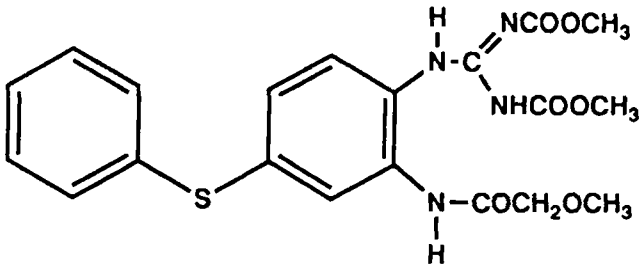
FEBANTEL

IDENTITY

Chemical: Dimethyl [[2-(2-dimethyloxyacetamido)-4-(phenylthio)phenyl]imidocarbonyl]dicarbamate

Synonyms: RINTAL; ORATEL ; CAS 58306-30-2

Structural formula:



Molecular formula: $C_{20}H_{22}N_4O_6S$

Molecular weight: 446.48

OTHER INFORMATION ON IDENTITY AND PROPERTIES

Pure active ingredient: 98.0-102.0% Febantel, based on dry substance.

Appearance: White crystals

Melting point: 129-130 °

Solubility: 1-2 ppm in water at pH 5-9

Octanol/Water Partition Coefficient: Log_{ow} 2.9, 2.8, 2.6 (pH 5, 7, 9)

RESIDUES IN FOOD AND THEIR EVALUATION

CONDITIONS OF USE

General

Febantel is a broad spectrum anthelmintic used for the treatment of gastrointestinal parasitism in cattle, sheep and swine. The recommended therapeutic doses for cattle, sheep and swine are 10, 5 and 5 mg/kg, respectively.

Dosages

Febantel is administered orally as a suspension, a paste (45.5%) or as a feed additive.

METABOLISM

General

For background studies on the metabolism of febantel in rats and sheep, the guanidino carbon was labelled with ^{14}C and ^{13}C. The specific activity was approximately 44 mCi/mM, and the radiochemical purity was greater than 98.5% as shown by thin-layer chromatography. (Weber, et al, 1976)

All other metabolism and total residue depletion studies were conducted with [^{14}C-phenylthio] febantel. Specific activity was approximately 40 mCi/mM, and the radiochemical purity was greater than 98.0%. Radiopurity of febantel of specific activity 39.99 mCi/mM showed no significant losses (> 98.0%) during an 8 month storage period. The specific activity of [^{14}C] febantel was lowered by addition of cold drug substance and subsequent repurification to give a specific activity of 10.1 mCi/mM and a radiopurity of > 99.0%. After storage of [^{14}C] febantel over a 4 month period at -70° as a solid or in ethanol, no significant loss of radiopurity was observed. (Waggoner, 1987a)

To support metabolite identification, reference standards corresponding to suspected metabolites and degradation products have been synthesized and their identity confirmed by spectral data. The reference standards were utilized in all metabolism studies with the target species and rats. (Waggoner, 1987b)

Febantel cyclizes rapidly in vivo to yield fenbendazole, oxfendazole and oxfendazole sulfone as its main metabolites. There is some evidence that parent febantel and febantel sulfoxide are present in liver tissue at short withdrawal times; however, their concentrations are too low for either to serve as a marker residue. Studies with ^{14}C-guanidino labelled febantel do not quantitatively measure the benzimidazole metabolites and should be used only to identify the metabolites.

Rat

The pharmacokinetics of febantel was investigated by administering either oral, intravenous or intraduodenal doses of [^{14}C-guanidino] febantel to Sprague-Dawley SPF male rats. Within 48 hours after oral dosing of 5 mg drug per kg body weight,

approximately 25 - 30% of the administered dose was excreted in urine and approximately 70% was excreted in feces. Rate of absorption was rapid as shown by peak levels in blood serum within one hour after dosing. From intravenous and intraduodenal doses, approximately 70% of the dose was eliminated via the bile. During the first 48 hours after an oral dose, excretion of residues was measured in the respired air. Less than l% of the dose was found, indicating that ^{14}C-guanidino carbon was not transformed to volatile products or carbon dioxide. Depletion of total drug-related residues was observed in tissue to a maximum of 10 days after a single oral dose of 5 mg/kg and is summarized in Table I. Liver contained the most residue at all withdrawal intervals. (Weber, et al, 1977a)

Table I. Total Drug-related residues (in μg/kg) in rat tissue after oral dosing.

Time (Days)	Liver	Kidney	Muscle	Fat
0.1	800	220	35	280
0.3	600	155	22	230
1	250	34	3	22
3	115	11	3	<4
6	60	5	<1	<4
10	18	3	<1	<4

For comparative metabolism purposes, four male and four female Charles River Wistar rats [Crl:(WI)Br] were administered [^{14}C-phenylthio] febantel, orally at a dose of 25 mg/kg/day for seven days. Urine was collected daily and pooled by sex for investigation of metabolic profile. Approximately 20 - 25% of residues in urine were conjugated as demonstrated by glucuronidase/ sulfatase enzyme studies. Components in urine were characterized by thin-layer chromatography and comparison of Rf values to those of authentic analytical standards. Urinary excretion in males and females accounted for approximately 17 and 24%, respectively, of the administered dose. A summary of components found in male and female urine is given in Table II. Those metabolites identified compared against authentic standards by thin-layer chromatography are given in Table III. No significant qualitative differences were observed between male and females. (Leeling et al, 1986a)

In another study 4 male and 4 female Wistar rats, of identical breeding to rats in the urine study, were administered [^{14}C-phenylthio] febantel as a single oral dose of 25 mg/kg. The animals were sacrificed one hour after dosing. Livers were removed, pooled by sex and investigated for residues of febantel. Livers were subjected to hydrolysis in the presence of glucuronidase/sulfatase prior to extraction and characterization by thin-layer chromatography. Results are summarized in Table II indicating the number and levels of metabolites found in liver. Identified metabolites are shown in Table IV. Residues of febantel, BAY h 5156 (fenbendazole) and BAY h 6817 (fenbendazole sulfone) were detected in liver but not urine for rats. (Leeling et al, 1986b)

Table II. Metabolites of febantel found in rat urine and liver.

% of Total Residue Found	Number of Metabolites			
	Urine		Liver	
	Male	Female	Male	Female
>10	3	3	3	2
5-10	1	3	2	2
1-5	9	2	7	7
<1	21	33	6	12
Total	35	41	18	23

Figure 1, below, is a schematic representation of febantel metabolites. The molecule has substituted groups at the X and R locations with the nature of these groups determining the metabolite. The R is either $COOCH_3$ or H and X is a sulfur, a sulfoxide or a sulfone. See the following tables III to IX.

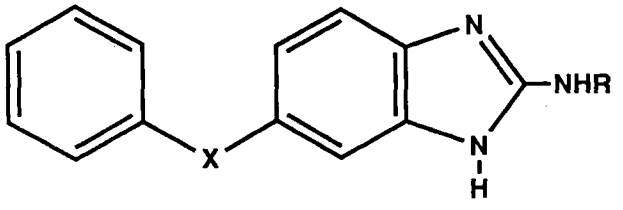


Figure 1

Table III. Identification of metabolites of febantel in rat urine.

Metabolite	R	X	% of Total Residue in Urine	
			Male	Female
BAY h 7828	$COOCH_3$	SO	27	36
BAY k 8449	H	SO_2	18	14
BAY k 1236[a]	$COOCH_3$	S	13	8
BAY k 7648	H	SO	10	13

[a] A para-hydroxy substituted phenyl metabolite.
(Note: See figure 1 for R and X)

Table IV. Identification of metabolites of febantel in rat tissue.

Metabolite	R	X	% of Total Residue in Liver Male	Female
Febantel[a]	-	-	3	1
BAY h 5156[b]	$COOCH_3$	S	22	37
BAY h 7828	$COOCH_3$	SO	3	3
BAY k 6817	$COOCH_3$	SO_2	12	8
BAY k 1236	$COOCH_3$	S[c]	27	23
BAY i 6057	H	S	0	<1
BAY k 7648	H	SO	0	0
BAY k 8449	H	SO_2	3	2

[a] Parent drug
[b] Fenbendazole
[c] Para-hydroxyphenyl substituted
(Note: See figure 1 for R and X)

Sheep

Studies were initially conducted to identify metabolites in urine, feces and blood serum. Three female lambs were administered a single 5 mg/kg dose intraruminally. Pooled samples of urine and feces, each collected over a 3 day interval after dosing, were investigated for metabolites. Blood serum was investigated for metabolite content at two intervals after dosing, 8 and 12 hours. Up to 30 components were found in urine. Approximately 30% of total residues in urine was conjugated as shown by glucuronidase/sulfatase enzyme hydrolysis. Results of components found in urine are shown in Table V. At least 15 components were found in feces. The major components are also summarized in Table V. Up to 18 components were found in blood serum, and the major ones are also summarized in Table V. (Maul, et al, 1977a)

Table V. Metabolites of febantel found in sheep urine, feces and blood serum.

Metabolite	R	X	% of Total Residue Found Urine	Feces	Blood Serum
Febantel[a]	-	-	0	30	0
BAY k 1236[b]	$COOCH_3$	S	20	15	0
BAY h 5156	$COOCH_3$	S	7	17	12[c]
BAY h 7828	$COOCH_3$	SO	11	3	35
BAY h 6817	$COOCH_3$	SO_2	4	6	35
BAY i 6057	H	S	0	0	0
BAY k 7648	H	SO	10	0	<1
BAY k 8449	H	SO_2	1	0	<1
Urea[d]	-	-	1	-	-

[a] Parent drug
[b] Para-hydroxyphenyl substituted
[c] Maximum level found
[d] Formed from guanidino carbon
(Note: See figure 1 for R and X)

Cattle

One male and one female beef calf were administered a single oral dose of [^{14}C-phenylthio] febantel at 7.5 mg drug per kg body weight. Tissues were collected after 0.75 day withdrawal. Liver was then investigated to determine a metabolic profile. Results are summarized in Table VI. Greater than 90% of total drug-related residues was readily extractable from liver. (Leeling et al, 1986c)

Table VI. Metabolites of febantel found in bovine liver -0.75 day withdrawal.

			% of Total Residue Found	
Metabolite	R	X	Male	Female
Febantel[a]	-	-	3	6
BAY h 5156[b]	$COOCH_3$	S	41	30
BAY h 7828	$COOCH_3$	SO	4	10
BAY h 6817	$COOCH_3$	SO_2	15	14
BAY k 6057	H	S	>1	>1
BAY k 7648	H	SO	3	>1
BAY k 8449	H	SO_2	>1	0
Unidentified	-	-	6	12

[a] Parent drug
[b] Fenbendazole
(Note: See figure 1 for R and X)

Liver from a female calf treated orally with a single 7.5 mg/kg dose and sacrificed at a 10 day withdrawal interval was investigated for the characterization of metabolites. A more rigorous extraction was needed in order to solubilize the residues. Nearly 90% of total residues were extracted by a soxhlet extraction utilizing acetic acid:water (1:9 v/v). Metabolites were then separated and detected by HPLC utilizing UV. Results for a 10 day withdrawal interval are given in Table VII. Values are for two separate samples. For kidney at a 10 day interval, BAY h 5156 and BAY k 8449 represented 10 and 16% of total residues, respectively, although each metabolite was less than 0.05 ppm. (Waggoner, 1986a)

Table VII. Metabolites of febantel found in bovine liver at 10 day withdrawal.

Metabolite	R	X	% of Total Residue Found
Febantel[a]	-	-	0
BAY h 5156[b]	$COOCH_3$	S	<1-4
BAY h 7828	$COOCH_3$	SO	3-9
BAY h 6817	$COOCH_3$	SO_2	1-13
BAY i 6057	H	S	0
BAY k 7648	H	SO	3-7
BAY k 8449	H	SO_2	12-35

[a] Parent drug
[b] Fenbendazole
(Note: See figure 1 for R and X)

Recovery of total drug-related residues for 10 day liver was found initially to be 70.1%. The 10 day liver sample was reinvestigated with an emphasis to account for all radioactivity in the sample. Utilizing an identical extraction procedure as described previously, aqueous 10% acetic acid followed with methanol, quantitative (actually 104%) accountability was obtained from liver containing 2.65 ppm total drug-related residues. (Waggoner, 1987c)

Metabolic profiles were also determined for 0.75 and 10 day bovine fat, containing 0.9 and 0.1 ppm, respectively, of total drug-related residues, and quantitative recovery was obtained. Results are summarized in Table VIII. (Waggoner, 1987c)

Table VIII. Metabolites of febantel found in bovine fat.

			% of Total Residue Found	
Metabolite	R	X	0.75 Days	10 Days
BAY h 5156	$COOCH_3$	S	6	28
BAY h 7828	$COOCH_3$	SO	6	82
BAY h 6817	$COOCH_3$	SO_2	11	0
BAY i 6057	H	S	8	0
BAY k 7648	H	SO	0	0
BAY k 8449	H	SO_2	12	0

(Note: See figure 1 for R and X)

Approximately 90% of total drug-related residues were readily solubilized from liver following a 0.75 day withdrawal interval, with solvents such as aqueous acetonitrile. In contrast, only 15% of total drug-related residues could be solubilized with the same solvent system following a 10 day withdrawal interval. Other attempts to extract 10 day liver with methanol or ethyl acetate solubilized only 15 - 20% of total residue. The best extraction procedure for solubilizing the highest amount of total residue was soxhlet extraction with aqueous 10% acetic acid using fresh solvent for each of three 24-hour

extractions which resulted in a maximum of 90% of total drug related residues solubilized from liver following a 10 day withdrawal interval; however, typical amounts of solubilized residue were actually in the 60 - 70% range. (Waggoner, 1987d)

It was therefore concluded that approximately 30 - 40% of total drug-related residues in 10 day liver was bound in some unknown way and required more vigorous conditions (heat, polar solvent and time) for the solubilization. Comparison of metabolic profiles for liver following a 0.75 day withdrawal interval were demonstrated between the two analytical procedures used; (I) aqueous acetonitrile extraction followed by thin-layer chromatographic detection of metabolites and (2) aqueous 10% acetic acid soxhlet extraction followed by HPLC radiochemical detection of metabolites. For both procedures, the metabolic profiles were qualitatively identical with small differences quantitatively. It was found that the levels of methylcarbamoyl ester fenbendazole metabolites were slightly lower from the acetic acid extraction, probably due to hydrolysis under the more drastic conditions, lower pH and higher temperatures. For example, levels of BAY k 8449 from aqueous acetic acid extraction represented 5 - 10% of total drug-related residues but only 0.3% from aqueous acetonitrile extraction. (Waggoner, 1986a)

In summary, approximately 80% of total drug-related residues were bound to tissue of 10 day liver, and normally, all but approximately 30% of total drug-related residue could be extracted with aqueous 10% acetic acid at elevated temperature. Unidentified residue consisted of many metabolites probably retaining intact benzimidazole ring structures that were reversibly bound to tissue. (Waggoner, 1986a)

Swine

A single dose of 5 mg/kg of the ^{14}C-(guanidino) febantel was administered orally to each of three animals. One animal was sacrificed at 10, 20 and 30 days. The serum concentrations calculated as febantel equivalents peak at about 1500 ppb within 1-2 hours. The concentrations declined slightly up to 24 hours after administration. Thereafter, the activity declined with a half life of 12 to 15 hours. In the interval up to 10 days after administration 30-50% of the activity was eliminated in the urine and 40-60% in the feces. (Weber, et al, 1977b)

In another study three animals were dosed with 5 mg/kg of ^{14}C-(guanidino) febantel. The major metabolites in urine, feces and serum were identified. The results are summarized in Table IX. All values represent the time of 0 - 48 hours. (Maul, et al, 1977b)

Table IX. Metabolites of febantel found in swine urine, feces and serum.

			% of Total Residue Found		
Metabolite	R	X	Urine	Feces	Blood Serum
Febantel[a]	-	-	0	24	-
BAY k 1236[b]	$COOCH_3$	S	3	8	-
BAY h 5156	$COOCH_3$	S	5	35	5
BAY h 7828	$COOCH_3$	SO	4	2	25
BAY h 6817	$COOCH_3$	SO_2	1	3	22
BAY i 6057	H	S	<1	4	-
BAY k 7648	H	SO	21	<1	6
BAY k 8449	H	SO_2	16	<1	13
Urea[d]	-	-	26	-	21

[a] Parent drug
[b] Para-hydroxyphenyl substituted
[c] Formed from guanidino carbon
(Note: See figure 1 for R and X)

In a study with ^{14}C-ring labelled febantel no obvious differences in major metabolites and amount (except for labelled urea) could be detected on comparing the metabolism after administration of (U-^{14}C-phenyl ring)- and (^{14}C)guanidino- labelled drug. As expected, radioactive urea was not detected after administration of the ring-labelled drug. (Maul, 1980)

TISSUE RESIDUE DEPLETION STUDIES

Radiolabelled Residue Depletion Studies

Sheep

Three female sheep were administered a single dose of [^{14}C-guanidino] febantel, BAY Vh 5757, intraruminally equivalent to 5 mg drug per kg body weight. Within 4 days, approximately 85% of the dose had been excreted, 19% in urine and 66% in feces. Maximum levels in urine occurred between 12 - 24 hours after dosing. Maximum levels of total residues in blood serum were 400 - 700 ppb occurring within 12 - 24 hours after dosing. After 15 days, levels had fallen to the 5 - 10 ppb range. One animal was sacrificed at each of the three withdrawal intervals; 4, 8 and 15 days. The levels of total drug-related residues are summarized in Table X.

Table X. Total Drug-Related Residues of Febantel in Sheep.

Time (Days)	Tissue (ppb) Liver	Kidney	Muscle	Fat
4	5,000	235	35	100
8	2,500	75	10	15
15	300	45	<10	<10

It was concluded from this study that the rate of absorption was relatively slow as shown by peak levels in blood serum occurring between 12 - 24 hours. The highest residues occurred in liver at all withdrawal intervals, and levels observed in the remaining tissues were generally less than one-tenth the amount found in liver at all withdrawal intervals. (Weber, et al, 1976)

Cattle

For all total residue depletion studies, animals were administered a single oral dose of drug, [^{14}C-phenylthio] febantel. Initial studies were conducted by dosing animals at two rates, 7.5 and 12.0 mg drug per kg body weight. Additional studies were then conducted by administering a 7.5 mg/kg dose and sacrifice of the treated animals at 0.75 and 10 days after dosing. For establishing a total residue depletion curve for beef cattle receiving a single oral dose of 7.5 mg/kg, data from all three studies were summarized in a single format. A summary of total drug-related residues in bovine tissue for 0.3 - 42 days withdrawal is given in Table XI. Total residue levels depleted to less than 0.1 ppm in kidney, muscle and fat within 14 days after dosing. The highest level of residues occurred in liver at all withdrawal intervals. The rate of absorption was relatively slow as shown by peak levels in all tissues found at the 0.75 day withdrawal interval. The rate of absorption in cattle compared favorably to that found previously for sheep. (Waggoner, 1986b)

Table XI. Total Drug-Related Residues of Febantel in Bovine Tissue

Time (Days)	ppm (SD)[a] Tissues Liver	Kidney	Muscle	Fat
0.3	3.4 (.3)	0.6 (.2)	0.1 (.04)	0.4 (.1)
0.75	10.7 (.2)	2.7 (.1)	0.6 (.1)	1.0 (.3)
10	2.5 (.9)	0.2 (.07)	<.1	<.1
14	0.8 (.3)	<.1	<.1	<.1
28	0.3 (.07)	<.1	<.1	<.1
42	0.1 (.02)	<.1	<.1	<.1

[a] Dose 7.5 mg/kg; SD= ± standard deviation

In another study eight beef cattle were administered a single oral dose of 7.5 mg febantel/kg body weight which corresponded to the maximum dose anticipated under normal use. Two animals (1 male and 1 female) were sacrificed at each of the following

withdrawal intervals: 10, 12, 14 and 16 days. Total drug-related residues were then determined in liver, kidney, muscle and fat. These data are summarized in Table XII. (Waggoner, 1987d)

Table XII. Total Drug-Related Residues of Febantel in Bovine

Time (Days)	ppm* Tissue Liver	Kidney	Muscle	Fat
10	1.9	0.1	<.1	0.2
12	1.5	<.1	<.1	<.1
14	1.1	<.1	<.1	<.1
16	0.9	<.1	<.1	0.1

* Each value is an average of one male and one female.

Swine

Tissue levels in swine were determined 10, 20 and 30 days following oral administration of 5 mg/kg of febantel. The results are summarized in Table HIII. (Weber et al, !977b)

Table XIII. Total drug-related residue of febantel in swine.

Time (Days)	ppm Ticsue Liver	Kidney	Muscle	Fat
10	.35	.06	<.01	-
20	.15	.02	<.01	-
30	.06	<.01	<.01	-

Other Residue Depletion Studies (with Unlabelled Drug)

Sheep

Residue levels of BAY h 5156, BAY h 7828 and BAY h 6817 were determined in plasma and tissue of sheep after receiving a 5 mg/kg dose of febantel intraruminally. Residue levels represented the sum of the above three metabolites expressed as "BAY h 5156 equivalents". Results are summarized in Table XIV. (Raemsch, 1977)

Table XIV. Residue levels of BAY h 5156 equivalents in sheep plasma and tissue (in ppm[a])

Sample	Days 1	3	7	14	21
Plasma[b]	0.2	0.02	-	-	-
Liver[c]	6.8	-	2.2	0.2	<.1
Kidney	1.2	-	<.1	<.1	<.1
Muscle	0.4	-	<.1	<.1	<.1
Fat	0.8	-	<.1	<.1	<.1

[a] Values are corrected for background and recovery from appropriate tissue.
[b] Average mean of nine animals per interval.
[c] Average mean of three animals per interval for each tissue.

Cattle

Residue levels of BAY h 5156, BAY h 7828 and BAY h 6817 were determined in tissue of cattle receiving a single oral dose of febantel at 10 and 20 mg/kg. Residue levels represent the total of the three metabolites and were expressed as "febantel equivalents". Results are summarized in Table XV.

Table XV. Residue levels of febantel (BAY Vh 5757) equivalents in bovine tissue ppm[a,b]

Tissue	Days 7	14	21
20 mg/kg dose			
Liver	1.5	0.2	<.1
Kidney	2	0.1	<.1
Muscle	0.1	0.2	<.1
Fat	0.2	<.1	<.1
10 mg/kg dose			
Liver	0.8	<.1	0.2
Kidney	0.1	<.1	<.1
Muscle	0.2	<.1	<.1
Fat	0.2	<.1	<.1

[a] Values are corrected for background and recovery for the appropriate tissue; average mean values for three animals per interval.
[b] Values represent measurement of BAY h 5156 equivalents converted to febantel equivalents using a molecular weight factor.

In another study bovine calves, male and female, were each administered a single oral dose of Febantel 45.5% Oral Paste corresponding to a dose of 7.5 mg drug/kg body weight, the maximum dose. Three animals each were sacrificed at the following withdrawal intervals; 4, 7, 9, 11, 14, 16 and 18 days. Livers were sampled and frozen immediately. Analyses livers were conducted for BAY k 8449 with a liquid chromatographic procedure. Results are summarized in Table XVI. (Waggoner, 1987e)

Table XVI. Residue levels of BAY k 8449 in bovine liver.

Withdrawal Interval (Days)	Animal Sex	ppm BAY k 8449 Individual	Mean
4	M	0.17	0.24
	F	0.32	
	M	0.22	
7	M	0.2	0.21
	F	0.25	
	F	0.18	
9	M	0.17	0.16
	F	0.21	
	M	0.11	
11	F	0.13	0.11
	F	0.11	
	M	0.10	
14	M	0.07	0.14
	F	0.12	
	F	0.22	
16	M	0.06	0.08
	F	0.08	
	F	0.09	
18	M	0.04	0.06
	F	0.07	
	M	0.07	

Swine

Three groups of 3 pigs were treated for 6 days with an oral dose of 5 mg/kg per day of febantel in a granulated formulation. Groups of 3 pigs were sacrificed 6, 14 and 28 days after the last administration and the concentrations of the main metabolites - fenbendazole, fenbendazole sulphoxide and fenbendazole sulphone - were measured in the edible tissues. The fenbendazole and fenbendazole sulphoxide were converted to the sulphone by selective oxidation. The results are summarized in Table XVII. (Raemsch and Dorn, 1982)

Table XVII. Individual tissue concentrations (ppb) in pigs 6, 14 and 28 days after the last oral dose.

Day	Pig No.	Liver	Kidney	Muscle	Fat
12	287	124.3	<5	<5	27.6
	291	105.3	<5	<5	12.8
	294	978.5	25.2	7.6	259.7
20	283	295.5	7.4	<5	40.9
	288	256.7	11.4	<5	8.6
	293	211.2	21.9	<5	8.6
28	284	51.8	<5	<5	10.1
	286	21.8	<5	<5	5.6
	289	98.0	<5	<5	7.3

METHODS OF ANALYSIS FOR RESIDUES IN TISSUES

Sheep

An analytical residue method was initially developed for measuring levels of fenbndazole sulfone (BAY h 6817) in liver. Two other metabolites of febantel were included in the procedure by oxidizing the corresponding sulfide (BAY h 5156, fenbendazole) and sulfoxide (BAY h 7828, fenbendazole sulfoxide) with aqueous permanganate to BAY h 6817 which was quantitated utilizing spectrophotometry. The method consisted of homogenization of tissue in pH 7.0 buffer, extraction of the three metabolites with benzene, partitioning of the metabolites into dilute acid, oxidation of metabolites to BAY h 8617, partitioning into chloroform and quantitation by fluorescence:298 nm excitation and 335 nm emission. Recoveries from kidney, muscle and fat fortified with 0.1 ppm BAY h 5156 (the sulfide) ranged from 66 - 90%. Recoveries from liver fortified with 0.5 ppm BAY h 5156 averaged 77%. Background levels for all tissues were less than 0.01 ppm. (Raemsch, 1976)

Cattle

The analytical residue method as described for sheep was also utilized for analyzing the same metabolites in bovine tissue; liver, kidney, muscle and fat. Recoveries from liver fortified with 0.5 ppm BAY h 5156 averaged 89%, whereas recoveries from the remaining tissues each fortified at 0.1 ppm BAY h 5156 ranged from 55 - 74%. (Hopkins and Rafferty, 1977)

A liquid chromatographic analytical residue method was developed in order to measure residues of BAY k 8449 in iiver. The method consists initially of a digestion of liver tissue in 12 N HCl at 90° for 16 hours, neutralization, partitioning of the desired residue into dichloromethane, cleanup by column chromatography over Sephadex LH-20, subsequent analysis of the desired eluate fraction from the column by HPLC utilizing detection by UV at 254 nm. Background levels for liver from untreated bovine were 0.01 ppm or less. Recoveries from liver fortified at 0.1 and 0.2 ppm were 41 46% (n = 3) and

51 - 59% (n ~ 5), respectively. Liver taken from each of three bovine, at a 10 day withdrawal interval, that had been administered a single oral 7.5 mg/kg dose of [l4C-phenylthio] febantel was analyzed for levels of BAY k 8449 utilizing the described procedure. For each liver sample, the following residue levels were found (total residues based on ^{14}C assay, BAY k 8449 as determined by liquid chromatography, both values in ppm); 2.8, .22; 3.2 .29 and 1.5, .15. It was concluded that approximately 10% of the total drug-related residues could be measured as BAY k 8449. (Waggoner and Bowman, 1986)

The HPLC conditions in the analytical procedure for BAY k 8449 were revised in order to shorten the retention time of febantel, febantel sulfoxide and febantel sulfone and to minimize tissue interferences. Liver from febantel-treated animals was analyzed, but none of the intact febantel residues were found at a sensitivity of 0.01 ppm. (Waggoner, 1987f)

Analyses of bovine liver were conducted for levels of BAY k 8449 from animals orally dosed with 7.5 mg [^{14}C] febantel/kg body weight and sacrificed at 10, 12, 14 and 16 days withdrawal interval. Two liver samples each from a male and female animal were analyzed at each withdrawal interval. Levels of BAY k 8449 found with the liquid chromatographic analytical residue method and corresponding levels of total drug-related residues determined by radioassay are summarized in Table XVIII. (Waggoner, 1987d)

Table XVIII. Total Drug-Related Residues and BAY k 8449 in Bovine Liver.

Withdrawal Interval (Days)		Total Drug-Related Residues (ppm)	BAY k 8449 Found ppm	BAY k 8449 % of Total Residue
10	Male	2.11	0.25	11.8
	Female	1.60	0.20	12.5
12	Male	1.63	0.15	9.2
	Female	1.39	0.16	11.5
14	Male	1.17	0.13	11.1
	Female	1.07	0.14	13.1
16	Male	0.76	0.08	10.5
	Female	0.98	0.13	13.3

Over a withdrawal period of 10 - 16 days, BAY k 8449 accounted for an average of 11.6% of total drug-related residues which remained constant over the withdrawal period investigated varying from 9.2 - 13.3%. Recovery of BAY k 8449 from liver fortified at 0.12 ppm averaged 74%.

A gas chromatographic procedure for separation of BAY k 8449 with a capillary column and detection by selected ion monitoring with mass spectrometry was reported. The procedure consisted of extraction, cleanup by solvent partitioning and column chromatography and initial separation and detection with HPLC/ UV following the analytical residue method in by liquid chromatography. For the confirmatory procedure, the BAY k 8449 fraction which was normally dissolved in mobile phase prior to analysis

by HPLC was, instead, dissolved in ethyl acetate for preparation of a trimethylsilyl derivative for subsequent analysis by mass spectrometry. (Waggoner, 1987g)

Fenbendazole

A cold residue study was also conducted by administering a single oral dose of fenbendazole as a 10% paste to each of six beef cattle at a dosage of 5 mg drug/kg body weight (5.2 - 6.7 mg/kg range). Three animals each were sacrificed at 4 and 8 days withdrawal after treatment. Livers were sampled from each animal and immediately stored frozen. Analyses for levels of BAY k 8449 were then conducted with the liquid chromatographic. Levels of BAY k 8449 in liver from a 4 day withdrawal interval ranged from 0.12 - 0.18 ppm and from an 8 day withdrawal interval ranged from .09 - 0.18 ppm averaging 0.15 and 0.13 ppm, respectively. It was, therefore, demonstrated that residues of BAY k 8449 can be found from either febantel or fenbendazole treated cattle. (Waggoner, 1987h)

APPRAISAL

(See Annex 2 for a combined appraisal of Febantel, Fenbendazole and Oxfendazole).

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