

US EPA ARCHIVE DOCUMENT



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

OFFICE OF CHEMICAL SAFETY
AND POLLUTION PREVENTION

November 23, 2010

MEMORANDUM

Subject: Name of Pesticide Product: SERGEANT'S FIPRONIL + CYPHENOTHRIN
SQUEEZE-ON FOR DOGS
EPA Reg. No. /File Symbol: 2517-RUN
DP Barcode: DP 380756
Decision No.: 435798
Action Code: R310
PC Codes: 129121 (Fipronil: 9.8%)
129013 (Cyphenothrin: 5%)

From: Byron T. Backus, Ph.D., Toxicologist *Byron T. Backus*
Technical Review Branch
Registration Division (7505P) *NOV-23-2010* *M. Hasler*

To: Bonaventure Akinlosotu/Richard Gebken RM 10
Insecticide Branch
Registration Division (7505P)

Registrant: SERGEANT'S PET CARE PRODUCTS, INC.

FORMULATION FROM LABEL:

| | | |
|------------------------------|-------|-----------------|
| <u>Active Ingredient(s):</u> | | <u>% by wt.</u> |
| 129121 Fipronil | | 9.8 |
| 129013 Cyphenothrin | | 5.0 |
| <u>Other Ingredient(s):</u> | | <u>85.2</u> |
| | TOTAL | 100.0 |

ACTION REQUESTED: The Risk Manager requests:

“For your review: MRIDs 481296-03 thru 08 for an R310, new EP...”

BACKGROUND:

The material received for review includes 5 acute toxicity studies (MRID 48129803: acute oral LD50; 48129604: acute dermal LD50; 48129606: primary eye irritation; 48129607: primary dermal irritation; 48129608: dermal sensitization) conducted on a formulation identified as “Dog Products,” a clear liquid with a density of 1.030 g/cm³, containing as active ingredients: Fipronil: 9.8%; S-Methoprene: 8.5%; Cyphenothrin: 5.5%; and Pyriproxyfen: 2%). There is a waiver request (MRID 48129605) for acute inhalation toxicity testing. In addition, there is a proposed label (signal word: WARNING) which indicates the following dosage rates: 0.023 fl. oz. (0.67 mL) for dogs and puppies of up to 22 lbs; 0.045 fl. oz. (1.34 mL) for dogs 23-44 lbs; 0.091 fl. oz. (2.68 mL) for dogs 45-88 lbs; and 0.136 fl. oz. (4.02 mL) for dogs 89-132 lbs. The statements are made on p. 5 of the label that: “Repeat application may be made if necessary, but do not apply more often than once every 4 weeks.” and “Do not reapply for 30 days.” The proposed label includes the statement: “DO NOT USE ON CATS.” In addition, this package includes a data matrix, a CSF (dated 06/18/2010) and a cover letter dated June 18, 2010.

COMMENTS AND RECOMMENDATIONS:

1. The five acute toxicity studies have been classified as acceptable. After taking into consideration the composition of the test material, and the formulation for 2517-RUN, TRB concludes that these studies can be used to satisfy the acute toxicity data requirements for the registration of 2517-RUN. However, the data matrix indicates (under 870.7200) a new submission for a dog companion animal safety study, which was not a part of this package. 17

2. TRB has concluded that based on the amount of formulation applied (0.5-4.5 mL), the packaging of these products (single-use containers or ampules), the non-volatile nature of the formulations, and the application method (directly to the skin of the dog or cat) which does not result in any respirable particles, a waiver for the acute inhalation toxicity study requirement is appropriate, and that these products (including 2517-RUN) can be assigned to EPA Toxicity Category IV by this exposure route.

3. The following is the acute toxicity profile for Sergeant’s Fipronil + Cyphenothrin Squeeze-On for Dogs (EPA File Symbol 2517-RUN), based on the results of the acute toxicity studies:

| | | | |
|---------------------------|----------|------------|-----------------|
| Acute oral toxicity | III | Acceptable | MRID 48129603 |
| Acute dermal toxicity | III | Acceptable | MRID 48129604 |
| Acute inhalation toxicity | (IV) | Waived | (MRID 48129605) |
| Primary eye irritation | II | Acceptable | MRID 48129606 |
| Primary dermal irritation | IV | Acceptable | MRID 48129607 |
| Dermal sensitization | Positive | Acceptable | MRID 48129608 |

4. Based on the acute toxicity profile above, and taking into consideration the proposed uses specified on the label and information in the CSF, the following would be the precautionary and first aid labeling for EPA File Symbol 2517-RUN [Sergeant’s Fipronil + Cyphenothrin Squeeze-On for Dogs] as obtained from the Label Review System:

PRODUCT ID #: 002517-00140

PRODUCT NAME: SERGEANT'S FIPRONIL + CYPHENOTHRIN SQUEEZE-ON FOR DOGS

PRECAUTIONARY STATEMENTS

SIGNAL WORD: WARNING

Hazards to Humans and Domestic Animals:

Restricted Use Pesticide due to toxicity categories. For retail sale to and use only by Certified Applicators or persons under their direct supervision and only for those uses covered by the Certified Applicator's certification. Child Resistant Packaging Required.

Causes substantial but temporary eye injury. Harmful if absorbed through skin. Harmful if swallowed. Do not get in eyes or on clothing. Wear protective eyewear (goggles, face shield, or safety glasses). Wash thoroughly with soap and water after handling and before eating, drinking, chewing gum, or using tobacco. Remove and wash contaminated clothing before reuse. Avoid contact with skin. Wear long-sleeved shirt and long pants, socks, shoes, and gloves.

Prolonged or frequently repeated skin contact may cause allergic reactions in some individuals.

First Aid:

If in eyes:

- Hold eye open and rinse slowly and gently with water for 15-20 minutes.
- Remove contact lenses, if present, after the first 5 minutes, then continue rinsing.
- Call a poison control center or doctor for treatment advice.

If on skin:

- Take off contaminated clothing.
- Rinse skin immediately with plenty of water for 15-20 minutes.
- Call a poison control center or doctor for treatment advice.

If swallowed:

- Call a poison control center or doctor immediately for treatment advice.
- Have person sip a glass of water if able to swallow.
- Do not induce vomiting unless told to by a poison control center or doctor.
- Do not give anything to an unconscious person.

NOTE TO PHYSICIAN: Note to PM/CRM/Registrant: The proposed label should contain a "Note to Physician". The following statements are suggested types of information that may be included, if applicable: - technical information on symptomatology; - use of supportive treatments to maintain life functions; - medicine that will counteract the specific physiological effects of the pesticide; - company telephone number to specific medical personnel who can provide specialized medical advice.

Have the product container or label with you when calling a poison control center or doctor or going for treatment. You may also contact 1-800-xxx-xxxx for emergency medical treatment information.

✓ According to the LRS, this product is a candidate for restricted use because it is in Toxicity Category II for eye exposure. However, TRB concludes that, based on the packaging (in ampules) and proposed product use (direct application to the skin of dogs or cats), the risk of eye exposure is adequately reduced, and a restricted use classification is not necessary.

5. The CSF (dated 06/18/2010) for 2517-RUN should also be reviewed and accepted by the TRB Chemistry Team.

STUDY TYPE: Acute Oral Toxicity – Rat; OPPTS 870.1100; OECD 425

TEST MATERIAL: “Dog Products,” a clear liquid with a density of 1.030 (p. 20 of MRID 48129603) or 1.037 (p. 22 of MRID 48129603) g/cm³, containing as active ingredients: Fipronil: 9.8%; S-Methoprene: 8.5%; Cyphenothrin: 5.5%; and Pyriproxyfen: 2%.

SYNONYMS: DOG SPOT ON SOUP

CITATION: Leoni, A. (2010) Acute Oral Toxicity Up and Down Procedure with Dog Products (in Rats): Final Report. Project Number: 100137B. Unpublished study prepared by BSL Bioservice Scientific Laboratories GmbH. May 3, 2010. 45 p. MRID 48129603.

SPONSOR: Sergeant’s Pet Care Products, Inc., Omaha, NE 68130-1703

EXECUTIVE SUMMARY: In an acute oral toxicity study (MRID 48129603), fasted (17.5-20 hrs) female WISTAR [albino] rats Crl: WI(Han)(Full-Barrier) (8-11 weeks of age; 154-185 g; source: Charles River, 97633 Sulzfeld, Germany) were orally gavaged with “Dog Products.” The rats dosed at 5000 mg/kg received undiluted test material; those dosed at less than 5000 mg/kg received dilutions of the test material in cottonseed oil, with a constant dose volume of 5 mL/kg.

Five animals were used in the initial limit test (5000 mg/kg), and 8 in the subsequent main test (doses ranging from 175 to 5000 mg/kg).

In the limit test, the initial animal dosed at 5000 mg/kg survived. The second was found dead on day 5, and the third was found dead on day 7. The fourth survived. The fifth was found dead on day 6, so a total of 2 out of 5 animals survived (indicating an LD₅₀ < 5000 mg/kg).

In the subsequent main test, there were no deaths at 175 or 550 mg/kg. One out of 2 animals dosed at 1750 mg/kg died; and 2 out of 3 animals dosed at 5000 mg/kg died.

All deaths occurred on days 5-7 with the exception of the one mortality at 1750 mg/kg (rat found dead on day 1).

Signs of toxicity in the one rat dosed at 175 mg/kg were moderately reduced spontaneous activity, kyphosis (curvature of the upper back) and slight to moderate piloerection, with recovery on day 2 (dosing was on day 1). One rat dosed at 550 mg/kg showed severely reduced spontaneous activity, kyphosis, moderate piloerection, anxiety and closure of eyes, with recovery on day 5; the other rat dosed at 550 mg/kg showed no signs of toxicity. Additional signs at 1750 mg/kg included tremor and ataxia; and at 5000 mg/kg included aggressiveness and weight loss.

Female rat LD₅₀ = 2722 mg/kg with a 95% PL confidence interval of 279.6 to >20,000 mg/kg.

Based on the acute oral LD₅₀ = 2722 mg/kg, "Dog Products," a clear liquid with a density of 1.030 (p. 20 of MRID 48129603) or 1.037 (p. 22 of MRID 48129603) g/cm³, containing as active ingredients: Fipronil: 9.8%; S-Methoprene: 8.5%; Cyphenothrin: 5.5%; and Pyriproxyfen: 2%, is in EPA Toxicity Category III for oral toxicity.

COMPLIANCE: Signed and dated GLP, Quality Assurance and [No] Data Confidentiality statements were provided.

RESULTS and DISCUSSION:

AOT425statpgm (Version: 1.0) Test Results and Recommendations
Acute Oral Toxicity (OECD Test Guideline 425) Statistical Program

Date/Time: Thursday, November 18, 2010, 10:54:29 AM
Data file name: work.dat
Last modified: 11/18/2010 10:54:24 AM

Test/Substance: Dog Products
Test type: Limit Test
Limit dose (mg/kg): 5000
Assumed LD50 (mg/kg): Default
Assumed sigma (mg/kg): 0.5

DATA:

| Test Seq. | Animal ID | Dose (mg/kg) | Short-term Result | Long-term Result |
|-----------|-----------|--------------|-------------------|------------------|
| 1 | 1 | 5000 | O | O |
| 2 | 2 | 5000 | O | X |
| 3 | 3 | 5000 | O | X |
| 4 | 4 | 5000 | O | O |
| 5 | 5 | 5000 | O | X |

(X = Died, O = Survived)

Dose Recommendation: The limit test is complete. A main test may be needed.

SUMMARY OF LONG-TERM RESULTS:

| Dose | O | X | Total |
|-----------|---|---|-------|
| 5000 | 2 | 3 | 5 |
| All Doses | 2 | 3 | 5 |

Statistical Estimates:

The LD50 is less than 5000 mg/kg.

AOT425statpgm (Version: 1.0) Test Results and Recommendations
Acute Oral Toxicity (OECD Test Guideline 425) Statistical Program

Date/Time: Thursday, November 18, 2010, 10:59:11 AM

Data file name: work.dat

Last modified: 11/18/2010 10:59:08 AM

Test/Substance: Dog Products

Test type: Main Test

Limit dose (mg/kg): 5000

Assumed LD50 (mg/kg): Default

Assumed sigma (mg/kg): 0.5

Recommended dose progression: 5000, 1750, 550, 175, 55, 17.5, 5.5, 1.75

DATA:

Test Animal Dose Short-term Long-term
Seq. ID (mg/kg) Result Result

| | | | | |
|---|----|------|---|---|
| 1 | 6 | 175 | O | O |
| 2 | 7 | 550 | O | O |
| 3 | 8 | 1750 | X | X |
| 4 | 9 | 550 | O | O |
| 5 | 10 | 1750 | O | O |
| 6 | 11 | 5000 | O | X |
| 7 | 12 | 5000 | O | O |
| 8 | 13 | 5000 | O | X |

(X = Died, O = Survived)

Dose Recommendation: The main test is complete.

Stopping criteria met: 3 at Limit Dose.

SUMMARY OF LONG-TERM RESULTS:

| Dose | O | X | Total |
|------|---|---|-------|
| 175 | 1 | 0 | 1 |
| 550 | 2 | 0 | 2 |
| 1750 | 1 | 1 | 2 |
| 5000 | 1 | 2 | 3 |

| | | | |
|-----------|---|---|---|
| All Doses | 5 | 3 | 8 |
|-----------|---|---|---|

Statistical Estimate based on long term outcomes:

Estimated LD₅₀ = 2722 (Based on maximum likelihood).
95% PL Confidence interval is 279.6 to Greater than 20,000.

- A. **Mortality**: In the limit test a total of 2 out of 5 animals survived dosage at 5000 mg/kg (indicating an LD₅₀ < 5000 mg/kg). In the subsequent main test there were no deaths at 175 (one rat) or 550 (two rats) mg/kg. One of 2 animals dosed at 1750 mg/kg died; and 2 out of 3 animals dosed at 5000 mg/kg died. All deaths occurred on days 5-7 (dosage was on day 1) with the exception of the mortality which occurred at 1750 mg/kg (rat found dead on day 1).
- B. **Clinical observations**: Signs of toxicity in the one rat dosed at 175 mg/kg were moderately reduced spontaneous activity, kyphosis (curvature of the upper back) and slight to moderate piloerection, with recovery on day 2 (dosing was on day 1). One rat dosed at 550 mg/kg showed severely reduced spontaneous activity, kyphosis, moderate piloerection, anxiety and closure of eyes, with recovery on day 5; the other rat dosed at 550 mg/kg showed no signs of toxicity. Additional signs at 1750 mg/kg included tremor and ataxia, and at 5000 mg/kg included aggressiveness and weight loss.
- C. **Gross necropsy**: There were no significant findings in any of the rats which survived the 14-day exposure period, except for one of the 5000 mg/kg rats in the limit test which showed piloerection. All rats that died showed lung congestion along with a number of other findings that included lateral position, red secretion at the nose, slight red salivation, brownish dried discharge around the mouth, foam in the mouth, dried foam around the mouth, and (in one rat dosed at 5000 mg/kg) bloody and small sized thymus. Two of the three rats which survived dosage at 5000 mg/kg had weight losses between day 1 and 8, but then had subsequent weight gains between day 8 and 15.
- D. **Reviewer's conclusions**: We can accept the findings of this study and the reported LD₅₀ value (female rat LD₅₀ = 2722 mg/kg) which places Dog Products in EPA Toxicity Category III for oral toxicity.

STUDY TYPE: Acute Dermal Toxicity – Rat; OPPTS 870.1200; OECD 402

TEST MATERIAL: “Dog Products,” a clear liquid with a density of 1.030 (p. 18 of MRID 48129604) g/cm³, containing as active ingredients: Fipronil: 9.8%; S-Methoprene: 8.5%; Cyphenothrin: 5.5%; and Pyriproxyfen: 2%.

SYNONYMS: DOG SPOT ON SOUP

CITATION: Leoni, A. (2010) Acute Dermal Toxicity (Limit Test) with Dog Products (in Rats): Final Report. Project Number: 100138B. Unpublished study prepared by BSL Bioservice Scientific Laboratories GmbH. March 23, 2010. 34 p. MRID 48129604.

SPONSOR: Sergeant’s Pet Care Products, Inc., Omaha, NE 68130-1703

EXECUTIVE SUMMARY: In an acute dermal toxicity study (MRID 48129604) 5 male and five female WISTAR [albino] rats Crl: WI(Han)(Full-Barrier) (males: 9 weeks of age; 234-247 g; females: 13 weeks of age; 220-235 g; source: Charles River, 97633 Sulzfeld, Germany) were dermally exposed to 2000 mg/kg undiluted “Dog Products,” a clear liquid with a density of 1.030 g/cm³, containing as active ingredients: Fipronil: 9.8%; S-Methoprene: 8.5%; Cyphenothrin: 5.5%; and Pyriproxyfen: 2%. The appropriate amount of test material was applied evenly on each rat over a dose area of approximately 10% of the body surface. The test material was kept in contact with the skin by a gauze dressing and non-irritating tape during the 24-hour exposure period. Residual test material was not removed from the skin at the end of the exposure period. The rats were observed for 14 days.

There were no deaths. There were no signs of systemic toxicity. From information on p. 15-16 of MRID 48129604 there was slight dermal irritation in one female and one male. From p. 27 the female had desquamation on days 6-8; from p. 28 the male had slight scratches on days 10 to 14. Three females had weight losses (from 6 to 18 g) between days 1 and 8; all rats gained weight between days 8 and 15, and all but one female gained weight between days 1 and 15.

No abnormalities were observed at gross necropsy.

LD₅₀ Males > 2000 mg/kg bw
LD₅₀ Females > 2000 mg/kg bw
LD₅₀ Combined > 2000 mg/kg bw

Based on the acute dermal LD₅₀ > 2000 mg/kg, “Dog Products,” a clear liquid with a density of 1.030 g/cm³, containing as active ingredients: Fipronil: 9.8%; S-Methoprene: 8.5%; Cyphenothrin: 5.5%; and Pyriproxyfen: 2%, is in EPA Toxicity Category III for dermal toxicity.

This study is classified as acceptable. It does satisfy the guideline requirements for an acute dermal study (OPPTS 870.1200; OECD 402) in the rat.

COMPLIANCE: Signed and dated GLP, Quality Assurance and [No] Data Confidentiality statements were provided.

RESULTS and DISCUSSION:

| Dose (mg/kg bw) | Mortality/Number Tested | | |
|--------------------|-------------------------|---------|----------|
| | Males | Females | Combined |
| 2000 | 0/5 | 0/5 | 0/10 |

- A. **Mortality:** There were no deaths.
- B. **Clinical observations:** There were no signs of systemic toxicity. From information on p. 15-16 of MRID 48129604 there was slight dermal irritation in one female and one male. From p. 27 the female had desquamation on days 6-8; from p. 28 one male had slight scratches on days 10 to 14. Three females had weight losses (from 6 to 18 g) between days 1 and 8; all rats gained weight between days 8 and 15, and all but one female gained weight between days 1 and 15.
- C. **Gross necropsy:** There were no observable abnormalities.
- D. **Reviewer's conclusions:** The acute dermal LD₅₀ for males, females (and the combined sexes) is greater than 2000 mg/kg bw. This places "Dog Products," a clear liquid with a density of 1.030 g/cm³ in EPA Toxicity Category III for dermal toxicity.

Reviewer: Byron T. Backus, Ph.D.
Risk Manager (EPA): 10

Date: November 22, 2010

STUDY TYPE: Acute Inhalation Toxicity – Rat; OPPTS 870.1300; OECD 403

TEST MATERIAL: Combinations of Fipronil + Cyphenothrin + S-Methoprene + Nylar used as spot-ons for dogs and cats.

CITATION: Hoskins, K. (2010) Waiver Request of Requirement for Further Testing for Acute Inhalation Toxicity for Following Sergeant's Products: Sergeant's Fipronil + Cyphenothrin Squeeze-On for Dogs, Sergeant's Fipronil + Cyphenothrin + Methoprene Squeeze-On for Dogs, Sergeant's Fipronil + Cyphenothrin + Nylar Squeeze-On for Dogs, Sergeant's Fipronil + Nylar Squeeze-On for Dogs, Sergeant's Fipronil + Etofenprox Spot-On for Cats, Sergeant's Fipronil + Etofenprox + Methoprene Spot-On for Cats, Sergeant's Fipronil + Etofenprox + Nylar Spot-On for Cats, Sergeant's Fipronil + Nylar Spot-On for Cats. Project Number: SERGEANTS/06/10/05. Unpublished study prepared by Sergeant's Pet Care Products, Inc. June 18, 2010. 6 p. MRID 48129605.

SPONSOR: Sergeant's Pet Care Products, Inc., Omaha, NE 68130-1703

EXECUTIVE SUMMARY: This is a waiver request for a number of products used as spot-ons for dogs and cats. A single application is 0.5-4.5 mL, these formulations have a non-volatile composition, and the application method (directly to the skin of the dog or cat) does not result in any respirable particles.

Based on the amount of formulation applied (0.5-4.5 mL), the packaging of these products (single-use containers or ampules), the non-volatile nature of the formulations, and the application method (directly to the skin of the dog or cat) which does not result in any respirable particles, TRB concludes that a waiver for the acute inhalation toxicity study requirement is appropriate, and that these products can be assigned to EPA Toxicity Category IV by this exposure route.

STUDY TYPE: Primary Eye Irritation – Rabbit; OPPTS 870.2400; OECD 405

TEST MATERIAL: “Dog Products,” Batch No. 091129-dog, a clear liquid with a density of 1.030 (p. 18 of MRID 48129606) g/cm³, containing as active ingredients: Fipronil: 9.8%; S-Methoprene: 8.5%; Cyphenothrin: 5.5%; and Pyriproxyfen: 2%, pH: not reported.

SYNONYMS: DOG SPOT ON SOUP

CITATION: Ahuja, V. (2010) Acute Eye Irritation/Corrosion with Dog Products (in Rabbits): Final Report. Project Number: 100140B. Unpublished study prepared by BSL Bioservice Scientific Laboratories GmbH. March 17, 2010. 33 p. MRID 48129606.

SPONSOR: Sergeant’s Pet Care Products, Inc., Omaha, NE 68130-1703

EXECUTIVE SUMMARY: In a primary eye irritation study (MRID 48129606), 0.1 mL of undiluted “Dog Products” (a clear liquid with a density of 1.030 g/cm³, containing as active ingredients: Fipronil: 9.8%; S-Methoprene: 8.5%; Cyphenothrin: 5.5%; and Pyriproxyfen: 2%) was instilled in the conjunctival sac of one eye of each of 3 New Zealand white female rabbits (age: approximately 14 weeks; weight: 2.5-2.9 kg; source: Charles River Deutschland, D-97633 Sulzfeld. The eyes were examined and scored for irritation at 1, 24, 48 and 72 hours and at 4, 5 and 6 days (and if necessary, at 7 and 8 days) after instillation.

At one and 24 hours, all eyes were positive for conjunctival irritation, with scores for conjunctival redness and chemosis ≥ 2 . At 24 hours two of the eyes could not be evaluated for corneal opacity or iritis; the remaining eye scored zero for these criteria. One eye was positive for corneal opacity from 48 hours through day 7, and another eye was positive for corneal opacity on days 4 and 5. All eyes were completely clear (all scores zero) by day 8. A Maximum Mean Total Score (MMTS) could not be calculated from the data as presented.

In this study, all 3 eyes were positive for conjunctival irritation effects at 24 hours, and two eyes showed corneal opacity, which did not clear until day 8 for one eye. “Dog Products” (containing Fipronil: 9.8%; S-Methoprene: 8.5%; Cyphenothrin: 5.5%; and Pyriproxyfen: 2%) is in EPA Toxicity Category II for primary eye irritation.

This study is classified as acceptable. It does satisfy the guideline requirements for a primary eye irritation study (OPPTS 870.2400; OECD 405) in the rabbit.

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

RESULTS and DISCUSSION:

| Observations | Number of eyes positive/Number treated | | | | | |
|-----------------|--|-------------------|-----|-----|------|-------------------|
| | Hours | | | | Days | |
| | 1 | 24 | 48 | 72 | 4 | 7 |
| Corneal Opacity | 0/3 | 0 ^a /1 | 1/3 | 1/3 | 2/3 | 1 ^b /3 |
| Iritis | 0/3 | 0 ^a /1 | 0/3 | 0/3 | 0/3 | 0/3 |
| Conjunctivae | | | | | | |
| Redness* | 3/3 | 3/3 | 2/3 | 2/3 | 0/3 | 0/3 |
| Chemosis* | 3/3 | 3/3 | 3/3 | 2/3 | 0/3 | 0/3 |
| Discharge** | 3/3 | 3/3 | 2/3 | 1/3 | 0/3 | 0/3 |

* Score of 2 or more required to be considered "positive"

** Discharge does not indicate a positive effect according to the grading scale

^a Two eyes could not be evaluated for corneal opacity or iritis at 24 hours due to swelling.

^b This eye was clear for corneal opacity on day 8.

- A. **Observations:** At one and 24 hours, all eyes were positive for conjunctival irritation, with scores for conjunctival redness and chemosis ≥ 2 . Because of the swelling at 24 hours two of the eyes could not be evaluated for corneal opacity or iritis; the remaining eye scored zero for these criteria. One eye was positive for corneal opacity from 48 hours through day 7, and another eye was positive for corneal opacity on days 4 and 5. All eyes were completely clear (all scores zero) by day 8.
- B. **Results:** A Maximum Mean Total Score (MMTS) could not be calculated from the data as presented.
- C. **Reviewer's conclusions:** The test material is in EPA Toxicity Category II for eye irritation.

STUDY TYPE: Primary Dermal Irritation – Rabbit; OPPTS 870.2500; OECD 404

TEST MATERIAL: “Dog Products,” Batch No. 091129-dog, a clear liquid with a density of 1.030 (p. 17 of MRID 48129607) g/cm³, containing as active ingredients: Fipronil: 9.8%; S-Methoprene: 8.5%; Cyphenothrin: 5.5%; and Pyriproxyfen: 2%, pH: not reported.

SYNONYMS: DOG SPOT ON SOUP

CITATION: Ahuja, V. (2010) Acute Dermal Irritation/Corrosion with Dog Products (in Rabbits): Final Report. Project Number: 100139B. Unpublished study prepared by BSL Bioservice Scientific Laboratories GmbH. March 4, 2010. 29 p. MRID 48129607.

SPONSOR: Sergeant’s Pet Care Products, Inc., Omaha, NE 68130-1703

EXECUTIVE SUMMARY: In a primary dermal irritation study (MRID 48129607), three young adult female New Zealand white rabbits (age: approximately 18 weeks; weight: 2.9-3.2 kg; source: Charles River Deutschland, D-97633, Sulzfeld) were dermally exposed to 0.5 mL undiluted “Dog Products” (a clear liquid with a density of 1.030 g/cm³, containing as active ingredients: Fipronil: 9.8%; S-Methoprene: 8.5%; Cyphenothrin: 5.5%; and Pyriproxyfen). The test material was applied to a gauze patch which was then applied to a 6 cm² application site on the left side of the dorsal area. The gauze was held in place with non-irritating tape during the 4-hour exposure period, followed by scoring at 1, 24, 48 and 72 hours after patch removal.

There was no irritation, with all scores (for both erythema and edema) zero at all times.

The Primary Dermal Irritation Index = 0.00. The mean irritation score at 72 hours was 0.0.

In this study, “Dog Products” (a clear liquid with a density of 1.030 g/cm³, containing as active ingredients: Fipronil: 9.8%; S-Methoprene: 8.5%; Cyphenothrin: 5.5%; and Pyriproxyfen) was a non-irritant and is classified in EPA Toxicity Category IV for primary dermal irritation.

This study is classified as acceptable. It does satisfy the guideline requirements for a primary dermal irritation study (OPPTS 870.2500; OECD 404) in the rabbit.

COMPLIANCE: Signed and dated GLP, Quality Assurance and [No] Data Confidentiality statements were provided.

RESULTS and DISCUSSION:

| Animal Number | Sex | Hours | | | |
|---|--------|------------------|---------|---------|---------|
| | | ½-1 | 24 | 48 | 72 |
| 1 | Female | 0/0 ^a | 0/0 | 0/0 | 0/0 |
| 2 | Female | 0/0 | 0/0 | 0/0 | 0/0 |
| 3 | Female | 0/0 | 0/0 | 0/0 | 0/0 |
| Severity of Irritation: Mean Score | | 0.0/0.0 | 0.0/0.0 | 0.0/0.0 | 0.0/0.0 |

^a erythema/edema.

- A. **Observations:** There was no irritation, with all scores (for both erythema and edema) zero at all times.
- B. **Results:** The primary irritation index (PII, average of scores at 1, 24, 48 & 72 hours) = 0.00. The mean irritation score at 72 hours was 0.0.
- C. **Reviewer's conclusions:** In this study, "Dog Products" (a clear liquid with a density of 1.030 g/cm³) was a non-irritant. This formulation is classified in EPA Toxicity Category IV for primary dermal irritation.

STUDY TYPE: Dermal Sensitization – Guinea Pig; OPPTS 870.2600; OECD 406

TEST MATERIAL: “Dog Products,” Batch No. 091129-dog, a clear liquid with a density of 1.030 (p. 19 of MRID 48129608) g/cm³, containing as active ingredients: Fipronil: 9.8%; S-Methoprene: 8.5%; Cyphenothrin: 5.5%; and Pyriproxyfen: 2%, pH: not reported.

SYNONYMS: DOG SPOT ON SOUP

CITATION: Stelter, D. (2010) Test for Sensitization (Guinea Pig Maximisation Test) with Dog Products (in Guinea Pigs): Final Report. Project Number: 100141B. Unpublished study prepared by BSL Bioservice Scientific Laboratories GmbH. April 6, 2010. 41 p. MRID 48129608.

SPONSOR: Sergeant’s Pet Care Products, Inc., Omaha, NE 68130-1703

EXECUTIVE SUMMARY: In a dermal sensitization study (MRID 48129608) 10 young adult female Crl: HA [Hartley albino] full-barrier guinea pigs (age: 5-6 weeks; weight: 304-363 g; source: Charles River, 97633 Sulzfeld, Germany) received induction treatments according to the Guinea Pig Maximization Test Protocol. An additional 5 guinea pigs were assigned to the negative control group. Based on preliminary testing the following concentrations were used in the main test: 5% (in cottonseed oil) for intradermal induction; 100% for dermal induction, and 100% for challenge and re-challenge applications.

On day 0 each of the induced group received 3 pairs of 0.1 mL injections consisting of the following: Injection pair 1: a 1:1 (v/v) mixture of Freund’s Complete Adjuvant (FCA): physiological (0.9%) saline. Injection pair 2: a 5% concentration of the test item in cottonseed oil. Injection pair 3: a 5% concentration of the test item formulated in a 1:1 (v/v) mixture FCA: physiological (0.9%) saline.

Guinea pigs in the negative control group were also given 3 pairs of 0.1 mL injections; injection pair 1 was the same as that of the induced group, injection pair 2 consisted of 100% cottonseed oil, and injection pair 3 was a 50% (v/v) formulation of cottonseed oil in 1:1 (v/v) FCA: physiological (0.9%) saline.

On day 6 the application areas of the 10 induced and 5 negative control guinea pigs were treated with 0.5 mL of 10% sodium lauryl sulphate in vaseline to create local irritation. On day 7 the induced animals were treated with 0.5 g of the test item which was applied to the application area and held in contact there for 48 hours; the negative controls were similarly treated with 0.5 g of vaseline.

For challenge, on day 20 a patch containing 0.5 g of the test material was applied to the left flank of each animal, and a patch containing 0.5 g of vaseline was applied to the right flank. These patches were held in contact with the skin for 24 hours.

Because the results of the first challenge test were borderline, a rechallenge was conducted on day 27. In addition to the test (induced group) and negative controls, an additional negative control group of 5 animals was also tested.

Following first challenge, one of the previously induced guinea pigs showed a response at 24 hours and 3 showed a response at 48 hours; however, one of these 3 also showed a response to the vehicle. None of the negative controls showed a response at 24 hours, but one showed a response to the test material at 48 hours. Following the second challenge, 3 of the previously induced guinea pigs showed a response to the test item at 24 hours, but 2 of these guinea pigs also showed a response to the vehicle. Four of the previously induced guinea pigs showed a response at 48 hours, but 3 also showed a response to the vehicle.

A concurrent positive control study with 2-Mercaptobenzothiazole gave appropriate results (4/5 positive responses at 24 hours and 5/5 positive responses at 48 hours to the positive control; no positive responses to the vehicle).

Based on the results of this study, it is concluded that “Dog Products,” Batch No. 091129-dog, a clear liquid with a density of 1.030 g/cm³, containing as active ingredients: Fipronil: 9.8%; S-Methoprene: 8.5%; Cyphenothrin: 5.5%; and Pyriproxyfen: 2%, is a mild dermal sensitizer.

This study is classified as acceptable. It does satisfy the guideline requirements for a dermal sensitization study (OPPTS 870.2600; OECD 406) in the guinea pig.

COMPLIANCE: Signed and dated GLP, Quality Assurance and [No] Data Confidentiality statements were provided.

PROCEDURE:

- A. Induction:** On day 0 the 10 females in this group received 3 pairs of 0.1 mL injections consisting of the following: Injection pair 1: a 1:1 (v/v) mixture of Freund’s Complete Adjuvant (FCA): physiological (0.9%) saline. Injection pair 2: a 5% concentration of the test item in cottonseed oil. Injection pair 3: a 5% concentration of the test item formulated in a 1:1 (v/v) mixture FCA: physiological (0.9%) saline. On day 6 the application sites were treated with 0.5 mL of 10% sodium lauryl sulphate in vaseline to create local irritation. On day 7 the induced animals were treated with 0.5 g “Dog Products” which was applied to application site and held in contact there for 48 hours.
- B. Challenge:** On day 20 a patch containing 0.5 g “Dog Products” was applied to the left flank of each guinea pig, and a patch containing 0.5 g of vaseline was applied to the right flank. These patches were held in contact with the skin for 24 hours. Reactions were scored at 24 and 48 hours after removal of the patches.
- C. Rechallenge:** Because the results of the first challenge test were borderline, a rechallenge was conducted on day 27.

D Negative controls: A group of 5 females received 3 pairs of 0.1 mL intradermal injections on day 0. Injection pair 1 was the same as that of the induced group; injection pair 2 consisted of 100% cottonseed oil, and injection pair 3 was a 50% (v/v) formulation of cottonseed oil in 1:1 (v/v) FCA: physiological saline. On day 6 the application sites were treated with 0.5 mL of 10% sodium lauryl sulphate in vaseline to create local irritation. On day 7 0.5 g of vaseline was applied to the application site and held in contact there for 48 hours. On Day 20 a patch containing 0.5 g “Dog Products” was applied to the left flank of each guinea pig, and a patch containing 0.5 g of vaseline was applied to the right flank, with 24-hour exposure. Reactions were scored at 24 and 48 hours after removal of the patches.

These guinea pigs, as well as an additional group of 5 unexposed guinea pigs, was also treated on day 27, using the same procedure as on day 20.

RESULTS and DISCUSSION:

- A. Reactions and durations:** Following challenge, 1/10 induced and 0/5 negative control guinea pigs showed a reaction at 24 hours. 3/10 induced guinea pigs showed a positive response at 48 hours, but one of these 3 animals also showed a response to the vehicle; 1/5 negative controls showed a positive response to the test material at 48 hours. Following the rechallenge, 3 of the previously induced guinea pigs showed a response to the test item at 24 hours, but 2 of these guinea pigs also showed a response to the vehicle.
- B. Positive control:** A concurrent positive control study with 2-Mercaptobenzothiazole gave appropriate results (4/5 positive responses at 24 hours and 5/5 positive responses at 48 hours, no positive responses to the vehicle). These results are acceptable.
- C. Reviewer’s conclusions:** Based on the results of this study, it is concluded that “Dog Products,” Batch No. 091129-dog, a clear liquid with a density of 1.030 g/cm³, containing as active ingredients Fipronil: 9.8%; S-Methoprene: 8.5%; Cyphenothrin: 5.5%; and Pyriproxyfen: 2%, is a mild dermal sensitizer.

1. **DP BARCODE:** 380756
2. **PC CODES:** 129121 (Fipronil); 129013 (Cyphenothrin); 105401 (S-Methoprene); 129032 (Pyriproxyfen)
3. **CURRENT DATE:** November 22, 2010
4. **TEST MATERIAL:** "Dog Products," Batch No. 091129-dog, a clear liquid with a density of 1.030 (p. 19 of MRID 48129608) g/cm³, containing as active ingredients: Fipronil: 9.8%; S-Methoprene: 8.5%; Cyphenothrin: 5.5%; and Pyriproxyfen: 2%, pH: not reported.

| Study/Species/Lab Study # / Date | MRID | Results | Tox. Cat. | Core Grade |
|--|----------|---|-----------|------------|
| Acute oral toxicity/rat BSL Bioservice Scientific Laboratories GmbH. Project No. 100137B / May 3, 2010 | 48129603 | Three out of 5 females dosed at 5000 mg/kg died in limit test; in subsequent main test Oral LD ₅₀ (female rats) = 2722 (95% PL confidence interval 279.6- >20,000) mg/kg | III | A |
| Acute dermal toxicity/rat BSL Bioservice Scientific Laboratories GmbH. Project No. 100138B / March 23, 2010 | 48129604 | LD ₅₀ Males > 2000 mg/kg bw LD ₅₀ Females > 2000 mg/kg bw LD ₅₀ Combined > 2000 mg/kg bw | III | A |
| Acute inhalation toxicity/rat Sergeant's Pet Care Products Sergeant's 06-10/05 / June 18, 2010 | 48129605 | Waiver request; application is 0.5-4.5 mL; non-volatile composition; application method does not result in any respirable particles. | (IV) | W |
| Primary eye irritation/rabbit BSL Bioservice Scientific Laboratories GmbH. Project No. 100140B / March 17, 2010 | 48129606 | All 3 eyes positive for conjunctival irritation at 1 & 24 hours; one eye was positive for corneal opacity on day 7. All eyes were completely clear (all scores zero) by day 8. | II | A |
| Primary dermal irritation/rabbit BSL Bioservice Scientific Laboratories GmbH. Project No. 100139B / March 4, 2010 | 48129607 | No irritation; all scores = 0. PII = 0.00; the mean irritation score at 72 hours was 0.0. Formulation is a non-irritant. | IV | A |
| Dermal sensitization – Maximization Test /guinea pig BSL Bioservice Scientific Laboratories GmbH. Project No. 100141B / April 6, 2010 | 48129608 | Formulation is a mild dermal sensitizer. | Positive | A |

Core Grade Key: A = Acceptable, S = Supplementary, U = Unacceptable, W = Waived