

US EPA ARCHIVE DOCUMENT

**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

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**Disclaimer**

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

\_\_\_\_\_  
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**Registration Action Branch 1, Health Effects Division (7509P)**

**Date:** \_\_\_\_\_

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**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-( $\alpha,\alpha,\alpha$ -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 Unpublished.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."

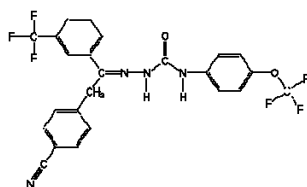
**This study is considered Unacceptable/Nonguideline. It may be upgraded to acceptable if the dosing rationale were explained and if it is determined that an appropriate dose was used. The solvent used for the study should also be described.**

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

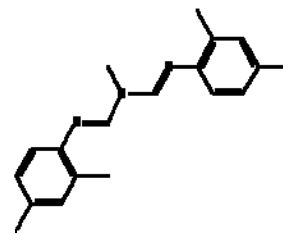
## I. MATERIALS AND METHODS:

### A. MATERIALS:

- 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

- 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

<b>Diet:</b>	300 g/day, Harlan Teklad 2021 dog food
<b>Water:</b>	Tap water, <i>ad libitum</i>
<b>Environmental conditions:</b>	<b>Temperature:</b> 17-20EC
	<b>Humidity:</b> 43-90%
	<b>Air changes:</b> Not reported
	<b>Photoperiod:</b> Not reported
<b>Acclimation period:</b>	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  $21.5 \pm 0.6$  mg/kg for amitraz and  $22.1 \pm 0.63$  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 centrifuged 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.