MAY 9 1996

DP Barcode: D221290 Case: 027494

MEMORANDUM

Subject: EPA File Symbol/EPA Reg. No.: 45735-2/Ro-Pel Animal, Rodent and Bird Repellant

From: Carol E. Glasgow, Ph.D., Toxicologist Carol
Precautionary Review Section
Registration Support Branch (7505W)
Registration Division (7505C)

To: Robert Forrest, PM 14
Insecticide-Rodenticide Branch
Registration Division (7505C)

Applicant: Burlington Bio-Medical & Scientific Corp.
222 Sherwood Avenue
Farmingdale. NY 11735-1718

FORMULATION:

Active Ingredient (s):	% by weight
Denatonium saccharide	0.065
Thymol	0.035
Inert ingredient(s)	99.900

BACKGROUND: Burlington Bio-Medical & scientific Corp. submitted six studies (acute oral, dermal, inhalation toxicities, primary eye and dermal irritations and dermal sensitization) as required for 8-month call-in for re-registration. Research was performed by Product Safety Labs on MRID numbers 436981-07 through -12. The studies were originally reviewed by Dynamac Corporation.

RECOMMENDATION: RSB/PRS findings are as follows:

All studies are Acceptable. Minor comments are given below.

Although individual observations for the entire day of dosing were not conducted for the acute oral, acute dermal or acute inhalation toxicity studies and treatment site irritation was not addressed for the acute dermal toxicity, these deficiencies had no effect on the results of the study.

The actual test atmosphere concentration for the acute inhalation study was determined gravimetrically, which is considered an unacceptable method for liquid aerosols. However, since gravimetrically-determined concentrations are typically biased-low (because they usually do not account for volatilization of a test substance), and since no mortality occurred in this study at the calculated concentration of 2.52 mg/L (>limit concentration), this deficiency does not alter the observed LC_{50} values or subsequent Toxicity Category of Ro-Pel Animal, Rodent, and Bird Repellent and is considered minor.

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The aerodynamic particle size should have been determined hourly during the exposure. This deficiency is considered minor since the size was determined once every 2 hours, and since the individual MMAD values were comparable and within a reasonable range (4.2 and 4.0 μm).

The oxygen level during exposure was not reported. However, since the mean chamber airflow was 55.5 L/min (equivalent to 22.2 turnovers/hour), it is unlikely that the oxygen level was below 19%, and this deficiency is also considered minor.

TOXICITY PROFILES

Acute oral toxicity	IV	Acceptable
Acute dermal toxicity	IV	Acceptable
Acute inhalation toxicity	IV	Acceptable
Primary eye irritation	IV	Acceptable
Primary skin irritation	IV	Acceptable
Dermal sensitization	No	Acceptable

LABELING: Based on the studies submitted, the signal word for this product is "Caution." There is no special precautionary language needed for this product label.

DATA REVIEW FOR ACUTE ORAL TOXICITY TESTING (§81-1)

CITATION: Wnorowski, G. (1995) Acute oral toxicity limit test. Product Safety Labs, East Brunswick, NJ. Laboratory Project Number 3633. May 23, 1995. MRID 43698107.

EXECUTIVE SUMMARY: In an acute oral toxicity study (MRID 43698107), a group of five young adult Sprague-Dawley albino rats/sex were given a single oral dose of Ro-Pel Animal, Rodent, and Bird Repellent (0.035% thymol and 0.065% benzyldiethyl[(2,6-xylylcarbamoyl)methyl]ammonium saccharide) at 5,000 mg/kg (limit concentration); the test substance was administered as received. Animals were observed for clinical signs and mortality for up to 14 days postdosing.

Oral LD_{50} Males = >5,000 mg/kg (observed)
Females = >5,000 mg/kg (observed)

Ro-Pel Animal, Rodent, and Bird Repellent is **TOXICITY CATEGORY IV** based on calculated LD_{50} values in both sexes.

All animals survived and appeared normal and healthy during the 14-day observation period. No treatment-related effect on body weight was observed, and necropsy of animals sacrificed after 14 days revealed no gross abnormalities.

This study is **acceptable**, and satisfies the guideline requirement for an acute oral study (81-1) in the rat.

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

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test Material: Ro-Pel Animal, Rodent, and Bird Repellent
   Description: Clear liquid
   Lot/Batch #: B2225A
   Purity: 0.035% Thymol and 0.065% benzyldiethyl-[(2,6-xylylcarbamoyl)methyl]ammonium saccharide
   CAS #: 89-83-8 and 90823-38-4, respectively

2. Vehicle: None employed

3. Test animals: Species: Rat
   Strain: Sprague-Dawley derived, albino
   Age: Young adult
   Weight: 211-232 g males; 199-229 g females

Source: Hilltop Lab Animals, Scottdale, PA
Acclimation period: 16 days
Diet: Purina Rodent Chow (#5012), unspecified amount/animal/day
Water: Filtered tap water, ad libitum

B. STUDY DESIGN and METHODS:

1. In-life dates: April 20-May 4, 1995

2. Animal assignment and treatment: Following a 19-hour fasting period, five young adult rats/sex were given a single oral dose of Ro-Pel Animal, Rodent, and Bird Repellent at 5,000 mg/kg (limit concentration) by gavage; the test substance was administered as received. The rats were observed for signs of toxicity and/or mortality at 1, 3, and 21 hours following administration, and at least once daily thereafter for the remainder of the 14-day study; body weights were recorded at 0 (prior to dosing), 7, and 14 days. At 14 days, the surviving animals were sacrificed, necropsied, and examined for gross pathological changes.

3. Statistics: Not applicable to this study.

II. **RESULTS AND DISCUSSION**:

A. Mortality: All animals survived the 14-day observation period.

Oral LD_{50} Males = >5,000 mg/kg (observed)
Females = >5,000 mg/kg (observed)

B. Clinical observations: All animals appeared normal and healthy during the 14-day observation period.

C. Body Weight: No treatment-related effect on body weight was observed, with overall (0-14 days) average increases of 53% for males and 24% for females.

D. Necropsy: Gross necropsy revealed no treatment-related gross pathological changes.

E. Deficiencies: Although individual observations for the entire day of dosing were not conducted, this deficiency has no effect on the results of the study and is considered minor.

DATA REVIEW FOR ACUTE DERMAL TOXICITY TESTING (§81-2)

CITATION: Wnorowski, G. (1995) Acute dermal toxicity limit test. Product Safety Labs, East Brunswick, NJ. Laboratory Project Number 3636. May 25, 1995. MRID 43698108.

EXECUTIVE SUMMARY: In an acute dermal toxicity study (MRID 43698108), five young adult Sprague-Dawley albino rats/sex were dermally exposed to Ro-Pel Animal, Rodent, and Bird Repellent (0.035% thymol and 0.065% benzyldiethyl[(2,6-xylylcarbamoyl)-methyl]ammonium saccharide) at 5,000 mg/kg (>2X limit concentration) for 24 hours; the test substance was applied as received to approximately 10% of the total body surface area. Animals were observed for clinical signs and mortality for up to 14 days postdosing.

Dermal LD_{50} Males = >5,000 mg/kg (observed)
Females = >5,000 mg/kg (observed)

Ro-Pel Animal, Rodent, and Bird Repellent is **TOXICITY CATEGORY IV** based on observed LD_{50} values in both sexes.

All animals survived and appeared normal and healthy during the 14-day observation period; the presence or absence of treatment site irritation was not addressed. No significant treatment-related effect on body weight was observed, and necropsy of animals sacrificed after 14 days revealed no gross abnormalities.

This study is **acceptable**, and satisfies the guideline requirement for an acute dermal study (81-2) in the rat.

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

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test Material: Ro-Pel Animal, Rodent, and Bird Repellent
   Description: Clear liquid
   Lot/Batch #: B2225A
   Purity: 0.035% Thymol and 0.065% benzyldiethyl-[(2,6-xylylcarbamoyl)methyl]ammonium saccharide
   CAS #: 89-83-8 and 90823-38-4, respectively

2. Vehicle: None employed

3. Test animals: Species: Rat
   Strain: Sprague-Dawley derived, albino
   Age: Young adult

Weight: 204-228 g males; 201-218 g females
Source: Hilltop Lab Animals, Scottdale, PA
Acclimation period: 7 days
Diet: Purina Rodent Chow (#5012), unspecified amount/animal/day
Water: Filtered tap water, ad libitum

B. STUDY DESIGN and METHODS:

1. In-life dates: May 1-15, 1995

2. Animal assignment and treatment: Fur from the dorsal trunk areas of five animals/sex was clipped 1 day prior to dermal administration of Ro-Pel Animal, Rodent, and Bird Repellent at 5,000 mg/kg (>2X limit concentration). The test substance was applied as received and spread evenly over an area of approximately 6-in^2 (10% of the total body surface area). Each application site was covered with an 6.75-in^2 adhesive-backed gauze patch, and the entire trunk of each animal was wrapped with Durapore tape. The coverings were removed 24 hours following application, and the test sites were gently wiped with water and a clean towel. The rats were observed for signs of toxicity and/or mortality at 0.25, 2, and 5 hours following application, and at least once daily thereafter for the remainder of the 14-day study; body weights were recorded at 0 (prior to dosing), 7, and 14 days. At 14 days, surviving animals were sacrificed, necropsied, and examined for gross pathological changes.

3. Statistics: Not applicable to this study.

II. **RESULTS AND DISCUSSION**:

A. Mortality: All animals survived the 14-day observation period.

Dermal LD_{50} Males = >5,000 mg/kg (observed)
Females = >5,000 mg/kg (observed)

B. Clinical observations: All animals appeared normal and healthy during the 14-day observation period. The presence or absence of treatment site irritation was not addressed.

C. Body Weight: No significant treatment-related effect on body weight was observed, with overall (0-14 days) average increases of 46% for males and 15% for females.

D. Necropsy: Gross necropsy revealed no treatment-related gross pathological changes.

E. Deficiencies: Although individual observations for the entire day of dosing were

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not conducted and treatment site irritation was not addressed, these deficiencies have no effect on the results of the study and are considered minor.

DATA REVIEW FOR ACUTE INHALATION TOXICITY TESTING (§81-3)

CITATION: Wnorowski, G. (1995) Acute inhalation toxicity limit test. Product Safety Labs, East Brunswick, NJ. Laboratory Project Number 3638. May 25, 1995. MRID 43698109.

EXECUTIVE SUMMARY: In an acute inhalation toxicity study (MRID 43698109), five young adult Sprague-Dawley albino rats/sex were exposed by whole-body inhalation to Ro-Pel Animal, Rodent, and Bird Repellent (0.035% thymol and 0.065% benzyldiethyl[(2,6-xylylcarbamoyl)methyl]ammonium saccharide) at 2.52 mg/L (>limit concentration) for 4 hours. Animals were observed for clinical signs and mortality for up to 14 days following exposure.

Inhalation LC_{50} Males = >2.52 mg/L (observed)
Females = >2.52 mg/L (observed)

Ro-Pel Animal, Rodent, and Bird Repellent is **TOXICITY CATEGORY IV** based on the observed LC_{50} values in both sexes.

All animals survived the 4-hour exposure and 14-day observation periods. Effects observed during exposure included irregular respiration, hunched posture, and hypoactivity. All effects subsided upon exposure completion, and all animals appeared normal and healthy during the 14-day observation period. The body weight of a single female animal decreased slightly between 7 and 14 days; otherwise, no treatment-related effect on body weight was observed. Necropsy of animals sacrificed after 14 days revealed no gross abnormalities.

This study is **acceptable**, and satisfies the guideline requirement for an acute inhalation study (81-3) in the rat.

COMPLIANCE: Signed and dated GLP and Data Confidentiality statements were provided. A signed Quality Assurance page was not included in the study, and the existence of the original microfiched page (at the EPA) needs to be verified.

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test Material: Ro-Pel Animal, Rodent, and Bird Repellent
   Description: Clear liquid
   Lot/Batch #: B2225A
   Purity: 0.035% Thymol and 0.065% benzyldiethyl-[(2,6-xylylcarbamoyl)methyl]ammonium saccharide
   CAS #: 89-83-8 and 90823-38-4, respectively

2. Vehicle and/or positive control: None employed

3. Test animals: Species: Rat

Strain: Sprague-Dawley derived, albino
Age: Young adult
Weight: 263-282 g males; 219-250 g females
Source: Hilltop Lab Animals, Scottdale, PA
Acclimation period: 27 days
Diet: Purina Rodent Chow (#5012), unspecified amount/day, except during exposure
Water: Tap water, ad libitum, except during exposure

B. STUDY DESIGN and METHODS:

1. In-life dates: May 1-15, 1995

2. Exposure conditions: A rectangular dynamic-flow perspex exposure chamber (150 L) was used; aside from "whole body", animal orientation within the chamber was not described.

Test atmosphere (liquid) was generated using a Spraying Systems 0.25-inch JCO atomizer operated using dry, compressed air. Measured test material was pumped via a Master Flex Pump into the atomization nozzle. The resultant aerosol was diluted with filtered, "conditioned" room air prior to entering the exposure chamber. The mean chamber airflow averaged 55.5 L/min (equivalent to 22.2 turnovers/hour). The times required for 90 and 99% equilibration were 6.2 and 12.4 minutes, respectively.

The nominal test atmosphere concentration was determined at the end of the exposure period by dividing the total amount of test material delivered to the chamber by the total air volume that passed through the chamber during the exposure time. The actual test atmosphere concentration was determined gravimetrically six times during the exposure period by drawing 12-L samples from the breathing zone of the animals through 25-mm Whatman glass fiber filters. The nominal and average gravimetric test concentrations were 71.56 and 2.52 mg/L, respectively.

Particle size was determined twice during the exposure period using an eight-stage Andersen cascade impactor. Samples were collected from the breathing zone of the animals. The calculated average mass median aerodynamic diameter (MMAD) and geometric standard deviation (GSD) were 4.1 and 1.91 µm. The average percentages of particles <9.0 and <3.3 µm were 96.2 and 21.5%, respectively.

During exposure, the temperature was 68-72 □F, and the relative humidity ranged from 42 to 100%; the oxygen level was not specified.

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3. Animal assignment and treatment: Five albino rats/sex were exposed via whole-body inhalation to Ro-Pel Animal, Rodent, and Bird Repellent at 2.52 mg/L (>limit concentration) for 4 hours. Following exposure, each animal was wiped free of excess test substance. The animals were observed for signs of toxicity and/or mortality at least every 30 minutes during exposure, upon chamber removal, and at least once daily thereafter for the remainder of the 14-day study. Body weights were recorded at 0 (prior to exposure), 7, and 14 days. After 14 days, the surviving animals were sacrificed, necropsied, and examined for gross pathological changes.

4. Statistics: Not applicable to this study.

II. RESULTS AND DISCUSSION:

A. Mortality: All animals survived the 4-hour exposure and 14-day observation periods.

Inhalation LC_{50} Males = >2.52 mg/L (observed)
Females = >2.52 mg/L (observed)

B. Clinical observations: Effects observed during exposure included irregular respiration, hunched posture, and hypoactivity. All effects subsided upon exposure completion, and all animals appeared normal and healthy during the 14-day observation period.

C. Body Weight: The body weight of a single female animal decreased slightly between 7 and 14 days; otherwise, no treatment-related effect on body weight was observed. Overall (0-14 days) all animals gained weight, with averaged increases of 34 and 17% for male and female animals, respectively.

D. Necropsy: Gross necropsy revealed no treatment-related gross pathological changes.

E. Deficiencies: The actual test atmosphere concentration was determined gravimetrically, which is considered an unacceptable method for liquid aerosols. However, since gravimetrically-determined concentrations are typically biased-low (because they usually do not account for volatilization of a test substance), and since no mortality occurred in this study at the calculated concentration of 2.52 mg/L (>limit concentration), this deficiency does not alter the observed LC_{50} values or subsequent Toxicity Category of Ro-Pel Animal, Rodent, and Bird Repellent and is considered minor.

The aerodynamic particle size should have been determined hourly during the exposure. This deficiency is considered minor since the size was determined once every 2 hours, and since the individual MMAD values were comparable and within a reasonable range (4.2 and 4.0 μm).

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The oxygen level during exposure was not reported. However, since the mean chamber airflow was 55.5 L/min (equivalent to 22.2 turnovers/hour), it is unlikely that the oxygen level was below 19%, and this deficiency is considered minor.

Although individual observations for the entire day of dosing were not conducted, this deficiency has no effect on the results of the study and is considered minor.

DATA REVIEW FOR PRIMARY EYE IRRITATION TESTING (§81-4)

CITATION: Wnorowski, G. (1995) Primary eye irritation. Product Safety Labs, East Brunswick, NJ. Laboratory Project Number 3634. May 23, 1995. MRID 43698110.

EXECUTIVE SUMMARY: In a primary eye irritation study (MRID 43698110), 0.1 mL of Ro-Pel Animal, Rodent, and Bird Repellent (0.035% thymol and 0.065% benzyldiethyl[(2,6-xylylcarbamoyl)-methyl]ammonium saccharide) was instilled into the conjunctival sac of one eye of three New Zealand White rabbits/sex. The animals were observed for up to 72 hours following instillation, and eye irritation was scored using the Draize scale.

Slight conjunctival redness was observed in 1/6 treated eyes 1 hour following instillation and in 3/6 eyes at 24 hours. No other ocular effects were observed.

In this study, **Ro-Pel Animal, Rodent, and Bird Repellent is a very slight conjunctival irritant**, and is **TOXICITY CATEGORY IV** for primary eye irritation based on no significant readings at 24 hours.

This study is acceptable, and satisfies the guideline requirement for a primary eye irritation study (81-4) in the rabbit.

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

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test Material: Ro-Pel Animal, Rodent, and Bird Repellent
   Description: Clear liquid
   Lot/Batch #: B2225A
   Purity: 0.035% Thymol and 0.065% benzyldiethyl-[(2,6-xylylcarbamoyl)methyl]ammonium saccharide
   CAS #: 89-83-8 and 90823-38-4, respectively

2. Vehicle and/or positive control: None employed

3. Test animals: Species: Rabbit
   Strain: New Zealand White
   Age: Adult
   Weight: Not specified
   Source: Davidson's Mill Farm, South Brunswick, NJ
   Acclimation period: 6 days
   Diet: Pelleted Purina Rabbit Chow (#5326), unspecified amount/animal/day
   Water: Filtered tap water, ad libitum

B. STUDY DESIGN and METHODS:

1. In-life dates: May 3-6, 1995

2. Animal assignment and treatment: A 0.1-mL aliquot of Ro-Pel Animal, Rodent, and Bird Repellent was instilled into the conjunctival sac of the right eye of three New Zealand White rabbits/sex. The treated eyes were held together for approximately 1 second to prevent loss of material and were not rinsed, and the animals were fitted with Elizabethan collars. The left eye of each animal served as an untreated control. The animals were observed for ocular irritation at 1, 24, 48, and 72 hours following instillation. At 24 hours, fluorescein dye was used to confirm the absence of corneal ulceration. Eye irritation was scored by the Draize scheme. In addition, each animal was observed for signs of toxicity and/or mortality at least once daily during the 3-day study.

II. **RESULTS AND DISCUSSION:**

A. Clinical observations: Slight conjunctival redness (score of 1) was observed in 1/6 treated eyes 1 hour following instillation and in 3/6 eyes at 24 hours. No other ocular effects were observed, and 3/6 treated eyes remained free of irritation during the 72-hour observation period. Based on the results of this study, Ro-Pel Animal, Rodent, and Bird Repellent is a very slight eye irritant.

All animals appeared normal and healthy during the 3-day study.

B. Deficiencies: Although individual observations for the entire day of dosing were not conducted, this deficiency does not alter the results of the study and is considered minor.

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DATA REVIEW FOR PRIMARY DERMAL IRRITATION TESTING (§81-5)

CITATION: Wnorowski, G. (1995) Primary skin irritation. Product Safety Labs, East Brunswick, NJ. Laboratory Project Number 3635. May 23, 1995. MRID 43698111.

EXECUTIVE SUMMARY: In a primary dermal irritation study (MRID 43698111), six adult female New Zealand White rabbits were dermally exposed to 0.5 mL of Ro-Pel Animal, Rodent, and Bird Repellent (0.035% thymol and 0.065% benzyldiethyl[(2,6-xylylcarbamoyl)methyl]ammonium saccharide) for 4 hours. The test substance was applied to a single intact 6-cm^2 site/animal. Animals were observed for dermal irritation for up to 72 hours following application, and irritation was scored by the Draize scheme.

No dermal irritation was observed during the 3-day observation period. In this study, **Ro-Pel Animal, Rodent, and Bird Repellent is not a dermal irritant, and is Toxicity Category IV** for primary dermal irritation.

This study is **acceptable**, and satisfies the guideline requirement for a primary dermal irritation study (81-5) in the rabbit.

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

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test Material: Ro-Pel Animal, Rodent, and Bird Repellent
   Description: Clear liquid
   Lot/Batch #: B2225A
   Purity: 0.035% Thymol and 0.065% benzyldiethyl-[(2,6-xylylcarbamoyl)methyl]ammonium saccharide
   CAS #: 89-83-8 and 90823-38-4, respectively

2. Vehicle: None employed

3. Test animals: Species: Rabbit
   Strain: New Zealand White
   Age: Adult
   Weight: Not specified
   Source: Davidson's Mill Farm, South Brunswick, NJ
   Acclimation period: 6 days
   Diet: Pelleted Purina Rabbit Chow (#5326), unspecified amount/animal/day
   Water: Filtered tap water, ad libitum

B. STUDY DESIGN and METHODS:

1. In-life dates: April 25-28, 1995

2. Animal assignment and treatment: Fur from the dorsal trunk areas of six adult female animals was clipped 1 day prior to dermal administration with 0.5 mL of Ro-Pel Animal, Rodent, and Bird Repellent. The test substance was applied to a single intact 6-cm^2 site/animal using a 6.75-cm^2 adhesive-backed gauze patch. The entire trunk of each animal was wrapped with Micropore tape, and the animals were fitted with Elizabethan collars. The coverings and collars were removed 4 hours following application, and the sites were gently wiped with water and a clean towel. The rabbits were observed for dermal irritation at 1, 24, 48, and 72 hours following patch removal. Erythema and edema were scored separately using the Draize scheme. In addition, signs of toxicity and/or mortality were monitored at least once daily during the 3-day study.

II. **RESULTS AND DISCUSSION:**

A. Clinical observations: No dermal irritation was observed, and all animals appeared normal and healthy during the 3-day observation period. Based on the results of this study, Ro-Pel Animal, Rodent, and Bird Repellent is not a dermal irritant.

B. Deficiencies: Although individual observations for the entire day of dosing were not conducted, this deficiency does not alter the results of the study and is considered minor.

DATA REVIEW FOR DERMAL SENSITIZATION TESTING (§81-6)

CITATION: Wnorowski, G. (1995) Dermal sensitization test - Buehler method. Product Safety Labs, East Brunswick, NJ. Laboratory Project Number 3637. May 26, 1995. MRID 43698112.

EXECUTIVE SUMMARY: In a dermal sensitization study (MRID 43698112) conducted with Ro-Pel Animal, Rodent, and Bird Repellent (0.035% thymol and 0.065% benzyldiethyl[(2,6-xylyl)carbamoyl)methyl]ammonium saccharide), ten young adult Hartley albino guinea pigs were tested using methods based on those derived by Buehler. For the induction phase, application was repeated three times a week for 3 weeks (nine total applications). A concurrent positive control study was conducted in the same manner using 1-chloro-2,4-dinitrobenzene.

No dermal irritation was observed following the single challenge treatment in either previously-induced or naive control animals. Acceptable positive control data were provided to validate the test methodology. Based on the results of this study, **Ro-Pel Animal, Rodent, and Bird Repellent is not a dermal sensitizer.**

This study is **acceptable**, and satisfies the guideline requirement for a dermal sensitization study (81-6) in the guinea pig.

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

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test Material: Ro-Pel Animal, Rodent, and Bird Repellent
   Description: Clear liquid
   Lot/Batch #: B2225A
   Purity: 0.035% Thymol and 0.065% benzyldiethyl-[(2,6-xylylcarbamoyl)methyl]ammonium saccharide
   CAS #: 89-83-8 and 90823-38-4, respectively

2. Vehicle and positive control: No test substance vehicle was employed.

   0.06% 1-Chloro-2,4-dinitrobenzene (DNCB; ≥98% purity) in 80% aqueous ethanol (induction phase) or 0.04% DNCB in acetone (challenge phase) was used for concurrent positive control data.

3. Test animals: Species: Guinea pig
   Strain: Hartley albino
   Age: Young adult
   Weight: 284-355 g males; 297-355 g females
   Source: Davidson's Mill Farms, South Brunswick, NJ

Acclimation period: 5 days
Diet: Pelleted Purina Guinea Pig Chow, unspecified amount/animal/day
Water: Filtered tap water, ad libitum

B. STUDY DESIGN and METHODS:

1. In-life dates: April 17-May 19, 1995

2. Animal assignment and treatment: The study was conducted using methods based on those derived by Buehler [Robinson, M., et al., Toxicology, 61:91-107 (1990); Ritz, H. and E. Buehler, Current Concepts in Cutaneous Toxicity, p. 25-40: Academic Press, NY (1980)]. Based on the results of preliminary testing conducted using either 0.4 mL of Ro-Pel Animal, Rodent, and Bird Repellent as received (100%) or 0.4 mL of 25, 50, or 75% distilled water dilutions, the test substance was used as received for both phases of the definitive study.

For the induction phase, fur on the dorsal and flank areas of ten young adult animals (seven male/three female) was clipped 1 day prior to dermal administration of 0.4 mL of Ro-Pel Animal, Rodent, and Bird Repellent. The test substance was applied to the left side of each animal using an occlusive 25-mm Hill Top Chamber secured with Durapore adhesive tape. An additional ten animals (seven male/three female) were treated in the same manner as described using 0.4 mL of 0.06% DNCB. Following a 6-hour exposure period, the chambers were removed, and any excess test substance was removed from the skin by gently wiping with water and clean towels. Application of the test substance was repeated three times weekly for 3 weeks (nine total applications). For the fourth and seventh DNCB induction treatments, the dosing site was moved to an adjacent area avoid application to necrotic skin.

For the challenge phase, a single treatment was applied using 0.4 mL of either Ro-Pel Animal, Rodent, and Bird Repellent or 0.04% DNCB in acetone in the same manner as described, to the previously untreated right side of each animal 12 days following the final induction treatment. To serve as naive controls, an additional five animals/test substance (three male/two female) were included for the challenge treatment. The guinea pigs were observed for dermal irritation 24 and 48 hours following each induction and challenge exposure. Skin reactions were scored according to the following scale:

0 - No reaction
0.5 - Very faint erythema, usually non-confluent
1 - Faint erythema usually confluent
2 - Moderate erythema
3 - Severe erythema with or without edema

Body weights of each animal were recorded prior to the first induction treatment and on the day following the challenge treatment.

II. **RESULTS AND DISCUSSION:**

A. Induction reactions and duration: No dermal irritation was observed during the induction phase.

B. Challenge reactions and duration: No dermal irritation was observed following the single challenge treatment in either previously-induced or naive control animals. Based on the results of this study, Ro-Pel Animal, Rodent, and Bird Repellent is not a dermal sensitizer.

No treatment-related effect on body weight was observed between test and naive control animals, with overall (approximately 33 days) average increases of 65-74% in males and 56-59% in females.

C. Positive control: Twenty-four hours following the first induction treatment, very faint to faint erythema (scores of 0.5-1) was observed at 6/10 test sites. In general, the incidence and severity of irritation increased with each successive application; 24 hours following the ninth induction application, moderate to severe erythema (scores of 2-3) was observed at all test sites with desquamation at 9/10 sites.

Twenty-four and 48 hours following the single challenge treatment to previously-induced animals, very faint to moderate erythema (scores of 0.5-2) was observed at 9/10 and 10/10 test sites, respectively. In contrast, 24 and 48 hours following treatment to naive control animals, very faint erythema was observed at 1/5 and 0/5 sites, respectively. These data confirm the adequacy of the test species and method employed.

D. Deficiencies: None.