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ACTH-(1–24) blocks the decompensatory phase of the haemodynamic response to acute hypovolaemia in conscious rabbits

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Abstract

Graded caval occlusion in conscious rabbits caused a biphasic cardiovascular response. Phase I was characterized by a fall in systemic vascular conductance so that arterial pressure was maintained. When cardiac output had fallen to $64 \pm 3\%$ of its baseline level, phase II supervened. During phase II, conductance rose abruptly and arterial pressure fell to a life-threatening level (< 40 mm Hg). Intravenous (i.v.) or central (fourth ventricular) administration of the adrenocorticotrophin (ACTH) fragment ACTH-(1–24) prevented the occurrence of phase II. The central dose of ACTH-(1–24) needed to block the occurrence of phase II was ~ 39 times less than the i.v. dose. Central administration of the δ_1-opioid receptor agonist [D-Pen^2,D-Pen^5]enkephalin (DPDPE) reversed this effect of both central and i.v. ACTH-(1–24). I.v. ACTH-(1–24) also lowered arterial pressure while raising cardiac output and vascular conductance. These effects were insensitive to propranolol and hyoscine methyl bromide, and were not mimicked by cortisol or adrenaline. It is concluded that ACTH-(1–24) has an acute, adrenal-independent, peripheral vasodilator effect as well as a central, anti-shock, effect.

Keywords: Hypovolemia; ACTH-(1–24); Hemorrhagic shock; (Conscious rabbit)

1. Introduction

The cardiovascular response to acute haemorrhage in unanaesthetized rabbits, as in all mammals, consists of two phases. In phase I, systemic vascular conductance falls pari passu with blood volume and cardiac output, so that arterial pressure is well maintained (Schadt et al., 1984; Ludbrook and Rutter, 1988). This compensatory vasoconstriction is chiefly attributable to the action of the arterial baroreceptor reflex (Ludbrook and Graham, 1984; Schadt and Gaddis, 1986). If acute blood loss exceeds about 30% of blood volume phase II commences abruptly. In phase II the compensatory vasoconstriction fails and blood pressure falls steeply (Schadt et al., 1984; Ludbrook and Rutter, 1988). A similar, biphasic response occurs when acute central hypovolaemia is produced by graded inflation of a cuff on the inferior vena cava (Evans et al., 1989a,b; Ludbrook et al., 1988). In rabbits, phase II is associated with a dramatic fall in sympathetic vasoconstrictor drive (Burke and Dorward, 1988), and is dependent on a brainstem (δ-opioid receptor mechanism (Evans et al., 1989a,b; Ludbrook and Ventura, 1994a).

Intravenous (i.v.) injection of adrenocorticotrophin (ACTH) fragments induces a potent and sustained reversal of otherwise fatal haemorrhagic hypotension in rats and dogs (for review see Bertolini et al., 1989). This effect is adrenal-independent, since many of the ACTH fragments used possess little or no adrenocorticotrophic activity, and the anti-shock action of ACTH-(1–24) has been reported in adrenalectomized rats (Bertolini et al., 1986a, 1989). There is also a suggestion that ACTH-(1–24) can restore blood pressure in haemorrhagic shock in humans (Bertolini et al., 1987; Noera et al., 1989, 1991; Pinelli et al., 1989).

In conscious rats, i.v. MSH/ACTH fragments can have acute cardioaccelerator and pressor effects (Gruber et al., 1984; Klein et al., 1985; Callahan et al., 1988; De Wildt et al., 1993). These appear to be mediated by catecholamines since they can be attenuated by α- and β-adrenoceptor antagonists (Gruber et

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al., 1984). At least some of this released catecholamine appears to come from sympathetic nerves since the effects of γ_2-melanocyte stimulating hormone (γ_2-MSH) can be attenuated by bretylium which prevents nerve terminal release of noradrenaline (Callahan et al., 1988). This action of ACTH is a peripheral one, since intracerebroventricular injection of up to 4 μg of ACTH in conscious rats has no observable effect on either heart rate or blood pressure. In contrast to these studies on rats, i.v. administration of ACTH-(1–24) decreased arterial pressure and increased heart rate and sympathetic nerve activity in pithed and anaesthetised rabbits (Szabo et al., 1989). Similar observations have also been reported in conscious and anaesthetised rats (De Wildt et al., 1994). In unanaesthetized lambs, α-MSH or ACTH-(4–10) decreased blood pressure and raised heart rate and cardiac output (Llanos et al., 1983). In conscious sheep, i.v. ACTH-(1–24) raises cardiac output, followed by a rise in heart rate and then blood pressure (Graham et al., 1980; Spence et al., 1989). This rise in blood pressure may be centrally mediated, since intracerebroventricular injection of ACTH-(1–24) caused a similar rise in blood pressure (Scoggins et al., 1984).

Circulating levels of ACTH rise during or soon after haemorrhage in conscious rats (Darlington et al., 1986; Grässler et al., 1990), anaesthetized cats (Bereiter et al., 1983, 1984, 1986), anaesthetized and conscious dogs (Lilly et al., 1983, 1986; Wood et al., 1982), conscious pigs (O'Benar et al., 1987), and fetal sheep (Wood et al., 1989). This ACTH release appears to occur mainly in the hypotensive phase II of haemorrhage. Since there is suggestive evidence that the amount of ACTH released is directly proportional to the fall in arterial pressure during haemorrhage (Darlington et al., 1986; Bereiter et al., 1984, 1986), ACTH release may be important in recovery from acute blood loss.

The experiments described in this paper were designed to investigate whether ACTH-(1–24) can prevent the decompensatory phase of acute hypovolaemia in conscious rabbits by a central or a peripheral action, whether this is a specific action of ACTH, and whether it acts through a brainstem δ-opioid receptor mechanism. A secondary aim of the experiments was to investigate how i.v. ACTH-(1–24) causes the acute haemodynamic changes that have been observed. A preliminary report of these experiments has recently been presented (Ludbrook and Ventura, 1994b).

2. Materials and methods

Nine New Zealand White rabbits were used, weighing 2.13–3.59 kg (mean 2.85 kg). The experiments were done in accordance with the Australian Code of Practice for the Care and Use of Animals for Scientific Purposes (1990), and were approved in advance by the Animal Ethics Committee of the Royal Melbourne Hospital.

2.1. Surgical procedures

Major procedures

These were performed under halothane anaesthesia after induction with i.v. thiopentone sodium (25 mg/kg) and endotracheal intubation. At a first operation, an inflatable cuff was placed around the thoracic inferior vena cava (caval cuff). Caval cuffs are hand made in our laboratory from plastic tubing, and in our experience remain operational when chronically implanted for at least 6 months. Two weeks later, an ultrasonic (transit time) flow probe (Transonic Systems, Ithaca, NY, USA; type 6S) was placed extrapericardially around the ascending aorta. A 0.3 mm o.d. polyvinyl chloride tube (Dural Plastics SV10) was introduced 2 weeks later through the atlanto-occipital ligament so that its tip lay in the fourth ventricle. Its dead space of 18 μl was filled with 154 mM NaCl.

Minor procedures on study days

These were done under local anaesthesia with 0.5% lignocaine HCl. The rabbit was placed in a $15 \times 17 \times 40$ cm box fitted with a wire mesh lid, 180 min before the beginning of the study. The tubes leading to the caval cuff and fourth ventricular catheter (when used), and the connecting plug for the flow probe, were retrieved from their subcutaneous positions. A catheter was inserted into a central ear artery and advanced to the root of the ear for measuring arterial pressure. For i.v. drug administration a catheter was inserted into a marginal ear vein.

2.2. Haemodynamic variables

Arterial pressure was measured by connecting the ear artery catheter to a Stratham P23Dc transducer which was placed at the level of the heart 50 mm above the floor of the rabbit's box. The flow probe was connected to a flowmeter (Transonic Systems, Ithaca, NY, USA; model T206) to measure ascending aortic flow (cardiac output). Heart rate was measured by a tachometer that was actuated by the flow pulse.

The signals were amplified and recorded on a Grass model 7 Polygraph, and sent to an Olivetti M24 computer equipped with an A-D converter which provided 10 s mean values for arterial pressure (mm Hg), heart rate (beats/min) and cardiac output (ml/min). The computer also calculated 10 s means for cardiac index (cardiac output/body weight in kg) and systemic vascular conductance index (100 × cardiac index/mean arterial pressure).

2.3. Graded central hypovolaemia

The caval cuff was gradually inflated by a micrometer-driven syringe so that cardiac index fell at a constant rate of 8.5 ± 0.1% of its baseline level per minute. This corresponds approximately to blood loss at a rate of 7% of blood volume per minute (Ludbrook et al., 1988). The caval cuff was deflated when mean arterial pressure had fallen to ≤ 40 mm Hg, or when cardiac index had fallen to ~ 34% of its baseline level, whichever occurred first.

2.4. Drugs

The drugs used were: the adrenoceptor agonist adrenaline (Astra), the δ_1-opioid receptor agonist [D-Pen^2,D-Pen^5]enkephalin (DPDPE) (Sigma), hydrocortisone sodium succinate (cortisol) (Solu-Cortef, Upjohn), the β-adrenoceptor agonist isoprenaline hydrochloride (Isuprel, Winthrop), the β-adrenoceptor antagonist *l*-propranolol hydrochloride (ICI), the muscarinic receptor antagonist (−)-scopolamine methyl bromide (hyoscine methyl bromide) (Sigma), sodium nitroprusside dihydrate (David Bull) and tetracosactrin (ACTH-(1–24)) (Synacthen, Ciba-Geigy). The drugs were dissolved and diluted to the required concentrations in sterile 154 mM NaCl.

2.5. Experimental protocol

Nine different experimental protocols were performed. Only one protocol was performed on one rabbit on any given day, and rabbits were studied at intervals of 2–7 days. During each study caval cuff inflation was repeated up to 6 times at intervals of 90 min. Loading doses of drugs were injected into the fourth ventricle in a volume of 15 μl over 1 min, 10 min before the commencement of caval cuff inflation. This was followed by a slow infusion of 0.75 μl/min until the caval cuff was deflated. In our extensive experience of fourth ventricular administration of drugs we have never observed any effect of administering vehicle at the same rate and volume as in this regimen.

Similarly, i.v. administered drugs were given as a loading dose in 0.2–0.3 ml over 1 min, 10 min before the commencement of caval cuff inflation, followed by slow infusion at 5% of the loading volume per minute. The first caval cuff inflation in each study was performed 90 min after either fourth ventricular or i.v. administration of saline vehicle.

Critical doses of ACTH-(1–24)

The aim was to determine the critical (threshold) doses of ACTH-(1–24) which prevented circulatory decompensation during caval cuff inflation, when administered either i.v. (protocol 1, nine rabbits) or into the fourth ventricle (protocol 2, nine rabbits). The first caval cuff inflation on each day was performed after fourth ventricular or i.v. administration of saline. Caval cuff inflations were then repeated at 90 min intervals after doses of fourth ventricular ACTH-(1–24) (dose range: 10 pmol–1 nmol) or i.v. ACTH-(1–24) (dose range: 0.1–3.4 nmol/kg) ascending in half-logarithmic units until a critical dose was reached. The critical dose was taken as the dose at which phase II of the cardiovascular response to caval cuff inflation did not occur. These two protocols were the first two studies conducted on each rabbit and were performed in a randomized order.

Effects of fourth ventricular DPDPE

In these studies the effect of fourth ventricular saline administration on the response to caval cuff inflation was first tested, then 90 min later the effect of fourth ventricular administration of DPDPE (50 nmol) on the response to caval cuff inflation was tested. After a further 90 min the effects of a combination of both the critical dose of fourth ventricular ACTH-(1–24) and DPDPE (50 nmol) were tested on the response to caval cuff inflation (protocol 3, four rabbits). Protocol 4 (two rabbits) followed a similar procedure except that prior to the third caval cuff inflation the critical dose of i.v. ACTH-(1–24) was administered in combination with the fourth ventricular DPDPE (50 nmol).

Effects of i.v. propranolol and i.v. hyoscine methyl bromide

The first caval cuff inflation was performed following i.v. administration of saline. 90 min later a second caval cuff inflation was performed following i.v. infusion of either propranolol (1 μmol/kg) (protocol 5, four rabbits) or a combination of propranolol (1 μmol/kg) and hyoscine methyl bromide (0.13 μmol/kg) (protocol 6, four rabbits). These doses of propranolol and hyoscine methyl bromide when administered i.v. to conscious rabbits have been shown to block peripheral β-adrenoceptors and muscarinic receptors, respectively (Lew et al., 1987). After a further 90 min, the same doses of i.v. propranolol and/or hyoscine methyl bromide were again administered along with the i.v. critical dose of ACTH-(1–24) followed 10 min later by caval cuff inflation.

Effects of i.v. cortisol, adrenaline or sodium nitroprusside

The first caval cuff inflation in each protocol was performed after i.v. administration of saline. Caval cuff inflation was then repeated at 90 min intervals after doses of cortisol (dose range: 0.1–100 μmol/kg) (protocol 7, two rabbits), adrenaline (dose range: 0.05–50 nmol/kg/min) (protocol 8, two rabbits) or sodium nitroprusside (dose range: 1–100 nmol/kg/min) (protocol 9, two rabbits), ascending in logarithmic units.

I.v. doses of adrenaline or sodium nitroprusside were given by constant infusion, because of the transient nature of the responses elicited by bolus doses.

All rabbits underwent protocols 1 and 2 in a randomized order, before undergoing from one to seven of the other protocols.

2.6. Analysis of results

The baseline levels of the haemodynamic variables before and after each dose of ACTH-(1–24) were compared and contrasted by repeated measures analysis of variance (ANOVA), using the Greenhouse-Geisser $\hat{\epsilon}$ adjustment to correct for multisample asphericity (Ludbrook, 1994). The within-rabbit interaction was used to test for differences in the pattern of response. Haemodynamic variables during caval cuff inflation were compared by ANOVA, the Dunn-Šidák correction being applied if multiple contrasts were made (Ludbrook, 1991). Levels of haemodynamic variables are tabulated as between-rabbit means ± 1 S.E.

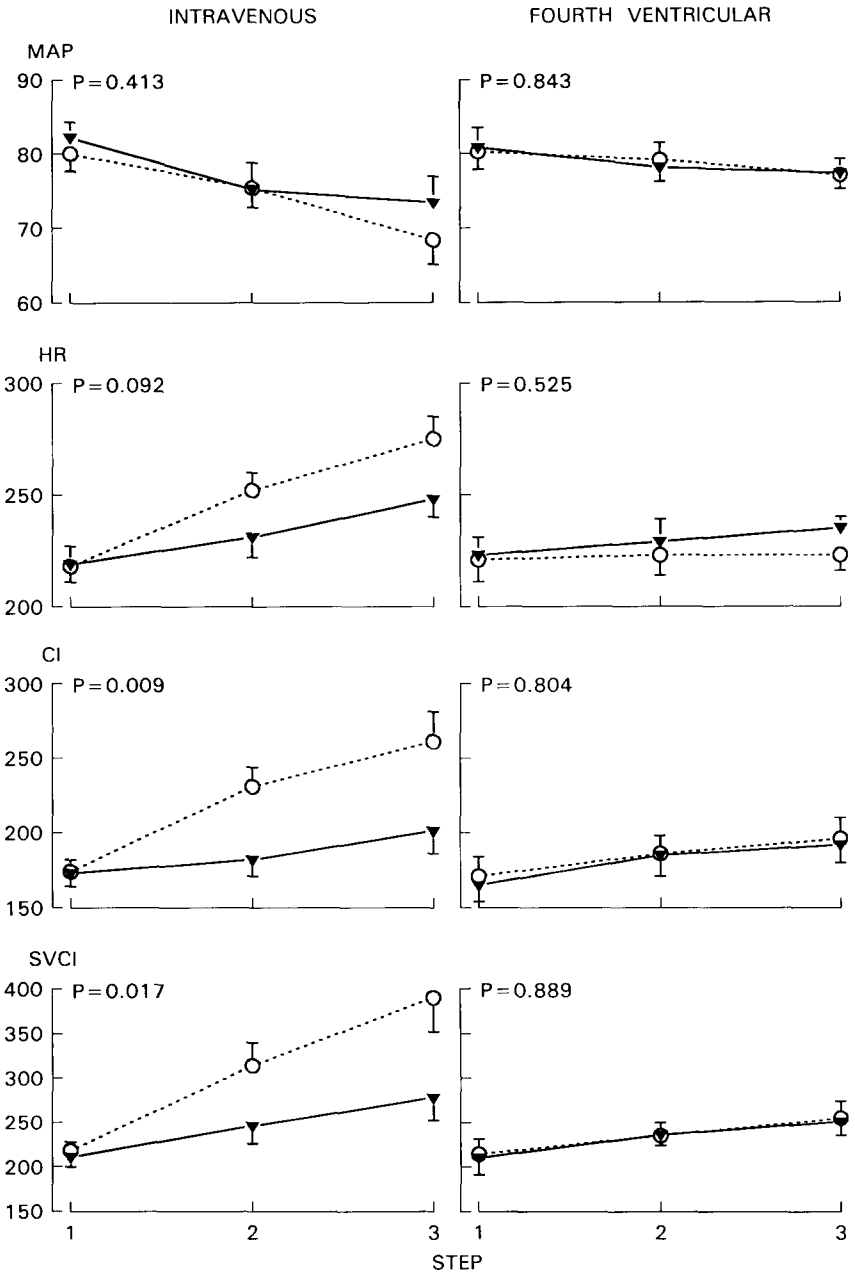

Fig. 1. Baseline haemodynamic variables before (▼) and after (○) fourth ventricular or i.v. administration of saline (step 1), the sub-critical dose of ACTH-(1–24) (step 2) or the critical dose of ACTH-(1–24) (step 3). The fourth ventricular critical dose was 0.17 nmol (range: 0.1–1), and the i.v. critical dose was 6.6 nmol (range: 2.2–9.8). MAP = mean arterial pressure (mm Hg), HR = heart rate (beats/min), CI = cardiac index (ml/min/kg) and SVCI = systemic vascular conductance index (100 × CI/MAP). *P* values are the difference in profile of the dose-response relation according to the route of administration (repeated measures ANOVA).

The critical doses of the drugs were logarithmically transformed to calculate between-rabbit geometric means with ranges in parentheses, on the assumption that the dose-response relationship is distributed log-normally.

Carry-over effects of fourth ventricular and i.v. administered ACTH-(1–24) from previous experimental protocols were analysed by one-way ANOVA.

3. Results

3.1. Effects of i.v. saline and ACTH-(1–24)

Resting haemodynamic variables

Before and after i.v. administration of saline, the levels of the haemodynamic variables in all nine rabbits were within the normal range for our laboratory (Fig.

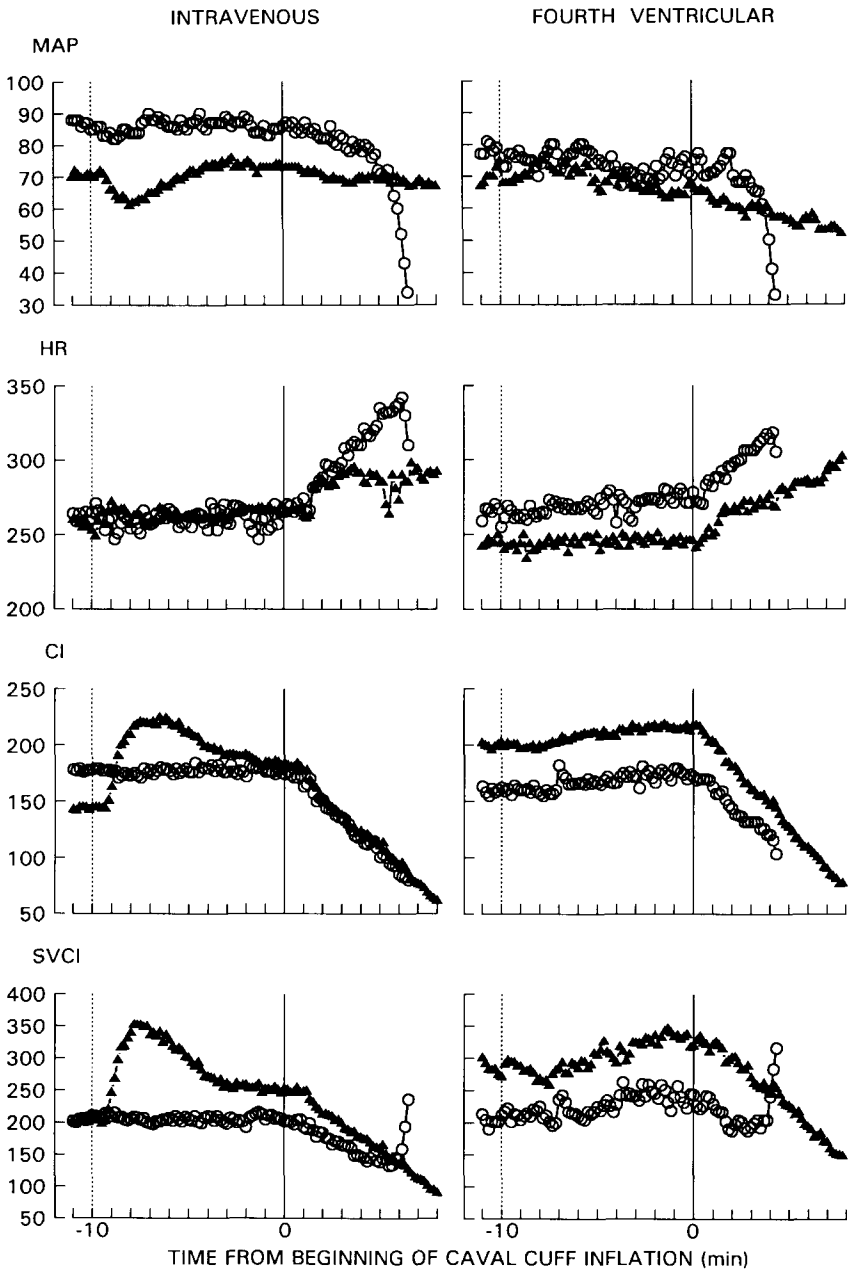

Fig. 2. Haemodynamic changes during graded caval cuff inflation after fourth ventricular or i.v. infusion of saline vehicle (○) or the critical dose of ACTH-(1–24) (▲) in one rabbit. In all nine rabbits the mean critical dose for fourth ventricular administration was 0.1 nmol (range: 0.1–1) and for i.v. administration was 6.6 nmol (range: 2.2–9.8). Symbols represent the mean estimate over 10 s. Bolus infusions were made 10 min before the beginning of caval cuff inflation which was commenced at time = 0. Abbreviated haemodynamic variables are the same as in Fig. 1.

1) (Evans et al., 1989a,b; Ludbrook and Ventura, 1994a). Rabbits which had undergone the fourth ventricular ACTH-(1–24) protocol 2–3 days previously showed no signs of carry-over effects in haemodynamic baseline levels, body weight or haematocrit ($P \geq 0.28$, $n = 4$). ACTH-(1–24) i.v. raised cardiac index and systemic vascular conductance in a dose-dependent fashion (Figs. 1 and 2), lowered mean arterial pressure and raised heart rate (Fig. 2). However, the mean arterial pressure and heart rate responses were only transient and these variables had returned to baseline levels by the commencement of the caval cuff inflation (see Figs. 1 and 2).

Haemodynamic responses to caval cuff inflation

After saline infusion the haemodynamic response to caval cuff inflation was biphasic (Fig. 2). During phase I, systemic vascular conductance fell steadily so that mean arterial pressure fell only slightly. Heart rate rose steadily during phase I. Phase II began when cardiac index had fallen to 63.6 ± 3.4% of its resting level, at which point systemic vascular conductance rose abruptly and mean arterial pressure fell precipitately (Fig. 2). Rabbits which had undergone the fourth ventricular ACTH-(1–24) protocol 2–3 days previously had no differences in the rates of change of haemodynamic variables during caval cuff inflation nor in the timing of the onset of phase II of the response to caval cuff inflation ($P \geq 0.56$, $n = 4$). The haemodynamic response to caval cuff inflation was not affected by sub-critical doses of i.v. ACTH-(1–24). Even though during infusion of sub-critical doses of i.v. ACTH-(1–24) phase II occurred at an earlier absolute value of cardiac index (see Table 1), the level of cardiac index at the commencement of the phase II was not consistently different from that during saline infusion when expressed as a percentage of the baseline level (see Table 1).

Higher doses of i.v. ACTH abolished the decompensatory phase II. That is, throughout the caval cuff inflation there was a steady fall in systemic vascular conductance and rise in heart rate, with only a small fall in mean arterial pressure (Fig. 2). The critical dose of i.v. ACTH-(1–24), needed to abolish phase II, was 2.6 nmol/kg (range: 1.0–3.4). This translates to a mean total dose of 6.6 nmol (range: 2.2–9.8).

3.2. Effects of fourth ventricular saline and ACTH-(1–24)

Resting haemodynamic variables

Before and after fourth ventricular infusion of saline all baseline haemodynamic variables were within the normal range for our laboratory in all nine rabbits (Fig. 1). Rabbits which had undergone the i.v. ACTH-(1–24) protocol 2–3 days previously showed no signs of carry-over effects in any of the baseline haemodynamic variables, body weight or haematocrit ($P \geq 0.55$, $n = 5$). Fourth ventricular infusion of ACTH-(1–24) did not affect any of the haemodynamic variables (see Fig. 1).

Haemodynamic responses to caval cuff inflation

After fourth ventricular saline infusion these were biphasic (see Fig. 2). Rabbits which had undergone the fourth ventricular ACTH-(1–24) protocol 2–3 days previously showed no change in the rates of change of any haemodynamic variables during caval cuff inflation nor in the time of onset of phase II of the response to caval cuff inflation ($P \geq 0.32$, $n = 5$). The haemodynamic responses to caval cuff inflation were unaffected by sub-critical doses of ACTH-(1–24) (Table 1). Higher doses of i.v. ACTH-(1–24) abolished the decompensatory phase II. That is, throughout the caval cuff inflation there was a steady fall in systemic vascular conductance and rise in heart rate, with only a small fall in mean arterial pressure (Fig. 2). The mean critical dose of fourth ventricular ACTH-(1–24), needed to abolish the onset of phase II of the response to caval cuff inflation, was 170 pmol (range: 100–1000). This was 39 times less (range: 7–95) than the critical i.v. dose.

3.3. Effects of DPDPE

In five rabbits, fourth ventricular administration of DPDPE (50 nmol) raised mean arterial pressure from 77 ± 2 to 98 ± 3 mm Hg ($P = 0.014$). Heart rate also rose, from 203 ± 8 to 267 ± 11 beats/min ($P = 0.005$). Cardiac index and systemic vascular conductance were not consistently affected ($P \geq 0.314$).

Fourth ventricular administration of DPDPE (50 nmol) did not affect the response to caval cuff inflation, but reversed the phase II-blocking effects of critical doses of both fourth ventricular ($n = 3$) and i.v. ($n = 2$) ACTH-(1–24).

3.4. Effects of propranolol and hyoscine methyl bromide

Resting haemodynamic variables

Propranolol i.v. (1 μmol/kg) in four rabbits had no consistent effect on either resting cardiac index, mean arterial pressure or systemic vascular conductance index ($P = 0.99$). Heart rate was reduced transiently. Simultaneous infusion of both propranolol (1 μmol/kg) and hyoscine methyl bromide (0.1 μmol/kg) caused an inconsistent rise in heart rate from 201 ± 19 to 234 ± 9 beats/min ($P = 0.131$, $n = 4$), but again had no effect on either resting cardiac index, mean arterial pressure or systemic vascular conductance index ($P \geq 0.110$, $n = 4$).

Blockade of β-adrenoceptors by i.v. propranolol (1 μmol/kg) was confirmed by the abolition of the in-

crease in heart rate and decrease in mean arterial pressure caused by the administration of a bolus dose of i.v. isoprenaline (4 nmol/kg).

Effects on haemodynamic effects of ACTH-(1–24)

The heart rate response to i.v. ACTH-(1–24) infusion was reduced but not abolished by i.v. propranolol infusion. The i.v. ACTH-(1–24)-induced increases in cardiac index and systemic vascular conductance and the fall in mean arterial pressure were unaffected by i.v. propranolol infusion. I.v. infusion of propranolol plus hyoscine methyl bromide abolished the heart rate response to i.v. ACTH-(1–24) but did not affect the ACTH-(1–24)-induced changes in cardiac index, systemic vascular conductance and mean arterial pressure.

Effects on responses to caval cuff inflation

Responses of cardiac index, mean arterial pressure and systemic vascular conductance to caval cuff inflation during both i.v. saline and i.v. ACTH-(1–24) infusion were unaffected by i.v. propranolol (1 μmol/kg) but the heart rate response was attenuated. The rise in heart rate during caval cuff inflation was abolished by i.v. infusion of a combination of propranolol (1 μmol/kg) and hyoscine methyl bromide (0.1 μmol/kg).

I.v. infusion of either propranolol (1 μmol/kg), hyoscine methyl bromide (0.1 μmol/kg), or both did not affect the onset of phase II during control caval cuff inflations, nor the phase II blocking effect of i.v. ACTH-(1–24).

3.5. Effects of adrenaline and sodium nitroprusside

I.v. infusion of adrenaline ($n = 2$) increased mean arterial pressure, heart rate and cardiac index, without having a consistent effect on systemic vascular conductance. Continuous infusion of adrenaline at doses up to 50 nmol/kg/min did not abolish the onset of the decompensatory phase II of the response to caval cuff inflation.

In contrast to adrenaline, sodium nitroprusside ($n = 2$) had similar effects to ACTH when infused i.v. Sodium nitroprusside lowered mean arterial pressure while increasing heart rate, cardiac index and systemic vascular conductance. Sodium nitroprusside when infused at rates of up to 100 nmol/kg/min did not prevent the onset of phase II.

3.6. Effects of cortisol

In two rabbits cortisol was infused i.v. as bolus doses of up to 100 μmol/kg. It had no effect on any of the resting haemodynamic variables, nor on the haemodynamic response to caval cuff inflation.

4. Discussion

Our experiments have resulted in two main findings. The first is that ACTH-(1–24) given i.v. has marked, dose-dependent, haemodynamic effects in conscious rabbits, but none when it is given centrally. The second is that ACTH-(1–24) can act within the brainstem to prevent the decompensatory phase II of acute central hypovolaemia.

4.1. Direct haemodynamic effects of i.v. ACTH-(1–24)

We have confirmed the blood pressure-lowering effects of i.v. ACTH-(1–24) reported in anaesthetized and pithed rabbits (Szabo et al., 1989) and in conscious and anaesthetized rats (De Wildt et al., 1994). Furthermore, we have shown that the fall in blood pressure is the net result of systemic vasodilatation and increased cardiac output (Fig. 1).

It is likely that the primary action of i.v. ACTH-(1–24) was as a direct vasodilator, as suggested by Szabo et al. (1989), and that the rise in cardiac output and heart rate were secondary effects. Support for a direct, rather than adrenally-mediated, vasodilator action of ACTH-(1–24) is given by our findings that the rise in systemic vascular conductance was unaffected by propranolol, adrenaline had a pressor rather than depressor effect, and i.v. cortisol had no haemodynamic effects. The rise in cardiac output was not dependent on cardioacceleration, since it was unaffected by propranolol and hyoscine methyl bromide. It most likely reflects the sensitivity of the rabbit left ventricle to afterload (Faris et al., 1981), as indicated by the cardiac output-raising effect of sodium nitroprusside. The cardioacceleration that we observed, and the increase in sympathetic drive reported by Szabo et al. (1989), are explicable as a baroreflex response to the fall in blood pressure.

4.2. Central effects of ACTH-(1–24)

When injected into the fourth ventricle, ACTH-(1–24) had no discernible effects on the baseline levels of the haemodynamic variables, but at a critical dose it prevented the occurrence of the decompensatory phase II during caval cuff inflation (Fig. 2; Table 1). The critical dose was 39 times less than that which had the same effect when given i.v., so this seems to be a genuine central effect of ACTH-(1–24). Its site of action is likely to have been the brainstem, because when dye is injected into the fourth ventricle at the same rate as in our protocol it stains only the surface of the pons and medulla and the cerebellar vermis (Evans et al., 1992). The lipophobic properties of ACTH make it unlikely that it penetrated deeply into the brainstem, but groups of neurones which play an important role in cardiovascular control are located

Table 1
Cardiac index at time of onset of phase II of caval cuff inflation

	V4 ACTH-(1–24)	i.v. ACTH-(1–24)
Saline vehicle		
ABSCI [a]	114 ± 17	112 ± 10
PERCI [b]	64.9 ± 4.6	63.6 ± 3.4
Sub-critical dose		
Dose range (nmol)	0.03 – 0.3	0.7 – 3
ABSCI [a]	114 ± 15	137 ± 12 [c]
PERCI [b]	60.4 ± 4.8	59.2 ± 3.4
Critical dose		
Dose range (nmol)	0.1 – 1	2 – 10
ABSCI [a]	64 ± 5 [cc]	90 ± 8
PERCI [b]	32.8 ± 0.8 [cc]	34.1 ± 0.6 [cc]

[a] ABSCI = absolute cardiac index at time of onset of phase II (ml/min/kg), [b] PERCI = percentage of baseline cardiac index at time of onset of phase II. Pairwise contrasts between ACTH-(1–24) and the vehicle were made by ANOVA applying the Dunn-Šidák correction for multiple contrasts. [c] $P < 0.05$, [cc] $P < 0.01$. The critical dose was defined as the minimum dose which blocked the onset of phase II of the response to caval cuff inflation. The sub-critical dose was the highest dose tested prior to the critical dose in which phase II still occurred. Values are the means ± S.E.M. from nine experiments.

relatively superficially in the nucleus tractus solitarius and ventrolateral medulla (Dampney, 1994). The lack of effect of ACTH-(1–24) on baseline levels of the haemodynamic variables suggests that the inhibitory pathways within which it acts are not tonically active.

The effect of i.v. ACTH-(1–24) in preventing the decompensatory phase II appears to be a specific one and independent of its peripheral vasodilator action since sodium nitroprusside, which caused similar haemodynamic changes, did not affect the response to caval cuff inflation. We cannot be certain that this effect of i.v. ACTH-(1–24) is mediated entirely by a brainstem mechanism, though it seems likely. The best evidence we have is that DPDPE abolished its protective effect, because a δ-opioid receptor mechanism in the brainstem is an essential element in the decompensatory phase II (Evans et al., 1989b). This interaction between ACTH-(1–24) and DPDPE is consistent with the proposal by Bertolini et al. (1986b) that there is an ACTH-opioid balance which is upset by haemorrhage. Neither can we be sure where i.v. ACTH-(1–24) crosses the blood-brain barrier, though the area postrema is a strong candidate because its neurones project to the nucleus tractus solitarius and rostral ventrolateral medulla (Dampney, 1994).

4.3. Implications of our findings

The acute vasodepressor actions of i.v. administered ACTH-(1–24) may be mediated by an ACTH-like receptor, possibly the previously described melanocortin-2 receptor (Gantz et al., 1993). It appears that other types of melanocortin receptor are not involved in this effect since these receptors tend to mediate vasoconstrictor actions (De Wildt et al., 1994). The central anti-shock effect of ACTH-(1–24) may be mediated by an ACTH receptor or one of the several other melanocortin receptors which are known to be located centrally (Tatro and Entwistle, 1994).

It seems that ACTH should be added to the list of peptides, such as arginine vasopressin and angiotensin II, that can modulate cardiovascular functions by crossing the blood-brain barrier at privileged sites and the area postrema in particular (Dampney, 1994). The fact that ACTH, like vasopressin, enters the bloodstream mainly in the decompensatory hypotensive phase II of acute hypovolaemia (see Introduction) suggests that it may contribute to the restoration of blood pressure by a central action. However, it is not yet established that the plasma concentration of ACTH in acute hypovolaemia can reach similar levels to those resulting from exogenous infusion in experiments such as ours. It is also still a matter for speculation whether ACTH-(1–24) may prove useful as an adjunct to blood volume replacement in clinical hypovolaemia, though its combined but opposite central and peripheral actions make this an attractive idea. The virtual lack of acute toxicity, and other clinical data (Bertolini et al., 1987; Noera et al., 1989,1991; Pinelli et al., 1989) make the idea of first-aid use of ACTH-(1–24) even more attractive.

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