Renal effects of Paraoxon in the Rat

by

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Experiments investigating the direct renal activity of anticholinesterase agents have failed to show any effects. LAVENDER et al (1965) failed to show any change in renal function due to the infusion of physostigmine, neostigmine or DEP into a renal artery. WILLIAMS AND PEARSON (1970) also reported no direct renal effects during the infusion of paraoxon intra-renally. However, studies of the parasympathomimetic drugs, pilocarpine CARTER et al (1961), WILLIAMS AND CARTER (1965) and the anticholinesterase, physostigmine CARTER and ATKINSON (1961), CARTER AND ATKINSON (1961) administered intravenously or subcutaneously, resulted in increases in electrolyte excretion in rats.

The purpose of this study is to investigate the chronic and acute renal effects of the cholinesterase inhibitor, paraoxon. Paraoxon [diethyl-4-nitrophenyl phosphate] is the active metabolite of the organophosphate pesticide, parathion, and is a prompt, potent acetylcholinesterase inhibitor MURPHY et al (1968). Because parathion is widely used in the control of agriculture and domestic pests, an investigation of its effects on renal function following acute and chronic exposure was warranted. DAVIES et al (1969) have reported changes in phosphate excretion in occupationally exposed spraymen.

Methods

Chronic Study

Female Holtzman rats weighing 200-250 grams were divided into two groups, housed in metabolism cages, and maintained on Purina Rat Chow. The experimental group received daily injections of 0.1 mg/kg body weight of paraoxon subcutaneously for 6 to 13 days. The control animals were injected daily with an equal volume of normal saline. Twenty-four hour urine samples were collected. The volumes were recorded and aliquots of the samples were analyzed for sodium, chloride, potassium, urea, inorganic phosphorus and osmolality. At the end of the experimental period the animals were sacrificed and the kidneys removed for histological studies.

Acute Study

Female Sprague Dawley rats weighing 140-160 grams were divided into a control and an experimental group and hydrated with tap water, 25 ml/kg body weight. The experimental rats were injected subcutaneously with paraoxon 0.15 mg/kg body weight and the con-

Bulletin of Environmental Contamination & Toxicology,
Vol. 11, No. 4 © 1974 by Springer-Verlag New York Inc.

TABLE 1. CHRONIC EFFECTS OF PARAOXON

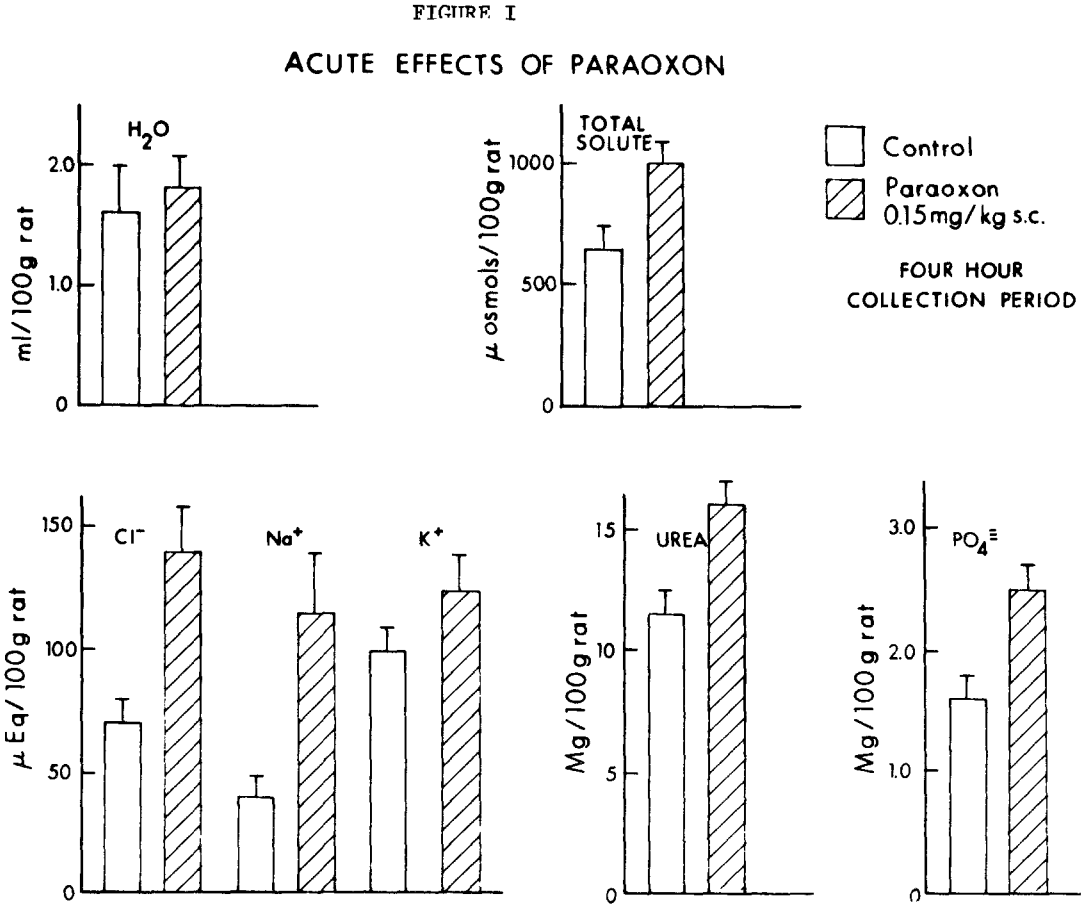
DAYS	1	2	3	4	5	6	7	8	9	10	11	12	13
						Volume cc/100gm Rat.							
Control	3.7	4.0	3.2	4.4	4.3	3.3	3.1	3.2	2.5	1.8			
No.	(23)	(23)	(20)	(22)	(15)	(10)	(5)	(5)	(5)	(5)			
Exptl.	5.0*	6.5*	6.3*	6.9*	7.7*	8.1*	9.4*	9.8*	9.0	5.7	7.2	6.2	6.1
No.	(23)	(23)	(21)	(21)	(19)	(13)	(7)	(5)	(5)	(5)	(5)	(5)	(5)
						Milliosmols/100 gm							
Control	5553	5470	5106	6258	6144	4753	5112	4829	4439	3665			
No.[1]	(23)	(23)	(20)	(22)	(15)	(8)	(5)	(5)	(5)	(5)			
Exptl.	5994	5926	5570	6691	6272	7301*	7134	7897*	6873	5510	6358	7139	6913
No.	(23)	(23)	(21)	(21)	(21)	(19)	(13)	(7)	(5)	(5)	(5)	(5)	(2)
						Sodium µeq/100 gm							
Control	456	453	372	519	502	380[2]	440	389	378	282			
Exptl.	506	443[3]	396	431	441	489	537	510	467	422	489	527	549
						Chloride µeq/100 gm							
Control	518	487	450	579	560	415[2]	484	495	437	336			
Exptl.	486	408	354	446	431	490	503	480	464	426	493	521	519
						Potassium µeq/100 gm							
Control	723	670	633	801[4]	763	588[2]	683	644	607	446			
Exptl.	681	546[5]	547	642	612	683	638	722	676	569	681	762	679
						Phosphate mg/100 gm							
Control	7.5	6.6	5.5	7.0	6.4	4.7[2]	5.4	5.3	5.1	3.5			
Exptl.	6.3	5.4	5.8	6.0	6.1	6.1	6.2	5.9	4.6	4.3	5.0	5.7	4.0
						Urea mg/100 gm							
Control	78.8	76.1	67.2	86.0	83.0	61.2[2]	78.8	69.2	68.2	44.5			
Exptl.	79.0	69.7	70.8	74.6	79.7	77.1	82.2	82.1	73.5	67.7	59.1	78.0	70.7

* indicates significant difference at level of $P \leq 0.05$

No. equals number of samples.

[1] number of samples identical for remaining analytical determinations except [2]=9, [3]=22, [4]=23, [5]=22.

FIGURE I



The acute effects of the anticholinesterase, paraoxon on renal function.

trol animals received an equal volume of normal saline. They were housed in metabolism cages and four hour urine samples were collected. The urine samples were handled as described in the chronic study.

RESULTS

Chronic Study

The daily subcutaneous administration of paraoxon, 0.1 mg/kg body weight, to female Holtzman rats resulted in no significant changes in electrolyte excretion (24 hour collection). After six to thirteen consecutive daily injections of paraoxon, urine sample analysis indicated no changes in sodium, chloride, potassium, phosphate or urea excretion when compared to control. There were, however, some significant changes in osmolalities (Table 1). Urine volume showed the only significant difference between control and experimental values in a daily sequence. As much as a three-fold increase in urine volume was observed after the adminintration of paraoxon. Histological examination of kidney tissues showed no significant changes between control and experimental.

Acute Results

The subcutaneous administration of paraoxon 0.15 mg/kg body weight, to hydrated female Sprague Dawley rats resulted in marked increases in sodium, chloride, urea, and phosphate excretions over a four hour period. Osmolalities also showed a marked increase after paraoxon injection. Changes in potassium excretion are less marked but significant. Paraoxon injection caused no significant changes in urine volume (Fig. I).

Discussion

Paraoxon given at doses of 0.15 mg/kg, body weight, subcutaneously resulted in the observable pharmacological effects of cholinergic stimulation. These were: muscle tremors, salivation, lacrimation, preening, and chewing. The dose effect response is rather sharp because 0.1 mg/kg subcutaneously of paraoxon seemed to cause much less effect and very few deaths due to acute poisoning. Therefore the animals were chronically exposed to 0.1 mg/kg of paraoxon for 6-13 days. Under our protocol of 24 hour collections there were no apparent renal effects with the exception of urine volume. This increase in daily urine volume was statistically significant. Sometimes as much as a three-fold increase was observed. However, there were no significant effects upon the renal excretion of total solutes of urea.

In contrast, the rats which were treated acutely with paraoxon following hydration responded differently. After a four-hour collection of urine there was a significant increase in sodium, chloride, phosphate, urea, and total solute. However, there was no increase in water excretion in the treated rats compared to the

controls. This pattern of electrolyte and water excretion in acute rats given paraoxon was identical to that which was reported for arecoline, a direct acting cholinergic stimulant which mimics the action of acetylcholine WILLIAMS and CARTER (1965). Since atropine blocked all the renal effects of arecoline upon the kidney it was suggested that arecoline acted like acetycholine by its muscarinic effect upon renal tubules. However, systemic cholinergic stimulation also reflexly releases catecholamines. The direct effect of norepinephrine upon the kidney causes salt retention, WILLIAMS, R.L. and CARTER, M.K., however, systemic levels of norepinephrine can cause salutesis by increasing the arterial blood pressure. The interpretation of the response is complicated by the release of catecholamines, norepinephrine from the sympathetic nevers and epinephrine from the adrenal medulla , WILLIAMS, R.L. and PEARSON, J.E.,JR. (1970). GREEN and SIM (1961) demonstrated that norepinephrine and epinephrine when administered subcutaneously to hydrated rats resulted in saluresis. It is concluded from this and other studies that the saluresis observed following acute exposure to paraoxon is a result of the cholinergic effect of acetylcholine upon the kidney and potentiation of this effect through an increase in systemic blood pressure caused by circulating catecholamines.

It is also concluded from these studies that the initial effect of paraoxon exposure is a saluresis, followed by a long period of salt retention. Because of the longer duration of action of catecholamines the 24-hour collection reveals no effect while a 4-hour period reveals the renal saluresis of cholinergic stimulation.

References

BURN, J.H.: The Autonomic Nervous System, Oxford. Blackwell Scientific Publications (1968).

CARTER, M.K. and ATKINSON, S.L.: Fed. Proc. 20 [I] 412 (1961).

CARTER, M.K., HEMSTREET, G.K. AND REID, J.W.: Pharmacologist 3:57 (1961).

DAVIES, J.E., MANN, J.B. AND TOCCI, P.M.: Annals, N.Y. Acad. Sc. 160: 323, (1969).

GREEN, A.F. AND SIM, M.F.: Brit, J. Pharma. Co.. Chemother. 17; 464 (1961).

LAVENDER, A.R.. AHO, I. and PULLMAN, T.N.: Proc. Soc. Exptl. Biol, Med.; 119: 887 (1965).

MURPHY, S.D., LAUWERYS, R.R. AND CHEEVER, K.L.: Toxicol. Appl. Pharmacol. 12: 22-35 (1968).

PEARSON, J.E. AND WILLIAMS, R.L.:Brit. J. Pharmacol. 33:223 (1968).

WILLIAMS, R.L. AND CARTER, M.K.: Arch. Int. Pharmacodyn. 157: (1965).

WILLIAMS, R.L. AND PEARSON, J.E., JR.: Arch. Int. Pharmacodyn. 184: 195 (1970).