

CASE REPORT

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Toxicologic Studies in a Fatal Overdose of 2,4-D, Mecoprop, and Dicamba

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ABSTRACT: A suicidal poisoning committed by a 61-year-old woman, who ingested an unknown quantity of Killex[®], containing in aqueous solution 100 g/L of (2,4-dichlorophenoxy)acetic acid (2,4-D), 50 g/L of mecoprop, and 9 g/L of dicamba as amine salts is described. Quantitation of chlorophenoxy acids was performed by extraction from an acidified mixture and concentration before high performance liquid chromatography analysis. All three herbicides were separated in a phosphate buffer/acetonitrile mixture at 280 nm on a RP-8 column. Concentrations of herbicides found were: in blood—520-mg/L 2,4-D, 530-mg/L mecoprop, and 170-mg/L dicamba; in urine—670-mg/L 2,4-D and 520-mg/L mecoprop; in bile—340-mg/L 2,4-D, 530-mg/L mecoprop, and 140-mg/L dicamba; and in liver—540-mg/Kg 2,4-D, 500-mg/Kg mecoprop, and < 100-mg/Kg dicamba. Liquid chromatography was found to be a reliable method for herbicide quantitation in biological tissues and fluids. The technique offered definite advantages over ultraviolet spectrophotometry and avoids the derivatization requirement for gas chromatography.

KEYWORDS: toxicology, poisons, suicide, chromatographic analysis

The chlorinated phenoxyacid derivatives (2,4,5-trichlorophenoxy)acetic acid (2,4,5-T) and (2,4-dichlorophenoxy)acetic acid (2,4-D) have been used extensively as herbicides over the past 30 years. Various chlorinated herbicides are often combined in commercial formulations as amine salts or esters [1].

Since their introduction, several reports in the literature document the pharmacokinetic and toxic concentrations of one or more chlorophenoxyacetic acid derivatives in fatal and non-fatal suicide attempts [2-8].

Previous toxicologic analysis of 2,4-D, 2,4,5-T, and other chlorinated phenoxy herbicides were generally performed using ultraviolet spectrophotometry at 282 nm or by gas chromatography after derivatization using flame ionization or electron capture detection.

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This report gives a detailed case history of a fatal overdose of Killex® contained in an aqueous solution of 100 g/L of (2,4-dichlorophenoxy)acetic acid (2,4-D), 50 g/L of mecoprop [2-(4-chloro-2-methylphenoxy) propionic acid], and 9 g/L of dicamba (3,6-dichloro-2-methoxybenzoic acid).

Analysis of body fluids and liver was by liquid chromatography with ultraviolet detection at 280 nm.

Case History

A 61-year-old female with a history of mental illness and chronic alcohol abuse was brought to the hospital at 8:00 a.m. Her son-in-law found her 1 h earlier, shortly after she ingested an unknown quantity of Killex (a mixture of 2,4-D, 100 g/L; mecoprop, 50 g/L; and dicamba, 9 g/L).

On initial examination, the woman was comatose and distressed with a blood pressure of 50/unobtainable, and a peculiar odor on her breath. She was breathing heavily and had marked distension of the abdomen.

Her laboratory results on admission were: blood pH 7.25, PCO₂ 38, PO₂ 63, and bicarbonate 17. All other blood chemical indices were within reference range limits except for a slight elevation in aspartate aminotransferase, 36 U/L (8 to 29 U/L). Hematological assays were all normal except for an elevated white blood count, 19.3 by 10⁹/L (4.5 to 10.5 by 10⁹/L). Routine urinalysis was normal. Microscopic examination showed an elevated white cell count of 10 to 20/high power field. Toxicological screening of blood and urine was negative for alcohols, acid, and neutral and basic drugs except for a trace of salicylates in the urine.

Gastric lavage was performed and 50 g of activated charcoal was administered. In the following 3 h, the patient vomited several times while her clinical course deteriorated. Her blood pressure was unrecordable at 12 p.m. Cardiopulmonary resuscitation was performed for 25 min. She was pronounced dead at 12:25 p.m.

Autopsy findings were nonspecific except for the odor of Killex mixture and a large volume of blood in the left pleural cavity. No blood was found in the gastrointestinal tract.

Toxicologic Analysis

High Performance Liquid Chromatography (HPLC)

Liquid chromatography was performed on a model 740 solvent delivery system by Spectra Physics, SF 770 variable wavelength detector by Schoeffel Instruments, and an Omniscribe recorder by Houston Instruments (all obtained from Technical Marketing Associates, Halifax, Nova Scotia). Analysis was performed at ambient temperature using a 250- by 4.6-mm RP-8 column with 5- μ m particle size (Brownlee Labs, Santa Clara, CA). Detector wavelength was set at 280 nm.

A buffer solution consisting of 0.01 mol/L of potassium dihydrogen phosphate adjusted to pH 3.2 with phosphoric acid was mixed with acetonitrile and *n*-nonylamine to form the mobile phase (550/450/0.6). The flow rate was 1.6 mL/min.

Osterloh recently reported [9] a fatal overdose of 2,4-D and mecoprop with analysis by HPLC. In their system however, mecoprop and the internal standard 2(2,4-dichlorophenoxy) propionic acid were not well resolved (retention times of 8.53 and 9.16 min, respectively).

Standards and Reagents

2,4-D, mecoprop, and dicamba as free acids were obtained from U.S. Environmental Protection Agency, Research Triangle Park, NC. Silvex [2-(2,4,5-trichlorophenoxy) propionic acid] was purchased from Polyscience Corp. Niles, IL.

A 100-mg/L solution of the internal standard was dissolved in methanol and diluted to volume with water.

Acetonitrile and methanol were HPLC grade and glass distilled (Caledon Laboratories Ltd., Georgetown, Ontario).

Analysis of Biological Fluids and Tissue

Specimens of blood, urine, bile, and blank blood (for standards) were diluted 1/10 with water. Ten grams of liver tissue was homogenized in 10 mL of water with a Brinkmann Polytron homogenizer. This mixture was diluted to a volume of 100 mL with water. Aliquots (1 mL) of the homogenate and diluted blood, urine, and bile were analyzed as follows: 1 mL of fluid was mixed with internal standard (1 mL), 1M hydrochloric acid (1 mL), and chloroform (5 mL). After mixing and centrifugation, the organic layer was evaporated under nitrogen in a hot water bath and reconstituted in 150 μ L of methanol. Ten microlitres were injected into the chromatograph.

Urine and liver specimens were not hydrolyzed since at least 77% of 2,4-D is excreted unchanged in the urine [10].

Quantitation of 2,4-D, mecoprop, and dicamba was determined by peak height ratios with the internal standard using aqueous standards, blank blood and liver standards, and standard addition for liver and blood specimens.

Urine and bile specimens were quantitated using aqueous standards. The comparative results for blood and liver by the three standardization methods agreed within 8%. Dicamba was not quantitated in urine because of an interfering peak on the chromatogram. Absolute recovery of all three herbicides from blank blood ranged from 90 to 95%.

Quantitation of 2,4-D was linear from 0.5 to 800 mg/L, mecoprop from 0.5 to 800 mg/L, and dicamba from 100 to 500 mg/L.

Results of toxicologic analysis are presented in Table 1. The chromatogram for the blood specimen is shown in Fig. 1.

Discussion

Animals killed by massive doses of 2,4-D are believed to die of ventricular fibrillation. At lower doses, various signs of neuromuscular involvement including stiffness, ataxia, paralysis, and eventually coma [8] are observed.

Reported lethal dose (LD₅₀) values for chlorophenoxy herbicides vary widely but range from 300 to 1000 mg/Kg. The relative toxicities of the Killex components (LD₅₀) are: 2,4-D, 750; mecoprop, 930; and dicamba, 1028 mg/Kg [11].

From literature reports [2,8], clinical symptomatology in herbicide ingestion is variable. The marked hypotension on initial examination is not a common finding in chlorophenoxy herbicide intoxications. Ingredients such as solvents and emulsifiers used in the various commercial formulations may have significant toxic effects of their own. In addition, the physical health and access to immediate medical treatment may help explain why one individual reached a concentration of 1030 mg/L (2,4-D) in serum and survived [12].

Total concentration of chlorinated herbicides in blood of fatalities have ranged from 58 to 1010 mg/L [2,3,13]. The total blood concentration of 2,4-D, mecoprop, and dicamba in this case was 1220 mg/L.

TABLE 1—Summary of toxicological findings.

Substance	Specimen Concentrations, mg/L or mg/Kg			
	Blood	Urine	Bile	Liver
2,4-D	520	670	340	540
Mecoprop	530	520	530	500
Dicamba	170	...	140	< 100

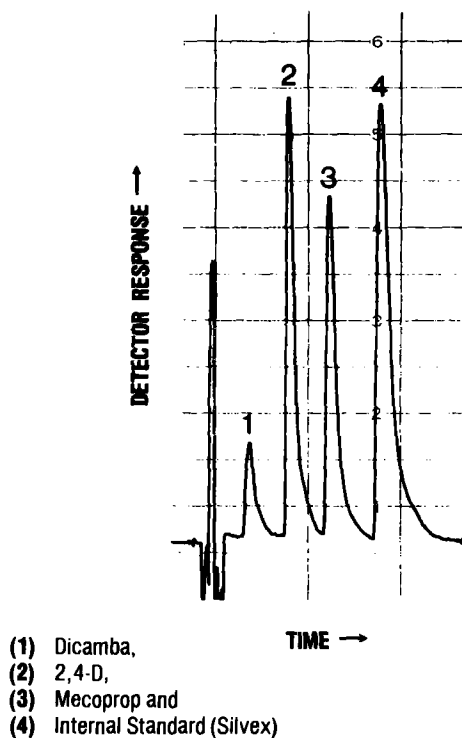


FIG. 1—High performance liquid chromatogram of blood extract: (1) dicamba 170 mg/L, (2) 2,4-D 520 mg/L, (3) mecoprop 530 mg/L, and internal standard Silvex (4).

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