

Lindane Toxicity to One Year Old Calves

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Lindane (the gamma isomer of 1,2, 3, 4, 5, 6 hexachlorocyclohexane) is still recommended and used as seed treatment and for control of pests especially against lice and ticks on sheep and cattle (but not on dairy cattle and laying hens).

Unlike others organochlorine pesticides, lindane does not persist in the environment or in living animals. Multitest data have established that it is not a highly toxic pesticide. About domestic animals toxicity, no exact data is available and LD50 is not exactly known. Values obtained after an accidental intoxication allow us to have some data on toxicity levels and on toxic origin symptoms.

During the last year, one case of intoxication of calves was investigated on 30 calves (sprayed against ectoparasite with lindane) in which 5 died. Results are reported in the present work.

MATERIALS AND METHODS.

During february 1989, "Departmental Veterinary Direction" called the laboratory about intoxication of 30 one year old Charolais calves (300 kg). The investigation into the death revealed that this intoxication has been induced by an accidental dermal application with approximatively 10g of lindane per calf. Treatment was rapidly followed by classical signs of chlorinated hydrocarbon poisoning such as muscular twitching, incoordination and important salivation. The day after, 2 calves died and two others during the following week. The laboratory was not contacted in the beginning but 17 days after intoxication and lindane déterminations in blood and fat were investigated. Five months later, one calf died and tissues and organs were collected for analysis.

Lindane was extracted from 30g of tissues and organs by using the method previously described by Venant and al. (1982). After fat extraction with 50ml hexane, 1g of melted fat was added to 6ml of a mixture acetonitrile-methylene chloride (75-25 V/V). Samples were mixed for 1 minute and then centrifuged at 2000 rpm for 10 minutes at a temperature of -10°C. Supernatants were transferred to a flask and extraction was repeated once more. Supernatants were evaporated to dryness and clean

up was carried out on a 5g florisil (deactivated with 2.5% water by weight) and 50 ml eluent (hexane - methylene chloride 80-20 V/V). Extracts were evaporated to 1-2 ml under reduced pressure at 30°C. Sample was then concentrated to dryness by evaporation using a stream of air. From blood samples, collected into heparinized vacutainers, plasma (5ml) was shaken with 20 ml hexane. Hexane was collected and extraction with 10 ml hexane was repeated once more. Organic phases were concentrated until 5 ml.

Lindane analysis was performed on a girdel 3000 and a varian 6000 serie vista gas liquid chromatograph equipped with an electron capture detector (Ni63). The girdel 3000 GC conditions were : column : 5% Dow II on 80-100 mesh chromosorb WAWDMCS (1.80 m long x 4 mm interne diameter) ; temperature : 180°C ; carrier gas flow (N₂) : 30 ml/min. The varian 6000 serie vista GC conditions were : column : DB608 wide bore capillary column (30 m x 0.53 mm id) ; programmed temperature : from 140°C to 250°C at 10°C min⁻¹ ; carrier gas (N₂) flow : 4 ml/min.

RESULTS AND DISCUSSION

This study was conducted on three calves (n° 1, 2 and 3) and during five months until lindane concentration in plasma was below determination limit (0.1 µg/l). At this time, 13 blood samples were collected to obtain larger results and 5 fat samples, taken by biopsy, were also analysed.

It was very difficult to obtain fat because animals were very skinny, they lost weight after intoxication and could not eat normal diet during the first four months after intoxication.

All results of fat and blood obtained from calf n° 1, 2 and 3 are shown in table 1. Lindane can be detected in plasma at very high levels at day J+17 (17 days after intoxication), at day J+26 and are approximatively 100 µg/l. The rate of disappearance of lindane from blood was rapid, plasma concentration being reduced by 70% between days 17 and 56 and by 90% by day 70.

Mean lindane concentrations obtained from plasma samples (calf n° 1, 2 and 3) are plotted on a curve shown in figure 1. No statistical study was performed but raw results were very close and it was able to obtain mean values. Statistical approach shows that results were on a linear regression curve (r value is 0.97) with $\beta = 0.05$. The rate of elimination from plasma was similar in all three calves. The elimination half-life was calculated for each animal and gave results between 13.3 days and 16.3 days with a mean half-life time of 14 days.

During this experiment, five analysis on fat were undertaken. Lindane concentrations found in fat samples are very high at day J+17 (19.10⁻³ µg/kg) showing a rapid accumulation of this compound. Ratio between plasma and fat is 0.33% for calf n° 3, 0.6% for calf n° 6 and less than 0.05% for calves n° 7 and 10 (Dargorn and al. - 1987 found 0.4% for sheep). Regarding fat samples, the elimination half-life is 11 days and β is 0.063.

Table 1 : Lindane concentrations in fat and plasma
Results in µg/l in plasma and in µg/kg in fat.

Calves number	1	2	3	6	4 and 16	7	10	13 and 15	11	17	26
Days after intoxication											
17	100	160	130								
28	150	100	150								
46	75	63	60 (19x10 ³ µg/kg in fat)								
56	33	31	25								
63	17	16	11								
70	12	10	6								
77	7	8	4								
92	2	2	2								
105	4	3	4								
126	2	0.7	0.7 (230µg/ kg in fat)	0.7 110µg/ kg in fat) 0.5	0.1	0.1 (220µg/ kg in fat	0.1 (370µg/ kg in fat	0.1	0.2	0.5	0.4
161	0.5	0.1	0.1								

Detection limit in plasma : < 0.1 µg/l.

Detection limit in fat : < 0.001 µg/kg.

Table 2 : Distribution of lindane in calf which died on july 17 th.

Organs	Results in µg/kg b.w	Results in µg/kg on fat basis
Visceral fat	-	188
Bladder	-	247
Pancréas	12	177
Intestine	-	226
Thyroid	20	204
Liver	2	58
Lung	1	93
Spleen	6	124
Heart	2	100
Muscle	2	138
Heart Fat	-	427
Plasma	10	-
Kidney	1	61
Kidney fat	-	140
Thorax fat	-	130
Brain	2	22
Thymus	33	166
Salivary gland	81	192

Five months after lindane poisoning one calf died and specimens of different organs were submitted to the laboratory for analysis. Results are included in table 2. Concentration found in liver, lung, kidney and brain are very low comparing with those obtained in fatty organs such as bladder, salivary gland, thyroid, intestine, visceral fat and heart fat according with results given by Frank R. (1984).

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