D_o} = \frac{D_e - D_o}{D_o} \quad \text{or} \quad \frac{D_e - D_o}{D_c}
\]

where \(D_p, D_o,\) and \(D_c\) are external diameters under passive, normal, and active conditions at each internal pressure.

Statistical analysis. All data were expressed as means ± SD. ANOVA was used to evaluate the dependence of dimensional and mechanical parameters on experimental periods. Actually, however, there were no significant temporal changes in all the parameters. Comparisons between the two groups (HP group and Sham group) at each experimental period were made with Student's t-test. Differences were considered to be significant for \(P < 0.05\) (5%).

RESULTS

Blood pressure measured immediately before the animal was killed was significantly higher in the left femoral vein (HP group; 16.5 ± 3.1, 16.9 ± 1.6, and 14.8 ± 1.3 mmHg at 1, 2, and 4 wk, respectively) than in the contralateral vein (Sham group; 8.0 ± 1.3, 9.1 ± 1.6, and 8.4 ± 1.5 mmHg) at all the periods (Fig. 3); blood pressures in HP group were almost twice as high as those in Sham group. In our preliminary experiments using several other animals, blood pressure measured soon after the venous constriction was almost the same as that measured at the time of the same animal being killed after 1 to 4 wk. Therefore, we estimated that high blood pressure in the left femoral vein was also maintained throughout the experimental periods.

At each period, there was no significant difference in the internal diameter of the vein at in vivo pressure (i.e., working blood pressure) between the two groups under normal conditions, although the HP group seemed to have a smaller diameter than the Sham group at 4 wk (Fig. 4). On the other hand, the wall thickness was larger in the HP group (44 ± 15, 50 ± 23, and 59 ± 17 μm at 1, 2, and 4 wk, respectively) than in the Sham group (24 ± 7, 32 ± 17, and 32 ± 12 μm); the differences were statistically significant at 1 and 4 wk. Therefore, the external diameter was increased by the elevation of blood pressure. Because of the wall hypertrophy, circumferential wall stresses in the HP group at in vivo pressures under normal conditions (36.8 ± 18.6, 42.7 ± 17.2, and 28.2 ± 14.4 kPa at 1, 2, and 4 wk, respectively) were almost the same as those in the Sham group (38.5 ± 17.3, 37.3 ± 11.0, and 28.6 ± 8.5 kPa) regardless of the experimental period (Fig. 5); that is, wall hoop stress was independent of in vivo working blood pressure. Although the stress should have been increased by the elevation of blood pressure soon after the operation, it was restored to normal levels within 1 wk and was kept at normal up to 4 wk.

Examples of the relations between internal pressure and distension ratio are demonstrated in Fig. 6, where the distension ratio \((D_d/D_o)\) is the ratio of external
diameter at each pressure under normal or active conditions (\(D_o\)) to that measured at 10 mmHg under passive conditions (\(D_s\)). The shape of the relations obtained under passive conditions was almost similar to that observed under normal conditions in either the HP group or the Sham group, as can be seen from the data of diameter response that are shown below (see Fig. 8). In the HP group, the administration of NE (active condition) contracted the vein and shifted the curve toward the left; however, it developed almost no contraction in the Sham group. The relations observed at 1 and 2 wk were almost the same as those obtained at 4 wk.

There were no significant differences in the vascular compliance and incremental elastic modulus at each in vivo pressure between the two groups under normal conditions at all the experimental periods (Fig. 7). However, the diameter responses of the vein at each pressure were larger in the HP group than in the Sham group, both under normal and active conditions; the differences were statistically significant below 10 mmHg under active conditions (Fig. 8). Similar results were also observed at 1 and 2 wk. In all experimental periods, the diameter response at in vivo pressure was larger in the HP group than in the Sham group, both under normal and active conditions; the differences between the two groups were significant under active conditions at 1 and 4 wk (Fig. 9).

**DISCUSSION**

Not only experimental studies (e.g., 8, 13, 26, 50, 54) but also clinical observations (e.g., 11, 40, 41) have shown that arteries change their dimensions and properties in response to the chronic increase of blood pressure. One of the specific biomechanical manifestations to arterial wall adaptation in response to hypertension is wall hypertrophy. Because of this manifestation, wall hoop stress is most likely restored to a normal value even under hypertension. In addition, arterial stiffness at in vivo working blood pressure changes to an optimal level. For example, Matsumoto and Hayashi (26, 27) have demonstrated in the rat with induced Goldblatt hypertension that the aortic hoop stress at in vivo pressure was rapidly returned to a physiologically normal level, i.e., within 2 wk after the induction of hypertension and remained at the level, thereafter, and that the elastic modulus of wall at the in vivo pressure became a normal value after a
relatively long period of time, i.e., at 16 wk after the induction of hypertension. In addition, Fridetz et al. (8, 9) have shown in a rat model with abdominal aortic ligation that the vascular smooth muscle cell, which is the sensing and effecting element of the adaptation process, is activated during the early phase of arterial adaptation to hypertension. Furthermore, there are many studies showing that arteries change the internal diameter in response to blood flow change keeping wall shear stress at normal, constant levels if the flow change is within a certain range (for example see Refs. 7 and 20).

Like these, a number of studies have been made on the phenomena of stress-dependent arterial adaptation. It has also been shown that vascular smooth muscle cells (8, 21, 37) and endothelial cells (22, 49) play important roles in the control of wall hoop stress and wall shear stress, respectively. On one hand, there are many studies on the response of in vitro cultured cells to stress, which have indicated that stress affects the proliferation and function of vascular smooth muscle cells (46, 48) and endothelial cells (14, 45, 47). However, because there are discrepancies between in vivo experimental and clinical studies and in vitro cell culture studies, the mechanisms of stress-dependent vascular remodeling have not been elucidated yet.

The present study showed that the vein also exhibits biomechanical response to the elevation of blood pressure in an essentially similar fashion to that observed in arteries. Vascular tone observed under normal conditions, vascular contractility under active condition (Figs. 8 and 9), and wall thickness (Fig. 4) were larger in the hypertensive vein (HP group) than in the normotensive one (Sham group). Because of the wall hypertrophy, there were almost no differences in the circumferential wall stress (Fig. 5) and vascular compliance (Fig. 7) at in vivo pressure between the two groups. Previously, we have reported that the circumferential wall stress and the vascular compliance in nontreated control femoral veins at in vivo pressure were $32.5 \pm 13.4$ kPa and $6.7 \pm 6.0 \times 10^{-5}$ Pa, respectively (34). These stress and compliance values are similar to those shown in Figs. 5 and 7, respectively. We did not obtain the initial stress immediately after the constriction, because no permanent changes occurred in wall dimensions at this stage. However, we can estimate the stress from the in vivo pressure in hypertensive animals (present study) and the wall dimensions in normal animals at this pressure (previous study), and it was $\sim 100$ kPa. These results indicate an optimal operation of the vein against the chronic elevation of blood pressure.

As mentioned in the introduction, to our knowledge, only Monos and his co-workers (29, 30, 32, 33, 43) have been doing detailed studies on the biomechanical response of venous wall to blood pressure elevation. For example, Monos et al. (30) studied the mechanical and morphological properties of the saphenous vein that was exposed to high blood pressure induced by tilting rats at a 45° angle (gravitational method) for 2 wk. The external diameter was significantly larger in the tilted animals than in normal controls at all pressures of up to 20 mmHg; however, no significant difference was observed in the wall thickness. As a result, the circumferential wall stress was significantly larger in the tilted rats than in controls, although there were no differences in the wall distensibility except for 7.5 mmHg. The results on the wall thickness and wall stress are different from the present study.

Later, Monos et al. (32) studied the biomechanical response of the rat saphenous vein to the elevation of blood pressure induced by the above-mentioned method (animal tilting method, for 2 wk) and by surgically resecting 75% of renal mass (volume-expansion method, for 6 wk). In both cases, active, smooth muscle-related responses, which are expressed by the difference in vascular diameter between the total response observed in normal Krebs-Henseleit solution and the passive response obtained in calcium-free, magnesium-rich solution were significantly larger compared with the age-matched controls. The membrane potential of smooth muscle cells measured with intracellular glass microelectrodes was greater in hypertensive veins than in normotensive ones. The hypertensive veins had a larger external diameter than the normal ones in both cases. Furthermore, wall hypertrophy was observed in the veins in which blood pressure was elevated by the volume-expansion method.

By in vitro experiments, significant myogenic tone was observed in the human saphenous vein that was exposed to high in vivo blood pressure, but it was not seen in the human cephalic vein where in vivo blood pressure was low (43). Furthermore, Monos et al. (33) observed in the rat that the above-mentioned animal tilting significantly increased nerve terminal density and synaptic microvesicle density in both saphenous veins and arteries, but this was not the case in brachial veins and arteries. These results imply that long-term gravitational load develops adaptive morphological and functional remodeling of sympathetic innervation in blood vessels of the extremities.

Milesi et al. (28) studied the mechanical properties of human saphenous veins from hypertensive patients.
(blood pressure exceeding 140/90 mmHg) and normotensive subjects by using isolated ring specimens. In tensile stress-extension ratio relations, stress was significantly larger in hypertensive than in normotensive rings in the region of the high extension ratio. We also observed similar results in the rabbit femoral vein at 4 wk after the elevation of blood pressure (data not shown). In addition, they demonstrated that the rings from hypertensive patients developed more contraction than the rings from normotensive subjects in response to KCl and norepinephrine. In hypertensive patients, blood pressure is elevated not only in arteries but also in veins (42), which may stimulate smooth muscle cells and increase vascular tone, leading to larger contraction. These observations are also very similar to ours (Figs. 8 and 9). Previously, we have shown that the diameter response (contraction) of the rat common carotid artery was increased by arterial hypertension (8, 9, 15). Furthermore, human saphenous veins display a much larger degree of myogenic tone compared with canine saphenous veins and femoral veins (2). This might be attributable to the fact that the human saphenous vein is subjected to substantial blood pressure variations due to changing orthostatic loads. This result also suggests that veins exposed to high blood pressure have high myogenic tone.

Pressure-diameter relations of the saphenous vein in spontaneously hypertensive rats (SHR) were studied by Szentivanyi et al. (44). The external radius was larger and the wall thickness was smaller in SHR than in normotensive Wistar-Kyoto (WKY) rats at the same pressures; the differences were statistically significant only in the radius at and above the pressure of 6 mmHg. Therefore, the circumferential wall stress was larger in SHR than in WKY if compared at the same pressures; significant differences were observed at 18 mmHg and above. Because the in vivo blood pressure in the vein is probably higher in SHR than in WKY as mentioned in the article by Szentivanyi et al. (44), the stress at the working pressure would be larger in SHR than in WKY. These results are different from the results obtained from the present study on induced venous hypertension.

Almost all the reported results obtained from, for example, the aorta, common carotid artery, and mesenteric artery show that the circumferential wall stress in SHR is similar to that in WKY if compared at each in vivo working pressure (6, 25, 38). This phenomenon is the same as that observed in the case of induced hypertension, as mentioned above. However, Szentivanyi et al. (44) reported that the circumferential wall stress in the saphenous artery was the same in SHR and WKY if compared at the same pressures; therefore, the stress at each in vivo working pressure is higher in SHR than in WKY. This result is different from many other reported ones. Szentivanyi et al. (44) did not directly measure wall thickness but estimated it from vascular weight, which may have yielded the different results from the others. Because the rat saphenous artery is small, the accuracy in the measurement of its weight is considered to be low. This problem would also be applicable to the saphenous vein that has much less weight than the saphenous artery. Thus the results obtained by Monos et al. (30, 32) may be different from the results obtained from the present study.

A slightly smaller internal diameter observed in the HP group at 4 wk (Fig. 4) may be ascribed to a decreased blood flow in the femoral vein caused by the constriction of the proximally located iliac vein. Many previous studies have shown that arteries chronically exposed to reduced blood flow decrease their internal diameter, keeping wall shear stress at normal level (for example, 14, 15, 20, 22).

Clinically, chronic venous diseases like iliofemoral deep vein thrombosis and valvular dysfunction, which are accompanied by varicosities, edemas, pigmentation, and so on, induce venous hypertension (1, 23, 36, 39, 52). These diseases are clinically rather common and, therefore, venous hypertension should occur frequently. Also, its effects on venous mechanics are of great importance, because pressure-induced changes in passive lumen capacity and myogenic response would contribute much to the maintenance of venous return and cardiac output (29, 35, 36). However, there have been almost no studies on this issue except for the above-stated work done by Monos et al. and the present experiments.

Autologous veins have been used to replace segments of diseased arteries, primarily due to atherosclerosis. After transplantation, vein grafts undergo great changes in the mechanical environment: from a steady flow, low-pressure condition to a pulsatile flow, high-pressure condition. Thus transplanted veins may remodel in response to these changes in the mechanical environment (31). Actually, the transplantation develops intimal hyperplasia and medial hypertrophy, possibly in response to increases in wall shear stress and wall hoop stress, respectively. These morphological changes are expected to occur so as to return the stresses to their normal values. However, vein grafts respond as a damaged tissue, with an associated reparative response to the changes in hemodynamic loads (51, 53). Indeed, Liu and Fung (24) reported that medial smooth muscle cells in vein grafts were damaged soon after transplantation by the step-wise increase in blood pressure and flow. Thus the remodeling phenomena observed in vein grafts are much different from those seen in the veins exposed to relatively small change in blood pressure within a physiological range at their in situ locations, like the femoral vein studied in the present experiment.

In conclusion, when the vein is chronically exposed to high blood pressure, circumferential wall stress is rapidly restored to control level due to wall thickening, vascular tone and contractility are increased, and wall elasticity and compliance are kept at normal level. These remodeling phenomena in the vein are very similar to those observed in hypertensive arteries.

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REFERENCES


