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Effect of the Selective A_1 Adenosine Antagonist 8-Cyclopentyl-1,3-dipropylxanthine on Acute Renal Dysfunction Induced by *Escherichia coli* Endotoxin in Rats

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Abstract—The effect of the selective A_1 adenosine antagonist, 8-cyclopentyl-1,3-dipropylxanthine (CPX), on *Escherichia coli* endotoxin-induced acute renal dysfunction was determined in anaesthetized rats. Bolus administration of endotoxin at doses of either 1 or 20 mg kg^{-1} evoked decreases in inulin clearance, renal blood flow, urine flow and excretion of sodium, potassium and chloride. The changes in renal function produced by 20 mg kg^{-1} endotoxin were more severe than those noted with 1 mg kg^{-1} toxin and, by contrast to this lower dose, renal function showed no signs of recovery. Intravenous administration of CPX (0·1 mg kg^{-1}) elicited a statistically significant, although modest, attenuation of the decline in inulin clearance, renal blood flow, urine output and electrolyte excretion induced by 20 mg kg^{-1} endotoxin. By contrast, treatment with 0·1 mg kg^{-1} CPX resulted in statistically significant protection against the falls in excretory function evoked by 1 mg kg^{-1} endotoxin but not against the reductions in renal blood flow and inulin clearance produced by the lower dose of toxin. These results suggest that adenosine may play a role, albeit not a major one, in the pathophysiology of endotoxaemic acute renal failure.

Acute renal failure (ARF) is a common and serious complication of Gram-negative sepsis, and this renal complication is associated with mortality rates in excess of 60% (Turney et al 1990). Many of the manifestations of Gram-negative sepsis appear to be due to release of endotoxin from the bacterial cell wall into the systemic circulation (Morrison & Ryan 1987). Consequently, studies of the effect of endotoxin on renal function in animals have been carried out to define the mechanisms which underlie ARF resulting from Gram-negative sepsis (see Kikeri et al 1986). One potential problem in such studies is investigation of renal function in the presence of endotoxin-induced hypotension. Species such as the goat, rabbit and dog are exquisitely sensitive to the cardiovascular effects of endotoxin which evokes a profound hypotension (see Kikeri et al 1986). The rat, by contrast, is relatively resistant to the systemic effects of endotoxin (Kikeri et al 1986). This species, therefore, allows a more rigorous evaluation of the renal effects of endotoxin without the complications of changes in systemic blood pressure.

One potential mediator of endotoxin-induced renal dysfunction is adenosine which has been shown to play an important role in the haemodynamic changes which occur in a number of forms of ARF (Churchill & Bidani 1990). Moreover, plasma concentrations of adenosine are known to be elevated 4- to 5-fold in sepsis, compared with levels in healthy volunteers (Bardenheuer & Thiel 1991). Micropuncture studies in the rat have shown that endotoxin induces vasoconstriction of afferent arterioles and a reduction in glomerular hydrostatic pressure (Conger et al 1981). Stimulation of adenosine A_1 receptors present in the afferent arteriole evokes constriction (Holz & Steinhausen 1987), and therefore blockade of these receptors may attenuate endotoxin-induced changes in renal haemodynamics. The aim of this study was to assess to what extent administration of 8-cyclopentyl-1,3-dipropylxanthine (CPX), a selective A_1 adenosine antagonist (Kellett et al 1989), could provide protection against endotoxin-induced renal dysfunction in the rat. Some of these findings have been presented to the British Pharmacological Society (Knight et al 1992).

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Materials and Methods

Materials

Escherichia coli endotoxin (serotype 055:B5) was obtained from Sigma Chemical Co., UK. [^{3}H(G)]Inulin (355·5 mCi g^{-1}), of stated radiochemical purity greater than 98%, was obtained from Du Pont NEN Research Products, UK. CPX was synthesized by Dr R. James of ICI Pharmaceuticals, UK.

Experimental protocol

Male albino Wistar rats, 250–350 g, were anaesthetized with thiobutabarbitone (180 mg kg^{-1}, i.p.) and cannulae inserted into the trachea to facilitate spontaneous ventilation, left jugular vein for saline infusion and drug administration, and right carotid artery for measurement of systemic blood pressure. Blood pressure was measured via a pressure transducer (Druck PDCR 75) with heart rate obtained electronically from the blood pressure signal. The abdomen was opened by a midline incision, a cannula inserted into the bladder for collection of urine and, in some experiments, an ultrasonic perivascular flow probe (model 2SB, Transonic Systems Inc., USA) was placed around the left renal artery. The probe was connected to a small-animal flowmeter (T206 Transonic Systems Inc.) to record mean renal blood flow (RBF). Body temperature was maintained at 37°C using a rectal thermometer and heating lamps. On completion of surgery, 2 mL saline (0·9% w/v NaCl) containing 0·35 μCi mL^{-1} [^{3}H]inulin were administered via the jugular vein. This solution was infused for the remainder of the experiment at a rate of 100 μL min^{-1}. A 60-min equilibration period was then allowed for stabilization of urine flow.

Each experiment consisted of seven 15-min clearance periods during which urine was collected into pre-weighed tubes, and a blood sample (0·5 mL) was taken at the midpoint of urine collection. Blood samples were centrifuged and the plasma separated for subsequent analysis. The erythrocytes were suspended in an equal volume of isotonic saline and transfused back into the animal. Following two control collection periods, rats received either *E. coli* endotoxin (20 mg kg^{-1}, i.v.) or saline (1 mL kg^{-1}, i.v.). Rats given endotoxin were injected with either saline (1 mL kg^{-1}), CPX (0·1 mg kg^{-1}) or the vehicle for CPX (1% v/v DMSO; 0·75 v/v 1 M NaOH in saline, 1 mL kg^{-1}), and animals which received saline were injected with either CPX (0·1 mg kg^{-1}) or a further dose of saline (1·0 mL kg^{-1}). Animals given saline only (2 × 1 mL kg^{-1}) are referred to as saline controls.

A separate series of experiments was conducted in which endotoxin was administered at a dose of 1 mg kg^{-1}. The experimental protocol was similar to that described above and included recording of left renal blood flow. As a result of the oliguria noted in experiments with 20 mg kg^{-1} endotoxin and the consequent difficulties of urine analysis, the clearance periods were extended to 30 min and reduced in number to five, of which only the first period acted as a control. By comparison with the experiments with 20 mg kg^{-1} endotoxin, a limited number of experimental groups were investigated, consisting of intravenous endotoxin administration followed by injection of either CPX (0·1 mg kg^{-1}) or its vehicle (1 mL kg^{-1}).

Urine and plasma analysis

Levels of [^{3}H]inulin in plasma and urine were determined by liquid scintillation counting. Plasma and urinary concentrations of sodium and potassium were determined by flame photometry and chloride levels were measured with a chloride meter (Corning 925).

Analysis of results

Standard formulae were used to calculate renal clearances of inulin (C_{IN}), sodium, potassium and chloride ions. Fractional excretion of the various ions was derived from: clearance/C_{IN} × 100. Results are expressed as mean ± s.e.m. Statistical comparisons were made within groups using a paired Student's *t*-test. Comparison of means between groups was made using either Student's *t*-test for unpaired data, or, where appropriate, by one-way analysis of variance with means compared by Scheffe's test.

Results

Effect of CPX on renal function

Administration of CPX to saline-treated rats had no significant effects ($P > 0·05$) on mean arterial blood pressure (MAP) or heart rate. For example, 5 min following its injection MAP was 96 ± 6 mmHg and heart rate 374 ± 11 beats min^{-1} (n = 8) compared with control levels of 97 ± 6 mmHg and 370 ± 11 beats min^{-1} (n = 8).

The effect of CPX on C_{IN}, urine flow and sodium excretion is shown in Figs 1a, 2a, 3a and Table 1. For brevity, data for the excretion of chloride and potassium have been omitted from this and subsequent experiments. However, changes in both absolute and fractional excretion of sodium were mirrored by changes in chloride excretion; but there was no statistically significant ($P > 0·05$) effect of CPX on the excretion rate or fractional excretion of potassium. These observations of the effect of CPX on electrolyte excretion are consistent with those previously reported (Knight et al 1993).

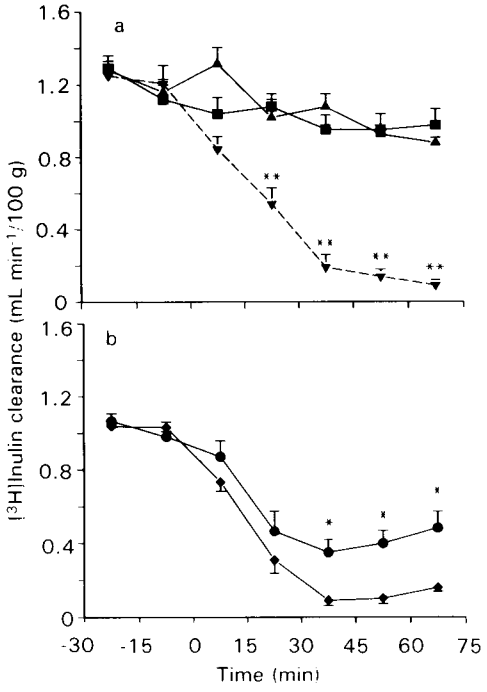

FIG. 1. Clearance of [^{3}H]inulin in anaesthetized male rats before and following intravenous administration at 0 min of a. saline (2 mL kg^{-1}) (■), saline (1 mL kg^{-1}) and CPX (0·1 mg kg^{-1}) (▲), and endotoxin (20 mg kg^{-1}) and saline (1 mL kg^{-1}) (▼). ** $P < 0·001$ compared with the value at the respective time point in the saline control group (unpaired *t*-test); b. endotoxin (20 mg kg^{-1}) and vehicle (1 mL kg^{-1}) (◆), and endotoxin (20 mg kg^{-1}) and CPX (0·1 mg kg^{-1}) (●). * $P < 0·05$ compared with the value at the respective time point in the endotoxin/vehicle group (analysis of variance). Each point represents mean ± s.e.m. (n = 8), except clearance periods 45–60 min and 60–75 min of the endotoxin/saline group and periods 15–30, 30–45 and 45–60 min of the endotoxin/vehicle group, where n = 5.

Effect of CPX on renal dysfunction induced by endotoxin (20 mg kg^{-1})

Administration of endotoxin (20 mg kg^{-1}, i.v.) followed by saline (1·0 mL kg^{-1}) produced an immediate fall in mean arterial pressure (MAP) of 28 ± 3 mmHg (n = 8) from control levels of 100 ± 5 mmHg (n = 8). Similarly heart rate fell by 83 ± 12 beats min^{-1} (n = 8) from control levels of 373 ± 16 beats min^{-1} (n = 8). Both MAP and heart rate returned to pre-endotoxin levels in less than 1 min, and there was no subsequent statistically significant change ($P > 0·05$) in either variable for the rest of the experiment. By contrast, endotoxin produced marked falls in C_{IN} and urine flow (Figs 1a, 2a). In addition, there were marked reductions in the excretion rates of sodium, potassium and chloride without any significant effect on the fractional excretion of these ions (Table 1, Fig. 3a sodium data only). The low volumes of urine produced during the later collection periods did not allow statistical comparisons to be made of sodium excretion

Table 1. Sodium excretion (μmol min^{-1}/100 g) in anaesthetized rats before and following intravenous injection of either saline (1 mL kg^{-1}) or endotoxin (20 mg kg^{-1}), followed by either saline (1 mL kg^{-1}), vehicle (1 mL kg^{-1}) or CPX (0·1 mg kg^{-1}).

	Control collection periods		Collection periods after injection				
Treatment	1 (−30–15 min)	2 (−15–0 min)	3 (0–15 min)	4 (15–30 min)	5 (30–45 min)	6 (45–60 min)	7 (60–75 min)
Saline controls	3·05 ± 0·61	3·13 ± 0·68	2·61 ± 0·72	1·88 ± 0·57	1·48 ± 0·48	1·31 ± 0·38	2·00 ± 0·62
Saline + CPX	3·50 ± 0·81	3·34 ± 0·79	8·37 ± 1·03**	7·91 ± 1·65*	7·84 ± 1·19**	6·33 ± 1·26*	4·49 ± 0·89*
Endotoxin + saline	2·32 ± 0·74	2·22 ± 0·42	2·82 ± 1·22	1·05 ± 0·21	0·30 ± 0·10*	0·13 ± 0·02	0·17 ± 0·00
Endotoxin + vehicle	3·37 ± 0·69	3·21 ± 0·41	2·99 ± 0·61	0·43 ± 0·06	0·08 ± 0·03	0·14 ± 0·00	0·19 ± 0·04
Endotoxin + CPX	3·74 ± 0·71	3·38 ± 0·58	5·71 ± 1·10	2·74 ± 0·90‡	1·03 ± 0·44	0·54 ± 0·16	0·51 ± 0·11†

Values are mean ± s.e.m. For each value n = 8, except periods 6 and 7 of the endotoxin-saline group and periods 5 and 6 of the endotoxin-vehicle group where n = 2. * $P < 0·05$, ** $P < 0·001$ compared with the respective collection period in the saline control group (unpaired t-test) and † $P < 0·05$, ‡ $P < 0·01$ compared with the respective collection period in the endotoxin/vehicle group (analysis of variance).

data obtained in endotoxin-injected animals (see Table 1, Fig. 3a). Results for C_{IN}, urine output and electrolyte excretion obtained from rats injected with endotoxin and saline were not significantly different ($P > 0·05$, using analysis of variance for all endotoxin-treated groups) from those obtained from rats given endotoxin and the vehicle for CPX. This is seen by a comparison of the results shown in parts a and b of Figs 1–3 and Table 1.

Injection of either CPX (0·1 mg kg^{-1}) or its vehicle (1·0 mL kg^{-1}) immediately following endotoxin injection had no significant ($P > 0·05$) effect on the transient changes in blood pressure or heart rate induced by endotoxin. By contrast, Figs 1b, 2b and 4 show that CPX attenuated the decline in C_{IN}, urine flow and RBF observed in endotoxin-treated rats. In addition, the marked reductions in the excretion of sodium, potassium and chloride, following endotoxin administration, were also significantly ($P < 0·05$) attenuated by co-administration of CPX (sodium data only is presented in Table 1). There was, however, no statistically significant difference ($P > 0·05$) in the fractional excretion of sodium, potassium or chloride ions between vehicle and CPX treatments in endotoxin-injected rats (Fig. 3b, sodium data only).

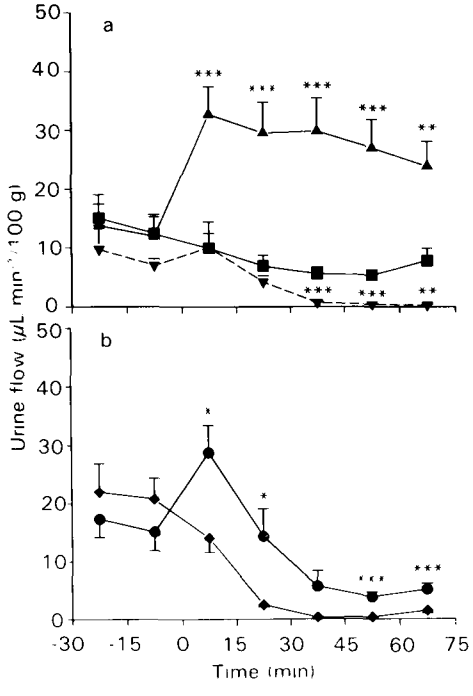

FIG. 2. Urine flow in anaesthetized male rats before and following intravenous administration at 0 min of a. saline (2 mL kg^{-1}) (■), saline (1 mL kg^{-1}) and CPX (0·1 mg kg^{-1}) (▲), and endotoxin (20 mg kg^{-1}) and saline (1 mL kg^{-1}) (▼). ** $P < 0·01$, *** $P < 0·005$ compared with the value at the respective time point in the saline control group (unpaired t-test); b. endotoxin (20 mg kg^{-1}) and vehicle (1 mL kg^{-1}) (◆), and endotoxin (20 mg kg^{-1}) and CPX (0·1 mg kg^{-1}) (●). * $P < 0·05$, *** $P < 0·001$ compared with the value at the respective time point in the endotoxin/vehicle group (analysis of variance). Each point represents mean ± s.e.m. (n = 8).

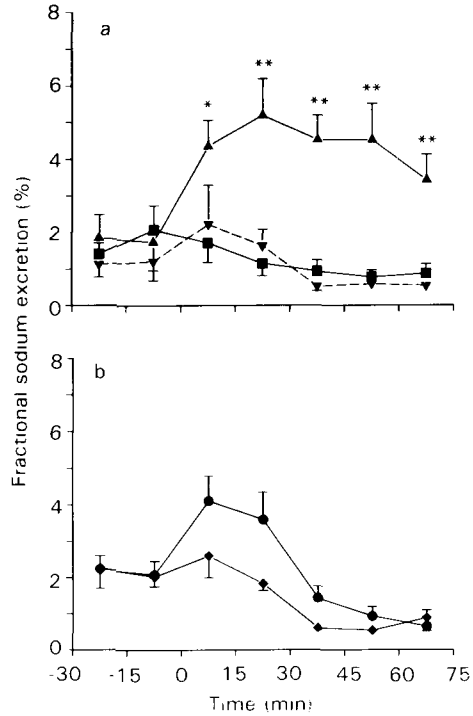

FIG. 3. Fractional sodium excretion in anaesthetized male rats before and following intravenous administration at 0 min of a. saline (2 mL kg^{-1}) (■), saline (1 mL kg^{-1}) and CPX (0·1 mg kg^{-1}) (▲), and endotoxin (20 mg kg^{-1}) and saline (1 mL kg^{-1}) (▼). * $P < 0·05$, ** $P < 0·01$ compared with the value at the respective time point in the saline control group (unpaired t-test); b. endotoxin (20 mg kg^{-1}) and vehicle (1 mL kg^{-1}) (◆), and endotoxin (20 mg kg^{-1}) and CPX (0·1 mg kg^{-1}) (●). Each point represents mean ± s.e.m. (n = 8), except clearance periods 45–60 and 60–75 min of the endotoxin/saline group and periods 30–45 and 45–60 min of the endotoxin/vehicle group where n = 2.

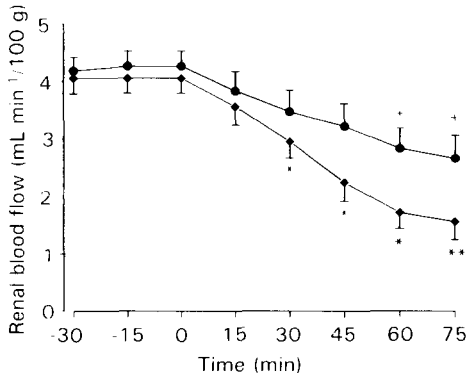

FIG. 4. Renal blood flow in anaesthetized rats before and following intravenous administration at 0 min of endotoxin (20 mg kg^{-1}) and vehicle (1 mL kg^{-1}) (◆), and endotoxin (20 mg kg^{-1}) and CPX (0·1 mg kg^{-1}) (●). Each point represents mean ± s.e.m. (n = 8). * $P < 0.05$, ** $P < 0.01$ compared with control period −15–0 min (paired t-test). † $P < 0.05$ compared with the value at the respective time point in the endotoxin/vehicle group (unpaired t-test).

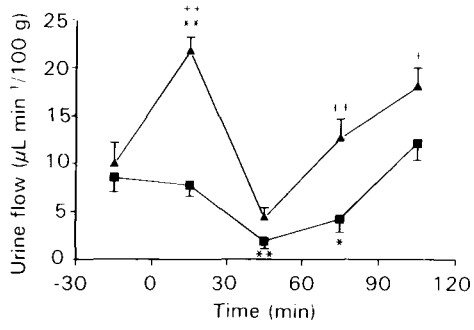

FIG. 6. Urine flow in anaesthetized rats before and following intravenous administration at 0 min of endotoxin (1 mg kg^{-1}) and vehicle (1 mL kg^{-1}) (■), and endotoxin (1 mg kg^{-1}) and CPX (0·1 mg kg^{-1}) (▲). Each point represents mean ± s.e.m. (n = 6). * $P < 0.05$, ** $P < 0.01$ compared with control period −30–0 min (paired t-test). $^{+}$ $P < 0.05$, $^{++}$ $P < 0.01$ compared with the value at the respective time point in the endotoxin/vehicle group (unpaired t-test).

Effect of CPX on renal dysfunction induced by endotoxin (1 mg kg^{-1})

Administration of a twentyfold lower dose of endotoxin in vehicle-treated rats resulted in transient falls in MAP and heart rate of 30 ± 4 mmHg and 78 ± 6 beats min^{-1} (n = 6), respectively from control levels of 99 ± 6 mmHg and 362 ± 14 beats min^{-1} (n = 6). As noted with 20 mg kg^{-1} of endotoxin, these systemic haemodynamic changes were unaffected by the treatment with CPX (0·1 mg kg^{-1}). C_{IN}, RBF and urine flow in rats which received endotoxin and vehicle, and endotoxin and CPX are shown in Figs 5 and 6. The urinary excretion of sodium, potassium and chloride were significantly reduced by endotoxin (1 mg kg^{-1}), with maximal depression recorded during the third collection period (30–60 min, Fig. 7a sodium data only), although, as noted with the higher dose of endotoxin (20 mg kg^{-1}), there was no statistically significant decrease in the fractional excretion of these ions (Fig. 7b, sodium data only). Administration of

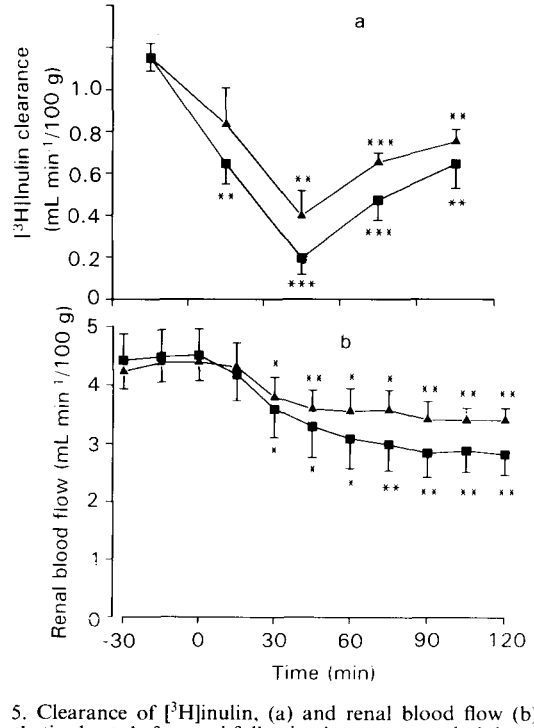

FIG. 5. Clearance of [^{3}H]inulin, (a) and renal blood flow (b), in anaesthetized rats before and following intravenous administration at 0 min of endotoxin (1 mg kg^{-1}) and vehicle (1 mL min^{-1}) (■ and endotoxin (1 mg kg^{-1}) and CPX (0·1 mg kg^{-1}) (▲). Each point represents mean ± s.e.m. (n = 6). * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$ compared with control period −30–0 min (paired t-test).

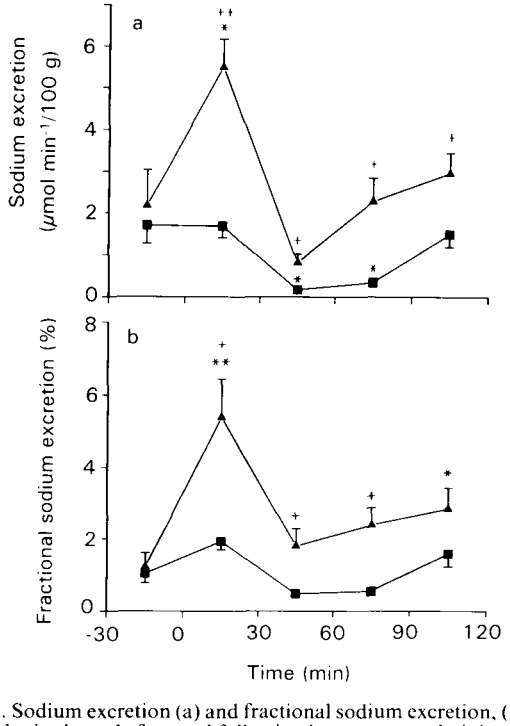

FIG. 7. Sodium excretion (a) and fractional sodium excretion, (b), in anaesthetized rats before and following intravenous administration at 0 min of endotoxin (1 mg kg^{-1}) and vehicle (1 mL kg^{-1}) (■), and endotoxin (1 mg kg^{-1}) and CPX (0·1 mg kg^{-1}) (▲). Each point represents mean ± s.e.m. (n = 6). * $P < 0.05$, ** $P < 0.01$ compared with control period −30–0 min (paired t-test). $^{+}$ $P < 0.05$, $^{++}$ $P < 0.01$ compared with the value at the respective time point in the endotoxin/vehicle group (unpaired t-test).

CPX to endotoxin-treated rats elicited an increase in the excretion rate of sodium, potassium and chloride ($P < 0.05$), during the first collection period after injection (0–30 min). The excretion of these ions subsequently fell, but remained significantly higher ($P < 0.05$) in the CPX-treated group than in the group given its vehicle (Fig. 7a, sodium data only). The administration of CPX to endotoxin-treated rats also resulted in significant ($P < 0.05$) increases in the fractional excretion of sodium and chloride, but not potassium, when compared with values from the endotoxin/vehicle group of rats (Fig. 7b sodium data only).

Discussion

The results show that in the anaesthetized rat, endotoxin administration produces oliguria and marked reductions in C_{IN} and RBF. The changes in renal haemodynamics occurred in the presence of a stable systemic blood pressure which indicates that endotoxin selectively increases renal vascular resistance. *E. coli* endotoxin evoked disproportionate changes in C_{IN} and RBF with greater reductions occurring in C_{IN}. This is consistent with an increase in the ratio of pre- to post-glomerular arteriolar resistance or contraction of mesangial cells with a subsequent decrease in the ultrafiltration coefficient. These observations on the renal haemodynamic actions of endotoxin concur with those of Conger et al (1981), who also described reductions in RBF and glomerular filtration rate in the rat. In the present study, the urinary excretion of sodium, potassium and chloride was also markedly reduced following exposure to endotoxin. However, the lack of any statistically significant effect of endotoxin on the fractional excretion of these ions, suggests that reabsorption of solutes within the tubule is preserved. These findings are in accord with those of Churchill et al (1987), who found that fractional excretion of water was unchanged after endotoxin administration, and that fractional excretion of sodium actually decreased, indicating enhanced sodium reabsorption by the nephron.

The mechanism which underlies endotoxin-induced renal dysfunction is at present unknown. Endotoxin is known to injure endothelial cells both in-vivo and in-vitro, and to induce the production of a myriad of vasoactive and inflammatory mediators such as anaphylatoxins, tumour necrosis factor, interleukin-1 and eicosanoids (Morrison & Ryan 1987). Churchill et al (1987) demonstrated, in rat, that theophylline, an alkylxanthine which shows no selectivity in blocking adenosine-receptor subtypes, attenuates the falls in the clearance of inulin and *p*-aminohippurate following endotoxin infusion. Those authors suggested that adenosine played a significant role as a mediator of the renal haemodynamic changes associated with the development of endotoxaemic ARF in the rat. In the present study, administration of the selective A_1 adenosine antagonist, CPX, simultaneously with endotoxin, afforded statistically significant protection against the reductions in C_{IN}, RBF, urine flow rate and urinary excretion of sodium, potassium and chloride ions induced by 20 mg kg^{-1} endotoxin. However, this protection provided by CPX against the acute effects of endotoxin was by no means complete. For example, in rats treated with 20 mg kg^{-1} endotoxin, C_{IN} fell to 35% of control values in rats given CPX, compared with a decline to < 10% in rats given its vehicle. This incomplete protective effect of CPX is unlikely to be due to inadequate adenosine receptor blockade, since we have shown in rats that 0·1 mg kg^{-1} CPX produces almost complete inhibition of adenosine-induced bradycardia for over 5 h (Knight et al 1991). By contrast with the present results, Churchill et al (1987) reported greater protective effects with theophylline. They found that this alkylxanthine, whilst attenuating endotoxin-induced decrease in the clearance of inulin, prevented any fall in the clearance of *p*-aminohippurate, a measure of renal plasma flow. In the study of Churchill et al (1987) endotoxin was administered by infusion (5 mg kg^{-1} h^{-1}) which resulted in decreases of 40 and 60% in the clearances of inulin and *p*-aminohippurate, respectively, whereas in our investigation the depression of renal function induced by a bolus dose of 20 mg kg^{-1} endotoxin was considerably more severe. It was possible, therefore, that the severity of renal dysfunction in our experiments masked the full protective potential of CPX. Consequently, experiments were conducted to establish whether a greater degree of protection could be achieved with a reduced dose of endotoxin.

Administration of endotoxin at a dose of 1 mg kg^{-1} to rats produced a similar pattern of changes to the 20 mg kg^{-1} dose, but there was a reduction in both the severity and duration of these changes. Reductions in urine flow, C_{IN} and electrolyte excretion reached a nadir at 30–60 min after 1 mg kg^{-1} endotoxin administration before starting to recover, whereas little recovery was seen in rats given 20 mg kg^{-1} endotoxin. CPX significantly improved urine output and electrolyte excretion in rats given endotoxin (1 mg kg^{-1}), without any statistically significant enhancement of C_{IN} or RBF. Thus a greater protective effect of CPX was not observed in rats given the lower dose of endotoxin.

The effects of CPX in rats given 1 mg kg^{-1} endotoxin are consistent with its reported diuretic effects (Knight et al 1993) and reproduced here in control rats. It is possible that the diuretic actions of CPX are responsible for the renal protective effects following endotoxin, since diuretic treatment has been shown to ameliorate other forms of ARF (Bailey et al 1973). However, CPX produced a significant and persistent amelioration of the decline in C_{IN} induced by 20 mg kg^{-1} endotoxin, whereas in normal animals injected with CPX, no significant elevation in C_{IN} was noted. Furthermore, in rats given 1 mg kg^{-1} endotoxin, the diuretic actions of CPX were evident yet there was no statistically significant improvement in C_{IN} or RBF. Moreover, Kikeri et al (1986) have shown that volume expansion with isotonic saline and subsequent diuresis and natriuresis had no protective effect on the development of endotoxaemic ARF.

CPX had no significant effect on the fractional excretion of sodium and chloride in rats given 20 mg kg^{-1} endotoxin in contrast to the significant elevations noted when CPX was administered to either normal animals or to rats injected with 1 mg kg^{-1} endotoxin. This may reflect the fact that as glomerular filtration rate is severely depressed following 20 mg kg^{-1} endotoxin, the filtered load of these ions would be considerably reduced. Consequently, any inhibition of transport mechanisms within the proximal tubule, the previously identified site of CPX's diuretic action (Knight et al 1993), may be compensated for by enhanced reabsorption along the subsequent nephron segments.

The lack of any statistically protective effects afforded by CPX with respect to the falls in C_{IN} or RBF in rats receiving 1 mg kg^{-1} endotoxin, contrasts with that in rats given the higher dose of endotoxin, where significant protection was evident. This difference in sensitivity to the renal protective action of adenosine antagonism supports the contention that the initiation of endotoxaemic ARF is mediated by other factors, such as leukotrienes and thromboxane A_2 (Badr et al 1986) or platelet-activating factor (Toplins et al 1984) and that adenosine only plays a role in significantly compromising renal haemodynamics when the endotoxaemic insult is severe enough to produce substantial release of the purine. The source of adenosine in a severe insult could be either damaged endothelial cells or renal parenchymal cells with depleted ATP levels.

In summary, a bolus injection of *E. coli* endotoxin induced acute reductions in C_{IN}, RBF, urine output and electrolyte excretion. Co-administration of the A_1 selective adenosine antagonist CPX resulted in a statistically significant attenuation of the reduction in C_{IN} and RBF in animals receiving high dose but not low dose endotoxin. The results suggest that adenosine may play a role, albeit not a major one, in the pathophysiology of endotoxaemic ARF.

References

Badr, K. F., Kelley, V. E., Rennke, H. G., Brenner, B. M. (1986) Roles for thromboxane A_2 and leukotrienes in endotoxin-induced acute renal failure. Kidney Int. 30: 474–480

Bailey, R. R., Natale, R., Turnbull, D. I., Linton, A. L. (1973) Protective effect of frusemide in acute tubular necrosis and acute renal failure. Clin. Sci. Mol. Med. 45: 1–17

Bardenheuer, H., Thiel, M. (1991) The biological role of adenosine for the cellular function during sepsis. In: Messmer, K. (ed.) Capillary Functions and White Cell Interaction. Progress in Applied Microcirculation. Vol. 18, Karger, Basel, pp 112–131

Churchill, P. C., Bidani, A. K. (1990) Adenosine and renal function. In: Williams, M. (ed.) Adenosine and Adenosine Receptors. Humana Press, Clifton, NJ, pp 335–380

Churchill, P. C., Bidani, A. K., Schwartz, M. M. (1987) Renal effects of endotoxin in the male rat. Am. J. Physiol. 253: F244–F250

Conger, J. D., Falk, S. A., Guggenheim, S. J. (1981) Glomerular dynamics and morphologic changes in the generalised Schwartzman reaction in post-partum rats. J. Clin. Invest. 67: 1334–1346

Holz, G., Steinhausen, M. (1987) Renovascular effects of adenosine receptor agonists. Renal Physiol. 10: 272–282

Kellett, R., Bowmer, C. J., Collis, M. G., Yates, M. S. (1989) Amelioration of glycerol-induced acute renal failure in the rat with 8-cyclopentyl-1,3-dipropylxanthine. Br. J. Pharmacol. 98: 1066–1074

Kikeri, D., Pennell, J. P., Huang, K. H., Jacobs, A. I., Richman, A. V., Bourgoignie, J. J. (1986) Endotoxemic acute renal failure in awake rats. Am. J. Physiol. 250: F1098–F1106

Knight, R. J., Collis, M. G., Yates, M. S., Bowmer, C. J. (1991) Amelioration of cisplatin-induced acute renal failure with 8-cyclopentyl-1,3-dipropylxanthine. Br. J. Pharmacol. 104: 1062–1068

Knight, R. J., Bowmer, C. J., Yates, M. S. (1992) The effect of 8-cyclopentyl-1,3-dipropylxanthine on *E. coli* endotoxin-induced renal dysfunction. Br. J. Pharmacol. 106: 18P

Knight, R. J., Bowmer, C. J., Yates, M. S. (1993) The diuretic action of 8-cyclopentyl-1,3-dipropylxanthine, a selective A_1 adenosine receptor antagonist. Br. J. Pharmacol. 109: 271–277

Morrison, D. C., Ryan, J. L. (1987) Endotoxins and disease mechanisms. Ann. Rev. Med. 38: 417–432

Toplins, J. P., Vercellotti, G. M., Wilkowske, M., Ha, B., Jacob, H. S., Raij, L. (1984) Role of platelet activating factor in endotoxemic acute renal failure in the rat. J. Lab. Clin. Med. 113: 316–324

Turney, J. H., Marshall, D. H., Brownjohn, A. M., Ellis, C. M., Parsons, F. M. (1990) The evolution of acute renal failure 1956–1988. Q. J. Med. 74: 83–104