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Lindane Toxicity to Four-Month-Old Calves

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Lindane (the gamma isomer of 1,2,3,4,5,6-hexachlorocyclohexane) was first recorded to be the most toxic of five isomers by Slade (1945) when he reported on oral LD_{50} of 190 mg/kg for rats. An extensive review on its toxicity by Herbst and Bodenstein (1972) revealed that domestic animals were more sensitive than rats to both oral and dermal dosages. Lindane has been recommended and used in Ontario for many years for the control of biting lice (*Bovicola bovis*) and sucking lice (*Haematopinus sp*) on beef but not dairy cattle and on animals over three months old (OMAF 1970-81). In the last year two cases of toxicity have been investigated. In one, adverse effects were observed on 15 calves aged 3-6 months in which two died.[1] The second case reported here involved a case history on two calves which were maliciously dosed with lindane and died.

MATERIALS AND METHODS

The investigation into the deaths of two holstein calves on a dairy farm in Middlesex County, Ontario, revealed that a 10% emulsifiable concentrate of lindane had been administered orally. The two calves, aged 16 and 18 weeks weighed respectively 85 and 82 kg, were housed indoors and fed a regular grain concentrate with hay *ad lib* (Table 1). The total quantity administered to the two calves was determined.

A post mortem was conducted on each of the two calves and weights and samples of alimentary tract contents and organs and tissues were collected for analysis. Plant and animal tissues (50 g each) were extracted by blending at high speed with 250 ml acetonitrile:water (2:1 vol /vol). The extracts were filtered and placed in a freezer at -10C until water and the bulk of the extracted lipids were "frozen out". Urine (50 ml) was extracted with acetonitrile by shaking manually. A measured aliquot of the acetonitrile extract was then partitioned with hexane according to procedures described in Analytical Methods for Pesticide Residues in Food (1973). Cleanup and isolation of lindane by adsorption chromatography was carried out according to the method of Mills *et al.* (1972).

[1] Personal communications from laboratory records

Lindane residues were determined by gas-liquid chromatography under the following conditions:

Column: 1.8m x 2mm i.d. glass packed with 2% OV-1/3% OV-210 on 80/100 mesh Gas Chrom Q
Detector: electron capture, ^{63}Ni source, constant current operation
Carrier gas: nitrogen at 60 ml/min
Temperatures: column, 150C; injector, 240C; detector, 375C
The limit of detection was 0.01 mg/kg

RESULTS AND DISCUSSION

Calf 'A' went into convulsions almost immediately after ingestion, collapsed and died within one hour (Table 1). The post mortem on this animal revealed no chemical changes. Calf 'B' developed convulsions and collapsed within 10 hours of ingestion. Convulsions became progressively more severe and the condition of the calf deteriorated until death occurred after 50 hours. Post mortem of the animal revealed that the esophagus anterior to the thorax was congested and the left lung was partially atelectate. The rumen contents were drier than normal and the urine was a dark amber color. The animal had not eaten since the administration of the dose.

Table 1. Details on two calves poisoned by lindane (10% emulsifiable concentrate)

Item	Calf 'A'	Calf 'B'
Age (weeks)	16	18
Sex	female	female
Breed	Holstein	Holstein
Death (hrs)	1	50
Weight at death (kg)	85	82
Main symptoms	convulsions	convulsions

It was determined that 3.6 g of lindane a.i. had been removed from the bottle and administered to the two calves. The analyses of samples taken from the two calves appear in Table 2. Since calf 'A' died so rapidly following ingestion it was concluded that the tissue analyses would reveal the approximate amount ingested. The rumen, reticulum, omasum and abomasum contents contained 2136 mg while the body tissues and organs sampled contained 119 mg and the urine 47 mg. This gave a total of 2302 mg of lindane. It was assumed, therefore, the calf ingested between 2300-2500 mg leaving between 1100 and 1300 mg ingested by calf 'B'. Calf 'A' died with a brain level of 15 mg/kg lindane or a total of 9 mg being present in that organ. Assuming the ingestion was between 2.3 and 2.5 g of lindane then the lethal dose for calf 'A' was between 27 and 29 mg/kg b.w.

Calf 'B' would appear to have ingested between 1.1 and 1.3 g. However, the amount found in the four stomachs amounted to only 12.1 mg and the total amount found in all parts of the body only amounted to 73.4 mg. This calf survived for 50 hours before

Table 2. Distribution of lindane in the calves at post mortem

Body Part	Calf 'A' (dose)			Calf 'B' (dose)		
	Weight (kg)	Lindane		Weight (kg)	Lindane	
		Conc. (mg/kg)	Total (mg)		Conc. (mg/kg)	Total (mg)
Rumen Contents	6.5	140	910	5.5	1.1	6.1
Reticulum & Omasum Con.	1.5	235	352	1.0	1.6	1.6
Abomasum Contents	2.3	380	874	2.1	2.1	4.4
Liver	1.7	27	46	1.7	0.6	1.0
Heart	0.6	37	22	0.6	0.6	0.4
Kidney	1.1	14	15	1.0	2.0	2.0
Muscle & Abdominal Fat	4.1	6.6	27	3.0	18	54.0
Brain	0.6	15	9	0.6	0.04	0.03
Urine (from bladder)	0.7	67	47	0.6	6.5	3.9
Total	19.5		2302	16.1		73.4
Whole Body	85		2300 to 2500			1100 to 1300

dying. However, the contents of the rumen and abomasum were dehydrated forming a mass that had moved very little. Lindane storage in the fat rose to 18 mg/kg by the time of death, the highest concentration of any tissue while the brain level was only 0.04 mg/kg, the lowest level of the tissues analyzed. Of the 1.1 to 1.3 g of lindane assumed to have been ingested, only 6% was recovered. The missing lindane could have been excreted in the urine, excreted in the feces and/or metabolized. In the 50 hrs the calf survived it could have excreted 2 to 3 liters of urine. However, the removal in the urine could only have accounted for a portion of this loss even if the concentration had risen to several hundred milligrams per liter based on the urine level of 67 mg/L in the calf that died after one hour and the 6.5 mg/kg in the urine of the calf dying after 50 hours. Since there appeared to be little movement of food through the alimentary canal, loss via the feces would appear to have been small. This leaves degradation as the major route for disappearance of the ingested lindane. Lindane could have been metabolized by the scheme outlined by Grover and Sims (1965).

In spite of the apparent removal of the major portion of lindane from the internal organs, the animal nevertheless collapsed in 10 hours and died in 50 hours. Based on the assumed ingestion of 1.1 to 1.3 g of lindane, a lethal dose of 13 to 16 mg/kg b.w. could be calculated.

Radeleff *et al.* (1955) reported on 1 to 2 week old Jersey calves that tolerated oral doses of 2.5 mg/kg b.w. (9 dosed, 9 unaffected), were affected by 5 mg/kg of b.w. (3 dosed, 2 died, 1 affected severely but recovered) and 4 of 5 died (1 unaffected) after consuming 15 mg/kg b.w. A one year old Hereford animal was unaffected by 10 mg/kg b.w. but a second died after receiving a single dose of 25 mg/kg. In a later document Radeleff and Bushland (1960) reported the maximum non-toxic oral dose in 1-2 week old calves was 2.5 mg/kg and the minimum toxic dose was 5.0 mg/kg b.w. The calves were considerably older than most of the reported intoxications indicating little increase in tolerance with age.

Two Holstein calves died within 1 and 50 hours after ingesting 3.6 g of lindane. The mean lethal dose was 22 mg/kg b.w. with a range of 13 to 29 mg/kg b.w. The calf dying in 1 hour had a brain level of 15 mg/kg. The calf dying in 50 hours appeared to have removed lindane from essential organs to relatively low levels but still succumbed.

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