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Metabolic Fate of the Herbicide Dicamba in a Lactating Cow

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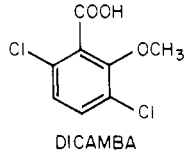
The herbicide dicamba (Banvel, 3,6-dichloro-2-methoxybenzoic acid) was rapidly absorbed, only partly metabolized, and rapidly excreted by a lactating cow after oral treatment during a 5-day period at a dose equivalent to 2.2 mg kg^{-1} day^{-1} or about 60 ppm of dietary dicamba. Within 6 h after the final [^{14}C]dicamba dose, about 89% of the administered ^{14}C had been excreted in the urine, <2% in the feces, and about 0.02% in the milk. The excreted radiocarbon was primarily unchanged dicamba (about 80%), but the compound was metabolized by aryl-O-demethylation to 3,6-dichloro-2-hydroxybenzoic acid (DCHBA), which was the major metabolite in urine and in extracts of feces and tissues and was the only radioactive component extracted from milk. Minor metabolites identified in urine were 2,5-dichlorophenol and the glucuronide conjugate of DCHBA. Results in these studies suggest that low-level exposure of ruminants to dicamba through contaminated feed, forage, or water will not result in appreciable retention of residues by edible tissues or their secretion into milk.

The herbicide dicamba (Banvel, 3,6-dichloro-2-methoxybenzoic acid) is widely used for control of many



DICAMBA

broadleaf weeds infesting corn, wheat, and rangeland grasses. The fate of dicamba has been studied in various components of the environment, including soils (Altom and Stritzke, 1973; Friesen, 1965; Smith, 1974), plants (Broadhurst et al., 1966; Chang and VandenBorn, 1971a,b; Robocker and Zamora, 1976), and animals (Chio and Sanborn, 1978; Ivie et al., 1974; St. John and Lisk, 1969; Tye and Engel, 1967). In the only previous study dealing with the fate of dicamba in ruminants in vivo (St. John and Lisk, 1969), unlabeled dicamba was tested; thus, results were not definitive regarding the occurrence and distribution of dicamba metabolites. The current studies were undertaken to more accurately define the metabolic fate of dicamba in ruminants and to evaluate the potential for appearance of dicamba or its metabolites in meat and milk of exposed animals.

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MATERIALS AND METHODS

Chemicals. Radiolabeled dicamba (17.06 mCi/mM, uniformly labeled with ^{14}C in the phenyl ring) was supplied for these studies by the Velsicol Chemical Corp., Chicago,

IL. Radiochemical purity was determined to be >99% by thin-layer chromatography (TLC) in solvent systems A and B (vide infra). Unlabeled dicamba and several potential metabolites were also supplied by Velsicol. These compounds included methyl 3,6-dichloro-2-methoxybenzoate (dicamba-Me), 3,6-dichloro-2-hydroxybenzoic acid (DCHBA) and its methyl ester, 3,6-dichloro-2,5-dihydroxybenzoic acid and its methyl ester, 3,6-dichloro-5-hydroxy-2-methoxybenzoic acid and its methyl ester, 1,4-dichloro-2-methoxybenzene, 2,5-dichloro-1,4-benzenediol, and 2,5-dichlorophenol.

Treatment and Sampling. A 411-kg lactating Jersey cow was obtained from a local diary and confined in an indoor stall several days before treatment began. A commercial dairy ration was fed at each milking (12-h intervals); water and alfalfa hay were available at all times. Immediately before treatment, the animal was stanchioned in a metabolism stall and catheterized. The radiolabeled chemical was mixed with a small amount of grain in a gelatin capsule and administered orally with a balling gun. Eleven doses of 450 mg of [^{14}C]dicamba (1.0 mCi) each were administered at 12-h intervals. This dosage was 2.2 mg of dicamba (kg of body weight)$^{-1}$ day^{-1} and was equivalent to a concentration of about 60 ppm of dicamba in a normal diet if we assume that the animal consumed an amount of feed equal to 3–4% of its body weight per day. The total [^{14}C]dicamba given was 4.95 g (11.0 mCi). The specific activity of 4900 dpm/μg allowed the detection of concentrations of dicamba in excreta, tissues, and milk equivalent to 0.01 ppm or above.

After treatment began, samples of urine and milk were collected at 12-h intervals. Duplicate samples of both milk (0.5 mL) and urine (0.2 mL) were immediately pipetted into vials for analysis by liquid scintillation counting (lsc), and larger aliquots were frozen for subsequent studies. Feces samples were taken at 24-h intervals. The animal was sacrificed 6 h after the 11th dose had been administered, and samples of various tissues were taken and immediately frozen.

Extraction and Analysis. Total radiocarbon in urine and milk was quantitated directly by lsc as described above, and ^{14}C in feces and tissue samples was quantitated by oxygen combustion of samples that had first been air-dried and pressed into pellets. The resulting combustion gases were bubbled through a solution containing 5 mL of CO_2 trapping solution (Packard Carbosorb II) and 5 mL of a toluene-based scintillation fluid. The trapped radiocarbon was then quantitated by addition of 10 more mL of scintillation fluid and subjecting the sample to lsc. One-gram samples of tissue and feces (wet weight) were analyzed by combustion, and 0.2-g samples of adipose tissues were analyzed.

Radiocarbon-labeled constituents in whole urine and in extracts of milk and feces were resolved by two-dimensional TLC. The resolved ^{14}C components were visualized by autoradiography (Kodak No-Screen) and then the gel corresponding to each metabolite was scraped from the plate and quantitated by direct lsc. The TLC plates spotted with whole urine were exposed to film for 2 weeks; those spotted with extracts of milk, feces, and tissues were exposed for 4 weeks.

Feces samples (5 g) were extracted four times with 20-mL parts of methanol. Each extraction was homogenized with a Polytron, and solids were separated by centrifugation. The radiocarbon not extracted from the feces residue was quantitated by combustion and subsequent lsc.

Milk samples (100 mL) were adjusted to pH 2.0 with hydrochloric acid and then extracted three times with 100-mL parts of ethyl acetate. Centrifugation was used to help break emulsions. The solvent was removed from the combined milk extract with a Buchi evaporator, and the resulting oily residue was taken up in hexane and partitioned three times with acetonitrile. Radiocarbon in each phase was quantitated by lsc, then the acetonitrile part was analyzed by TLC.

Kidney (50 g) was homogenized in 150 mL of acetonitrile and then filtered. The extraction was repeated three more times in 100 mL of acetonitrile. After the solvent was removed, the residue was taken up in hexane and partitioned three times with acetonitrile. Radiocarbon in each phase was quantitated; then the acetonitrile phase was concentrated and analyzed by TLC. The extracted kidney residue was dried, combusted, and analyzed by lsc in an effort to determine the amount of unextracted radiocarbon.

Liver samples (5 g) were extracted by homogenizing the tissue with 25 mL of methanol, centrifuging the homogenate, and decanting the supernatant. The extraction was repeated five more times. After the solvent was removed, the liver extract was taken up in a small amount of hexane and subjected to cleanup on a 20-g, 2-cm diameter Florisil column, which was developed in sequence with 100 mL of hexane, 100 mL of ethyl acetate, and 100 mL of 1% acetic acid in methanol. Essentially all of the radiocarbon eluted with the methanol fraction; and after solvent removal, this fraction was taken up in hexane and partitioned three times with acetonitrile. As with the milk and kidney extracts, the acetonitrile part was then analyzed by TLC. Radiocarbon in the hexane phase was quantitated but not further studied; radiocarbon in the extracted liver residue was quantitated by combustion analysis.

In efforts to obtain quantities of urine metabolites sufficient for spectral studies, 100-mL samples of urine were adjusted to pH 0.5 with hydrochloric acid and then extracted five times with 100-mL portions of ethyl acetate. Both phases were analyzed by lsc for radiocarbon content. The organic phase was dried over anhydrous sodium sulfate, concentrated, and subjected to preparative TLC with solvent system A (vide infra). After autoradiography, the gel corresponding to the radiolabeled components was scraped from the plates, and the components were eluted with methanol. Each metabolite was subjected to a second (solvent system B), and one metabolite to a third (solvent system H) cleanup by TLC. In each cleanup, the products were recovered by elution with methanol. Metabolites suspected to be glucuronides were subjected to acid and enzyme hydrolysis procedures. Acid hydrolysis involved heating (100 °C) the metabolites in 1 N hydrochloric acid for 1 h, then extracting with ethyl acetate, and subsequent TLC analysis. Enzyme hydrolysis involved incubating the metabolites for 12 h in 5.0 mL of 0.1 M sodium acetate buffer (pH 4.5) with 2.1 IU of β-glucuronidase. After incubation, the solution was adjusted to pH 2.0, extracted with ethyl acetate, and subjected to further analysis.

Thin-Layer Chromatography. Metabolites in urine, feces, milk, kidney, and liver were resolved and characterized by TLC. Precoated silica gel chromatoplates (Silplate-F22, 0.25- and 2.0-mm gel thickness; Brinkman, Westbury, NY) were used. Metabolites were usually separated by developing the plates two-dimensionally, first in solvent system A (25:25:1, methanol/ethyl acetate/acetic acid) and then in B (50:1, ethyl acetate/acetic acid). Other solvent systems used were as follows: C (50:1, acetone/acetic acid), D (50:1, acetonitrile/acetic acid), E (90:30:1, benzene/dioxane/acetic acid), F (85:10:5, chloroform/ethanol/acetic acid), G (93:7:2, benzene/ethanol/acetic acid), and H (1:1, hexane/methylene chloride). Radioac-

Table I. Thin-Layer Chromatography (TLC) Values of Dicamba and Its Metabolites in a Cow[a]

metabolite	R_f in indicated TLC system[b]							
	A	B	C	D	E	F	G	H
dicamba	0.57	0.30	0.31	0.23	0.17	0.37	0.18	
DCHBA[c]	0.63	0.14	0.40	0.25	0.03	0.17	0.06	
2,5-dichlorphenol	0.72	0.63	0.75	0.71	0.53	0.67	0.48	0.50
DCHBA glucuronide	0.19	0.02	0.03	0.03	0.01	0.02	0.01	

[a] Brinkman Silplate F-22, 0.25-mm gel thickness. [b] Solvent systems as follows: A, 25:25:1, methanol/ethyl acetate/acetic acid; B, 50:1, ethyl acetate/acetic acid; C, 50:1, acetone/acetic acid; D, 50:1, acetonitrile/acetic acid; E, 90:30:1, benzene/dioxane/acetic acid; F, 85:10:5, chloroform/ethanol/acetic acid; G, 93:7:2, benzene/ethanol/acetic acid; and H, 1:1, hexane/methylene chloride. [c] 3,6-Dichloro-2-hydroxybenzoic acid.

Table II. Radiocarbon Excretion by a Cow Given 11 Successive 450-mg Oral Doses of [^{14}C]Dicamba at 12-h Intervals[a]

days after 1st treatment	urine		feces[b]		milk	
	%[c]	ppm[d]	%[c]	ppm[d]	%[c]	ppm[d]
0.5	5.52	48.9			0.0004	0.01
1.0	6.73	51.3	0.13	0.7	0.0014	0.02
1.5	7.55	54.7			0.0009	0.02
2.0	8.06	62.7	0.12	1.5	0.0016	0.03
2.5	10.61	65.7			0.0015	0.04
3.0	9.49	46.5	0.55	2.3	0.0029	0.04
3.5	8.58	82.0			0.0021	0.03
4.0	10.26	72.3	0.36	1.7	0.0030	0.03
4.5	9.06	113.6			0.0018	0.02
5.0	7.55	70.0	0.30	1.4	0.0023	0.02
Sac[e]	5.43	81.4	0.07	1.1		
total	88.84		1.53		0.0179	

[a] Dosage equivalent to 2.2 mg kg^{-1} day^{-1} and approximately 60 ppm of dietary dicamba. [b] Feces samples collected at 24-h intervals. [c] Percentage of total administered dose. [d] Ppm dicamba equivalents. [e] Samples collected when the cow was sacrificed 6 h after the final dose.

tive areas were located by autoradiography, and unlabeled standards were detected by viewing the fluorescent plates under short-wavelength ultraviolet light. Identification of metabolites was based on coincidence of radioactive areas and the known standards in each of several of the solvent systems used. TLC R_f values for dicamba and its metabolites in the cow in each of these solvent systems are given in Table I.

Gas–Liquid Chromatography/Mass Spectroscopy. The identifications of metabolites established by TLC were confirmed, where possible, by interfaced gas liquid chromatography (GLC)/mass spectroscopy. The instrumentation included a Varian-MAT-CH-7 spectrometer coupled with a Varian 2700 gas chromatograph and a 620 L Varian computer. The gas chromatograph was equipped with a 1.8 m × 2 mm i.d. glass column packed with 3% SE 30 on 100–120-mesh Varaport 30. Operating parameters were as follows: injector, 210 °C, column temperature as specified; ion source, 275 °C; helium flow, 50 mL/min; ionizing voltage, 70 eV. Compounds that required derivatization before GLC/mass spectral analysis were methylated with an ether solution of diazomethane. Retention times and mass spectra of metabolites or their derivatives were compared with those of the appropriate authentic standards to establish identity.

RESULTS

Radiocarbon Excretion and Tissue Residues. Of the total radiocarbon given the cow during the 5-day treatment period, about 89% was excreted in the urine, 1.5% in the feces, and less than 0.02% in the milk (Table II).

Table III. Radiocarbon Residues in Tissues of a Cow Given 11 Successive 450-mg Doses of [^{14}C]Dicamba at 12-h Intervals[a,b]

tissue	ppm[c]	tissue	ppm[c]
blood	0.40	liver	0.30
brain	0.01	muscle[d]	0.02
fat, omental	0.02	muscle[e]	0.03
fat, renal	0.02	ovary	0.25
heart	0.09	spleen	0.05
kidney	2.59	tongue	0.09

[a] Dosage equivalent to 2.2 mg kg^{-1} day^{-1} and approximately 60 ppm of dietary dicamba. [b] Tissues collected 6 h after the final dose. [c] Ppm dicamba equivalents. [d] Longissimus dorsi. [e] Triceps.

Table IV. Distribution of Dicamba and Its Metabolites in Urine and Feces of a Cow Given 11 Successive 450-mg Oral Doses of [^{14}C]Dicamba at 12-h Intervals[a]

days after 1st treatment	% radiocarbon as indicated metabolite				
	dicamba	DCHBA[b]	DCHBA glucuronide	2,5-dichlorophenol	other
			Urine		
1	81	15	2	2	<1[c]
2	79	14	3	4	<1[c]
3	77	18	2	3	<1[c]
4	79	17	2	2	<1[c]
5	80	16	2	2	<1[c]
			Feces		
1	84	9	0	0	7[d]
2	84	8	0	0	8[d]
3	77	11	0	0	12[d]
4	77	11	0	0	12[d]
5	75	13	0	0	12[d]

[a] Dosage equivalent to 2.2 mg kg^{-1} day^{-1} and approximately 60 ppm of dietary dicamba. [b] 3,6-Dichloro-2-hydroxybenzoic acid. [c] Radiocarbon remaining at origin after TLC in solvent systems A × B. [d] Radiocarbon not extracted from the feces residue.

In the milk, the levels of radioactivity reached their maximum concentration (0.04 ppm) 2.5 days after the first dose. After this time, milk radiocarbon levels appeared to decline slightly (Table II). Residues of radiocarbon in all tissues analyzed were also very low, even though the animal was sacrificed only 6 h after the last treatment, when tissue residues would be expected to be at or near their highest levels (Table III). As might be predicted on the basis of the large amounts of radiocarbon excreted in the urine, kidney samples contained higher concentrations of ^{14}C residues than any of the other tissues studied.

Metabolite Resolution and Characterization. *Urine.* Two-dimentional TLC of whole urine in solvent systems A and B resolved the urinary radiocarbon into five components. The major radioactive product in urine, comprising 77–81% of the total ^{14}C present (Table IV), was identified as unmetabolized dicamba on the basis of TLC

cochromatography with authentic dicamba in solvent systems A–G, and on the basis of GLC/mass spectral analysis (170 °C column, retention time, 2.05 min) of the methylated product from urine in comparison with the properties of authentic dicamba-Me. Major ions observed both in the authentic standard and the methylated urine component were as follows (Cl = 35): m/e 234 (M^+), 203 (base peak, $M^+ - OCH_3$), 188 ($M^+ - OCH_3$, CH_3), and 175 ($M^+ - CO_2CH_3$).

The major metabolite in urine was 3,6-dichloro-2-hydroxybenzoic acid (DCHBA) generated by hydrolysis of the aryl-*O*-methyl group. The metabolite comprised 14–18% of the radiocarbon eliminated in the urine, depending on the sample (Table IV). Its structure was established by TLC cochromatography with authentic DCHBA (A-G). Structure was confirmed upon GLC/mass spectral analysis of the fully methylated metabolite, which established that the methylated product was indeed dicamba-Me. The other relatively nonpolar metabolite was characterized as 2,5-dichlorophenol. The metabolite from urine behaved the same on TLC (A–G) as the authentic dichlorophenol standard; and upon GLC/mass spectral analysis (110 °C column; retention time, 2.75 min), the metabolite gave the same retention time and mass spectrum as authentic 2,5-dichlorophenol. Major ions were as follows (Cl = 35): m/e 162 (base peak, M^+), 133 (M^+ – COH), 126 (M^+ – Cl, H), 99 (M^+ – CO, Cl), 98 (M^+ – COH, Cl).

One of the two relatively polar metabolites in urine was represented by a very low amount of ^{14}C that remained at the origin after TLC in A × B. In all samples, this product(s) comprised <1% of the total radiocarbon present (Table IV), and no attempt was made to define its chemical nature. The other metabolite was also quite polar, but did migrate slightly above the origin upon TLC in A and B. The high polarity of the compound suggested that it might be a conjugate, and acid and β-glucuronidase hydrolysis confirmed that the product was a glucuronide of DCHBA. Both hydrolysis procedures resulted in >80% transformation of the metabolite into a product that was identical on TLC (A–G) with authentic DCHBA. GLC/mass spectral analysis of the methylated aglycon confirmed its structure as the methyl ester of dicamba and thus that the aglycon was DCHBA. Although we did not determine experimentally whether the sugar moiety was attached at the phenolic or carboxylic acid sites, very likely the site of conjugation was the aryl hydroxyl group. This conclusion is based primarily on the observation that relatively large amounts of dicamba per se were eliminated in the urine with no apparent glucuronide conjugation.

Feces. Extraction of feces samples with methanol recovered 88–93% of the total radiocarbon present (Table IV), and TLC analysis of the extracts showed that only dicamba and DCHBA were present. Dicamba was by far the major radioactive component in feces (75–84% of total feces radiocarbon), while DCHBA represented 8–13% of the feces radioactivity. The structure of both products were confirmed by TLC (A–G) comparisons with authentic standards.

Milk. The milk contained only very low amounts of radiocarbon, and studies with the milk were confined to the 3-day sample, which contained the highest residues. Ethyl acetate extraction of the acidified whole milk recovered 70% of the radiocarbon present, and further hexane–acetonitrile cleanup resulted in 84% of the extracted ^{14}C partitioning into acetonitrile. Subsequent TLC analysis of the acetonitrile phase (A × B) showed that the only detectable radioactive component present was DCHBA. The metabolite from milk behaved the same on TLC (A–G) as authentic DCHBA, but it was not obtained in sufficient quantity for study by GLC/mass spectroscopy. The nature of the very low amounts of radiocarbon in the extracted aqueous phase of milk or that in the hexane phase from cleanup was not defined.

Tissues. In liver, 51% of the total radiocarbon was dicamba, 21% was DCHBA, 3% was almost equally divided between two unidentified metabolites that migrated slightly higher than dicamba on TLC (C, E, F, and G), and 5% remained at the origin after TLC. The hexane phase from cleanup contained only negligible levels of radiocarbon. About 20% of the radiocarbon in liver remained in the residue after extraction. Radiocarbon in kidney samples consisted of dicamba (70%) and DCHBA (11%). Only about 1% of the kidney radiocarbon partitioned into the hexane during the cleanup step, and 18% remained in the extracted residue. The identities of dicamba and DCHBA in the liver and kidney extracts were confirmed by TLC (A–G) cochromatography with the appropriate standards. The metabolites were not available in sufficient quantities for confirmation of structure by GLC/mass spectral analysis.

DISCUSSION

The current studies have established that dicamba is rapidly absorbed, only partly metabolized, and rapidly excreted after oral exposure to a ruminant. For comparative purposes with the data in Table II, we can consider that because the cow was treated with 11 equal [^{14}C]dicamba doses at 12-h intervals, the animal received about 9% of the total dose every 12 h. Thus, examination of Table II indicates that by the time the second [^{14}C]dicamba dose was administered, the animal had already excreted about 60% of the radiocarbon given only 12 h earlier. The data in Table II also indicate that within 2–3 days after treatment began, the rate of excretion equaled the rate of intake. At the time the cow was sacrificed 6 h after the 11th and final [^{14}C]dicamba dose, the animal had eliminated about 90% of the total administered ^{14}C. We can reasonably assume that most of the ^{14}C unaccounted for in the excreta can be attributed to that remaining within the digestive tract or circulatory system of the animal at death.

Dicamba is rapidly and almost entirely absorbed from the digestive tract of ruminants, as evidenced by the fact that only very low amounts of ^{14}C were eliminated in the feces. This absorption would also suggest a minor role for biliary excretion of dicamba and its metabolites unless enterohepatic circulation was extensive.

Dicamba showed very little tendency toward secretion into milk or retention by tissues of the treated animal. In fact, no dicamba per se was detected in the milk samples analyzed; the only radioactive constituent identified in the milk was DCHBA.

Only about 20% of the [^{14}C]dicamba given the cow during the 5-day treatment period was metabolized, and by far the major transformation was O-demethylation to DCHBA. Although unmetabolized dicamba was the major radioactive component in all excreta and tissue samples studied, except milk, DCHBA was the major dicamba metabolite in urine, the only metabolite in extracts of feces and kidney, and the only radioactive component in extract of milk. In addition to dicamba and DCHBA, extract of liver contained two other, relatively nonpolar, unidentified metabolites. These products were apparently generated in exceedingly minor quantity because they did not appear in detectable amounts in urine, feces, or milk. The only other dicamba metabolites identified in the cow were

DCHBA glucuronide and 2,5-dichlorophenol (Table IV). The phenol was apparently generated by decarboxylation of DCHBA.

The appearance of DCHBA as a metabolite in feces may have been the result of O-demethylation of dicamba within the digestive tract or it may have resulted from limited biliary excretion of the metabolite. Earlier work in these laboratories has shown, however, that O-demethylation of dicamba does not occur in ovine rumen fluid in vitro (Ivie et al., 1974).

Results in studies on the fate of [^{14}C]dicamba in the rat have shown that the compound is excreted primarily in the urine in the form of free dicamba, with lesser amounts of dicamba glucuronide (Tye and Engel, 1967). It was not established whether or not DCHBA was a dicamba metabolite in the rat. Thus, dicamba metabolism by ruminants may differ from that of laboratory rodents in at least two ways: The conjugation of dicamba with glucuronic acid in rats but not in cows and O-methylation with subsequent glucuronide conjugation or decarboxylation in cows but possibly not in rats.

Our studies with [^{14}C]dicamba in the cow are more definitive than, but are certainly consistent with, an earlier report on the fate of unlabeled dicamba in ruminants (St. John and Lisk, 1969). These workers showed that oral treatment of lactating dairy cattle with dicamba resulted in rapid elimination of unmetabolized dicamba in the urine and no detectable dicamba residue in milk. Our work confirms these earlier observations, but also indicates that dicamba is, in fact, metabolized to a limited extent in ruminants and that although dicamba itself is not secreted into milk, its metabolite, DCHBA, does have the potential for appearance in milk at trace levels.

On the basis of results in the current studies, exposure of livestock to low levels of dicamba through contaminated forage, feed, or water likely will be followed by rapid elimination from the body with little or no retention of residues by edible tissues. The secretion of appreciable levels of residues into the milk of exposed animals seems highly unlikely.

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In Vitro Rumen Metabolism of ^{14}C-Labeled Oxamyl and Selected Metabolites of Oxamyl

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The metabolism of radiolabeled oxamyl, methyl *N',N'*-dimethyl-*N*-[(methylcarbamoyl)oxy]-1-thiooxamimidate-*1*-^{14}*C*, by rumen microorganisms, in vitro, was rapid. Essentially all (99%) of the oxamyl had been metabolized within 6 h. The major metabolites after 24 h of incubation were methyl *N*-hydroxy-*N',N'*-dimethyl-1-thiooxamimidate and *N,N*-dimethyl-1-cyanoformamide (DMCF), which accounted for 80% of the residual radioactivity. Separately, the in vitro rumen metabolism of metabolite A, the glucose conjugate of methyl *N*-hydroxy-*N',N'*-dimethyl-1-thiooxamimidate, yielded DMCF. In turn, DMCF was found to metabolize to *N,N*-dimethyloxamide, *N,N*-dimethyloxamic acid, and *N*-methyloxamic acid. All the metabolites isolated and characterized, except for *N,N*-dimethyloxamide, were identical with those previously demonstrated in the rat.

Oxamyl, the generic name for methyl *N',N'*-dimethyl-*N*-[(methylcarbamoyl)oxy]-1-thiooxamimidate, is the active ingredient in Du Pont's Vydate oxamyl insecticide/nematicide. It was formerly known as DPX-1410.

Information relative to the metabolic fate of oxamyl in a variety of crop plants, as well as in the rat, and the characterization of the metabolites found has been reported by Harvey et al. (1978) and by Harvey and Han (1978a). Also, the degradation of oxamyl in water and its decomposition and movement in soil under laboratory and field conditions have been reported (Harvey and Han, 1978b).

The findings reported in this study describe the metabolism of oxamyl, *N,N*-dimethyl-1-cyanoformamide, and the glucose conjugate of methyl *N*-hydroxy-*N',N'*-dimethyl-1-thiooxamimidate (metabolite A) in the presence of rumen microorganisms.

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