readily absorbed from the rectal compartent. Surgical disruption of the lymphatic route had a lesser impact on the adjuvant plasma profiles compared with the model drug compounds. Plasma profiles indicate an approximate 60% reduction in AUC of sodium 5-methoxysalicylate and sodium salicylate as a result of the thoracic duct cannulation.

For each compound studied, the total amount recovered from the thoracic duct after rectal administration did not totally account for the reduction in plasma AUC. This apparent discrepancy in the data has been attributed to technical problems with the surgical procedure. Some of the collected lymph was invariably lost through leakage around the catheter insertion site. This leakage was often compounded by back pressure in the catheter from lymphatic coagulation.

These experiments have shown that the gut-associated lymphatic system plays an important role in the transport of water-soluble compounds after rectal absorption. Drug access to this transport mode could help explain the reported instances of apparent direct systemic drug delivery from the rectal compartment. In this study, the rectal bioavailability of one model drug, cefoxitin, was reduced more than 90% by interrupting lymphatic return. Chemical modification of drug substances in conjunction with the use of certin absorption adjuvants may offer a means of targeting drug transport after rectal absorption. The application of directed lymphatic transport may not only permit avoidance of first-pass liver exposure but may also allow site-specific delivery of drugs targeted toward gut-associated lymphoid tissue.

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Effect of methysergide on renal function

Y. L. CHAN*, W. R. JACOBS, *Department of Physiology and Biophysics, University of Illinois Medical Center, Chicago, Illinois 60612. U.S.A.*

5-Hydroxytryptamine (5-HT) has been implicated as a possible regulator of renal function (Erspamer 1966). While its antidiuretic effect has been demonstrated in rats (Del Grego et al 1956) and dogs (Spinazzola & Sherod 1957) the mechanism by which this antidiuresis occurs remains obscure. The intravenous infusion of 5-HT has been shown to decrease renal cortical blood flow with a subsequent decrease in glomerular filtration rate and urine (Erspamer & Ottolenghi 1953). However, other studies have shown that its infusion can produce an antidiuresis in the absence of significant changes in either renal blood flow or glomerular filtration rate (Little et al 1961). In the latter study the antidiuresis was characterized by a decrease in urine flow rate and sodium excretion. Similar reductions in sodium excretion and urine flow rate have been observed during animal surgery (Maddox et al 1977). It has been proposed that the decreases in sodium excretion and urine flow rate may be due, in part, to the release of neurotransmitters which presumably affect renal function (Maddox et al 1977). Our recent report has shown that methysergide, a 5-HT antagonist, could produce a diuresis in Sprague-Dawley rats receiving large doses of horseradish peroxidase, a protein known to cause vascular leakage and antidiuresis in rats (Chan & Straus 1980). The present study was undertaken to further elucidate the effect of methysergide on renal function.

* Correspondence.

Methods

Male Sprague-Dawley rats 180 to 250 g, were maintained on a regular rat pellet diet and tap water, were anaesthetized with 5-ethyl-5-(L-methylpropyl)-2-thiobarbituric acid (Inactin), 100 mg kg^{-1} i.p. They were placed on a thermostatically controlled animal table where their body temperatures were maintained at 37 °C. The surgical procedures were similar to those of preparing the animal for micropuncture described previously (Chan 1976). Briefly, after tracheotomy, the external jugular vein was cannulated for saline infusion and drug administration. The left carotid artery was cannulated for blood pressure recording as well as for blood sampling. The left kidney was exposed laterally and then immobilized in a double-cup (W. Hampel, Frankfurt, Germany). The left renal artery was exposed to allow arterial injection of the drug through a 30 gauge needle, avoiding constriction of the artery. Pre-warmed mineral oil flowed over the kidney throughout the experiment to maintain the kidney temperature at 37 °C. The ureter was catheterized to allow unhindered flow and for

Table 1. *Effect of methysergide on haemodynamics and renal sodium excretion.*

	Saline			Methysergide		
	Control		Experimental	Control		Experimental
FE_{Na} %	0·21 ± 0·017	$P > 0·05$	0·20 ± 0·23	0·19 ± 0·015	$P < 0·01$	0·38 ± 0·032
RBF ml min^{-1}/100 g b. wt	6·3 ± 0·36	$P > 0·05$	6·4 ± 0·56	6·9 ± 0·26	$P < 0·05$	7·8 ± 0·32
RPF ml min^{-1}/100 g b. wt	3·2 ± 0·42	$P > 0·05$	3·3 ± 0·18	3·5 ± 0·29	$P < 0·05$	4·0 ± 0·23
HCT %	42 ± 2·4	$P < 0·05$	49 ± 2·30	43 ± 1·6	$P > 0·05$	45 ± 2·1

Saline (0·3 ml), methysergide (0·3 ml, 10^{-7} mol/100 g) FE: Fractional excretion of sodium, RBF: Renal blood flow, RPF: Renal plasma flow, HCT: Haematocrit. Control samples were taken after the 60 min equilibration time. Experimental samples were taken 30 min after injection of either saline or methysergide. Data are expressed as mean ± s.e. 4–6 rats. $P < 0·05$ is significantly different.

collection of urine. A 3% solution of inulin in 0.9% NaCl (saline) was infused at a rate of 1.8 ml h^{-1} through the jugular vein. An equilibration period of 60 min was allowed before any urine collection was started. Urine samples were collected for 10 min under mineral oil in pre-weighed glass vials and the urine volume was determined gravimetrically. Blood samples were collected from the carotid artery during the mid-portion of each clearance period. Blood and urine samples were analysed for inulin, PAH, Na^+, and K^+. Inulin was assayed by the method of Führ et al (1955), Na^+ and K^+ were determined by flame photometry. Clearance of inulin was used to determine glomerular filtration rate (GFR). Electrolyte excretion was expressed in both absoluted terms and as fractional excretion (clearance of the electrolyte corrected for GFR and expressed as percentage). Blood samples were also collected from the renal vein to permit calculation of renal blood flow by determining the renal extraction of inulin. Clearance of PAH was used to estimate renal plasma flow (RPF). PAH was determined by auto-analyser according to Smith et al (1945).

To test the effect of the methysergide, a single dose in 0·3 ml of either saline or saline plus drug was injected into individual animals after three 10 min control urine collections had been made. Six more clearance periods of 10 min each were carried out following the drug administration.

Data are presented as mean ± standard error. Statistical significance of differences between experimental data was evaluated by Student's *t*-test.

Results

Control rats (4) received i.v. 0·3 ml saline without methysergide. There were no significant changes in mean arterial blood pressure, urinary flow rate, urinary excretion of sodium, GFR, and RPF before and after the injection (Fig. 1, Table 1). The slight gradual reduction in urine flow and Na^+ excretion during the 90 min collection period may be the result of a slight vascular dehydration as indicated by an increase of haematocrit (Table 1) or an anaesthetic effect.

In contrast, intravenous administration of 10^{-7} mol/100 g methysergide in six additional rats resulted in a significant natriuresis and diuresis with little change in renal blood flow and GFR.

During the third 10 min experimental period following the drug injection, urine flow increased by 124% ($P < 0·001$) with a simultaneous increase in Na^+ excretion of 99% ($P < 0·001$) over control values. In the same period GFR and RPF increased slightly, 9·3% ($P < 0·05$) and 13% ($P < 0·05$) respectively; mean arterial blood pressure was not affected; the fractional excretion of Na^+ increased by 105% ($P < 0·001$) indicating a significant natriuresis independent of the slightly increased GFR.

Fig. 1 shows the time course for the response to methysergide injection. The maximum changes in urine flow rate, Na^+ excretion and fractional excretion of Na^+ occurred 30 min after methysergide injection with most values returning to near control levels at the end of 60 min. The notable exception is the slight increase in GFR at 30 min which continued to increase throughout the experimental period. Similar results were observed when methysergide was administered into the renal artery.

Discussion

The results from this investigation demonstrate that methysergide, a peripheral antagonist of 5-HT, has a natriuretic and diuretic effect on the rat kidney. The data also show that the effect of methysergide on renal function was not mediated by changes in systemic blood pressure, RPF or GFR. The observation that methysergide greatly increased the fractional excretion of Na^+, while only slightly increasing GFR and RPF, indicates that the drug caused a decrease in tubular Na^+ and water reabsorption. The mechanism by which methysergide acted to decrease water and Na^+ reabsorption is not clear from the present data, however, several possibilities exist. Of particular interest is the possibility that methysergide antagonizes the action of 5-HT on the kidney.

Previous investigators have shown that the infusion of 5-HT results in an antidiuresis (Alder 1977). While most of these studies indicated that the effect of 5-HT was to

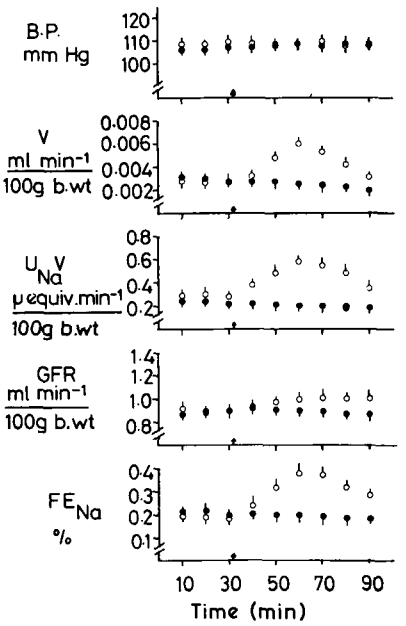

FIG. 1. Effect of methysergide on systemic blood pressure and renal function on non-diuretic rats. Methysergide or saline was administered intravenously (0·3 ml) after three control clearance periods as indicated by arrows. Close circles represent experiments with saline injection. Open circles represent experiments with methysergide injection. B. P.: mean arterial blood pressure. V: urine flow rate. $U_{Na}V$: urinary excretion of sodium. GFR: glomerular filtration rate. FE_{Na}: fractional excretion of sodium ($U_{Na}V/GFR \times P_{Na}$). Abscissa: Urine collection period for every 10 min in sequence after equilibration period for inulin infusion. Data are mean ± s.e. for 4–6 rats.

decrease renal cortical blood flow and GFR, resulting in an antidiuresis, several investigators reported that low dose infusion could induce an antidiuresis without changing GFR or renal blood flow (Little et al 1961; Park et al 1968). Little et al (1961) reported that low dose 5-HT infusion in dogs reduced the renal excretion of Na^+ without changing urine flow while Park et al (1968) showed that slightly higher doses of 5-HT reduced both the renal excretion of sodium and urine flow. It has recently been reported that 5-HT stimulates a slight but significant increase in the content of cAMP in suspensions of renal cortical tubules (Shaw et al 1979), suggesting that there may be 5-HT receptors directly on the renal tubules. However, our preliminary micropuncture studies (Chan & Krantz 1978) have demonstrated that there is only a slight, but not statistically significant decrease in proximal tubular water reabsorption after methysergide injection, indicating that methysergide may act on the renal tubule at some site beyond the proximal segment.

While it is apparent that the primary effect of methysergide injection was on renal function it should also be noted that changes in extracellular fluid distribution may also have occurred and may be a contributing factor to the observed renal effects of the drug. During control experiments the arterial haematocrit rose an average of 7%. The increase in haematocrit during animal surgery has been reported previously (Maddox et al 1977) and indicates a loss of circulating plasma volume. However, during experiments with methysergide the arterial haematocrit rose only an average of 2% indicating that methysergide may have a protective effect on the vascular volume changes associated with surgery. This protective effect of methysergide was also observed in a previous study in which large doses of horseradish peroxidase induced a dramatic increase in haematocrit (Chan & Straus 1980), presumably by stimulating the release of 5-HT from intestinal mast cells (Cotran & Karnovsky 1967) which increased vascular permeability. It was found that pretreatment of the rat with methysergide prevented the obligatory increase in haematocrit associated with horse radish peroxide injection by presumably antagonizing the effect of 5-HT on vascular permeability. While the data from this investigation indicate that 5-HT may play a role in the obligatory decrease in circulating plasma volume, plasma 5-HT levels were not measured.

In conclusion, the data show that methysergide has natriuretic and diuretic effects on rat kidneys and support the view that 5-HT has an effect on tubular water and sodium reabsorption independent of changes in renal haemodynamics and GFR.

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