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Myocardial and circulatory effects of *E. coli* endotoxin; modification of responses to catecholamines

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Summary

1. The predominant acute effect of *E. coli* endotoxin in anaesthetized, ventilated cats was pulmonary hypertension resulting from a 8–12 fold increase in pulmonary vascular resistance. This was followed by decreases in left ventricular (LV) and systemic arterial pressures and in LV dP/dt max. Recovery occurred within 2–4 min and was dependent upon increased sympathetic drive; recovery did not occur in cats treated with the β-adrenoceptor blocking drug alprenolol.

2. The pulmonary vasoconstriction was reduced in cats given compound 48/80 and evidence is presented that it results primarily from histamine release.

3. Over the 2–3 h period following endotoxin injection, systemic arterial pressure tended to decrease and heart rate and myocardial metabolic heat production to increase. Myocardial blood flow and LV dP/dt remained fairly stable until the terminal stages of shock.

4. The predominant delayed effects of *E. coli* endotoxin in cats were a markedly reduced stroke volume, an increase in peripheral vascular resistance and a severe metabolic acidosis (arterial base excess—20 mEq/litre). Arterial pO_2 and pCO_2 were not significantly affected. It is concluded that myocardial contractility is maintained at this time through the release of catecholamines and that endotoxin itself depresses contractility.

5. The effects of adrenaline and noradrenaline infusions on systolic and diastolic blood pressures, heart rate, cardiac output, myocardial blood flow and LV dP/dt max were markedly reduced in the period 2–3 h after endotoxin. In a few animals some recovery of the response to noradrenaline occurred and was associated with a general circulatory improvement and a reduced metabolic acidosis.

Introduction

There is still considerable disagreement regarding the relative roles of myocardial and peripheral vascular failure in irreversible shock. This is particularly true of circulatory collapse resulting from infection with gram-negative bacteria. Thus 'haemodynamic findings in various species provide no evidence for an important effect of endotoxin on myocardial function, the drop in cardiac output being almost solely due to impaired venous return' (Gilbert, 1960). In dogs, apart from an initial decrease in contractile force (Maxwell, Castillo, Crumpton, Afonso, Clifford & Rowe, 1960; Kadowitz & Yard, 1970) ventricular contractile capacity is maintained during the first three hours after endotoxin (Goodyer, 1967), and indeed often

until the preterminal stage (Alican, Dalton & Hardy, 1962). The reason for this maintained myocardial contractility is probably increased sympathetic drive, since ventricular function can be impaired by endotoxin in dogs given propranolol (Goodyer, 1967).

In contrast to these findings, Solis & Downing (1966), in a modified cat heart-lung preparation, showed that following endotoxin administration there was progressive myocardial deterioration, as indicated by a shift in the ventricular function curve to the right (i.e. decreased stroke volume at a constant left ventricular end-diastolic pressure). There is also evidence that endotoxin causes subendocardial haemorrhage and necrosis of myocardial fibres (Palmerio, Ming, Frank & Fine, 1962).

It seemed possible that the reason for these differences in myocardial function might be variations in cardiac sympathetic drive or endotoxin-induced variations in the responsiveness of cardiac and vascular smooth muscle to sympathetic neurotransmitters. The purpose of the present experiments was therefore to study the haemodynamic responses to endotoxin, particularly relating changes in myocardial contractility and output to simultaneously measured changes in myocardial tissue perfusion; an examination of effects on the pulmonary vascular bed and on systemic and mixed venous blood gases was included. It was hoped that this would provide the necessary background information for an experimental model which could be used for the examination of a number of pharmacological agents in shock. This paper also includes an examination of the relative responsiveness of the heart and peripheral vessels to catecholamines in the terminal stages of endotoxin shock.

Methods

Thirty-one cats weighing between 1·5 and 4·3 kg were anaesthetized with sodium pentobarbitone (30 mg/kg i.p.) and respired with room air using a Palmer positive pressure ventilation pump (rate 20/min; stroke volume 40–60 ml). Systemic arterial blood pressure was recorded with a capacitance transducer (Elema–Schönander EMT 35) from a catheter inserted, by way of a femoral or carotid artery, such that the tip lay in the descending aorta or aortic arch. Mean pressure was obtained by electronic integration. Systolic ejection time was measured (in ms) from the beginning of the upstroke of the central aortic pulse to the trough of the incisural notch.

Left ventricular pressure was recorded with a capacitance transducer (Elema–Schönander EMT 34) from a catheter inserted by way of the right common carotid artery or by direct left ventricular puncture with a wide bore steel needle. To permit the accurate determination of end-diastolic pressure (LVEDP) the pressure record was cut off above 25 mmHg (1 mmHg$\equiv$1·333 mbar) using a high gain recording. The rate of change of left ventricular pressure with time (dP/dt) was continuously determined with an analogue differentiator circuit. Right atrial pressure was recorded with a third capacitance transducer (EMT 33) from a catheter inserted by way of the right external jugular vein. This catheter was also used for the injection of 1·8 ml of 0·9% w/v NaCl (saline) at room temperature (18–24° C) in order to measure cardiac output by a thermodilution technique (Hosie, 1962). A 36 s.w.g. copper-constantan thermocouple, sheathed (except for the recording junction itself) in polyethylene tubing, was inserted by way of the right femoral artery such that the thermocouple junction lay in the aortic arch or in the upper portion of the descending aorta. The cold (reference) junction was placed in the

rectum together with a direct recording thermocouple (Ellab, Copenhagen). The output from the thermocouple circuit was fed directly into a Kipp & Zonen BD5 recorder (50 μV for a full-scale of 20 cm=1·2° C) at a paper speed of 200 mm/minute. The area under the thermodilution curve was calculated by the method of Williams, O'Donovan & Wood (1966) or with a Kipp & Zonen BCl integrator.

In fifteen of the cats the chest was opened and a heated thermocouple recorder implanted in the region supplied by the terminal branches of the anterior descending left coronary artery. The principle and general methodology for assessing myocardial tissue blood flow and metabolic heat production from heated thermocouples has been fully described in previous publications (Grayson & Mendel, 1961 ; Grayson & Parratt, 1966 ; McInnes & Parratt, 1969). In the present experiments, however, the recorder design was changed so that the temperature recording junction (made from 36 s.w.g. copper and constantan) was at the tip rather than 4 mm back (as in the original Grayson recorder). The heating element consisted of 25 turns of very fine (42 s.w.g.) enamelled copper wire wound around the copper lead, commencing 0·5 mm from the thermojunction. The entire recorder is considerably smaller than the one previously used and it is easier to judge the depth of insertion into ventricular muscle, so allowing comparisons to be made of deep (endocardial) and superficial (epicardial) flows. The second modification of the method outlined in a previous publication (McInnes & Parratt, 1969) concerns the electronic switch gear unit which supplies current cyclically to the heater leads. Currents and timing were reduced and the cycle used in the present experiments was: 0·00A (10 s), 0·387A (0·15A^2, 20 s), 0·592A (0·35A^2, 10 s) and 0·500A (0·25A^2, 20 s). The output from the heated thermocouple (with reference (unheated) thermojunction in the aortic arch) was fed directly into a second Kipp & Zonen recorder (100 μV for a full-scale deflection of 20 cm=2·5° C).

In seven experiments pulmonary artery (' downstream ') pressure was recorded from a needle inserted directly into the pulmonary artery ; in an additional experiment, a pulmonary lobular artery was catheterized with polyethylene tubing and pressure recorded normally (i.e. ' end pressure ') from this vessel. When ' downstream ' pulmonary pressure was measured, no attempt was made to correct for the kinetic energy artifact. Left ventricular pressure, left ventricular end-diastolic pressure (LVEDP), arterial pressure, pulmonary artery pressure, right atrial pressure, left ventricular (LV) dP/dt and the electrocardiogram (standard limb lead 11) were recorded on an eight-channel ink jet writing recorder (Elema-Schönander Mingograph 81).

Anaerobic blood samples (usually 1·0 ml) were taken at intervals from an artery and from either the right atrium or the pulmonary artery. Oxygen and carbon dioxide tensions and pH were recorded with appropriate electrode systems (Radiometer, Copenhagen). The pH electrode was calibrated by means of standard buffers and the oxygen and carbon dioxide electrodes with gas mixtures, the oxygen and carbon dioxide concentrations of which had been measured with a modified Lloyd-Haldane apparatus. Oxygen and carbon dioxide tensions (mmHg) and pH were corrected for any temperature difference between the electrode system and the animals mid-oesophageal temperature by the use of the blood gas calculator described by Severinghaus (1966).

The effects of infusions of adrenaline and noradrenaline (1·0 (μg/kg)/min as base), given by way of a femoral vein, were examined before and at various times

after the intravenous administration (slowly, over a period of 30–45 s) of 2 mg/kg of *E. coli* endotoxin (Difco Laboratories, 055:B5), suspended in 0·9% w/v sodium chloride solution. This dose is slightly below the LD50 in this species (Kadowitz & Yard, 1970). In some experiments the effects of endotoxin were examined after the histamine liberator compound 48/80 (2–25 μg/kg i.v. in repeated doses) and were also compared with the effects of intravenous infusions of histamine (5 (μg/kg)/min as base).

Results

Acute effects of E. coli *endotoxin*

Within 5–20 s of the end of the injection, right atrial and pulmonary artery pressures began to increase and this was followed (Fig. 1) by marked decreases in left ventricular pressure, LV dP/dt max and in systemic arterial pressure, often to levels below 50 mmHg. There was often a transient bradycardia and multiple ventricular extrasystoles also occurred, effects which can also be seen in Figure 1. The maximum effect on pulmonary artery pressure was observed 3–4 min after injection. The mean values at this time were, systolic pulmonary artery pressure (SPAP) 48 ± 7 mmHg and diastolic pulmonary artery pressure (DPAP) 35 ± 6 mmHg. These are significantly ($P<0{\cdot}01$; 5 experiments) higher than the control pre-endotoxin values of 20 ± 1 and 11 ± 1 mmHg respectively. This represents a highly significant (8–12 fold) increase in pulmonary vascular resistance (mean PAP–LVEDP/cardiac output) and in the pulmonary vascular pressure gradient (mean PAP–LVEDP), which was increased by 2 to 4 times. This vasoconstrictor action on the pulmonary vascular bed was by far the most pronounced effect of endotoxin in the acute (0–20 min) phase and was indeed still evident 1–2 h later. The mean pressures at this time were 34 ± 5 mmHg (SPAP) and 22 ± 5 mmHg (DPAP).

Pulmonary hypertension, associated with acute pulmonary vasoconstriction, led to the death of four of the animals from pulmonary oedema. This predominant

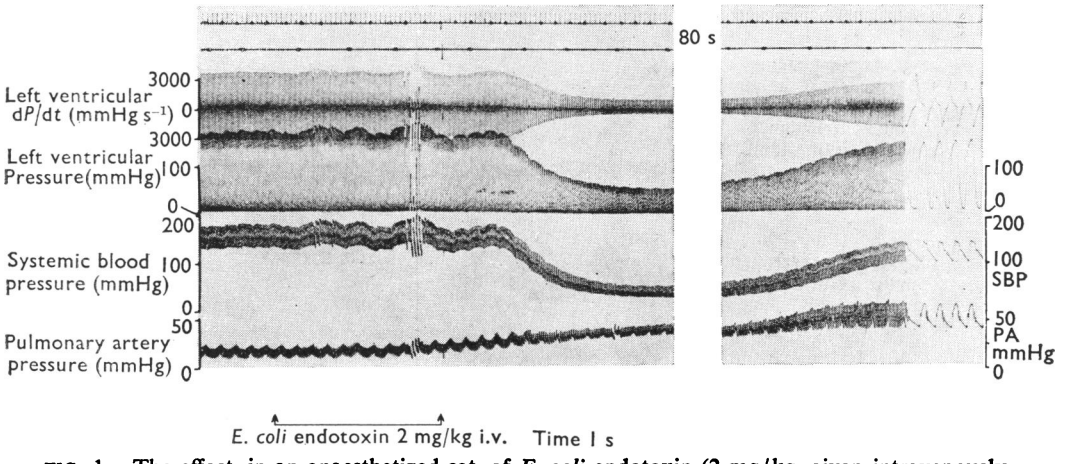

FIG. 1. The effect, in an anaesthetized cat, of *E. coli* endotoxin (2 mg/kg, given intravenously over a period of about 30 s) on (from above) left ventricular (LV) dP/dt, left ventricular pressure, systemic arterial pressure and pulmonary artery pressure. Endotoxin caused a marked increase in pulmonary artery pressure and secondary decreases in LV and systemic arterial pressures and in LV dP/dt. Recovery began within 2–3 min but pulmonary artery pressure remained elevated. Ventricular extrasystoles and bradycardia occurred during, or soon after, the injection. There was an 80 s interval between the two panels. Time 1 s.

effect of endotoxin on the pulmonary vasculature is probably due partly to the release of histamine. The evidence for this from the present study is as follows:

1. Infusions of histamine (5–10 (μg/kg)/min) increased pulmonary artery pressure (three experiments) and decreased systemic arterial pressure and a LV dP/dt max (Fig. 2).

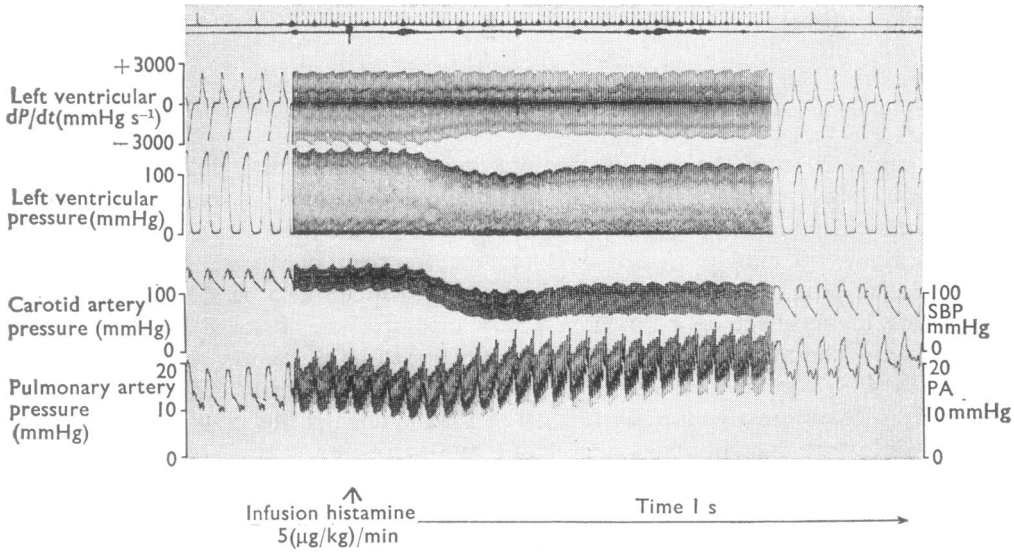

FIG. 2. The effect of an infusion of histamine 5 (μg/kg)/min, commencing at the arrow on (from above) LV dP/dt, LV pressure, carotid artery pressure and pulmonary artery pressure in an anaesthetized cat. Time 1 s.

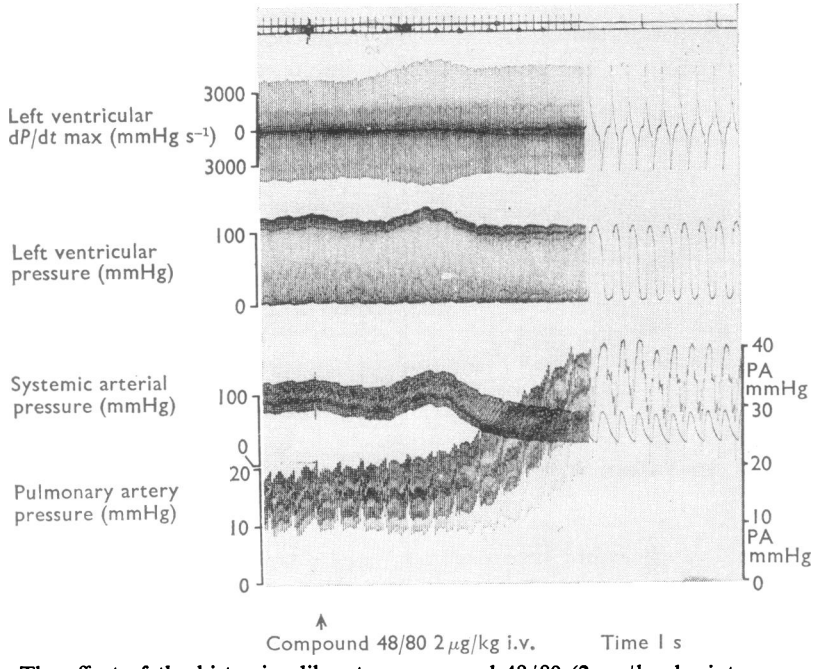

FIG. 3. The effect of the histamine liberator compound 48/80 (2 μg/kg, by intravenous injection) on (from above) LV dP/dt, LV pressure, systemic arterial pressure and pulmonary artery pressure in an anaesthetized cat. Time 1 s.

2. Release of histamine *in vivo* by compound 48/80 (2–5 (μg/kg)/min), also markedly increased pulmonary artery pressure (Fig. 3) and pulmonary vascular resistance.

3. The circulatory effects of endotoxin were much less marked when administered to cats pre-treated with compound 48/80 (2–25 (μg/kg)/min in divided doses, three experiments ; Fig. 4).

It is clear from these results that the right ventricular failure that occurred in some cats after administration of *E. coli* endotoxin was secondary to an increase in the pulmonary vascular pressure gradient rather than to an initial direct myocardial depressant effect. In most of the animals, left ventricular pressure and systemic artery pressure had returned to slightly below control levels within 2–4 min of endotoxin administration (Fig. 1). In some cases LV dP/dt max was increased at this time and local myocardial blood flow was unchanged (myocardial thermal conductivity increment, Δk $(6{\cdot}6 \pm 0{\cdot}9) \times 10^{-4}$ c.g.s. units (i.e. $0{\cdot}00066 \pm 0{\cdot}00009$ calories/cm s °C or $(6{\cdot}6 \pm 0{\cdot}9) \times 10^{-4} \times 418{\cdot}68$ Jm/m²s °C) before endotoxin and $(6{\cdot}3 \pm 0{\cdot}9) \times 10^{-4}$ c.g.s. units, 5 to 10 min later). There was thus no evidence of a primary effect of endotoxin of the myocardial vasculature. This would be expected if the acute effects of endotoxin are indeed primarily mediated through the release of histamine, since this amine dilates the blood vessels of the myocardial microcirculation (Parratt, 1969).

That the effects of endotoxin on LV dP/dt max and on systemic arterial pressure are so transient (Fig. 1) is probably due to a reflex increase in sympathetic discharge. In three experiments, in which endotoxin was administered after β-adrenoceptor blockade with alprenolol (0·5 mg/kg, i.v.), no such recovery in myocardial contractility was observed (Fig. 5) and all three animals died of right ventricular failure 4 to 6 min later.

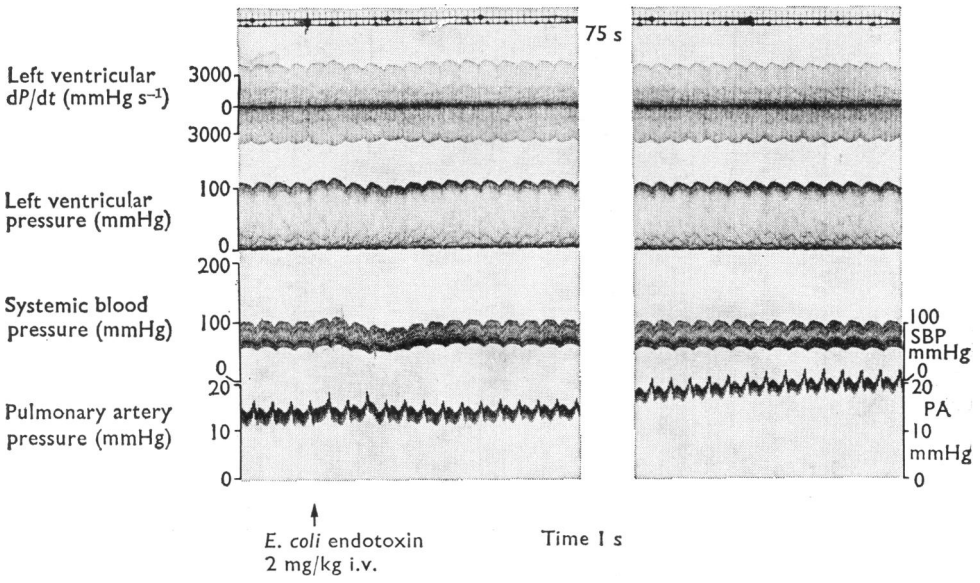

FIG. 4. The effect of an intravenous injection of *E. coli* endotoxin (2 mg/kg) on (from above) LV dP/dt, LV pressure, systemic arterial pressure and pulmonary artery pressure in a cat which had been given repeated injections of compound 48/80. The animal was the same as that in Fig. 3. There was a 75 s interval between the two panels. Time 1 s.

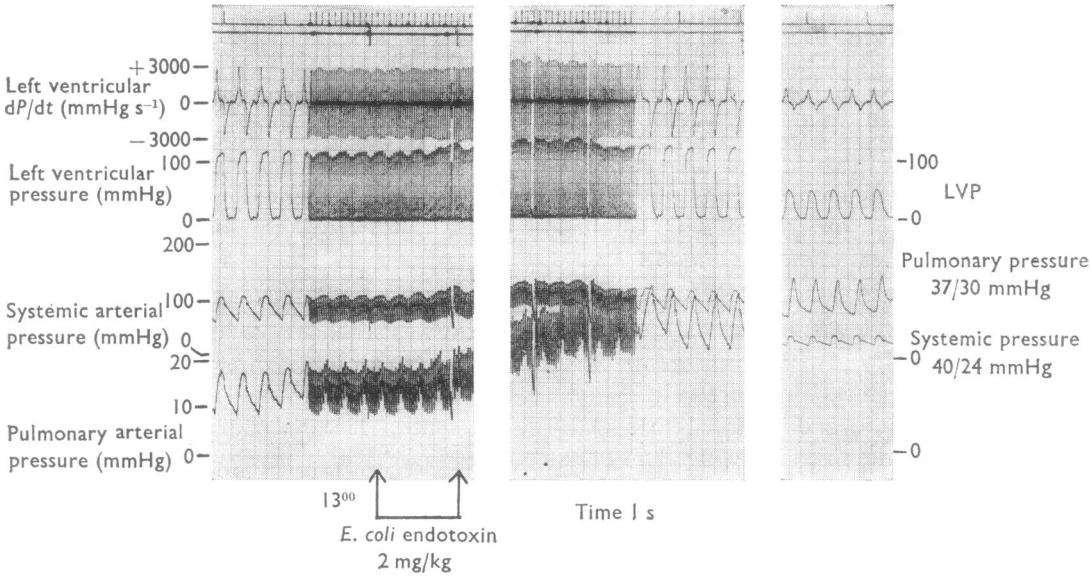

FIG. 5. Haemodynamic effects of *E. coli* endotoxin (2 mg/kg, intravenously) in a cat given alprenolol (0·5 mg/kg) 30 min previously. There was a 20 s interval between the first and second panels and a 48 s interval between the second and third panels. The animal died in right ventricular failure at 13·04. Time 1 s.

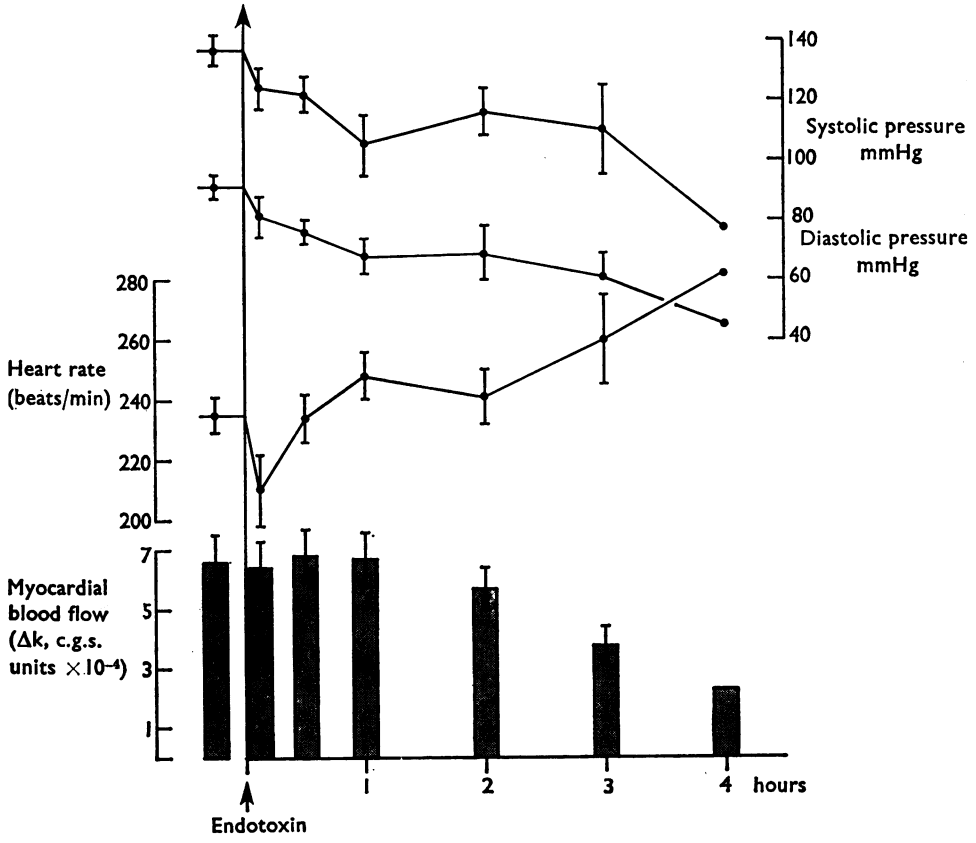

FIG. 6. Systemic and diastolic arterial pressures (mmHg), heart rate (beats/min) and myocardial blood flow (as myocardial thermal conductivity, $\Delta k \times 10^{-4}$ c.g.s. units) before, and at various times after, the intravenous administration of *E. coli* endotoxin (2 mg/kg) in anaesthetized cats. Values are means ± S.E.M.

Delayed effects of E. coli *endotoxin administration*

Over the 2–3 h period following endotoxin injection, systemic arterial pressure tended to decrease, heart rate to increase and myocardial blood flow remained fairly stable (Fig. 6). The main cardiovascular effects at this time (2 to 3 h after endotoxin) are summarized in Table 1. There was a highly significant decrease in stroke volume despite evidence (increased heart rate, LV dP/dt max and myocardial metabolic heat production) of increased sympathetic discharge. External cardiac work was decreased at this time (from $0{\cdot}42 \pm 0{\cdot}2$ kg m min^{-1} before endotoxin to $0{\cdot}29 \pm 0{\cdot}3$ kg m min^{-1}) and peripheral vascular resistance was increased by a mean of 27%. Analysis of arterial and right atrial blood showed (Table 2) that there was a marked metabolic acidosis, the arterial base excess being −20 mEq/litre. In pentobarbitone anaesthetized and ventilated cats before endotoxin, it ranged from −3 to −5 mEq/litre.

In none of the cats was there electrocardiographic evidence of myocardial ischaemia but the P wave was often enlarged. This is indicative of right atrial enlargement, perhaps associated with right ventricular strain.

Most of the cats either died in circulatory failure, or the experiment was terminated 3–4 h after endotoxin administration. Some of the animals were allowed to recover from the shock phase and these received a second dose of endotoxin (2 mg/kg) 3–5 h after the first injection. The 'acute' haemodynamic phase of the endotoxin response (pulmonary vasoconstriction) was considerably less than with the initial administration.

TABLE 1. *Cardiovascular effects of* E. coli *endotoxin in cats (mean*±S.E.M., 12–20 *experiments)*

	Control	2–3 hours after endotoxin
Systolic blood pressure (mmHg)	137±5	113±15‡
Diastolic blood pressure (mmHg)	91±4	65±9†
Right atrial pressure (mmHg)	3·8±0·6	3·7±0·6
Heart rate (beats/min)	235±6	259±15‡
Cardiac output (ml/min)	276±19	208±11†
Stroke volume (ml)	1·20±0·09	0·78±0·05*
Systolic injection time (ms)	98±5	79±5†
Left ventricular dP/dt max (mmHg s^{-1})	4,400±450	5,074±510†
LVEDP (mmHg)	4·8±0·7	4·5±0·8
Myocardial thermal conductivity increment (blood flow Δk, $\times 10^{-4}$, c.g.s. units)	6·6±0·9	5·6±0·7‡
Myocardial metabolic heat production (change; corrected temperature °C)		+0·21±0·08

* $P<0{\cdot}001$; † $P<0{\cdot}01$; ‡ $P<0{\cdot}05$.

TABLE 2. *Arterial and mixed venous pH, pO_2 and pCO_2, in cats ventilated with room air, before, and 2–3 hours after,* E. coli *endotoxin (mean*±S.E.M., 11 *experiments)*

	Control		2–3 hours after endotoxin	
	Arterial†	Right atrial	Arterial	Right atrial
pO_2 (mmHg)	107±3	47±3‡	96±5	41±3
pCO_2 (mmHg)	27±1	34±3	25±3	40±5
pH (units)	7·439±0·02	7·367±0·05	7·163±0·05*	7·084±0·05*

* Significantly different from control, $P<0{\cdot}001$. † Values for spontaneously breathing cats (11 experiments) were pO_2 (mmHg) 105±2, pCO_2 (mmHg) 33±1 and pH 7·388±0·03 units. ‡ Pulmonary artery pO_2 40±2 (mmHg).

Cardiovascular effects of adrenaline before and after endotoxin

The results are summarized in Table 3. Adrenaline increased systemic arterial pressure, heart rate, myocardial blood flow, cardiac output and left ventricular dP/dt by amounts similar to those observed in a previous study using an essentially similar preparation (Parratt & Wadsworth, 1970). All these effects of adrenaline were considerably reduced in the 2–3 h period after endotoxin administration.

TABLE 3. *The cardiovascular effects of adrenaline* (1·0 (μg/kg)/*min by intravenous infusion*) *before, and* 2–3 *hours after,* E. coli *endotoxin* (*change from control, mean*±S E.M., 8–16 *experiments*)

	Before endotoxin	After endotoxin
Systolic blood pressure (mmHg)	+27±4	+15±3†
Diastolic blood pressure (mmHg)	+18±4	+6±2†
Heart rate (beats/min)	+13±4	+4±3‡
Cardiac output (ml/min)	+87±16	+40±16†
Stroke volume (ml)	+0·3±0·06	+0·07±0·15
Left ventricular dP/dt max (mmHg s^{-1})	+2,332±459	+779±416‡
Myocardial thermal conductivity increment (blood flow; Δk, $\times 10^{-4}$, c.g.s. units)	+1·8±0·4 (+27%)	+0·95±0·4 (+17%)

† Significantly different from the change before endotoxin at level of $P<0·01$; ‡ $P<0·05$.

Cardiovascular effects of noradrenaline before and after endotoxin

These effects are summarized in Table 4. Before endotoxin noradrenaline significantly increased systemic arterial pressure, left ventricular pressure, LV dP/dt max and myocardial blood flow. As with adrenaline, the effects on myocardial contractility, as well as the vasoconstrictor effect, were reduced in the endotoxin 'shock' phase (see Fig. 7). In the experiments in which recovery from the delayed effects of endotoxin took place, some recovery of the response to noradrenaline also occurred. This was associated with a general circulatory improvement and a reduced metabolic acidosis. For example, in one of the experiments the noradrenaline induced increase in LV dP/dt max, prior to the administration of endotoxin, was +1,645 mmHg s^{-1} and the arterial pH was 7·513 units. Ninety min after endotoxin, the response was only +551 mmHg s^{-1} (arterial pH, 7·300); 130 min after endotoxin, the increase in LV dP/dt max in response to noradrenaline was +1,400 mmHg s^{-1} (pH 7·263) and 200 min after it was +2,252 mmHg s^{-1}. At this time the arterial pH had recovered to 7·372.

TABLE 4. *The cardiovascular effects of noradrenaline* (1·0 (μg/kg)/*min by intravenous infusion*) *before, and* 2–3 *hours after,* E. coli *endotoxin* (*change from control, mean*±S.E.M., 9–15 *experiments*)

	Before endotoxin	After endotoxin
Systolic blood pressure (mmHg)	+41±4	+18±6*
Diastolic blood pressure (mmHg)	+26±4	+14±3†
Heart rate (beats/min)	−10±7	+1±3
Cardiac output (ml/min)	+19±16	+15±20
Stroke volume (ml)	+0·12±0·06	+0·06±0·06
Left ventricular dP/dt max (mmHg s^{-1})	+2,030±310	+551±242†
Myocardial thermal conductivity increment (blood flow; Δk, $\times 10^{-4}$, c.g.s. units)	+1·17±0·5 (+18%)	+1·08±0·2 (+20%)

* Significantly different from change before endotoxin at level of $P<0·001$; † $P<0·001$.

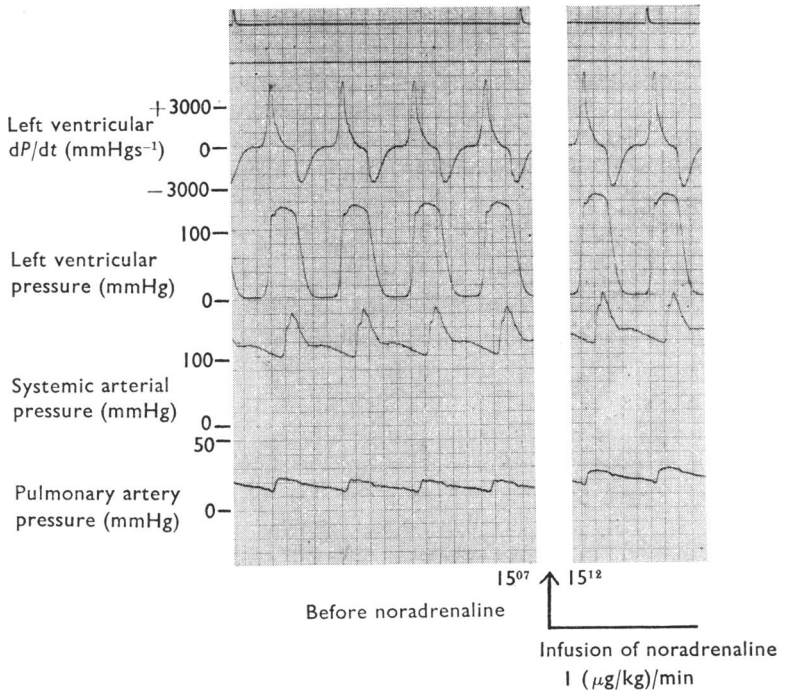

FIG. 7. The effect of an intravenous infusion of noradrenaline (1·0 (μg/kg)/min) 150 min after the intravenous injection of *E. coli* endotoxin. From above LV dP/dt, LV pressure, femoral artery pressure and pulmonary artery pressure. On the left are the control (pre-noradrenaline) values and on the right are the responses towards the end of the 5 min infusion of noradrenaline. There are only slight increases in each cardiovascular parameter.

Discussion

Although there have been a number of haemodynamic studies of the effects of *E. coli* endotoxin in the cat (Kuida, Hinshaw, Gilbert & Visscher, 1958; Kuida, Gilbert, Hinshaw, Brunson & Visscher, 1961; Kadowitz & Yard, 1970; Greenway & Murthy, 1971), usually only a few parameters have been measured. There have been no simultaneous measurements, in an intact preparation, of myocardial contractility, output, heat production, myocardial tissue perfusion and pulmonary artery pressure. Detailed information is also not available in this species of the effects of endotoxin on arterial and mixed venous blood gases. There are clearly two quite distinct effects of *E. coli* endotoxin in the cat. The 'acute' response (pulmonary hypertension) results from a massive (8–12 fold) increase in pulmonary vasoconstriction and leads to pulmonary oedema and right ventricular failure. The 'delayed' or 'shock' response is characterized by an increasingly severe metabolic acidosis and usually leads to death from circulatory failure 3 to 4 h later. Greenway, Lautt & Stark (1969) have shown that it is possible to separate these two responses by subjecting endotoxin to mild alkaline hydrolysis. This abolishes the 'acute' response without modifying the delayed, lethal effect. There is evidence in other species (dog and monkey; for references see Hinshaw, 1964) that endotoxin can release histamine as a result of a complex reaction between endotoxin, platelets and a plasma constituent (Vick, 1960). The present study suggests that histamine release is probably the cause of the initial pulmonary vasoconstriction in the cat. Histamine, whether infused intravenously, or released by compound 48/80, causes pulmonary vasoconstriction, particularly on the venous

side (see de Burgh Daly & Hebb, 1966). The fact that the rise in pulmonary arterial pressure occurred with a delay of about 30 s, that it was largely prevented by pre-treatment with 48/80 and that a second dose of endotoxin (administered 3 to 4 h after the first) had little effect on the pulmonary circulation all suggest a major contribution by endogenously released histamine. That the acute response of endotoxin was reduced by compound 48/80 also suggests that the source of this released histamine in this species is the mast cell and that 5-hydroxytryptamine is not involved to any major extent; mast cells in the cat do not contain significant amounts of 5-hydroxytryptamine (Parratt & West, 1957).

There was a substantial recovery of systemic arterial pressure and left ventricular dP/dt max within 1–3 min of the initial systemic hypotension (see Fig. 1). This recovery was not observed in the three animals pre-treated with the β-adrenoceptor blocking agent alprenolol (see Fig. 5), which all died in right ventricular failure within six minutes of the administration of endotoxin. This suggests that recovery from the acute haemodynamic effects of endotoxin depends upon increased sympathetic discharge. There is direct evidence for this. In baboons, Cavanagh, Rao, Sutton, Bhagat & Bachmann (1970) have shown a 60% increase in blood noradrenaline levels 3 min after endotoxin and in cats, and to a lesser extent in dogs, catecholamine blood levels are very markedly increased within 5 min of endotoxin administration (Hall & Hodge, 1971). This is the result of adrenal medullary stimulation (Nykiel & Glaviano, 1961; Hökfelt, Bygdeman & Sekkenes, 1962). There are probably a number of factors involved in this release of catecholamines. Histamine (which the present experiments have implicated in the acute response) is known to release catecholamine from the adrenal medulla, whilst systemic hypotension, right ventricular strain and, in the later stages, the decrease in blood pH, are probably also involved. Whatever the initial stimulus, circulating catecholamine levels remain high well into the 'delayed' phase of shock. In the experiments of Hall & Hodge (1971) for example, levels were still increasing 90 min after the initial endotoxin injection. This increased sympathetic discharge would account for the elevated heart rate, LV dP/dt max, peripheral vascular resistance and myocardial metabolic heat production observed in the present experiments. It is also probably responsible for the maintained myocardial tissue blood flow. Myocardial contractility is thus maintained during the 'delayed' phase of endotoxin shock as a result of the massive discharge of adrenaline and noradrenaline.

The most pronounced finding during the 2–3 h post-endotoxin period was a severe metabolic acidosis (Table 2). This is presumably due primarily to lactic acid which is elevated, by a factor of 3–5 times, 2–3 h after endotoxin administration in cats (Lucas & Kitzmiller, 1972). Cellular hypoxia, which results in lactic acidosis, is secondary to the reduced perfusion pressure. This decrease in systemic pressure is associated with peripheral vasoconstriction induced by catecholamines. Angiotensin, which is also released in endotoxin shock in the cat (Hall & Hodge, 1971) may also be involved. At the microcirculatory level, both precapillary arterioles and postcapillary venules initially constrict in response to catecholamine stimulation (α-adrenoceptors). This results in 'ischaemic anoxia'. As shock progresses, the precapillary resistance vessels dilate as a result of the release of vasoactive metabolites such as lactic acid. Since the postcapillary (venular) resistance vessels are more resistant to a reduction in pH, fluid leaves the circulation for the tissues (Mellander & Lewis, 1963) and contributes to the decrease in venous return which occurs during endotoxin shock in the cat (Kuida *et al.*, 1961). The predominant sites of

pooling and of fluid loss in the cat have yet to be clearly defined but the fact that pulmonary vascular resistance remained elevated throughout the entire shock period suggests that some circulatory fluid loss occurs in the lungs. Pulmonary oedema was certainly an invariable post-mortem finding in the present study.

The evidence from the present experiments suggests that the increased sympathetic discharge maintains myocardial contractility (increased dP/dt max at the same LVEDP) well into the shock period but that output is reduced because of the lowered venous return. Although there is some disagreement in the literature (see **Introduction**), some recent evidence appears to demonstrate that endotoxin does have a direct myocardial depressant effect (Solis & Downing, 1966; Wyler, Neutze & Rudolph, 1970). *In vivo*, this appears to be balanced by the effect of released catecholamines; if the myocardial effects of catecholamines are prevented by β-adrenoceptor blockade, at least in the early stage of shock, myocardial contractility is not maintained and the animals die in right ventricular failure. The present experiments thus indirectly support the hypothesis that endotoxin does have a myocardial depressant effect. Whether this is direct, or secondary, to the release of a myocardial depressant factor from the ischaemic splanchnic viscera (Wangensteen, Geissinger, Lovett, Glenn & Lefer, 1971) is uncertain.

The release of catecholamines during the delayed, 'shock' phase, though enhancing contractility, has detrimental effects on the peripheral circulation. The reduced tissue perfusion leads to stagnant anoxia and metabolic acidosis. This would indicate that drugs which increase tissue perfusion (providing myocardial contractility is maintained), for example α-adrenoceptor blocking agents, might improve survival in endotoxic shock. There is some evidence that this is indeed so (Longerbeam, Lillehei, Scott & Rosenberg, 1962; Nickerson & Gourzis, 1962; Phillips & Vick, 1971).

Two to three hours after the administration of endotoxin the *in vivo* cardiac and vascular effects of adrenaline and noradrenaline are significantly depressed (Tables 3 and 4). This extends the findings of Cavanagh and his colleagues that atria removed from guinea-pigs (Bhagat, Cavanagh, Merrild, Rana & Rao, 1970) and baboons (Cavanagh *et al.*, 1970) are less responsive to noradrenaline and, in the baboon, also to tyramine. This reduced responsiveness to noradrenaline has now been found to apply also to adrenaline. Furthermore, it has been demonstrated that vascular smooth muscle, as well as cardiac muscle, is less responsive to catecholamines. Effects on systolic and diastolic blood pressure (results of α-adrenoceptor stimulation) and on LV dP/dt max, heart rate and cardiac output (β-adrenoceptor stimulation) are all markedly reduced. The reason for this reduced responsiveness to noradrenaline and adrenaline is not known. Since it occurs *in vitro*, as well as in the intact animal, it is presumably not due to the reduction in blood pH or to desensitization caused by high circulating levels of endogenous catecholamines. One possible explanation is that since endotoxin inhibits ATP-dependent calcium uptake by preparations of cardiac sarcoplasmic reticulum (Hess & Briggs, 1971), ultimately less calcium would be available for the contraction process. If this occurred *in vivo*, it would explain not only depression of cardiac contractility by endotoxin, but also the reduced response to catecholamines since there is evidence (Shinebourne, Hess, White & Hamer, 1969) that the effect of noradrenaline on myocardial contractility is correlated with an *increased* rate of uptake of calcium by the sarcoplasmic reticulum.

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