O.S.A. Mohamed [1]

N.I. Eldirdiri [1]

M.A. Karrar [2]

S.E.I. Adam [2]

Toxicity of chlorpyrifos in Nubian goats

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L'administration par voie orale de chlorpyrifos à des chèvres de Nubie, aux doses de 1 200, 600 et 300 mg/kg, a occasionné des troubles nerveux et la mort au bout de 15 minutes à deux jours. Une dose orale de 150 mg/kg a été toxique mais non fatale pour les chèvres. Les animaux ayant reçu du chlorpyrifos, par voie orale, à des doses journalières de 300, 150 et 75 mg/kg, ont montré des signes de toxicité et sont morts après deux à sept jours de traitement. Les modifications pathologique, biochimique et hématologique sont décrites dans cette étude.
Mots clés : Toxicité - Chlorpyrifos - Chèvre - Soudan.

INTRODUCTION

Exposure to pesticides is a widespread hazard during field applications, in the factory and in other places where they are frequently used. Chlorpyrifos (Dursban 4EC®) is an organophosphate compound marketed in numerous formulations as plant or livestock insecticide.

The concentrations used and the frequency of application are dependent upon the insect involved. There are few references related to blood cholinesterase activity in chlorpyrifos-poisoned cattle (2, 5). In the Sudan, chlorpyrifos is used for control of pest complex on cotton fields in irrigated areas. Aerial application is very common and consequently pollution of water resources and contamination of vegetables are likely to occur and threaten both human and animal lives. The purpose of the present study was to assess the toxicity of chlorpyrifos in Nubian goats.

MATERIALS AND METHODS

Animal and dosing

Eighteen Nubian goats of both sexes 6 to 48-month old were used. The animals were housed in pens and fed on forage sorghum and water *ad libitum*.

1. Central Veterinary Research Laboratory, POB 8067, Khartoum, Sudan.

2. Department of Veterinary Medicine, Pharmacology and Toxicology, University of Khartoum, POB 32, Khartoum North, Sudan.

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Chlorpyrifos (0,0-diethyl-0-(3, 5, 6-trichloro-2-pyridyl) phosphorothionate) manufactured as Dursban® by Dow Chemicals Co., USA, was administered orally in single doses of 1200 mg/kg to goats 23, 24 (group 1), 600 mg/kg to goats 25, 26 (group 2), 300 mg/kg to goats 27, 28 (group 3) and 150 mg/kg to goats 29 and 30 (group 4). Oral doses of chlorpyrifos were given daily at the rate of 300 mg/kg per day to goats 31, 32, 33 (group 5), 150 mg/kg per day to goats 34, 35, 36 (group 6) and 75 mg/kg per day to goats 37 and 38 (group 7). Goats 39 and 40 (group 8) were untreated controls.

Blood sampling

All goats were bled from the jugular vein prior to the treatment and thereafter at appropriate intervals for serum analysis and haematology. Blood samples were allowed to clot, serum was separated and stored at - 20 °C until analysis.

Chemical methods

The concentrations of serum magnesium (9), calcium (10) and inorganic phosphate, sodium and potassium (11) were determined by standard methods. The concentrations of creatinine and urea and the activity of aspartate amino-transferase were estimated using commercial kits (Boehringer Mannheim GmbH Diagnostica, West Germany). The concentration of total protein was measured using a refractometer (Atago, Japan).

Haematological methods

The packed cell volume was measured in a microhaematocrit centrifuge. Haemoglobin concentration (Hb) was determined by the cyanmethaemoglobin technique. Red and white blood cells (RBC and WBC) were counted with a haemocytometer.

Differential leucocyte count was made by the battlement method (8). Mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) were calculated. Tissues were fixed in 10 % formol saline and paraffin sections were stained with haematoxylin and eosin (H & E).

Statistical analysis significance was assessed by Student's t-test (6).

O.S.A. Mohamed N.I. Eldirdiri M.A. Karrar S.E.I. Adam

RESULTS

The dosing schedule and time of death of chlorpyrifos-poisoned goats are given in table I.

TABLE I Details of goats given chlorpyrifos by drench.

Group	Goat no.	Age (months)	Sex	Oral dose of dursban (mg/kg)	Time of death
	23	8	M	1 200 (single dose)	15 minutes
	24	8	M	1 200 (single dose)	24 hours
2	25	48	F	600 (single dose)	24 hours
	26	48	F	600 (single dose)	24 hours
3	27	6	M	300 (single dose)	36 hours
	28	6	M	300 (single dose)	2 days
4	29	8	F	150 (single dose)	7 days (slaughtered)
	30	7	M	150 (single dose)	7 days (slaughtered)
5	31	9	F	300 (repeated doses)	2 days
	32	9	M	300 (repeated doses)	3 days
	33	9	M	300 (repeated doses)	3 days
6	34	7	M	150 (repeated doses)	4 days
	35	7	F	150 (repeated doses)	5 days
	36	7	M	150 (repeated doses)	5 days
7	37	6	M	75 (repeated doses)	6 days
	38	6	M	75 (repeated doses)	7 days

Clinical signs

Goats 23, 24 (group 1), 25, 26 (group 2), 27 and 28 (group 3), showed hyperexcitability, dyspnoea, profuse salivation, inappetance, lachrymation, bloat, staggering, tremors, paresis of the hind limbs, urination and defaecation, blood-tinged nasal discharge, clonic convulsions, recumbency and forward extension of the head and neck. These signs developed immediately after the treatment was applied and the goats died within 15 minutes to 48 hours. In goats 29 and 30 (group 4) receiving single doses of 150 mg/kg, the signs of toxicity developed immediately after the dosing and lasted for about 4 hours. These animals gradually regained appetite and were slaughtered on day 7. Goats 31, 32, 33 (group 5), 34, 35, 36 (group 6), 37 and 38 (group 7) showed signs of toxicity (photo 1) and death occurred on days 2, 3, 3, 4, 5, 5, 6 and 7, respectively. There were no clinical changes in control goats 39 and 40 (group 8).

Post mortem findings

Results are summarized in table II. In goats of groups 1, 2, 3 and 5, there was severe haemorrhage and congestion in the brain, meninges, trachea, lungs, heart, liver, kidneys, abomasum and intestine, catarrhal enteritis, pulmonary oedema, emphysema and cyanosis with fatty

Photo 1 : Goat 32 (group 5), orally treated with 300 mg/kg/day of chlorpyrifos for 3 days showing profuse salivation and dyspnoea.

change and necrosis of the liver and kidneys. These lesions were less marked in goats of groups 4, 6 and 7. Abomasitis and small abomasal erosions were seen in goats of groups 2, 4 and 6. The control goats 39 and 40 (group 8) showed no significant lesions.

Histological findings

There were varying degrees of haemorrhage in the hepatic parenchyma, renal cortex and medulla, pulmonary alveoli and cardiac muscle fibres of goats in groups 1, 2, 3, 4, 5 and 6. Cytoplasmic fatty vacuolation and necrosis of the centrilobular hepatocytes and pulmonary oedema and emphysema were observed in poisoned goats. The renal convoluted tubules contained acidophilic homogeneous material. The mucous membranes of the abomasum and small intestines showed inflammatory changes. The exudate consisted of mucus, desquamated epithelial cells, and leucocytes. In some places, however, the exudate contained erythrocytes. Aggregates of lymphocytes were seen in the liver, renal

TABLE II Summary of the post-mortem findings in chlorpyrifos-poisoned goats.

Site	Finding	Groups						
		1	2	3	4	5	6	7
Brain	Congestion	+ + +	+ + +	+ + +	+ +	+ + +	+ +	+
Trachea	Congestion, haemorrhage and frothing	+ + +	+ + +	+ +	+ +	+ +	+ +	+
Lungs	Congestion and haemorrhage	+ + +	+ + +	+ + +	+ +	+ + +	+ +	+
	Oedema	+ + +	+ + +	+ + +	+ + +	+ + +	+ + +	+
	Emphysema	+ + +	+ + +	+ + +	+ +	+ + +	+ +	+ +
	Cyanosis	+ + +	+ + +	+ + +	+ +	+ + +	+ +	+ +
Heart	Congestion and haemorrhage	+ +	+ +	+ +	+ +	+ +	+ +	+
Liver	Congestion and haemorrhage	+ +	+ + +	+ + +	+ +	+ + +	+ +	+
	Fatty change and necrosis	+	+ +	+ + +	+ +	+ + +	+ +	+ +
Kidneys	Congestion and haemorrhage	+	+ +	+ +	+ +	+ +	+ +	+
	Degeneration and necrosis	(−)	+ +	+ +	+ +	+ +	+ +	+ +
Abomasum	Congestion and haemorrhage	+	+ +	+ +	+	+	+	+
	Abomasitis and erosions	(−)	+ +	(−)	+	(−)	+	(−)
Intestines	Congestion and haemorrhage	+ +	+ + +	+ + +	+ +	+ + +	+ +	+ +
	Enteritis	+	+ +	+ + +	+ +	+ + +	+ +	+ +

+ ... + + + : Increasing severity of lesions. (−) Absence of lesions.

cortex, *lamina propria* of both abomasum and small intestines and around the bronchioles. In goats of group 7, the lesions were less marked and there was no damage of the abomasal mucosa.

Changes in serum constituents

There were no signficant changes in the concentrations of magnesium, calcium, inorganic phosphate, sodium and creatinine in the serum of any goat. There were significant differences ($P < 0.05 - 0.001$) in the concentrations of urea, GOT and total protein between the control group and test groups (table III). The concentration of serum potassium tended to decrease after treatment with chlorpyrifos and significant difference ($P < 0.01 - 0.001$) was recorded for groups 5 and 7 when compared with the control group.

TABLE III Changes in the concentrations of some serum constituents in chlorpyrifos-poisoned goats.

Group	GOT (iu)	Total protein (g/100 ml)	Urea (mg/100 ml)	Potassium (mg/100 ml)
4	35.67 ± 18.64 P < 0.05	4.78 ± 0.75 P < 0.05	36 ± 19.17 P < 0.05	10.8 ± 0.93 NS
5	52.83 ± 8.61 P < 0.001	4.37 ± 0.65 P < 0.01	86.67 ± 10.8 P < 0.001	8.13 ± 0.54 P < 0.001
6	59.5 ± 7.29 P < 0.001	4.3 ± 0.37 P < 0.01	65.83 ± 17.15 P < 0.001	9.7 ± 0.65 NS
7	48.67 ± 7.58 P < 0.001	5.17 ± 0.26 P < 0.05	67.5 ± 25.23 P < 0.001	8.95 ± 0.61 P < 0.01
8	19.13 + 2.39	5.65 + 0.43	17.29 + 0.98	10.28 + 0.62

Group 4 = chlorpyrifos at 150 mg/kg ; group 5 = 300 mg/kg/day ; group 6 = 150 mg/kg/day ; group 7 = 75 mg/kg/day ; group 8 = control. NS : not significant. P : level of significance above or below control values.

Haematological changes

There were significant increases ($P < 0.05 - 0.001$) in the values of Hb, PCV and RBC between the control and test goats at days 2 and 4. MCH and MCHC values did not change. MCV values increased and leucocytosis occurring on days 2 and 4 was due to an increase in the number of neutrophils.

DISCUSSION

The results o. this investigation show that chlorpyrifos is toxic and fatal to goats at single doses of 300 mg/kg and above. The daily oral administration of chlorpyrifos to goats at doses of 150 and 75 mg/kg per day caused toxic manifestations and death between days 4 and 7.

The clinical, biochemical and pathological data were indicative of effects on the central nervous system, liver, kidneys, lungs and gastrointestinal tract. The main clinical manifestations were restlessness, grinding of the teeth, hypersalivation, dyspnoea, clonic convulsions, diarrhoea,

bloat, incoordination of movements, ataxia and collapse. Similar clinical effects have been produced by poisoning of sheep and calves with other members of organophosphorous compounds (1, 3). The development of nervous signs might have been due to the stimulation of the nervous system with the concomitant inhibition of cholinesterase activity. It is known that chlorpyrifos is a cholinesterase inhibitor (4, 5, 7).

The pathological changes in the brain and meninges of goats poisoned by chlorpyrifos were slight and none of the animals showed demyelination of the peripheral nerves and white matter of the spinal cord.

The lesions in the liver of chlorpyrifos-poisoned goats consisted of congestion, haemorrhage, fatty changes and centrilobular necrosis. The renal tubules were degenerated and/or necrotic. The increase in the activity of GOT and in the concentration of urea and decrease in the concentration of total protein in serum add further evidence of hepatorenal damage.

The decrease in the concentration of serum potassium suggests an alternative route of excretion. It is possible that potassium ion might have been excreted in the profuse saliva or it might have passed via saliva into the rumen with eventual excretion in the faeces.

The pulmonary lesions including cyanosis, congestion, haemorrhage and oedema probably contributed to the development of dyspnoea in the experimental animals. Diarrhoea may be a consequence of the muscarinic effect or catarrhal abomasitis and enteritis or both. The widespread haemorrhage is attributed to an endotheliotoxic action.

The increase in Hb, PCV and RBC values was probably due to haemoconcentration.

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A single oral administration of chlorpyrifos to Nubian goats at 1200, 600 and 300 mg/kg caused nervous signs and death within 15 minutes to 2 days of treatment. An oral dose of the compound at 150 mg/kg was toxic, but not fatal to goats. Animals given chlorpyrifos orally at daily doses of 300, 150 and 75 mg/kg showed signs of toxicity and died within 2 to 7 days of treatment. Pathological, biochemical and haematological changes are described. *Key words* : Toxicity - Chlorpyrifos - Goats - the Sudan.

MOHAMED (O.S.A.), ELDIRDIRI (N.I.), KARRAR (M.A.), ADAM (S.E.I.). Toxicidad del clorpirifos en cabras de Nubia. *Revue Élev. Méd. vét. Pays trop.*, 1990, **43** (4) : 431-434

Dosis de 1 200, 600 y 300 mg de clorpirifos admínistradas por vía oral provocaron trastornos nerviosos y la muerte al cabro de 15 minutos a dos días. Una dosis de 150 mg/kg fué tóxica pero fatal para las cabras. Los animales tratados por vía oral con dosis diarias de 300, 150 y 75 mg/kg de clorpirifos mostraton síntomas de toxicidad y murieron después de dos a siete días de tratamiento. Se describen las modificaciones patologica, bioquímica y hematológica. *Palabras claves* : Cabra - Clorpirifos - Toxicidad - Sudán.

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