UNITED STATES ENVIRONMENTAL PROTECTION AGENCY

WASHINGTON, D.C. 20460

OFFICE OF
PREVENTION, PESTICIDES
AND TOXIC SUBSTANCES

Date: 15-AUG-2006

MEMORANDUM

SUBJECT: Flufenoxuron: Human-Health Risk Assessment for Proposed Tolerances on Apples, Pears, Grapes, Oranges and Livestock Commodities Imported into United States (U.S.). PC Code: 108203. PP#: 8E4943. DP#: D313998. Decision#: 342691.

FROM: Jennifer R. Tyler, Chemist
Guruva B. Reddy, D.V.M., Ph.D., Toxicologist
Registration Action Branch (RAB1)
Health Effects Division (HED; 7509P)

THROUGH: P.V. Shah, Ph.D., Senior Toxicologist
Felecia Fort, Acting Branch Chief
RAB1/HED (7509P)

Christina Swartz, Chemist
John Liccione, Toxicologist
Risk Assessment Review Committee (RARC; 7509P)

TO: Mark Suarez/Richard Gebken, PM Team 03
Insecticide Branch
Registration Division (RD; 7505P)

The HED of the Office of Pesticide Programs (OPP) is charged with estimating the risk to human health from exposure to pesticides. The RD of OPP has requested that HED evaluate hazard and exposure data and conduct dietary, occupational, residential and aggregate exposure assessments, as needed, to estimate the risk to human health that will result from proposed tolerances for the new active ingredient flufenoxuron on imported apples, grapes, pears, oranges and livestock commodities. A summary of the findings and an assessment of human risk resulting from the proposed tolerances for flufenoxuron is provided in this document. The risk assessment, the residue chemistry data review, and the dietary risk assessment were provided by Jennifer Tyler (RAB1), and the hazard characterization and endpoint selection by Guruva Reddy (RAB1).

Table of Contents

1.0 Executive Summary . . . . . Page 4 of 64

2.0 Ingredient Profile . . . . . Page 6 of 64
- 2.1 Summary of Registered/Proposed Uses . . . . . Page 6 of 64
- 2.2 Structure and Nomenclature . . . . . Page 8 of 64
- 2.3 Physical and Chemical Properties . . . . . Page 9 of 64

3.0 Metabolism Assessment . . . . . Page 9 of 64
- 3.1 Comparative Metabolic Profile . . . . . Page 9 of 64
- 3.2 Nature of the Residue in Foods . . . . . Page 10 of 64
- 3.3 Rat Metabolism . . . . . Page 10 of 64
- 3.4 Tabular Summary of Metabolites and Degradates . . . . . Page 12 of 64
- 3.5 Toxicity Profile of Major Metabolites and Degradates . . . . . Page 14 of 64
- 3.6 Summary of Residues for Tolerance Expression and Risk Assessment . . . . . Page 14 of 64

4.0 Hazard Characterization/Assessment . . . . . Page 14 of 64
- 4.1 Hazard Characterization . . . . . Page 14 of 64
- 4.2 FQPA Hazard Considerations . . . . . Page 20 of 64
- 4.3 Recommendation for a Developmental Neurotoxicity Study . . . . . Page 24 of 64
- 4.4 Hazard Identification and Toxicity Endpoint Selection . . . . . Page 24 of 64
- 4.5 FQPA Safety Factor . . . . . Page 26 of 64
- 4.6 Endocrine Disruption . . . . . Page 27 of 64

5.0 Exposure Characterization/Assessment . . . . . Page 27 of 64
- 5.1 Dietary Exposure/Risk Pathway . . . . . Page 27 of 64
- 5.2 Water Exposure/Risk Pathway . . . . . Page 32 of 64
- 5.3 Residential (Non-Occupational) Exposure/Risk Pathway . . . . . Page 32 of 64

6.0 Aggregate Risk Assessments and Risk Characterization . . . . . Page 32 of 64

7.0 Cumulative Risk Characterization/Assessment . . . . . Page 33 of 64

8.0 Occupational Exposure/Risk Pathway . . . . . Page 33 of 64

9.0 Data Needs and Label Requirements . . . . . Page 33 of 64

10.0 References . . . . . Page 35 of 64

Appendices . . . . . Page 36 of 64

1.0 Executive Summary

Background: Flufenoxuron is a new benzoylurea type acaricide/insecticide which inhibits chitin biosynthesis (MOA Group 15) in nymphal mites and caterpillars. There are currently no food/feed uses or tolerances for flufenoxuron in the U.S. There are also no registered or proposed residential uses of flufenoxuron in the U.S. BASF Corporation is supporting the use of flufenoxuron on apples, grapes and oranges grown for export to the U.S. in selected European and South American countries. Flufenoxuron is formulated as 50 and 100 g/L emulsifiable concentrates (EC), dispersible concentrate (DC) or flowable concentrate (FlC) under the trade name Cascade®. Depending on the crop and the country of use, these formulations are proposed for one to four foliar applications per season to the above crops at rates equivalent to 0.036-0.22 lb ai/A/application.

Hazard Characterization: The database is adequate for establishment of tolerances on the requested commodities, except for a developmental toxicity study in rat which was classified as unacceptable/guideline, however, this study provides adequate information to conclude that flufenoxuron was not fetotoxic at doses up to the limit dose. As acute toxicity studies are not required for tolerances on imported commodities, only an acute oral toxicity study in rat was submitted. Based on this study, flufenoxuron was placed in Toxicity Category IV.

The critical toxicological effects of flufenoxuron in mammals are on the hemopoeitic system. These effects include shifts in red blood cell parameters (decreased in hemoglobin, hematocrit, red blood cells and erythroid: myloid ratio) resulting in mild anemia and enhanced levels of methemoglobin and sulfhemoglobin in dogs and rats. It is likely that a rat metabolite (aniline metabolite (4-[2-chloro, α, α, α-trifluoro-p-tolyoxy]-2-fluoroaniline) is responsible for the formation of methemoglobin and sulfhemoglobin. Two oncogenicity studies in mice and one in rat were conducted at doses exceeding 4-7.5x limit dose. One of the mouse studies conducted at 7.5x limit dose had an increased incidence of vascular tumors in males and females. The mouse oncogenicity study was repeated with the high dose exceeding 1.5x limit dose; and there was no increased incidence of any tumors. Therefore, the increased incidences of vascular tumors are considered high dose effects only. Flufenoxuron exhibited a negative response in various genotoxicity screening assays. Based on the lack of evidence of carcinogenicity in both rats and mice carcinogenicity studies and negative response for genotoxicity in various assays, flufenoxuron is considered "not likely to be carcinogenic to humans."

Flufenoxuron was not teratogenic in rats and rabbits at doses up to the limit dose (1000 mg/kg/day). It did not affect reproductive parameters in a 2-generation reproduction study at doses up to 10, 000 ppm (771.6 mg/kg/day). There was no evidence of increased susceptibility for flufenoxuron in the developmental toxicity study in rats. No adverse effects were observed in either dams or offspring at the limit dose. There was slight evidence of increased susceptibility observed in the developmental toxicity study in rabbits in which decreased fetal weight was observed in the absence of maternal toxicity at the limit dose. There is a well-characterized NOAEL protecting fetuses from the high dose toxicity. Decreased in pup body weight was observed in the absence of maternal toxicity in a 2-generation reproduction study in rats indicating increased susceptibility. However, there is no concern for this increased susceptibility since there is a well characterized NOAEL established in the study, and the endpoints were used

for risk assessment. Based on this discussion, there is no concern for increased susceptibility of offspring following exposure to flufenoxuron.

Dose-Response Assessment and Food Quality Protection Act (FQPA) Decision: The risk assessment team selected endpoints for risk assessment and evaluated the potential for increased susceptibility of infants and children from exposure to flufenoxuron according to the February 2002 OPP 10X guidance document. The FQPA safety factor (SF) was reduced to 1x based on toxicological considerations by the team, the conservative residue assumptions used in the dietary and exposure risk assessment, and the completeness of the residue chemistry database.

Risk assessments were conducted for the chronic dietary exposure scenario only. The chronic reference dose (cRfD) was calculated by dividing the no-observed-adverse-effect-level (NOAEL) by 100 (10X for interspecies extrapolation, 10X for intraspecies variation). Since the FQPA SF has been reduced to 1X, the chronic population adjusted dose (cPAD) is equal to the cRfD.

Exposure Scenario	Dose	Endpoint	Study/Effect
Chronic dietary	NOAEL = 3.75 mg/kg/day	cRfD and cPAD = 0.0375 mg/kg/day	2-generation reproduction study in rats. The lowest-observed-adverse-effect-level (LOAEL) of 14.33/16.0 (M/F) mg/kg/day was based on decreased body weights during lactation during days 4-21.

Dietary-Exposure Assessment: An unrefined, chronic dietary exposure assessment was performed for the general U.S. population and various population subgroups assuming tolerance-level residues and 100% crop treated (%CT) for all commodities. As there are no proposed domestic uses of flufenoxuron, drinking water was not incorporated into the dietary assessment. The chronic dietary exposure estimates are below HED's level of concern (<100% cPAD) for the general U.S. population (14% cPAD) and all population subgroups. The most highly-exposed population subgroup is children 1-2 years old at 63% cPAD.

Aggregate Risk Assessment: The current action is for tolerances on commodities imported in the U.S. only; therefore, an aggregate risk assessment is not warranted.

***HED Recommendations*:** Pending submission of the revised Sections B and F as specified in Section 9.0 of this risk assessment, the available toxicology and residue chemistry databases support the establishment of the following time-limited tolerances for residues of flufenoxuron *per se*:

apple . . . . . 0.50 ppm
pear . . . . . 0.50 ppm
grape . . . . . 0.70 ppm
grape, raisin . . . . . 2.0 ppm
orange . . . . . 0.30 ppm
orange, oil . . . . . 60 ppm
milk . . . . . 0.20 ppm
milk, fat . . . . . 4.0 ppm
meat * . . . . . 0.10 ppm
meat byproducts * . . . . . 0.50 ppm
fat * . . . . . 4.5 ppm

* of cattle, goats, horses, and sheep.

Conversion from time-limited to permanent tolerances is contingent upon satisfaction of the data needs requirements as listed in Section 9.0 of this risk assessment.

2.0 Ingredient Profile

2.1 Summary of Registered/Proposed Uses

Flufenoxuron is a benzoylurea type acaricide/insecticide which inhibits chitin biosynthesis (MOA Group 15) in nymphal mites and caterpillars. There are currently no food/feed uses or tolerances for flufenoxuron in the U.S. BASF Corporation is supporting the use of flufenoxuron on apples, grapes and oranges grown for export to the U.S. in selected European and South American countries. Flufenoxuron is formulated as 50 and 100 g/L EC, DC or FlC under the trade name Cascade®. Depending on the crop and the country of use, these formulations are proposed for one to four foliar applications per season to the above crops at rates equivalent to 40-250 g ai/ha/application (0.036-0.22 lb ai/A/application).

Table 2.1.1. Summary of Directions for Use of Flufenoxuron.

Country	Application Type, and Equipment	Form. [1]	Applic. Rate (g ai/ha) *[lb ai/A]*	Max. No. Applic. per Season	Max. Seasonal Applic. Rate (g ai/ha) *[lb ai/A]*	PHI[3] (days)	Use Directions and Limitations
Apple and Pear							
Argentina	Broadcast foliar; Ground Equipment	100 g/L EC	240 *[0.21]*	1	240 [0.21]	90 (80) [2]	Maximum rate based on maximum application volume of 2000 L/ha.
Brazil	Broadcast foliar; Ground or aerial equipment	100 g/L EC	200 *[0.18]*	NS (3)	600 [0.54]	35	Only for use on apple. Apply aerial applications in 20-40 L/ha and low volume ground applications in 5-50 L/ha
Chile	Broadcast foliar; Ground Equipment	100 g/L DC	113 (150) *[0.10]*	1	113 (150) *[0.10]*	60	Maximum rate based on maximum application volume of 1500 L/ha.

Table 2.1.1. Summary of Directions for Use of Flufenoxuron.

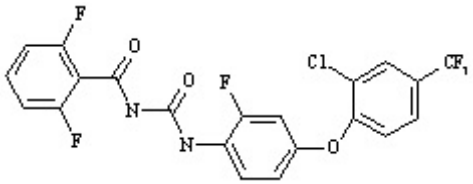
Country	Application Type, and Equipment	Form. [1]	Applic. Rate (g ai/ha) *[lb ai/A]*	Max. No. Applic. per Season	Max. Seasonal Applic. Rate (g ai/ha) *[lb ai/A]*	PHI[3] (days)	Use Directions and Limitations
France	Broadcast foliar; Ground equipment	100 g/L DC	100 (75) *[0.89]*	NS (1)	100 (75) *[0.89]*	119	Maximum rate based on maximum application volume of 1000 L/ha.
Greece	Broadcast foliar; Ground equipment.	100 g/L DC	200 *[0.18]*	3	600 *[0.54]*	28	Maximum rate based on maximum application volume of 2000 L/ha.
Italy	Broadcast foliar; Ground equipment	50 g/L DC	150 *[0.13]*	2	300 *[0.26]*	90	Do not apply by air
Spain	Broadcast foliar; application equipment not specified	100 g/L DC	150 *[0.13]*	NS (2)	300 *[0.26]*	28	Maximum rate based on maximum application volume of 1500 L/ha.
Grape							
France	Broadcast foliar; Ground equipment	100 g/L DC	40 *[0.036]*	NS (3)	120 *[0.11]*	28	Maximum rate based on maximum application volume of 1000 L/ha.
Greece	Broadcast foliar; Ground equipment	100 g/L DC	75 [0.067]	2	150 *[0.134]*	28	Maximum rate based on maximum application volume of 1500 L/ha.
Italy	Broadcast foliar; Ground equipment	50 g/L DC	100 *[0.089]*	2	200 *[0.18]*	30	Do not apply by air.
Spain	Broadcast foliar; application equipment not specified	100 g/L DC	75 [0.067]	NS (2)	150 *[0.134]*	28	Maximum rate based on maximum application volume of 1000 L/ha.
Orange							
Brazil	Broadcast foliar; Ground or aerial equipment	100 g/L EC	90 *[0.080]*	NS (2)	180 *[0.16]*	28 (15)	Apply aerial applications in 20-40 L/ha and low volume ground applications in 5-50 L/ha
Chile	Broadcast foliar; Ground equipment	100 g/L DC	113 (150) *[0.089]*	1	113 (150) *[0.089]*	60	Label allows application to "citrus". Maximum rate based on maximum application volume of 1500 L/ha.
Greece	Broadcast foliar; Ground equipment	100 g/L DC	200 *[0.18]*	4	800 *[0.72]*	28	Maximum rate based on maximum application volume of 4000 L/ha.
Italy	Broadcast foliar; Ground equipment	50 g/L DC	150 *[0.13]*	2	300 *[0.26]*	75	Includes use on Tangerines and Clementines. Do not apply by air.
Spain	Broadcast foliar; application equipment not specified	100 g/L DC	250 *[0.22]*	NS (3)	750 *[0.66]*	28	Maximum rate based on maximum application volume of 5000 L/ha.

[1] No formulations are currently being proposed for use in the U.S. The DC formulation appears to be equivalent to an FlC type formulation.
[2] When there is a discrepancy between the information on the label and in the petitioner's summary table, the values from the summary tables are listed in parentheses.
[3] PHI = preharvest interval.
NS = Not specified on translated label.

HED Conclusions. The use directions provided by the petitioner are adequate to allow evaluation of the residue data relative to the proposed uses, although discrepancies were noted between application rates on the translated labels and the summary table provided by the petitioner. The petitioner has stated that the application rates provided in the summary table are the actual proposed rates (via e-mail message from M. Suarez to J. Tyler; 1/11/06). Therefore, the rates provided in the summary table provided by the petitioner were assumed. The most recent versions of BASF flufenoxuron labels should be submitted along with translations.

2.2 Structure and Nomenclature

Table 2.2.1. Test Compound Nomenclature

Chemical Structure	
Empirical Formula	$C_{21}H_{11}ClF_6N_2O_3$
Common name	Flufenoxuron
Company experimental names	CL 811,678 WL 115110
IUPAC name	1-[4-(2-chloro-α,α,α-trifluoro-*p*-tolyloxy)-2-fluorophenyl]-3-(2,6-difluorobenzoyl)urea
CAS name	N-[[[4-[2-clair-4-(trifluoromethyl)phenoxy]-2-fluorophenyl]amino]carbonyl]-2,6-difluorobenzamide
CAS Registry Number	101463-69-8
End-use product/EP	50 or 100 g/L FlC and EC
Chemical Class	benzoylurea
Known Impurities of Concern	none

2.3 Physical and Chemical Properties

Table 2.3.1. Physicochemical Properties of Flufenoxuron

Parameter	Value		Reference
Melting point	169-172°C		MRID 44448404
pH	6.1		
Density	1.57 g/mL		
Water solubility (25°C)	1.36 x 10^{-6} g/L		
Solvent solubility (g/L at 25°C)	hexane: 0.012 octan-1-ol: 1.12 methanol: 3.55 dichloromethane: 18.7	cyclohexane: 96.4 dimethyl sulfoxide: 287 dimethylformamide: 287 acetone: 73.6	
Vapor pressure at 20°C	6.52 x 10^{-12} Pa		
Dissociation constant (pK_a)	10.1		
Octanol/water partition coefficient Log(K_{OW}) at 25°C	4.01		

3.0 Metabolism Assessment

3.1 Comparative Metabolic Profile

The nature of the flufenoxuron residues in fruit, ruminants and rats is understood based on the acceptable tomato, apple, grape, lactating goat and rat metabolism studies.

In rats, flufenoxuron (when labeled in both the aniline ring and the 2,6-difluorobenzene ring labels) is extensively metabolized primarily via hydrolysis. The metabolism of flufenoxuron proceeds via hydrolysis to a benzoic acid metabolite, a phenyl urea metabolite (4-[2-chloro, α, α, α-trifluoro-p-tolyoxy]-2-fluorophenyl urea), an aniline metabolite (4-[2-chloro, α, α, α-trifluoro-p-tolyoxy]-2-fluoroaniline), and subsequently several minor components. Benzoic acid was the major urinary metabolite following administration of the benzoyl ring labeled test substance, accounting for 10-12% of the administered dose. Both [4-(2-chloro, α, α, α-trifluoro-p-tolyoxy)-2-fluorophenyl urea] and [4-(2-chloro, α, α, α-trifluoro-p-tolyoxy)-2-fluoroaniline] were detected in the feces and urine following administration of the aniline ring labeled test article, but together at levels of <5% of applied dose. Tissue burdens generally represented less than 2% of the administered dose, 80% of which was identified as the parent.

The results of the available plant and ruminant metabolism data indicate that flufenoxuron is not metabolized in tomatoes, apples, grapes, and lactating goats. In all studies, parent was the only compound detected, accounting for 50-91% of total radioactive residues (TRR) in plants, and 76-115% in milk and ruminant tissues.

3.2 Nature of the Residue in Foods

3.2.1. Plant Metabolism

The results of acceptable tomato, apple, and grape metabolism studies indicate that the metabolism of flufenoxuron in/on fruits and fruiting vegetables is minimal and that translocation of flufenoxuron from leaves to fruits is limited. Residues are largely confined to the fruit surface and are composed almost entirely of parent, even at longer post-treatment intervals. Flufenoxuron accounted for 91% of the TRR in tomatoes and apples at 28 and 99 days after treatment (DAT), respectively, and 50-55% of the TRR in grapes by 28 DAT.

3.2.2 Livestock Metabolism

Although numerous deficiencies were noted, the available goat metabolism study is adequate for delineating the nature of the residues in ruminants for purposes of this petition, and a poultry metabolism study is not required as there are no poultry feed items associated with the proposed crop uses. The goat study indicates that there is the potential for bioaccumulation of flufenoxuron in ruminant tissues and that the metabolism of flufenoxuron in ruminants appears to be minimal with parent being excreted primarily in the feces and to a lesser extent in the milk. Following 4 days of dosing with [^{14}C]flufenoxuron at levels equivalent to ~2.8 ppm in the diet, TRR levels were highest in fat (1.59 ppm) and were considerably lower in liver (0.373 ppm), kidney (0.130 ppm), and muscle (0.076-0.103 ppm). Residues in milk increased over time to 0.372 ppm by Day 4 (p.m.) and were primarily associated with the cream fraction (82-93% TRR). High-performance liquid chromatography (HPLC) analyses of purified acetonitrile (ACN) fractions from milk and tissues (76-115% TRR) detected only parent compound.

3.3 Rat Metabolism

The metabolic fate of flufenoxuron was determined using two radiolabeled positions (aniline and difluorobenzene ring). Flufenoxuron exhibited a dose-dependant absorption following a single low dose (3.5 mg/kg) or single high dose (350 mg/kg). At the high dose, saturated absorption was observed. Approximately 86% of the low dose and 1% of the high dose was absorbed in 168 hours, the majority of which occurred within 48 hours. For the difluorobenzene ring test article, urine was a major route of excretion in the low-dose group, but not in the high-dose group (<1%). Conversely, 93-102% of the high dose and 4-19% of the low dose was eliminated in feces. Elimination via expired air was insignificant. Biliary excretion using the aniline ring label showed that all the radioactivity in the feces of females and 40% of that in males are biliary excretion products. Although, the majority of both urinary and fecal excretion occurred within 48 hours, excretion by both routes was biphasic, with slower phase occurring throughout the post-exposure, resulting in accumulation in adipose tissue. This phenomenon was probably due to entero-hepatic circulation. Accumulation of radioactivity in muscle and adipose tissue 4 hours post dosing with 3.5 mg/kg benzyl label was 30% and 42%, respectively. At 168 hours post dose, these values were 6% and 19%, respectively, suggesting an accumulation in the adipose tissue. High doses of both labels resulted in negligible tissue burden (<0.3%) indicating saturation absorption.

For the flufenoxuron aniline ring test article, the parent and a total of 10 urinary metabolites accounted for approximately 5% of the administered dose, and were considered non-significant. Fecal excretion of metabolites was quantitatively greater with parent compound accounting for the greatest portion of radioactivity. However, most fecal metabolites represented < 1% of the administered dose. Both [4-(2-chloro, α, α, α-trifluoro-p-tolyoxy)-2-fluorophenyl urea] and [4-(2-chloro, α, α, α-trifluoro-p-tolyoxy)-2-fluoroaniline] were detected in the feces and urine following administration of the aniline ring labeled test article. Unextractable residues accounted for 7-8% of the dose. The major urinary metabolite of [^{14}C- 2,6-difluorobenzene]flufenoxuron was the corresponding benzoic acid which, over 48 hours, accounted for 10-12% of the administered dose. Difluorobenzamide (<1%) was also detected in the urine along with unknown components all of which individually represented <1% of the dose. The only component detected in the feces of rats given the 2,6-difluorobenzene label was the parent compound. The results of the metabolism characterization studies with both label positions suggest that metabolism of flufenoxuron proceeds via hydrolysis to a benzoic acid metabolite, a phenyl urea metabolite (4-[2-chloro, α, α, α-trifluoro-p-tolyoxy]-2-fluorophenyl urea), an aniline metabolite (4-[2-chloro, α, α, α-trifluoro-p-tolyoxy]-2-fluoroaniline), and subsequently several minor components.

3.4 Tabular Summary of Metabolites and Degradates

Table 3.4.1. Tabular Summary of Metabolites and Degradates.

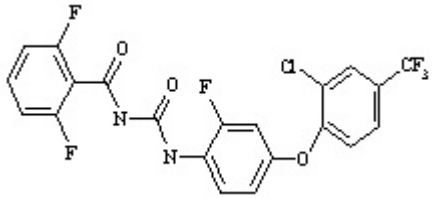
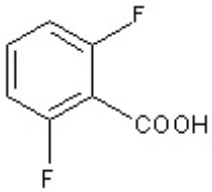
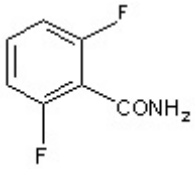
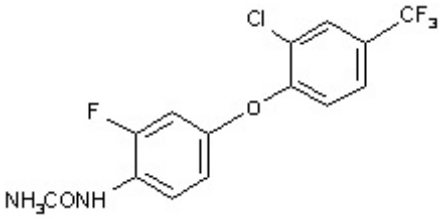
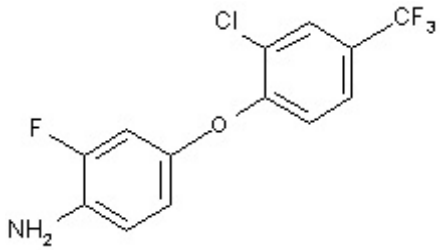
Chemical Name	Commodity	Percent TRR (PPM)[1]		Structure
		Matrices - Major Residue (>10%TRR)	Matrices - Minor Residue (<10%TRR)	
Flufenoxuron	Tomato	91.0		
	Apples	96.5		
	Grapes	94.2		
	Poultry	Liver 82.7 Kidney 82.1 Muscle 75.8 Fat 114.8 Milk, Day 1 81.1		
	Rat	Benzoyl label Feces (low dose) 6-12% (0-24 hours) Feces (high dose) 68-78 at 0-24 hours 22-13 (24-48 hours)	Benzoyl label Feces (low dose) 2-3 (24-48 hours) Aniline label Urine <1	
2,6,-difluorobenzoic acid	Rat		Benzoyl label Urine 8.0-8.6 (0-24 hours) 2.1-3.5 (24-48 hours)	
2,6,-difluorobenzamide	Rat		Benzoyl label Urine 0.2 (0-24 hours) <0.01-0.01 (24-48 hours)	

Table 3.4.1. Tabular Summary of Metabolites and Degradates.

Chemical Name	Commodity	Percent TRR (PPM)[1]		Structure
		Matrices - Major Residue (>10%TRR)	Matrices - Minor Residue (<10%TRR)	
4-(2-chloro,α, α, α-trifluoro-p-tolyoxy)-2-fluorophenyl urea (WL129183)	Rat		Aniline label Urine <1 Feces <1	
4-(2-chloro,α, α, α-trifluoro-p-tolyoxy)-2-fluoroaniline (WL15096)	Rat		Aniline label Urine <1 Feces <1	

Tomato: MRID No. 44448423; 0.125 kg ai/ha (0.11 lb ai/A); 0.21x the maximum proposed seasonal rate for any crop; fruit development; 0- and 28-day PHI.
Apple: MRID No. 44448424; 9.266 mg ai/tree; fruit development; 0-, 46-, and 99-day PHI.
Grape: MRID No. 46482204; 0.8 kg ai/ha; 1.3x; fruit development; 15-, 28-day PHI.
Lactating Goat: MRID No. 44448426; 2.8 ppm; 3.5x and 7x the MTDB for beef and dairy cattle, respectively; 4 days of dosing; 24 hour PSIl.
Rat Metabolism: MRID No. 44448422; 3.5 mg/kg or 350 mg/kg; Fischer-344 rats

3.5 Toxicity Profile of Major Metabolites and Degradates

Although no toxicity data on the rat metabolites are available, HED postulates that the aniline metabolite [4-(2-chloro,α, α, α-trifluoro-p-tolyoxy)-2-fluoroaniline] might be of concern because of methemo- and sulfhemoglobin formation. However, this metabolite was seen at low levels in the urine and feces (<1%). In addition, as this metabolite was not seen in the available plant or livestock studies, it is not of concern for tolerance or risk assessment purposes for this petition only.

3.6 Summary of Residues for Tolerance Expression and Risk Assessment

For this petition only, the residue of concern for tolerance and risk assessment purposes in plant and ruminant commodities is flufenoxuron *per se*. This conclusion applies only to the use of flufenoxuron on fruits and fruiting vegetables. Any future uses on other crops, such as leafy vegetables, legumes, or cereal grains, may require the submission of additional metabolism data, and any new uses having a substantial impact on livestock diets should include a new ruminant metabolism study, and potentially, a poultry metabolism study.

3.6.1 Tabular Summary

Table 3.6.1.1. Summary of Metabolites and Degradates to be included in the Risk Assessment and Tolerance Expression

Matrix		Residues included in Risk Assessment	Residues included in Tolerance Expression
Plants	Primary Crop	Parent only	Parent only
Livestock	Ruminant	Parent only	Parent only

3.6.2 Rationale for Inclusion of Metabolites and Degradates

Although several metabolites were found in the rat metabolism study, all were found at low levels in the urine and feces. In addition, only parent was seen in the plant and ruminant studies, accounting for the majority of the TRR in plants, milk and ruminant tissues.

4.0 Hazard Characterization/Assessment

4.1 Hazard Characterization

The database is adequate for establishment of tolerances on the requested commodities, except for a developmental toxicity study in rat which was classified as **unacceptable/guideline.** However, this study provides adequate information to conclude that flufenoxuron was not fetotoxic at doses up to the limit dose. Only an acute oral toxicity study in rat was submitted, which was placed in Toxicity Category IV. Acute toxicity studies are not required for import tolerance.

The critical toxicological effects of flufenoxuron in mammals are on the hemopoeitic system. These effects include shifts in red blood cell parameters (decreased in hemoglobin, hematocrit, red blood cells and erythroid: myloid ratio) resulting in mild anemia and enhanced levels of methemoglobin and sulfhemoglobin in dogs and rats. Dogs appears to be more susceptible to the hematopoetic effects described above. It is likely that a metabolite (aniline metabolite (4-[2-chloro, α, α, α-trifluoro-p-tolyoxy]-2-fluoroaniline) is responsible for the formation of methemoglobin and sulfhemoglobin. Two oncogenicity studies in mice and one in rat; were conducted at doses exceeding 4 - 7.5x limit dose. One of the mouse studies conducted at 7.5x limit dose had an increased incidence of vascular tumors in males and females. Total incidence of vascular tumors (combined hemangiosarcomas of the liver and spleen) among high-dose females (16%) was significantly increased over controls (0%) and was slightly outside the published data for historical controls (mean: 3.7%; range: 0%-14%). The overall increase in splenic hemangiosarcomas in high-dose females (14%, $p<0.01$) and the increase in total vascular tumors in the liver of high-dose males (20% vs 12% in controls) occurred at doses that exceeded the limit dose. The mouse oncogenicity study was repeated with the high dose exceeding 1.5x limit dose; and there was no increased incidence of any tumors. Therefore, the increased incidences of vascular tumors are considered high dose effects only. Flufenoxuron exhibited a negative response in various genotoxicity screening assays. Based on the lack of evidence of carcinogenicity in both rats and mice carcinogenicity studies and negative response in various genotoxicity assays, flufenoxuron is considered "not likely to be carcinogenic to humans."

Flufenoxuron was not teratogenic in rats and rabbits at doses up to the limit dose (1000 mg/kg/day). It did not affect reproductive parameters in a 2-generation reproduction study at doses up to 10, 000 ppm (771.6 mg/kg/day). There was no evidence of increased susceptibility for flufenoxuron in the developmental toxicity study in rats. No adverse effects were observed in either dams or offspring at the limit dose. Although fetal external examination data were not provide in the study report and have been requested, their absence does not effect the current risk assessment. Evidence of increased susceptibility was observed in the developmental toxicity study in rabbits. Specifically, decreased fetal weight was observed in the absence of maternal toxicity; however, fetal effects were observed at the limit dose, and the NOAEL, which is one order of magnitude lower, is considered protective of this high-dose effect. In the 2-generation reproduction study, increased susceptibility of offspring was observed in the form of decreased body weight, since this effect was observed at a lower dose than the maternal NOAEL. However, a NOAEL for this effect in offspring was also observed, and it is considered protective of any effects at the offspring LOAEL. Based on this discussion, there is no concern for increased susceptibility of offspring following exposure to flufenoxuron.

Table 4.1.1. Acute Toxicity of Flufenoxuron

Guideline No.	Study Type	Results (LD50/LC50)	Toxicity Category
870.1100 (81-1)	Acute Oral (rat)	LD_{50}> 5000 mg/kg (M) LD_{50} > 5000 mg/kg (F)	IV
870.1200 (81-2)	Acute Dermal	NA	NA
870.1300 (81-3)	Acute Inhalation (rat)	NA	NA
870.2400 (81-4)	Primary Eye Irritation	NA	NA
870.2500 (81-5)	Primary Skin Irritation	NA	NA
87.2600 (81-6)	Dermal Sensitization (guinea pig)	NA	NA

Table 4.1.2. Toxicity Profile of Flufenoxuron Technical.

Guideline No./ Study Type	MRID No. (year)/ Classification /Doses	Results
870.3100 90-Day oral toxicity rodents (rat)	MRID 44448406 (1987) **Acceptable/guideline** 0, 50, 500, 5,000, 50,000 ppm M & F: 0, 2.5, 25, 250, 500, 2500 mg/kg/day	NOAEL = 25 mg/kg/day (M/F) LOAEL = 250 mg/kg/day (M/F), based on decreased hemoglobin levels and hematocrit levels, and red cell counts in females and decreased erythroid:myeloid ratios in males.
870.3100 90-Day oral toxicity rodents (mouse)	MRID 44448408 (1991) **Acceptable/Guideline** 0, 50, 500, 5,000, 50,000 ppm (Limit dose) M& F: 0, 7.5, 75, 750, 1500, 7500 mg/kg/day	NOAEL = 7500 mg/kg/day (M/F) LOAEL was not established.
870.3150 90-Day oral toxicity in nonrodents (dog)	MRID 44448409 (1987) **Acceptable/Guideline** in conjunction with the chronic dog study (MRID 44448411) 0, 0.05% (500 ppm), 0.5% (5000 ppm), 5% (50,000 ppm) in the diet 0, 38, 375, 3750 (3.5x Limit dose) mg/kg/day	NOAEL = 7.5 mg/kg/day (M/F; based on chronic toxicity study in dog). LOAEL = 38 mg/kg/day (M/F), based on decreased hemoglobin, hematocrit levels, and erythrocyte counts in males and increased absolute liver weights, bone marrow hyperplasia andmethemoglobinemia in males and females.
870.3200 21/28-Day dermal toxicity (rat)	NA[1]	NA
870.3250 90-Day dermal toxicity	NA	NA
870.3465 90-Day inhalation toxicity	NA	NA

Table 4.1.2. Toxicity Profile of Flufenoxuron Technical.

Guideline No./ Study Type	MRID No. (year)/ Classification /Doses	Results
870.3700a Pre-natal developmental in rodents (rat)	MRID 44448415 (1991) **Unacceptable/Guideline, upgradable upon review of stability and certain fetal external examination data.** 0, 10, 100, 1000 mg/kg/day (Limit dose) , GD 6 - 16, inclusive	**Maternal** NOAEL = 1000 mg/kg/day LOAE was not observed. **Developmental** NOAEL = 1000 mg/kg/day LOAEL was not observed.
870.3700b Pre-natal developmental in nonrodents (rabbit)	MRID 44448416 (1991) **Acceptable/Guideline** 0, 10, 100, 1000 mg/kg/day (Limit dose) GD 6-18, inclusive	**Materna**l NOAEL 1000 mg/kg/day LOAEL was not established. **Developmenta**l NOAEL = 100 mg/kg/day LOAEL = 1000 mg/kg/day, based on delayed fetal growth.
870.3800 Reproduction and fertility effects (rats)	MRID 44448417 (1991) **Acceptable/Guideline** 0, 50, 190, 710, 10,000 ppm M: 0, 3.75, 14.33,53.64, 771.6 mg/kg/day F: 0, 4.26, 16.0, 60.98, 907.4 mg/kg/day	Parental Toxicity NOAEL = 771.6/907.4 mg/kg/day (M/F) LOAEL was not established. Rep. Toxicity NOAEL = 771.6/907.4 mg/kg/day (M/F) LOAEL was not established. Offspring Toxicity NOAEL = 3.75/4.26 mg/kg/day (M/F) LOAEL = 14.33/16.0 mg/kg/day, based on lower pup body weight.
870.4100b Chronic toxicity dogs	MRID 44448411 (1989) 0, 10, 100, 500, 50,000 ppm M & F: 0, 0.75, 7.5, 37.5, 3750 mg/kg/day **Acceptable/Guideline**	NOAEL = 7.5 mg/kg/day (M/F) LOAEL = 37.5 mg/kg/day (M/F), based on decreased erythrocyte counts, and mean hemoglobin concentration and increased mean cell volume, platelet counts in males, methemoglobinemia and sulfhemoglobinemia in males and females.
870.4300 Combined chronic toxicity/carcinogenicity rodents (rat)	MRIDs 44448410 & 44448412 (1990) **Acceptable/Guideline** Chronic phase: 0, 1, 5, 50, 500, 5000, 50000 (> Limit dose) ppm M: 0, 0.044, 0.226, 2.21, 22.03, 232.5, 2470.6 mg/kg/day F: 0, 0.05, 0.279, 2.82, 28.33, 301.0, 3205.6 mg/kg/day Carcinogenicity phase: 0, 500, 5000, 50000 (> Limit dose) ppm M: 0, 21.57, 217.5, 2289.8 mg/kg/day F: 0, 25.92, 276.4, 2900.9 mg/kg/day	NOAEL = 22.03/28.33 mg/kg/day (M/F) LOAEL = 232.5/301 mg/kg/day (M/F), based on decreased body weight and weight gain. **No evidence of carcinogenicity**.

Table 4.1.2. Toxicity Profile of Flufenoxuron Technical.

Guideline No./ Study Type	MRID No. (year)/ Classification /Doses	Results
870.4300 Carcinogenicity mice	MRID 44448414 (1992) **Acceptable/Guideline** 0, 500, 5,000, 50,000 (> Limit dose) ppm M: 0, 56, 559, 7356 mg/kg/day F: 0, 73, 739, 7780 mg/kg/day $B_6C_3F_1$ strain	NOAEL = 559/73 mg/kg/day (M/F) LOAEL = 7356/739 (M/F), based on decreased body weight and body weight gain and liver lesions in males and females and spleen lesions in males. **Dosing was adequate (> 7.5x the limit dose). Total incidence of vascular tumors (combined hemangiosarcomas of the liver and spleen) among high-dose females (16%) was significantly increased over controls (0%) and was slightly outside the published data for historical controls (mean: 3.7%; range: 0%-14%). The overall increase in splenic hemangiosarcomas in high-dose females (14%, p<0.01) and the increase in total vascular tumors in the liver of high-dose males (20% vs 12% in controls) occurred at doses that exceeded the limit dose.**
870.4300 Carcinogenicity mice	MRID 44448413 (1996) **Acceptable/Guideline** 0, 100, 1,000, 10,000 (> limit dose) ppm M: 0, 15.3, 152, 1592 mg/kg/day F: 0, 17.4, 187, 1890 mg/kg/day $B_6C_3F_1$ strain	NOAEL = 1592/187 mg/kg/day (M/F) LOAEL = 1890 mg/kg/day (F), based on decreased body weight and body weight gains. LOAEL for males not established. **No evidence of carcinogenicity**
Gene Mutation 870.5100 reverse gene mutation assay in bacteria	44448419 (1991) **Unacceptable/guideline** *Salmonella typhimurium* strains TA98, TA100, TA1535, and TA1538; *Escherichia coli* strain WP2 were exposed to flufenoxuron at doses 0, 31.25, 62.5, 125, 250, 500, 1000, 2000, and 4000 µg/plate. Inappropriate positive controls for strains TA1535 and WP2 with +S9 and TA100, TA1537, TA1538, and TA98 -S9.	No evidence of induced mutant colonies over background.
Gene Mutation 870.5300 forward gene mutation assay in mammalian cells	46607201 (1986) **Acceptable/guideline** CH V79 cells exposed to flufenoxuron at concentrations up to 1350 µg/mL (±S9)	There was no evidence that flufenoxuron induced mutant colonies over background in the ± S9 activation.
Cytogenetics 870.5375 *in vitro* mammalian cytogenetic assay	46607202 (1987) **Acceptable/guideline** CHO cells exposed to flufenoxuron at doses up to 150 µg/mL (±S9).	No evidence of increased chromosomal aberrations in the presence of S9.
Cytogenetics 870.5375 *in vitro* mammalian cytogenetic assay	46607204 (1988) **Unacceptable/guideline** Rat liver epithelioid cells exposed to flufenoxuron at doses up to 450 µg/mL (-S9) and/or 160 µg/mL (+S9).	Not clastogenic with or without S9 activation, at any dose tested; however, the negative results were not followed-up experiment with a much longer harvest time and in the absence of S9.
Cytogenetics 870.5375 *in vitro* mammalian cytogenetic assay	46607205 (1992) **Acceptable/guideline** Human lymphocytes cultures exposed to flufenoxuron at doses up to 160 µg/mL (±S9)	No evidence of increased chromosomal aberrations in the ±S9 activation.

Table 4.1.2. Toxicity Profile of Flufenoxuron Technical.

Guideline No./ Study Type	MRID No. (year)/ Classification /Doses	Results
Cytogenetics 870.5385 *in vivo* mammalian cytogenetic assay	46607206 (1986) **Acceptable/guideline** CD rats oral dosed with flufenoxuron at doses up to 4000 mg/kg (>limit dose)	No increase in aberrant cells were seen in the bone marrow chromosomal aberration assay.
Other Effects 870.5395 *in vivo* mammalian cytogenetic assay	46607208 (1993) **Acceptable/guideline** 0, 500, 1000, and 2000 mg/kg (limit dose)	Did not induce micronucleated polychromatic erythrocytes (PMCEs) in bone marrow at any dose
Other Genotoxic Effects 870.5550 UDS synthesis in mammalian cell culture	46607207 (1988) **Unacceptable/guideline** 0, 188, 375, 750, and 1500 mg/kg oral gavage No justification for upper dose tested.	Did not induce UDS at any dose
Other Genotoxic Effects 870.5575 Mitotic gene conversion in *Saccharomyces cerevisiae*	44448419 (1991) **Acceptable/guideline** 0, 0.01, 0.1, 0.25, 0.5, and 1.0 mg/mL ± S9.	No increase in the mitotic gene conversion at either histidine or tryptophan loci ± S9.
Non-guideline Replicative DNA synthesis in rat hepatocytes	46607209 (1992) **Unacceptable/non-guideline** 0, 2000 and 4000 mg/kg oral gavage. No positive control data.	No increased in the incidence replicative DNA synthesis over untreated controls.
870.6200a Acute neurotoxicity screening battery	NA	NA
870.6200b 4-Week Subchronic neurotoxicity screening battery	46482203 (2003) **Acceptable/non-guideline** because of duration 0, 1000, 5000, 20000 ppm M: 0, 88.3, 435.4, 1774.6 mg/kg/day F: 0, 94.9, 474.5, 1934.4 mg/kg/day	NOAEL= 88.3/94.9 mg/kg/day (M/F) LOAEL = 435.4/474.5 mg/kg/day (M/F), based on decreased body weight and body weight gains in males.
870.6300 Developmental neurotoxicity	NA	NA
870.7485 Metabolism and pharmacokinetics (rat)	44448422 (1992) **Acceptable/guideline** M & F: 3.5 or 350 mg/kg single oral dose (labeled the aniline and difluorobenzene ring) , -single gavage dose of 5 mg/kg for 14 days followed by labeled 5 mg/kg,-in bile duct cannulated, single dose of 5 mg/kg	Metabolic fate of flufenoxuron was determined using two label positions (aniline and difluorobenzene ring). At the high dose, saturated absorption was observed. Approximately 86% of the low dose and 1% of the high dose was absorbed in 168 hours; the majority of which occurred within 48 hours. For the low dose ring, urine (9-27%) was major route of excretion (much less was in the urine at the high dose). Conversely, 93-102% of the high dose and 4-19% of the low dose was eliminated in feces. Elimination via expired air was insignificant. Biliary excretion using the aniline ring label showed that all the radioactivity in the feces of females and 40% of that in males are the biliary excretion products. Accumulation of radioactivity in muscle and adipose tissue 4 hours post dosing with 3.5 mg/kg benzyl label was 30% and 42%, respectively. At 168 hours post dose, these values were 6% and 19%, respectively, suggesting an accumulation in the adipose tissue. High doses of both labels resulted in negligible tissue burden (<0.3%), indicating saturation absorption. Urinary samples showed 10 urinary metabolites and parent accounted for ~ 5% of the dose in the urineand was considered non-significant. Fecal analysis indicated that parent compound accounting for the greatest portion of radioactivity. Most fecal metabolites represented < 1% of the administered dose. The results of the metabolism characterization studies with both label positions suggest that

Table 4.1.2. Toxicity Profile of Flufenoxuron Technical.

Guideline No./ Study Type	MRID No. (year)/ Classification /Doses	Results
		metabolism of flufenoxuron proceeds via hydrolysis to a benzoic acid metabolite, a phenyl urea metabolite (4-[2-chloro, α, α, α-trifluoro-p-tolyoxy]-2-fluorophenyl urea), an aniline metabolite (4-[2-chloro, α, α, α-trifluoro-p-tolyoxy]-2-fluoroaniline), and subsequently several minor components.
870.7600 Dermal penetration	NA	NA

[1] NA = not applicable.

4.2 FQPA Hazard Considerations

4.2.1 Adequacy of the Toxicity Database

The toxicology database for flufenoxuron is adequate, except for the developmental toxicity in rat. However, this study provides adequate information to conclude that flufenoxuron was not fetotoxic at doses up to the limit dose. The following acceptable studies are available:

- Developmental toxicity study in rabbits
- 2-generation reproduction study in rats

4.2.2 Evidence of Neurotoxicity

There is no concern for neurotoxicity resulting from exposure to flufenoxuron. In a 4-week subchronic neurotoxicity study, no evidence of neurotoxicity was observed. No evidence of clinical signs and neuropathology was observed in the available data from multiple studies; therefore, the chemical is not considered neurotoxic.

28-Day Neurotoxicity Study in Rats - In a subchronic neurotoxicity study (MRID 46482203), BAS 307 I (Flufenoxuron; 99.2% a.i., Batch #: SNPE 38/00) was administered in the diet to 10 Wistar rats/sex/group at doses of 0, 1000, 5000, or 20,000 ppm (equivalent to 0/0, 88.3/94.9, 435.4/474.5, or 1774.6/1934.4 mg/kg/day [M/F]) for 4 weeks. Functional observational battery (FOB) and motor activity were evaluated on Days -1 (prior to dosing) and 27. At termination, 5 rats/sex/group were perfused *in situ,* and tissues from the control and 20,000 ppm groups were examined microscopically. Positive control data were provided.

No treatment-related effects on mortality, clinical signs, food or water consumption, food efficiency, FOB, locomotor activity, brain weight, and gross or neuropathology parameters were observed.

The neurotoxicity LOAEL was not observed. The neurotoxicity NOAEL is 20,000 ppm (equivalent to 1774.6/1934.4 mg/kg/day [M/F]; exceeds the limit dose).

There was no evidence of neurotoxicity at any dose tested.

Systemic effects were limited to decreases (p<=0.05) in body weight (decr 6-10%) and body weight gain (decr 16-19%) in the >=5000 ppm males.

The systemic LOAEL is 5000 ppm (equivalent to 435.4/474.5 mg/kg/day [M/F]) based on decreased body weight and body weight gain in the males. The systemic NOAEL was 1000 ppm (equivalent to 88.3/94.9 mg/kg/day [M/F]).

The study is classified as **acceptable/non-guideline** and does not satisfy the guideline requirements (870.6200b) for a subchronic neurotoxicity study in the rat.

4.2.3 Developmental Toxicity Studies

Developmental Toxicity Study in rabbits: In a developmental toxicity study (MRID 44448416) flufenoxuron (Batch #16, 94.7% a.i.) was administered to 15 mated female New Zealand White rabbits/dose by gavage in 0.5% carboxymethylcellulose at dose levels of 0, 10, 100, or 1000 (limit dose) mg/kg bw/day on gestation days (GDs) 6 through 18, inclusive. On GD 29, the surviving does were sacrificed and necropsied. Gravid uterine weight, corpora lutea count, and the number and position of live fetuses, "fetal deaths," "early embryonic deaths," and "late embryonic deaths" were recorded. All fetuses were examined externally, and live fetuses were weighed and sexed internally. Approximately two thirds of the live fetuses from each litter were examined for both visceral and skeletal alterations, and the remaining one third were subjected to a visceral examination which included evaluation of the internal structures of the head.

There were no treatment-related effects on maternal survival, clinical signs, body weight, food consumption, or gross pathology. **The maternal toxicity NOAEL is greater than or equal to 1000 (limit dose) mg/kg bw/day. The maternal toxicity LOAEL for flufenoxuron in New Zealand white rabbits is not identified.**

There were no treatment-related effects on live litter size, fetal death, early or late embryonic deaths, post-implantation loss, or fetal sex ratio. The mean fetal weight for the combined sexes was slightly lower for the high-dose group compared with that of the control group. There was a slight treatment-related effect on fetal ossification at the highest dose level. This was seen as increased litter incidences of several ossification parameters (retarded hyoid arch; partially unossified tibial, femoral, and/or humeral epiphyses; unossified long bone epiphyses; and retarded pelvic bones) and decreased fetal and litter incidences of ossified olecranon(s).

The developmental toxicity LOAEL for flufenoxuron in New Zealand white rabbits is 1000 mg/kg bw/day, based on delayed fetal growth. The developmental toxicity NOAEL is 100 mg/kg bw/day.

This developmental toxicity study in the rabbit is classified **Acceptable/Guideline** and satisfies the guideline requirement for a developmental toxicity study in the rabbit [OPPTS 870.3700b; OECD 414]. It should be noted that the study was conducted prior to the implementation of the current guideline (see deficiency section).

Developmental Toxicity Study in Rats: In a developmental toxicity study (MRID 44448415) flufenoxuron (94.7% a.i., batch # 16) was administered to 26 female Sprague-Dawley rats/dose by gavage at dose levels of 0, 10, 100, or 1000 mg/kg bw/day from days 6 through 16 of gestation. On GD 20, dams were sacrificed, subjected to gross necropsy, and all fetuses examined externally (external examination data not provided). Approximately one-half of the fetuses were examined viscerally (by Wilson's technique), and the remaining one-half of the fetuses were examined for skeletal malformations/variations and were examined grossly for visceral abnormalities. The total numbers of fetuses examined (number of litters) were 327(25), 280(21), 296(23), and 301(23) for the 0, 10, 100, and 1000 mg/kg bw/day groups, respectively.

No adverse, treatment-related maternal effects were noted in clinical signs, absolute body weight, body weight gain, feed consumption, or gross necropsy findings.

Therefore, the maternal toxicity NOAEL for flufenoxuron in rats is ≥1000 mg/kg/day (limit dose), and the maternal toxicity LOAEL could not be established.

No treatment-related, statistically significant effects on pregnancy rates, number of corpora lutea, pre- or post-implantation losses, resorptions/dam, fetuses/litter, or fetal sex ratios were observed in the treated groups compared with the controls. No dam had complete litter resorption. No treatment-related visceral or skeletal malformations/variations were observed in any groups. External examination data were not provided.

Therefore, the developmental toxicity NOAEL for flufenoxuron in rats is ≥1000 (limit dose) mg/kg/day, and the developmental toxicity LOAEL could not be established.

The developmental toxicity study in the rat is classified **Unacceptable/Guideline** and does not satisfy the guideline requirement for a developmental toxicity study (OPPTS 870.3700; OECD 414) in the rat. It is upgradable if the following information is supplied: fetal external examination data.

4.2.4 Reproductive Toxicity Study

In a 2-generation reproduction study (MRID 44448417 & 44448418) WL115110 (97.0% a.i., batch #7016) was administered to 28 CrL: CD® (SD) BR VAF/Plus rats/sex/dose in the diet at levels of 0, 50, 190, 710 or 10,000 ppm. Premating doses for the F_0 parental animals were 0, 3.75, 14.33, 53.64, or 771.6 mg/kg bw/day for males and 0, 4.26, 16.0, 60.98, or 907.4 mg/kg bw/day for females. The premating interval was 70 days for the F_0 generation and 84 days for the F_1 generation. Two litters were produced for each generation. F_1 parental generation animals were selected from the B litter.

Due to pup mortality in the 2-generation study, a cross-fostering study (MRID 44448418) was initiated in which WL115110 (97.0% a.i., batch # 7016, ST87/129) was administered to 25 male and 50 female CrL: CD® (SD) BR VAF/Plus rats/dose in the diet at levels of 0 or 20,000 ppm (equivalent to 0 or 1633 mg/kg bw/day). The females received the test article during a 10-week premating period, then through mating and gestation. Treatment of all females was discontinued at parturition. The males were maintained on basal diet for the first nine weeks and were treated

for one week prior to, and during, mating. As soon as possible after parturition, the young in each litter were culled to 8 pups (ideally 4 males and 4 females) and were cross-fostered so that the control dam nursed the litter of the treated dam and *vice-versa*. Also included were groups of control and treated dams that nursed their own pups either throughout lactation or were sacrificed on lactation day 1, 7, 14, or 21 for analysis of Flufenoxuron content in milk and abdominal fat.

Minimal parental effects were evidenced by statistically significant changes ($p<0.05$ or 0.01) in body weight adjusted for weight at week 0 as covariate at 710 and 10,000 ppm during premating week 8 for the F_0 females and at weeks 4, 8, and 12 at 190-10,000 ppm for the F_1 males, but the differences from the control weight were less than 8%, not biologically significant. In the cross-fostering study, a slightly lower body weight of the treated females throughout premating ($p \leq 0.05$ or 0.01, reductions of about 3 to 4%) was not considered treatment-related. The minimal decreases in body weight, combined with a lack of treatment-related effects on food consumption and food efficiency values, indicate that the changes in body weight are not treatment-related. At necropsy, no treatment-related effects were revealed by macroscopic or microscopic examination. Absolute weight of the adrenals from females of both generations was significantly increased at 10,000 ppm ($p<0.05$, increase of 10.7% [F_0]; $p<0.01$, increase of 15.3% [F_1]), but microscopic examination was not performed to further evaluate the organ weight changes.

The parental systemic toxicity NOAEL for flufenoxuron in male and female rats is ≥10,000 ppm (≥771.6 mg/kg/day for males and ≥907.4 mg/kg/day for females). The parental systemic toxicity LOAEL was not identified.

Decreased survival was observed in the two generations of offspring during lactation, as evidenced by the increased number of total litters lost among the four groups of offspring at 710 and 10,000 ppm (losses of 8 and 13 litters, respectively, compared to the loss of one control litter). Mean litter size was statistically decreased ($p<0.05$ or 0.01) post-culling in the F_{1a}, F_{2a}, and F_{2b} pups during lactation at 10,000 ppm; the lactation index for these treated litters was 89.2, 73.3, and 80.8%, respectively, compared with 98.7, 97.4, and 98.7%, respectively, for the controls.

At 10,000 ppm, body weight for the F_{1a}, F_{2a}, and F_{2b} pups was significantly decreased at the end of lactation ($p<0.05$, 0.01, or 0.001) due to an overall weight gain 88, 89, and 86%, respectively, of the control level. In the F_{1a} pups, body weight was also significantly decreased at 190 and 710 ppm on days 12 and 21. In the 190-, 710-, and 10,000-ppm groups weight gain by the pups of both litters and generations was decreased to 86-96% of the control level.

In cross-fostered offspring, no effects were seen on offspring viability or body weight.

The offspring toxicity LOAEL for flufenoxuron in male and female rats is 190 ppm (14.33 mg/kg/day for males and 16.0 mg/kg/day for females), based on lower pup body weight. The offspring toxicity NOAEL for male and female rats is 50 ppm (3.75 mg/kg/day for males and 4.26 mg/kg/day for females).

Mating performance and fertility of the males and females of the F_0 and F_1 parental generations were not affected by treatment with Flufenoxuron. In the cross-fostering study when treatment

was discontinued during lactation, the test article content in milk and fat decreased rapidly from 449 and 973 ppm, respectively, on day 1 to 9.5 and 48.5 ppm, respectively, on day 21. The half-life was about 2.3 days in milk and about 7.6 days in abdominal fat.

The reproductive toxicity NOAEL for flufenoxuron in male and female rats is ≥10,000 ppm (≥771.6 mg/kg/day for males and ≥907.4 mg/kg/day for females). The reproductive toxicity LOAEL was not identified.

This study is **Acceptable/Guideline** and satisfies the guideline requirement for a 2-generation reproductive study (OPPTS 870.3800; OECD 416) in rats.

4.2.5 Pre-and/or Post-natal Toxicity

There was no evidence of increased susceptibility for flufenoxuron in the developmental toxicity study in rats. No adverse effects were observed in either dams or offspring at the limit dose. Although fetal external examination data were not provide in the study report and have been requested, their absence does not effect the current risk assessment. Evidence of increased susceptibility was observed in the developmental toxicity study in rabbits. Specifically, decreased fetal weight was observed in the absence of maternal toxicity; however, fetal effects were observed at the limit dose, and the NOAEL, which is one order of magnitude lower, is considered protective of this high-dose effect. In the 2-generation reproduction study, increased susceptibility of offspring was observed in the form of decreased body weight, since this effect was observed at a lower dose than the maternal NOAEL. However, a NOAEL for this effect in offspring was also observed, and it is considered protective of any effects at the offspring LOAEL. Based on this discussion, there is no concern for increased susceptibility of offspring following exposure to flufenoxuron. If adverse effects are observed after submission of fetal external examination data for the developmental toxicity study in rats, this conclusion may be revised.

4.3 Recommendation for a Developmental Neurotoxicity Study

As no evidence of clinical signs and/or neuropathology was observed in subchronic or chronic toxicity studies, flufenoxuron is not considered to be neurotoxic. Therefore, a developmental neurotoxicity study is not required for flufenoxuron.

4.4 Hazard Identification and Toxicity Endpoint Selection

4.4.1 Acute Reference Dose (aRfD)

An acute dietary endpoint for child bearing females (females 13-49 years old) and the general U.S. population, including infants and children, was not established since an endpoint of concern attributable to a single dose was not identified. Quantification of acute risk for child bearing females (females 13-49 years old) and general U.S. population, including infants and children, is not required.

4.4.2 Chronic Reference Dose (cRfD)

A chronic dietary dose of 0.0375 mg/kg/day was determined on from the 2-generation reproduction toxicity in the rat. The LOAEL of 14.33 mg/kg/day was based on lower pup body weight during lactation days 4-21. This study provided the lowest NOAEL (3.75 mg/kg/day) in the database. The chronic PAD of 0.0375 mg/kg/day is based on interspecies extrapolation (10x), intraspecies variability (10x), and the FQPA 1x safety factor.

4.4.3 Incidental Oral Exposure (Short- and Intermediate-Term)

Endpoints for these scenarios were determined from the 2-generation reproduction toxicity in the rat. The LOAEL of 14.33 mg/kg/day was based on lower pup body weight during lactation days 4-21. This study provided the lowest NOAEL (3.75 mg/kg/day) in the database. This endpoint is appropriate for the route, duration of exposure (1-30 days) and population of concern (infants and children).

4.4.4 Dermal and Inhalation Exposure (Short, Intermediate and Long Term)

Endpoints for these scenarios were determined from the 2-generation reproduction toxicity in the rat. The LOAEL of 14.33 mg/kg/day was based on lower pup body weight during lactation days 4-21. The endpoint is appropriate for the duration of exposure. Since an oral study was selected for dermal and inhalation exposure assessment, a dermal penetration factor and/or inhalation factor of 100% oral equivalents were used.

4.4.5 Margins of Exposure

The level of concern for residential and occupational exposure is for MOEs < 100. However, as there are no registered or proposed domestic uses of flufenoxuron, no residential or occupational exposure assessments were conducted.

4.4.6 Recommendation for Aggregate Exposure Risk Assessments

The current action is for tolerances on commodities imported in the U.S. only; therefore, an aggregate assessment is not warranted.

4.4.7 Classification of Carcinogenic Potential

Based on lack of evidence of carcinogenicity in both rats and mice carcinogenicity studies, the chemical is considered as “not likely to be carcinogenic to humans.”

Table 4.4.7.1. Summary of Toxicological Dose and Endpoints for Flufenoxuron for Use in Human Risk Assessment[1]

Exposure Scenario	Dose Used in Risk Assessment, UF	FQPA SF and Level of Concern for Risk Assessment	Study and Toxicological Effects
Acute Dietary females 13-50 years of age	An endpoint of concern attributed to single dose effect was not identified in the database. Quantification of acute risk to general population including infants and children is not required.		
Acute Dietary general population including infants and children	An endpoint of concern attributed to single dose effect was not identified in the database. Quantification of acute risk to general population including infants and children is not required.		
Chronic Dietary all populations	NOAEL= 3.75 mg/kg/day UF = 100 **Chronic RfD** = 0.0375 mg/kg/day	FQPA SF = 1X **cPAD** = chronic RfD / FQPA SF = 0.0375 mg/kg/day	2-Generation Reproduction Toxicity - Rat LOAEL =14.33/16.0 (M/F) mg/kg/day, based on decreased body weights during lactation days 4-21.
Oral - All Durations[2] (Residential)	NOAEL = 3.75 mg/kg/day	**Residential LOC for MOE** = 100	2-Generation Reproduction Toxicity - Rat LOAEL = 14.33/16.0 (M/F) mg/kg/day, based on decreased body weights during lactation days 4-21.
Dermal - All Durations[2] (Occupational/ Residential)	oral NOAEL= 3.75 mg/kg/day (dermal absorption rate = 100%)	**Occupational LOC for MOE** = 100 **Residential LOC for MOE** = 100	2-Generation Reproduction Toxicity - Rat LOAEL = 14.33/16.0 (M/F) mg/kg/day, based on decreased body weights during lactation days 4-21.
Inhalation - All Durations[2] (Occupational/ Residential)	oral NOAEL= 3.75 mg/kg/day (inhalation absorption rate = 100%)	**Occupational LOC for MOE** = 100 **Residential LOC for MOE** = 100	2-Generation Reproduction Toxicity - Rat LOAEL = 14.33/16.0 (M/F) mg/kg/day, based on decreased body weights during lactation days 4-21.
Cancer (oral, dermal, inhalation)	"not likely to be carcinogenic to humans"		

[1] UF = uncertainty factor, FQPA SF = FQPA safety factor, NOAEL = no observed adverse effect level, LOAEL = lowest observed adverse effect level, PAD = population adjusted dose (a = acute, c = chronic) RfD = reference dose, MOE = margin of exposure.
[2] As there are no registered or proposed domestic uses of flufenoxuron, residential or occupational exposure assessments were not conducted. However, if in the future domestic uses are proposed that would result in occupational and/or residential exposure to fluflenoxuron, these endpoints will be applicable.

4.5 FQPA Safety Factor

The flufenoxuron risk assessment team evaluated the quality of the toxicology and exposure data; and, based these data, recommended that the FQPA SF be reduced to 1x. The recommendation is based on the following:

- There is no evidence of increased susceptibility in the developmental study in rats.
- In the rabbit developmental study, there is evidence of increased susceptibility; however,

the effects are well characterized and clear NOAELs and LOAELs are established. Since the effects occurred at the limit dose, the delayed fetal growth may be considered a high dose effect.

- In the 2-generation reproduction study in rat, there is evidence for the increased susceptibility; however, the effects were well characterized, clear NOAELs and LOAELs were established for offspring toxicities, and the endpoints were used for risk assessment. Therefore, there is no residual uncertainty for pre- and/or post-natal susceptibility.
- The toxicological database is complete for FQPA assessment.
- The chronic dietary food exposure assessment utilizes proposed tolerance level residues and assumes 100% CT information for all commodities. By using these screening-level assessments, actual exposures/risks will not be underestimated.

4.6 Endocrine Disruption

EPA is required under the Federal Food Drug and Cosmetic Act (FFDCA), as amended by FQPA, to develop a screening program to determine whether certain substances (including all pesticide active and other ingredients) "may have an effect in humans that is similar to an effect produced by a naturally occurring estrogen, or other such endocrine effects as the Administrator may designate." Following the recommendations of its Endocrine Disruptor Screening and Testing Advisory Committee (EDSTAC), EPA determined that there was scientific bases for including, as part of the program, the androgen and thyroid hormone systems, in addition to the estrogen hormone system. EPA also adopted EDSTAC's recommendation that the Program include evaluations of potential effects in wildlife. For pesticide chemicals, EPA will use FIFRA and, to the extent that effects in wildlife may help determine whether a substance may have an effect in humans, FFDCA has authority to require the wildlife evaluations. As the science develops and resources allow, screening of additional hormone systems may be added to the Endocrine Disruptor Screening Program (EDSP).

When the appropriate screening and/or testing protocols being considered under the Agency's EDSP have been developed, flufenoxuron may be subjected to additional screening and/or testing to better characterize effects related to endocrine disruption.

5.0 Exposure Characterization/Assessment

The residue chemistry data submitted in support of proposed petitions were summarized in the HED-memorandum dated 8/15/06 (J. Tyler; DP# 310518). The chronic dietary exposure assessment was completed in a HED-memorandum dated 8/15/06 (J. Tyler, DP# 330630).

5.1 Dietary Exposure/Risk Pathway

5.1.1 Residue Profile

Nature of the Residue in Plants and Livestock Commodities: The nature of flufenoxuron residues in the subject crops and ruminant commodities is understood based on the acceptable tomato, apple, grape and goat metabolism studies. For purposes of this petition, the residue of concern in plant and ruminant commodities is flufenoxuron *per se*. This conclusion applies only

to the use of flufenoxuron on fruits and fruiting vegetables. Any future uses on other crops, such as leafy vegetables, legumes, or cereal grains, may require the submission of additional metabolism data, and any new uses having a substantial impact on livestock and poultry diets should include a new ruminant metabolism study, and, potentially, a poultry metabolism study.

Analytical Methodology: An adequate HPLC/ultraviolet (UV) method (SAMS 432-3) and liquid chromatography (LC)/mass spectrometry (MS)/MS method (BASF Method 544/0) are available for collecting data on flufenoxuron residues in/on plant commodities. The limit of quantitation (LOQ) for flufenoxuron in/on plant commodities is 0.05 ppm for the HPLC/UV method and 0.01 ppm for the LC/MS/MS method. Method SAMS 432-3, which is also the proposed enforcement method for plant commodities, has been radiovalidated and undergone a successful independent laboratory validation (ILV) trial. However, the method, as currently written, is not adequate for tolerance enforcement as the confirmatory HPLC/UV analysis is not sufficiently distinct from the primary analytical method. The petitioner should revise the method to include a confirmatory analysis using LC/MS/MS, which has been shown to adequately detect and quantify flufenoxuron in BASF Method 544/0. As a successful ILV trail has already been conducted, the method has been forwarded to the Analytical Chemistry Branch of the Biological & Economics Analysis Division (ACB/BEAD) for a petition method validation (PMV) (Memo, J. Tyler, 8/10/05; D320112).

Adequate HPLC/UV methods are also available for collecting data on flufenoxuron residues in milk (Method SAMS 486-1) and livestock tissues (Method SAMS 457-2). The validated LOQ for flufenoxuron is 0.01 ppm in milk, 0.3 ppm in fat, and 0.1 ppm in other tissues. Method SAMS 486-1, which is the proposed enforcement method for milk, does not require radiovalidation (due to the similarity between the extraction procedures in the proposed method and the extraction procedures used in the metabolism studies) and has undergone a successful ILV. This method has been forwarded to ACB/BEAD for a PMV trial (Memo, J. Tyler, 8/10/05; DP# 320112). Although a successful ILV trial was conducted on HPLC/UV method SAMS 458-1 using fat samples, this method is distinct from SAMS 457-2 and is only for the analysis of fat. Therefore, a separate ILV trial should be conducted on Method SAMS 457-2 using samples of liver and muscle. Any proposed HPLC/UV method must also be revised to include directions for a confirmatory analysis using an analytical method that is distinct from the primary analytical method. In addition, radiolabeled method validation data are required for the proposed enforcement method using tissue samples from the goat metabolism study to ensure that the method will adequately extract endogenous flufenoxuron residues.

Storage Stability: Adequate storage stability data are available indicating that flufenoxuron is stable at -20°C in apples, grapes, oranges and cottonseed for up to 36 months and in milk and tissues for up to 12 months. These data adequately support the available crop field trials, which had a maximum storage interval of 22 months, and the cattle feeding study, in which samples were stored for up to 4-8 months. However, to support the apple and grape processing studies, storage stability data are required for frozen juice (apple or grape), wet apple pomace, and raisins stored for up to 9 months.

Magnitude of the Residue in Livestock: An adequate cattle feeding study is available reflecting dosing of dairy cattle at levels of 1.75, 5.25, and 17.5 ppm in the diet for up to 90 days. Based on the calculated maximum theoretical dietary burdens (MTDB) of 0.169 ppm for beef and dairy cattle, these dietary levels are equivalent to 10x, 31x and 100x the MTDB for beef and dairy cattle. Fractionation of a bulk milk sample indicated that flufenoxuron residues concentrate substantially (5.1x) in milk fat. The distribution of flufenoxuron residues among tissues was similar at each dose level, with residue levels being highest in fat and lowest in muscle. For the 1.75-ppm dose group (10x), maximum flufenoxuron residues were 6.95 ppm in fat, 0.83 ppm in liver, 0.44 ppm in kidney, and 0.28 ppm in muscle, and average residues were 2.30 ppm in fat, 0.75 ppm in liver, 0.34 ppm in kidney, and 0.14 ppm in muscle. After cessation of dosing at 17.5 ppm (100x), residues in milk declined slowly with an elimination half-life of 16 days, and substantial residues remained in fat (3.46-7.60 ppm) at 40 days post-treatment. The available data indicate that flufenoxuron residues transfer primarily to fatty tissues or fractions in cattle.

Using the calculated MTDBs for beef and diary cattle and the regression equations for residues in tissues and milk, estimated flufenoxuron residues in livestock at a 1x dose level are 0.13 ppm in milk, 0.41 ppm in liver, 0.15 ppm in kidney, 0.079 ppm in muscle, and 4.08 ppm in fat. Estimated residues in milk fat would be 3.5 ppm based on a 25x concentration factor from whole milk and the estimated milk residues. These levels support the time-limited tolerances for residues of flufenoxuron *per se* in the following livestock commodities: milk (0.20 ppm); milk, fat (4.0 ppm); and meat (0.10 ppm), meat byproducts (0.50 ppm), and fat (4.5 ppm) of cattle, goats, horses and sheep.

Magnitude of the Residue in Plants: The petitioner has submitted crop field trial data from various countries in Europe and South America to support the use of flufenoxuron on apples, pears, grapes (Europe only), and oranges grown for export to the U.S. Available residue decline data indicate that residues of flufenoxuron decline steadily at longer post-treatment intervals in apples, pears and grapes. The residue decline data indicate the residues in orange peel do not decline at longer post-treatment intervals. All residues were <LOQ in orange pulp; therefore, residue decline could not be determined.

Apples: The available field trial data are adequate to support time-limited tolerances on apples imported from Europe and South America. Although use patterns used in the apple field trials varied considerably from the use rates actually being proposed, the rates used in the field trials were generally higher than the proposed rates, and samples from the field trials were generally collected at shorter intervals than the preharvest intervals (PHIs) listed on the proposed labels. Although the overall HAFT was 0.61 ppm, due to the use patterns in the field trials, the available residue data are adequate to support a **tolerance of 0.50 ppm for residues of flufenoxuron *per se* on apple.**

Pears: The available field trial data are adequate to support the proposed uses on pears in France, Italy and Spain as the two submitted field trials were conducted at rates equivalent to 1-3x the maximum use rates in these countries and the samples were collected at the minimum PHI of 28 days. Considering the amount of pears imported from each country for which this use is proposed, and the type of formulation, maximum seasonal use rate and PHI being proposed for each country, additional pear field trial data are requested to support the uses in South America

(largest exporter of pears to U.S.). Therefore, the data are not adequate to support a permanent tolerance. However, the available residue data on apples are adequate to support a time-limited tolerance of 0.50 ppm for residue of flufenoxuron *per se* on pear. For a permanent tolerance, three additional trials should be conducted in Argentina (2 trials) and Chile (1 trial). These trials should be conducted at the maximum seasonal rate being proposed for the given country using the appropriate formulation (e.g. EC in Argentina). Duplicate samples of whole fruit should be collected from each trial at the minimum PHI.

Grapes: The available field trial data are adequate to support the proposed uses on grapes in Europe. The majority of the trials were conducted at rates well below (≤0.7x) the proposed maximum seasonal rates of 0.12 kg ai/ha for France, 0.15 kg ai/ha for Spain and Greece, and 0.20 kg ai/ha for Italy. The labels originally submitted in support of this petition are from 1995-1996; whereas, the most recent and complete, grape field trial data are from 1998-2003. HED notes that the discrepancy between the maximum label rates and field trials use rates may reflect changes in the label direction since 1996. The petitioner has stated that the application rates provided in the summary table are the actual proposed rates (via e-mail message from M. Suarez to J. Tyler; 1/11/06). The available grape field trial data indicate that the appropriate tolerance for residues of flufenoxuron *per se* on grapes is 0.70 ppm.

Oranges: The available orange field trial data are not adequate to support permanent tolerance on oranges. The European field trials were conducted at rates well below (0.1-0.7x) the proposed maximum use rate, and samples of whole oranges, which is the regulated raw agricultural commodity (RAC), were analyzed in only 3 of the 9 trials submitted. The European data indicate that residues of flufenoxuron concentrate in the peel, and very little seems to translocate to pulp. In addition, in the Brazilian trials, whole oranges were analyzed only in the 1992 study, but the actual application rate can not be determined. However, the Brazilian residue data also indicate that residues remain in the peel. The European and Brazilian data together are adequate to support a time-limited tolerance of 0.30 ppm (see Section 860.1550 Proposed Tolerances). To support the proposed use on oranges, a new set of field trials should be conducted. Based on the amount of orange products imported from the countries for which this use is being proposed, a total of 12 field trials are requested. Considering the level of imports from each country, and the type of formulation, maximum seasonal use rate and PHI being proposed for each country, the requested trials should be conducted in Brazil (6 trials), Spain (4 trials) and Greece (2 trials). These trials should be conducted at the maximum seasonal rate being proposed for a given country and use the appropriate formulation (e.g. EC in Brazil). Duplicate samples of whole fruit should be collected from each trial at the minimum PHI.

Magnitude of the Residue in Processed Commodities: Both of the apple processing studies had quantifiable residues of flufenoxuron in whole apples (0.02 and 0.029 ppm). In both studies, processing factors were <0.3-<0.5x for juice, 1.6-10.9x for dry pomace, and 0.6-2.1x for wet pomace. Average processing factors from both treatments were 0.4x for juice, 6.25x for dry pomace, and 1.35x for wet pomace. Because none of the apple field trials were conducted according to label directions, the recommended tolerance was use instead of the overall HAFT to determine potential residues in processed apple commodities. Based on average processing factors and recommended tolerance of 0.50 ppm, the maximum expected residues would be 0.2 ppm in juice and 0.675 ppm in wet pomace. As the maximum residues in apple juice were lower

than the recommended tolerance for apples (0.50 ppm), a separate tolerance will not be required for apple juice. In addition, as the use is on apples for importation into the U.S., wet apple pomace would not be imported into the U.S. Therefore, a separate tolerance is not required for wet apple pomace.

Seven of the nine grape processing studies had readily quantifiable residues in/on grape RAC samples (0.108-1.83 ppm). In these studies, processing factors for flufenoxuron residues were 0.4x in juice, <0.02-<0.09x in wine, and 1.26-3.76x in raisins. Average processing factors were 0.4x for juice, 0.04x in wine and 2.98x for raisins. Based on the 2.98x processing factor for raisins and HAFT residues of 0.66 ppm from the grape field trials, the maximum expected residues in raisins would be 1.97 ppm, which is above the recommended 0.70 ppm tolerance for grapes. Therefore, a separate tolerance should be established for grape, raisin at 2.0 ppm.

In the orange processing studies, residue data were not provided on dried pulp and finished oil, which are both regulated commodities of citrus. Data on the oil emulsion concentrate indicate that residues are likely to concentrate substantially (≥50x) in the final oil fraction. (It should be noted that, according to the study report, samples of cold pressed oil were taken, but residue data were not provided.) In addition, a description of the method used to analyze the concentrated oil emulsion was not provided, and there are no data supporting the stability of flufenoxuron in processed citrus fractions. Although inadequate, the available study does suggest that residues will not concentrate in juice (<0.5x), but will concentrate in dried peel (7.14x). The orange processing data do support a time-limited tolerance on 60 ppm on orange, oil.

5.1.2 Chronic Dietary Exposure and Risk

A chronic dietary exposure (general U.S. population and all population subgroups) assessment was conducted using the Dietary Exposure Evaluation Model - Food Commodity Intake Database™ (DEEM-FCID™; ver. 2.03) program which incorporates consumption data from the United Stated Department of Agriculture's (USDA's) Continuing Surveys of Food Intakes by Individuals (CSFII), 1994-1996 and 1998. The 1994-96, 98 data are based on the reported consumption of more than 20,000 individuals over two non-consecutive survey days. Foods "as consumed" (e.g., apple pie) are linked to EPA-defined food commodities (e.g. apples, peeled fruit - cooked; fresh or N/S; baked; or wheat flour - cooked; fresh or N/S, baked) using publicly available recipe translation files developed jointly by USDA/ARS and EPA. Consumption data are averaged for the entire U.S. population and within population subgroups for chronic exposure assessment.

For chronic exposure and risk assessment, an estimate of the residue level in each food or food-form (e.g., orange or orange juice) on the food commodity residue list is multiplied by the average daily consumption estimate for that food/food form. The resulting residue consumption estimate for each food/food form is summed with the residue consumption estimates for all other food/food forms on the commodity residue list to arrive at the total average estimated exposure. Exposure is expressed in mg/kg body weight/day and as a percent of the cPAD. This procedure is performed for each population subgroup.

HED's level of concern is when the exposure is greater than 100% of the PAD. That is, estimated exposures above this level are of concern, while estimated exposures at or below this level are not of concern. The DEEM-FCID™ analysis estimates the dietary exposure of the U.S. population and various population subgroups. The results reported in Table 5.1.2.1 are for the U.S. Population, all infants (<1 year old), children 1-2 years old, children 3-5 years old, children 6-12 years old, youth 13-19 years old, females 13-49 years old, males 20-49 years old, and adults 50+ years old.

An unrefined, chronic dietary exposure assessment was performed for the general U.S. population and various population subgroups assuming tolerance-level residues and 100% CT for all commodities. As there are no proposed domestic uses of flufenoxuron, drinking water was not incorporated directly into the dietary assessment. This assessment concludes that the chronic dietary exposure estimates are below HED's level of concern (<100% cPAD) for the general U.S. population (15% cPAD) and all population subgroups. The most highly-exposed population subgroup is children 1-2 years old at 63% cPAD.

Table 5.1.2.1 Summary of Dietary Exposure and Risk for Flufenoxoron.

Population Subgroup	Chronic Dietary		
	cPAD (mg/kg/day)	Exposure (mg/kg/day)	% cPAD
General U.S. Population	0.0375	0.005262	14
All Infants (< 1 yr)		0.008469	23
Children 1-2 yrs		**0.023448**	**63**
Children 3-5 yrs		0.016271	43
Children 6-12 yrs		0.008996	24
Youth 13-19 yrs		0.004562	12
Adults 20-49 yrs		0.003334	8.9
Adults 50+ yrs		0.002961	7.9
Females 13-49 yrs		0.003227	8.6

5.2 Water Exposure/Risk Pathway

As there are currently no registered or proposed domestic uses for flufenoxuron, a drinking water assessment was not conducted.

5.3 Residential (Non-Occupational) Exposure/Risk Pathway

As there are currently no registered or proposed residential uses for flufenoxuron, a residential exposure assessment was not conducted.

6.0 Aggregate Risk Assessments and Risk Characterization

The current action is for tolerances on commodities imported in the U.S. only; therefore, an aggregate assessment is not warranted.

7.0 Cumulative Risk Characterization/Assessment

Unlike other pesticides for which EPA has followed a cumulative risk approach based on a common mechanism of toxicity, EPA has not made a common mechanism of toxicity finding as to flufenoxuron and any other substances, and flufenoxuron does not appear to produce a toxic metabolite produced by other substances. For the purposes of this tolerance action, therefore, EPA has not assumed that flufenoxuron has a common mechanism of toxicity with other substances. For information regarding EPA's efforts to determine which chemicals have a common mechanism of toxicity and to evaluate the cumulative effects of such chemicals, see the policy statements released by EPA's OPP concerning common mechanism determinations and procedures for cumulating effects from substances found to have a common mechanism on EPA's website at http://www.epa.gov/pesticides/cumulative/.

8.0 Occupational Exposure/Risk Pathway

As there are currently no registered or proposed domestic uses for flufenoxuron, an occupational exposure assessment was not conducted.

9.0 Data Needs and Label Requirements

9.1 Toxicology

- Confirmatory data on external fetal observations from the developmental toxicity study in rat should be submitted.

9.2 Residue Chemistry

- The most recent versions of BASF flufenoxuron labels should be submitted along with translations.

- The proposed tolerance enforcement method for plant commodities (HPLC/UV Method SAMS 432-3) should be revised to include an alternate confirmatory analysis using LC/MS/MS, which has been shown to adequately detect and quantify flufenoxuron in BASF Method 544/0.

- The proposed tolerance enforcement method for milk (HPLC/UV Method SAMS 486-1) should be revised to include an alternate confirmatory analysis that is distinct from the primary analytical method.

- To support the use of Method SAMS 457-2 as an enforcement method, an ILV trial should be conducted with this method using samples of liver and muscle fortified at the method LOQ and the recommended tolerances for meat byproducts (0.8 ppm) and meat (0.2 ppm). Any proposed HPLC/UV method must also be revised to include a confirmatory analysis using a analytical method that is distinct from the primary analytical method. In addition, radiolabeled method validation data are requested for the proposed enforcement method using tissue samples from the goat metabolism study.

- Completion of adequate PMV by the ACB.

- The EPA National Pesticide Standards Repository has requested that additional reference standard be submitted for flufenoxuron.

- To support the available grape and apple processing studies, data are requested demonstrating the stability of flufenoxuron in frozen samples of juice (apple, grape or orange), wet apple pomace, raisins, dried pulp and orange oil for up to 9 months.

- Three additional pear field trials should be conducted in Argentina (2 trials) and Chile (1 trial) using the appropriate formulation at the maximum seasonal rate being proposed for the given country. Duplicate samples of whole fruit should be collected from each trial at the minimum PHI.

- New orange field trials are requested. Based on the amount of orange products (fresh fruit and juice) imported from each country, and the type of formulation, maximum seasonal use rate and PHI being proposed for each country, a total of 12 field trials should be conducted, with the trials being distributed between Brazil (6 trials), Spain (4 trials) and Greece (2 trials). These trials should be conducted at the maximum seasonal rate being proposed for a given country and use the appropriate formulation (e.g. EC in Brazil). Duplicate samples of whole fruit should be collected from each trial at the minimum PHI.

- Processing data on dried pulp and finished orange oil are requested.

- Revised Section F to include the following HED-recommended tolerances and the correct commodity definitions: apple (0.50 ppm); grape (0.70 ppm); grape, raisin (2.0 ppm); cattle, meat (0.10 ppm); cattle, fat (4.5 ppm); cattle, meat byproducts (0.50 ppm); goat, meat (0.10 ppm); goat, fat (4.5 ppm); goat, meat byproducts (0.50 ppm); horse, meat (0.10 ppm); horse, fat (4.5 ppm); horse, meat byproducts (0.50 ppm); sheep, meat (0.10 ppm); sheep, fat (4.5 ppm); sheep, meat byproducts (0.50 ppm); milk (0.20 ppm); milk, fat (4.0 00m); orange (0.30 ppm); orange, oil (60 ppm); and pear (0.50 ppm).

10.0 References

DP#: 310518
Title: Flufenoxuron. PP#8E4943. Request for Tolerances on Apples, Pears, Grapes, Oranges and Livestock Commodities Imported in the United States (U.S.). Summary of Analytical Chemistry and Residue Data.
From: J. Tyler
To: M. Suarez
Date: 8/15/06

DP#: 330630
Title: Flufenoxuron. Chronic Dietary Exposure and Risk Assessment for New Active Ingredient.
From: J. Tyler
To: J. Tyler
Date: 8/15/06

cc: J. Tyler, G. Reddy
RDI: RAB1 (8/2/06), RARC [C. Swartz (8/10/06), J. Liccione (8/10/06)], F. Fort (8/15/06), PV Shah ((8/15/06)
J. Tyler:S-10943:(703)305-5564:7509P:RAB1

Appendices

1.0 TOXICOLOGY DATA REQUIREMENTS

The requirements (40 CFR 158.340) for Food Use for Flufenoxuron are presented below. Use of the new guideline numbers does not imply that the new (1998) guideline protocols were used.

Test		Technical	
		Required	Satisfied
870.1100	Acute Oral Toxicity	yes	yes
870.1200	Acute Dermal Toxicity	no	no
870.1300	Acute Inhalation Toxicity	no	no
870.2400	Primary Eye Irritation	no	no
870.2500	Primary Dermal Irritation	no	no
870.2600	Dermal Sensitization	no	no
870.3100	Oral Subchronic (rodent)	yes	yes
870.3150	Oral Subchronic (nonrodent)	yes	yes
870.3200	21-Day Dermal	no	no
870.3250	90-Day Dermal	no	no
870.3465	90-Day Inhalation	no	no
870.3700a	Developmental Toxicity (rodent)	yes	no[1]
870.3700b	Developmental Toxicity (nonrodent)	yes	yes
870.3800	Reproduction	yes	yes
870.4100a	Chronic Toxicity (rodent)	yes	yes
870.4100b	Chronic Toxicity (nonrodent)	yes	yes
870.4200a	Oncogenicity (rat)	yes	yes
870.4200b	Oncogenicity (mouse)	yes	yes
870.4300	Chronic/Oncogenicity	yes	yes
870.5100	Mutagenicity—Gene Mutation - bacterial	yes	no
870.5300	Mutagenicity—Gene Mutation - mammalian	yes	yes
870.5375	Mutagenicity—Structural Chromosomal Aberrations	yes	yes
870.5395	Mutagenicity—Other Genotoxic Effects	yes	yes
870.6100a	Acute Delayed Neurotox. (hen)	no	-
870.6100b	90-Day Neurotoxicity (hen)	no	-
870.6200a	Acute Neurotox. Screening Battery (rat)	no	-
870.6200b	90 Day Neuro. Screening Battery (rat)	no	-
870.6300	Develop. Neuro	no	-
870.7485	General Metabolism	yes	yes
870.7600	Dermal Penetration	no	no
Special Studies for Ocular Effects			
	Acute Oral (rat)	no	-
	Subchronic Oral (rat)	no	-
	Six-month Oral (dog)	no	-

[1] upgradable; information was used for risk assessment.

2.0 NON-CRITICAL TOXICOLOGY STUDIES

MRID No.: **Citation**

44448406. Esdaile, D.(1987). WL 115110: A 90-day feeding study in rats. Shell Research Limited, Sittingbourne Research Centre, Sittingbourne, Kent, ME9 8AG, England. Report No. SBGR.86.256. June 9, 1987. Unpublished.

44448407 Esdaile, D.(1986). WL 115110: A 28-day feeding study in rats. Shell Research Limited, Sittingbourne Research Centre, Sittingbourne, Kent, ME9 8AG, England. Report No. SBGR.86.085. October 30, 1986. Unpublished.

EXECUTIVE SUMMARY: In a 90-day oral toxicity study (MRID 44448406) flufenoxuron (91.5-96.6% a.i., Batch no. 16) was administered to groups of 10 Fischer 344 rats/sex/dose in the diet at concentrations of 50, 500, 5000, 10,000 or 50,000 ppm (equivalent to 2.5, 25, 250, 500 and 2500 mg/kg/day, respectively, based on a food conversion factor of 0.05). Twenty control rats/sex were maintained on basal diet without the test compound.

There were no toxicologically significant compound-related effects based on the assessment of mortality, clinical signs, body weight, food consumption, urinalysis, ophthalmic examination, gross necropsy and histopathology. Evidence of regenerative anemia, which included statistically significant ($p \leq 0.01$) decreases in red blood cell count, hematocrit and hemoglobin levels and significant increases in reticulocytes ($p \leq 0.01$), hemoglobin variant production, and spleen weights ($p \leq 0.01$) was seen in females only between 5000 and 50,000 ppm. Further, evidence of anemia was associated with a significant increase in methemoglobinemia in males and females at 5000 and 50,000 ppm. A significant ($p \leq 0.01$) increase in medullary hematopoiesis and a two-fold decrease in the myeloid to erythroid ratio was seen at 50,000 ppm in both males and females.

The LOAEL = 5000 ppm (~250 mg/kg/day) based on a two-fold increase in a hemoglobin variant production and increased RBC turnover in females and increased methemoglobinemia in males and females. The NOAEL = 500 ppm (25 mg/kg/day) in males and females.

This 90-day oral toxicity study in the rat is **Acceptable/Guideline** and satisfies the guideline requirement for a 90-day oral toxicity study (OPPTS 870.3100; OECD 408) in the rat.

44448408 Esdaile, D.J. (1991) WL115110: A 90 day feeding study in mice and Flufenoxuron (WL115110): A 28 day range-finding feeding study in mice. Shell Research Limited, Sittingbourne Research Centre, Sittingbourne, Kent, England. Reports Nos. SBGR. 86.257 and SBGR.91.179. September 16, 1991. Unpublished.

EXECUTIVE SUMMARY: In a 90-day oral study (MRID 44448408), flufenoxuron (96.6% a.i., batch/lot # ST86/165) was administered in the diet to groups of 10 male and 10 female mice in the treated groups and 20 male and 20 female mice in the control groups. An additional subgroup of ten mice/sex/dose group in the treated groups and twenty mice/sex for controls were used to provide blood for clinical chemistry. The concentrations administered were 0 (controls), 50, 500, 5,000, 10,000, or 50,000 ppm (equivalent to 0, 7.5, 75, 750, 1500 or 7500 mg/kg/day, respectively, based on a food conversion factor of 0.15). Doses were based on a 28-day range finding study conducted prior to the 90 day study by the same laboratory.

Five animals either died or were sacrificed prematurely during the study. These included two females in the 5,000 ppm group, two males in the 10,000 ppm group and one female in the 50,000 ppm subgroup used for clinical chemistry bleeding. The causes of death were not determined in necropsy and did not appear to be treatment-related. The only clinical sign observed in any group, treated or controls, was the presence of pale feces in the highest-dosed group in both males and females throughout the study which appeared to be unabsorbed compound.

Mean body weight was decreased in the highest-dose (50,000 ppm) males compared to controls throughout the study, although these decreases were <10% of controls. Mean body weight gain was also decreased in the 50,000 ppm males starting at week three and ranged from 14-28% less than controls without a time-response. Overall, the high-dose males had a total weight gain that was 22% less than controls. Treated females had body weight and weight gain comparable to controls throughout the study. Food consumption data were marginally collected. From the limited data endpoints, males did not exhibit any treatment-related differences and no conclusions could be made on the females.

A statistically significant ($p \leq 0.01$) decrease in some erythrocyte parameters including: red blood cell count, erythrocyte cell size variability (red cell distribution width), mean hemoglobin, and mean hematocrit, were observed in high-dose males. High-dose females also demonstrated a statistically significant ($p \leq 0.01$) decrease in red cell width distribution. Bilirubin concentrations were statistically increased ($p \leq 0.01$) in both the treated male and female mice compared to controls at doses above 50 ppm and exhibited a dose response, although the increases were slight and did not increase to the extent expected to correspond with the increase in dose concentration. In males, other differences observed were decreases in urea nitrogen starting at 500 ppm and decreases in triglycerides starting at 5000 ppm; however, these did not have a dose-response. Females had a statistically significant decrease in urea in the 10,000 and 50,000 ppm groups that was dose-responsive. While some of the differences in the clinical chemistry and hematology results support a treatment-related anemia, additional parameters such as methemoglobulin values, reticulocyte counts, myeloid/erythroid ratios and splenic weights and appearances were within normal range.

A statistically significant ($p \leq 0.05$ or 0.01) increase in liver organ weight when adjusted to the terminal body weight was observed in the male and female mice beginning at the 500 ppm dose group; although there were not any corresponding hepatic lesions on gross or histopathological examination.

Body weight changes, hematology parameters and bilirubin levels in males and females at the highest dose tested were marginal and were within biological variation for this strain and of mice, therefore, considered to be no biological significance.

The NOAEL = 50,000 ppm 50,000 ppm (7500 mg/kg/day, HDT) which is 7.5x the limit dose. LOAEL was not established.

This 90-day study in the mouse is **Acceptable/Guideline** and satisfies the guideline requirement for an oral feeding study [OPPTS 870.3100; OECD 451] in mice.

44448409 Greenough, R.J., Duffen, J., and Goburdhun, K.R. (1987) WL 115110: 13 week oral toxicity study in dogs. Inveresk Research International Limited, Musselburgh, EH21 7UB, Scotland. November 10, 1987. Unpublished.

EXECUTIVE SUMMARY: In a 90-day oral toxicity study (MRID 44448409), WL 115110 (98 % a.i., batch # 6001) was administered to four beagle dogs/sex/dose in the diet at dose levels of 0, 0.05% (500 ppm), 0.5% (5000 ppm), or 5% (50,000 ppm) [0, 38, 375 or 3750 (> 3.5 times **limit dose**) mg/kg/day, respectively]. A mistake in the dose preparation of the 0.5% group was corrected in the second week of the study, and therefore the study was extended to 15 weeks.

There were no differences between the controls and the WL 115110 treated groups in body weights, food and water consumption, ophthalmology, and electrocardiography. There was no mortality during the study. The clinical observations were confined to pallor of the gums or sclera which occurred late in the study in the mid-and high-dose groups.

In all treated males, the levels of methemoglobin were increased in a dose-related manner and were significantly higher ($p<0.001$) in the mid-and high-dose groups than the controls. The effect was seen in males of the 50,000 ppm group by Week 2 ($p<0.05$). The levels of sulfhemoglobin were also increased ($p<0.05$ and $p<0.001$) in the mid-and high-dose groups. These changes were associated with the decreases in hemoglobin, hematocrit and erythrocyte counts in the mid-and high-dose groups at Week 9, with recovery by Week 15. In addition, these changes also correlated with the increase in mean cell volume of red blood cells in the mid-dose group (at Week 9) which persisted to Week 15 in the high dose group. The effect of WL 115110 on hematology parameters at doses $\geq$ 0.05% was first observed at Week 9, followed by some compensatory effect and recovery in the low-and mid-dose groups. The effect persisted to Week 15 in male beagle dogs that received the mid- and high doses. Similar effects of WL 115110 were observed in the treated females but to a lesser extent than males in the mid-and high-dose groups. In females, the presence of methemoglobinemia in the mid-and high-dose groups had no effect on red blood cells parameters. The destruction of red blood cells by WL 115110 was compensated by increased red blood cells production and was associated with the increased

incidence of yellow pigment deposition in liver Kupffer cells and in the bone marrow. Histologically, bone marrow hyperplasia was observed in the males and females at doses ≥ 0.05% of WL 115110.

Males showed increases in absolute and relative liver weight. Males in the high dose group exhibited a slight reduction in body weight gain. In contrast, no remarkable changes were found in females. There were a number of sporadic changes in clinical chemistry analyses found, such as the elevated cholesterol levels in mid-and high-dose groups of males. No remarkable changes were found in the females.

The systemic toxicity LOAEL is 38 mg/kg/day (500 ppm) based decreased hemoglobin, hematocrit levels and erythrocyte counts in males; increased absolute liver weights, bone marrow hyperplasia and methemoglobinemia in males and females. The systemic toxicity NOAEL was not established.

This study by itself is classified as **Unacceptable/guideline**, however, in combination with the chronic toxicity study (MRID 44448411) in dogs **satisfy** the guideline requirement for a 90-day oral toxicity study (OPPTS 870.3150; OECD 409) in dogs.

44448411 Greenough, R.J., R. Goburdhun, and C. Parkinson. (1989) WL 115110: 52 week oral toxicity study in dogs. Inveresk Research International, Musselburgh, EH21 7UB, Scotland. Laboratory Project Identification IRI635474, February 16, 1989. Unpublished.

EXECUTIVE SUMMARY: In a 52-week oral chronic toxicity study (MRID 44448411), WL 115110 (98 % a.i., batch # 7005) was administered to four beagle dogs/sex/dose in the diet at dose levels of 0, 10, 100, 500, or 50,000 (> limit dose) ppm (0, 0.75, 7.5, 37.5 or 3750 mg/kg/day).

There were no differences between the controls and the WL 115110 treated groups in body weights, food and water consumption, ophthalmology, electrocardiography, clinical chemistry, or urinalysis. There were no treatment related mortalities and no treatment related clinical observations. One female dog in the 500 ppm group diagnosed with "autoimmune hemolytic anemia" was humanely sacrificed.

Males that received 50,000 ppm WL 115110 showed increases in absolute liver weight of 136% of the control ($p<0.001$).

In males in the 50,000 ppm group, the levels of methemoglobin were increased from 208 to 296% of the control ($p<0.05$ or $p<0.01$) during Weeks 5 to 40. The levels of sulfhemoglobin were also increased from 214 to 680% of the control ($p<0.05$ and 0.001) in the same group through out the study period. In addition, in the 500 ppm group, there were increases of methemoglobin (159% of the control, $p<0.05$) but only in Week 27 and sulfhemoglobin (267 to 286% of the control, $p<0.05$) during Weeks 27 and 40. These changes were associated with a decrease in hemoglobin and erythrocyte values in the 50,000 ppm group and to a lesser extent, in

the 500 ppm group. In addition, these changes also correlated with an increase in mean cell volume of red blood cells in the 50,000 ppm group in Weeks 13 to 40 and in the 500 ppm group at Week 27. The effects of WL 115110 on hematology parameters at 50,000 ppm was first observed at Week 5 and continued. The destruction of red blood cells by WL 115110 was compensated by the increase of red blood cell production and platelets. These effects on the blood also correlated with increases in brown pigment deposition in the liver Kupffer cells, hemorsiderin deposition in the spleen, and in bone marrow hypercellular activity. Increased reticulocyte and platelet counts ranged from 213 to 350% and 151 to 191% of the controls, and attained statistical significance for dogs that received 50,000 ppm from Week 5 to the end of the study, respectively. In females, similar hematological effects as seen in males , were observed, however, to a lesser extent.

The LOAEL is 37.5 mg/kg/day (500 ppm), based on decreased erythrocytes counts, and mean cell hemoglobin concentration and increased mean cell volume, platelet counts in males, methemoglobinemia and sulfhemoglobinemia in males and females. The NOAEL is 7.5 mg/kg/day (100 ppm).

This chronic study is **Acceptable/Guideline** and satisfies the requirement for a chronic oral study [OPPTS 870.4100, OECD 452] in the dog.

44448410 Esdaile, D. 1990. WL115110: a 2 year chronic toxicity feeding study in rats. Shell Research, Ltd., Sittingbourne Research Centre, Sittingbourne, Kent, ME9 8AG, England. Laboratory Report No. SBGR.89.150, May 9, 1990. Unpublished

44448412 Esdaile, D. and P. Berry. 1992. WL115110: a two year oncogenicity feeding study in rats. Sittingbourne Research Centre, Sittingbourne, Kent, ME9 8AG, England. Laboratory Report No. SBGR.89.135, March 2, 1992. Unpublished.

EXECUTIVE SUMMARY: In chronic toxicity and carcinogenicity studies (MRID 44448410 and 44448412, respectively), WL115110 (flufenoxuron) (97.6% a.i., lot/batch # WB16867, 7003, ST87/002) was administered to groups of male and female Fisher 344 rats as described below. In the chronic study, groups of 20 male and 20 female Fisher 344 rats were administered the test material in feed at dietary concentrations of 1, 5, 50, 500, 5000, or 50,000 ppm for up to 2 years and 40 male and 40 females receiving the basal diet alone served as controls. The doses were 0, 0.044, 0.226, 2.21, 22.03, 232.5, and 2470.6 mg/kg/day, respectively, for males and 0, 0.05, 0.279, 2.82, 28.33, 301.0, and 3205.6 mg/kg/day, respectively, for females. In addition, groups of 20 (controls) or 10 (treated groups) rats of each sex were administered the same diets and sacrificed at 12 months for interim evaluation. In the carcinogenicity study, groups of 50 male and 50 female rats were administered the test article in feed at dietary concentrations of 0, 500, 5000, or 50,000 for 2 years. The doses were 0, 21.57, 217.5, and 2289.8, mg/kg/day, respectively, for males and 0, 25.92, 276.4, and 2900.9 mg/kg/day, respectively, for females.

No treatment-related effects on survival, clinical signs, or incidence of ophthalmoscopic abnormalities were observed in male or female rats receiving any dose of the test material. Sufficient numbers of rats survived to study termination for evaluation of late-developing effects.

No treatment-related effects were observed on any parameter in rats receiving the test material at concentrations of 1-500 ppm in the chronic or carcinogenicity study. Body weight and weight gain were decreased in males and females at 5000 and 50,000 ppm. Body weight was significantly ($p<0.01$ or <0.05) decreased by 2-10% in males from weeks 12-96 and 3-13% in female rats from weeks 3-28 and 88-104 at 50,000 ppm. At 5000 ppm, body weight was decreased by 2-8% in males from weeks 28-80 and by 3-7% in females at a few time points between week 20 and 104. Weight gain by both sexes was similar to that of controls during the first 13 weeks but was reduced by 28% and 31% in males and 26% and 24% in females from weeks 13-52 at 5000 and 50,000 ppm, respectively. Weight gain for male rats over the entire study was similar between the 5000- and 50,000-ppm groups and the controls due to almost no weight gain by controls during the second year of the study. Weight gain over the entire study was decreased by 10% and 14% in females at 5000 and 50,000 ppm, respectively compared with that of controls. In addition, weight gain was reduced by 22 and 25% and 33 and 26% in 5000- and 50,000-ppm group male and female rats, respectively, from week 16-52 for the interim sacrifice group. Food consumption was generally increased in males and females at 5000 and 50,000 ppm in the chronic and carcinogenicity studies. Food efficiency was not calculated.

Statistically significant changes in erythrocyte parameters provided evidence of mild anemia in male and female rats at 5000 and 50,000 ppm and was accompanied by significantly elevated serum bilirubin concentrations in 5000- and 50,000-ppm males at week 25 and at all intervals in 5000- and 50,000-ppm females. The incidence of lymphoid erythrophagocytosis, which was increased in 50,000-ppm males in the chronic study but not in the carcinogenicity study, may have been associated with the anemia in males. No evidence of erythrocyte hemolysis was observed in either study; therefore, the mild anemia is not considered adverse. Statistically significant changes in clinical chemistry parameters were not considered treatment-related or adverse.

Postmortem examination revealed no changes in organ weight (spleen, kidney, and liver in males and spleen, adrenals, liver, and ovaries in females) in the chronic and carcinogenicity studies that were considered adverse and related to treatment with the test material. No treatment-related increase in the incidences of gross or non-neoplastic microscopic lesions were observed in male or female rats receiving any dose of the test material.

The lowest-observed-adverse-effect level (LOAEL) for flufenoxuron in rats is 5000 ppm for male and female rats (232.5 and 301.0 mg/kg/day, respectively) based on decreased body weight and weight gain. The no-observed-adverse-effect level (NOAEL) is 500 ppm for both sexes (22.03 and 28.33 mg/kg/day, respectively).

At the doses tested, there was no treatment related increase in the incidence of tumors at any anatomical site compared to controls. Mononuclear cell leukemia, which occurred at a high incidence in male and female rats in the control group showed a negative trend in treated rats. Dosing was considered adequate based on decrease in body weight and/or weight gain in male and female rats. The dose at 50,000 ppm was two to three times the limit dose of 1000 mg/kg/day.

These chronic toxicity and carcinogenicity studies in the rat are **Acceptable/Guideline** when

evaluated together and satisfy the guideline requirement for a chronic toxicity/carcinogenicity study (OPPTS 870.4300; OECD 453) in the rat.

44448414 Esdaile, D.J. and P.H. Berry (1992) WL115110. A 2 year oncogenicity feeding study in mice. Shell Research Limited, Sittingbourne Research Centre, Sittingbourne, Kent, England. Laboratory report number SBGR.89.151. March 6, 1992. Unpublished

44448408 Esdaile, D.J. (1991) WL115110: A 90 day feeding study in mice and Flufenoxuron (WL115110): A 28 day range-finding feeding study in mice. Shell Research Limited, Sittingbourne Research Centre, Sittingbourne, Kent, England. Reports Nos. SBGR. 86.257 and SBGR.91.179. September 16, 1991. Unpublished

EXECUTIVE SUMMARY: In a carcinogenicity study (MRID 44448414 & 44448408), Flufenoxuron (97.6% a.i., Batch #WB16867; 7003; ST87/002) was administered in the diet to 60 $B_6C_3F_1$ mice/sex/dose at dietary concentrations of 0, 500, 5000, or 50,000 ppm (equivalent to 0, 56, 559, or 7356 mg/kg/day for males and 0, 73, 739, or 7780 mg/kg/day for females, respectively). Ten animals of each sex per group were killed at 12 months for interim evaluation. The remaining surviving animals were treated for 104 weeks.

There were no treatment-related effects on clinical signs of toxicity or measured hematological parameters. Treatment-related statistically significant ($p<0.05$ or $p<0.01$) decreases in body weight were noted among high-dose males (2%-12%) throughout most of the study and among females (5%-20%) from week 20 to week 104 of the study. Over 104 weeks of treatment, the body weight gain at the high-dose level was 21% lower for males and 30% lower for females compared to controls. In addition, there was a decrease in final body weight and body weight gain of 8% and 14%, respectively, for mid-dose females. Survival among treated male groups was comparable with the controls, with hepatocellular tumors the most common cause of death among high-dose males. Among mid- and high-dose females survival was lower (62% and 60%, respectively) compared to controls (70%) but did not reach statistical significance.

Statistically significant ($p<0.01$) increases in absolute liver weight were seen in high-dose females at 52 weeks (16%) and in high-dose males and females at 104 weeks (46%-56%) compared to respective controls. Statistically significant increases in liver weight in both sexes and spleen weight (43%, $p<0.01$) in high-dose females were related to histopathological changes. At the 50,000 ppm concentration, histopathological changes included increased occurrences of Kupffer cell aggregates in the liver of both sexes at 52 weeks (90% in both sexes compared with 0% in the controls) and at 104 weeks (94% and 88% in males and females, respectively, compared with 0% in the controls) as well as increased incidences of syncytial macrophages in the spleen (37% and 20% in males and females, respectively, compared with 0% in the controls). At study termination, liver changes at the high-dose consisted of individual cell necrosis (84% and 76% in males and females, respectively), hepatocellular hypertrophy (62% and 70% in males and females, respectively) and inflammation (24% and 20% in males and females, respectively), indicating chronic damage to the liver. An increased occurrence of Kupffer cell aggregates

(22%) was also noted in mid-dose females at 104 weeks. This finding was not associated with any other lesions of the liver. Statistically significant ($p<0.05$ or $p<0.01$) increases in the occurrence of ulcer-associated changes in the forestomach were noted in mid- (6/23, 26%) and high-dose (6/48, 13%) males. Not all animals in the mid-dose groups were subjected to examination.

The lowest-observed-effect-level (LOAEL) for flufenoxuron in female mice is 5000 ppm in the diet (739 mg/kg/day) based on decreased final body weight and weight gain and lesions of the liver (Kupffer cell aggregates); the LOAEL for male mice is 50,000 ppm in the diet (7356 mg/kg/day) based on decreased final body weight and weight gain and lesions of the liver (Kupffer cell aggregates, hepatocellular hypertrophy and individual cell necrosis) and spleen (syncytial macrophage aggregates). The NOAELs for female and male mice are 500 and 5000 ppm in the diet, respectively (73 mg/kg/day and 559 mg/kg/day, respectively).

Except for the low-dose group of males, the incidences of hepatocellular adenomas in this study (6%, 22%, 20% , respectively vs 30% in controls) were within the range of neoplasms for historical controls (mean: 30%; range: 14%-58%). Among male mice there were statistically significant increases (by pair-wise comparison) in the 500, 5000 and 50,000 ppm dose groups compared with the controls for hepatocellular carcinoma (38%, 30% and 30%, respectively vs 6% in controls); however, a dose-response relationship was not evident. The combined incidences of hepatocellular adenomas/carcinomas in the low-, mid- and high-dose male groups (44%, 52% and 50% , respectively) were not statistically significantly different from the control (36%) and were within the historical control range (14%-58%). Therefore, based on the lack of dose-response, and within the historical control incidence, the increased incidence of hepatocellular carcinoma in treated male groups in the present study was considered incidental to treatment.

Total incidence of vascular tumors (combined hemangiosarcomas of the liver and spleen) among high-dose females (16%) was significantly increased over controls (0%) and was slightly outside the published data for historical controls (mean: 3.7%; range: 0%-14%). The overall increase in splenic hemangiosarcomas in high-dose females (14%, $p<0.01$) and the increase in total vascular tumors in the liver of high-dose males (20% vs 12% in controls) occurred at doses that exceeded the limit dose.

The dosing was considered adequate and exceeded the limit dose of 1000 mg/kg/day at the high dose, resulting in significant decreases in body weight in high-dose males and females (12 and 19%, respectively) and body weight gain in high-dose males (21%) as well as in mid- and high-dose females (14% and 30%, respectively).

This carcinogenicity study in mice is **Acceptable/Guideline** and satisfies the guideline requirement for a carcinogenicity study [OPPTS 870.4200; OECD 451] in mice.

44448413 Broadmeadow, A. (1996) WL115110: Oncogenicity study by dietary administration to $B_6C_3F_1$ mice. Huntingdon Life Science Ltd., P.O. Box 2, Huntingdon, Cambridgeshire, PE18 6ES, Suffolk, England. Laboratory report number 95/0875. April 12, 1996. MRID . Unpublished.

44448414 Esdaile, D.J. and P.H. Berry (1992) WL115110. A 2 year oncogenicity feeding study in mice. Shell Research Limited, Sittingbourne Research Centre, Sittingbourne, Kent, England. Laboratory report number SBGR.89.151. March 6, 1992. Unpublished.

EXECUTIVE SUMMARY: In a carcinogenicity study (MRID 44448413 & 44448414), Flufenoxuron (97.8% a.i., Batch # 7003; ST87/002) was administered in the diet to 50 $B_6C_3F_1$ mice/sex/dose at concentrations of 0, 100, 1000, or 10,000 ppm (equivalent to 0, 15.3, 152, or 1592 mg/kg/day for males and 0, 17.4, 187, or 1890 mg/kg/day for females, respectively) for 104 weeks. There were no interim sacrifices made. All surviving mice were sacrificed after Week 104.

There were no treatment-related adverse effects on clinical signs, food consumption, food efficiency, hematological parameters or organ weight in either sex. Mortality was slightly higher among mid- and high-dose females (36% each group vs 22% in controls). However, these differences were not statistically significant by pairwise comparison with the controls. In each group at least 76% of males and 64% of females survived to study termination. A treatment-related statistically significant ($p<0.05$) decrease in body weight was observed in high-dose females at Week 104 (10%). In addition, statistically significant ($p<0.05$) decreases in body weight gain in high-dose females were noted from Week 13-26 (25%) leading to an overall 15% decrease by Week 104.

The survival of animals in this study was adequate to assess the carcinogenic potential of the test compound. The administration of flufenoxuron in the diet at concentrations above the limit dose (1592 mg/kg/day for males and 1890 mg/kg/day for females) did not influence the tumor incidence or the time of tumor onset in $B_6C_3F_1$ mice.

In conclusion, the lowest-observed-effect level (LOAEL) for systemic effects is 10,000 ppm in the diet for female mice (1890 mg/kg/day) based on decreased final body weight (10%) and body weight gain (15%); the LOAEL for males was not identified. The corresponding no-observed-effect levels (NOAELs) are 1000 ppm (187 mg/kg/day) and ≥10,000 ppm (≥1592 mg/kg/day), respectively.

There was no evidence of potential carcinogenicity of the test material in this study. At the doses tested, there was no treatment related increase in tumor incidence in any male or female treated groups. Dosing was considered adequate in females at the high dose based on a significant decrease in body weight gain (15%). However, no apparent toxicity was noted among males at the high dose which exceeded the limit dose of 1000 mg/kg/day.

This carcinogenicity study in the mouse is **Acceptable/Guideline** and satisfies the guideline requirement for a carcinogenicity study [OPPTS 870.4200; OECD 451] in mice.

44448419 Brooks, T.M. and D.E. Wiggins (1991) Microbial mutagenicity studies with WL115110. Shell Research Limited, Sittingbourne Research Centre, Sittingbourne, Kent, ME9 8AG, England. Report No.: SBGR.86.026, February 28, 1991. Unpublished.

EXECUTIVE SUMMARY: In a reverse gene mutation assay in bacteria (MRID 44448419), strains TA98, TA100, TA1535, TA1537 and TA1538 of *Salmonella typhimurium* and strain WP2(uvrA pKM101) of *Escherichia coli* were exposed to WL115110 [Batch No. 9 (Ref. 2014/092, 99 ± 2% a.i.)] in DMSO at concentrations of 0, 31.25, 62.5, 125, 250, 500, 1000, 2000 or 4000 µg/plate with and without metabolic activation (S9-mix). Two independent assays were performed using the standard plate assay procedure and all plating was in triplicate. A gene conversion assay at the histidine and tryptophan loci in *Saccharomyces cerevisiae* JD1 was also included. The cells were exposed in suspension cultures to WL115110 in DMSO at concentrations of 0, 0.01, 0.1, 0.25, 0.5 and 1.0 mg/mL with and without S9-mix. Exposure times were 16 hours without S9-mix and 2 hours with S9-mix. Cell viability was not affected at any tested concentration. The S9-fraction was obtained from Aroclor induced rat liver (no additional information provided).

WL115110 was tested to upper concentrations limited by solubility. In the Ames test, the test material formed a fine precipitate in the top agar at 31.25 µg/plate, a milky suspension at 1000 µg/plate and lumps of precipitate at higher concentrations. No cytotoxicity was seen at any concentration to any of the bacterial strains. The upper dose in the second assay was reduced to 2000 µg/plate because of excessive precipitation at 4000 µg/plate in the first assay. Inappropriate positive controls were used in the assays with bacterial strains TA1535 and WP2 with S9-mix and TA100, TA1537, TA1538 and TA98without S9-mix; therefore, the results from these assays were uninterpretable. However, the number of revertants per plate was not increased over the concurrent solvent control value at any test material concentration in bacterial strains TA100, TA1537, TA1538 and TA98 with S9-mix; or TA1535 and WP2 without S9-mix. No increase in the rate of mitotic gene conversion was seen at any test material concentration at either loci at any test material concentration with or without S9-mix in *S. cerevisiae*. The solvent control values were appropriate. The positive controls, where used appropriately, produced values that were desirable for the respective strains. Not all bacterial strains were exposed to both a direct and an indirect acting positive control. **There was no evidence of induced mutant colonies over background in the number of prototrophs per 10^6 surviving cells in the gene conversion assay.**

This study is classified as **Unacceptable/Guideline** and does not satisfy the intent of Test Guideline OPPTS 870.5100; OECD 471 for *in vitro* mutagenicity (bacterial reverse gene mutation) data because all bacterial strains were not exposed to both a direct and indirect acting positive control. This study can not be upgraded. This study is classified as **Acceptable/Guideline** and satisfies the intent of Test Guideline OPPTS 870.5575; OECD 481 for mitotic gene conversion in *Saccharomyces cerevisiae*.

46607201 Clare, M.G.(1986) *In vitro* mutagenicity studies with WL115110 (Insecticide) using cultured Chinese hamster V79 cells. Shell Research Limited, London. Sittingbourne Research Centre, Sittingbourne, Kent, ME9 8AG, England. Report No. SBGR.86.047, Experiment No. 3287, May 16, 1986. Unpublished.

EXECUTIVE SUMMARY: In a mammalian cell gene mutation assay to measure the induction of 6-thioguanine resistance (MRID 46607201), Chinese hamster V79 cells cultured *in vitro* were exposed to flufenoxuron (99 ± 2 % a.i., Batch No. 10B, dissolved in DMSO) at concentrations of 50, 150, 450, 900 and 1350 µg/mL in the presence, and 50, 150, 450 and 1350 µg/mL in the absence of S9 microsomes from rat liver for 4 hours.

Flufenoxuron was tested at concentrations ranging from 50 to 1350 µg/mL, which resulted in cell survivals of ~90 to 100% at the lowest dose and 10 to 25% survival at the highest dose. Small increases in the gene mutation frequency of treated cells relative to the control mutation frequency were occasionally seen. However, the variations were within the range normally encountered in this assay, the elevated values were not dose-related, were usually confined to a single flask and were not reproducible from one experiment to another. It was concluded that flufenoxuron did not increase the mutation frequency to 6-thioguanine resistance in Chinese hamster V79 cells under the experimental conditions employed in the study. The positive controls did induce the appropriate response. **There was no evidence of induction of mutant colonies over background.**

This study is classified as **Acceptable/Guideline** and satisfies the guideline requirement for Test Guideline OPPTS 870.5300, OECD 476 for *in vitro* mutagenicity (mammalian forward gene mutation) data. No data on the pH and osmolality of the treatment medium was presented.

46607202 Meyer, A.L.(1987) Genotoxicity studies with WL115110: *In vitro* chromosome studies with WL115110. Shell Research Limited, London. Sittingbourne Research Centre, Sittingbourne, Kent, ME9 8AG, England. Report No. SBGR.86.216, April 15, 1987. Unpublished.

EXECUTIVE SUMMARY: In a mammalian cell cytogenetics chromosome aberration assay (MRID 46607202), CHO cell cultures were exposed to flufenoxuron (96.6 % a.i., Batch No. 6001, Drum 2, dissolved in DMSO) at concentrations of 15, 75 and 150 µg/mL in the presence, and absence of S9 microsomes from rat liver for 3 hours.

Flufenoxuron was tested at concentrations of 15, 75 and 150 µg/mL, with the high concentration resulting in ~50% cell survival. Flufenoxuron did not increase chromosome aberrations in CHO cells in the absence of S9 activation, but in the presence of S9 the frequency of aberrations in the treated cells at all concentration levels ranged from 3.3 to 7.1 times the frequency of aberrations seen in the solvent control cells when gaps were not included. When gaps were included, the frequency of aberrations in the treated cells at all concentration levels ranged from 2.4 to 4.1 times the frequency of aberrations seen in the solvent control cells. Most of the aberrations seen were chromatid gaps, chromatid breaks and exchange figures. The solvent and positive control values were appropriate. **There was evidence that flufenoxuron increased chromosome**

aberrations in CHO cells over background in the presence of S9 mix.

This study is classified as **Acceptable/Guideline** and satisfies the guideline requirement for Test Guideline *In vitro* mammalian cytogenetics in Chinese hamster ovary cells OPPTS 870.5375, OECD 473.

46607204 Meyer, A.L., and D.E. Wiggins (1988) Genotoxicity Studies With WL115110: *In vitro* chromosome studies with WL115110 using rat live (RL4) cell line. Sittingbourne Research Centre, Sittingbourne, Kent, England ME9 8AG. Sponsor's Study No.: SBGR.87.118, January 29, 1988. Unpublished.

EXECUTIVE SUMMARY: In a mammalian cell cytogenetics assay (Chromosome aberration) (MRID 46607204), rat liver epithelioid cell cultures were exposed for three hours to WL115110 (Lot ST86/176, batch 6001, drum 2, 96.6% a.i.) in dimethyl sulfoxide (DMSO) at concentrations of 0, 45, 225, or 450 µg/mL without metabolic activation (S-9 mix) and at concentrations of 0, 16, 80, or 160 µg/mL with metabolic activation (S-9 mix). Cells were harvested 5, 9, and 21 hours following termination of treatment both with and without S-9 mix. The S9-fraction was obtained from Aroclor-1254 induced rat liver.

WL115110 was tested up to concentrations limited by cytotoxicity. The total cell count was reduced to 45.9% of the solvent control value at a concentration of the test chemical of 450 µg/mL in the absence of S-9 mix, and it was reduced to 61.7% of the solvent control value at a concentration of the test chemical of 160 µg/mL in the presence of S-9 mix. Because significant levels of toxicity were observed at these two concentration levels, they were chosen as the maximum test concentrations. There were no statistically significant increases in the percentages of cells with structural aberrations, including or excluding gaps, over the solvent control values at any test material concentration with or without S9-mix. There was also no biologically significant increase in polyploidy. The solvent and positive control values were appropriate, except for the earliest harvest time in the absence of S-9 mix, for which the positive control induced no chromosomal aberrations. That short harvest time clearly does not provide a valid test. Nothing was said about the laboratory's historical control range. **There was no evidence of chromosome aberrations induced over background.**

This study is classified as **Unacceptable/Guideline** and does not satisfies the guideline requirement for Test Guideline *in vitro* mammalian cytogenetics (chromosomal aberrations) OPPTS 870.5375; OECD 473. The test is unacceptable because the negative result should have prompted a follow-up experiment with a much longer treatment time and without S-9 mix. The experiment, as it was conducted, may not have provided a test that was stringent enough to yield a firm conclusion.

46607205 McEnaney, S. (1992) Flufenoxuron: Study to evaluate the chromosome damaging potential of WL115110 by its effects on cultures human lymphocytes using an *in vitro* cytogenetics assay. Hazleton UK, Otley Road, Harrogate, North Yorkshire, England HG3 1PY and Shell Research Limited, Sittingbourne Research Centre,

Sittingbourne, Kent, England ME9 8AG. Sponsor's Study No.: IAU-5567, September 23, 1992. Unpublished.

EXECUTIVE SUMMARY: In a mammalian cell cytogenetics assay (Chromosome aberration) (MRID 46607205), human lymphocyte (whole blood) cultures were exposed to WL115110 (Lot SNPE8/91-1, 98.1% a.i.) in dimethyl sulfoxide (DMSO) at concentrations of 0, 78.4, 112, or 160 µg/mL for three hours with and without metabolic activation (S-9 mix). Cells were harvested 17 hours following termination of treatment both with and without S-9 mix. Cells were also harvested 41 hours following termination of treatment in another experiment at the highest concentration with S-9 mix. An additional assay was conducted by exposing cells to test material concentrations of 0, 78.4, 112, or 160 µg/mL without S9-mix for 20 hours with immediate post-treatment harvest. Yet another assay was conducted by exposing cells to test material concentrations of 0 or 160 µg/mL without S9-mix for 44 hours with immediate post-treatment harvest. Blood was obtained from a healthy male donor. The S9-fraction was obtained from Aroclor 1254 induced male Sprague-Dawley rat liver.

WL115110 was tested up to concentrations limited by solubility. The maximum mitotic inhibition detected was a decrease of 42% in the mitotic index following the 20-hour treatment at the highest concentration in the absence of S-9 mix. Precipitate was seen in the culture media at all concentrations tested. There were no statistically significant increases in the percentage of cells with structural aberrations including or excluding gaps or in polyploidy over the solvent control values at any test material concentration with or without S9-mix. The solvent and positive control values were appropriate, and solvent control values were within the testing laboratory's historical control ranges in all assays. **There was no evidence of chromosome aberrations induced over background.**

This study is classified as **Acceptable/Guideline** and satisfies the guideline requirement for Test Guideline *in vitro* mammalian cytogenetics (chromosomal aberrations) OPPTS 870.5375; OECD 473.

46607206 Allen, J.A., R.J. Proudlock and P.C. Brooker (1986) Genotoxicity studies with WL115110: *In vivo* chromosome studies with rat bone marrow cells. Huntingdon Research Centre Ltd., Huntingdon, Cambridgeshire, PE18 6ES., England. Report No.: SSL 86/8621, April 23, 1986. Unpublished.

EXECUTIVE SUMMARY: In a CD rat bone marrow chromosomal aberration assay (MRID 46607206), five rats/sex/harvest time were treated once via gastric intubation with WL 115110 (99.0 ± 2% a.i., Batch No. 10B) in corn oil at doses of 0 or 4000 mg/kg body weight. Bone marrow cells were harvested at 6, 24 and 48 hours following treatment. The positive control group was sacrificed 24 hours after i.p. injection of cyclophosphamide.

WL 115110 was tested at an acceptable dose as determined in a preliminary toxicity test in male and female rats at concentrations up to 4000 mg/kg. No mortality was seen during the in-life part of the cytogenetic study. Slight diarrhoea starting about two hours after treatment and lasting about four hours occurred in both sexes following treatment with either the vehicle or WL

115110. Slight piloerection, hunched posture and ptosis was seen in both sexes starting about four to six hours following exposure to WL 115110 and lasting until sacrifice. There was no statistically or biologically significant increase in the frequency of cells with structural aberrations, excluding gaps, over the concurrent solvent control value at any harvest time. Numerical aberrations were not reported. The solvent and positive controls induced the appropriate responses. **There was no indication that WL 115110 induced chromosomal aberrations in rat bone marrow cells as tested in this study.**

This study is classified as **Acceptable/Guideline** and satisfies the guideline requirement for Test Guideline OPPTS 870.5385; OECD 475 for *in vivo* cytogenetic mutagenicity data.

46607207 Cifone, M.A. (1988) In the *In vivo/in vitro* rat primary hepatocyte unscheduled DNA synthesis assay. Hazleton Laboratories America, Inc., 5516 Nicholson Lane, Suite 400, Kensington, Maryland 20895. Laboratory Project ID.: HLA Study No. 10342-0-494, November 22, 1988. Unpublished.

EXECUTIVE SUMMARY: In an *in vivo/in vitro* unscheduled DNA synthesis (UDS) assay (MRID 46607207) in male Fischer 344 rat hepatocytes, WL 115110 (97.4%, a.i.; batch no. 7016) at doses of 0, 188, 375, 750 or 1500 mg/kg body weight, was administered to three male rats per test group by oral gavage. The test material was delivered in corn oil. Hepatocytes were isolated at four hours post-treatment from test material, positive control, and vehicle control treated rats and cultured for about 18 hours in the presence of tritiated thymidine. Grain count was determined using the autoradiographic procedure.

The upper dose of 1500 mg/kg was selected based on the reported oral LD_{50} in rats of >3000 mg/kg and on the ability to prepare a useable suspension of the test material in corn oil. No toxicity data were presented. The investigators did not report any clinical signs in the rats after dosing and the test material did not appear to be cytotoxic to the hepatocytes. The volume of test material suspension administered to the rats did not exceed 5.0 mL/kg. A larger volume is well tolerated in rats with 10 mL/kg commonly used (see Allen, J.A., R.J. Proudlock and P.C. Brooker (1986) Genotoxicity studies with WL115110: *In vivo* chromosome studies with rat bone marrow cells. Huntingdon Research Centre Ltd., Huntingdon, Cambridgeshire, PE18 6ES. Report No.: SSL 86/8621, April 23, 1986. MRID 46607206. Unpublished). The upper dose tested was thus not justified on the basis of toxicity/cytotoxicity or on the inability to administer a larger volume to the rats. No statistical analysis was performed; however, little difference was seen between the net nuclear grain counts in hepatocytes from WL 115110 treated rats and those from the solvent control group. The mean net nuclear grain counts were below zero in all test material treated groups and none of the testing laboratory's criteria for a positive response were met. The solvent and positive control values were appropriate. **There was no evidence that WL 115110 increased the incidence of UDS over the solvent control value indicating no induction of DNA damage.**

This study is classified as an **Unacceptable/Guideline** study. It does not satisfy the guideline requirement in OPPTS 870.5550 [§84-2] because the upper dose tested was not justified by toxicity/cytotoxicity or solubility limitations. However, if the submitter can provide data

justifying the level selected for the high dose, then the study may be upgraded to acceptable.

46607208 Nishitomi, T. (Study Director) (1993) Micronucleus test on WL 115110 in mice. Mitsubishi-Kasei Institute of Toxicological and Environmental Sciences, Minato-ku, Tokyo, Japan. Study No.: 2L283, July 20, 1993. Unpublished.

EXECUTIVE SUMMARY: In an ICR mouse bone marrow micronucleus assay (MRID 46607208), six male mice/dose were treated twice via i.p. injection (24 hours between doses) with WL 115110 (98.2% a.i., w/w, Lot # 16/91-1) in corn oil at doses of 0, 500, 1000 or 2000 mg/kg body weight. Bone marrow cells were harvested at 24 hours following the second treatment and scored for the incidence of micronucleated polychromatic erythrocytes (PCE) and for the PCE/total erythrocyte ratio.

WL 115110 was tested to the limit dose for the assay. No mortality or clinical signs were seen in either the preliminary toxicity test or in the micronucleus assay. There was no biologically or statistically significant increase in the incidence of micronucleated PCEs or in the PCE/total erythrocyte ratio compared with the solvent control value at any test material dose. The vehicle and cyclophosphamide positive control values were appropriate. **There was no statistically significant increase in the frequency of micronucleated polychromatic erythrocytes in mouse bone marrow seen in this study.**

This study is classified as **Acceptable/Guideline**. It satisfies Test Guideline OPPTS 870.5395; OECD 474 for *in vivo* cytogenetic mutagenicity data.

46607209 Miyagawa, M. (Study Director) (1992) Replicative DNA syntheses (RDS) rat livers on WL 115110. Yokohama Laboratory, 1000 Kamoshida-cho, Midori-ku, Kanagawa, Japan. Laboratory Study No.: 2B300, September 4, 1992. Unpublished.

EXECUTIVE SUMMARY: In an *in vivo* Replicative DNA synthesis (RDS) assay (MRID 46607209) in male Fischer 344 rat hepatocytes, WL 115110 (98.2%, a.i., Lot # 16/91-1) at doses of 0, 2000 or 4000 mg/kg body weight, was administered to four male rats per test group by oral gavage in the volume of 10 mL /kg. The test material was delivered in corn oil. Hepatocytes were isolated at 24, 39 and 48 hours post-treatment and cultured for four hours in the presence of (methyl-^{3}H)-thymidine. The incidence of RDS was determined in 2000 cells per rat using the autoradiographic procedure.

The upper dose of 4000 mg/kg was determined to be the MTD and was selected based on the results of a previous acute study in rats showing an LD_{50} >5000 mg/kg and considering the technical limit dose. No mortality or clinical signs were seen during the study. No increase in the percentage of cells undergoing RDS was seen at either dose at any harvest time when compared with the value from the untreated control group. No vehicle or positive controls were used. **There was no evidence that WL 115110 increased the incidence of RDS over the untreated control value.**

This is not a guideline study and is classified as **Unacceptable/non-guideline** because no positive control was used. At best it would be considered supplementary with respect to genotoxicity. There is no OPPTS guideline requirement for this assay.

44448422 Huckle, K.R., Hutson, D.H., Mayo, B.C., Richardson, K.A., Baldwin, M.K., Kirkpatrick, D. (1992). Flufenoxuron (WL115110): general metabolism in the rat. Huntingdon Research Centre, Ltd., P.O. Box 2, Huntingdon, Cambridgeshire PE18 6ES, England and Shell Research Limited, Sittingbourne Research Centre, Sittingbourne, Kent, ME9 8AG, England. September 29, 1992. Report No. SBGR.87.186, SBGR.86.104, HRC/SLL 193/911344, SBGR.88.032, SBGR.88.104, SBGR.88.179, HRC/SLL 249/921154. Unpublished.

EXECUTIVE SUMMARY: A metabolism study (MRID 44448422) was conducted in which groups of male and female Fischer-344 rats (2-9/group depending on the specific experiment) were given a single 3.5 mg/kg or 350 mg/kg oral dose of [^{14}C]-WL115110 (flufenoxuron) (aniline ring label: lot no. 2712-038, >99.4% radiochemical purity; benzoyl ring label: lot no. 005/90, 99.6% radiochemical purity). Additional studies (SBGR.88.032, SBGR.88.179, and HRC/SLL 249/921154 included as appendices to MRID 44443822) examined tissue residues following 28-day and 100-day dietary exposures, and assessed biliary excretion of a 3.5 mg/kg dose of [aniline-^{14}C]WL115110.

This study describes the metabolism and disposition of WL115110 (flufenoxuron) in male and female rats. There were no overt signs of test article-induced toxicity in treated rats. Mass balance was an acceptable 93-104% depending on the specific experiment. Results of the experiments clearly demonstrated dose-dependent absorption. Saturated absorption was observed at the high dose (350 mg/kg) as shown by tissue/carcass burdens, recovery of radioactivity in the excreta, and AUC values. Elimination via the urine (9-27% of the low dose) was the major route of excretion for [^{14}C-2,6-difluorobenzene]WL115110. Conversely, fecal elimination was notably greater for the high dose (93-102% for high dose vs 4-19% for low dose). Elimination via expired air was insignificant. The excretory pattern for the [^{14}C]- aniline ring label varied somewhat from that of the difluorobenzene ring label in that the feces represented the major excretory route (21-24% of dose) and urinary excretion was secondary (5% of dose). Results of biliary excretion experiments (HRC/SLL 249/921154 cited as an appendix to MRID 44443844) using a single 3.5 mg/kg dose of the aniline ring label showed that nearly all of the radioactivity in the feces of female rats and approximately 40% of that in male rats was the result of biliary excretion products. Although the majority of both urinary and fecal elimination of radioactivity occurred within 48 hours, excretion by both routes was biphasic, with a slower phase occurring throughout the post-exposure observation period. An accumulation in adipose tissue and possible enterohepatic circulation were likely explanations. At four hours following a 3.5 mg/kg dose of the difluorobenzene label, muscle and adipose tissue contained up to 30% and 42% of the administered radioactivity, respectively. At 168 hours post-dose these values were approximately 6% and 19%, respectively, indicating an accumulation in adipose tissue. High-doses of either labeled form exhibited saturated absorption and negligible tissue burdens (<0.3%).

Experiments using two label positions (aniline and 2,6-difluorobenzene ring) allowed for thorough determination of the metabolic fate of flufenoxuron. For the aniline ring, 10 urinary

metabolites and parent compound represented in total ~5% of the dose and were quantitatively insignificant. Fecal excretion of metabolites was quantitatively greater with parent compound accounting for the greatest proportion of radioactivity. Most fecal metabolites represented <1% of the administered dose. Both [4-(2-chloro, α, α, α-trifluoro-p-tolyoxy)-2-fluorophenyl urea] and [4-(2-chloro, α, α, α-trifluoro-p-tolyoxy)-2-fluoroaniline] were detected in the feces and urine following administration of the aniline ring labeled test article. Unextractable residues accounted for 7-8% of the dose which is generally considered acceptable for this type of study. The major urinary metabolite of [^{14}C- 2,6-difluorobenzene]Flufenoxuron was the corresponding benzoic acid metabolite which, over 48 hours, accounted for 10-12% of the administered dose. Difluorobenzamide (<1%) was also detected in the urine along with unknown components all of which individually represented <1% of the dose. The only component detected in the feces of rats given the 2,6-difluorobenzene label was parent compound. The results of the metabolism characterization studies with both label positions are consistent with the investigators' contention that metabolism of flufenoxuron proceeds via hydrolysis to a benzoic acid metabolite, a phenyl urea metabolite (4-[2-chloro, α, α, α-trifluoro-p-tolyoxy]-2-fluorophenyl urea), an aniline metabolite (4-[2-chloro, α, α, α-trifluoro-p-tolyoxy]-2-fluoroaniline), and subsequently several minor components.

This study (MRID 44448422) on the metabolism and disposition of WL115110 (flufenoxuron) is classified **Acceptable/Guideline** and satisfies the 85-1 Guideline Requirement for a metabolism study [OPPTS 870.7485, OECD 417]. The main study along with earlier studies included as appendix material have provided a thorough assessment of the metabolism of both [^{14}C-2,6-dibenzofuran]WL115110 and [^{14}C-aniline]WL115110 by male and female rats.

3.0 METABOLISM CONSIDERATIONS

1.01 Team Proposal

For purposes of this petition, the residue of concern for tolerance and risk assessment purposes in plant and ruminant commodities is flufenoxuron *per se*. This conclusion applies only to the use of flufenoxuron on fruits and fruiting vegetables. Any future uses on other crops, such as leafy vegetables, legumes, or cereal grains, may require the submission of additional metabolism data, and any new uses having a substantial impact on livestock should include a new ruminant, and, potentially, a poultry metabolism study.

2.0 Nature of the Residue Studies in Plants

The petitioner has submitted three acceptable plant metabolism studies delineating the nature of flufenoxuron residues in tomato, apple, and grapes following one or two foliar applications. The nature of the flufenoxuron residues in fruits is understood based on these studies. These studies indicate that the metabolism of flufenoxuron in/on fruits and fruiting vegetables is minimal and that translocation of flufenoxuron from leaves to fruits is limited. Residues are largely confined to the fruit surface and are composed almost entirely of parent, even at longer post-treatment intervals. Flufenoxuron accounted for 91% of the TRR in tomatoes and apples at 28 and 99 DAT, respectively, and 50-55% of the TRR in grapes by 28 DAT.

2.1.1 Executive Summary of Tomato Study

[^{14}C]Flufenoxuron, uniformly labeled in the aniline ring [An-^{14}C], was applied as a single broadcast foliar application to tomato plants during fruit development at a rate equivalent to 0.125 kg ai/ha (0.11 lb ai/A). Tomato fruit were harvested at 0 and 28 DAT; foliage samples were not collected.

TRR in/on fruit declined from 0.38 ppm at 0 DAT to 0.17-0.20 ppm by 28 DAT. For analysis, fruit samples were surface washed with ACN:water (7:3, v/v), and the washed fruit were then extracted with ACN:water. Solubilized ^{14}C-residues were concentrated, diluted with water, and partitioned with dichloromethane (DCM), and the organic phase was analyzed by thin-layer chromatography (TLC).

Surface washes with ACN:water recovered >92% of the TRR from 0- and 28-DAT fruit samples, and solvent extraction of washed fruits released an additional 0.5-0.9% TRR. Nonextractable residues accounted for 1.1-5.8% TRR in all samples.

Flufenoxuron (parent) was the only compound identified in tomato fruits, accounting for ~88% of the TRR from the 28-DAT samples. No metabolites were identified. The petitioner provided a TLC figure depicting the peak corresponding to parent and stated that parent accounted for at least 98% of the radioactivity. Therefore, the reported 88% of the TRR and 0.176 ppm are estimates that were not quantified precisely.

2.1.2 Tabular Summary of Tomato Study

Summary of Characterization and Identification of ^{14}C-Residues in Tomato Fruit Following a Single Foliar Application of [An-^{14}C]Flufenoxuron at 0.125 kg ai/ha (0.111 lb ai/A).

Compound [1]	**Tomatoes (28 DAT) Sample II**	
	TRR = 0.20 ppm	
	% TRR	**ppm**
Flufenoxuron (parent)	91.0 [2]	0.182 [2]
Total identified [2]	**91.0**	**0.182**
Total characterized	91.0	0.182
Total extractable	**94.2**	**0.188**
Unextractable (PES) [2]	5.8	0.012
Accountability [3]	100	

1 Parent was identified by TLC and confirmed by MS. Structures and codes are found in Table B.2.1.
2 The petitioner provided a TLC figure depicting the peak corresponding to parent and stated that parent accounted for at least 99% of the TLC radioactivity. Therefore, the reported 91% of the TRR and 0.182 ppm are estimates that were not quantified precisely.
3 Post-extraction solids (PES); residues remaining after exhaustive extractions.
4 Accountability = (Total extractable + Total unextractable)/(TRR from TABLE C.2.1) * 100.

2.1.3 Executive Summary of Apple Study

[An-^{14}C]Flufenoxuron was applied as a single broadcast foliar application to apple trees during fruit development at a rate equivalent 97.4 mg ai/L, which was calculated to be 9.266 mg ai/tree. The trees were sprayed to "run off" with the diluted test substance. Immature apple fruits were harvested at 0 and 46 DAT and mature fruits were harvested at 99 DAT.

TRR in/on fruit declined from 15.1 ppm at 0 DAT to 10.0 ppm at 99 DAT. Fruit samples were surface washed sequentially with ACN and hexane, and then extracted with ACN:water (7:3, v/v). Sample extracts were combined, filtered and concentrated. Surface wash and fruit subsamples extracts were diluted with water and partitioned with DCM, and the organic phase was concentrated and analyzed by TLC. For metabolite quantitation, surface wash and fruit subsamples from the 0-DAT and 99-DAT intervals were analyzed by TLC and/or HPLC.

Initial ACN and hexane surface washes and ACN:water extractions solubilized >97% of the TRR from 0- and 99-DAT fruit samples. The majority of radioactivity was in the surface wash from the 0-DAT samples (96% TRR) and 99-DAT samples (77% TRR). An additional 3.7% TRR (0 DAT) and 23% TRR (99 DAT) was extracted from the washed fruit. Nonextractable residues accounted for 0.3-2.5% TRR.

Flufenoxuron (parent) was the only compound detected in apple fruit at both sampling intervals, accounting for 91-97% of the TRR. No metabolites were identified, indicating that the metabolism of flufenoxuron in apples was minimal.

2.1.4 Tabular Summary of Apple Study

Summary of Characterization and Identification of ^{14}C-Residues in Apple Fruit Following a Single Foliar Application of [An-^{14}C]Flufenoxuron at 9.266 mg ai/tree (0.67 lb ai/A).

Compound [1]	Apples (0 DAT)		Apples (99 DAT)	
	TRR = 15.069 ppm		TRR = 10.001 ppm	
	% TRR	ppm	% TRR	ppm
WL115110 (parent)	96.5	14.545	90.9	9.083
Total identified	**96.5**	**14.545**	**90.9**	**9.083**
Total characterized	96.5	14.545	90.9	9.083
Total extractable	**97.6**	**14.707**	**97.0**	**9.701**
Unextractable (PES) [2]	0.3	0.045	2.5	0.250
Accountability [3]	97.9		99.5	

[1] Metabolites were identified by TLC and/or HPLC.
[2] Post-extraction solids (PES); residues remaining after exhaustive extractions.
[3] Accountability = (Total extractable + Total unextractable)/(TRR from combustion analysis; see TABLE C.2.1) * 100.

2.1.5. Executive Summary of Grape Study

[^{14}C]Flufenoxuron, uniformly labeled in either the aniline [An-^{14}C] or in the benzamide [Bz-^{14}C] ring, was applied to grape vines as two broadcast foliar applications during fruit development at 0.04 kg ai/ha (0.036 lb ai/A), for a total of 0.08 kg ai/ha (0.072 lb ai/A; 0.4x the maximum

proposes seasonal application rate for grapes). The first application was made at BBCH stage ES 59, and the second was applied 40 days later. Samples of grape leaves were harvested 15 and ~28 DAT, and grape clusters were harvested ~28 DAT, and separated into stalks and fruit.

TRR levels for the various matrices were similar for the two ^{14}C-labels. ^{14}C-Residues in/on leaves declined from 2.29-2.67 ppm at 15 DAT to 1.42-1.82 ppm at ~28 DAT. At maturity (28 DAT), TRR were 0.012-0.014 ppm in grapes and 0.106-0.163 ppm in stalks. Solvent extraction with methanol (MeOH) and water released 95-97% of the TRR from all grape matrices, and residual solids accounted for ≤4.9% of the TRR. Solubilized ^{14}C-residues were characterized and identified by HPLC.

For both ^{14}C-labels, flufenoxuron (parent) was identified as the major component in leaves (86.2-96.9% TRR), stalks (94.5-96.3% TRR), and fruit (49.7-54.6% TRR). Analyses of purified solvent fractions also indicated that levels of parent may be higher in fruit then indicated by the initial analysis of the MeOH fraction. Two to three polar unknowns were also detected in fruit, each at 9.9-28.8% of the TRR; however, each of these unknowns was present at ≤0.004 ppm. Based on the above data, the metabolism of flufenoxuron in grapes was minimal. In addition, transport of flufenoxuron from leaves to fruit appears to be limited given the low levels of radioactivity in fruit relative to foliage.

2.1.6. Tabular Summary of Grape Study

Distribution of the ^{14}C-Residues in Grape Matrices Following Two Foliar Applications of [An-^{14}C]Flufenoxuron Totaling 0.08 kg ai/ha (~0.071 lb ai/A).

Metabolite Fraction [1]	Leaves (15 DAT)		Leaves (28 DAT)		Grapes (28 DAT)		Stalks (28 DAT)	
	TRR = 2.285 ppm		TRR = 1.424 ppm		TRR = 0.012 ppm		TRR = 0.106 ppm	
	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm
MeOH Extracts (HPLC analyses)	94.2	2.153	95.0	1.353	94.3	0.012	94.5	0.100
Parent	94.2	2.153	95.0	1.353	54.6	0.007	94.5	0.100
Polar Unknowns	ND	--	ND	--	39.7 [2]	0.005	ND	--
Hexane	23.2 [3]	0.531	53.8 [3]	0.766	94.3 [3]	0.012	89.4 [3]	0.095
EtOAc	75.6 [3]	1.727	46.4 [3]	0.660	4.6	<0.001	7.8	0.008
Aqueous	3.3 [3]	0.075	5.3 [3]	0.076	3.6	<0.001	1.3	0.001
Aqueous Extract	1.0	0.023	0.8	0.012	0.8	<0.001	0.8	0.001
Residual Solids	4.7	0.108	4.2	0.059	4.9	<0.001	4.7	0.005

[1] ^{14}C-Residues were identified and quantified by HPLC.
[2] Two polar metabolites were detected at 10.9 and 28.8% if the TRR. These fractions were not further analyzed as each metabolite was present at ≤0.004 ppm.
[3] Indicated fractions were analyzed by HPLC, and only parent was detected in all organic fractions and aqueous fractions.
ND = not detected

3.0 Nature of the Residue in Livestock

Poultry or swine metabolism studies were not submitted with the current petition as there are no poultry or swine feed items associate with the crops for which uses are being proposed. However, processed fractions associated apple and oranges are utilized as feed items for ruminants;

therefore, a goat metabolism study was submitted with the current petition and is discussed below.

Although numerous deficiencies were noted, the available goat metabolism study is adequate for delineating the nature of the residues in ruminants for purposes of this petition, and a poultry metabolism study is not required as there are no poultry feed items associated with the proposed crop uses. The goat study indicates that there is the potential for bioaccumulation of flufenoxuron in ruminant tissues and that the metabolism of flufenoxuron in ruminants appears to be minimal with parent being excreted primarily in the feces and to a lesser extent in the milk. Following 4 days of dosing with [^{14}C]flufenoxuron at levels equivalent to ~2.8 ppm in the diet, TRR levels were highest in fat (1.59 ppm) and were considerably lower in liver (0.373 ppm), kidney (0.130 ppm), and muscle (0.076-0.103 ppm). Residues in milk increased over time to 0.372 ppm by Day 4 (p.m.) and were primarily associated with the cream fraction (82-93% TRR). HPLC analyses of purified ACN fractions from milk and tissues (76-115% TRR) detected only parent compound.

3.1.1 Executive Summary of Ruminant Study

In a ruminant metabolism study, a single dairy goat was dosed orally once a day for 4 consecutive days via gavage with [U-^{14}C-aniline]flufenoxuron at 10.23 mg/day. Based on an estimated feed consumption of 3.65 kg dry wt/day, this dosing level is equivalent to ~2.8 ppm in the feed. Urine and feces were collected daily and milk was collected twice daily during the dosing period. The goat was sacrificed within 24 hours of the final dose, and samples of liver, kidney, muscle (leg and dorsal), fat (renal and omental), bile, blood, and intestinal contents were collected. All samples were stored frozen (-20°C) prior to analysis. Based on the experimental start and completion dates, samples were potentially stored frozen for up to 11 months. Data from reanalysis of fat samples indicate that ^{14}C-residues were stable for at least 3-4 months of frozen storage. No additional storage stability data are required as only residues of parent were detected in milk and tissue samples.

Excretion of radioactivity was limited and occurred primarily via the feces (18.3% dose) and to a lesser extent in the milk (8.3% dose). A substantial portion of the dose was estimated to have remained in fatty tissues (43-57% dose) at sacrifice, based on concentrations of radioactivity in fat samples. Radioactivity in liver and kidney accounted for 1.15 and 0.06% of the dose, respectively.

TRR in milk showed a general increase from 0.268 ppm on Day 1 (pm) to 0.372 ppm by Day 4 (pm), with the cream fraction (1.27-4.68 ppm) accounting for 82-93% of the TRR in milk. Concentrations of radioactivity in tissues were also highest in fat (1.59 ppm) and were considerably lower in liver (0.373 ppm), kidney (0.130 ppm), and muscle (0.076-0.103 ppm).

Solvent extraction released 91.4-99.8% of the TRR from milk, fat, liver, and kidney, and 66.2% of the TRR from muscle. ^{14}C-Residues were purified by solvent partitioning and the final ACN fraction from each matrix (75.8-114.8% TRR) was analyzed by HPLC. Although no quantitative data were provided, the HPLC analyses detected only a single peak of radioactivity that co-chromatographed with flufenoxuron in milk and all tissues, suggesting that parent accounted for approximately 76-100% of the TRR in milk and tissues. No secondary method was used to confirm the identity of parent. However, 1D TLC analyses of fecal extracts (87-89% TRR) from

Study Days 1 and 4 using two solvent systems also detected only ^{14}C-parent.

The available data suggest that there is the potential for bioaccumulation of flufenoxuron in ruminants and that the metabolism of flufenoxuron in ruminants appears to be minimal with parent being excreted primarily in the feces and to a lesser extent in the milk.

3.1.2 Tabular Summary of Ruminant Study

Distribution of ^{14}C-Residues in Tissues and Milk from a Goat Dosed for 4 Days with [^{14}C-An]-Flufenoxuron at 10.23 mg ai/day, equivalent to ~2.8 ppm in the diet.

Fraction/ Metabolite	Liver (TRR = 0.373 ppm)		Kidney (TRR = 0.130 ppm)		Muscle (TRR = 0.103 ppm)		Fat (TRR = 1.59 ppm)		Milk Day 1 (TRR = 0.268 ppm)	
	%TRR	ppm	% TRR	ppm	% TRR	ppm	%TRR	ppm	%TRR	ppm
Solvent extracts[1]	93.4	0.348	91.4	0.119	66.2	0.068	99.8	1.59	98.3	0.263
Hexane	3.5	0.013	14.7	0.019	6.5	0.007	1.5	0.02	0.0	--
ACN (HPLC)	82.7	0.308	82.1	0.106	75.8	0.078	114.8	1.83	81.1	0.217
Parent[2]	82.7	0.308	82.1	0.106	75.8	0.078	114.8	1.83	81.1	0.217
Residual solids (PES)	NR	--	NR	--	NR	--	NR	--	NR	--

[1] The various solvent extracts for each tissue and milk were combined, concentrated and partitioned between hexane and ACN. The ACN fractions were then analyzed by HPLC.

[2] No quantitative data were provided for the HPLC analyses, but chromatograms for all milk and tissue fractions showed only a single peak of radioactivity that co-chromatographed with parent.

NR = not reported.

4.0 Confined Rotational Crop Studies

As the proposed uses are for perennial crops grown outside the U.S., rotational crop studies are not required for this petition.

5.0 Tabular Summary of Analytical Methodology

Method Name	Applicable Commodities	Analytes	Extraction Solvent(s)	Clean-up Step(s)	Determinative Step	LOQ (ppm)	LOD (ppm)
SAMS 432-3	Plants	Flufenoxuron	Homogenization with DCM and sodium sulfate	Filtered and dried, redissolved in hexane and cleaned up on Bond-elut cartridge, then cleaned up using reversed-phase HPLC	Normal-phase HPLC with UV detection at 254 nm	0.05	0.01

Method Name	Applicable Commodities	Analytes	Extraction Solvent(s)	Clean-up Step(s)	Determinative Step	LOQ (ppm)	LOD (ppm)
SAMS 457-2	Cattle tissues	Flufenoxuron	Homogenization with hexane:ACN (1:4, v/v) and sodium sulfate, with partitioning into the ACN fraction	Normal and/or reverse phase HPLC. Normal-phase cleanup uses a NH_2 column with a mobile phase of hexane:ethanol:acetic acid (97:3:0.2, v/v/v), and reverse-phase cleanup uses a ODS2 column with a mobile phase of ACN:water:triethylamine (80:20:0.2, v/v/v)	HPLC with UV detection at 254 nm	fat - 0.3 all other tissue- 0.1	0.03
SAMS 486-1	Milk	Flufenoxuron	Homogenization with 5% aqueous potassium oxalate, the extraction with ethanol, diethyl ether, and hexane (5:8:4, v/v/v)	Normal-phase HPLC using a Spherisorb CN column with an isocratic mobile phase of hexane:ethanol:acetic acid (95:5:0.2, v/v/v)	HPLC using reverse-phase column (Spherisorb ODS2) and isocratic mobile phase of ACN:water (70:30, v/v) with UV detection at 254 nm	0.01	0.001

6.0 Tabular Summaries of Magnitude of Residue (MOR) Studies

6.1 Plants

The petitioner has submitted crop field trial data from various countries in Europe and South America to support the use of flufenoxuron on apples, pears, grapes (Europe only), and oranges grown for export to the U.S. The results of these field trials are summarized in the tables below.

Summary of Residue Data from Apple Field Trials Conducted in South America and Europe using Flufenoxuron (FlC).

Country	Formulation	Total Rate (kg ai/ha) *[lb ai/A]*	PHI (days)[1]	Residues (ppm)[2]						
				n	Min.	Max.	HAFT[3]	Median	Mean	Std. Dev.
Argentina	FlC	0.525 *[0.47]* {2.2x}	0	16	0.26	0.57	0.42	0.35	0.37	0.08
			30	16	0.11	0.55	0.33	0.32	0.31	0.12
			60	12	0.09	0.35	0.23	0.22	0.22	0.08
			90	16	0.05	0.18	0.13	0.11	0.12	0.03
France	FlC	0.3 *[0.27]* {3x}	21, 28	4	0.17	0.28	0.28	0.22	0.22	0.05
			42	4	0.06	0.21	0.21	0.12	0.13	0.06
		0.4 *[0.36]* {4x}	0	4	0.09	0.34	0.34	0.16	0.19	0.11
			7	4	0.08	0.26	0.26	0.13	0.15	0.08
			14	4	0.11	0.32	0.32	0.17	0.19	0.09
			21	4	0.05	0.58	0.58	0.12	0.22	0.25
		0.5-0.6 *[0.53]* {5x-6x}	21, 28	4	0.17	0.45	0.45	0.32	0.31	0.13
			42	4	0.01	0.24	0.24	0.10	0.11	0.11

Summary of Residue Data from Apple Field Trials Conducted in South America and Europe using Flufenoxuron (FlC).										
Country	Formulation	Total Rate (kg ai/ha) *[lb ai/A]*	PHI (days)[1]	Residues (ppm)[2]						
				n	Min.	Max.	HAFT[3]	Median	Mean	Std. Dev.
		0.8 *[0.71]* {8x}	0	4	0.07	0.56	0.56	0.24	0.28	0.22
			7	4	0.10	0.53	0.53	0.25	0.28	0.19
			14	4	0.02	0.46	0.46	0.16	0.20	0.19
			21	4	0.04	0.24	0.24	0.21	0.18	0.09
Chile	FlC	0.22-0.44 *[0.20-0.40]* {~2x-4x}	35	3	0.43	0.52	0.52	0.45	0.47	0.05
			77	3	<0.05	0.08	0.08	0.03	0.04	0.03
			113, 126	6	<0.05	<0.05	0.05	0.025	0.025	0
		0.18-0.36 *[0.16-0.32]* {1.6x-3x}	15	2	0.28	0.46	0.46	0.36	0.36	0.13
			36	2	0.07	0.17	0.17	0.12	0.12	0.07
			49	2	<0.05	0.09	0.09	0.06	0.06	0.05
			85	2	<0.05	0.12	0.12	0.07	0.07	0.07
			98	2	<0.05	0.05	0.05	0.04	0.04	0.02
		0.156-1.15 *[0.14-1.0]* {1.5x-10x4}	113-149	28	<0.05	<0.05	0.05	0.025	0.025	0
Brazil	FlC	~0.45[4] *[0.40]* {0.75x}	15	2	0.34	0.41	0.41	0.38	0.38	0.05
			30	1	0.40	0.40	0.40	0.40	0.40	0
		~0.90[4] *[0.80]* {1.5x}	15	2	0.40	0.61	0.61	0.51	0.51	0.15
			30	1	0.20	0.20	0.20	0.20	0.20	0

1 PHI = preharvest interval.
2 The limit of quantitation (LOQ) is 0.05 ppm; the limit of detection (LOD) was 0.01 ppm. For calculation of the median, mean, and standard deviation, ½LOQ was used for samples with residues <LOQ.
3 HAFT = Highest-Average Field Trial.
4 Actual rates in terms of kg ai/ha were not reported for the Brazilian trials, the listed rate is the maximum possible rate calculated by the reviewer.

Summary of Pear Residue Data.										
Commodity	**Formulation**	**Total Rate (kg ai/ha)**	**PHI (days)[1]**	**Residues (ppm)[2]**						
				n	**Min.**	**Max.**	**HAFT[3]**	**Median**	**Mean**	**Std. Dev.**
Pear fruit	100 g/L FlC	0.3 (3x)	28	2	0.10	0.16	0.16	0.13	0.13	0.04

1 PHI = preharvest interval..
2 The limit of quantitation (LOQ) is 0.05 ppm for flufenoxuron; the limit of detection (LOD) was 0.01 ppm. For calculation of the median, mean, and standard deviation, ½LOQ was used for samples with residues <LOQ.
3 HAFT = Highest-Average Field Trial.
NA = not applicable

Summary of Grape Residue Data.										
Commodity/ Country	**Formulation**	**Total Rate (kg ai/ha)[1]**	**PHI (days)[2]**	**Residues (ppm)[3]**						
				n	**Min.**	**Max.**	**HAFT[4]**	**Median**	**Mean**	**Std. Dev.**
Grapes/ France	100 or 50 g/L FlC	0.050-0.100 (0.4x-0.8x)[5]	21-61	18	<0.01	0.15	0.15	0.025	0.049	0.038
Grapes/ Spain		0.133-0.147 (0.9x-1x)[5]	30	2	0.20	0.37	NA	0.285	0.285	NA
Grapes/ France, Italy, Greece	100 or 50 g/L FlC	0.040-0.046 (0.3x, 0.3x, 0.2x)[6]	0	4	<0.05	0.28	0.28	0.075	0.144	0.115
			7	4	<0.05	0.19	0.19	0.038	0.073	0.079
			15	4	<0.05	0.09	0.09	0.028	0.043	0.032
			21	4	<0.02	0.06	0.06	0.025	0.030	0.021
			28	4	<0.02	0.06	0.06	0.025	0.030	0.021
Grapes/ France, Germany, Italy, Spain, Greece	100 or 50 g/L FlC	0.080-0.100 (0.7x, 0.5x, 0.4x)[6]	-0	5	<0.02	0.17	0.17	0.025	0.064	0.067
			0	10	0.03	0.50	0.50	0.105	0.186	0.165
			7	4	<0.05	0.36	0.36	0.055	0.124	0.159
			14-15	5	0.03	0.50	0.50	0.060	0.202	0.218
			21	10	<0.02	0.31	0.31	0.065	0.097	0.096
			27-29	10	<0.02	0.66	0.66	0.075	0.129	0.192
			35	6	0.02	0.53	0.53	0.080	0.142	0.192
Grapes/ Spain, Greece	100 g/L FlC	0.118-0.130 (1x, 0.8x, 0.6x)[6]	-0	3	0.11	0.25	0.25	0.250	0.203	0.081
			0	3	0.21	0.38	0.38	0.250	0.280	0.089
			14	3	0.15	0.38	0.38	0.310	0.280	0.118
			21	3	0.13	0.40	0.40	0.180	0.237	0.144
			28	3	0.10	0.31	0.31	0.150	0.187	0.110
			35	3	0.15	0.22	0.22	0.170	0.180	0.036

[1] The maximum total use rate for grapes is 0.15 kg ai/ha for Greece and Spain, 0.12 kg ai/ha for France, and 0.20 kg ai/ha for Italy.
[2] The proposed PHI for grapes in Europe is 28-30 days.
[3] The LOQ for flufenoxuron in/on grapes is 0.05 or 0.02 ppm for the HPLC/UV methods and 0.01 ppm for the LC/MS/MS method. For calculation of the median, mean, and standard deviation, ½LOQ was used for samples with residues <LOQ.
[4] HAFT = Highest Average Field Trial.
[5] Use rates in terms of maximum proposed rate for study where trial was conducted.
[6] The maximum use rates used in the field trials are also reported in terms of the maximum use rates proposed in France, Spain/Greece, and Italy, respectively.
NA = not applicable.

Summary of Residue Data from Orange Field Trials Conducted in Europe using Flufenoxuron (FlC).											
Commodity	Formulation	Location	Total Rate (kg ai/ha)[1]	PHI (days)[2]	Residues (ppm)[3]						
					n	Min.	Max.	HAFT[4]	Median	Mean	Std. Dev.
Whole Fruit	100 or 50 g/L FlC	Spain	0.08-0.138 (0.11x-0.18x)	31	4	0.05	0.08	0.08	0.07	0.075	0.013
			0.208 (0.28x)	31	2	0.11	0.16	0.16	0.135	0.135	0.035
Peel	100 or 50 g/L FlC	Spain	0.138 (0.18)	31	1	0.12	0.12	0.12	0.12	0.12	NA
			0.208 (0.28x)		1	0.33	0.33	0.33	0.33	0.33	NA
			0.508 (0.68x)	28	1	0.57	0.57	0.57	0.57	0.57	NA
		Italy	0.140 (0.19x)	28	1	0.16	0.16	0.16	0.16	0.16	NA
		Greece	0.179-0.250 (0.24x-0.33x)	28	4	0.23	0.73	0.73	0.315	0.40	0.22
Pulp	100 or 50 g/L FlC	Spain	0.138 (0.18)	31	1	<0.05	<0.05	<0.05	0.025	0.025	NA
			0.208 (0.28x)		1	<0.05	<0.05	<0.05	0.025	0.025	NA
			0.508 (0.68x)		1	<0.05	<0.05	<0.05	0.025	0.025	NA
		Italy	0.508 (0.68x)	28	1	<0.05	<0.05	<0.05	0.025	0.025	NA
		Greece	0.179-0.250 (0.24x-0.33x)	28	4	<0.05	<0.05	<0.05	0.025	0.025	NA

[1] The maximum proposed total use rate for oranges is 0.80 kg ai/ha for Greece, 0.30 kg ai/ha for Italy, and 0.75 kg ai/ha for Spain.
[2] The proposed PHI is 28 days for Greece and Spain and 75 days for Italy.
[3] The LOQ is 0.05 ppm for flufenoxuron; the LOD was 0.01 ppm. For calculation of the median, mean, and standard deviation, ½LOQ was used for samples with residues <LOQ.
[4] HAFT = Highest Average Field Trial.
NA = not applicable.

6.2 Livestock

An adequate cattle feeding study is available reflecting dosing of dairy cattle at levels of 1.75, 5.25, and 17.5 ppm in the diet for up to 90 days. Based on the calculated MTDB of 0.169 ppm for beef and dairy cattle, these dietary levels are equivalent to 10x, 31x and 100x the MTDB for beef and dairy cattle. In each dose group, flufenoxuron residues in milk increased steadily over time, plateauing between 56 and 70 days at averages of 0.59, 1.70, and 5.32 ppm for the three groups. Fractionation of a bulk milk sample indicated that flufenoxuron residues concentrate substantially (5.1x) in milk fat. The distribution of flufenoxuron residues among tissues was similar at each dose level, with residue levels being highest in fat and lowest in muscle. For the 1.75 ppm dose group (10x), maximum flufenoxuron residues were 6.95 ppm in fat, 0.83 ppm in liver, 0.44 ppm in kidney, and 0.28 ppm in muscle, and average residues were 2.30 ppm in fat, 0.75 ppm in liver, 0.34 ppm in kidney, and 0.14 ppm in muscle. Over the dose range tested, flufenoxuron residues in cattle increased linearly with dose level in milk and tissues (r^2=9833-0.9998), although the relationship was not linear for fat (r^2=0.8695). After cessation of dosing at 17.5 ppm (100x),

residues in milk declined slowly with an elimination half-life of 16 days, and substantial residues remained in fat (3.46-7.60 ppm) at 40 days post-treatment. The available data indicate that flufenoxuron residues transfer primarily to fatty tissues or fractions in cattle.

Summary of Milk and Tissue Residue Data from Cattle Feeding Study with Flufenoxuron.[1]

Matrix	Feeding Level (ppm)	Flufenoxuron Residues (ppm) [2]					
		n	Min.	Max.	Median	Mean	Std. Dev.
Whole Milk	1.75 (1x)	12	0.47	0.77	0.57	0.59	0.09
	5.25 (3x)	12	1.30	2.20	1.60	1.70	0.26
	17.5 (10x)	30	4.10	6.90	5.30	5.32	0.68
	17.5 (10x)[3]	3	0.61	1.30	0.84	0.92	0.35
Fat (Peritoneal)	1.75 (1x)	6	0.56	6.95	1.09	2.30	2.51
	5.25 (3x)	6	6.36	24.92	20.96	17.22	8.24
	17.5 (10x)	6	14.26	47.14	27.59	29.36	13.72
	17.5 (10x)[3]	6	2.16	12.05	8.46	7.60	4.35
Fat (Subcutaneous)	1.75 (1x)	6	0.12	2.93	0.39	0.86	1.08
	5.25 (3x)	6	3.59	15.27	10.20	9.45	5.13
	17.5 (10x)	6	3.07	18.20	7.54	8.57	5.71
	17.5 (10x)[3]	6	0.43	6.88	3.44	3.46	2.74
Kidney	1.75 (1x)	6	0.23	0.44	0.36	0.34	0.09
	5.25 (3x)	6	0.98	2.67	1.44	1.62	0.69
	17.5 (10x)	6	2.87	7.16	3.61	4.27	1.70
	17.5 (10x)[3]	6	0.14	1.50	0.66	0.70	0.54
Liver	1.75 (1x)	6	0.64	0.83	0.75	0.75	0.08
	5.25 (3x)	6	1.85	2.31	2.18	2.15	0.17
	17.5 (10x)	6	7.46	10.62	8.62	8.69	1.19
	17.5 (10x)[3]	6	0.23	1.64	1.02	0.93	0.58
Muscle	1.75 (1x)	6	0.08	0.28	0.10	0.14	0.08
	5.25 (3x)	6	0.28	1.19	0.58	0.65	0.35
	17.5 (10x)	6	1.28	1.94	1.67	1.62	0.26
	17.5 (10x)[3]	6	0.03	0.48	0.27	0.25	0.18

[1] Cattle were dosed for 90 days. Unless otherwise specified, tissue samples were collected within 24 hours of the final dose (Day 91). For calculation of average residues in milk, data from Days 56, 70, 86, and 90 were used, as these samples are representative of steady state kinetics.

[2] The validated method limit of quantitation (LOQ) for flufenoxuron is 0.01 ppm in milk, 0.3 ppm in fat, and 0.1 ppm in other tissues, and the reported limit of detection (LOD) is 0.001 ppm in milk and 0.03 ppm in tissues.

[3] Residues in milk and tissues following a 40-day depuration period.

7.0 International Considerations

There are currently no established or proposed Canadian, Mexican or Codex maximum residue limits (MRLs) for flufenoxuron. There are European Union (EU) MRLs for flufenoxuron for apples, pears, and grapes at 0.5 ppm; and for citrus at 0.3 ppm.

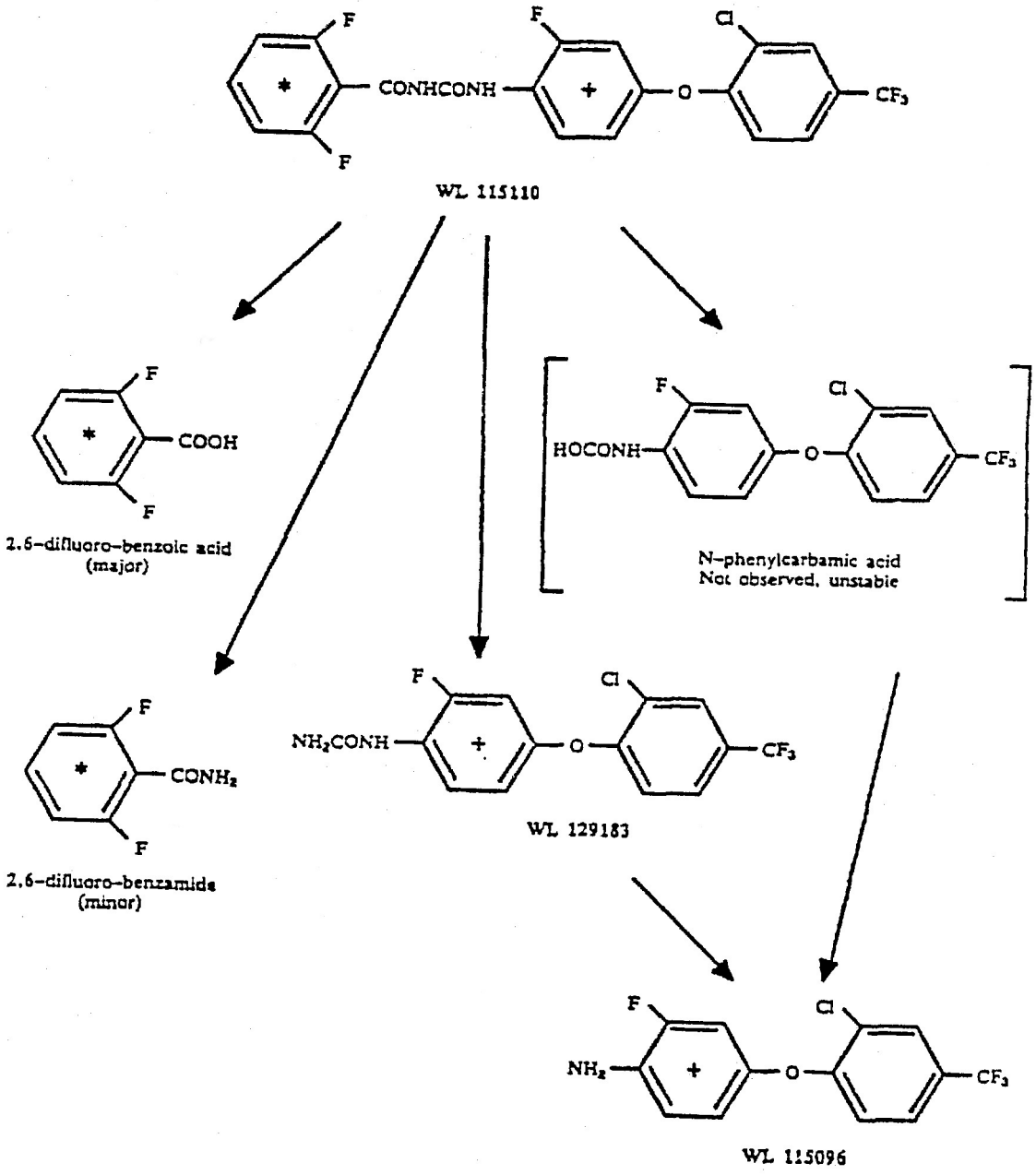



Figure 1. Proposed metabolic pathway for WL115110 (Flufenoxuron) in the rat.