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91/414/EEC
REVIEW OF PARAQUAT DICHLORIDE
UNDER REGULATION 3600/92

PARAQUAT DICHLORIDE

DOCUMENT M-II, Section 3

TOXICOLOGICAL AND METABOLISM STUDIES

APPENDIX A: THE EMETIC PP796

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APPENDIX A

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1. **RATIONALE BEHIND THE ADDITION OF AN EMETIC AGENT TO FORMULATIONS OF PARAQUAT**

Over 30 years of practical use has demonstrated that paraquat does not present a risk to human health when handled and used according to normal agricultural practice.

Human poisonings have however occurred through oral ingestion of paraquat formulations. Efforts to reduce the incidence of poisonings and fatalities resulting from the accidental oral ingestion of paraquat formulations have been an on-going priority for ICI (now ZENECA).

Two complementary approaches have been taken towards the inclusion of formulation additives to reduce the incidence of poisoning resulting from the accidental oral ingestion of paraquat formulations. These formulation approaches are themselves complementary to additional company product stewardship initiatives which have included improvements in packaging and labelling and emphasis on farmer education and training.

The Use of Alerting Agents

The first approach was to ensure that paraquat formulations could not be mistaken for drinks suitable for human consumption. This has resulted in the addition of a distinctive smell and a colour. A stenching agent must not be too unpleasant for the legitimate user of the product but must be sufficient to clearly differentiate the product from beverages with which it might otherwise be confused. The stenching agent used in liquid paraquat formulations satisfies these requirements.

Aqueous formulations of paraquat are dark brown in the absence of a dye. The 'natural' colour of such formulations is not unlike that of certain common beverages such as tea, coffee or soft drinks. To clearly differentiate paraquat formulations from beverages a distinctive blue dye is added resulting in a blue/green liquid. Few human drinks (or foodstuffs) are blue in colour and the addition of both stench and dye together help limit any potential for confusion with drinks intended for human consumption.

The Use of a Potent Emetic

The second approach was to incorporate an emetic with the objective of inducing rapid emesis such that insufficient paraquat is retained in the gastrointestinal tract to cause poisoning following accidental oral ingestion (and secondarily, to alert the person or those in the vicinity that the product has been ingested in order to improve the likelihood of the person receiving rapid medical treatment).

The principal criteria for the selection of an appropriate (effective) emetic were as follows:

SPEED AND MODE OF ACTION

The emetic must produce a rapid vomiting response prior to the absorption of toxic amounts of paraquat. It should act centrally and should not produce its action via an irritant effect on the gastric system; irritancy could facilitate the absorption of paraquat.

SPECIFICITY

The agent must be able to act in the presence of paraquat, i.e. appropriate animal experiments must provide evidence that paraquat does not interfere with the mode or speed of action of the emetic.

HUMAN SAFETY

The emetic should be toxicologically acceptable and harmless to the user of the product in which it is incorporated.

ENVIRONMENTAL SAFETY

The emetic should not have a harmful effect on the environment.

STABILITY

The emetic must be stable in the presence of paraquat and vice versa. The emetic should not interfere with the herbicidal properties of paraquat.

An assessment was made of the following candidate emetic agents, most of which were excluded on the basis that they did not meet one or more of the necessary criteria.

Matricaria

This is the ground flower heads of the camomile plant. It is irritant in action, unreliable and, being insoluble in water, would be of no practical value for use in paraquat formulations.

Mustard

This is also highly irritant and insoluble.

Salts of heavy metals (eg. copper, antimony and zinc)

These agents have been firmly rejected by modern medical opinion because of their high toxic risk. They would also be unacceptable from an environmental perspective.

Sodium chloride

This emetic, commonly used in domestic poisoning incidents, is no longer generally recommended by the medical profession. Its efficacy is variable and a number of deaths have occurred following its use.

Apomorphine

This agent, although highly effective, can only be administered by intramuscular injection. It is unstable in air. It is therefore clearly of no practical value for inclusion in paraquat formulations.

Ipecacuanha

This agent is widely used as an orally administered emetic, it acts on the central nervous system but is also a gastric irritant. When administered in excessive amounts (approximately 10x the emetic dose) fatalities have occurred. This emetic was seriously considered by ICI for inclusion in paraquat formulations but a study in cynomolgus monkeys found that the response was unpredictable and only effective at doses associated with toxic symptoms.

2. THE EMETIC PP796**History**

PP796 is a triazolo-pyridine originally discovered by ICI Pharmaceuticals. It was extensively studied as a potential drug for the relief of asthma. Mammalian toxicology studies were completed to the satisfaction of the UK Committee for the Safety of Medicines which granted a Clinical Trials Certificate, enabling human clinical trials to take place. However during these trials it became clear that PP796 was of unexpectedly high emetic potency in humans.

(Bayliss, 1973)

It was therefore withdrawn from further development as a drug. Its emetic properties did, however, indicate considerable potential for use with paraquat formulations, and studies were therefore undertaken to establish whether PP796 could match the criteria established for a suitable emetic.

Mode of action

Following administration of an oral dose, PP796 is rapidly absorbed in mammals, peak plasma levels are observed in man within an hour of administration.

(Bayliss, 1973)

In species which vomit such as pig, dog, monkey and man (rodent species do not vomit) the rise in plasma level is associated with the onset of vomiting. This generally occurs within 15 minutes of dosing. Vomiting may be repeated four or five times within the first hour. Thereafter the effect ceases, probably as a consequence of the rapid metabolism and excretion of the compound. Evidence for the action of PP796 being centrally mediated is provided by:

- the rapid onset of vomiting and absence of irritant effects;
- the production of vomiting in dogs following intravenous administration at plasma levels similar to those producing the effect after oral administration.

(Case & Dunlop, 1977)

The proposed pharmacological rationale for the action of PP796 involves the inhibition of phosphodiesterase.

(Foulkes, 1978)

Dose required

PP796 is an extremely potent oral emetic, in a study to determine the vomiting response of cynomolgus monkeys (*Macaca fascicularis*) an oral dose of 2 mg PP796/kg bodyweight was sufficient to cause vomiting in five out of seven animals within 30 minutes of administration. The remaining two animals both vomited within approximately one hour of administration.

(Purser et al., 1978)

The emetic response of dogs to a range of single oral dose levels of PP796 (0, 0.1, 0.5, 1.0, 3.0, 10.0 or 20.0 mg/kg bodyweight) has been investigated. Emesis, with subdued behaviour, was produced at dose levels in the range 0.5 to 30 mg PP796/kg bodyweight, no effects occurred at 0.1 mg PP796/kg bodyweight. The onset of vomiting and the duration and severity of effects were all found to be dose-related. Vomiting occurred at 0.5 and 3 mg/kg bodyweight but was more rapid at 10 or 20 mg PP796/kg bodyweight (within five minutes of dosing). Severe vomiting occurred at dose levels of 3 mg PP796/kg bodyweight and above. Diarrhoea and defecation of mucus were present in some of the animals given 1 mg PP796/kg bodyweight and above. No other treatment-related effects were observed and all dogs had recovered within six hours of dosing.

The minimal effective dose level was considered to be 0.5 mg PP796/kg bodyweight, 20 mg PP976/kg bodyweight was considered to be the maximum tolerated dose (based on the severity of the clinical effects seen). The time interval between administration of PP796 and vomiting was reduced to within ten minutes by increasing the dose to 3 mg PP796/kg bodyweight and to within five minutes following doses of 10 or 20 mg PP796/kg bodyweight. However the reduction in the time taken to the initiation of vomiting was also associated with an increase in the severity of the clinical effects seen at these dose levels.

(Brammer & Robinson, 1986)

Efficacy in the presence of paraquat

The emetic action of PP796 is not impaired by the presence of paraquat. Investigations in cynomolgus monkeys (*Macaca fascicularis*) have demonstrated that the induction of vomiting by PP796 is unaffected by the presence of paraquat and that PP796 can reduce the toxicity of paraquat (Table 1).

TABLE 1: The Effect of PP796 on the Toxicity of Paraquat to the Cynomolgus Monkey

{PRIVATE }No. of Animals	Paraquat Dose PP796 Dose		No. Surviving
	(mg/kg bw)	(mg/kg bw)	
8	100	-	0
8	100	2	6

(Cobb et al., 1979)

A further study in cynomolgus monkeys (*Macaca fascicularis*) in which the doses of both paraquat and PP796 were increased has demonstrated that PP796 is capable of effectively producing an approximately three-fold increase in the LD₁₀₀ of paraquat to the cynomolgus monkey (Table 2).

TABLE 2: The Effect of PP796 on the Toxicity of Paraquat to the Cynomolgus Monkey

{PRIVATE }No. of Animals	Paraquat Dose PP796 Dose		No. Surviving
	(mg/kg bw)	(mg/kg bw)	
2	100	2	2
2	250	5	2
2	350	7	0
2	500	10	0

(Cobb et al., 1979)

In an additional study, PP796 was orally administered at 100 mg/kg bodyweight to ten male cynomolgus monkeys (*Macaca fascicularis*). Nine animals vomited between four and forty-three minutes after dosing; vomiting usually occurring twice within one hour of dosing. The remaining animal failed to vomit. Four of the ten animals (including the animal which failed to vomit) died, all within twenty-four hours of dosing.

(Purser et al., 1978)

The effects of different oral doses of PP796 have been investigated in dogs dosed orally with 20 mg paraquat/kg bodyweight. Groups of three male dogs received an oral dose of 20 mg paraquat/kg bodyweight plus 0, 0.5, 3.0 or 20 mg PP796/kg bodyweight.

The effects of paraquat administration were assessed as follows:

- measurement of peak plasma paraquat concentrations,

- measurement of the area under the plasma paraquat concentration/time curve (AUC) and,
- grossly observable paraquat-related lung lesions at necropsy eight days after dosing.

There was a marked decrease in the peak plasma paraquat concentration, the area under the curve (AUC) and the severity of paraquat-related lung lesions of dogs dosed with either 0.5 or 3.0 mg PP796/kg bodyweight plus paraquat when compared with dogs dosed paraquat alone. These reductions were dose-related. The response in dogs dosed 20 mg PP796/kg bodyweight plus paraquat was variable, some dogs showing a reduction in the effects of paraquat whilst others showed no decrease. One dog showed evidence of increased effects of paraquat, these were considered to be due to an increased amount of paraquat having been systemically absorbed as a result of inhalation of part of the dose following regurgitation.

The effective dose range of PP796 in dogs in terms of its action in reducing the amount of paraquat which is systemically absorbed following oral ingestion, and hence its toxic effects, is considered to be between 0.5 and 3.0 mg PP796/kg bodyweight. High doses of PP796 provide no advantages over a dose of 3.0 mg PP796/kg bodyweight and may, in some dogs, be contraindicated.

(Robinson & Brammer, 1986)

3. THE EFFECT OF PP796 ON THE TOXICITY OF PARAQUAT TO EXPERIMENTAL ANIMALS

3.1 THE EFFECT OF PP796 ON GASTRIC EMPTYING

PP796 is a potent inhibitor of gastric emptying as well as being a potent oral emetic. The inhibition of gastric emptying has been demonstrated in the mouse, rat and cynomolgus monkey. The results are presented in the following table. The animals were fasted for 24 hours and then dosed with either phenol red (monkeys) or radiolabelled chromium in the form of sodium chromate (mice and rats) in the presence or absence of PP796 (administered orally or by subcutaneous injection). The animals were sacrificed after one hour and the amount of phenol red or ⁵¹Cr remaining in the stomach was determined (Table 3).

TABLE 3: The Effects of PP796 on Gastric Emptying in the Mouse, Rat and Cynomolgus Monkey

{PRIVATE }Species	Treatments	% Remaining in the Stomach
Mouse*	<u>Oral</u>	
	Control	3.9 ± 0.65
	2.5 mg PP796/kg bw	44.8 ± 4.2
	<u>Subcutaneous</u>	
Rat*	Control	7.1 ± 1.88
	1.0 mg PP796/kg bw	49.4 ± 4.0
	<u>Oral</u>	
	1.0 mg PP796/kg bw	16.2 ± 1.8
Cynomolgus monkey **	<u>Subcutaneous</u>	62.4 ± 3.8
	1.0 mg PP796/kg bw	72.5 ± 2.3
	<u>Oral</u>	
	Control	33.7 ± 6.4
	0.2 mg PP796/kg bw	85.7 ± 4.7

* = non-vomiting species
** = the dose of PP796 used was 1/10th of the dose required to cause emesis and did not cause vomiting in any of the animals.

The mouse and the rat both lack the vomiting response, the effects of gastric emptying were therefore measured using doses causing vomiting in the dog and the monkey.

The inhibition of gastric emptying in monkeys was substantial, despite having been evaluated using a relatively low, sub-emetic dose.

(Wright *et al.*, 1979)

3.2 THE EFFECT OF PP796 ON THE SYSTEMIC ABSORPTION OF PARAQUAT

In rats, the administration of PP796 together with paraquat significantly delays the onset of the appearance of paraquat in the plasma and eliminates the early high peak plasma concentration which is otherwise seen in the first hour. This phenomenon results solely from the action of PP796 in inhibiting gastric emptying since the rat does not vomit. This finding therefore demonstrates that paraquat is not readily absorbed from the stomach. When PP796 and paraquat are administered to dogs and monkeys, as well as the inhibition of gastric emptying, vomiting occurs and concentrations of paraquat in the plasma are consequently much reduced.

(Wright *et al.*, 1979; Rose, 1978)

An analysis of the relationship between the concentration of paraquat present in plasma and mortality in humans reveals that those patients with paraquat blood plasma concentrations above a critical value (approximately 3 µg paraquat/ml, 24 hours after ingestion) have died. The survival of patients who accidentally (or deliberately) ingest paraquat is therefore critically dependent on the concentration of paraquat in the blood plasma during the first 24 hours after ingestion.

The inclusion of PP796 in paraquat formulations leads to a considerable reduction in the concentration of paraquat in the blood plasma of dogs and monkeys and should also therefore make paraquat formulations less orally toxic to humans.

3.3 THE EFFECT OF PP796 ON THE TOXICITY OF PARAQUAT TO DOGS AND MONKEYS

A formulation containing 200 g paraquat/litre ('Gramoxone') and 0.05% w/v PP796 has been demonstrated to be between two and five times less orally toxic than a similar non-emeticised formulation when administered to dogs. The median lethal dose of the formulation containing PP796 was approximately 0.5 ml formulation/kg bodyweight (approximately 100 mg paraquat/kg bodyweight), the median lethal dose of the formulation without the emetic was between 0.1 and 0.25 ml formulation/kg bodyweight (approximately 20 to 50 mg paraquat/kg bodyweight).

(Parkinson & Lefevre, 1977)

In a similar study carried out in the cynomolgus monkey (*Macaca fascicularis*) the presence of an emetic dose of PP796 raised the LD₅₀ of paraquat (previously determined as approximately 60 to 70 mg paraquat/kg bodyweight, Purser *et al.*, 1975) to between 250 and 500 mg paraquat/kg bodyweight, thus reducing the toxicity of paraquat approximately five-fold.

(Rose, 1976)

4. EVALUATION OF THE EFFECTIVENESS OF THE ADDITION OF PP796 TO PARAQUAT FORMULATIONS ON THE TOXICITY OF PARAQUAT TO HUMANS

4.1 ESTIMATION OF THE EMETIC DOSE REQUIRED TO INDUCE VOMITING IN HUMANS

When PP796 is included in paraquat formulations in amounts sufficient to cause emesis within one hour in dogs and in monkeys, the toxicity of the formulation is significantly reduced (see Section 3.3). In order to achieve a reduction in the toxicity of paraquat formulations to humans, it is necessary to add sufficient PP796 to cause emesis, in a volume of formulation concentrate that would normally be lethal if accidentally ingested.

Human data indicates that a volume of 10 ml of a non-emeticised formulation containing 200 g paraquat/litre ('Gramoxone') represents the minimum volume which is potentially lethal to humans. It has been concluded that a concentration of 5 mg PP796 in 10 ml of 'Gramoxone' (i.e. 0.05% w/v) should be added to the formulation in order to ensure that a person ingesting the lowest potentially lethal volume (10 ml) receives an effective dose of emetic. This dose is estimated to induce

vomiting within an hour in a person ingesting 10 ml or more of 'Gramoxone'.

(Rose, 1977)

4.2 ROLE OF PP796 IN THE REDUCTION OF FATALITIES ARISING FROM THE ACCIDENTAL ORAL INGESTION OF PARAQUAT FORMULATIONS

For obvious reasons it is not possible to carry out a study to specifically determine the effect of the addition of PP796 on the toxicity of paraquat formulations to humans. Epidemiological data, can however provide reliable information on the effectiveness of the emetic when such data is based on accurate records of cases involving paraquat poisoning. The UK National Poisons Information Service (NPIS) provides a reliable source of information on paraquat poisonings in the UK.

Published data from the NPIS based on UK incident data collected in the early 1980s suggested that the presence of PP796 in paraquat formulations may be associated with some reduction in mortality rates following the ingestion of paraquat formulations.

(Denduyts-Whitehead et al., 1985)

The UK NPIS data has been reviewed more recently with the following conclusions. Paraquat formulations containing PP796 cause both a more reliable and an increased incidence of vomiting within half an hour of ingestion than paraquat formulations without emetic. The addition of PP796 to paraquat formulations may therefore have some value in preventing serious poisoning when the amounts ingested are relatively small (< 25 ml 'Gramoxone'), i.e. in those cases involving accidental ingestion. However, in cases of deliberate oral ingestion with suicidal intent, circumstances in which relatively large (>25 ml 'Gramoxone') volumes are swallowed, a beneficial effect of the inclusion of the emetic on survival rates could not be demonstrated. This is presumably because even after emesis has occurred the amount of paraquat present in the gastrointestinal tract is still sufficient to result in a fatality.

Suicides, by whatever means are a tragic occurrence but for paraquat and other chemical means (agrochemicals, pharmaceuticals and other household chemicals) this represents gross abuse of a product which when used for its legitimate purpose brings significant benefits to society.

ZENECA Agrochemicals' experience in the UK and indeed, worldwide, is that the combination of preventive measures adopted, including the formulation additives of stench, dye and an effective emetic have resulted in a significantly decreased incidence of fatalities occurring as a result of the accidental oral ingestion of paraquat formulations.

(Calderbank, 1992)

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