서울醫大學術誌 제24권 제2호
The Seoul Journal of Medicine
Vol. 24, No. 2, June 1983

Factors Influencing the Peak Acceleration of Ventricular Ejection Blood Flow in the Conscious Dog

Hee C. Kim and Byoung G. Min

Department of Biomedical Engineering, College of Medicine, Seoul National University

INTRODUCTION

Rushmer suggested that the ventricle acts as an impulse generator and the "initial ventricular impulse" can be used as a useful index of myocardial performance(Rushmer, 1964). This view was further supported by Noble et al. (Noble, 1966) from the observation of the high sensitivity of the maximum acceleration of the blood flow in the early ejection phase to intracoronary injection of stimulating drugs, prior to any changes in stroke volume. Based upon these observations, the peak acceleration of the blood flow in the ascending aorta was proposed as an index of the contractile state of the left ventricle.

On the other hand, Wilcken and his co-investigators observed that the peak acceleration was changed with the alterations in aortic impedance (Wilcken, 1964). In this case, the peak acceleration cannot represent the intrinsic left ventricular contractile state.

To evaluate the above conflicting results, we performed animal experiment using a conscious dog, and then, studied quantitatively the hemodynamic factors influencing the peak acceleration of aortic flow using the equivalent circuit analysis of the left ventricle—systemic circulation model.

† 접수일자 : 1983. 3. 9.
* 이 논문은 1983년도 서울대학교 병원 특진연구비 보조로 이루어진 것임.
** 본 동물실험은 미국 Duke 대학 Cardiology Lab. 에서 행하여졌음.

EXPERIMENT AND RESULTS

To study the afterload dependency of the peak acceleration in the conscious dog, we performed the following experiment. The experimental protocol was similar to Noble et al.'s (Noble, 1966) and Wilcken et al.'s (Wilcken, 1964) except that the present measurement was performed on the conscious dog without anesthesia.

Animal Preparation. On the first day of experiment, the measuring transducers were implanted on a mongrel dog after anesthesia. An electromagnetic flow probe was fitted around the root of the ascending aorta, and a later balloon was looped around the descending aorta. This balloon was used to produce a partial aortic occlusion after the balloon inflation. A pacing catheter was used, and a saline-filled catheter was located in the left ventricle for measurement of L.V. pressure. Ultrasonic transducers for a sonocardiometer were attached to the epicardium of the left ventricle for measurement of the L.V. dimension. Ten days after the above transducer implantation, the experiment was performed in conscious state without anesthetization, and these data were used for the analysis.

Measurements and Data Processing. Aortic flow signal was used as an input to an electronic derivative circuit for measuring the peak acceleration of aortic flow. A four channel paper recorder was used for recording of L.V. pressure, the aortic root flow, the acceleration of aortic

flow, and the L.V. dimension. A Houston Instrument HI-PAD™ digitizer was used to digitize the paper recorded data as the input to a PDP MINC-11 computer for further analysis. The ejection phase was selected using the points where aortic flow crossed zero level. Then, the two points on the L.V. pressure waveform corresponding to the onset and the ending of the ejection phase were used as the arterial end-diastolic pressure (Pad) and the arterial end-systolic pressure (Pas). Between the two points of Pas and Pad, aortic pressure was connected by a mono-exponential curve, and its time constant was computed from the values of Pas, Pad, and the diastolic duration. Left ventricular mean pressure and aortic mean flow was calculated during the ejection period, and the equivalent input resistance, Req, was computed as the ratio of the mean pressure to the mean flow. Source pressure, Pg, was estimated using Fast Fourier Series from the two adjacent beats using the data of L.V. pressure and aortic flow (Min, 1976). A correlation coefficient between the peak acceleration and Pad was also calculated.

RESULTS

The results of the present study were basically similar to the experimental results reported by Wilcken et al. (Wilcken, 1964). The peak acceleration diminished by about 17% after aortic occlusion, and it increased by 33% after release of the aortic occlusion. We could also observe that the elevation of the peak acceleration after balloon deflation was greater than its diminution after balloon inflation. This result was also consistent with other investigators' observations (Wilcken, 1964).

A. Effects of sudden increase in aortic impedance after partial aortic occlusion: When the aortic impedance increased after aortic occlusion, the left ventricular pressure increased in the following beat, and the aortic flow and its peak accceleration decreased temporarily. During the period of two consecutive beats, there was no significant alteration in the performance of the left ventricle as observed by stable L.V. dimension waveform in the consecutive beats. The arterial end-diastolic pressure, Pad, and the end-systolic arterial pressure, Pas, increased linearly after occlusion. The time constant of the exponential decay curve of the diastolic arterial pressure has increased. Also, the equivalent resistance, Req, increased, but the ejection period decreased slightly. After these transient phenomena, the aortic flow, the accloration, and the left ventricular dimension returned to the pre-occlusion state. With the partial occlusion remaining for about three minutes, all these hemodynamic data increased significantly, presumably due to the autonomic nerve activity.

B. Effects of sudden reduction in aortic impedance after release of aortic occlusion: After three minutes of the above partial aortic occlusion, the balloon was deflated abruptly to observe the changes of the peak acceleration after diminution of aortic input impedance. With the release of aortic occlusion, there was significant increases of aortic flow and the peak acceleration together with a large reduction in left ventricular pressure in the first two or three beats. During this period, the data of Pad, Pas, the time constant, and Req decreased significantly, and the ejection time increased slightly. Following these transient changes, L.V. pressure and aortic flow returned to its normal level gradually.

The hemodynamic data are summarized in Table 1 and 2 for pre- and post-aortic occlusion. The correlation between the peak acceleration and the end-diastolic arterial pressure is shown in Table 3.

Table 1. Hemodynamic parameters before and after impedance elevation caused by aortic occlusion

Heart Beat No.	Experimental Procedure	Pad (mmHg)	Pas (mmHg)	Mean Pressure (mmHg)	Mean Flow (L/min)	Req (mmHg/L/min)	Time Constant (sec.)	Systolic Time (msec.)	Diastolic Time (msec.)
1	Pre-Occlusion	76.8	99.8	122.9	3.53	34.8	2.06	49.8	64.6
2		85.1	120.4	129.5	3.47	37.3	2.53	47.1	68.8
3	Post-Occlusion	105.0	136.5	142.6	2.93	48.7	3.07	47.1	70.3
4		121.7	144.4	147.5	3.01	49.0		41.8	

Table 2. Hemodynamic parameters after impedance diminution caused by release of the aortic occlusion

Beat	Experimental Procedure	Pad (mmHg)	Pas (mmHg)	Mean Pressure (mmHg)	Mean Flow (L/min.)	Req (mmHg/L/min)	Time Constant (sec.)	Systolic Time (msec.)	Diastolic Time (msec.)
1	Pre-Release	100.9	126.4	143.2	4.09	35.0	0.73	54.8	58.6
2		84.2	128.5	139.2	4.06	34.3	0.70	55.5	59.7
3		83.6	112.1	134.9	4.37	30.9	0.34	57.0	56.3
4	Post-Release	48.0	90.8	110.8	6.16	18.0	0.43	55.9	59.7
5		54.0	77.3	106.4	5.98	17.8		56.3	

Table 3. The relationship between the peak acceleration and the end-distolic arterial pressure, Pad

Experimental Procedure.	Beat No.	1	2	3	4	5	Correlation Coefficient.
After aortic occlusion from the control state	Peak Acceleration (L/min^2)	1.353	1.393	1.191	1.12		
	Pad (mmHg)	76.8	85.1	105.0	121.7		−0.951
After release of aortic occlusion	Peak Acceleration (L/min^2)	1.592	1.559	1.604	1.949	2.077	
	Pad (mmHg)	100.9	84.2	83.6	48.0	45.0	−0.943

THEORETICAL ANALYSIS

To evaluate quantitatively the present experimental results of the changes of the peak acceleration in response to the changes of aortic input impedance, we have used the equivalent circuit analysis on the left ventricle-systemic circulation model of other investigators (Abel, 1966, 1971; Buoncristiani, 1973; Elzinga, 1973, 1974, 1976; Westerhof, 1973). In theoretical analysis, the left ventricle is considered to consist of a voltage source (*Pg*) and a series source impedance (*Zg*) in Fig. 1. The afterload of left ventricle is simulated in the model as a combination of a diode of aortic valve in series with an intertance(L), and a resistance(*Ra*), representing aortic charateristic impedance. For the peripheral circulation, the arterial capacitance (*C*) is connected in parallel with a peripheral resistance (*Rp*) as shown in Fig. 1. In the model, the voltages are equivalent to

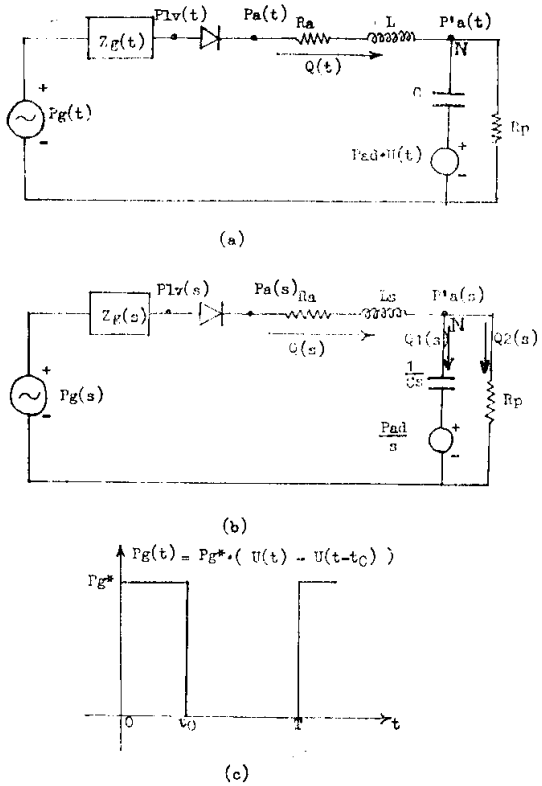


Fig. 1. (a) The equivalent circuit model of the ventricle-arterial circulatory system,
(b) The circuit model represented by Laplace transform with initial conditions,
(c) Waveform of source pressure, $Pg(t)$.

the pressure components, and the current represents the blood flow.

For the sake of convenience, the present analysis will be restricted to the ejection phase where the aortic valve opens at $t=0$, and it will be assumed that the ventricular pressure, $Plv(t)$, equals to root aortic pressure, $Pa(t)$, at all times when the aortic valve is open.

In Fig. 1, the aortic flow during ejection phase is related to the source parameter and the vascular input impedance, as well as the arterial end-diastolic pressure, acting as the capacitance's initial stored voltage. Thus, in circuit analysis, Pad is replaced by an initial condition generator, $Pad \cdot U(t)$, as shown in Fig. 1-(a).

Applying Kirhhoff's current law at the node N for the ejection period, the Laplace transform of aortic flow, $Q(s)$, becomes

$$Q(s)=Q1(s)+Q2(s) \tag{1}$$

$$\text{or } Q(s)=\frac{Pg(s)-P'a(s)}{Zg(s)+Ra+Ls}$$

$$=Cs\cdot\left(P'a(s)-\frac{Pad}{s}\right)+\frac{1}{R_p}\cdot P'a(s) \tag{2}$$

where $P'a$ is the pressure at node N, and (s) represents the Laplace transformation.

Eliminating $P'a(s)$ from Eq. (2), the ejection flow transform, $Q(s)$, can be related as follows;

$$\left\{1+\left(Cs+\frac{1}{R_p}\right)\cdot(Zg(s)+Ra+Ls)\right\}\cdot Q(s)$$

$$=\left(Cs+\frac{1}{R_p}\right)\cdot Pg(s)-C\cdot Pad \tag{3}$$

Representing source pressure $Pg(t)$ as an impulse with finite duration as shown in Fig. 1-(c), based upon Rushmer's observations (Rushmer, 1964) that the pumping action of the ventricles can be represented in terms of initial impulse, defined as the product of force and time, we can assume for the analysis that the maximum acceleration occurs at the early onset phase of ejection close to the instant of $t=0^+$. This analysis is also related to the report (Rushmer, 1964) that the maximum rate of myocardial shortening occurs at the onset of contraction so that initial velocity is the greatest velocity during contraction. Then, using the properties of Laplace transform, related to the differentiation with respect to time and the initial-value property, the peak acceleration can be obtained as follow;

$$\left.\frac{dQ(t)}{dt}\right|_{\max}=\left.\frac{dQ(t)}{dt}\right|_{t=0^+}\leftrightharpoons\lim_{s\to\infty}s\cdot[sQ(s)],$$

and from Eq. (3), (4)

$$\lim_{s\to\infty}s\cdot[sQ(s)]$$

$$=\lim_{s\to\infty}\frac{s^2[(Cs+1/R_p)\cdot Pg(s)-CPad]}{(Cs+1/R_p)\cdot(Zg(s)+Ra+Ls)+1}$$

where "$\leftrightharpoons$" represents a Laplace transform pair.

At the infinite limit of the right side of Eq.

(4), the lower power terms of "s" can be deleted, and result becomes;

$$\left.\frac{dQ(t)}{dt}\right|_{max} \Leftrightarrow \lim_{s\to\infty} \frac{s^2 \cdot Pg(s) + 1/R_pC \cdot s\, Pg(s) - Pad \cdot s}{Ls + Zg(s)} \quad (5)$$

For the source impedance, $Zg(s)$, as it consists of passive lumped-elements, the degree of its numerator polynomial may be the same as the degree of the denominator polynomial, or they can be different only by unity. Then, the limit value of $Zg(s)$ becomes;

$$\lim_{s\to\infty} Zg(s) =$$

$$\begin{cases} \text{Constant including zero,} \\ \text{when degree of numerator} \leqq \text{degree of denominator.} \\ Zg^* \cdot s, \\ \text{when degree of numerator} > \text{degree of denominator, and } Zg^* \text{ is constant.} \end{cases} \quad (6)$$

From Fig. 1-(c), the Laplace transform of source pressure, $Pg(t)$, can be represented as,

$$Pg(s) = \frac{Pg^*}{s}\,(1 - \exp(-st_0)), \quad (7)$$

where Pg^* is constant.

From Eqs. (6) and (7), the peak acceleration can be described as follows;

$$\left.\frac{dQ(t)}{dt}\right|_{t=0+}^{max} = \begin{cases} \dfrac{Pg^* - Pad}{L}, & \text{when } \lim_{s\to\infty} Zg(s) = \text{constant.} \\ \dfrac{Pg^* - Pad}{L + Zg^*}, & \text{when } \lim_{s\to\infty} Zg(s) = Zg^* \cdot s. \end{cases} \quad (8)$$

The final result of Eq. (8) shows that the arterial end-diastolic pressure, Pad, as well as Pg^* can affect the peak acceleration for the constant values of L and Zg^* during ejection phase.

Then, we have sutdied the factors influencing Pad. When the aortic valve is closed, the peripheral pressure, $P'a(t)$, is equal to the aortic root pressure, $Pa(t)$, and it decreases exponentially from the aortic (or arterial) pressure at the valve closing time, Pas, down to Pad with a time constant of R_pC during the diastolic phase in Fig. 1-(a). Thus, the arterial end-diastolic pressure is determined by the properties of the arterial tree (time constant, R_pC), the factor related to the preceding beat (Pas), and the diastolic time duration, $(T - t_0)$. Using the voltage division rule in Fig. 1-(b), the transform of the arterial pressure, $P'a(t)$, before and after valve closing time, $t = t_0^+$, can be represented as follows;

$$P'a(s) = \frac{\dfrac{1}{(1/R_p + Cs)}}{(Zg(s) + Ra + Ls) + \dfrac{1}{(1/R_p + Cs)}} \cdot Pg(s), \quad (9)$$

$$\text{or} = \frac{R_p}{R_p + (1 + R_pC \cdot s)\,(Zg(s) + Ra + Ls)} \cdot Pg(s)$$

for $0^+ \leq t \leq t_0$,

and

$$P'a(s) = \frac{\exp(-st_0)}{s + R_pC} \cdot Pas,$$

or (10)

$$P'a(t) = Pas \cdot \exp\left[-(t - t0)/R_pC\right]$$

for $t_0 \leq t \leq T$.

Eq. (9) can be simplified to Eq. (11) using an approximation of the term $(1 + R_pC \cdot s)$ to unity for typical values of R_p and C.

$$P'a(s) = \frac{R_p}{R_p + (Zg(s) + Ra + Ls)} \cdot Pg(s)$$

for $0^+ \leq t \leq t_0$. (11)

For the two consecutive beats before and after aortic occlusion, we can expect that with constant $Pg(s)$ and $(Zg(s) + Ra + Ls)$, and with the increases of peripheral resistance Rp, each harmonics of $P'a(t)$ grows parabolically to those of $Pg(t)$. The above analysis shows that aortic pressure at the time of valve closing, Pas, increases to source pressure Pg at the time of $t = t_0$ with an increase of Rp.

As a result of circuit analysis, we can conclude that, for the same Pg, the peak acceleration of ejected blood changes as a function of the arterial end-diastolic pressure, Pad. On the other hand, Pad is determined by Pas, the

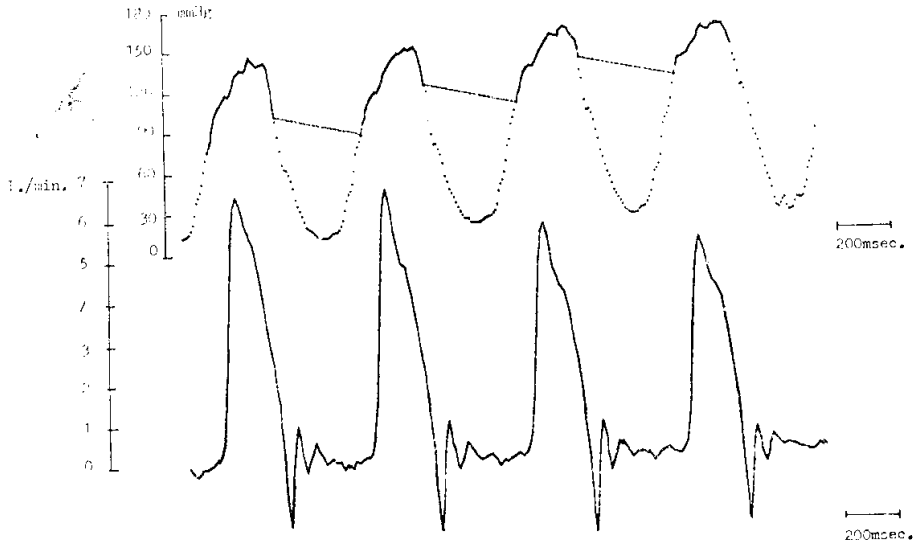


Fig. 2-(a). Changes of aortic pressure (upper; line), L.V. pressure (upper; dot) and aortic flow (lower) waveforms before (first two beats) and after partial aortic occlusion.

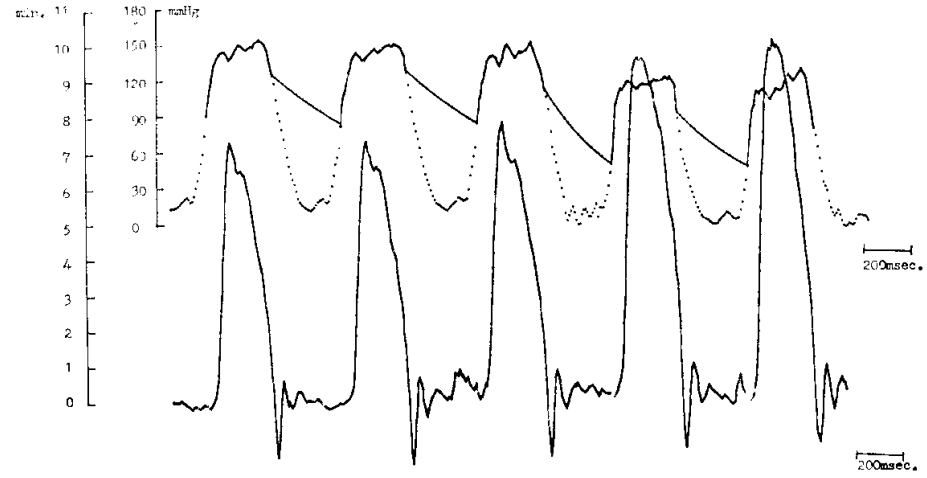


Fig. 2-(b). Same waveform before (first three beats) and after release of the partial aortic occlusion.

time constant(RpC), and the diastolic period. Also, it is shown that the end-systolic arterial pressure, Pas, represnets a portion of the source pressure, *Pg*, with its magnitude varies depending upon the ratio of the peripheral resistance to the sum of the source and characteristic impedences.

DISCUSSION

Theoretical analysis and animal experiment in the conscious dog with the chronally implanted transducers were performed to study the factors influencing the peak acceleration of left ventricular blood flow. As a result of circuit analysis of Eq. (8), the peak acceleration is

shown to be related to the difference between the source pressure and the end-diastolic arterial pressure, Pad. The experimental data also supported this analysis result with high correlation coefficient between the peak acceleration and the end-diastolic pressure.

In the second-order mechanical system consisting of a mass(M) only, the applying force (F) is related to the acceleration of the mass by Newton's second law; $F=M\cdot a$. This relationship can be expressed in terms of electrical circuit elements in direct analogue form by the equation; $v(t)=L\cdot\frac{di(t)}{dt}$ or $\frac{di(t)}{dt}=\frac{1}{L}\cdot v(t)$. Since Noble et al. assumed that at the time of maximum acceleration, only the inertiance component dominates the opposition to left ventricular ejection(Noble, 1966), the peak acceleration of flow was shown to be directly related to the afterload independent cardiac force, which can be represented by Pg^*/L in the circuit analysis.

However, the left ventricle and its peripheral circulatory system can be more accurately represented by being consisted of a force generator (pressure source) which the source impedance, and they are connected to combinations of mass (inertia), resistance component, and the elastance component. The present analysis shows that even the peak acceleration at the onset of the impulse-like pressure source is dependent upon the factors including source and peripheral impedances. In our present study and other investigators' experimental study, the peak acceleration was affected by the alteration of input impedance. Especially, in our circuit analysis, Pad was shown to be an important parameter, where Pad was affected by the changes of the peripheral resistance and arterial capacitance in addition to the source parameters. Changes of the input impedance can alter the time constant of arterial pressure waveform during diastolic phase and the end-systolic arterial pressure Pas, which, in turn, led to the alteration of Pad. The present experiment confirmed the above theoretical results by showing very high correlation coefficients between the two parameters (peak acceleration and Pad) in the ranges of -0.95. In the present study, we have assumed that the source pressure acts as an impulse with finite duration, and thus, the peak acceleration occurs at the onset of ejection. These are based upon Rushmer's conclusion (Flanklin, 1962; Rushmer, 1964) and other authors' observations (Sonnenblick, 1962; 1962; Wilcken, 1964; Noble, 1966).

Another assumption of the present study is that, for two or three beats just after aortic occlusion or release of occlusion, the source pressure has not changed. This assumption is based on the present observations of stable ventricular dimensional waveform in two beats, and also, other investigators' observations (Levine, 1964; Noble, 1972; Noble, 1966; Min, 1976), suggesting that the changes in impedances between two consecutive beats do not result in variations in the contractile state.

As shown in Fig. 3 and Table 1 and 2, the computed source pressure, representing the contractile state of the ventricle has increased, when the partial occlusion state continued for three minutes. From the above results, we may conclude that the changes of the peak acceleration just after the changes of impedance condition are not caused by changes of the source pressure but due to the variation in the input impedance. Also, the differences in hemodynamic parameters and its changes for the balloon inflation case (Table 1) and the balloon deflation case (Table 2) may be mainly due to the difference of the source pressure between the two states.

Based upon the above results, the peak acceleration of left ventricular blood flow may be

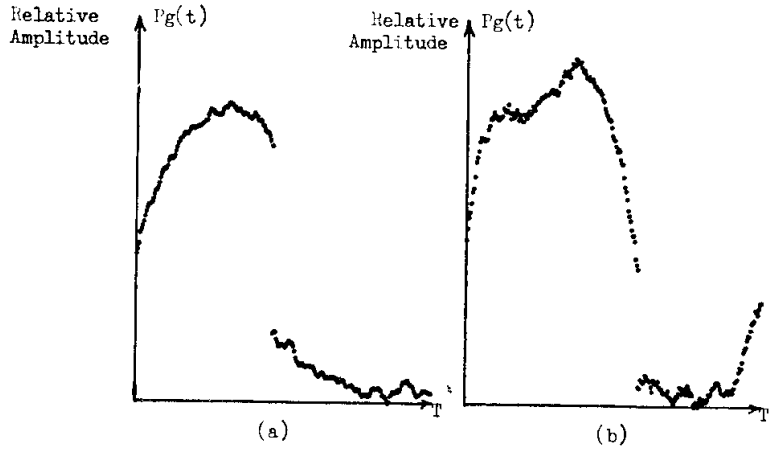
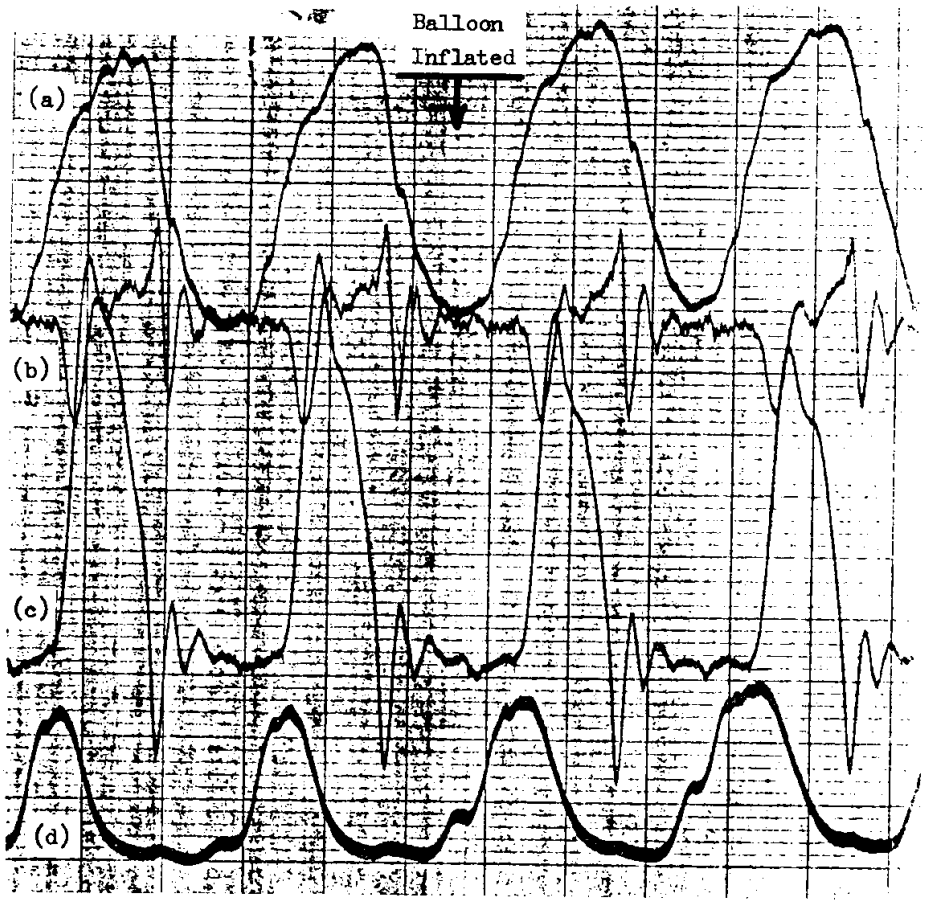
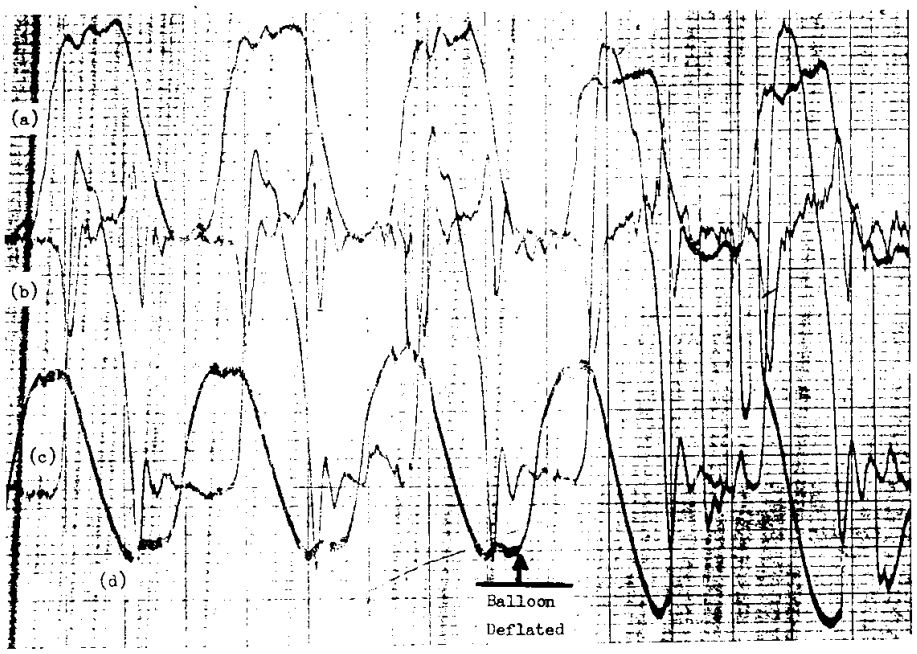


Fig. 3. Computed source pressure in control state (a); and after three minutes of continuous aortic occlusion (b).



Appendix I. Four channel paper recorded waveforms of (a) L.V. pressure, (b) acceleration of aortic flow, (c) aortic flow and (d) L.V. dimension before and after partial aortic occlusion.



Appendix II. Same waveforms before and after release of partial aortic ocelusion.

used as an index of the left ventricular contractile state, only when we compare the ventricular functions at the constant input impedance conditions. In the case of the altered arterial system, the peak acceleration may not be a reliable index of left ventricular contractile performance.

=國文抄錄=

左心室 最大血流加速度에 影響을 주는 生理變數에 관한 硏究

金 喜 贊·閔 丙 九
서울大學校 醫科大學 醫工學科

심근의 수축력(contractility)을 나타내는 생리변수로서 이미 여러 논문을 통해 그 중요성이 주장되어 온 "좌심실 최대혈류 가속도(Maximum Acceleration of the Left Ventricular Ejection flow)에 대해 등가모델을 이용한 정량적 해석 및 동물실험을 통한 분석을 실시하였다. 등가모델해석 결과로 최대혈류 가속도가 고유한 좌심실 심근의 수축상태(Contractile State)를 반영하기 위한 조건 및 이것에 영향을 미치는 요소들을 알 수 있었으며, 동물실험으로 이러한 이론적인 예상을 확인할 수 있었다. 특별히 본 연구의 동물실험에서는 좌심실 내의 혈류압력, 대동맥에서의 혈류속도 및 가속도, 그리고 좌심실 외벽간의 길이 변화등을 연속적으로 관찰할 수 있는 측정기들을 부착시킨 개(Dog)를 사용하여 의식상태에서의 데이타를 수집, 분석하였다. 본 연구의 결과, 좌심실 최대혈류가속도는 좌심실 확장 말기때의 동맥압(End-diastolic Arterial Pressure)에 따라 변하는 것을 알 수 있었으며, 이 두 변수들간의 상관관계가 −0.9 이상으로 매우 높게 나타났다. 그런데 심실확장말기의 동맥압은 좌심실에 걸리는 부하(Afterload)의 변동에 따라 변하게 되어 고유한 심근수축상태를 나타낼 수 없다는 것을 알 수 있었다. 그러나 한편으로 좌심실 최대혈류가속도는 좌심실 고유의 기능(Performance)을 나타내는 변수와도 비례하게 되어 적절한 조건하에서는 좌심실 수축상태를 반영할 수도 있게 됨을 알 수 있었다.

REFERENCES

Abel, F.L.: *Fourier analysis of left-ventricular performance. Circ. Res.*, 28:*119-135, 1971.*

Abel, F.L.: *An analysis of left ventricles as a pressure and flow generator in the intact systemic circulation. IEEE Trans. Biomed. Eng.*, 13:*182-*

188, 1966.

Buoncristiani, J.F., Liedtke, A.J., Strong, R.M. and Urschel, C.W.: *Parameter estimates of a left-ventricular model during ejection. IEEE Trans. Biomed. Eng.*, **20**:*110-114, 1973.*

Elzinga, G. and Westerhof, N.: *Pressure and flow generated by the left ventricle against different impedances. Circ. Res.*, **32**:*178-186, 1973.*

Elzinga, G. and Westerhof, N.: *End-diastolic volume and source impedance of the heart. In The physiological basis of Starling's law of the heart, Ciba Foundation Symposium.*, **24**:*251-255, 1974.*

Elzinga, G. and Westerhof, N.: *The pumping ability of the left heart and the effect of coronary occlusion. Circ. Res.*, **38**:*297-302, 1976.*

Franklin, D.L., Citters, R.L.V. and Rushmer, R.F. *Left ventricular function described in physical terms. Circ. Res.*, **14**:*702-711, 1962.*

Levine, H.F. and Britman, N.A.: *Force-velocity relation in the intact dog heart. J. Clin. Invest.*, **43**:*1383-1396, 1964.*

Min, B.G., Fich, S., Kostis, J.B., Doblar, D. and Kuo, P.T.: *Sensitivity and afterload independence of zero-load aortic flow. Annals of Biomed. Eng.*, **4**:*330-342, 1976.*

Nole, M.I.M., Trenchard, D. and Guz, A.: *Left ventricular ejection in conscious dogs: 1. Measurement and significance of the maximum acceleration of blood from the left ventricle. Circ. Res.*, **19**: *139-147, 1966.*

Noble, M.I.M., Stubbs, J., Trechard, D., Else, W., Eisele, J.H. and Guz, A.: *Left-ventricular performance in the conscious dog with chronically denervated heart. Cardiovasc. Res.*, **6**:*457-477, 1972.*

Rushmer, R.F.: *Initial ventricular impulse: A potential key to cardiac evaluation. Circulation*, **29**:*268-283, 1964.*

Sonnenblick, E.H.: *Force velocity relations in mammalian heart muscle. Am. J. Physiol.*, **202**:*931, 1962.*

Sonnenblick, E.H.: *Implications of muscle mechanics in the heart. Federation Proc.*, **21**:*975, 1962.*

Westerhof, N., Elzinga, G. and van den Bos, G.C.: *Influence of central and peripheral changes on the hydraulic iuput impedance of the systemic arterial tree. Med. Biol. Eng.*, **11**:*710-722,*

Wilcken, D.E.L., Charlier, A.A., Hoffman, J.I.E. and Guz, A.: *Effects of alterations in aortic impedance on the performance of the ventricle. Circ. Res.*, **16**:*283-293, 1964.*