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

JAN 30 1989
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OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: PP#6F3380/6H5502. Glyphosate (Roundup®) in or on Soybeans. Amendment of 7/22/88 and Registration Standard Data Follow-up (Acc. #405320-1, -2, -3, -4 and #405413-1. -2. -3; DEB #4285 and #4286).

FROM: W. T. Chin, Ph. D., Chemist W. T. Chin
Tolerance Petition Section III
Dietary Exposure Branch
Health Effects Division (TS-769)

THRU: Philip V. Errico, Section Head
Tolerance Petition Section III
Dietary Exposure Branch
Health Effects Division (TS-769)

TO: Robert J. Taylor, PM #25
Registration Division (TS-767)

and

Toxicology Branch
Health Effects Division (TS-769)

BACKGROUND

Monsanto Agricultural Products Company has proposed to increase the tolerances established under 40 CFR 180.364 for the combined residues of the herbicide glyphosate, N-(phosphonomethyl)glycine, and its metabolite aminomethylphosphonic acid (AMPA) for soybeans to 20 ppm from 6 ppm and for soybeans straw to 200 ppm from 15 ppm; and under 40 CFR 561.253 for soybeans hulls to 100 ppm from 20 ppm based on preharvest applications. DEB has recommended against the proposed tolerances for the deficiencies specified in Conclusions 2a, 2b, 2c, 3b, 5, and 6a of W. T. Chin's 7/6/87 memo.

SUMMARY OF DEFICIENCIES REMAINING TO BE RESOLVED

All the deficiences identified in W. T. Chin's 7/6/87 memo in connection with this petition have been resolved.

The petitioner has also satisfied the data requirements identified under §171-4, §158.125 and §158.150 of the Guidance for the Registration of Pesticide Products Containing Glyphosate as the Active Ingredient (June,1986).

RECOMMENDATION

TOX considerations permitting, DEB recommend for the petitioner's request to increase the tolerances established under 40 CFR 180.364 for the combined residues of the herbicide glyphosate, and its metabolite AMPA for soybeans to 20 ppm from 6 ppm and for soybeans straw to 200 ppm from 15 ppm; and under 40 CFR 561.253 for soybeans hulls to 100 ppm from 20 ppm based on preharvest applications.

CURRENT CONSIDERATIONS

In response to the deficiencies identified above, Timothy J. Long of Monsanto Co. submitted an amendment which includes a cover letter dated 7/22/88 to Robert J. Taylor of EPA; revising Sections B and F and together with seven volumes of documents (Acc. #405320-1, -2, -3, -4 and #405413-1, -2, -3). The deficiencies specified above are restated below, followed by the petitioner's responses and DEB's comments/conclusions.

Deficiency "2a"

"The petitioner is requested to submit detailed information regarding specific rates and application instructions for the current petition. The complete Roundup® label should be provided."

The Petitioner's Response to Deficiency "2a"

The petitioner has submitted a label which was approved by the Agency on May 28, 1986. On pp. 94-95 of this label, detailed instructions and restrictions of preharvest applications of glyphosate on soybean crop are indicated.

DEB's Comment/Conclusion on the Petitioner's Response to Deficiency "2a"

DEB concludes that deficiency "2a" has been resolved.

Deficiency "2b"

"In the submitted "Directions for Use", the sentence "Do not feed or graze treated areas within 25 days after preharvest application" should be modified to read "Do not graze or harvest treated crop for livestock feed within 25 days of last preharvest application."

The Petitioner's Response to Deficiency "2b"

The statement "Do not graze or harvest treated crop for livestock feed within 25 days of last preharvest application" has been added to the label as requested. The alternate verbiage has been deleted.

DEB's Comment/Conclusion on the Petitioner's Response to Deficiency "2b"

DEB concludes that deficiency "2b" has been resolved.

Deficiency "2c"

"The so-called "soybean hay" which the petitioner is proposing a tolerance at 200 ppm is understood to be "soybean straw", the dried plant residue remaining on the ground after harvest of the soybeans. The petitioner is requested to clarify this point by submitting a revised Section F."

The Petitioner's Response to Deficiency "2c"

The petitioner submitted a revised Section F in which a tolerance has been requested for "Soybeans, straw" at 200 ppm. The established tolerances for soybean hay and forage at 15 ppm are still adequate because the proposed use requiring the higher tolerances on soybeans, soybean hulls and soybeans straw is a harvest aid to be applied after pods loose their green color.

RCB's Comment/Conclusion on the Petitioner's Response to Deficiency "2c"

DEB concludes that deficiency "2c" has been adequately resolved.

Deficiency "3b"

"As indicated in Glyphosate Registration Standard (7/15/85), metabolism studies using ruminants and poultry are required. Animals must be dosed for at least three days with ^{14}C-glyphosate at a concentration in the total diet which will result in sufficient residues in the tissues, milk, and eggs for characterization. Animals must be sacrificed within 24 hours of the final dose (milk and eggs must be collected twice daily). The distribution, characterization, and quantification of residues must be determined in eggs, milk, muscle, fat, kidney and liver."

The Petitioner's Response to Deficiency "3b"

The petitioner submitted the following two metabolism studies:

1. Acc. #405413-1: Metabolism Study of Synthetic $^{13}C/^{14}C$-Labeled Glyphosate and Aminomethylphosphonic Acid (AMPA) in Lactating Goats (Feb, 1988).

   Briefly: A mixture of $^{13}C/^{14}C$ (9:1)-labeled glyphosate and AMPA (272.2 mg of the disodium salt of glyphosate and 28.7 mg of the monosodium salt of AMPA) was orally administered to three lactating goats for 5 days at 120 ppm of the daily diet equivalent to 3X of the expected exposure levels based upon previous crop residue studies. A control goat received capsules containing sucrose. The two treated and the control animals were sacrificed 22 and 24 hours after the last dose and blood, muscle, kidneys, liver, fat and gastrointestinal tract contents were collected for radioanalysis. One treated animal was maintained in a depuration phase for 5 days after the last dose with continued collection of milk, urine and faces. The depurated animal was sacrificed on day-10 and blood, muscle, kidney, liver, fat and gastrointestinal tract contents were collected for radioanalysis by combustion and liquid scintillation counting.

   The total recovery of radioactivity in the samples collected ranged from 81.1% to 86.7% with 20.2% to 23.8% in urine and pan rinse, 60.2% to 66.5% in feces, rumen contents and intestinal contents combined. Less than 0.01% was found in milk. The primary route of elimination was urine and feces. The rate of elimination approached a constant rate at the end of the 5-day depuration period.

   Most of the ^{14}C residues in tissues were water extractable. These residues were determined by two HPLC methods and confirmed by GC/MS as being glyphosate and AMPA. There was no evidence of any further metabolism of glyphosate and AMPA in any of the tissues.

   Among the edible tissues for the nondepurated animals, the ^{14}C residues were determined as follows: kidney (3.49 - 10.5 ppm), liver (0.457 - 0.529 ppm), fat and muscle (0.009 - 0.011 ppm), milk (0.019 - 0.086 ppm).

2. Acc. #405413-2: Metabolism Study of Synthetic $^{13}C/^{14}C$-Labeled Glyphosate and Aminomethylphosphonic Acid (AMPA) in Laying Hens (Feb, 1988).

   Briefly: Twenty five 27-week old White Leghorn laying hens were dosed with a mixture of $^{13}C/^{14}C$ (9:1)-labeled glyphosate and AMPA orally for 7 days. Five groups of 5 animals each were dosed at 0 ppm (group 1), 120 ppm (=3X, groups 2, 3 and 5) and 400 ppm (group 4) levels. Excreta and eggs were collected daily. Hens were sacrificed 22 or 24 hours or 10 days after the last dose. Kidney, liver, thigh and breast muscle, ovaries, fat, gazzard, the remaining digestive tract with contents and whole blood were collected and pooled by treatment group for radioanalysis by combustion and liquid scintillation counting.

The total recovery of radioactivity in the samples collected ranged from 82.4% to 90.5% with 81.0% to 90.5% in excreta, Less than 0.01% to 2.11% in the gastrointestinal tract contents, less than 0.02% in the total egg production and less than 0.1% in other tissues combined. Approximately 66.6% to 78.4% of the daily dose was eliminated in excreta within the first 24 hours after dosing.

For the nondepurated groups dosed at 120 ppm, the ^{14}C residues were determined as follows: kidney (1.75 - 1.81 ppm), liver (0.511 - 0.560 ppm), fat and muscle (<0.026 ppm), gizzard (0.352 - 0.361 ppm). The 400 ppm group showed three to four times higher residue levels in all tissues and eggs than the 120 ppm groups. Most of the ^{14}C residue in eggs (less than or equal to 0.244 ppm) were in the yolk. The majority of the ^{14}C residues in eggs and tissues were water extractable. These residues were determined by two HPLC methods and the ^{14}C residues in the liver of Group 4 (400 ppm level) were confirmed by GC/MS as being glyphosate and AMPA which were rapidly eliminated during depuration.

DEB's Comment/Conclusion

Results of the above metabolism studies in lactating goats and laying hens are consistent with the metabolism studies in rats, rabbits and cows (see PP#3F2809 P. Perfetti's 4/1/83 memo). The majority residues are the parent compound and AMPA in eggs, milk and animal tissues. DEB concludes that the above metabolism studies are adequate and, therefore, deficiency "3b" is resolved.

Deficiency 5

"Since the field trials were conducted in 1979 and the dates of analysis of the samples are not given, RCB cannot determine the storage periods of the samples analyzed. Therefore, RCB is unable to determine the adequacy of the residue data submitted for the requested tolerance changes without the support of adequate storage stability data. The petitioner should submit information on the conditions and period of sample storage."

The Petitioner's Response to Deficiency 5

Monsanto has submitted storage stability data for soybean grain from 9 to 45 months and for soybean hay from 9 to 46 months under 0°F. Monsanto is in the process of conducting a crop storage stability study and a progress report was submitted on 1/29/88 (Acc. #405260-5). The results of this study to date suggest that glyphosate residues in crops are stable between 10 and 44 months. For soybean forage, it was shown that there was no decline in residue levels between 32 and 57 months of storage. Data of 71 and 83 months storage will be reported later.

In a report (Acc. #405320-4) entitled "Storage Stability Study of Glyphosate and AMPA in Swine Tissue, Dairy Cow Tissues and Milk, Laying Hen Tissues and Eggs" indicates that glyphosate and AMPA are stable at -20°C for 2 years.

DEB's Comment/Conclusion on the Petitioner's Response to Deficiency 5

DEB concludes that the information provided in the above storage stability studies is adequate to satisfy this petition. Therefore, deficiency 5 has been resolved.

Deficiency "6a"

"Previous feeding studies on cattle, poultry and swine using a 3:1 ratio of glyphosate and AMPA at dietary levels of 10, 30 and 100 ppm indicated that no detectable (<0.025 ppm) residues of glyphosate and AMPA were found in milk or eggs and none (<0.05 ppm) were found in muscle or fat of cattle, swine or poultry from the 100 ppm feeding level (PP#5F1536). However, if the metabolism studies requested in Conclusion "3b" above identify additional residues of toxicological concern, new feeding studies may be needed."

The Petitioner's Response to Deficiency "6a"

Monsanto indicates that based upon the results of the goat (Acc.#405413-1 and chicken (Acc. #405413-2) metabolism studies submitted, no additional feeding studies are needed.

DEB's Comment/Conclusion on the Petitioner's Response to Deficiency "6a"

In both of the goat and chicken metabolism studies, glyphosate and AMPA were found to be the major residues in milk, eggs and tissues and no further metabolism of these two compounds were observed. Therefore, DEB concludes that the petitioner's response is acceptable and deficiency "6a" is adequately resolved.

OTHER CONSIDERATIONS: REGISTRATION STANDARD DATA FOLLOW-UP

1. Data Requirement: §171-4, Residue Analytical Method

Validation of An Analytical Method for the Determination of Glyphosate Residues in Animal Tissues (Feb., 1988, Acc. #405413-4)

Briefly: Residues in animal samples are extracted with water followed by iron-loaded Chelex® 100 resin, and eluted with HCl through an anion exchange resin. The sample is then evaporated to dryness and reconstituted in distilled, deionized water. Separation and quantitation are achieved by cation exchange HPLC, post-column reaction with fluoraldehyde® and fluorescence detection. The limit of detection of this method is 0.010 ppm for glyphosate and 0.012 ppm for AMPA. Recoveries for glyphosate and AMPA averages 87.9% and 87.3%, respectively in all tissues. Examples of calculation and chromatograms are adequately submitted.

DEB's Comment/Conclusion On This Method

DEB concludes that this method has been adequately valided by the petitioner. DEB will submit this method for validation. Therefore, the data requirement identified on page 54 under §171-4 of Guidance for the Registration of Pesticide Products Containing Glyphosate as the Active Ingredient (June, 1986) has been satisfied.

2a. Data Requirement: §158.125, Residues in Meat, Milk, Poultry, Eggs

Residue Determination of Glyphosate and AMPA in Laying Hen Tissues and Eggs Following A 28-day Feeding Study (Nov., 1987, Acc. #405320-1).

Briefly: One hundred single-comb White Leghorn laying hens were divided into four groups (one control and 3 dosed) and fed chicken chow containing a mixture of glyphosate and AMPA (9:1) at 0, 40(1X), 120 and 400 ppm levels. Each of the groups was divided into five-bird subsets. One-half of the animals from each group were sacrificed after feeding for 28 days and samples of fat, muscle, liver and kidney were collected for analysis. The treated feed diets were then replaced with non-treated diet. One-quarter of the animals were sacrificed 7 days later and the remaining animals were sacrificed 28 days later and the same samples were collected for analysis. Eggs were collected for analysis.

Results of analysis in tissues and eggs are corrected for recovery and summarized in Tables 1 and 2, respectively below:

Table 1. Average Residues of Glyphosate and AMPA in Chicken Tissues

Tissue	Treatment level	Residues (ppm) Determined in Different Treatment Days					
		Day-28		Day-35		Day-56	
		Glyphosate	AMPA	Glyphosate	AMPA	Glyphosate	AMPA
Fat	1X	-	-	-	-	-	-
	3X	-	-	-	-	-	-
	10X	<0.06*	-	-	-	-	-
Muscle	1X	-	-	-	-	-	-
	3X	-	-	-	-	-	-
	10X	-	-	-	-	-	-
Liver	1X	0.07	-	-	-	-	-
	3X	0.20	0.10	-	-	-	-
	10X	0.78	0.38	0.15	0.15	-	-
Kidney	1X	0.38	-	0.06	-	-	-
	3X	1.17	0.06	0.23	-	0.17	-
	10X	4.54	0.33	0.35	-	0.08	-

- denotes <0.05 ppm.
* represents averages of measurable and <0.05 ppm residues in tissues from the multiple animals in the study.

Table 2. Average Residues of Glyphosate and AMPA in Chicken Eggs

Treatment level	Day	Residues Determined (ppm) Day 1 to 28 Glyphosate	Day 1 to 28 AMPA	Day 29 to 56 Glyphosate	Day 29 to 56 AMPA
1X	1	-	-	-	-
	2	-	-	-	-
	4	-	-	-	-
	7	-	-	-	-
	14	-	-	-	-
	21	-	-	-	-
	28	-	-	-	-
3X	1	-	-	-	-
	2	-	-	-	-
	4	-	-	-	-
	7	-	-	-	-
	14	-	-	-	-
	21	-	-	-	-
	28	-	-	-	-
10X	1	-	-	0.09	-
	2	-	-	0.09	-
	4	-	-	0.07	-
	7	0.08	-	-	-
	14	0.09	-	-	-
	21	0.10	-	-	-
	28	0.09	-	-	-

- denotes <0.05 ppm.

DEB's Comments/Conclusions

The residue data shown in Tables 1 and 2 indicate that at 40 ppm (1X) feeding level, the glyphosate and AMPA residues in chicken tissues and eggs are less than 0.50 ppm. These data adequately support the 0.5 ppm tolerance established under 40 CFR 180.364(b) for glyphosate in poultry kidney and liver. DEB concludes that the data requirement identified by footnote 72 on page 65 of Guidance for the Registration of Pesticide Products Containing Glyphosate as the Active Ingredient (June, 1986) has been satisfied.

2b. Data Requirement: §158.125, Residues in Meat, Milk, Poultry, Eggs

Residue Determination of Glyphosate and AMPA in Swine Tissues Following A 28-day Feeding Study (Sept. 1987; Acc. #405320-2).

Beiefly: Sixteen cross-bred swine were divided into four groups (one control and 3 dosed) and fed a diet containing a mixture of glyphosate and AMPA (9:1) at 0, 40(1X), 120 and 400 ppm levels. One male and one female animal from each group were sacrificed after feeding for 28 days and samples of fat, liver, muscle and kidney were collected for analysis. The treated feed diets were then replaced with non-treated diet and the remaining animals were sacrificed 28 days later and the same samples were collected for analysis. Results of analysis in tissues are summarized in Table 3 below:

Table 3. Average Residues of Glyphosate and AMPA in Swine Tissues

Tissue	Treatment level	Residues Determined (ppm) Day-28 Glyphosate	Day-28 AMPA	Day-56 Glyphosate	Day-56 AMPA
Fat	1X	-	-	-	-
	3X	-	-	-	-
	10X	-	-	-	-
Muscle	1X	-	-	-	-
	3X	-	-	-	-
	10X	0.06	-	-	-
Liver	1X	-	-	-	-
	3X	0.21	0.12	-	-
	10X	0.75	0.39	-	-
Kidney	1X	0.37	0.07	-	-
	3X	2.59	0.29	0.08	-
	10X	7.81	0.96	0.18	-

- denotes <0.05 ppm

DEB's Comments/Conclusions

The residue data shown in Table 3 indicate that at 40 ppm (1X) feeding level, the glyphosate and AMPA residues in swine tissues are less than 0.50 ppm. These data adequately support the 0.5 ppm tolerance established under 40 CFR 180.364(b) for glyphosate in hogs kidney and liver. DEB concludes that the data requirement identified by footnote 72 on page 65 of Guidance for the Registration of Pesticide Products Containing Glyphosate as the Active Ingredient (June, 1986) has been satisfied.

2c. Data Requirement: §158.125, Residues in Meat, Milk, Poultry, Eggs

Acc. #405320-3: Residue Determination of Glyphosate and AMPA in Diary Cow Tissues Following A 28-day Feeding Study (Sept. 1987)

Briefly: Nineteen lactating cattle were divided into four groups (one control of four and 3 dosed of five) and fed with a diet containing a mixture of glyphosate and AMPA (9:1) at 0, 40(1X), 120 and 400 ppm levels. Three animals from each group were sacrificed after feeding for 28 days and samples of fat, liver, muscle and kidney were collected for analysis. The treated feed diets were then replaced with non-treated diet. One animal of each group were sacrificed 7 days and one from each group were sacrificed 28 days later and the same samples were collected for analysis. Results of analysis in tissues are summarized in Table 4. Milk samples were also collected at day 1, 2, 4, 7, 14, 21 and 28. However, no residues were ever detected in milk samples.

Table 4. Average Residues of Glyphosate and AMPA in Cow Tissues

Tissue	Treatment level	Residues (ppm) Determined in Different Treatment Days					
		Day-28		Day-35		Day-56	
		Glyphosate	AMPA	Glyphosate	AMPA	Glyphosate	AMPA
Fat	1X	-	-	-	-	-	-
	3X	-	-	-	-	-	-
	10X	-	-	-	-	-	-
Muscle	1X	-	-	-	-	-	-
	3X	-	-	-	-	-	-
	10X	-	-	-	-	-	-
Liver	1X	0.06	-	-	-	-	-
	3X	0.07	-	-	-	-	-
	10X	0.24	0.17	0.13	0.08	-	-
Kidney	1X	0.26	0.07	-	-	-	-
	3X	0.79	0.21	-	-	-	-
	10X	3.16	0.94	0.06	0.08	-	-

- denotes <0.05 ppm.

DEB's Comments/Conclusions

The residue data shown in Table 4 indicate that at 40 ppm (1X) feeding level, the glyphosate and AMPA residues in cattle tissues are less than 0.50 ppm. These data adequately support the established 0.5 ppm tolerance established under 40 CFR 180.364(b) for glyphosate in cattle kidney and liver. DEB concludes that the data requirement identified by footnote 72 on page 65 of Guidance for the Registration of Pesticide Products Containing Glyphosate as the Active Ingredient (June, 1986) has been satisfied.

cc: R.F., Circu., PP#6F3380/6H5502, Glyphosate Reg. Std., W.T.Chin, TAS, and PMSD-ISB

RDI: P.V.Errico(1/24/89), R.D.Schmitt(1/24/89)
TS-769: RCB: CM#2, RM812,557-4352, W.T.Chin,wc(1/24/89)