JACC Vol. 25, No. 6
May 1995:1428–35

Interaction Between Afterload and Contractility in the Newborn Heart: Evidence of Homeometric Autoregulation in the Intact Circulation

ROBERT JOHANNES MENNO KLAUTZ, MD, DAVID FRANK TEITEL, MD,*
PAUL STEENDIJK, PhD, FRANK VAN BEL, MD, JAN BAAN, PhD

Leiden, The Netherlands and San Francisco, California

Objectives. **We undertook the present study to determine whether afterload and contractility interact in the hearts of newborn lambs. We specifically investigated whether stepwise increases in afterload increase contractility.**

Background. **Several studies in the isolated and intact adult dog heart have shown that afterload and contractility are not independent determinants of cardiac performance; rather, they interact. Afterload and contractility are unlikely to interact in the newborn heart because the factors that may mediate the interaction in the adult are missing in the newborn.**

Methods. **We measured contractility at different steady state levels of afterload in seven newborn lambs under complete anesthesia. Contractility was measured by three different indexes: end-systolic pressure–volume relations (slope and volume position); preload-corrected first derivative of left ventricular pressure (dP/dt_{max}); and preload-corrected stroke work. Left ventricular pressure and volume were measured with a micromanometer and conductance catheter, respectively. Preload and afterload were manipulated by inflating or deflating a balloon catheter in the inferior vena cava and descending thoracic aorta, respectively. Data are expressed as mean value ± 1 SD.**

Results. **Stepwise increases in afterload increased contractility, independent of which of the three indexes was used. The slope of the end-systolic pressure–volume relation increased from a mean baseline value of 4.44 ± 2.43 to 6.69 ± 2.89 kPa/ml at the highest level of afterload. Concomitantly, volume at 14 kPa of the end-systolic pressure–volume relation decreased from 3.34 ± 1.52 ml at baseline to 1.12 ± 0.83 ml at the highest afterload. The other two indexes showed qualitatively similar changes. Beats selected from unloading interventions on the basis of the same end-diastolic volume for each level of afterload showed no difference in stroke volume.**

Conclusions. **This study in newborn lambs demonstrates that stepwise increases in afterload increase contractility considerably and that this enables the heart to maintain stroke volume at different levels of afterload. This forms direct evidence for the existence of homeometric autoregulation in the intact newborn heart.**

(J Am Coll Cardiol 1995;25:1428–35)

The function of the heart is to maintain adequate blood flow to all tissues of the body and, thus, cardiac output. Because cardiac output is determined by the amount of blood ejected by the ventricle per beat and the frequency of contraction, regulation of stroke volume and heart rate are of paramount importance. Whereas contraction frequency is mainly regulated by extracardiac, neurohumoral reflexes, stroke volume can be regulated by the heart itself. Frank and Starling formulated the famous law of the heart, which states that as more blood fills the ventricle in diastole, more blood is ejected during systole. Thus, there is a direct relation between stroke volume and end-diastolic volume. However, stroke volume can also increase by a decrease in end-systolic volume, which is generally assumed to represent an increase in contractility because an end-systolic pressure–volume point to the left of the control end-systolic pressure–volume relation (1) is reached (2). Long before the end-systolic pressure–volume relation was advocated to represent contractility of the ventricle (1), Sarnoff et al. (3) in 1960 described how stroke volume is maintained against higher afterloads in isolated ventricles and called it "homeometric autoregulation." However, it implies that contractility has to increase with increases in afterload, a phenomenon that has been doubted for many years to exist in the intact ventricle. Contractility has been defined as being independent of loading conditions.

Several studies (3–10) of the isolated and intact adult dog heart, using the end-systolic pressure–volume relation, have shown that afterload and contractility are not independent but, rather, interact. This interaction goes against the traditional concept of cardiac performance, in which afterload, preload, heart rate and contractility are four independent determinants. For this reason, the validity and usefulness of the end-systolic

From the Cardiac Physiology Laboratory, Departments of Pediatrics and Cardiology, Leiden University Hospital, Leiden, The Netherlands; and *Cardiovascular Research Institute, University of California, San Francisco, California. This study was presented in part at the 43rd Annual Scientific Session of the American College of Cardiology, March 1994, Atlanta, Georgia. Dr. Teitel was the recipient of a Senior International Fellowship for this project from the National Institutes of Health, Bethesda, Maryland, and was Visiting Professor at the Cardiac Physiology Laboratory in Leiden during this study. This study was supported in part by a grant from the Gisela Thier Foundation, Leiden, The Netherlands.

Manuscript received October 2, 1993; revised manuscript received December 9, 1994, accepted December 15, 1994.

Address for correspondence: Dr. Jan Baan, Cardiac Physiology Laboratory, Department of Cardiology, Leiden University Hospital, P.O. Box 9600, 2300 RC Leiden, The Netherlands.

0735-1097/95/$9.50
0735-1097(94)00562-5

pressure–volume relation to characterize ventricular contractility has been questioned by some investigators (4–9,11).

Whether afterload and contractility also interact in the newborn heart is unknown. There are indications that the interaction between afterload and contractility is of less importance in the newborn because the factors that probably mediate the interaction in the adult are missing in the newborn. These factors are alterations in calcium flux across the sarcoplasmic reticulum, alterations in myofilament sensitivity to available calcium and alterations in beta-adrenoreceptor activity. However, in the newborn the sarcoplasmic reticulum is relatively immature and thus may not be as sensitive to changes in afterload as that in the adult (12,13). Also, the number of myofilaments in the myocytes is low, and their geometry is immature (14), so altered sensitivity may not be translated efficiently into allied force generation. Last, the newborn heart has little beta-adrenergic contractile reserve to be drawn on by alterations in afterload (15,16).

We undertook the present study to determine whether afterload and contractility interact in the hearts of newborn lambs. We specifically investigated whether stepwise increases in afterload increase contractility. To this end, we measured various indexes of contractility and stroke volume in response to stepwise increases in afterload, induced by occluding the descending aorta, in seven newborn lambs. This design of our study, which differed considerably from a previous study in newborn lambs from our laboratory (17), permitted us specifically to evaluate not only the load dependency of the end-systolic pressure–volume relationship, but also the behavior of stroke volume with increased afterload at matched preload conditions, comparable to the study by Sarnoff et al. (3). In this way we were able to contribute to the discussion on the validity of the end-systolic pressure–volume relation to characterize contractility as well as to address the issue of whether shortening deactivation causes the load dependency of this relation.

Methods

The surgical and experimental procedures used were reviewed and approved by the animal research committee of the State University Leiden.

Surgical protocol. Seven newborn lambs, weighing 3.4 to 5.5 kg, were studied between 3 and 14 days of age. Ketamine hydrochloride was used for premedication (3 mg/kg body weight intravenously) and to maintain general anesthesia (8 to 30 mg/kg per h intravenously) throughout the study. The lambs were intubated and ventilated with an oxygen–air mixture using a pressure-regulated ventilator. Ventilation was adjusted to maintain arterial oxygen and carbon dioxide pressures in the normal range throughout the study. Pancuronium (0.2 mg/kg) was administered as needed to maintain adequate muscle relaxation. An intravenous infusion of 10% dextrose in 0.5 N sodium chloride solution was continued throughout the study at ~10 ml/kg per h, occasionally supplemented with sodium bicarbonate to maintain a normal base deficit (≤5 mmol/liter).

To facilitate the insertion of catheters, 6F to 7F sheaths were placed in both right and left femoral arteries and veins using a percutaneous technique. Under fluoroscopic guidance, 5F atrioseptostomy balloon catheters (American Edwards Laboratories) were advanced in a femoral artery to the midthoracic aorta and in a femoral vein to the inferior vena cava–right atrial junction to increase afterload and decrease preload, respectively. A 6F, eight-electrode (5.5-mm interelectrode distance) pigtail conductance catheter (Webster Laboratories) was advanced through the other femoral artery to the apex of the left ventricle to measure instantaneous left ventricular volume. In addition, a 5F Berman angiographic catheter (American Edwards Laboratories) was advanced through the other femoral vein to the main pulmonary artery for determination of the calibration factor (parallel conductance volume) of the volume catheter using the hypertonic salt injection method (18). The conductance catheter was connected to a Sigma-5 signal conditioner processor (CardioDynamics, Rijnsburg, The Netherlands) to convert instantaneous conductance measurements to volume. A 5F micromanometer catheter (Braun Medical, Best, The Netherlands) was advanced through a 6F sheath in a carotid artery (dissected after a small incision in the neck) into the left ventricle just below the aortic valve to measure left ventricular pressure. Aortic pressure was measured from the fluid-filled sideport of the sheath in the neck using a pressure transducer. Blood samples were drawn from a sideport of one of the arterial sheaths for measurement of arterial blood gases and hemoglobin concentration.

After completion of the surgical preparation, a 30-min period was allowed for the lambs to reach hemodynamic stability. During the study the lambs were kept under parasympathetic blockade with atropine (0.1 mg/kg intravenously) to prevent changes in heart rate secondary to changes in aortic pressure.

All analog signals were displayed on a paper recorder and screen for continuous monitoring. The left ventricular pressure and total left ventricular volume signals were also displayed on a X–Y oscilloscope for continuous monitoring of pressure–volume loops. All analog signals were digitized with 12-bit accuracy on an IBM-compatible microcomputer (Epson AT) at a sampling rate of 200 Hz and saved on a hard disk for subsequent analysis.

Study protocol. To determine whether stepwise increases in afterload increase contractility, we measured contractility at four to five steady state levels of afterload. The afterload steps were induced in random order by manual inflation of the balloon catheter in the descending aorta. Partial inflation of the balloon catheter was performed so as to ensure that the afterload steps differed from each other by at least 1 kPa in aortic pressure, registered on a digital instrument. In six of the seven lambs we were able to induce five afterload steps; in the seventh lamb we induced only four steps. Thus, we had a total of 34 afterload steps. The average step size was 1.82 ± 1.13 kPa (mean ± SD). The lowest step (with the balloon completely deflated) was taken as baseline. Hemodynamic conditions were allowed to stabilize for a few minutes after reaching a new step,

Table 1. Values of Parallel Conductance at Five Different Afterload Steps in Newborn Lambs

Afterload Step	V_c (mean ± SD)
Control	7.35 ± 2.18
1	7.58 ± 2.67
2	7.61 ± 2.42
3	8.10 ± 3.11
4	8.18 ± 3.33

V_c = parallel conductance.

and preload was subsequently reduced by rapidly (10 to 15 s) inflating the balloon in the inferior vena cava. Parallel conductance for calibration of left ventricular volume was measured at each level of afterload once. It was repeated if parallel conductance could not be computed in a reliable fashion (19). The results of these parallel conductance measurements are summarized in Table 1. (Note that in the tables the afterload steps are ordered from lowest to highest afterload.)

At each level of afterload, we assessed contractility using three different indexes: end-systolic pressure–volume relation (slope and volume intercept); first derivative of left ventricular pressure (dP/dt_{max}) corrected for preload (end-diastolic volume); and stroke work corrected for preload. In addition, we obtained stroke volume and standard hemodynamic variables. All these measurements were performed at end-expiration with the ventilator turned off.

Calculations. The three indexes of contractility were calculated from left ventricular pressure and volume measurements during a rapid unloading intervention (inferior vena cava occlusion). To calculate the preload-corrected dP/dt_{max} and stroke work, we selected beats from the unloading interventions that had the same end-diastolic volume at each afterload level within each animal. Stroke volume at constant preload was calculated in a similar manner.

The application, validation and calibration of the conductance catheter for measuring left ventricular volume and its use to measure the end-systolic pressure–volume relation as an index of contractility was described earlier by our group (4,5,17–19). Briefly, left ventricular pressure and volume are recorded during gradual inflation of the inferior vena cava balloon to restrict venous return and decrease the preload of the heart. During this unloading intervention, a line (end-systolic pressure–volume relation) is constructed through the end-systolic pressure–volume points (upper left-hand corners of the pressure–volume loops). End-systole is defined as the point in the cardiac cycle of maximal elastance [maximal $P/(V - V_0)$, where P = pressure, and V = volume]; V_0 is calculated by an iterative technique (4). The slope of the line and its position at 14 kPa represent intrinsic measures of contractility; an increase in slope or a leftward shift of the line, or both, has been shown to represent an increased contractile state (1,11). Calibration of the conductance signal to obtain absolute volume was performed by the hypertonic saline method. No attempt was made to correct conductance-derived volume values for the "slope factor" alpha, known to have a value of ~0.9 in small hearts (20). It was assumed to be 1 because we were only interested in intraindividual changes of left-ventricular volume and the quantities derived from it.

Systemic vascular resistance (kPa/ml per min) was approximated as mean aortic pressure (kPa) divided by cardiac output (ml/min); venous pressure was assumed to be negligible. Cardiac output (ml/min) was calculated as stroke volume (ml) multiplied by heart rate (beats/min).

All calculations were performed on a personal computer using a special application software program, and statistical calculations were performed on an Apple Macintosh computer using StatView 4.0 (Abacus Concepts, Inc.) software.

Statistical analysis. We analyzed the effects of afterload on contractility using a multiple linear regression implementation of analysis of variance (21). We designed a model in which we coded afterload with four dummy variables to represent the five afterload steps and incorporated interanimal variability also as dummy variables. The regression equation was

$$Y = a_0 + \sum_{k=1}^{4} a_{d_k} D_k + \sum_{k=1}^{6} a_{l_k} L_k ,$$

where Y represents the dependent variable of interest (e.g., slope of end-systolic pressure–volume relation, volume at 14 kPa of end-systolic pressure–volume relation, stroke volume), D_1 through D_4 code for the afterload steps and L_1 through L_6 code for the seven lambs (17). Because we wanted to compare the different afterload steps with baseline, we used reference coding for the afterload dummy variables and effects coding for the animal dummy variables (21). Briefly, we assigned a set of (0,0,0,0) to the control condition, (1,0,0,0) to the first step, (0,1,0,0) to the second step, (0,0,1,0) to the third step and (0,0,0,1) to the fourth step. This coding technique makes the intercept of the equation, a_0, the mean value of the dependent variable in the control situation and makes each coefficient the incremental value of the dependent variable at that step. To evaluate the overall effect of afterload on the dependent variable, an F test was performed for the set of afterload dummy variables by dividing the mean square of the set by the mean square of the residual error. Interanimal variability was considered only as a set of dummy variables. A p value ≤ 0.05 was considered statistically significant.

Results

The effects of afterload on the steady state hemodynamic variables, before the preload intervention, are listed in Table 2. Note that the values for stroke work and dP/dt_{max} in Table 2 are given only for the steady state situation. As explained earlier (see Methods), these values were computed for matched preload from the preload intervention along with the slope and volume position at 14 kPa of the end-systolic pressure–volume relation. Stepwise increases in afterload increased contractility, independent of which of the three indexes was used. For example, for the end-systolic pressure–

Table 2. Hemodynamic Variables at Five Steady State Afterload Steps in Newborn Lambs

	Afterload Step (mean ± SD)				
	Control	1	2	3	4
End-systolic pressure (kPa)	10.63 ± 2.30	12.52 ± 2.22	14.30 ± 1.87	16.33 ± 2.27	18.73 ± 2.09
End-systolic volume (ml)	2.03 ± 1.05	1.82 ± 0.86	1.92 ± 0.71	1.83 ± 0.63	1.94 ± 0.57
End-diastolic volume (ml)	4.20 ± 1.85	4.01 ± 1.81	4.07 ± 1.63	4.20 ± 1.41	4.39 ± 1.52
Stroke volume (ml)	2.77 ± 1.08	2.86 ± 1.14	2.83 ± 1.08	3.05 ± 1.32	3.00 ± 1.35
Stroke work (kPa·ml)	24.41 ± 11.77	27.80 ± 13.13	30.01 ± 10.41	36.81 ± 14.75	38.80 ± 14.33
Heart rate (beats/min)	186 ± 30	188 ± 28	199 ± 36	197 ± 30	194 ± 25
dP/dt_{max} ($kPa \cdot s^{-1}$)	432.1 ± 131.0	441.3 ± 103.9	513.2 ± 141.0	553.9 ± 131.8	600.4 ± 129.6

dP/dt_{max} = first derivative of left ventricular pressure.

volume relation we found that its slope increased, and its position at 14 kPa decreased, both indicating increased contractility (Fig. 1). The slope of the end-systolic pressure–volume relation increased from a baseline value of 4.44 ± 2.43 kPa/ml (mean ± SD) to 6.69 ± 2.89 kPa/ml at the highest level of afterload. Concomitantly, volume at 14 kPa of the end-systolic pressure–volume relation decreased from 3.34 ± 1.52 ml at baseline to 1.12 ± 0.83 ml at the highest afterload. The other two indexes, dP/dt_{max} and stroke work, both corrected for preload, showed similar changes: dP/dt_{max} increased from a baseline value of 402.0 ± 148.7 to 539 ± 96.3 kPa/s at the highest level of afterload; stroke work increased from 17.7 ± 6.9 to 26.8 ±14.2 kPa·ml.

The results of the multiple linear regression model, which allowed stepwise assessment of the effect of afterload, show that contractility increased at each stepwise increase in afterload (Table 3). Changes were significant for all indexes of contractility, although the step of afterload at which each index reached significance was not always the same.

As shown in Table 3, the beats selected for matched preload indeed fulfilled this criterion (end-diastolic volume was not significantly different for the five afterload steps). It is important to note that these beats also did not differ in heart rate (Table 3). These beats also did not differ in stroke volume, giving direct evidence for the existence of homeometric autoregulation in the newborn heart. A typical example of this phenomenon is shown in Figure 2. In other words, as the heart contracts against a higher afterload, it is able to maintain its stroke volume, even starting from the same end-diastolic volume. This is only possible is if contractility is increased.

Discussion

In the present study we found that stepwise increases in afterload increase contractility in newborn lambs. Each index of contractility showed similar changes: With an increase in systemic resistance of 58% (from 21.57 to 34.01 kPa/ml per min), the slope of the end-systolic pressure–volume relation was predicted to increase 47%, volume at 14 kPa of the end-systolic pressure–volume relation to decrease 63%, dP/

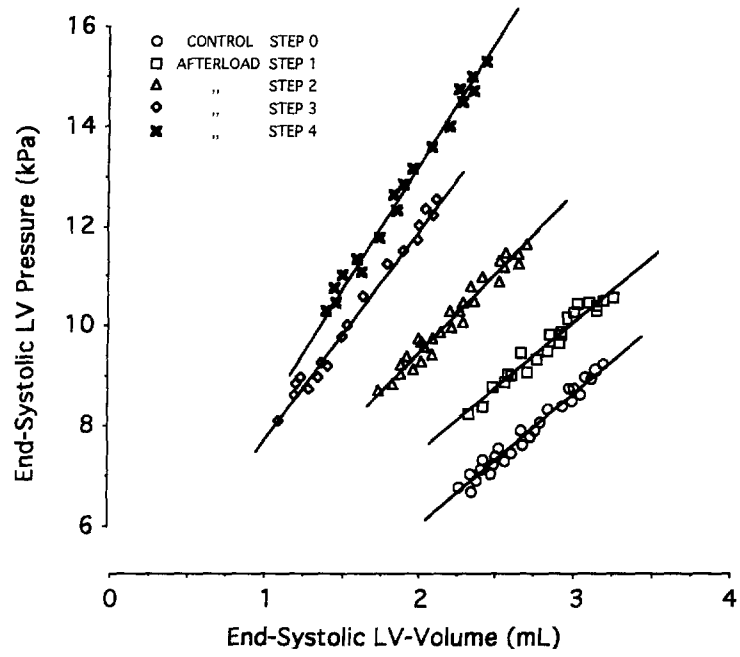


Figure 1. Typical effect of stepwise increases in afterload on contractility, as indicated by the end-systolic pressure–volume relation in one lamb. Afterload was increased from control (step 0) to four levels by inflating a balloon catheter in the descending aorta. The levels of afterload differed by at least 1 kPa of aortic pressure before vena cava occlusion was applied. End-systolic pressure–volume relations were constructed by linear regression of the end-systolic points during each inferior vena cava occlusion. With each step increase in afterload, the end-systolic pressure–volume relation became steeper and shifted to the left, indicating increased contractility. LV = left ventricular.

Table 3. Changes in Several Variables With Stepwise Increases in Afterload: Results of Multiple Linear Regression Implementation of Analysis of Variance in Which Afterload Is Coded by Discrete Dummy Variables

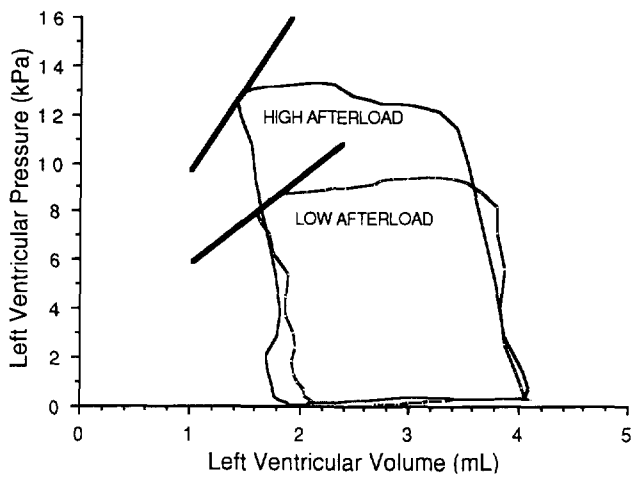
	Regression Coefficient of Equation	Set of Four Afterload Dummy Variables		Predicted Values of Variable at Different Afterload Steps				
		F Value	p Value	Control	Step 1	Step 2	Step 3	Step 4
E_{cs} (kPa/ml)	0.92	6.40	0.005	4.44	4.82	6.06*	6.44*	6.54*
V_{14} (ml)	0.89	18.07	0.005	3.34	2.39*	1.96*	1.49*	1.24*
HR (beats/min)	0.91	2.05	NS	184	187	199	197	196
R_{syst} (kPa/ml per min)	0.97	10.39	0.005	21.57	23.67	27.43*	30.35*	34.01*
Matched preload†								
V_{ed} (ml)	0.99	0.19	NS	3.53	3.54	3.52	3.52	3.56
SW (kPa·ml)	0.89	7.0	0.005	17.74	22.06	23.46	27.06*	28.43*
dP/dt_{max} (kPa/s)	0.91	8.1	0.005	402.0	420.5	499.2*	523.4*	538.0*
P_{es} (kPa)	0.90	21.4	0.005	9.42	11.55*	12.96*	14.1*	14.82*
V_{es} (ml)	0.85	0.9	NS	1.76	1.56	1.66	1.45	1.54
SV (ml)	0.93	1.2	NS	2.32	2.52	2.41	2.65	2.61
HR (beats/min)	0.88	2.7	NS	180	183	200*	196	197

*p = 0.05, significantly different from control. †See Results. dP/dt_{max} = first derivative of left ventricular pressure; E_{cs} = slope of end-systolic pressure (P_{es})–volume relation; HR = heart rate; R_{syst} = systemic vascular resistance; SV = stroke volume; SW = stroke work; V_{14} = volume at 14 kPa of end-systolic pressure–volume relation; V_{ed} (V_{es}) = end-diastolic (end-systolic) volume.

dt_{max} to increase 34% and stroke work 60%, the latter two variables corrected for changes in preload. Also, stroke volume, measured at a constant preload, did not change at higher levels of afterload. This last finding implies that the increase in contractility enables the ventricle to maintain its output despite increases in the load against which it contracts, better known as homeometric autoregulation.

Results from other studies. Our finding that contractility and afterload are interdependent rather than independent determinants of cardiac performance in the newborn heart is in agreement with a recent study from our laboratory (17) in which we showed that in newborn lambs afterload interventions yield higher values of the slope of the end-systolic pressure–volume relation than preload interventions, suggesting a positive inotropic effect of afterload. However, from that study it is difficult to draw conclusions pertinent to the present one because preload and afterload interventions were performed in different ranges of end-diastolic volume, making it impossible to assess the effect of afterload on stroke volume at constant preload. Furthermore, the afterload interventions consisted of continuous changes in afterload rather than steady state changes in which we were interested in this study.

Figure 2. Typical effect of an increase in afterload on the pressure–volume loop at matched preload conditions. The pressure–volume loop is constructed by plotting instantaneous left ventricular pressure against volume during one cardiac cycle. Afterload was increased by inflating a balloon in the descending aorta. Loops were selected from inferior vena cava occlusions on the basis of an end-diastolic volume of 4 ml. Increased afterload caused beats ejecting from the same preload to reach a higher stroke work, end-systolic pressure and peak developed pressure, whereas the end-systolic volume and, thus, stroke volume, remained constant. **Lines** indicate end-systolic pressure–volume relations.



The interaction of afterload and contractility has also been shown in the adult heart (4–9,22). Two of those studies (4,22) found that afterload interventions gave significantly steeper end-systolic pressure–volume relations than preload interventions did. Initially, several investigators argued that the end-systolic pressure–volume relation does not reflect a myocardial property at all (23) or that it is impossible to measure (24). Another explanation given for the behavior of the end-systolic pressure–volume relation was that during ejection, as the ventricular muscle fibers shorten, calcium flux changes, resulting in less developed force (4,5,25). This phenomenon, known as shortening deactivation, was first described in isolated muscle (26–29). However, the evidence for homeometric autoregulation shows that shortening cannot fully explain our findings: Beats with similar end-diastolic volumes, ejecting against different afterloads, maintain stroke volume and therefore undergo the same amount of shortening. Moreover, other indexes of contractility, derived before shortening has taken place (dP/dt_{max}–end-diastolic volume relation) and during shortening (stroke work–end-diastolic volume relation) revealed the same afterload dependence of contractility (4,5,11). It can therefore be concluded that the end-systolic pressure–

volume relation is not inaccurate in reflecting contractility but that afterload and contractility are interdependent.

Homeometric autoregulation. A different proof of this interdependence, not invoking the end-systolic pressure–volume relation or its derivatives, is the presence of homeometric autoregulation, also referred to as the Anrep effect. We found that beats with the same preload (end-diastolic volume) selected from unloading interventions at different afterloads develop the same stroke volume. That is, within the wide range of afterloads that we studied, the newborn heart was capable of maintaining its ventricular output by immediately increasing its contractility (homeometric autoregulation). Of published reports on the adult heart, there are several that confirm this finding, both in whole-heart experiments (3,30–32) and in isolated muscle fiber experiments (33–35). From those last experiments in particular, it became clear that the amount of homeometric autoregulation depends on experimental conditions, such as calcium concentration, temperature and rate of stimulation. This might explain why the effect was not always found under all conditions. Elzinga et al. (36), for instance, found neither a positive nor a negative effect of increased afterload on contractility in elegant studies in cat papillary muscle preparations as well as in denervated chronically instrumented dogs. One explanation for the fact that they did not find homeometric autoregulation in these dogs may be that they did not measure end-diastolic volume directly. Another explanation might be that the awake state is associated with a decreased presence of homeometric autoregulation (32).

The presence of homeometric autoregulation in the newborn heart was not found in another study in newborn lambs (37) in which increases in afterload were accompanied by a decrease in ventricular output. However, in that study, preload (which was expressed indirectly as end-diastolic pressure) was not manipulated as in our study, which made it impossible to assess output at a constant preload. Our findings of homeometric autoregulation in the newborn heart also differ from studies in fetal ventricles (38). The fetal heart decreases its output considerably in the face of higher afterloads. A possible explanation for this finding might be that the fetal ventricle is relatively immature, especially with respect to the regulation of calcium transport in the myocyte. From a teleologic point of view, homeometric autoregulation might be very important for the newborn heart to maintain output because it has much less beta-adrenergic contractile reserve to draw on compared with that in the fetal and adult heart (14,15) and only a limited chronotropic reserve.

We studied only relatively early changes in afterload and not late or chronic changes as exist in arterial hypertension and aortic stenosis. In addition, we only studied changes in afterload by proximal aortic occlusion. However, we believe that there is no difference with respect to the type of afterload intervention that we applied and effect of contractility, as we previously showed (4). In that study, proximal aortic occlusion had the same effect on contractility as increasing peripheral resistance with angiotensin.

Interaction of afterload and contractility. From our study as well as those of many others, it is evident that contractility cannot be considered independent of afterload, as is required in the traditional concept of cardiac function. This interaction also calls into question the search for so-called load-insensitive indexes of contractility that in the traditional concept, are critical for evaluation of the intrinsic contractile properties of the myocardium. In the traditional concept, the four independent determinants of ventricular performance are heart rate, preload, afterload and contractility. An index of contractility should be sensitive to changes in intrinsic contractile properties of the myocardium but insensitive to changes in heart rate, preload and afterload. However, the dependence of contractility on heart rate has long been appreciated (39,40), and its dependence on preload has been demonstrated in isolated muscle (41) and intact hearts (42). In fact, the Frank-Starling relation has recently been described entirely in terms of length-dependent activation (41). That is, the entire effect of increased preload on performance can be ascribed to increases in contractility induced by increased calcium availability to the myofilament or increased sensitivity of the myofilament to calcium, or both. Thus, it is not surprising that the effects of afterload on performance may also be in part mediated by early alterations in activation. However, it remains to be investigated whether this positive effect of increased afterload is also operative in the heart with impaired pump function.

An important confounding effect in this respect is the possibility of history-dependent effects. There is experimental evidence (43) that there is intrinsic interaction between single-beat contractility and both afterload and preload. This would have implications for our results if starting conditions would vary greatly. However, Table 2 shows that the steady state starting conditions (before any preload intervention) were not very different at all. End-diastolic volume was almost the same, and stroke volume increased with afterload rather than decreased; however, this change in stroke volume was not significant. These data by themselves show evidence of homeometric autoregulation, independent of preload interventions and thus of history-dependent effects.

Study limitations. There are some potential limitations to the present study. 1) Our results may have been influenced by our technique of measuring left ventricular volume. Although the conductance catheter has proved very reliable in measuring stroke volume, there is some doubt about its ability to accurately measure absolute volume (e.g., end-diastolic volume). To ensure that our absolute volume measurements were accurate in relation to one another at each level of afterload, we measured the parallel conductance volume at each level. Furthermore, it has been shown by our laboratory (4) as well as by others (44) that changes in hemodynamic conditions are reflected equally by conductance catheter- and by sonomicrometer crystal-derived volumes. It is also important to point out that the presence of homeometric autoregulation was proved by stroke volume measurements at the same end-diastolic volume for each lamb, which makes it unlikely that there is a systematic error in our measurements.

2) It is important to ensure that afterload did not change parasympathetic or sympathetic tone, resulting in changes in contractility secondary to these neurohumoral changes. Parasympathetic reflexes were unlikely because the lambs received atropine in adequate dosages to block all parasympathetic reflexes. Sympathetic reflexes are not easy to block, but we think that they were of minor importance for several reasons. Increased sympathetic tone would have increased heart rate independent of atropine blockade, and we did not find any change in heart rate at any level of afterload. In a previous study (17) using the same preparation, we found that the newborn lamb is very well able to increase its heart rate in response to adrenergic stimulation. b) Studies in adult dogs (4) have shown that the effect of afterload on contractility is reduced by beta-adrenergic stimulation rather than enhanced. c) We recently showed (45) that surgical sympathectomy does not change the effect of afterload on contractility. Thus, we believe that the changes that we observed in contractility were not caused by neurohumoral changes.

3) Another limitation is the small number of animals studied over a relatively wide age range (3 to 14 days). It has been shown (15) that newborn lambs demonstrate a maturational change in contractile reserve. However, in that study the major increase in contractile reserve occurred in the third and fourth weeks after birth, whereas in the first and second weeks after birth the lambs showed similar limitations in contractile reserve. In the present study the increases in contractility with increases in afterload were very similar in the youngest versus the oldest animals. However, the small size of the study makes it difficult to draw definite conclusions about age-related differences.

Conclusions. The present study in newborn lambs has shown that stepwise increases in afterload increase contractility considerably and that this is reflected by the maintenance of stroke volume at higher afterloads. The close relation between afterload and contractility forces us to reevaluate our concept of ventricular performance. Indexes of contractility, such as the end-systolic pressure–volume relation, do reflect contractile properties of the myocardium, but increases in afterload and in frequency also affect these properties. The mechanism through which this occurs is likely to reside in the intracellular calcium turnover (e.g., availability, utilization, sensitivity, reuptake) in much the same way as calcium sensitivity appears to be the major factor responsible for the Starling mechanism. Although it may be useful to investigate these factors further on the cellular level, it should be recognized that they also play an important role in the macroscopic evaluation of ventricular function in the normal as well as the diseased heart.

References

1. Suga H, Sagawa K. Instantaneous pressure–volume relationships and their ratio in the excised, supported canine left ventricle. Circ Res 1974;35:117–26.
2. Baan J, Van Der Velde ET, Steendijk P. Ventricular pressure–volume relations in vivo. Eur Heart J 1992;13 Suppl E:2–6.
3. Sarnoff SJ, Mitchell JH, Gilmore MS, Remensnyder JP. Homeometric autoregulation in the heart. Circ Res 1960;8:1077–91.
4. Baan J, Van Der Velde ET. Sensitivity of left ventricular end-systolic pressure–volume relation to type of loading intervention in dogs. Circ Res 1988;62:1247–58.
5. Van Der Velde ET, Burkhoff D, Steendijk P, Karsdon J, Sagawa K, Baan J. Nonlinearity and load sensitivity of end-systolic pressure–volume relation of canine left ventricle in vivo. Circulation 1991;83:315–27.
6. Spratt JA, Tyson GS, Glower DD, et al. The end-systolic pressure–volume relationship in conscious dogs. Circulation 1987;75:1295–309.
7. Sodums MT, Badke FR, Starling MR, Little WC, O'Rourke RA. Evaluation of left ventricular contractile performance utilizing end-systolic pressure–volume relationship in conscious dogs. Circ Res 1984;54:731–9.
8. Kass DA, Maughan WL, Guo ZM, Kono A, Sunagawa K, Sagawa K. Comparative influence of load versus inotropic states on indexes of ventricular contractility: experimental and theoretical analysis based on pressure–volume relationships [published erratum appears in Circulation 1988;77:559]. Circulation 1987;76:1422–36.
9. Burkhoff D, De Tombe P, Hunter WC, Kass DA. Contractile strength and mechanical efficiency of left ventricle are enhanced by physiological afterload. Am J Physiol 1991;H569–78.
10. Van Den Bos GC, Elzinga G, Westerhof N, Noble MIM. Problems in the use of indices of myocardial contractility. Cardiovasc Res 1973;7:834–48.
11. Little WC, Cheng CP, Peterson T, Vinten-Johansen J. Response of the left ventricular end-systolic pressure–volume relation in conscious dogs to a wide range of contractile states. Circulation 1988;78:736–45.
12. Kaufman TM, Horton JW, White DJ, Mahony L. Age-related changes in myocardial relaxation and sarcoplasmic reticulum function. Am J Physiol 1990;259:H309–16.
13. Mahony L. Maturation of calcium transport in cardiac sarcoplasmic reticulum. Pediatr Res 1988;24:639–43.
14. Friedman WF. The intrinsic physiologic properties of the developing heart. In: Friedman WF, Lesch M, Sonnenblick EH, editors. Neonatal Heart Disease. New York: Grune & Stratton, 1973:21–49.
15. Teitel DF, Sidi D, Chin T, Brett C, Heymann MA, Rudolph AM. Developmental changes in myocardial contractile reserve in the lamb. Pediatr Res 1985;19:948–55.
16. Anderson PA, Glick KL, Manring A, Crenshaw CJ. Developmental changes in cardiac contractility in fetal and postnatal sheep: in vitro and in vivo. Am J Physiol 1984;247:H371–9.
17. Teitel DF, Klautz RJM, Steendijk P, Van Der Velde ET, Van Bel F, Baan J. The end-systolic pressure–volume relationship in the newborn lamb: effects of loading and inotropic interventions. Pediatr Res 1991;29:473–82.
18. Baan J, Van Der Velde ET, De Bruin HG, et al. Continuous measurement of left ventricular volume in animals and humans by conductance catheter. Circulation 1984;70:812–23.
19. Baan J, Van Der Velde ET, Steendijk P, Koops J. Calibration and application of the conductance catheter for ventricular volume measurement. Automedica 1989;11:357–65.
20. Cassidy SC, Teitel DF. The conductance volume catheter technique for measurement of left ventricular volume in young piglets. Pediatr Res 1992;31:85–90.
21. Glantz SA, Slinker BK. Primer of Applied Regression and Analysis of Variance. New York: McGraw-Hill, 1990:777.
22. Nishioka O, Maruyama Y, Ashikawa K, et al. Load dependency of end-systolic pressure–volume relations in isolated, ejecting canine hearts. Jpn Heart J 1988;29:709–22.
23. Elzinga G, Westerhof N. "Pressure–volume" relations in isolated cat trabeculae. Circ Res 1981;49:388–94.
24. Su JB, Crozatier B. Preload-induced curvilinearity of left ventricular end-systolic pressure–volume relations. Effects on derived indexes in closed-chest dogs. Circulation 1989;79:431–40.
25. Suga H, Kitabatake A, Sagawa K. End-systolic pressure determines stroke volume from fixed end-diastolic volume in the isolated canine left ventricle under a constant contractile state. Circ Res 1979;44:238–49.
26. Brady AJ. Onset of contractility in cardiac muscle. J Physiol 1967;184:560–80.
27. Brutsaert DL, Sonnenblick EH. Force–velocity–length–time relations of the contractile elements in heart muscle of the cat. Circ Res 1969;24:137–49.
28. Jewell BR. A reexamination of the influence of muscle length on myocardial performance. Circ Res 1977;40:221–30.

29. Leash JK, Brady AJ, Skipper BJ, Millis DL. Effects of active shortening on tension development of rabbit papillary muscle. Am J Physiol 1980;238:H8–13.
30. Clancy RL, Graham TJ, Ross J Jr, Sonnenblick EH, Braunwald E. Influence of aortic pressure-induced homeometric autoregulation on myocardial performance. Am J Physiol 1968;214:1186–92.
31. Monroe RG, Gamble WJ, LaFarge CG, et al. The Anrep effect reconsidered. J Clin Invest 1972;51:2573–83.
32. Vatner SF, Monroe RG, McRitchie RJ. Effects of anesthesia, tachycardia, and autonomic blockade on the Anrep effect in intact dogs. Am J Physiol 1974;226:1450–6.
33. Donald TC, Peterson DM, Walker AA, Hefner LL. Afterload-induced homeometric autoregulation in isolated cardiac muscle. Am J Physiol 1976;231:545–50.
34. Wahler GM, Swayze CR, Fox IJ. A Ca^{2+}-dependent mechanism for the positive inotropic response to an increase in afterload in cat papillary muscle. Can J Physiol Pharmacol 1984;62:296–301.
35. Nichols CG, Hanck DA, Jewell BR. The Anrep effect: an intrinsic myocardial mechanism [published erratum appears in Can J Physiol Pharmacol 1989;67:172]. Can J Physiol Pharmacol 1988;66:924–9.
36. Elzinga G, Noble MIM, Stubbs J. The effect of an increase in aortic pressure upon the inotropic state of cat and dog left ventricles. J Physiol 1977;273:597–615.
37. Van Hare G, Hawkins JA, Schmidt KG, Rudolph AM. The effects of increasing mean arterial pressure on left ventricular output in newborn lambs. Circ Res 1990;67:78–83.
38. Hawkins J, Van Hare GF, Schmidt KG, Rudolph AM. Effects of increasing afterload on left ventricular output in fetal lambs. Circ Res 1989;65:127–34.
39. Bowditch HP. Über die Eigentümlichkeiten der Reizbarkeit, welche die Muskelfasern des Herzens zeigen. Ber Konigl Sachs Ges Wissen 1871;23:652.
40. Freeman GL, Little WC, O'Rourke RA. Influence of heart rate on left ventricular performance in conscious dogs. Circ Res 1987;61:455–64.
41. Lakatta EG. Starling's law of the heart is explained by an intimate interaction of muscle length and myofilament calcium activation. J Am Coll Cardiol 1987;10:1157–64.
42. Tucci PJF, Bregagnollo J, Cicogna AC, Ribeiro MCL. Length dependence of activation studied in the isovolumic blood-perfused dog heart. Circ Res 1984;55:59–66.
43. Slinker BK, Glantz SA. Beat-to-beat regulation of left ventricular function in the intact cardiovascular system. Am J Physiol 1989;R962–75.
44. Applegate RJ, Cheng CP, Little WC. Simultaneous conductance catheter and dimension assessment of left ventricle volume in the intact animal. Circulation 1990;81:638–48.
45. Schipper IB, Steendijk P, Klautz RJM, Van Der Velde ET, Baan J. Cardiac sympathetic denervation does not change the load dependence of the left ventricular end-systolic pressure/volume relationship in dogs. Eur J Physiol 1993;425:426–33.