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RESEARCH REPORT

Report NO: CTL/R/391

1-76

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REPORT NO: CTL/R/391 721.11 Pq Toxicity

THE EFFECT OF ADMINISTRATION OF AN EMETIC (PP 796) ON PARAQUAT TOXICITY IN DOG AND MONKEY

11-76

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Summary

When paraquat was administered orally to dogs (20 or 30 mg of cation/kg body weight) or monkeys (100 mg of cation/kg body weight) together with a potent emetic (PP 796), the animals vomited. Dogs that vomited within the first hour of dosing survived an approximate LD75 dose of paraquat. Monkeys that vomited within the first hour of dosing survived an otherwise lethal dose of paraquat. The concentration of paraquat in the plasma of animals administered both paraquat and emetic was markedly lower than that of animals given paraquat alone. The toxicity to monkeys of paraquat formulated in the presence of an emetic dose of PP796 was estimated to be lowered by a factor of approximately 5.

INTRODUCTION

Death from paraquat poisoning has occurred in man as a consequence of both accidental and deliberate ingestion of paraquat formulations (Fletcher, 1974). The LD50 in man has been estimated to be approximately 30 mg of paraquat cation/kg body weight which is equivalent to a volume of 10-15 ml of the 20% (w/v) paraquat concentrate to an adult man.

The addition of stenching agents to the formulation is currently being carried out as a possible way of reducing the risk of accidental ingestion. However, in order to increase the chances of preventing accidents or suicidal ingestion, it would be desirable to reduce the toxicity of the formulation. The addition of an effective emetic agent might be one way of achieving this, but such an emetic would have to be a) potent following oral administration b) of low toxicity c) stable in a mixture with paraquat and d) effective as an emetic in the presence of paraquat.

An ICI Pharmaceuticals Division compound, ICI 63197 [2-amino-6-methyl-5-oxo-4-n-propyl-4,5-dihydro-s-triazolo(1,5-a)pyrimidone] which was developed as a phosphodiesterase inhibitor with anti-bronchoconstrictor effects, was shown after clinical trials in man to have a potent emetic action (Bayliss, 1973). This compound, which has been reclassified as PP796 by ICI Plant Protection Division, appears to have the necessary spectrum of properties required for an emetic that could be added to the paraquat formulation.

In the work to be described here, this emetic, together with paraquat has been administered to dogs and monkeys in order to assess the effect on paraquat absorption and toxicity.

METHODS

Animals:

Male cynomolgus monkeys (Macaca fasicularis), body weights in the range 3.5-5 kg were used. Male beagle dogs, body weights in the range 9-12 kg were used.

Experimental:

In the case of monkeys, the paraquat dosing solution consisted of the appropriate volume of "Gramoxone" (formulation no. JF1423/A) diluted into a total volume of 20 ml such that animals received 100 mg paraquat cation/kg body weight. This dosing solution also contained 10 g Complan (Glaxo-Farley Foods Ltd, Plymouth, Devon) in an attempt to reduce the irritancy of paraquat to the stomach. The emetic (PP796) when added was present such that the animals received 2mg/kg body weight. In the case of dogs, the total dosing volume was 50 ml containing 6 g "Complan" and the required volume of "Gramoxone" (formulation no. JF1423/B) such that animals received either 20 mg or 30 mg paraquat cation/kg body weight and either 2 or 3 mg of emetic/kg body weight.

Animals were dosed by stomach tube with paraquat either with emetic or without, and the animals were then observed and the time taken to vomit recorded.

Blood samples (approximately 5 ml) were taken at intervals from a jugular vein (dogs) or femoral vein (monkeys). Paraquat was measured in these samples using the gas chromatographic method of Draffen et al (1976).

Two experiments were carried out in each species, each experiment involving 8 animals.

The LD50 to monkeys of paraquat formulations containing wetters (10%) stench (1% pyridine bases) and an emetic dose of PP796 was determined by dosing undiluted formulations followed by 100 ml water via a stomach tube, using pairs of animals at each dose level.

RESULTS

Monkeys dosed with 100 mg of paraquat cation/kg body weight died within the first few days of dosing, despite vomiting between 3 and 5 hours of being dosed (Table 1). Most of the monkeys dosed with the same amount of paraquat containing PP796 vomited within an hour and these animals survived (Table 1).

The concentration of paraquat in the plasma of monkeys dosed with paraquat plus emetic was considerably less than that in the plasma of monkeys dosed with paraquat alone (Table 2).

None of the dogs dosed with either 20 or 30 mg of paraquat cation/kg body weight vomited and 3 out of a group of 4 died between 4 and 8 days after dosing (Table 3). All dogs dosed with these levels of paraquat containing PP796 vomited within one hour of dosing and survived (Table 3).

The concentration of paraquat in the plasma of dogs dosed with paraquat plus emetic was considerably less than that in the plasma of dogs dosed with paraquat alone (Table 4).

The LD50 of paraquat formulated with 10% wetting agent, 1% pyridine stench and an emetic dose of PP796 was in the range 250-500 mg/kg body weight in monkeys (Table 5).

DISCUSSION

PP796 [2-amino-6-methyl-5-oxo-4-n-propyl-4,5-dihydro-s-triazolo (1,5-a)pyrimidine] has been shown to have emetic properties in experimental animals (monkeys, dogs and pigs) and in man (Tables 1 and 3; Farrell, 1970; Davies and Hepworth, 1969; Broome, 1972; Bayliss, 1973).

2-3 mg of PP796/kg body weight is clearly an emetic dose to monkeys and dogs, usually causing vomiting within an hour of dosing (Tables 1 and 3) and this action does not appear to be affected by the presence of a large excess of paraquat (between a 15 and 50 fold excess).

As a consequence of vomiting, the absorption of paraquat is considerably reduced (Tables 2 and 4) and animals survive an otherwise lethal dose of paraquat.

The administration to monkeys of paraquat formulation (containing no added emetic) also resulted in vomiting after a delay of many hours (Table 1), but this did not lead to the survival of the animals. Thus, for emesis to be effective, it must occur within an hour of

administration of the paraquat.

The LD50 of paraquat to monkeys has been shown to be approximately 60-70 mg/kg body weight (Purser et al, 1975). The presence of an emetic dose of PP796 raises this value to 250-500 mg/kg body weight, thus reducing the toxicity of paraquat by a factor of approximately 5.

In conclusion, therefore, it is clear that the inclusion of an emetic dose of PP796 to a formulation of paraquat reduces the absorption of paraquat and thus significantly reduces the toxicity of the formulation.

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TABLE 1

*Survival of Monkeys

	Animal no.		Time to vomit (hours)	Time to death (days)	Survival
Expt. 1	63	paraquat	3.75	2	0/4
	95	paraquat	5	0.3	
	293	paraquat	6	1	
	295	paraquat	4.5	2	
	297	paraquat + emetic	0.9	Survived	3/4
	299	paraquat + emetic	0.5	Survived	
	301	paraquat + emetic	0.4	Survived	
	303	paraquat + emetic	7.5	14	
Expt. 2	313	paraquat	-	0.25	0/4
	315	paraquat	5	1	
	317	paraquat	5	1.3	
	319	paraquat	5	2	
	305	paraquat + emetic	0.3	Survived	3/4
	307	paraquat + emetic	4.75	2	
	309	paraquat + emetic	0.3	Survived	
	311	paraquat + emetic	0.4	Survived	

* This work was carried out at the Huntingdon Research Centre, Huntingdon, England under the supervision of Dr D Purser and will be described in detail in a subsidiary report.

TABLE 2

Monkeys	Hour	Plasma Paraquat Concentrations (micrograms/ml) Paraquat	Paraquat + Emetic
Expt. 1	1.0	8.6 ± 4.0 (4)	1.5 ± 0.5 (4)
	2.0	13.4 ± 8.6 (4)	1.1 ± 0.7 (4)
	3.0	15.0 ± 6.2 (4)	1.4 ± 1.2 (4)
	4.0	18.1 ± 13.7 (4)	1.6 ± 1.3 (4)
	6.0	12.8 ± 13.9 (4)	1.5 ± 1.0 (4)
	8.0	3.2 ± 2 (3)	1.4 ± 1.0 (4)
	10.0	3.2 ± 1.7 (3)	0.8 ± 0.6 (4)
Expt. 2	1.0	4.3 ± 1.5 (4)	1.5 ± 0.8 (4)
	2.0	8.0 ± 3.0 (4)	2.7 ± 2.3 (3)
	3.0	9.6 ± 6.0 (4)	1.0 ± 2.4 (4)
	4.0	7.5 ± 2.7 (3)	1.1 ± 0.9 (4)
	6.0	4.0 ± 0.4 (3)	0.9 ± 0.9 (4)
	8.0	2.3 ± 0.8 (3)	1.0 ± 1.4 (4)
	10.0	3.0 ± 2.7 (3)	0.8 ± 1.6 (4)

Values are expressed as means ± standard deviation with the number of determinations in parentheses.

TABLE 3

Survival of Dogs

	Animal no.		Time to vomit (hours)	Time to death (days)	Survival
Expt. 1	20	paraquat	-	4	1/4
	93	paraquat	-	4	
	990	paraquat	-	7	
	986	paraquat	-	Survived	
	86	paraquat + emetic	0.2 - 0.5	Survived	4/4
	936	paraquat + emetic	0.2 - 0.5	Survived	
	21	paraquat + emetic	0.2 - 0.5	Survived	
	95	paraquat + emetic	0.2 - 0.5	Survived	
Expt. 2	27	paraquat	-	8	1/4
	92	paraquat	-	6	
	611	paraquat	-	5	
	9	paraquat	-	Survived	
	28	paraquat + emetic	0.1 - 0.2	Survived	4/4
	51	paraquat + emetic	0.1 - 0.2	Survived	
	91	paraquat + emetic	0.1 - 0.2	Survived	
	626	paraquat + emetic	0.1 - 0.2	Survived	

TABLE 4

Dogs	Hour	Plasma Paraquat Concentrations (micrograms/ml) Paraquat	Paraquat + Emetic
Expt. 1	0.5	4.3 ± 3.4 (4)	-
	0.75	-	2.9 ± 3.0 (4)
	2.5	8.9 ± 2.8 (4)	2.4 ± 2.6 (4)
	5.0	1.9 ± 1.3 (4)	0.6 ± 0.5 (4)
	7.0	0.9 ± 0.6 (4)	0.5 ± 0.4 (4)
Expt. 2	0.5	1.3 ± 2.0 (4)	0.3 ± 0.2 (4)
	1.0	5.6 ± 4.4 (4)	0.3 ± 0.3 (4)
	2.0	8.3 ± 5.8 (4)	0.3 ± 0.4 (4)
	4.0	6.2 ± 3.3 (4)	0.2 ± 0.3 (4)
	6.0	2.8 ± 2.1 (4)	N D*

* ND = none detected

Values are expressed as means ± standard deviation with the number of determinations in parentheses.

TABLE 5

*Survival of monkeys given paraquat in a formulation containing an emetic dose of PP796

Dose:	Animal no.	Died	Survived	Total survival
100 mgPQ^{2+}/kg	351	-	✓	$^{2}/_{2}$
	353	-	✓	
250 mgPQ^{2+}/kg	363	-	✓	$^{2}/_{2}$
	365	-	✓	
500 mgPQ^{2+}/kg	377	✓	-	$^{0}/_{2}$
	379	✓	-	

* This work was carried out at the Huntingdon Research Centre, Huntingdon, England under the supervision of Dr D Purser and will be described in detail in a subsidiary report.

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