1-2-96

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

January 2, 1996

OFFICE OF
PREVENTION, PESTICIDES, AND
TOXIC SUBSTANCES

MEMORANDUM:

SUBJECT: Atrazine (080803), Reregistration Case No. 0062 and Special Review. Hydroxyatrazine Metabolism in Goats. CBRS No. 12607, DPBarcode No. D195321, MRID 42925601. CBRS No. 15061, DPBarcode No. D211558, MRID 43508501.

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In response to an Agency DCI of 10/90, registrant Ciba-Geigy Corporation submitted data on the nature of the residue when hydroxyatrazine is fed to goats (MRID 42925601), and subsequently submitted a supplement (MRID 43508501). Assignment instructions are to review the submissions. Conclusions and Recommendations below pertain only to data in the present submissions. We note that the initial assignment also included product chemistry submissions, which have been transferred to DPBarcode D221089.

Tolerances are established for residues of the herbicide atrazine, 2-chloro-4-ethylamino-6-isopropylamino-*s*-triazine, in or on agricultural commodities (40 CFR 180.220(a)), and for combined residues of atrazine and its metabolites 2-amino-4-chloro-6-ethylamino-*s*-triazine (G-28279), 2-amino-4-chloro-6-isopropylamino-*s*-triazine (G-30033), and

2-chloro-4,6-diamino-*s*-triazine (G-28273), in or on specified plant commodities (40 CFR 180.220(b)); see Figure 1 for structures. Atrazine is a List A Chemical. The Residue Chemistry Chapter was issued 7/25/83; the Registration Standard (Guidance Document) was issued 9/85; a Second Round Review (SRR) Residue Chemistry Chapter was issued 10/18/88. Special Review has been initiated on triazine herbicides, including atrazine (59 FR 60412, 11/23/94, PD1).

Conclusions

1. Assignments of residues in urine are supported by the data provided (see Figure 2 for structures).

2. Assignments of metabolites in edible commodities were not confirmed by a second technique. However, even with this consideration, the assignment of residues in ruminant commodities is conservative for the purposes of risk assessment. Combined free hydroxy metabolites represented 27% of TRR in liver, and over 59% TRR in kidney, milk, meat, and fat (Table 3). Each of the other residues assigned also contained an intact triazine ring.

3. With the present submission, the nature of the residue in ruminants is adequately understood.

4. For the purposes of exposure assessment, data in the present submission on hydroxyatrazine can be translated to transfer ratios, residues in ruminant tissue:residues in feed, for combined free hydroxy metabolites in feed items (see Table 4).

5. No data are available on the transfer of free hydroxy residues in feed items to poultry commodities. However, available data on poultry are sufficient for exposure assessment under conservative assumptions. While a poultry metabolism study with hydroxyatrazine could allow refinements in exposure assessment, the value of such a study might be marginal. Accordingly, the requirement in reserve for a new poultry metabolism study with hydroxyatrazine, or any other individual metabolite, is withdrawn. The nature of the residue in poultry is adequately understood.

Recommendations

In accordance with Conclusions 3 and 5, the nature of the residue in livestock (Guideline 171-4b) is adequately understood. In accordance with Conclusions 4 and 5, data in the present submission and other available data can be used for exposure assessment.

We recommend that the Registrant be provided a copy of this memo.

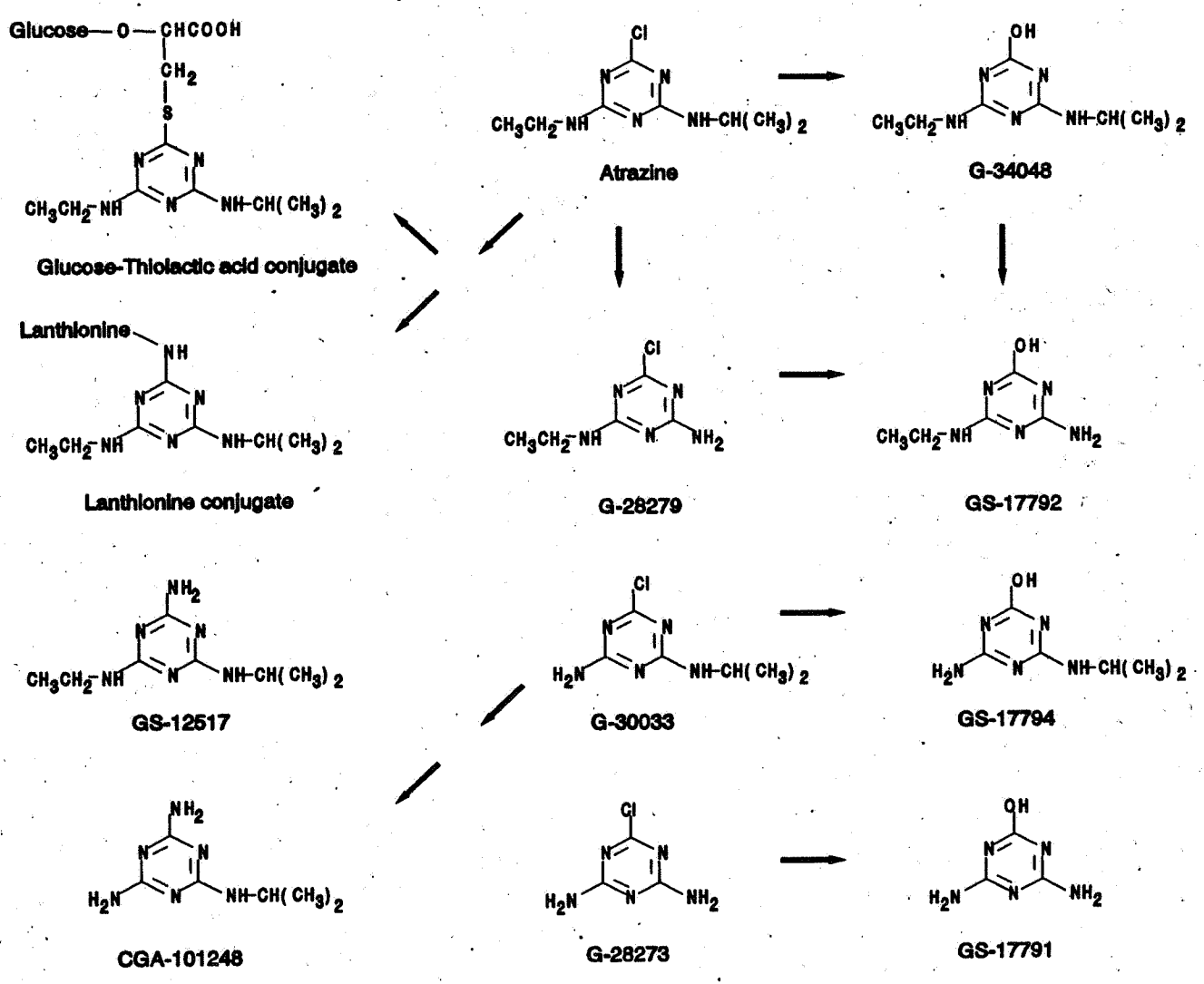

Figure 1. Atrazine metabolites identified in plants.

DETAILED CONSIDERATIONS

Background

A 1990 residue chemistry review of atrazine data noted that all metabolites containing the intact triazine ring were considered of toxicological concern, and data requirements should be revised so that all such metabolites are determined for all commodities for which atrazine is registered (DEB 5783, 5/3/90, M.S. Metzger). The Agency subsequently issued a DCI, received by registrant in 10/90, which superseded the residue chemistry data requirements of all previous DCIs and any other agreements entered into with the Agency pertaining to such requirements. The 10/90 DCI required additional data on livestock metabolism.

Registrant Ciba-Geigy's response to this DCI was reviewed and recommendations for conducting studies were provided (CBRS 9167, 1/22/92, M.S. Metzger). That review noted that available data were sufficient to conclude that metabolism of parent atrazine in livestock was well understood. However, additional data were necessary on the metabolism of hydroxyatrazine in ruminants and poultry (Ibid.). Subsequent review of a request to waive the requirement for additional poultry metabolism data concluded that a poultry metabolism study should be conducted by feeding a specific ratio of triazine ring-containing metabolites corresponding to that likely to be consumed by poultry. Consequently, the requirement for additional poultry metabolism data was reserved pending the outcome of radiolabel field trials for poultry feeds. (CBRS 10919, 12/4/92, M.S. Metzger)

Subsequent to the 10/90 DCI, the HED Metabolism Committee decided that exposure assessment for dietary cancer risk from atrazine would be conducted on the basis of total radioactive residues from radiolabel field trials, or if such data were not available, then with the best data available for estimating total triazine ring residues. The Committee noted that this decision could be altered, depending on the results of the cancer/chronic feeding study with hydroxyatrazine (Memo, 8/7/92, M.S. Metzger).

In response to the Agency DCI of 10/90, registrant Ciba-Geigy submitted data on radiolabeled field metabolism studies. Review of studies on corn and sorghum (CBRS 10980, 6/3/93, and CBRS 13059, 5/22/95, J. Abbotts) and on sugarcane and rotational crops (CBRS 12889, 6/29/95, J. Abbotts) found these submissions acceptable. As part of its comments on the PD1, the Registrant submitted additional metabolism data on corn and sorghum (CBRS 15633, 7/6/95, J. Abbotts) and on sugarcane (CBRS 15632, 7/6/95, J. Abbotts). Additional data on the metabolism of hydroxyatrazine in goats, which had not yet been reviewed, were summarized in comments on the PD1, and the summary was evaluated (CBRS 15634, 8/1/95, J. Abbotts).

With the additional data on plant metabolism and submissions on hydroxyatrazine toxicology, the HED Metabolism Committee revisited its provisional conclusions and recently issued two decisions pertaining to triazine chemicals. With regard to atrazine, the first decision was that the residues of concern for cancer dietary risk are parent and chloro metabolites (Memo,. 9/29/95, J. Abbotts); the chloro metabolites are those in the central pathway in Figure 1. The second decision for atrazine was that for chronic non-cancer dietary risk, evaluations should be performed on two different sets of residues. One evaluation should be based on anticipated residues of combined free hydroxy metabolites, using the RfD for hydroxyatrazine; this RfD will be assigned by TOX for the purposes of dietary risk assessment. The second evaluation should be based on anticipated residues for all other metabolites (total radioactive residues minus free hydroxy metabolites), using the RfD for parent atrazine. (Memo, 11/28/95, J. Abbotts) The free hydroxy metabolites are those on the right side of Figure 1; hydroxyatrazine is designated G-34048. The overall effect of the Metabolism Committee decisions is that three separate dietary risk evaluations should be carried out for atrazine, using different subsets of residues and different toxicological end points.

In response to the 10/90 DCI, the Registrant provided the following documents:

Metabolism of [Triazine-^{14}C]-Hydroxy-Atrazine in Lactating Goats, Ciba-Geigy Corporation, Environmental Health Center, Farmington, CT, Project F-00123, May 6, 1993 (MRID 42925601).

Addendum 1 to F-00123 (MRID 43508501) transmitted January 9, 1995

Review of the present submissions will be conducted with recognition of the recent decisions of the Metabolism Committee.

Animal Protocol

Animal handling and determination of total radioactive residues were carried out at Ciba-Geigy Corporation, Vero Beach Research Center, Vero Beach, FL. The performing laboratory for residue analysis was Ciba-Geigy Corporation, Environmental Health Center, Farmington, CT. The test substance was [^{14}C]-hydroxyatrazine, labeled uniformly in the triazine ring. Radiochemical purity of the test substance was 97%. The test substance was diluted with unlabeled hydroxyatrazine to a specific activity of 46.3 μCi/mg.

The test substance was placed in capsules and administered by balling gun to two lactating goats daily for four consecutive days at dose rates of 143 ppm and 83 ppm (average 113 ppm). Goat No. 1 was described as a finicky eater, and its feed consumption was below normal. Goats were milked twice daily, in the morning and afternoon, and urine and feces were collected once daily.

Animals were sacrificed approximately 6 h after receiving the last dose. Samples of tenderloin, leg muscle, heart, liver, kidney, omental fat, perirenal fat, and whole blood were collected. Samples were shipped frozen to the Environmental Health Center and stored at approximately -20°C until analysis.

Total radioactive residues (TRR) in tissue samples were determined by homogenizing samples, combustion, and liquid scintillation counting (LSC); milk and urine samples were mixed with scintillant and counted directly. Residues are summarized in Table 1. Maximum milk residues were 0.717 ppm (Day 2, PM) for Goat No. 1, and 0.457 (Day 3, PM) for Goat No. 2.

Table 1. Total radioactive residues in goat.

Tissue	Total radioactive residue, ppm:		
	Goat No. 1	Goat No. 2	Average
Milk	0.717	0.457	0.587
Leg muscle	0.20	0.20	0.20
Muscle, tenderloin	0.21	0.21	0.21
Liver	1.06	1.29	1.18
Kidney	2.28	2.86	2.57
Perirenal fat	0.25	0.08	0.16
Omental fat	0.12	0.06	0.09

The Registrant determined that radioactivity in milk and tissues accounted for slightly more than 1% of the administered dose. Radioactivity in urine and feces accounted for 57% or more of the administered dose.

Extraction

Extraction protocols were most extensive for liver, kidney, and tenderloin. Samples were homogenized and extracted in methanol:water (90:10, v/v) four times. The remaining solid was then extracted once with water, and once with acetone. The first three methanol:water extracts, containing most of the extracted radioactivity, were combined and concentrated. The tenderloin extract was passed through a C18 solid phase extraction (SPE) column, combined with a methanol wash, and concentrated. Concentrated extracts were filtered or centrifuged to remove solids, brought to pH 7, and then analyzed by HPLC and LSC.

Perirenal fat was dissolved in hexane, and the hexane was extracted with methanol, four times. The methanol extract was

concentrated and passed through a C18 SPE column. The column effluent was concentrated and analyzed by HPLC and LSC.

Milk samples were extracted by the addition of methanol, mixing, and centrifuging. The supernatant was decanted, and the pellet was washed with methanol and discarded. The methanol wash and first supernatant were combined, concentrated, and partitioned with hexane. Aqueous methanol was separated in a separatory funnel, concentrated, then passed through a C18 SPE column. The column effluent was combined with a methanol wash, concentrated, and analyzed by HPLC and LSC. Urine was adjusted to pH 7 with dilute formic acid, mixed with methanol, and the mixture was analyzed by HPLC and LSC. Table 2 summarizes the distribution of radioactivity in goat tissues into liquid extract and solids:

Table 2. Distribution of radioactive residues in goat tissues.

Sample [TRR, ppm for Goat 1, Goat 2]	% TRR in extract Goat 1, Goat 2	% TRR in solids Goat 1, Goat 2	% TRR, total recovery Goat 1, Goat 2
Urine, day 4 [213, 99]	103.7, 101.4		103.7, 101.4
Milk, day 4 [0.648, 0.430]	99.6, 95.4		99.6, 95.4
Liver [1.06, 1.29]	95.4, 93.3	2.16, 3.77	97.6, 97.1
Kidney [2.28, 2.86]	92.6, 93.2	0.45, 0.50	93.0, 93.7
Tenderloin [0.21, 0.21]	93.2, 96.0	0.73, 0.80	93.9, 96.8
Perirenal fat [0.25, 0.08]	Methanol: 98.9, 89.9 Hexane: 0.35, 0.94	1.80, 2.20	101.0, 93.0

Table note: Extraction protocols were as described in the text.

Residue Analysis

Concentrated extracts were analyzed by HPLC. Two different systems used a Partisil PAC column in series with a Spherex strong cation exchanged (SCX) column. Solvents were acetonitrile as a weak solvent A; 0.1 M ammonium formate, pH 7.0 as a strong solvent B; and methanol as a strong solvent C. With System 1, the mobile phase was isocratic in 88% A, 7% B, and 5% C. With system 2, the columns were eluted with a linear gradient from 94:6 A:B, to 70:20:10 A:B:C. System 3 was reverse phase HPLC with a Partisil ODS-3 column, eluted with a linear gradient from 10% to 30% acetonitrile in 0.1 M ammonium acetate. Residues were detected by radioactivity monitoring and by uv. Some unknowns were assigned based on comparison or co-chromatography with standard compounds.

HPLC analysis of extracts indicated similar chromatographic profiles across tissues. Although the relative proportions of individual metabolites varied with tissue, eight peaks were seen consistently. Table 3 summarizes the levels of metabolites detected in tissue extracts.

The same eight metabolites were observed during HPLC analysis of urine extract. Because levels of radioactivity were much higher in urine, these extracts were used to identify metabolites. Metabolites in urine were isolated using HPLC or thin layer chromatography (TLC). For TLC, silica gel plates were developed 2-3 times to achieve separation of components, in ethyl acetate:chloroform:acetic acid (3:4:3, by v). When metabolites 7 and 8 were treated with ß-glucosidase, a change in their chromatographic profiles was observed. Isolated metabolites were compared to standards by HPLC, and analyzed by gas chromatography/mass spectrometry (GC/MS), using a Finnigan mass spectrometer or a Finnigan triple quad mass spectrometer. Mass spectral analysis was by positive ion (thermospray), electron impact, and/or ammonium DCI. Mass spectra of metabolites were compared to those of standards. Some metabolites were also analyzed by proton nuclear magnetic resonance. Based on combined data from HPLC, GC/MS, and enzyme or chemical treatment, urine metabolites were assigned structures as indicated in Figure 2. Metabolite 3 was not identified because of low levels in edible tissues.

Addendum 1 to the study (MRID 43508501) provided supporting storage stability data. During the metabolism study, goats were sacrificed and samples collected and stored frozen in June 1991. Samples were shipped frozen to Environmental Health Center, Farmington CT, in July 1991, and then were stored at -20°C. The data presented include HPLC profiles of extracts from liver of Goat 2, and day 4 milk from Goat 1. These same samples were re-extracted, and HPLC profiles from November 1992 were shown. Registrant claimed no significant degradation of metabolites, which is supported by the HPLC profiles. As noted above, identification of metabolites was carried out on residues from urine extracts. The dates on mass spectra of the metabolites range from January 1992 to November 1992.

Table 3. Metabolites assigned in goat tissues.

Metabolite Number	Metabolite, % TRR (ppm), in extract of:				
	Milk	Liver	Kidney	Tenderloin	Perirenal fat
1	3.9 (0.02)	44.0 (0.50)	6.4 (0.15)	5.4 (0.01)	4.3 (0.01)
2a	16.8 (0.09)	8.6 (0.10)	5.3 (0.13)	8.1 (0.02)	7.1 (0.01)
3	7.4 (0.04)	5.6 (0.06)	3.4 (0.08)		
4 (G-34048)	46.5 (0.26)	20.2 (0.24)	47.7 (1.18)	54.8 (0.12)	49.2 (0.09)
5 (GS-17794)	4.6 (0.03)	3.3 (0.04)	3.0 (0.08)	3.9 (0.01)	3.1 (0.005)
6 (GS-17792)	9.0 (0.05)	3.3 (0.04)	8.4 (0.21)	13.8 (0.03)	11.8 (0.02)
7			8.6 (0.22)	2.0 (0.004)	9.1 (0.02)
8		2.2 (0.03)	6.3 (0.16)		5.1 (0.01)
Combined free hydroxy metabolites [%TRR]	[60.1]	[26.8]	[59.1]	[72.5]	[64.1]
Total assigned to known compounds	80.8	81.6	85.7	88.0	89.7

Table note: Metabolite levels were based on HPLC analysis. Blank spaces indicate residues not detected. For milk, extracts from goat 1 for days 1 and 4 were combined; for other tissues, extracts from both goats were combined.
See Figure 2 for structures assigned.

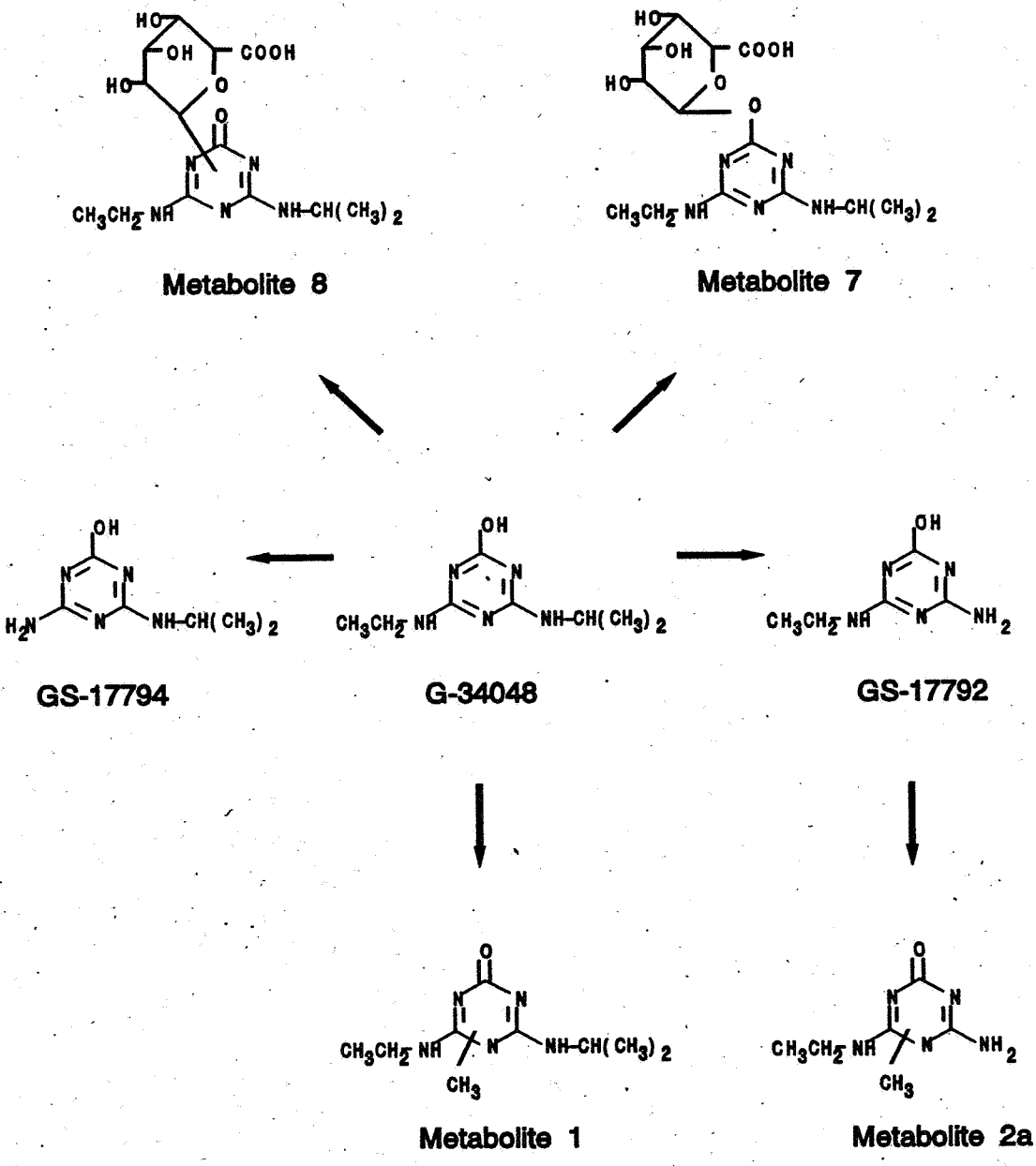

Figure 2. Assignment of hydroxyatrazine (G-34048) metabolites in goat.

CBRS Comments, Residue Analysis

Assignments of residues in urine were well-supported by the data provided. Although extracts from edible commodities showed the same general HPLC profile as urine extracts, no data were provided to show that peaks from tissues co-chromatographed with their corresponding urine peaks using a second technique. Ordinarily, such confirmation by a second technique would be required for metabolites representing greater than 10% TRR and greater than 0.05 ppm.

Despite this apparent deficiency, the recent decisions of the HED Metabolism Committee allow a simplified review. As noted in the Background section above, these decisions direct that for non-cancer chronic dietary risk assessment, one evaluation should be based on combined residues of free hydroxy metabolites, using the RfD for hydroxyatrazine; and a second evaluation should be based on all other residues (total radioactive residues minus free hydroxy metabolites), using the RfD for parent atrazine. The equivalent RfD for hydroxyatrazine is about 3.5 times lower than the RfD for parent atrazine (Metabolism Committee Briefing Memo, 10/10/95, J. Abbotts). Therefore, assignment of residues that overstated the contribution from free hydroxy metabolites would be conservative for risk assessment.

In the goat study, HPLC profiles were compared with the mobilities of free hydroxy metabolites, including GS-17791, as standards. If peaks assigned as free hydroxy metabolites were in fact other compounds, then the assignment would overstate the levels of these metabolites. As it is, unmetabolized hydroxyatrazine represents about half of TRR in most tissues, and combined residues assigned to free hydroxy metabolites represent over 70% TRR in meat (Table 3). The assignment of residues in goats therefore seems conservative, even if potential deficiencies are taken into account. The other metabolites assigned contain intact triazine rings, which supports the assumption of the Metabolism Committee that TRR represents total triazine ring residues (Memo, 9/11/95, J. Abbotts). These considerations lead to the following comments:

Conclusion 1: Assignments of residues in urine are supported by the data provided (see Figure 2 for structures).

Conclusion 2: Assignments of metabolites in edible commodities were not confirmed by a second technique. However, even with this consideration, the assignment of residues in ruminant commodities is conservative for the purposes of risk assessment. Combined free hydroxy metabolites represented 27% of TRR in liver, and over 59% TRR in kidney, milk, meat, and fat (Table 3). Each of the other residues assigned also contained an intact triazine ring.

As noted above, previous review concluded that metabolism of parent atrazine in animals was well understood, but additional data were necessary on the metabolism of hydroxyatrazine in ruminants and poultry (CBRS 9167, 1/22/92, M.S. Metzger). The present submission is sufficient for hydroxyatrazine data in ruminant.

Conclusion 3: With the present submission, the nature of the residue in ruminants is adequately understood.

Transfer Ratios to Animal Tissues

The data in the present submission are relevant to dietary exposure assessment in accordance with the recent decisions of the HED Metabolism Committee. As noted above, those decisions direct that assessment of non-cancer chronic risk be based on two sets of residues. Once the magnitude of these residue sets is determined in feed items, it will be necessary to determine four residue subsets in animal commodities: free hydroxy metabolites in feed items transferred to animal tissues as free hydroxy metabolites; all residues other than free hydroxy metabolites ("other residues") in feed items transferred as free hydroxy metabolites; free hydroxy residues in feed items transferred as other residues; other residues in feed items transferred as other residues. The present submission contains data on transfer of hydroxyatrazine residues in feed items to animal commodities. Table 4 is constructed using data from Tables 1 and 3:

Table 4. Transfer ratios for hydroxyatrazine, residues in animal tissue:residues in feed.

Tissue	Residues in animal tissues, ppm [transfer ratio]:		
	TRR, ppm	Combined free hydroxy residues, ppm (% TRR)	"Other" residues (TRR minus combined free hydroxies), ppm
Milk	0.587	0.352 (60.1) [0.0031]	0.234 [0.0021]
Meat	0.21	0.152 (72.5) [0.0013]	0.058 [0.0005]
Liver	1.18	0.316 (26.8) [0.0028]	0.863 [0.0076]
Kidney and other meat byproducts	2.57	1.52 (59.1) [0.013]	1.05 [0.0093]
Fat	0.16	0.103 (64.1) [0.001]	0.057 [0.0005]

Table notes: TRR data are taken from Table 1. Data on % TRR represented by combined free hydroxy metabolites are taken from Table 3. Transfer ratios were calculated based on an average feeding level of 113 ppm hydroxyatrazine.

Chemistry Branch previously determined anticipated residues for atrazine, including anticipated residues in animal commodities, using metabolism and/or feeding data then available. Anticipated residues were initially determined for atrazine and chloro metabolites (DEB 3688-3703, 3756, 9/14/88, M.S. Metzger). Anticipated residues were subsequently revised by assuming that total radioactive residues represented total triazine ring residues (DEB 5783, 5/3/90, M.S. Metzger). That revision used data from studies in which livestock were fed items containing "biosynthesized" atrazine residues; these were crops treated with ^{14}C-atrazine, then harvested and fed to livestock. The nature of the biosynthesized residues was not determined, but it was assumed that these residues were more representative of actual residues in feed items than parent or any individual metabolite alone. Data from cows, goats, and poultry used in that revision are summarized in Tables 5, 6, and 7, respectively, and transfer ratios have been calculated:

Table 5. Transfer ratios from previous studies with cattle.

Labeled chemical	Dose in feed, ppm	Total radioactive residues in animal tissues, ppm [Ratio, residues in tissue:residues in feed]					
		Liver	Kidney	Meat	Fat	Heart	Milk
Atrazine	28	3.58 [0.128]		1.09 [0.039]	0.26 [0.073]		0.67 [0.024]
	6.8	0.87 [0.128]		0.24 [0.035]	0.15 [0.022]		0.12 [0.018]
	0.62	0.11 [0.177]		0.02 [0.032]	0.01 [0.016]		0.01 [0.016]
G-34048	0.62	0.007 [0.011]	0.004 [0.006]	0.0006 [0.001]	<0.0005 [<0.001]	0.0008 [0.001]	0.003 [0.005]

Table notes: Data are taken from DEB 5783, 5/3/90, M.S. Metzger. Transfer ratios have been calculated here. Radiolabel was ^{14}C in all cases. See Figure 1 for structures of metabolites. Blank spaces indicate residues not determined in given tissues.

Table 6. Transfer ratios from previous studies with goats.

Labeled chemical	Dose in feed, ppm	Total radioactive residues in animal tissues, ppm [Ratio, residues in tissue:residues in feed]					
		Liver	Kidney	Meat	Fat	Heart	Milk
Atrazine	44	4.6 [0.105]	3.45 [0.078]	1.13 [0.026]		1.05 [0.024]	0.70 [0.016]
	33	5.16 [0.156]	3.32 [0.101]	0.95 [0.029]	0.10 [0.003]		0.63 [0.019]
	27.2	3.03 [0.111]	1.76 [0.065]	0.54 [0.020]	0.06 [0.002]	0.54 [0.020]	0.89 [0.033]
	5	1.26 [0.252]	0.81 [0.162]	0.13 [0.026]	0.06 [0.012]	0.15 [0.030]	0.16 [0.032]
G-28273	5.8	1.3 [0.224]	0.98 [0.169]	0.34 [0.059]	0.09 [0.016]		0.172 [0.030]
Biosyn, corn silage	0.95	0.01 [0.011]	0.003 [0.003]	0.0008 [0.0008]	0.0008 [0.0008]	<0.0006 [<0.0006]	0.004 [0.004]
Biosyn, corn grain	0.012	0.0006 [0.05]	<0.0006 [<0.05]	<0.0006 [<0.05]	<0.0006 [<0.05]	0.0006 [0.05]	0.0001 [0.008]
Biosyn, sorghum fodder	1.47	0.068 [0.046]	0.015 [0.010]	0.002 [0.001]	<0.001 [<0.001]	0.003 [0.002]	0.003 [0.002]
Biosyn, corn grain	0.32	0.036 [0.112]	0.01 [0.031]	<0.006 [<0.019]	<0.006 [<0.019]	<0.006 [<0.019]	0.003 [0.009]

Table notes: Data are taken from DEB 5783, 5/3/90, M.S. Metzger. Transfer ratios have been calculated here. Radiolabel was ^{14}C in all cases. "Biosyn" describes residues on feed items indicated from crops treated with labeled atrazine. See Figure 1 for structures of metabolites. Blank spaces indicate residues not determined in given tissues.

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Table 7. Transfer ratios from previous studies with chickens.

Labeled chemical	Dose in feed, ppm	Total radioactive residues in animal tissues, ppm [Ratio, residues in tissue:residues in feed]					
		Liver	Kidney	Meat	Fat	Heart	Eggs: White/Yolk
Atrazine	58	3.32 [0.057]	4.62 [0.079]	2.76 [0.048]	1.77 [0.031]	2.4 [0.041]	1.4/2.6 [0.024/0.045]
	50	3.15 [0.063]		3.40 [0.068]			1.15/2.5 [0.023/0.05]
G-28273	5	0.55 [0.11]	0.90 [0.18]	0.50 [0.10]	0.04 [0.008]		0.21/0.36 [0.042/0.072]
Biosyn, corn grain	0.047	0.013 [0.277]	0.009 [0.191]	ND	ND		0.008/0.01 [0.170/0.213]

Table notes: Same as Table 6.
ND=residues not detected, limit of detection not provided.

In the three tables above, only one study with hydroxyatrazine (G-34048) is included (Table 5). When the transfer ratios from that study in cattle are compared with transfer ratios for TRR in goat from the present submission (obtained by adding both transfer ratios in Table 4 for each tissue), one observes that the ratios are nearly identical for milk and liver, and the transfer ratios in Table 4 are slightly higher for meat, kidney, and fat. Because of the high feeding level used for Table 4, it was possible that transfer ratios could be artificially low because of plateau effects. The fact that the ratios are similar to those in Table 5, at a much lower feeding level for hydroxyatrazine, indicates that this consideration is not of concern. Previous review noted that for the same feeding levels in cattle, residues from hydroxyatrazine were about an order of magnitude lower in tissues, and 3-fold lower in milk, than residues from feeding atrazine (DEB 5783, 5/3/90, M.S. Metzger). The data from the present submission (Table 4) reinforce this conclusion, and the transfer ratios are conservative compared to the previous data. For the purposes of exposure assessment, and in the absence of data on transfer of other hydroxy metabolites, the hydroxyatrazine data will be used to represent transfer of combined free hydroxy metabolites. These considerations lead to the following comment:

Conclusion 4: For the purposes of exposure assessment, data in the present submission on hydroxyatrazine can be translated to transfer ratios, residues in ruminant tissue:residues in feed, for combined free hydroxy metabolites in feed items (Table 4).

The data in Tables 4 through 6 indicate that when ruminants were fed biosynthesized residues, the transfer ratios were closer to those for hydroxyatrazine than for atrazine or the chloro metabolite G-28273. Because the biosynthesized residues were not characterized, it is difficult to draw conclusions from these studies about transfer ratios for residues other than those fed alone.

In contrast, transfer ratios for poultry were generally higher with biosynthesized residues than with atrazine or G-28273 in the feed (Table 7). This observation leads to consideration of the need for additional poultry studies: As indicated in the Background section above, additional data were previously required on the metabolism of hydroxyatrazine in poultry (CBRS 9167, 1/22/92, M.S. Metzger). Review of a request to waive this requirement concluded that a poultry metabolism study should be conducted by feeding a specific ratio of triazine ring-containing metabolites corresponding to that likely to be consumed by poultry. Consequently, the requirement for additional poultry metabolism data was reserved pending the outcome of radiolabel field trials for poultry feeds (CBRS 10919, 12/4/92, M.S. Metzger).

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Figure 1 indicates a simplified version of atrazine metabolism in plants. The pathways shown are dealkylation of atrazine to produce chloro metabolites (central pathway); conversion of the chloro metabolites to free hydroxy metabolites (right pathway); and conjugation with glutathione followed by rearrangement and/or additional metabolism (left pathway; multiple arrows indicate putative intermediates not isolated). Figure 1 depicts metabolites identified in mature sugarcane cane; in immature sugarcane leaves, nine additional metabolites were identified, most of them associated with the glutathione pathway; all 21 metabolites together represented 45% of TRR, and only one individual metabolite represented as much as 11% (CBRS 15632, 7/6/95, J. Abbotts). Additional metabolism data indicated that the assumption that total radioactive residues represents total residues containing an intact triazine ring is reasonable (CBRS 15634, 8/1/95, J. Abbotts).

The fact that atrazine metabolism in plants is extensive, and no single residue represents a major portion of TRR across crops, indicates that determining the precise ratio of specific triazine ring residues in poultry feed items would be impractical. In the determination of atrazine anticipated residues, poultry feed items in national commerce with atrazine uses are corn grain and sorghum grain (DEB 5783, 5/3/90, M.S. Metzger; Memo, 6/7/93, J. Abbotts). In these commodities, combined free hydroxy metabolites represent about 13% and 7% TRR, respectively (Metabolism Committee Briefing Memo, 10/10/95, J. Abbotts). Therefore, although data are not available to meet the Metabolism Committee's specifications for exposure assessment, a poultry metabolism study with hydroxyatrazine or another free hydroxy metabolite would not be particularly informative.

Data with biosynthesized residues (Table 7, bottom row) appear to provide the most realistic representation of residue transfer from poultry feed items treated with atrazine. A major drawback in these data is a lack of transfer ratios for meat and fat, because limits of detection were not provided. However, the study with biosynthesized residues fed to goat at 0.32 ppm in the feed (Table 6, bottom row), and the poultry study with biosynthesized residues were submitted (MRIDs 41209808 and 41209807, respectively) and reviewed at the same time (DEB 5783, 5/3/90, M.S. Metzger). For both studies, animals were fed grain from corn treated preemergence with ^{14}C-atrazine at 3.0 lb ai/A. An examination of MRID 41209808 indicates that the limit of detection in goat tissues was 0.006 ppm, based on average background counts and the amount of sample combusted. According to both MRIDs, the same SOP for combustion was used in each study, and blood samples combusted were 0.2 g in each study. It therefore seems reasonable to assume that the limit of detection in poultry tissues was also 0.006 ppm. If this value is used for meat and fat in Table 7, the corresponding transfer ratios for TRR are $\leq$0.128 for each commodity. This value for meat is

comparable to the ratio for G-28273, and for fat is higher than ratios for atrazine or G-28273 (see Table 7).

Using data from the poultry feeding study with biosynthesized residues for exposure assessment therefore seems a conservative approach. The available data allow use of transfer ratios on at least a TRR:TRR basis. Anticipated TRRs determined in poultry commodities could be applied to either residue subset of combined free hydroxy residues, or "other" residues. These considerations lead to the following comments:

Conclusion 5: No data are available on the transfer of free hydroxy residues in feed items to poultry commodities. However, available data on poultry are sufficient for exposure assessment under conservative assumptions. While a poultry metabolism study with hydroxyatrazine could allow refinements in exposure assessment, the value of such a study might be marginal. Accordingly, the requirement in reserve for a new poultry metabolism study with hydroxyatrazine, or any other individual metabolite, is withdrawn. The nature of the residue in poultry is adequately understood.

cc:Circ, Abbotts, RF, Atrazine List A File, Atrazine SF
RDI:ARRathman:12/5/95:RBPerfetti:12/15/95:EZager:12/19/95
7509C:CBII-RS:JAbbotts:CM-2:Rm805A:305-6230:1/2/96
JA15\atrazine.18