Regional arterial stress-strain distributions referenced to the zero-stress state in the rat

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1Institute of Experimental Clinical Research, Aarhus University, DK-8200 Aarhus; 2Institute of Clinical-Medicine Science, China-Japan Friendship Hospital, 100013 Beijing, China; 3Center of Sensory-Motor Interaction, Aalborg University, DK-9220 Aalborg; and Department of Abdominal Surgery, Aalborg Hospital, DK-9000 Aalborg, Denmark

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Zhao, Jingbo, Judd Day, Zhuang Feng Yuan, and Hans Gregersen. Regional arterial stress-strain distributions referenced to the zero-stress state in the rat. Am J Physiol Heart Circ Physiol. 282: H622–H629, 2002; 10.1152/ajpheart.00620.2000.—Morphometric and stress-strain properties were studied in isolated segments of the thoracic aorta, abdominal aorta, left common carotid artery, left femoral artery, and the left pulmonary artery in 16 male Wistar rats. The mechanical test was performed as a distension experiment where the proximal end of the arterial segment was connected via a tube to the container used for applying pressures to the segment and the distal end was left free. Outer wall dimensions were obtained from digitized images of the arterial segments at different pressures as well as at no-load and zero-stress states. The results showed that the morphometric data, such as inner and outer circumference, wall and lumen area, wall thickness, wall thickness-to-inner radius ratio, and normalized outer diameter, as a function of the applied pressures, differed between the five arteries (P < 0.01). The opening angle was largest in the pulmonary artery and smallest in thoracic aorta (P < 0.01). The absolute value of both the inner and outer residual strain and the residual strain gradient were largest in the femoral artery and smallest in the thoracic aorta (P < 0.01). In the circumferential and longitudinal direction, the arterial wall was stiffer in the femoral artery and in the thoracic aorta, respectively, and most compliant in the pulmonary artery. These results show that the morphometric and biomechanical properties referenced to the zero-stress state differed between the five arterial segments.

MATERIALS AND METHODS

Experimental Procedures

Sixteen 4-mo-old male Wistar rats (354 ± 34 g body wt) were used in this study. The rats were anesthetized with an intraperitoneal injection of pentobarbital sodium (50 mg/kg). The right femoral artery was cannulated for systemic blood

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Fig. 1. Diagram of distended, no-load, and zero-stress states of arterial segments. The outer diameter and microbead (+) displacements at different pressures (P), inner (Ln-i) and outer (Ln-o) circumference length, thickness, and area in no-load and zero-stress state, and the opening angle of the arterial rings could be directly measured from the digitized images.

Fig. 2. Means ± SE of the morphometric parameters of different arteries. T, thoracic aorta; A, abdominal aorta; F, femoral artery; C, common carotid artery; P, pulmonary artery. The sample size was 16. The inner (A) and outer (B) circumferential length of no-load state and zero-stress state, wall area and lumen area (C–E) were largest in thoracic aorta and smallest in femoral artery (P < 0.01). The wall was thickest in thoracic aorta and thinnest in pulmonary aorta (P < 0.01). The wall thickness-to-inner radius ratio (F) was largest in the femoral artery and smallest in the abdominal aorta (P < 0.01).
pressure measurement. The blood pressure was 109 ± 4 mmHg. The thoracic aorta, abdominal aorta, left femoral artery, left common carotid artery, and the left pulmonary artery were dissected, excised, and placed immediately into a calcium-free Krebs solution with 6% dextran and EGTA and aerated with 95% O_2-5% CO_2. At the time of testing, the vessels were taken out of the bath and dried gently with a piece of absorbent paper. The surface was sprayed with microbeads (60–125 μm stainless steel spheres, Duke Scientific) that easily adhere to the tissue for determination of changes in length. For the distension experiments, the proximal end of the arterial segment was connected to a water column. The other end was ligated and left free in the axial direction. Branches were ligated. Stepwise pressurization was carried out by increasing the level of the column to induce pressures of 20, 40, 60, 80, 100, 120, and 140 mmHg for systemic arteries and 5, 10, 15, 20, 25, and 30 mmHg for the pulmonary arteries. Each step lasted at least 2 min until steady state was reached (no change in outer diameter over a 20-s period). The outer diameter and position of the microbeads were recorded with the use of a Sony charge-coupled device camera.

To obtain data on the no-load state and zero-stress state, one arterial ring was excised from the middle region of the various arterial segments and placed in Krebs solution. A photograph was taken of the cross-section of the ring in the no-load state. A radial cut was then made in the ring, which opened into a sector (22). Photographs were taken after ~20 min to allow viscoelastic creep to take place (11).

**Mechanical Data Analysis**

The morphometric data were measured from the digitized images of the arterial segments at the preselected pressures and at no-load and zero-stress states (Fig. 1). The no-load state was defined as that with no transmural pressure or axial loads. The zero-stress state was the stress-free configuration obtained by cutting vessel rings into sectors. The following parameters were measured with the use of Optimas image analysis software: 1) outer diameter and microbead displacement at different pressures, 2) inner and outer circumferential length of arterial rings in no-load and zero-stress state, and 3) opening angle of the cut-open sectors. These measures were used for computation of the biomechanical parameters.

**Residual strain of inner and outer wall.** Residual strain (E) was computed according to Green's formula using the inner (i) and outer (o) wall circumferences, respectively, measured at zero-stress state (L_{z-i} and L_{z-o}) and no-load state (L_{n-i} and L_{n-o}). This was computed at the inner wall

\[
E_{n-i} = \frac{1}{2} \left( L_{n-i}^2 - L_{z-i}^2 \right) / L_{z-i}^2
\]

and at the outer wall

\[
E_{n-o} = \frac{1}{2} \left( L_{n-o}^2 - L_{z-o}^2 \right) / L_{z-o}^2
\]

where L is the midwall length.

**Circumferential and longitudinal wall stress.** Average wall stresses were computed as Kirchhoff stresses assuming a

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**Fig. 3.** Means ± SE of the outer diameter (A) and the normalized outer diameter (B; the diameter at 0 pressure subtracted from the diameter at pressurized conditions) of the thoracic aorta (T), abdominal aorta (A), femoral artery (F), common carotid artery (C) and pulmonary artery (P) at different pressures. The sample size was 16. Outer diameter of arteries before applying pressure was largest in thoracic aorta and smallest in the femoral artery (P < 0.01). The normalized outer diameter was biggest in the pulmonary artery and smallest in femoral artery (P < 0.01).
circular cylindrical vessel geometry and a homogenous wall. The circumferential Kirchhoff stress was computed as

\[ S_c = \frac{\Delta P r_i}{h^2} \]  

and the longitudinal stress was computed as

\[ S_l = \frac{\Delta P r_i^2}{h^2 (r_o + r_i)} \]  

where \( \Delta P \) was the transmural pressure, i.e., the difference between the height of the pressure column and the fluid level in the organ bath in our experiments. The luminal radius \( r_i \) at various distension pressures was computed assuming circular geometry. The wall volume was computed from the wall area and length of arterial segment in no-load state. By assuming incompressibility, \( h \) as the arterial wall thickness could be calculated at various pressures.

The stretch ratio referenced to the zero-stress state. Radial stress components were ignored in this study.

Circumferential and longitudinal strains. Circumferential strain was computed from the midwall circumference referenced to the zero-stress state. The midwall circumferential Green strain was computed as

\[ E_c = \frac{1}{2} \left( \frac{C_p^2}{C_z^2} - \frac{C_z^2}{C_p^2} \right) \]  

where \( C_p \) was the midwall circumference at any of the pressure loads and \( C_z \) was the midwall circumference at the zero-stress state.

The longitudinal Green strain was computed from the midwall lengths at each pressure \( (L) \) and at the no-load state \( (L_o) \) as

\[ E_{\phi} = \frac{1}{2} \left( \frac{L^2 - L_o^2}{L_o^2} \right) \]  

The no-load state was used as reference in the longitudinal direction because midwall residual strains in longitudinal direction were negligible (H. Gregersen, unpublished data). \( L \) and \( L_o \) were measured directly from the displacement of two microbeads on the surface of the arterial segments.

We adopted the exponential strain energy function by Fung (11) for analyzing the data further. This function has the form

\[ W = \frac{1}{2} \left( a_1 E_c^2 + a_2 E_{\phi}^2 + a_4 E_{c_{\phi}}^2 \right) + a_4 E_{c_{\phi}}^2 \]  

where \( a_1, a_2, \) and \( a_4 \) are nondimensional material constants and \( C \) is a material constant with the unit of stress. \( \phi \) and \( \theta \) refer to the longitudinal and circumferential direction. \( E_{c_{\phi}} \) and \( E_{\phi_{c}} \) are strains corresponding to an arbitrarily selected pair of stresses. The meaning of the constants was discussed previously (11).

**Fig. 4.** Means ± SE of the opening angle (A), residual strain (B), residual strain gradient (C), and residual strain gradient-to-wall thickness ratio (D). The opening angle was largest in the pulmonary artery and smallest in the thoracic aorta \( (P < 0.01) \). The absolute values of both the inner and outer residual strain were largest in the femoral artery and smallest in the thoracic aorta \( (P < 0.01) \). The residual strain gradient and gradient-to-wall thickness were largest in the femoral artery and smallest in the thoracic aorta \( (P < 0.01) \). The sample size was 16.
Statistics

The data were representative of a normal distribution and accordingly the results are expressed as means ± SE. Analysis of variance was used for statistical analysis. The results were considered significant if $P < 0.05$.

RESULTS

Morphometric Data

Figure 2 shows the basic morphometric parameters of the arteries. The inner and outer circumferential length, wall and lumen area, wall thickness, and wall thickness-to-inner radius ratio differed between the arteries ($P < 0.01$). The inner and outer circumference at no-load state and zero-stress state, wall area, and lumen area in no-load state were largest in the thoracic aorta and smallest in the femoral artery. The wall thickness at no-load state was greatest in the thoracic aorta and thinnest in the pulmonary aorta. The wall thickness-to-inner radius ratio was largest in the femoral artery and smallest in the abdominal aorta.

The outer diameter and normalized diameter (diameter at a given pressure minus the diameter at zero pressure) as a function of pressure are shown in Fig. 3. The no-load diameter was largest in the thoracic aorta and smallest in the femoral artery. The normalized diameter was largest in the pulmonary artery and smallest in the femoral artery. Significant differences were found between all arteries ($P < 0.01$).

Biomechanical Data

The opening angle and residual strain of the arteries are shown in Fig. 4. A significant difference was found in both opening angle and residual strain between the vessels ($P < 0.01$). The opening angle was largest in the pulmonary artery and smallest in the thoracic aorta. The absolute value of both the inner and outer residual strain was largest in the femoral artery and smallest in the thoracic aorta. The residual strain gradient and residual strain gradient normalized with the wall thickness were also shown in Fig. 4. Both measures were biggest in the femoral artery and smallest in the thoracic aorta ($P < 0.01$).

The relations between stress and strain are shown in Fig. 5 for the circumferential (A) and longitudinal (B) direction. Stress in the femoral artery was higher at a given strain than in the others and that in the pulmonary artery was lower whereas in the longitudinal direction, the thoracic aorta had the higher stress at a

![Fig. 5](image-url)

Fig. 5. Means ± SE of the relation between circumferential (A) and longitudinal (B) stress and strain in different arteries. The sample size was 16.
The mechanical properties of arteries are important determinants of hemodynamics. The aorta, pulmonary artery, and large distributing arteries are distended rapidly during ventricular ejection, transiently accommodating 50% or more of the stroke volume (8). These vessels then retract during diastole. Because of these dimensional changes, the viscoelastic properties of the walls of these large vessels are factors determining instantaneous arterial pressure. The large arteries also transmit the pressure pulse and contribute the dynamic resistance to the oscillatory components of blood flow (8). Finally, certain baroreceptor areas of the arterial tree monitor blood pressure by distension, relaying this information to the central nervous system. All of these functions are modulated by the mechanical properties of the arterial wall.

Many studies have been conducted on stress and strain in the aorta (1, 6, 16–21, 24–25), femoral artery (2, 11), carotid artery (4, 8, 9), and pulmonary artery (3, 21, 22). Most studies before 1983 used the no-load state as reference for the strain analysis. However, it is now well documented that the vascular system expresses residual strain in the no-load state. This effect can be demonstrated by making a radial cut in a ring of tissue. It was demonstrated that this behavior is a mechanism that prestresses the vessel, thus reducing the concentration of circumferential stress at the inner wall. The zero-stress state provides the reference state for calculation of strain and the standard morphological state to describe vascular tissue. The zero-stress state is sensitive to tissue remodeling because the tissue is not deformed by stress. Fung and colleagues (11, 19, 20)

![Fig. 6. Means ± SE of the relation between strain and pressure in the thoracic aorta referenced to the no-load state (n) and the zero-stress state (z). Strains were computed at the inner surface (inner), midwall (mid), and outer surface (outer). The midwall strain referenced to the zero-stress state did not differ from that referenced to the no-load state, whereas the inner and outer surface strains were lower and higher when referenced to the zero-stress state compared with the no-load state. A similar effect was found in all five segments. The sample size was 16.](http://ajpheart.physiology.org/)

### Table 1. Constants from analysis of stress-strain curves

<table>
<thead>
<tr>
<th></th>
<th>Thoracic Aorta</th>
<th>Abdominal Aorta</th>
<th>Femoral Artery</th>
<th>Carotid Artery</th>
<th>Pulmonary Artery</th>
</tr>
</thead>
<tbody>
<tr>
<td>(a_1)</td>
<td>0.323 ± 0.074</td>
<td>0.521 ± 0.047</td>
<td>0.989 ± 0.266</td>
<td>0.178 ± 0.076</td>
<td>0.631 ± 0.083</td>
</tr>
<tr>
<td>(a_2)</td>
<td>0.235 ± 0.058</td>
<td>0.251 ± 0.072</td>
<td>0.076 ± 0.036</td>
<td>0.031 ± 0.013</td>
<td>0.202 ± 0.063</td>
</tr>
<tr>
<td>(a_3)</td>
<td>0.197 ± 0.0512</td>
<td>0.391 ± 0.029</td>
<td>0.782 ± 0.206</td>
<td>0.140 ± 0.059</td>
<td>0.476 ± 0.060</td>
</tr>
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Values are means ± SE for 16 rats. The nondimensional material constants \(a_1\), \(a_2\), and the material constant with a unit of stress, \(C\), were obtained from the strain energy function. \(P < 0.05\), significant difference between the arterial segments was found for each of the constants.
studied the zero-stress state of the aorta and its main branches, small blood vessels and the pulmonary artery of rats. They found that the opening angle varied along the rat aorta and its branches. The opening angle of the pulmonary artery showed axial variation (21, 22). We measured the opening angle of the arterial segments of thoracic aorta, abdominal aorta, common carotid artery, femoral artery, and pulmonary artery. The results in our experiment are concordant with those obtained by Fung and colleagues. We also computed the residual strain distribution on the inner and outer wall of the arterial segments. We demonstrate that the absolute value of residual strain both at inner and outer surface was largest in the femoral artery and smallest in the thoracic aorta. For the residual strain distribution in the thoracic aorta, the common carotid artery and the femoral artery, the results of our experiment were similar to the results reported by Li and Hayashi (18) in the same arterial segments in rabbits. The residual strain was largest in the femoral artery and smallest in the thoracic aorta among three arteries (15). Thus the same trend seems to exist between different species. We further computed the residual strain gradient and residual strain gradient-to-wall thickness ratio. The result showed that the residual strain gradient and residual strain gradient-to-wall thickness ratio were largest in the femoral segment and smallest in the thoracic aortic segment. These results suggest structural differences between the various arteries. However, these were not studied in greater detail in this study, and, in general, correlations between structure and residual strains have been inconclusive. This is likely due to the structural complexity of the arterial wall.

A volume-pressure method based on a step test was used to generate the data for computation of the stress and strain distribution of the five arteries. In the analysis, it was assumed that the arterial wall was homogeneous, incompressible, and elastic. Furthermore, stress and strain gradients throughout the arterial wall thickness were ignored. The analysis resulted in determination of material constants for the arterial segments. The exact meaning of these constants is discussed (11). The results (see Fig. 5 and Table 1) showed that in the circumferential direction, the arterial wall was stiffest in the femoral artery and softest in the pulmonary artery. In the longitudinal direction, the arterial wall was stiffest in the thoracic aorta and most compliant in the pulmonary artery. Biaxial testing of arteries has been done before (8, 10, 17), though the former studies did not consider the zero-stress state. Qualitatively, however, the same differences in elasticity as shown in this study were found previously.

The main structural components of blood vessels are elastin fibers, collagen fibers, and smooth muscle cells. The proportion of those components varies throughout the circulation system. The passive elements are prevalent in the large vessels whereas in small vessels at least 50% of the vessel wall may consist of smooth muscle cells. The mechanical characteristics of blood vessels are determined by both passive and active tissue components. When the vascular smooth muscle cells are inactivated, the elastic modulus is determined by connective tissue, primarily elastin and collagen fibers. Despite the complexity of the connections among elastin fibers, collagen fibers, and smooth muscle cells, the properties of the vessel are represented by an element (the parallel elastic element) in parallel with the series combination of elements representing the force generators in the smooth muscle cells (the contractile element) and the structures connecting them (the series elastic element). Because the structure and the proportion of the elements in the thoracic aorta, abdominal aorta, common carotid artery, femoral artery, and pulmonary artery differ, the strain distribution in these arterial segments will also differ. Despite the fact that the thoracic aorta is an elastic artery, it was stiffest in the longitudinal direction when pressurized. Furthermore, the femoral artery is a muscular artery, but it was stiffest in the circumferential direction when pressurized. The components determining the circumferential or longitudinal vascular stress-strain distributions and the physiological significance of these findings still need further study.

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REFERENCES


