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

OFFICE OF PREVENTION,
PESTICIDES AND TOXIC
SUBSTANCES

21 August 2006

MEMORANDUM

Subject: Name of Pesticide Product: PROMERIS SPOT-ON FOR DOGS
EPA Reg. No. /File Symbol: 80490-E
DP Barcode: D327883
Decision No.: 351841
PC Codes: 106201 (Amitraz); 281250 & 281251 (Metaflumizone)

From: Byron T. Backus Ph.D., Toxicologist
Technical Review Branch
Registration Division (7505P)

Byron T. Backus 8/21/2006
KJW 8/22/06

To: John Hebert, RM Team 07
Insecticide-Rodenticide Branch
Registration Division (7505P)

Registrant: FORT DODGE ANIMAL HEALTH

FORMULATION FROM LABEL:

Active Ingredient(s):	% by wt.
281250 & 281251 Metaflumizone	14.34%
106201 Amitraz	14.34%
Other Ingredient(s):	71.32%
Total:	100.00%

ACTION REQUESTED: The Risk Manager requests:

"Please conduct a secondary review of the pharmacokinetic study in dog. The primary review has been prepared by a contractor…

BACKGROUND:

TRB has previously reviewed a 6-pack of acute toxicity studies and two (adult dog and puppy) companion animal studies on this product. In the previous reviews of the companion animal safety studies, TRB expressed concerns regarding indications that there is a less-than-5X margin of safety between the use exposure level and that at which symptoms of toxicity occur.

In order to at least partially address the concerns, the registrant has had several additional dog studies conducted. One of these is the study in MRID 46672101 titled: "Pharmoacokinetics of R-28153 and amitraz after a single topical application to dogs at 20 mg/kg of R-28153 and amitraz." As TRB's resources are limited, and the study was somewhat beyond the usual type of studies reviewed by TRB, it was routed to HED for a primary (contract) review, but, at the request of HED, it was agreed that TRB would conduct the secondary review.

COMMENTS AND RECOMMENDATIONS:

EPA Reviewer's Comment: The maximum level of R-28153 observed in the plasma was 138.3 ng/mL (dog 150311 on day 42). One reference (www.cals.wisc.edu/research/rarc/pt5technique.PDF) gives the approximate blood volume in the dog as 86.2 mL/kg, with 50% of this plasma; 43.1 mL of plasma would then have contained 5.96 µg of R-28153, and, in terms of the original dose (22 mg/kg), this would be (5.96 /22000) = 0.00027 or 0.027%. As the rate of clearance (or half-life) of R-28153 from plasma is not known, no conclusions can be made regarding the total amount (or percentage of the original 22 mg/kg dose) that was actually absorbed by this one dog.

The following is the executive summary from the DER:

In a dermal pharmacokinetic study (MRID 46672101), R28153 (15% w/v) and amitraz (15% w/v) were applied once as a mixture to the intrascapular area of three male and three female adult beagle dogs. Each dog received a ~22 mg/kg dose of each test material. Blood was collected from each dog 5 and 10 hours after dosing and on study days 1, 2, 3, 5, 7, 10, 14, 21, 28, 42, and 56.

Plasma concentrations of amitraz were below the limit of quantification (50 ng/mL) throughout the 56-day study. It was detected in the plasma of two female dogs 24 and 48 hours post-treatment, respectively. Amitraz was below the limit of detection (3.2 ng/mL) at all other times,

R28153 was above the limit of quantification (50 ng/mL) in three male dogs and one female dog during the study. The test material was quantifiable in the plasma of one male on day 7 through the end of the study, in the second male from day 21 through day 42, and in the third male on days 28 and 42. R-28153 was quantifiable in one female dog's plasma on day 42 only. The maximum levels (range: 58.8 to 138.3 ng/mL) observed in these four dogs all occurred on day 42. Because the recovery of the test materials was mostly at or below the limit of quantification, pharmacological profiles of R-28153 and amitraz were not possible. The report includes the statement that: "The data do not allow a calculation of the pharmacokinetic profile of amitraz and R-28153 in dogs following application of the spot-on."

This study is considered Unacceptable/Nonguideline. It may be upgraded to acceptable if the dosing rationale were explained and if it is determined that an appropriate dose was used. The solvent used for the study should also be described.

DATA EVALUATION RECORD

METAFLUMIZONE and AMITRAZ
NONGUIDELINE
STUDY TYPE: DERMAL PENETRATION PHARMACOKINETICS
MRID 46672101

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1801 Bell Street
Arlington, VA 22202

Prepared by

Toxicology and Hazard Assessment Group
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. 125-2006

Primary Reviewer:
H. T. Borges, Ph.D., MT(ASCP), DABT

Signature: H T Borges
Date: MAR 1 : 2006

Secondary Reviewers:
R.A. Young, Ph.D., D.A.B.T.

Signature: Robert A. Young
Date: MAR 1 : 2006

Robert H. Ross, M.S., Group Leader

Signature: Robert H. Ross
Date: MAR 1 : 2006

Quality Assurance:
Eric B. Lewis, M.S

Signature: Eric B. Lewis
Date: MAR 1 : 2006

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory managed and operated by UT-Battelle, LLC., for the U.S. Department of Energy under Contract No. DE-AC05-00OR22725.

EPA Reviewer: Byron T. Backus, Ph.D.
Technical Review Branch, Registration Division (7505P)
EPA Secondary Reviewer: P.V. Shah, Ph.D.
Registration Action Branch 1, Health Effects Division (7509P)

Signature: Byron T. Backus
Date: 8/21/06
Signature: P.V. Shah
Date: 8/21/06

TXR#: 0053824

Template version 02/06

DATA EVALUATION RECORD

STUDY TYPE: Pharmacokinetics Following Dermal Application [Canine] Nonguideline.

PC CODE: 281250 and 281251 (R-28153); 106201 (Amitraz) **DP BARCODE:** D 322975

TEST MATERIAL (PURITY): R-28153 (purity = 96.5%) and Amitraz (purity = 98.8%)

SYNONYMS: R-28153: Metaflumizone; BAS 320i; (*EZ*)-2'-[2-(4-cyanophenyl)-1-(α,α,α-trifluoro-*m*-tolyl)ethylidene]-4-(trifluoromethoxy)carbanilohydrazide; 2-[2-(4-cyanophenyl)-1-[3-(trifluoromethyl)phenyl]ethylidene]-*N*-[4-(trifluoromethoxy)phenyl]hydrazinecarboxamide

Amitraz: Mitaban; Preventic; Aazdieno; Acarac; Amitraze; Baam; Edrizan; Mitac; Maitac; Triatox; Triatix; Vapcozin Taktic; Triazid; Topline; Tudy; Ectodex; Garial; Danicut; Ovidrex; Acadrex; Bumetran; Ovasyn; *N*-methylbis(2;4-xylyliminomethyl)amine; *N'*-(2;4-dimethyl phenyl)-*N*-[[(2;4-dimethylphenyl)imino]methyl]-*N*-methylmethanimidamide

CITATION: Mezzasalma; T. (2005). Pharmacokinetics of R-28153 and amitraz after a single topical application to dogs at 20 mg/kg of R-28153 and amitraz. Avagadro, Parc de Genibrat, 31470 Fontenilles, France. Study No. A04904. May 11, 2005. MRID 46672101. Unpublished.

SPONSOR: Fort Dodge Animal Health, P.O. Box 5366 Princeton, NJ 08543-5366.

EXECUTIVE SUMMARY: In a dermal pharmacokinetic study (MRID 46672101), R28153 (15% w/v) and amitraz (15% w/v) were applied once as a mixture to the intrascapular area of three male and three female adult beagle dogs. Each dog received a ~22 mg/kg dose of each test material. Blood was collected from each dog 5 and 10 hours after dosing and on study days 1, 2, 3, 5, 7, 10, 14, 21, 28, 42, and 56.

Plasma concentrations of amitraz were below the limit of quantification (50 ng/mL) throughout the 56-day study. It was detected in the plasma of two female dogs 24 and 48 hours post-treatment, respectively. Amitraz was below the limit of detection (3.2 ng/mL) at all other times,

R28153 was above the limit of quantification (50 ng/mL) in three male dogs and one female dog during the study. The test material was quantifiable in the plasma of one male on day 7 through

the end of the study, in the second male from day 21 through day 42, and in the third male on days 28 and 42. R-28153 was quantifiable in one female dog's plasma on day 42 only. The individual maximum levels (range: 58.8 to 138.3 ng/mL) observed in these four dogs all occurred on day 42. Because the recovery of the test materials was mostly at or below the limit of quantification, pharmacological profiles of R-28153 and amitraz were not possible. The report includes the statement that: "The data do not allow a calculation of the pharmacokinetic profile of amitraz and R-28153 in dogs following application of the spot-on."

This study is considered Unacceptable/Nonguideline. It may be upgraded to acceptable if the dosing rationale were explained and if it is determined that an appropriate dose was used. The solvent used for the study should also be described.

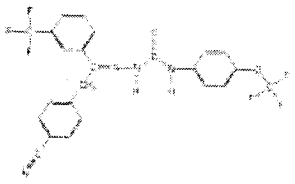
COMPLIANCE: Signed and dated GLP, Quality Assurance, and Data Confidentiality statements were provided.

I. MATERIALS AND METHODS:

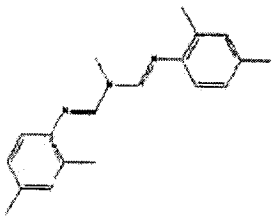
A. MATERIALS:

1. **Test material:** R-28153 (15% w/v) and Amitraz (15% w/v)

Description:	Clear yellow liquid
Lot/batch #:	0481704
Purity:	R-28153 = 7.4% Z isomer, 89.1% E isomer, total purity = 96.5%, Amitraz = 98.8%
Compound stability:	Expiration date: November 2004
CAS # for TGAI:	R-28153: 139968-49-3; Amitraz: 33089-61-1
Structure:	



R-21853



Amitraz

Vehicle/Solvent used:	Not reported
Source:	Fort Dodge Animal Health

2. **Relevance of test material to proposed formulation(s):** R-28153 and amitraz were mixed to provide a solution of 15% w/v each. The solvent was not reported.

3. **Test animals:**

Species:	Canine
Strain:	Beagle
Age/weight at study initiation:	Males – 10.3 ± 3 kg; Females – 9.0 ± 0.9 kg
Source:	Harlan, Zi le Malcourlet, 03800 Gannat, France
Housing:	Individually in stainless-steel cages with slatted stainless-steel floors
Diet:	300 g/day, Harlan Teklad 2021 dog food

Water:	Tap water, *ad libitum*	
Environmental conditions:	**Temperature:**	17-20EC
	Humidity:	43-90%
	Air changes:	Not reported
	Photoperiod:	Not reported
Acclimation period:	8 days	

B. STUDY DESIGN:

1. **In-life dates:** Start: September 28, 2004; End: November 23, 2004. At completion of the study, the dogs were returned to the study laboratory's stock supply.

2. **Dose:** A dose selection rationale was not provided. Each dog received a single 0.134 mL/kg body weight dose of test material formulation. The actual dose to each dog was 21.5 ± 0.6 mg/kg for amitraz and 22.1 ± 0.63 mg/kg for R28153.

3. **Animal preparation and dose application:** Before treatment, the appropriate volume for each dog was drawn into a disposable syringe. The test material was applied to a single spot on the midline between the shoulder blades by parting the hair, placing the syringe on the skin, and slowly dispensing the test material. A total of six dogs were treated, three male and three female. Elizabethan collars were placed on all the dogs and remained in place throughout the study. The dispensing syringes were weighed pre- and post-dosing to determine volume administered.

4. **Sample collection:** Heparinized whole blood was collected before dosing, approximately 5 and 10 hours after dosing and on study days 1, 2, 3, 5, 7, 10, 14, 21, 28, 42, and 56. The blood was centrifuges to obtain plasma which was aliquoted into three samples that were stored frozen until time of assay.

5. **Clinical observation:** All dogs were observed ~4 and 8 hours after treatment and daily thereafter for clinical signs of toxicity.

6. **Analytical technique:** The plasma concentrations of R28153 and amitraz were determined by HPLC. The samples were analyzed on a Zorbax SB-C18 column under gradient conditions with acetonitrile, methanol, and 0.1% formic acid. The effluent was monitored at 284 nm on a Waters UV detector. Appropriate calibration curves (six calibration points) for each test material and three levels of quality control plasma (75, 300, and 800 ng/mL) were used for analyses. The lower limit of quantification for amitraz in dog plasma was 50 ng/mL and the lower limit of detection was 3.2 ng/mL. The lower limit of quantification for R28153 was 50 ng/mL with a lower limit of detection of 1.0 ng/mL. Earlier studies had shown plasma amitraz samples were stable for three weeks frozen or for 30 minutes at room temperature.

II. RESULTS:

A. CLINICAL SIGNS OF TOXICITY:

No treatment-related clinical signs of toxicity were observed. Periodic body weights were not reported.

B. PLASMA CONCENTRATIONS:

Amitraz was below the limit of quantification throughout the 56-day study. It was detected in the plasma of two female dogs 24 and 48 hours post-treatment, respectively. At all other times, amitraz was below the limit of detection.

R28153 was above the limit of quantification in three male dogs and one female dog during the study. The test material was detected in the plasma of one male on day 7 through the end of the study, in the second male from day 21 through day 42, and in the third male on days 28 and 42. The individual maximum levels (range: 58.8 to 138.3 ng/mL) observed in these four dogs all occurred on day 42. R28153 was detected in one female dog's plasma on day 42 only. R-28153 was below the limit of quantification at all other times.

III. DISCUSSION AND CONCLUSIONS:

A. **INVESTIGATORS' CONCLUSIONS**: Based on the results, the study author stated that the bioavailabilities of amitraz and R-28153 are low when applied to the skin. The author also suggested that plasma levels of R28153 may be slightly higher in males than females.

B. **REVIEWER COMMENTS**: Because the amount of test material recovered was at or below the limit of quantification, pharmacological profiles of R28153 and amitraz were not possible. Assumptions concerning the bioavailability of R28153 being higher in male dogs are not valid because of the low concentrations recovered and the few animals used to conduct the study.

In the four dogs in which serum R-28153 was quantifiable, the highest individual levels (range: 58.8 to 138.3 ng/mL) all occurred on day 42.

The reviewer considers this study to be Unacceptable/Nonguideline. It may be upgraded to acceptable if the dosing rationale were explained and if it is determined that a suitable dose was used. The solvent used for the study should also be described.

C. **STUDY DEFICIENCIES:** The dosing rationale, dose selection, and dosing solvent should be described.