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## MEASUREMENTS OF YOUNG'S MODULUS OF ELASTICITY OF THE CANINE AORTA WITH ULTRASOUND

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### **Abstract**

We have developed an ultrasonic technique for determining the dynamic Young's modulus of elasticity ( $E$ ) of the canine aorta *in vivo*. Young's modulus was measured in the descending thoracic aorta (DTA) and the abdominal aorta (AA) of 12 dogs over a range of mean blood pressures from 40-200 mmHg. The vessels were excised and dynamic moduli were determined *in vitro* postmortem from pressure-volume curves. The data so obtained were compared to the *in vivo* values. *In vivo* and *in vitro* moduli increased exponentially with mean distending pressure ( $P$ ). The equation of best fit for these data was of the form  $E = E_0 \exp(aP)$ . Constants  $E_0$  and  $a$  depended on the site of measurement (AA or DTA) and upon the particular animal. *In vivo* and *in vitro* moduli were not significantly different in the AA (AA: *in vivo*  $E_0 = 667 \pm 382$  mmHg,  $a = 0.017 \pm 0.004$  mmHg<sup>-1</sup>; *in vitro*  $E_0 = 888 \pm 367$ ,  $a = 0.016 \pm 0.002$ ). However, *in vivo* moduli exceeded *in vitro* moduli in the DTA. (DTA: *in vivo*  $E_0 = 687 \pm 241$ ,  $a = 0.016 \pm 0.004$ ; *in vitro*  $E_0 = 349 \pm 64$ ,  $a = 0.018 \pm 0.003$ ). The increased stiffness of the DTA compared to the AA *in vivo* may be due to the *in situ* tethering of the aorta to the spine by the parietal pleura.

**Key words:** Arterial compliance; arterial elasticity; pulse wave velocity

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## Introduction

The normal mechanical function of the aorta is storage of part of the systolic outflow of the heart as elastic energy in the aortic wall, which is released to provide flow during diastole. However, stiffening of arteries occurs with age and may be associated with symptomatic and life threatening cardiovascular disease. An increase in aortic stiffness increases the work load of the heart.

The fundamental elastic property of the aortic wall is Young's modulus of elasticity. Young's modulus is an inherent property of any solid or elastomer, and is defined as the ratio of tensile stress to tensile strain. Studies of the mechanical/elastic properties of the aorta have been limited because there have been few methods of measuring the modulus of elasticity of the aorta in vivo. Studies of excised vessels are of limited validity. This paper describes a new ultrasonic method for the in vivo measurement of Young's modulus in the intact, living dog and compares in vivo measurements with in vitro values determined immediately postmortem in these same aortic segments. By comparing the in vivo and in vitro values, differences between aortic elasticity in the living animal and in excised vessels can be discerned.

## Methods and materials

In the present study, the in vivo Young's modulus of elasticity (E) was calculated from the aortic pulse wave velocity ( $V_p$ ), the diameter (d), thickness (t) of the aorta, and the density of blood ( $\rho$ ). The mathematical relationship describing these quantities is the Moens-Korteweg equation [1],

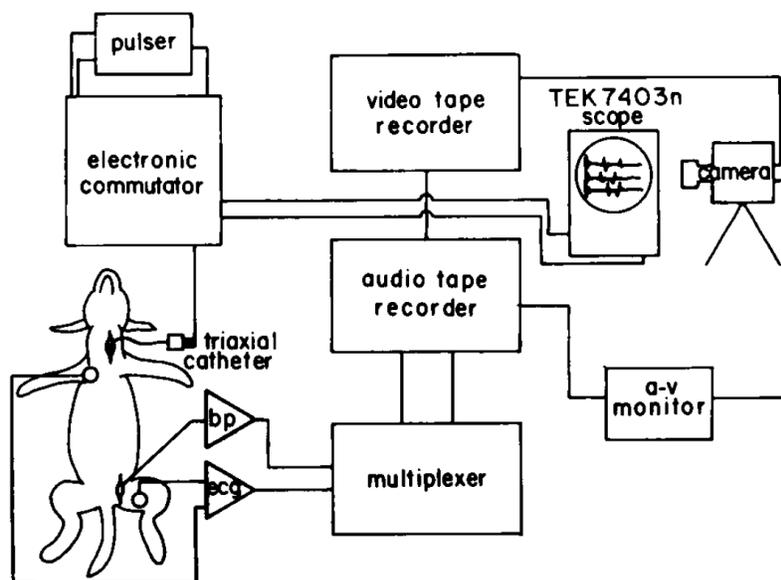
$$V_p = \frac{Et}{\rho d}. \quad (1)$$

Aortic diameter and thickness were measured with a triaxial ultrasound catheter. The density of blood was determined from the hematocrit [3]. The pulse wave velocity was measured in the aorta by determining the difference in pressure pulse arrival time with a catheter-tip pressure transducer at two sites a known distance apart. Due to the small relative diameters of the catheters with respect to the diameter of the aorta, the presence of the catheters in the vessel was not expected to modify blood flow. Using this technique, we determined the modulus of elasticity in the thoracic and abdominal aortae of 12 mongrel dogs. In 8 of the 12 animals, Young's modulus was measured in segments of both the thoracic and abdominal aortae. In each of the 12 dogs, after the in vivo measurements were completed, the aortae were excised and moduli were determined in vitro by a pressure-volume technique.

### *a. In vivo measurements*

Twelve dogs anesthetized with sodium pentobarbital (30 mg/kg i.v.) served as subjects. In each animal, measurements of aortic pulse wave velocity and aortic diameter and thickness were made over a wide range of mean aortic blood pressure (40-200 mmHg). Blood pressure was elevated by intravenous injections of phenylephrine hydrochloride (0.1 mg/kg), a short-acting peripheral vasoconstrictor. Blood pressure was lowered by intravenous doses of sodium nitroprusside (0.1 mg/kg), a short acting peripheral vasodilator. Atropine sulfate (1 mg, i.v.) was given prior to phenylephrine, and propranolol hydrochloride (1 mg, i.v.) was given prior to nitroprusside to blunt the action of autonomic reflexes.

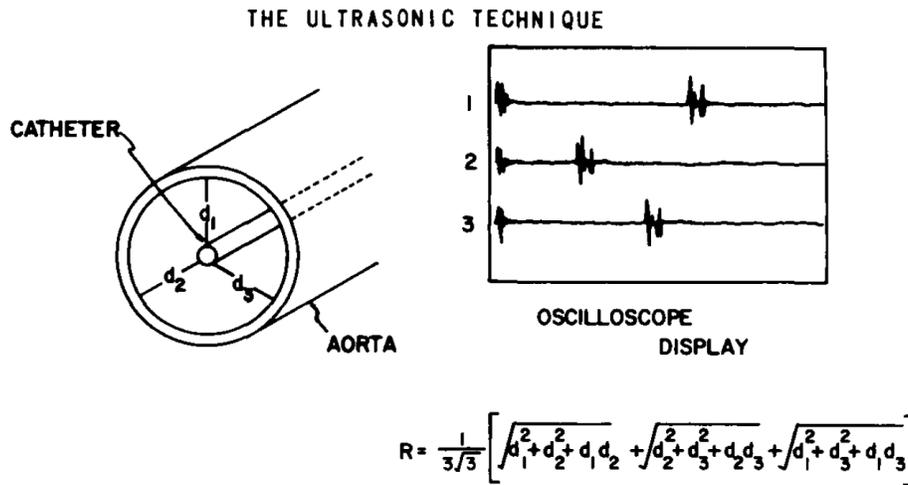
A specially designed ultrasonic catheter (developed with the Indianapolis Center for Advanced Research, Indianapolis, Indiana) was employed to measure aortic diameter and thickness. This catheter contained three piezoelectric elements spaced equiangularly (120 degrees) around the catheter tip in a plane perpendicular to its long axis. The catheter (7 French) was 60 cm long. The piezoelements were excited by a Panametrics (Waltham, Massachusetts) 5050PR pulser, which also provided preamplification for the echo signals received from the aortic walls. A specially designed electronic commutator was used to excite each piezoelectric element sequentially and display the echo patterns on a high-frequency oscilloscope. A pulse repetition frequency of 1 kHz was employed. This ultrasonic catheter system has been described in detail [2]. Figure 1 diagrams the experimental apparatus for the in vivo measurements. Echo patterns on the oscilloscope were recorded on videotape for later analysis. The videotape recordings allowed determination of aortic pressures and radii every 33 ms [2].



*Figure 1. Diagram of the experimental apparatus for the in vivo measurements.*

To determine aortic dimensions in vivo, the triaxial ultrasonic catheter was advanced down the right carotid artery into the desired position in the aorta. In eight of the twelve animals, measurements were made at two sites, one in the descending thoracic aorta (DTA) distal to the aortic arch at the mid-thoracic level, and the other in the infrarenal abdominal aorta (AA).

Figure 2 illustrates the technique of measuring aortic diameter from ultrasonic times-of-flight (tof). Three aortic inner- and outer-wall echoes were displayed on an oscilloscope screen, the image of which was recorded on video cassette tape for later analysis. By replaying the video recorded echo signals on a stop-frame video cassette player (Sony V0-2850), ultrasonic times-of-flight to and from the inner- and outer- walls of the aorta could be measured. Using previously determined values for the speed of 10 MHz ultrasound in canine blood [3], and in canine aortic wall [4], the distances from each piezoelement to the inner- and outer-wall were then computed, as shown in Figure 2. By applying the polygon law and the law of cosines to an inscribed triangle whose vertices were the points of intersection of the ultrasonic paths with the circular vessel walls, the inner- and outer-radii of the aorta were computed. Since only veins collapse to any significant extent, the assumption of circular aortic section for distending pressures greater than 40 mmHg should not hinder the application of the polygon toward calculation of vessel dimensions.



*Figure 2. Diagram of catheter location in the aortic lumen, typical A-scan display of echoes, and the equation relating vessel radius to intravascular distances.*

To facilitate calculation of mean, systolic and diastolic diameters in the aorta, a blood pressure channel was displayed by the oscilloscope and recorded on video tape, along with ultrasonic echoes. Mean inner diameter,  $D_m$ , was calculated in a manner analogous to mean blood pressure,  $P_m$  from systolic and diastolic pressures ( $P_s, P_d$ ) and diameters ( $D_s, D_d$ ):

$$P_m = P_s + K(P_s + P_d) \quad (2)$$

$$D_m = D_s + K(D_s + D_d).$$

The parameter  $K$  was evaluated from strip chart records of blood pressure. The difference between mean inner- and outer-wall radii was interpreted as the mean wall thickness,  $t$ .

The pulse wave velocity is the remaining quantity needed to calculate Young's modulus using Eq. (1). Pulse wave velocity was measured using differential pulse arrival times at two sites a known distance apart. The R-wave of the electrocardiogram was used as the reference time. This method, described by Patel et al. [5], measures the velocity at which the upstroke of the blood pressure waveform travels. Pulse arrival was detected by strain gage pressure transducer mounted at the tip of a 120 cm long #5 French catheter placed adjacent to the ultrasound catheter-tip transducer.

To calculate pulse wave velocity at the site of ultrasonic measurement, the Millar blood pressure catheter was translated 5 cm cephalad from the tip of the ultrasonic catheter for a few cardiac cycles and then moved 5 cm caudad. By using the R-wave of the ECG (lead II) as a timing reference, measuring the difference in pulse arrival times,  $T$ , over the 100 mm excursion gave the values necessary to compute the pulse wave velocity,  $V_p$ :

$$V_p = \frac{100 \text{ mm}}{T \text{ ms}} . \quad (3)$$

Initial adjacent location of the two catheter tips was verified by echoes received from the blood pressure catheter with one of the ultrasound transceivers. Postmortem examination of catheter positions also confirmed adjacency of the two catheter tips. In theory, blood flow velocity must be subtracted from the calculated pulse wave velocity to obtain the true pulse wave velocity. However, since the flow velocity is of the order of 0.25 m/s compared to 5 m/s for the pulse wave velocity [6], this correction was not made in this study. For calculation of in vivo moduli, values of the pulse wave velocity at specific mean pressures were taken from parabolic regression functions fitted to the pulse wave velocity data [7].

### ***b. In vitro measurements***

After the in vivo data were obtained, the animal was sacrificed and specimens of the DTA and AA were excised for determination of the in vitro modulus of elasticity. The excised specimens coincided with the points of ultrasonic measurement. Before excision, the in situ length,  $L$ , was determined carefully. The specimens were tethered to this length in a room temperature water bath for determination of Young's modulus by the pressure-volume method shown in Figure 3. An infusion pump was used to pressurize the vessel with a continuous infusion of saline loaded with India ink. The ink particles inhibited leakage from the vasa vasorum. The rate of infusion was constant at  $0.5 \text{ cm}^3/\text{s}$ . Transmural pressure ( $P_t$ ) was recorded with a strain gauge pressure transducer. Movement of the infusion pump shaft engaged a linear potentiometer to record the volume infused. Both transmural pressure and infused volume were recorded on an x-y plotter to produce a pressure-volume curve.

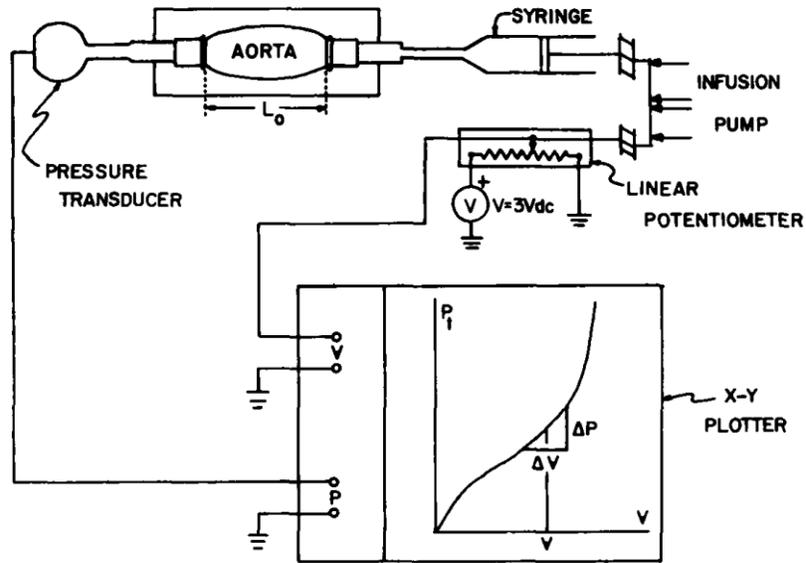
Careful calculation of the internal volume of the vessel ( $V_0$ ) at zero transmural pressure allowed determination of vessel volume,  $V$ , at any pressure from the infused volume. The vessel wall thickness at zero transmural pressure,  $t_0$ , was measured directly by a special tissue micrometer, by applying Archimedes Principle and knowing vessel dimensions and by using a magnifying optical gradicule. In each case, the vessel was tethered to the in situ length. Then wall thickness for any distending volume was calculated, assuming constant cross sectional area of the vessel wall (Poisson's ratio = 0.5) from the expression

$$t(V) = t_0 \frac{V_0}{V}. \quad (4)$$

Young's modulus was calculated from Eq. (5), as described by Posey [6],

$$E = \frac{\Delta P}{\Delta V} \cdot \frac{2}{t} \sqrt{\frac{V^3}{\pi L}}. \quad (5)$$

In this expression,  $\Delta P/\Delta V$  denotes the slope of the P-V curve,  $t$  is wall thickness,  $V$  is the internal volume, and  $L$  is the in vivo length of the vessel.

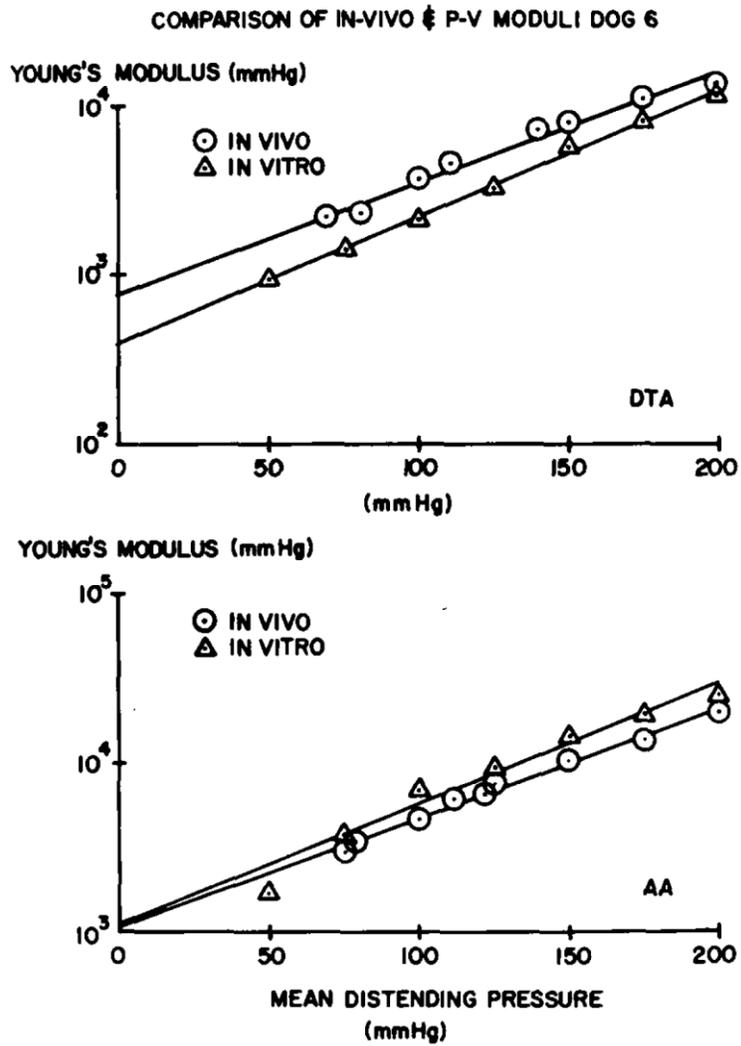


*Figure 3. Diagram of the apparatus used in the in vitro measurements.*

## Results

Typical in vivo and in vitro values of Young's modulus are represented by the semi-log plot in Figure 4. The linear relationship obtained suggested an exponential representation of Young's modulus,  $E$ , in terms of mean distending pressure,  $P$ . This relationship was described by the expression

$$E = E_0 \exp(aP). \quad (6)$$



*Figure 4. Young's modulus in vivo and in vitro for the thoracic (a) and abdominal (b) aortae.*

From semi-log treatment of these data, the constants  $E$  and  $a$  were determined for each of the 12 dogs. The mean values,  $E_0$  and  $a$ , are shown in Table 1, along with the ratio of in vivo to in vitro results. The correlation coefficients obtained for these exponential fits were in excess of 0.95.

Table 1

	$E_0$ (mm Hg)		
	<u>In vivo</u>	<u>In vitro</u>	<u>In-vivo/In-vitro</u>
DTA	687 ± 241	349 ± 64	1.969
AA	667 ± 382	888 ± 367	0.751
	$a$ (mm Hg) <sup>-1</sup>		
	<u>In vivo</u>	<u>In vitro</u>	<u>In-vivo/In-vitro</u>
DTA	.016 ± .0036	.0182 ± .0026	0.885
AA	.017 ± .0043	.0158 ± .0019	1.101

## Discussion

The Moens-Korteweg equation, Eq. (1), was derived in 1878 from the wave equation for propagation of a pressure impulse within a thin walled, perfectly elastic, cylindrical tube [1]. Details of the derivation were reviewed by Hardung [8]. Measurement of Young's modulus of the canine aorta in vivo was made on the basis of this relationship. The moduli so evaluated are necessarily dynamic, that is, they depend on the frequency of application of the distending force. In this investigation, the distending frequency was the heart rate which ranged from 2.0 to 2.5 Hz (120-150/min) due to phenylephrine and nitroprusside. Bergel [9] found that the dynamic modulus was independent of distending frequency for frequencies greater than 2 Hz and less than 10 Hz.

The in vitro measurements of Young's modulus represent values obtained by tethering the aorta to its in vivo length. This tethering prevents longitudinal extension of the vessel, a natural phenomenon in vivo, but the introduction of such end effects is minimized by tethering at the in vivo length [6].

The moduli evaluated in vitro represent dynamic moduli due to the nature of the distension achieved by employing the infusion pump. By setting the rate of infusion at about 0.5 cm<sup>3</sup>/s,

(yielding a  $dP/dT$  of about 200 mmHg/s), the rate of distension in vitro was equivalent to the in vivo rate of distension.

Because Young's modulus of elastomeric materials, such as blood vessels, is known to be strain dependent, the choice of an independent variable for the in vivo measurements was carefully considered. The circumferential elasticity of vessels is primarily related to radius rather than pressure [10]. However, since mean blood pressure and vessel radius are intimately related for the aorta [11], and because mean blood pressure is directly measurable, mean blood pressure was used as the independent variable for determining the pressure-dependent terms in the Moens-Korteweg equation.

### *a. Exponential elasticity*

Others have described the nonlinear elastic behavior of blood vessels [6, 8, 10, 12-14]. However, we found the modulus of elasticity to be exponentially dependent upon mean distending pressure (Figure 4). Our studies indicate that this nonlinear elastic behavior of the aorta, described by Eq. (6), applies to both in vivo and in vitro situations. Constants  $E_0$  and  $a$  were the intercept and slope, respectively, of the line resulting from a semi-log plot of  $E$  vs.  $P$ . Therefore, the aorta can be elastically characterized by  $E_0$  and  $a$  over the range of mean pressure.

A similar dependence of the dynamic modulus upon distension was obtained in vitro by Hardung [8]. Subjecting his data to a semi-logarithmic representation yields an exponential equation of best fit:

$$E = 22 \cdot 10^5 \exp(2.9 \Delta \ell / \ell), \quad (7)$$

where  $\Delta \ell / \ell$  is the initial extension of the of thoracic aorta,  $\ell$  being the length of the aortic strip along the direction of extension. The correlation coefficient obtained was 0.975.

Bergel [15] measured an incremental (or static) Young's modulus in the thoracic aorta of a dog and found a nonlinear dependence with pressure. We have applied an exponential fit to his data with a resulting equation of best fit

$$E = 717 (\text{mmHg}) \exp(0.016 P). \quad (8)$$

The correlation coefficient for this fit was 0.99. Others have compared measurements of the static and dynamic modulus of the canine aorta in vivo and in vitro [6,16,17]. Table II summarizes the results reported here and elsewhere. The range of values reported herein compare well with the results of these studies.

Table II. Comparison of values of Young's modulus reported by several investigators (after [19]).

Authors	Artery	Pressure Range (mm Hg)	Young's modulus (mm Hg)	
			Static	Dynamic
Bergel, [15]	DTA	100-200	3230-13600	
	AA	100-220	6680-13520	
Bergel, [9]	DTA	100		3310-3680
	AA	100		6920-8270
Nickerson & Drozic, [17]	DTA	100	9000	28700
Gow & Taylor, [16]	DTA	87-130		1650-5120
	AA	96-130		3450-11030
Posey, [6]	DTA	80-200	5900-44300	
	AA	80-200	10300-48700	
This study	DTA	40-200	(in vivo)	1020-30540
			(in vitro)	700-23650
	AA	40-200	(in vivo)	1020-24690
			(in vitro)	1250-24520

Our technique of measuring aortic diameter and wall thickness, unlike many previous methods, was conducted without dissection of the connective tissue surrounding the aorta [2]. In studies of pressure-diameter relationships in dogs, other investigators disturbed the intact vascular geometry to enable implantation of transducers for aortic diameter measurement [1, 2, 7].

### *b. In vivo and in vitro elasticity*

From comparison of the ratios given in Table I, it can be seen that the in vivo moduli in the thoracic aortae were greater than their in vitro counterparts over the same pressure range. The in vivo and in vitro moduli of the abdominal aortae were approximately the same. We speculated that this in vivo excess might be attributable to the differences in dynamic moduli reported by Bergel [15], due to the frequency dependence of the dynamic modulus. The ratio of the dynamic-to-static modulus for the thoracic aorta increases rapidly from unity at 0 Hz to about 1.07 at 2 Hz. For higher frequencies, this ratio remains essentially constant. Since the ratio of static-to-dynamic moduli was as high as 3.3 (dog no. 3) and consistently above 1.07 in our experiments, the differences were not expected to be due to this frequency effect alone.

A test of statistical significance of the greater magnitude of the modulus in vivo in the DTA was made by applying a paired Students t-test to the values of the zero-pressure modulus,  $E_0$ , and to values of Young's modulus calculated in vivo and in vitro at pressures of 75, 100, 125 and 150 mmHg [20]. For the zero-pressure modulus, the mean difference between in vivo and in vitro moduli was 338 mmHg and this excess was significant at the  $p < 0.005$  level. At 75 mmHg, the mean excess was 721 mmHg and the significance level was  $p < 0.01$ . Similarly, at 100 mmHg, the mean excess was 1298 mmHg and the level of significance  $p < 0.005$ . At 125 mmHg, the mean excess was 1661 mmHg and the significance level was calculated to be  $p < 0.01$ . Finally, at 150 mmHg, the mean excess was 2131 mmHg and it was significant at the  $p < 0.02$  level.

The difference between in vivo and in vitro moduli in the thorax may be attributable to another factor yet unaccounted for. In our thoracic surgery, we noted that the thoracic aorta was tightly tethered to the spine by the parietal pleura and intercostal arteries plus surrounding connective tissue. The abdominal aorta lacks such tethering. This tethering may account for the increased stiffness in vivo. The indication is for further investigation, repeating the in vivo measurement with the connective tissue bed and intercostal arteries dissected away.

### **Summary and conclusions**

Young's modulus of elasticity was measured in the canine aorta in vivo, in the intact animal, and in vitro, in excised vessels of 12 dogs over a mean pressure range of 40-200 mmHg. Measurements were made by determining the values of the various terms appearing in the Moens-Korteweg equation. Aortic diameter-to-thickness ratio was determined ultrasonically and invasively. The pulse wave velocity was determined from pulse arrival times at a catheter-tip pressure transducer. These same aortae were excised postmortem for evaluation of in vitro moduli by a standard pressure-volume technique.

Both in vivo and in vitro moduli were found to increase exponentially with mean distending pressure. These moduli were found to compare very well with previously reported moduli. The in vivo moduli of the descending thoracic aortae were in excess of the in vitro moduli of the thorax, perhaps due to the tethering of the DTA to the parietal pleura in vivo.

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