Distensibility of Arteries in Human Hypertension. (30836)

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This study concerns the distensibility of an arterial segment in human "essential" hypertension. This property was measured by a direct technique in living, intact, and unanesthetized hypertensive and normotensive humans. The results demonstrate that distensibility of a segment of brachial artery is significantly diminished in the hypertensive human.

Methods. Subject material. The subjects include 11 humans, 6 females and 5 males, with normal systemic arterial pressure (Cases 1 to 11) and 12 humans, 10 females and 2 males with persistent systemic arterial "essential" hypertension (Cases 12 to 23) (pressures greater than 140/90 mm Hg). Ages ranged from 31 to 52 years in each group. They were in the hospital for at least 2 weeks prior to the investigative studies, during which time clinical data were obtained and antihypertensive therapy was avoided. All were ambulatory and received a regular hospital diet, except those patients with impaired myocardial function who were given a low salt (2 to 4 g daily) diet. Resting supine blood pressure was measured twice daily by a single observer. All indirect blood pressure readings were confirmed by direct needle measurements. All subjects were volunteers who were informed about the procedure and the nature of the study.

Experimental technique. The present studies were performed after an overnight fast. The patients were in the resting, recumbent position and received 120 mg phenobarbital and 25 mg promethazine* intramuscularly 30 to 45 minutes before the procedure. Room temperature was maintained at 75° to 80°F. The left arm was placed comfortably on a padded extension of the table approximately at a 45° angle from the body and at heart level. A 4.0 cm segment of brachial artery along the medial aspect of the arm midway between the axilla and medial epicondyle was then exposed. The selection of this artery segment was based upon the following considerations: 1. It is relatively superficially located, lying deep to the subcutaneous tissue and in the plane between the biceps and brachialis muscles. It is readily accessible and simply exposed. 2. This artery is musculo-elastic and contains significant amounts of smooth muscle, elastic tissue, and collagen tissue, each of which contributes importantly and differently to the physical properties of an artery. 3. A segment length of at least 3.0 cm is available in this area. Initially, the feasibility of using a shorter segment was studied in the dog. It became apparent that a segment shorter than 3.0 cm was difficult to handle, was readily traumatized by placement of a needle, and did not permit satisfactory measurements.

Sterile surgical techniques were used to expose the vessel. A 4.0 to 5.0 cm skin incision was made along the course of the brachial artery after *intradermal* infiltration with 2% procaine. Four cm of artery between the profunda brachial and ulnar collateral branches were exposed gently. Tiny arterial branches from this segment were ligated. In 5 subjects (Cases 1, 13, 18, 20, 21) some pain was felt. In these cases, the local area was first bathed with 2% procaine, and exposure of the artery was then completed. The influence of this maneuver upon the experimental procedure is described below.

An indwelling cannula (standard 18 gauge Cournand needle) was then inserted 0.5 cm proximal to the lower margin of the 4.0 cm segment, and its blunt tip was advanced superiorly for a distance of 1.0 cm. Since the superior 3.0 cm of the segment was to be used for distensibility measurements, the cannula lay in the lower 0.5 cm of this 3.0 cm segment. This site of insertion was selected to avoid direct needle-puncture in the

* Phenergan®.

segment to be studied. A vaselined tape ligature was placed around the artery 0.5 cm above the insertion of the cannula (at the lower limit of the 3.0 cm segment). In the interim, a 3-way stopcock arrangement was prepared. A 10 cm length of polyethylene tubing (PE 200, I. D. 0.055″, O. D. 0.075″) was placed into each of the 3 arms of the 3-way stopcock, using special polyethylene adapters. The system was checked to ensure that it was leakproof. One PE tubing (a) was used to connect to the indwelling cannula, another (b) to connect to a strain gauge transducer and an oscillographic photographic recorder, and the third tubing (c) to connect to a 1.0 ml syringe, calibrated into 0.01 ml divisions. The stopcock and the PE tubing were filled with T-1824-tinged normal saline at 37°C.

The segment was then isolated. A nontraumatic Potts vascular clamp was applied to the proximal end of the 3.0 cm segment. Then, the vaselined tape ligature at the lower end of this segment was gently tightened around the cannula by means of a hemostat applied to the tape as it surrounded the vessel. At no time was the hemostat directly in contact with the artery.

Distensibility of the isolated artery segment was measured by determining pressure-volume relationships. First, the PE tubings were connected as the $(a)\overset{\text{S}}{\underset{\substack{| \\ (c)}}{\text{———}}}(b)$ arrangement described above. S denotes 3-way stopcock. The point of zero reference for measurement of pressure was taken at the level of the vessel segment. The arms (a) and (c) were then connected and the occluded segment was carefully emptied of its blood by gently applying suction to arm (c). This syringe was discarded and the 1.0 ml syringe, mentioned above, containing 1.0 ml sterile T-1824-tinged normal saline at 37°C, was attached to arm (c). T-1824-tinged fluid was used to facilitate detection of leakage from the vessel segment. Immediately thereafter, fluid was added to the segment in stepwise increments of 0.05 ml by connecting arms (a) and (c). At zero volume and at each increment of 0.05 ml, pressure in the occluded segment was recorded by turning the stopcock to connect arms (a) and (b). Stepwise increments of fluid were added until the segment pressure reached approximately 150 to 200 mm Hg. One pressure-volume run required about 60 to 120 seconds. Then the Potts clamp and vaselined tape ligature were loosened, and arterial flow restored. During any run, leakage from the segment could be noted by several means: 1. T-1824-tinged fluid outside the vessel; 2. failure of segment pressure to remain constant during the brief period (approximately 10 seconds) required for its measurement at any volume, and 3. failure to return to the 1.0 ml syringe the total volume of fluid previously added to the segment. If leakage occurred, that run was discarded.

In the 5 subjects (Cases 1, 13, 18, 20, 21) who felt pain during the initial exposure of the artery, topical application of procaine was required to complete the procedure. Then, the area was covered with a warm, moist, sterile gauze for 75 to 90 minutes, after which time pressure-volume measurements were obtained. These data are believed to be reliable since local sensory function returned by 75 to 90 minutes in each subject, indicating a disappearance of the procaine effect.

Results. Table I lists pressures in mm Hg at each progressive increase of 0.05 ml fluid volume within the isolated arterial segment in each subject. Fig. 1 demonstrates pressure-volume relationships graphically. There is a distinct separation of the pressure-volume relationships between 10 (Cases 2 to 11) of the 11 normotensive subjects and 11 (Cases 12 to 16, 18 to 23) of the 12 hypertensive subjects. The curves in the hypertensive subjects are distinctly shifted toward the pressure axis (less distensibility) as compared with those of the normotensive subjects, except for 2 patients, Cases 1 and 17. This separation begins with the smallest segment volume, 0.05 ml, at which segment pressures in the former subjects are 0 to 3 mm Hg and 5 to 28 mm Hg in the latter subjects. At segment volume 0.10 ml, the separation is greater; segment pressures in the normotensive subjects are 1 to 8 mm Hg and 12 to 58 mm Hg in the hypertensive subjects. This

TABLE I. Pressure-Volume Relationships in a 3.0 cm Segment of Brachial Artery in Normotensive and Hypertensive Humans.

Segment volume (ml) →	0	.05	.10	.15	.20	.25	.30	.35	.40	.45	.50	.55	.60	.65	.70
Normotensive patients	P	P	P	P	P	P	P	P	P	P	P	P	P	P	P
1. 116/67, .39 cm	0	1	12	41	78	137									
2. 130/90, .50	3	1	6	12	18	25	33	42	53	63	78	81	132		
3. 108/75, .65	0	0	1	2	4	8	14	19	30	48	50	64	94	144	
4. 118/70, .48	–4	1	2	5	12	19	27	42	62	107	210				
5. 118/75, .40	–4	1	5	15	20	37	59	66	85	104	117	140	160	182	207
6. 134/65, .40	1	2	7	15	24	38	62	108	164						
7. 142/84, .51	0	2	5	10	16	25	39	58	100						
8. 120/70, .45	0	2	8	15	25	39	60	80	113						
9. 135/76, .40	1	0	5	13	25	44	80	138							
10. 127/84, .40	1	3	8	14	22	51	65	78	145						
11. 101/53, .48	0	2	5	9	15	25	41	68	150						
Hypertensive patients															
12. 191/112, .47 cm	–2	6	17	32	53	90	168								
13. 142/90, .39	0	15	34	94	192										
14. 173/108, .40	–2	5	12	25	44	96	275								
15. 140/115, .50	0	–	47	–	120	–	240								
16. 144/93, .40	2	6	26	49	83	137	225								
17. 142/100, .48	0	2	4	8	14	25	46	90	164						
18. 163/90, .40	–1	21	58	116	210										
19. 218/108, .54	0	28	54	91	113	166									
20. 185/115, .55	0	15	40	89	182										
21. 203/104, .60	5	27	43	59	79	98	127	195							
22. 150/90, .41	1	5	20	45	90	166									
23. 184/106, .45	0	19	44	56	83	116									

Numbers listed horizontally for each subject are pressures (P) in mm Hg within the isolated segment after each 0.05 ml increment of fluid into that segment. Volumes of fluid within the segment after each 0.05 ml increment are listed horizontally in the line, segment volume (ml). After each patient number are listed the blood pressure (mm Hg) and the external diameter (cm) of the brachial artery segment prior to its isolation and measurement of pressure-volume relationships. Each segment was 3.0 cm in length.

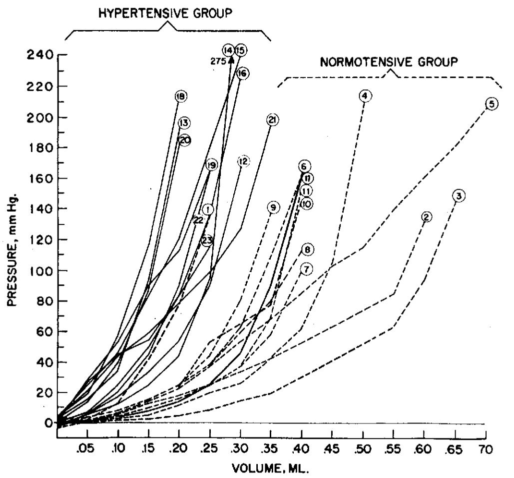

FIG. 1. Pressure-Volume Relationships in a 3.0 cm Segment of Brachial Artery in Normotensive and Hypertensive Humans. Continuous lines represent curves of hypertensive subjects, interrupted lines represent curves of normotensive subjects.

difference in segment pressures between the 2 groups persists, and even becomes greater, at increased volumes.

The distensibility curves relating pressure to volume in the isolated arterial segments are curvilinear with convexities toward the volume axis (Fig. 1). This indicates that the more the arterial walls are stretched, the more they resist further stretch.

Table I lists the average systolic and diastolic pressures (direct needle pressures) for each subject during the experimental period prior to initial isolation of the artery segment. Figure 2 is a scattergram relating these pressures to distensibility of the isolated artery segment in the hypertensive subjects. For convenience in relating distensibility to various physiologic indices, segment pressures at 2 segment volumes, 0.10 and 0.20 ml, are presented as representative of the lower and upper parts of the distensibility curve. At volumes greater than 0.20 ml, measurable portion of the curve ends in some subjects because of excessively high pressures. There is a wide scatter of segment pressures at these volumes when related to blood pressures (140 to 218/90 to 115 mm Hg) in these subjects. This indicates that there is no correlation between the severity of hypertension (as measured by levels of systolic and diastolic pressures) and the extent to which arterial distensibility is altered in hypertension (as measured by segment pressures at the 2 volumes selected).

Discussion. Distensibility of a vessel is defined as the volume-pressure relationships of that vessel. It varies directly with the vessel radius and inversely with the thickness and the mechanical properties of its wall. In the present study, distensibility of an isolated segment of brachial artery was measured directly for the first time in the living, unanesthetized human. It was demonstrated that this vessel was less distensible in hypertensive humans than in humans with normal blood pressure. This investigation did not elucidate the factors responsible for this abnormality. The technique described here lends itself well to further investigations of this problem. Experiments in which distensibility

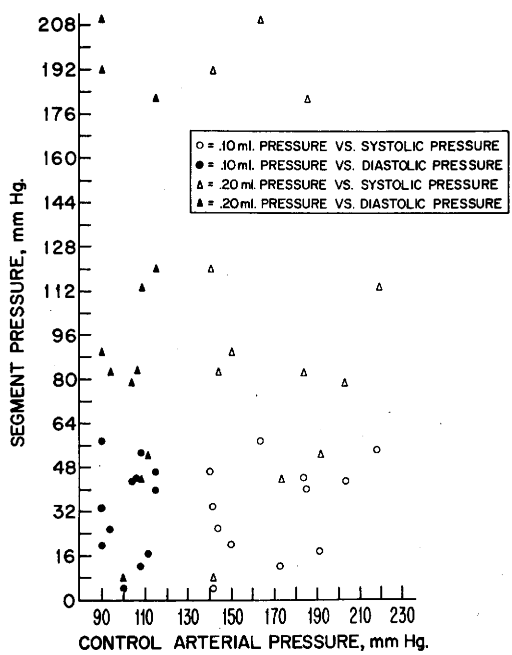

FIG. 2. Scattergram Relating Control Arterial Pressures to Distensibility of the Isolated Artery Segment in Hypertensive Subjects. Segment pressures at 2 segment volumes, 0.10 and 0.20 ml, are presented as representative indices of the total distensibility curve as a convenient means of relating distensibility to another physiologic parameter. ○, segment pressure at 0.10 ml segment volume correlated with systolic pressure. ●, segment pressure at 0.10 ml volume correlated with diastolic pressure. △, segment pressure at 0.20 ml volume correlated with systolic pressure. ▲, segment pressure at 0.20 ml volume correlated with diastolic pressure.

of the arterial segment is measured during a variety of experimental conditions, *viz.*, the application of vasoactive stimuli, administration of agents which influence salt and water metabolism, etc., are in progress in the attempt to elucidate the mechanisms responsible for the decreased arterial distensibility in hypertension.

These experiments confirm the work of other investigators who used indirect techniques *in vivo* or who measured elasticity in excised segments of human arteries postmortem(1,2). Conway and Smith(3) found that humans with hypertension could be divided into two groups, according to their response to amyl nitrite inhalation, *i.e.*, those with "elastic" and those with "inelastic" arteries. Conway(4) also observed that forearm vascular resistance was increased during reactive hyperemia in some hypertensive subjects, suggesting that blood vessels may be abnormal in hypertension.

Animal experiments suggested that changes exist in the arterial wall in hypertension. Wiggers(5) applied formalin to central arteries of hypertensive dogs. Pulse pressure widened as systolic pressure rose and diastolic pressure fell. He suggested that human hypertension was associated with decreased distensibility of large arteries. Feigl, Peterson, and Jones(6) found that distensibility of an intact segment of femoral artery was decreased in the dog with renal hypertension.

The pressure-volume relationships of the brachial artery segment in the present study formed a curvilinear slope with the upward convexity toward the volume axis. This confirms the work of other investigators in animal experiments using excised arterial segments(7-10). It is characteristic of arteries that they do not obey Hooke's Law, but that they resist further stretch more strongly the more they are stretched. Roach and Burton (7) indicated that this property was due to the heterogenous nature of the artery wall. In their experiments with postmortem human iliac artery segments, they demonstrated that resistance to stretch at low pressures was almost all due to elastin fibers, but as pressure increased, more and more collagen fibers reached their unstretched length. At high pressures, collagen fibers, which have elastic moduli greater than those of elastin fibers, were responsible for resistance to further stretch. Krafka(8) compared elastic moduli of the aortas of various animals with tendons and ligaments. The data suggested that the straight line portion of the elastic diagram of the aorta (at high pressure) was dependent upon the high moduli of the collagen fibers.

Summary. Distensibility of an isolated segment of brachial artery was measured directly in 11 normotensive and 12 hypertensive humans *in vivo*. The brachial artery segment between the profunda brachial and ulnar collateral branches was exposed by sterile surgical techniques. An indwelling cannula was inserted into the segment from below. The segment was then isolated and gently emptied. A distensibility curve was constructed by adding fluid (T-1824-tinged normal saline at 37°C) to the segment in stepwise increments of 0.05 ml and recording segment pressure at each fluid increment by the use of a 3-way stopcock arrangement connected to the cannula.

There was a distinct separation of the pressure-volume relationships of the brachial artery between the two groups of subjects. Curves from the hypertensive subjects were shifted toward the pressure axis, indicating that this vessel is less distensible in hypertensive humans than in humans with normal blood pressure. The mechanisms responsible for this phenomenon were not elucidated.

The pressure-volume relationships of the brachial artery segment formed a curvilinear slope with the upward convexity toward the volume axis. Artery segments do not obey Hooke's Law, *i.e.*, they resist further stretch more strongly the more they are stretched.

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Leukemia in Germfree Rats.* (30837)

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Leukemia in mice is a virus disease which is manifested either spontaneously or subsequent to whole-body exposure of the host to X-rays(1). The agent(s) of mouse leukemia induces leukemic disease in rats provided that they are inoculated shortly after birth(2,3, 4); however, the viruses are not considered indigenous to rat populations. Spontaneous leukemia has occurred infrequently in rats (5,6); and induction of leukemia in rats by a variety of chemical and physical agents has been of relatively insignificant incidence(7, 8). When spontaneous leukemia did appear in rats, the etiological agent(s) was not clearly identifiable(9).

Rats and mice have been maintained and propagated under defined germfree conditions for several years(10,11); and they have become at least comparable in quality to the conventional stock from which they were derived. One anatomical anomaly observed in germfree animals has been the development of very large fluid-filled, thin-walled ceca, which, in some instances, become so large as to occupy half of the abdominal cavity and thereby interfere with normal function. Also, the reticuloendothelial system is underdeveloped, but functionally intact. Virus-like particles have been observed in individual thymic tissues derived from the 6 strains of germfree mice; and leukemia has been induced in large numbers of mice among all of the strains by whole-body X-irradiation(12, 13). It is assumed that at least some of the observed particles are leukemia virus. In addition, virus-like particles have been observed in thymic tissues of preleukemic and leukemic germfree AKR mice(14). Virus associated with leukemia is the only agent thus far detected in germfree mice.

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Spontaneous leukemia has not been observed during the past 4 years among the 4000 germfree rats which have been propagated at Lobund Laboratory. While a microbial agent has not yet been detected in germfree rats, the search continues. Germfree rats develop neoplastic diseases(15). Spontaneous breast tumors have been observed in aged germfree Wistar strain rats. Breast tumors have been induced in germfree Sprague-Dawley rats which had been fed 7, 12 dimethylbenzanthracene (DMBA) when 50 days of age. Fibrosarcomas were induced in 3 strains of germfree rats by a single subcutaneous injection of sterilized 3-methylcholanthrene in olive oil. In all instances, the lesions which developed were indistinguishable from those which were induced by the same procedures in conventional counterpart rats.

It was of significant interest to determine (a) if the tissues of germfree rats contain virus-like particles, (b) if leukemia could be induced in germfree rats by X-irradiation, and (c) if leukemia could be induced in them by a strain of mouse leukemia virus.

Methods. Animals. The germfree rats used in this study were of the Wistar, Fischer, and Sprague-Dawley strains. The Wistar strain was in the 20th germfree generation, the Fischer in 6th, and Sprague-Dawley in 3rd generation by random brother-sister mating. They were propagated in steel isolator units,