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Metabolism of Glyphosate in Sprague–Dawley Rats: Tissue Distribution, Identification, and Quantitation of Glyphosate-Derived Materials following a Single Oral Dose[1]

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Metabolism of Glyphosate in Sprague–Dawley Rats: Tissue Distribution, Identification, and Quantitation of Glyphosate-Derived Materials following a Single Oral Dose. BREWSTER, D. W., WARREN, J., AND HOPKINS, W. E., II (1991). *Fundam. Appl. Toxicol.* **17,** 43–51. Five groups of male Sprague–Dawley rats were orally administered a mixture of [^{14}C]- and [^{12}C]-glyphosate (*N*-phosphonomethylglycine) at a dose level of 10 mg/kg body weight. The majority of radioactivity 2 hr after administration was associated with the gastrointestinal contents and small intestinal tissue. Approximately 35–40% of the administered dose was absorbed from the gastrointestinal tract, and urine and feces were equally important routes of elimination. The total body burden 7 days after administration was approximately 1% of the administered dose and was primarily associated with the bone. Total recovery for this study ranged from 95 to 102% of the administered dose. Metabolic profiles of tissues containing greater than 1% of the administered dose at various times after administration indicated that nearly 100% of the body burden of radioactivity was present as unmetabolized parent glyphosate. A minor component constituting <0.1% of the administered dose (<0.4 ppm) was observed in colon tissue from animals 2 hr after the administration of glyphosate and was also present in the GI contents of one animal 28 hr after administration of the radiolabel. The retention time for this metabolite was similar, but not identical, to the retention time for AMPA (aminomethylphosphonic acid), the major bacterial metabolite of glyphosate found in soil. Tissue extraction efficiency was always greater than 90% and stability assays indicated no significant effect of storage on either parent glyphosate or AMPA. The results from this study indicate that virtually no toxic metabolites of glyphosate were produced since there was little evidence of metabolism and essentially 100% of the body burden was parent compound with no significant persistence of material. © 1991 Society of Toxicology.

Glyphosate (*N*-phosphonomethylglycine) is a widely used broad spectrum postemergence herbicide which has been commercially available since 1974 and is commercially formulated as Roundup® Herbicide which contains glyphosate as the isopropylamine salt and a surfactant vehicle. Since glyphosate competitively inhibits enolpyruvyl-shikimate-phosphate synthase (EPSP-synthase), an enzyme which is absent in animals, it is selectively toxic to plants and relatively nontoxic to animals (acute oral rat LD50 ~5.6 g/kg; Street *et al.*, 1979; Monsanto, 1989). EPSP-synthase catalyzes the conversion of shikimate 3-phosphate to 5 - enolpyruvyl - shikimate - 3 - phosphate which is the precursor for essential aromatic amino acids (Mousdale and Coggins, 1984; Rubin *et al.*, 1984; Malik *et al.*, 1989). There is very little metabolism of glyphosate by plants (Newton *et al.*, 1984); however, it is

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readily degraded in soil where it is metabolized to aminomethylphosphonic acid (AMPA) and CO_2 (Sprankle *et al.*, 1975; Rueppel *et al.*, 1977; Mueller *et al.*, 1981).

Results from a previous study (Duerson and Sipes, 1987) indicated that glyphosate is rapidly eliminated from the body and that the total body burden is less than 2% of the administered dose 24 hr after administration. The purposes of this study were to determine the tissue distribution of glyphosate at selected times following a single oral dose and to determine the nature of the glyphosate-derived materials in tissues containing greater than 1% of the administered dose.

MATERIALS AND METHODS

Test Material

The test material for this study was prepared using ^{14}C-labeled and unlabeled (^{12}C) glyphosate (*N*-phosphonomethylglycine) mixed in proportion such that the specific activity was 8.08 mCi/mmol. The chemical purities were greater than 99% and the radiochemical purity was greater than 98% as determined by HPLC. The test material was synthesized by Monsanto Agricultural Co. (St. Louis, MO).

Animal Husbandry

Male CRL:CD (SD) Br rats were purchased from Charles River Breeding Laboratories (Portage, MI) in the weight range of 115–125 g and were approximately 5 to 6 weeks of age at time of dosing. The animals were housed in stainless-steel suspension metabolism cages with a screen bottom to separate urine from feces and kept on a 12-hr light/12-hr dark cycle. The rats were supplied Certified Purina Rodent Chow pellets from Ralston Purina (St. Louis, MO) and tap water *ad libitum* throughout the study.

Experimental Design

All animals were fasted for approximately 16–19 hr prior to dosing and were administered glyphosate (100 μCi) dissolved in saline (pH 7; 5 ml/kg) via oral intubation. Food was returned to the animals approximately 6 hr after dosing.

At approximately 2, 6.3, 28, 96, and 168 hr after administration the animals were terminated by CO_2 asphyxiation and blood and tissues were collected. These time points were chosen based upon a previous pharmacokinetic study of glyphosate in the rat (W.P. Ridley, personal communication, 1990, Monsanto Agricultural Co., St. Louis, MO) and correspond to the time of maximum blood concentration (C_{max}, 2 hr), $t_{\frac{1}{2}}$ of the α phase (6.3 hr), inflection point between the α and the β phases (28 hr), midpoint between the end point and inflection point (96 hr), and the end point (168 hr).

Radiochemical Analysis of Excreta and Tissues

Urine and feces were collected at 2 and 6.3 hr and at 24-hr intervals up to 168 hr after administration. Fresh urine samples were prepared for liquid scintillation counting (LSC) by adding ammonium bicarbonate (0.5 M) and Instagel or Ultima Gold. The fecal samples were combined with distilled water, homogenized, and weighed. Duplicate portions of the resulting fecal homogenates were combusted in a Packard Tricarb B306 sample oxidizer (Downers Grove, IL) and processed for LSC. Combustion efficiencies ranged from 95 to 100%.

Blood samples were collected from all animals at termination and a portion of the sample was separated into plasma and cells by centrifugation. The contents of the stomach, small intestine, cecum, and large intestine were extruded from their respective organs and combined into a single tared vial. The GI tract was rinsed two times with 10–30 ml of saline which was added to the combined gastrointestinal contents.

Duplicate samples of tissues were combusted and processed for LSC. Individual carcasses were frozen, freeze-fractured, lyophilized, and ground in a blender, and a minimum of five aliquots of the resulting powder were combusted for LSC. Blood, abdominal adipose tissue, and testicular adipose tissue weights were estimated to be 8, 5.5, and 5.5% of the body weight, respectively (Brewster and Birnbaum, 1987). Bone was estimated to compose approximately 8% of the body weight.

All samples were counted with a Mark III liquid scintillation spectrometer (Model 81, TM Analytic) or a Packard Tricarb 460 CD liquid scintillation system (Packard Instrument Co., Downers Grove, IL). Counting efficiencies were determined by external standardization and disintegrations per minute were calculated by the Mark III DPM calculation accessory using a set of Searle quench standards. Aqueous samples prepared in Instagel were counted with typical efficiencies of 82–85%, while combusted samples had typical efficiencies of 65–75%.

Statistical Procedures

The data are presented as the mean ± standard error of the mean for the number of animals indicated. Tissue, urine, fecal, and whole body elimination was analyzed as a function of time and the half-life was calculated by nonlinear regression using RS-1 Fit Function where the half-

TABLE 1

SUMMARY OF RECOVERED RADIOACTIVITY FROM RATS AFTER ORAL ADMINISTRATION OF [^{14}C]GLYPHOSATE[a]

	Hours after administration				
	2	6.3	28	96	168
Urine[b]	3.46 ± 1.00	17.96 ± 0.40	41.59 ± 5.31	39.13 ± 10.31	36.29 ± 3.91
Feces[b]	NS[c]	NS	34.58 ± 8.93	49.41 ± 10.33	50.72 ± 6.21
Tissues	39.04 ± 2.27	26.86 ± 0.89	3.93 ± 0.38	1.86 ± 0.53	1.13 ± 0.05
CW[d]	NS	1.52 ± 1.08	9.28 ± 1.60	6.55 ± 1.89	6.59 ± 3.04
GI cont.[e]	51.33 ± 1.7	49.23 ± 0.15	7.03 ± 0.33	0.09 ± 0.02	0.06 ± 0.05
Carcass	3.30 ± 0.10	5.94 ± 1.16	2.40 ± 0.21	1.12 ± 0.19	0.91 ± 0.13
Total recovery[f]	97.13 ± 0.29	101.50 ± 1.47	98.81 ± 5.06	98.15 ± 4.15	95.71 ± 2.63

[a] Percent of administered dose, mean ± SEM of three to four animals/time period.
[b] Cumulative excretion.
[c] No sample at this time point.
[d] Cages rinsed with water.
[e] Gastrointestinal contents.
[f] This number reflects total recovery from all tissues, organs, and excreta analyzed for these animals including the carcasses.

life is equal to -0.693/elimination rate constant (decay rate) and the decay rate is equal to the slope of the line (Klaassen, 1986).

Metabolite Identification

Tissue extraction. Tissues (with the exception of bone) having greater than 1% of the administered dose were analyzed for metabolites. Tissues were stored frozen until extracted with equal amounts of 1 N HCl and chloroform and all extractions were stored refrigerated until analyzed. Stability samples were prepared to verify that glyphosate did not degrade over time in selected tissue matrices.

HPLC analyses. The HPLC instrumentation used in this study consisted of a Varian 5500 high performance liquid chromatograph with detection by a Beckman 171 radioisotope detector. Cation exchange HPLC of the tissue samples was conducted on a Bio-Rad A-9 4.6 × 300-mm HPLC column (Cat. No. 999-1143-300) preceded by a Brownlee 4.6 × 30-mm PRP-1 guard column. The column was maintained at 50°C. The mobile phase consisted of 0.005 M KH_2PO_4 in 96:4 water:methanol with 0.02% EDTA adjusted to pH 2.1 with 85% phosphoric acid. The flow rate of the mobile phase was 0.7–0.8 ml/min. The HPLC effluent was mixed with DuPont Atomflow (scintillant:HPLC effluent ratio of 3:1) and passed through a 1.0-ml RAD cell.

Metabolite Characterization

Metabolite and parent glyphosate material were identified by comparing retention times to known standards. Glyphosate and its major metabolite, AMPA, and a minor contaminant in some glyphosate samples, MAMPA (*N*-methylaminomethylphosphonic acid), were synthesized by Monsanto Agricultural Co.

RESULTS

Material Balance

Animals were administered 10.44 ± 0.09 mg glyphosate/kg body weight containing 1.42 ± 0.04 × 10^8 dpm, as determined by aliquots of the dosing solution at the time of dosing (data not shown). At this dose glyphosate produced no adverse effects on body weight gain and no other signs of toxicity were observed. The total recovery for glyphosate-derived radioactivity was always greater than 95% (Table 1).

Absorption and Elimination

Approximately 36 and 51% of the administered dose was eliminated in the urine and feces, respectively, over the 7-day observation period (Table 1). Since material appearing in the urine after oral administration must rep-

TABLE 2

URINE, FECAL, AND WHOLE BODY PHARMACOKINETIC ELIMINATION PARAMETERS OF GLYPHOSATE DERIVED MATERIAL AFTER SINGLE ORAL ADMINISTRATION OF 10 mg GLYPHOSATE/kg BODY WEIGHT

	Pool size (% dose)	Decay rate (hr^{-1})	Half-life (hr)
Urine	1.14 ± 0.05	−0.0118 ± 0.0006	58.7
Feces	1.45 ± 0.07	−0.0147 ± 0.0008	47.1
Whole body	1.62 ± 0.05	−0.0134 ± 0.0005	51.7

resent absorbed material these results indicate that a minimum of 36% of an oral dose was absorbed from the gastrointestinal tract. This is comparable to previous studies using iv and oral administrations where 30–35% of the administered dose was estimated to be absorbed (W. P. Ridley, personal communication, 1990, Monsanto Agricultural Co.).

Urine and fecal elimination best fit a one-compartment model and the whole body half-life was calculated to be approximately 2 days (Table 2). Fecal elimination seemed to have a greater impact on whole body elimination than did elimination of material in the urine.

Tissue Distribution

The majority of radioactivity 2 hr after administration was associated with the gastrointestinal (GI) contents and tissues (Table 1). With time, greater than 85% of the radioactivity appeared in the urine and feces. The only tissues containing greater than 1% of the administered dose at any time period were the small intestine, bone, colon, and kidney (Table 3). The major tissue depot for glyphosate-derived radioactivity was the small intestine which contained greater than 34% of the dose 2 hr after administration. The majority of the activity in this organ was believed to be associated with intestinal cells since specific procedures were implemented to demonstrate that this radioactivity was not associated with

TABLE 3

TISSUE DISTRIBUTION (% ADMINISTERED DOSE) OF GLYPHOSATE DERIVED RADIOACTIVITY AT SELECTED TIME INTERVALS AFTER ORAL ADMINISTRATION OF 10 mg [^{14}C]GLYPHOSATE/kg BODY WEIGHT[a]

	Hours after administration				
Tissue/organ[b]	2	6.3	28	96	168
Abdominal fat[c]	0.15 ± 0.07	0.16 ± 0.02	0.15 ± 0.13	<0.02	<0.02
Blood[c]	0.38 ± 0.04	0.33 ± 0.00	0.06 ± 0.03	<0.02	<0.02
Bone[c]	2.03 ± 0.13	4.69 ± 0.22	2.72 ± 0.49	1.69 ± 0.04	1.06 ± 0.04
GI contents	51.33 ± 1.70	49.23 ± 0.15	7.03 ± 0.33	0.09 ± 0.02	0.06 ± 0.05
Colon	0.73 ± 0.45[d]	1.29 ± 0.40	0.20 ± 0.03	0.02 ± 0.00	<0.02
Carcass	3.30 ± 0.10	5.94 ± 1.16	2.40 ± 0.21	1.12 ± 0.19	0.91 ± 0.13
Kidney	0.73 ± 0.07	1.29 ± 0.08	0.13 ± 0.01	<0.02	<0.02
Liver	0.12 ± 0.05	0.17 ± 0.01	0.12 ± 0.01	0.06 ± 0.02	0.02 ± 0.00
Sm. intestine[e]	34.34 ± 2.30	18.48 ± 1.10	0.51 ± 0.01	0.05 ± 0.02	0.02 ± 0.01
Stomach	0.11 ± 0.03	0.13 ± 0.02	<0.02	<0.02	<0.02
Testicular fat[c]	0.39 ± 0.25	0.27 ± 0.11	0.02 ± 0.03	<0.02	<0.02
Total body burden	91.21	76.04	10.94	1.91	1.16

[a] Mean ± SEM of three to four animals.

[b] Less than 0.02% of the applied dose was found in the brain, heart, lungs, spleen, and testes at each time point.

[c] Abdominal fat, blood, bone, and testicular fat were estimated to be 5.5, 8, 8, and 5.5% of the body weight, respectively.

[d] The colon of one animal contained less than 1% of the administered dose.

[e] Tissue washed with saline. Data represent that activity associated with tissue and not intestinal contents.

the intestinal contents or mucous layers. Levels of radioactivity in the small intestine declined rapidly with time (Fig. 1).

Bone was also observed to contain significant amounts of radioactivity and approximately 5% of the dose was associated with the bone 6.3 hr after administration (Table 3). In contrast, the colon and kidney contained less than 1.5% of the administered dose at 6.3 hr. Up to 6% of the administered dose was detected in the carcass at 6.3 hr after dosing and less than 1% of the dose was observed in all other tissues examined. The liver to fat ratio was approximately 1:1 at all time periods.

The small intestine, kidney, bone, and colon were found to have the highest tissue to blood ratios (Table 4). Tissue to blood ratios significantly in excess of one are indicative of tissue deposition and there were few tissues with tissue to blood ratios significantly higher than unity at all time points. The tissue to blood ratio increased in the bone with time suggesting a slower elimination from the bone compared to the blood. At the early time periods only the small intestine and colon contained greater than 15 ppm (data not shown).

TABLE 4

TISSUE TO BLOOD RATIOS OF GLYPHOSATE DERIVED RADIOACTIVITY AT SELECTED TIMES AFTER ORAL ADMINISTRATION OF 10 mg [^{14}C]GLYPHOSATE/kg BODY WEIGHT

	Hours after administration				
Tissue/organ	2	6.3	28	96	168
Abdominal fat	0.57	0.71	2.34	0.91	0.58
Blood plasma	1.81	2.01	1.03	0.60	0.45
Bone	5.51	14.20	89.40	173.00	131.00
Brain	0.05	0.19	0.86	3.11	1.98
Colon	14.10	35.20	50.90	16.40	9.83
Heart	0.40	0.36	0.70	1.48	1.09
Kidney	14.80	31.40	34.70	13.30	5.55
Liver	0.52	0.99	5.33	9.08	2.51
Lung	0.70	0.71	2.01	5.30	3.88
Red cells	0.33	0.41	0.63	1.97	4.44
Small intestine	285.00	220.00	59.40	20.00	9.67
Spleen	0.26	0.49	3.01	3.93	1.97
Stomach	3.32	5.48	3.92	5.14	2.35
Testes	0.23	0.39	0.88	1.03	0.86
Testicular fat	1.42	1.17	0.96	1.02	0.57

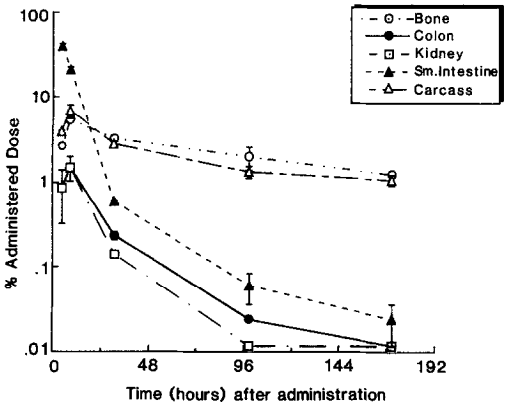

FIG. 1. Elimination of glyphosate-derived radioactivity from various tissues containing >1% of the administered dose after a single oral administration of 10 mg glyphosate/kg body weight (each point represents the mean ± SEM of three to four animals). The calculated pharmacokinetic elimination parameters were as follows:

	Decay rate (hr^{-1})		Half-life (hr)	
Tissue	α	β	α	β
Bone	−0.122 ± <0.001	−0.008 ± <0.001	5.7	91.6
Colon	−0.099 ± <0.001	−0.011 ± <0.001	7.0	60.6
Kidney	−0.116 ± <0.001	−0.012 ± <0.001	6.0	56.9
Sm. int.	−0.081 ± 0.023	−0.035 ± 0.036	8.6	20.0

Tissue Elimination

Other than in the small intestine, glyphosate-derived material appeared to reach maximal tissue levels at 6.3 hr after administration. Tissue levels declined rapidly with time in all tissues except bone (Fig. 1). Elimination from these tissues followed a two-component decay process consisting of a relatively short α phase followed by a much longer β phase. The α phase of elimination for all tissues ranged from 1.3 hr in the liver to 10.3 hr in the blood while the slower β phase ranged from 20 hr in the small intestine to over 90 hr in the bone.

Storage Stability

When GI contents and kidney tissue from untreated animals were homogenized and spiked with a glyphosate:AMPA mixture (~80:20) then analyzed at various times throughout the course of the study, no loss in stability of either component in these tissue matrices was noted (Fig. 2).

Metabolite Analysis

A mixture of glyphosate and AMPA standards was analyzed by HPLC as described under Materials and Methods. Under the conditions employed in this study, glyphosate had a retention time of 8–9 min and AMPA had a retention time of 16–17 min (Fig. 3). The retention time of MAMPA was 14–15 min.

Under these conditions all tissue extracts, except the colons from two animals 2 hr after administration and the GI contents of one animal 28 hr after the administration of glyphosate, had retention times similar to parent glyphosate (Table 5). Parent glyphosate always constituted at least 94% or greater of the tissue radioactivity and the HPLC recovery ranged from 92 to 105%. A carcass sample containing

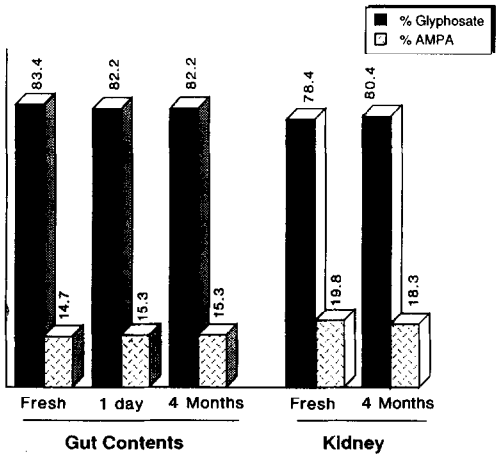

FIG. 2. Stability of glyphosate in gastrointestinal contents and kidney samples at various times after spiking. Gut contents and kidney tissue from untreated animals were homogenized and spiked with 80–85% glyphosate and 15–20% AMPA, then extracted and analyzed by HPLC as described under Materials and Methods. HPLC recovery (determined by dpm recovered from column/dpm applied to column ×100) was always greater than 97%.

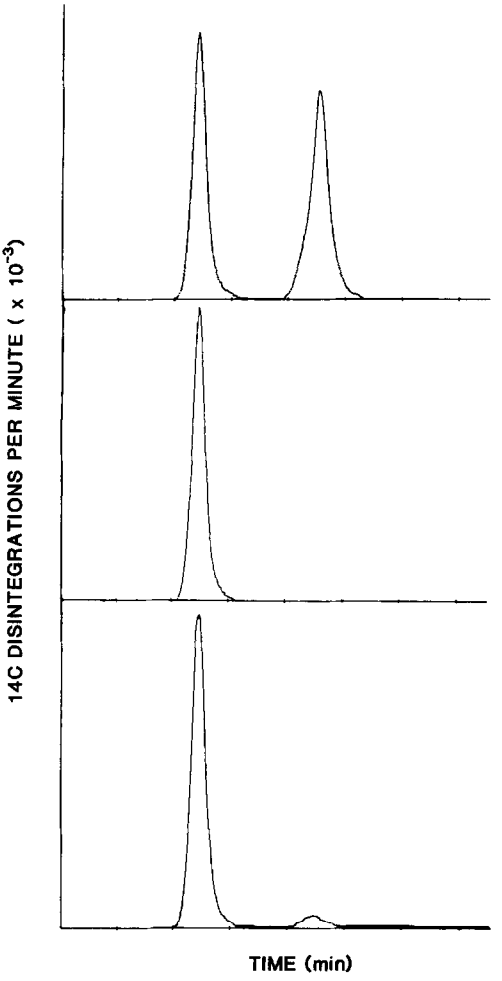

FIG. 3. HPLC chromatogram of a standard mixture of glyphosate and AMPA (top panel); kidney extracts 6 hr after administration of glyphosate (middle panel); and colon extracts of two animals 2 hr after the administration of glyphosate (bottom panel). The HPLC chromatograms of extracts from all other tissues containing greater than 1% of the administered dose at all other time points consisted of a single peak, as in the middle panel, with retention times similar to parent glyphosate (see Table 5). Extracts of the gastrointestinal contents from one animal 28 hr after the administration of glyphosate yielded a chromatogram similar to the profile in the bottom panel. In all cases extraction efficiency was greater than 91%.

TABLE 5

COMPARISON OF RETENTION TIMES BETWEEN TISSUE EXTRACTS AND GLYPHOSATE OR AMPA STANDARDS[a]

Sample	Retention time (min) Peak 1	Retention time (min) Peak 2	% Glyphosate	% AMPA	HPLC recovery[b]
2 hours[c]					
Standard[d]	9.0 ± 0.1	15.9 ± 0.2	—	—	—
Small intestine	8.8 ± <0.1	ND[e]	100.0 ± 0.0	ND	98.1 ± 0.5
Gut contents	8.8 ± <0.1	ND	100.0 ± 0.0	ND	100.5 ± 1.6
Standard	9.4 ± <0.1	16.5 ± <0.1	—	—	—
Colon[f]	9.2 ± 0.1	15.9 ± 0.2	94.1 ± 1.2	5.9 ± 1.2	98.6 ± 6.8
Standard	9.4 ± 0.1	18.6 ± <0.1	—	—	—
Carcass	9.5 ± 0.1	ND	99.0 ± 1.7	ND	98.2 ± 0.5
6.3 hours					
Standard	9.4 ± 0.3	15.9 ± 0.2	—	—	—
Small intestine	9.1 ± 0.1	ND	100.0 ± 0.0	ND	92.2 ± 2.1
Gut contents	9.5 ± 0.1	ND	100.0 ± 0.0	ND	110.9 ± 12.6
Colon	9.1 ± 0.1	ND	100.0 ± 0.0	ND	95.2 ± 4.6
Kidney	9.3 ± 0.1	ND	100.0 ± 0.0	ND	93.7 ± 2.3
Standard	9.8 ± 0.7	22.3 ± 2.8	—	—	—
Carcass	9.8 ± 0.3	ND	100.0 ± 0.0	ND	100.3 ± 2.6
28 hours					
Standard	8.2 ± 0.8	14.4 ± 1.7	—	—	—
Carcass	8.6 ± 0.4	ND	100.0 ± 0.0	ND	104.2 ± 3.1
Gut contents[g]	9.0 ± <0.1	15.8 ± 0.3	96.8 ± 2.1	3.2 ± 2.1	93.2 ± 0.8
96 hours					
Standard	8.3	15.1	—	—	—
Carcass	8.1 ± 0.4	ND	100.0 ± 0.0	ND	105.0 ± 6.5

[a] Mean ± SEM for three to four animals.
[b] Recovery determined by dpm recovered from column/dpm applied to column × 100.
[c] Hours after administration.
[d] Mixture of glyphosate and AMPA standards; peak 1 corresponds to glyphosate and peak 2 to AMPA.
[e] ND, not detectable.
[f] Two animals. The colon of one animal contained < 1% g the dose and was not extracted.
[g] Second peak present in one animal only, duplicate injections.

a single radioactive peak was spiked with radiolabeled glyphosate and the resulting HPLC profile indicated coelution of the radioactive material into one symmetrical peak. The extraction efficiency of tissues containing greater than 1% of the administered dose of radiolabeled glyphosate was always greater than 91%.

HPLC profiles of extracts of colons from animals administered glyphosate 2 hr previously and the gut contents of one animal 28 hr after the administration of glyphosate indicated the presence of a minor component (~6 and 3% of the tissue radioactivity, respectively) which seemed to coelute with AMPA (Table 5). Since this metabolite was present in very low levels (<0.4 ppm, <0.1% of the dose) and was of a transitory nature (present only at 2 hr after administration in the colon) further attempts of identification were not performed.

DISCUSSION

The purpose of this study was to evaluate the metabolite profile in tissues containing greater than 1% of an administered dose of radiolabeled glyphosate. Metabolite charac-

terization indicated that greater than 94% of the extractable body burden was parent glyphosate. The only metabolite observed (<0.04% administered dose) was detected in the colon 2 hr after administration and in the gut content of one animal 28 hr after administration. The production of this metabolite could have been the result of intestinal microbial action and was most likely AMPA which is known to be a microbial metabolic product of glyphosate (Rueppel *et al.*, 1977; Mueller *et al.*, 1981). It was less likely to be the synthetic impurity, MAMPA, since this was not detected in other samples. Absorbed material detected in the kidney was present as parent glyphosate. These results agree with that of other Monsanto studies where AMPA was found to be formed as the sole metabolite in very low levels (R. K. Howe, personal communication, 1990, Monsanto Agricultural Co.) and they indicate that glyphosate does not induce its own metabolism. In fact, at high levels glyphosate produced a moderate inhibition of microsomal monooxygenases (Hietanen *et al.*, 1983) and it had little effect on peroxisomal β oxidation and GSH activity (Vainio *et al.*, 1983).

Within 7 hr after oral administration, almost 40% of the absorbed material had been eliminated in the urine as parent material. The remaining 50% was still associated with the small intestinal tissue but after 7 days nearly all of the absorbed material had been eliminated from the body. These results suggest that both the urinary and fecal pathways are important clearance mechanisms for this chemical and that little metabolism occurs.

At 168 hr after administration less than 1.1% of the administered dose remained associated with the bone. The concentration had decreased to approximately 1 ppm. Previous studies indicate no evidence of adverse effects to either bone structure or function after prolonged exposures to glyphosate. Based upon the metabolite profile from other tissues it would be expected that this material is nonmetabolized parent glyphosate which has complexed with Ca^{2+} ions in the bone matrix.

Tissue to blood ratios significantly in excess of unity indicate tissue deposition. Of considerable interest was the increasing tissue to blood ratio observed in the bone with time. This increase suggests that elimination from the bone was slower than that from other tissues and paralleled what was observed in the carcass. The similarities between carcass levels and bone levels as well as the elimination patterns in the carcass and bone are indicative of the fact that the majority of radioactivity in the carcass was due to material retained by the bone.

In conclusion, total recovery for this study was 100 ± 10% for the animals studied, and the tissue retention and whole body retention times were relatively short. No toxic metabolites were produced from glyphosate. The samples were stable in a tissue matrix, tissue extraction efficiency was very good, and the vast majority of the body burden was unmetabolized parent glyphosate.

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