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

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

April 5, 2005

MEMORANDUM

Subject: Acute Toxicity Review for EPA Reg. No.: 3008-ON/ ORD-X372
DP Barcode: D312543

To: Adam Heyward, PM 34 / Renae Whitaker
Regulatory Management Branch
Antimicrobials Division (7510C)

From: Ian Blackwell, Biologist Ian D. Blackwell
Efficacy Evaluation Team
Product Science Branch
Antimicrobials Division (7510C)

Through: Karen Hicks, Team Leader
Chemistry and Toxicology Team
Product Science Branch
Antimicrobials Division (7510C)

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Product Science Branch
Antimicrobials Division (7510C)

Applicant: Osmose, Inc.

FORMULATION FROM LABEL:

Active Ingredient(s):	% by wt.
Copper carbonate	35.37
Didecyl dimethyl ammonium carbonate and didecyl Dimethyl ammonium bicarbonate	12.80
Other Ingredient(s):	51.83
Total:	100%

I BACKGROUND: Osmose, Inc., has submitted a set of five acute toxicity studies to support the registration of their new product, "ORD-X372". The studies were conducted by Product Safety Laboratories.

No primary eye irritation study was included in this submission. The registrant has requested a waiver of the primary eye irritation study as the primary skin irritation study was assigned toxicity category I.

These studies were primarily reviewed by AD/EPA contractor DynCorp/ CSC. A brief secondary was conducted by CTT/PSB/AD.

II RECOMMENDATIONS: PSB findings are:

1. The five submitted studies are acceptable.

2. The waiver of the primary eye irritation study is granted. It is noted that the single subject tested in the primary skin irritation study developed dermal corrosion only a single hour after the four hour exposure period. This very severe irritation (one hour after exposure) is not common. This waiver is in accordance with EPA regulations of 40 CFR §158.340.

The acute toxicity profile for EPA File Symbol 3008-ON is currently:

Study	MRID Number	Toxicity Category	Acceptability
acute oral toxicity	464381-03	III	Acceptable
acute dermal toxicity	464381-04	III	Acceptable
acute inhalation toxicity	464381-05	II	Acceptable
primary eye irritation	none	I	Waived
primary skin irritation	464381-06	I	Acceptable
dermal sensitization	464381-07	Nonsensitizer	Acceptable

III LABELING:

1. The signal word is "DANGER", based upon the results of the primary skin irritation study and the toxicity category assignment of the primary eye irritation study.

2

2. The Precautionary Statements should state:

"Corrosive. Causes irreversible eye damage and skin burns. May be fatal if inhaled. Harmful if swallowed or absorbed through skin. Do not get in eyes, on skin, or, on clothing. Do not breathe vapor or spray mist. Wear goggles, face shield, or, safety glasses. Wear coveralls worn over a long-sleeved shirt, long pants, socks, chemical resistant footwear, and, chemical resistant gloves. Wear a respirator with an organic-vapor removing cartridge with a prefilter approved for pesticides (MSHA/NIOSH approval number prefix TC-23C), or, a NIOSH approved respirator with an organic vapor (OV) cartridge or canister with any N, R, P or HE prefilter. Wash thoroughly with soap and water after handling and before eating, drinking, chewing gum, or, using tobacco. Remove and wash contaminated clothing before reuse."

3. The First Aid statements should state:

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 inhaled:

- Move person to fresh air.
- If person is not breathing, call 911 or an ambulance, then give artificial respiration, preferably mouth-to-mouth, if possible.
- 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 do so by a Poison Control Center or doctor.
- Do not give anything by mouth to an unconscious person.

3

Have the product container or label with you when calling a Poison Control Center or doctor, or, going for treatment.

4. This product satisfies the EPA requirements for Restricted Use pesticides, according to the 40 CFR §152.70. The PM Team should decide if restricted use classification is necessary or if alternative labeling will allay the requirement for restricted use classification.

5. This product also satisfies the EPA requirements for Child-Resistant packaging according the 40 CFR §157.22. The PM Team should decide if Child-Resistant Packaging is necessary or if alternative labeling will allay the requirement for restricted use classification.

DATA REVIEW FOR ACUTE ORAL TOXICITY TESTING (§ 81-1, 870.1100)
(UP AND DOWN PROCEDURE)

Product Manager: 34
MRID No.: 464381-03

Reviewer: Ian Blackwell
Study Completion Date: November 1, 2004
Report No.: 16029

Testing Laboratory: Product Safety Laboratories
Author: Daniel J. Merkel, B.S.

Quality Assurance (40 CFR §160.12): A Quality Assurance statement was included. A statement of Good Laboratory Practice (GLP) compliance also was included stating that the study meets the requirements of 40 CFR Part 160 and OECD specifications, except that specific information related to the stability, characterization, identity and verification of the test substance concentration as received and tested is the responsibility of the study sponsor.

Test Material: ORD-X372 / Lot #FN1-22B / light blue liquid dispersion
Dosage: Main Test: 150, 470, 1500 and 5000 mg/kg (administered as received)
Species: 10 Sprague-Dawley derived, albino rats
Sex: Female - nulliparous and nonpregnant)
Age: Young adult (9 - 12 weeks)
Weight: 171 - 226 grams at experimental start
Source: Ace Animals, Inc., Boyertown, PA
Housing: Temperature Range: 19 - 25 °C
Relative Humidity: information not provided
Photoperiod: 12-hour light/dark cycle
Acclimation: 7 - 28 days

Conclusion:

1. **LD_{50} (mg/kg)**: Females = 1,500 mg/kg
(95 % C.I. 634.7 to 3180 mg/kg)

2. **Tox. Category**: III **Classification**: Acceptable

Procedure (Deviations from §81-1): No deviations were reported by the laboratory. The relative humidity of animal housing was not provided.

Results:

Main Test - Reported Mortality

Dosing Sequence	Animal No.	Dose Level (mg/kg)	Short-Term Outcome	Long-Term Outcome
1	7219	150	S	S
2	7275	470	S	S
3	7303	1,500	S	S
4	7433	5,000	D	D
5	7460	1,500	S	S
6	7481	5,000	D	D
7	7517	1,500	D	D
8	7555	470	S	S
9	7674	1,500	S	D
10	7914	5,000	D	D

S - Survival D - Death

The test substance was administered in sequence as presented above. The decision to proceed with the next animal was based on the survival of the previous animal following dosing.

Observations:

150 mg/kg Dose Level (1 animal): This animal survived, gained body weight, and appeared active and healthy. There were no signs of gross toxicity, adverse clinical signs, or abnormal behavior.

470 mg/kg Dose Level (2 animals): Both animals survived and gained body weight during the study. Following administration, one animal exhibited piloerection and hypoactivity. However, this animal recovered by Day 4, and along with the animal, appeared active and healthy for the remainder of the 14-day observation period.

1,500 mg/kg Dose Level (4 animals): Two animals died within six days of test substance administration. Toxic signs noted prior to death included ano-genital staining, diarrhea, hypoactivity, and distended abdomen. Surviving animals exhibited similar clinical signs as well as facial staining and reduced fecal volume. However, the surviving animals recovered from these symptoms by Day 5 and appeared active and healthy for the remainder of the study, gaining body weight over the entire 14-day observation period.

5,000 mg/kg Dose Level (3 animals): All animals died within one day of test substance administration. Toxic signs noted prior to death included ano-genital staining, diarrhea, hypoactivity, and hunched posture.

Gross Necropsy Findings: No gross abnormalities were noted for any of the euthanized animals dosed at 150, 470 and 1,500 mg/kg dose levels when necropsied at the conclusion of the 14-day observation period. Gross necropsy of the two decedents at the 1,500 mg/kg dose level revealed discoloration of the intestines and lungs, and gaseous distention of the stomach. Gross necropsy of the decedents at the 5,000 mg/kg dose level revealed discoloration of the intestines.

Statistical Analysis: The *Acute Oral Toxicity (Guideline 425) Statistical Program* (Weststat, version 1.0, May 2001) was used for all data analyses including: dose progression selections, stopping criteria determinations and/or LD_{50} and confidence limit calculations.

DATA REVIEW FOR ACUTE DERMAL TOXICITY TESTING (§81-2, 870.1200)
(LIMIT TEST)

Product Manager: 34
MRID No.: 464381-04

Reviewer: Ian Blackwell
Study Completion Date: November 1, 2004
Report No.: 16030

Testing Laboratory: Product Safety Laboratories
Author: Daniel J. Merkel, B.S.

Quality Assurance (40 CFR §160.12): A Quality Assurance statement was provided. A statement of Good Laboratory Practice (GLP) compliance also was included stating that the study meets the requirements of 40 CFR Part 160 and OECD specifications, except that specific information related to the stability, characterization, identity and verification of the test substance concentration as received and tested is the responsibility of the study sponsor.

Test Material: ORD-X372 / Lot #FN1-22B / light blue liquid dispersion

Species: 10 Sprague-Dawley derived, albino rats
Sex: 5 / sex; females were nulliparous and nonpregnant
Age: Young adult (9-10 weeks)
Weight : Males: 292 - 338 grams at experimental start
Females:190 - 228 grams at experimental start
Source: Ace Animals, Inc. Boyertown, PA
Housing: Temperature Range: 19 - 23 °C
Relative Humidity: information not provided
Photoperiod: 12-hour light/dark cycle
Acclimation: 14 days

Summary:

1. **LD_{50} (mg/kg):** Males > 2,000 mg/kg
Females > 2,000 mg/kg
Combined > 2,000 mg/kg

2. **The estimated LD_{50} is** > 2,000 mg/kg

3. **Tox. Category:** III **Classification:** Acceptable

Procedure (Deviations From §81-4): No deviations were reported by the laboratory. The relative humidity of animal housing was not provided.

Results:

Reported Mortality

DOSAGE (mg/kg)	DEATHS / number tested		
	Males	Females	Total
2,000	0 / 5	1 / 5	0 / 10

8

Observations: One female rat died within five days of test substance application. Toxic signs noted prior to death included hunched posture, hypoactivity, and dermal irritation (erythema, edema, and hyperkeratosis) at the dose site. Surviving animals exhibited similar clinical signs as well as irregular respiration, eschar and / or reduced fecal volume. Light blue staining on the dose site was noted for all animals. With the exception of the dermal effects noted above, all survivors recovered by Day 6 and appeared active and healthy, gaining body weight over the 14-day observation period.

Gross necropsy findings: Gross necropsy of the decedent revealed discoloration of the lungs and intestines. No gross abnormalities were noted for any of the surviving animals when necropsied at the conclusion of the 14-day observation period.

DATA REVIEW FOR ACUTE INHALATION TOXICITY TESTING (§81-3, 870.1300)

Product Manager: 34
MRID No.: 464381-05

Reviewer: Ian Blackwell
Study Completion Date: November 1, 2004
Report No.: 16031

Testing Laboratory: Product Safety Laboratories
Author: Daniel J. Merkel, B.S.

Quality Assurance (40 CFR §160.12): A Quality Assurance Statement was provided. A statement of Good Laboratory Practice (GLP) compliance also was included stating that the study meets the requirements of 40 CFR Part 160 and OECD specifications except that specific information related to the stability, characterization, identity and verification of the test substance concentration as received and tested is the responsibility of the study sponsor.

Test Material: ORD-X372 / Lot #FN1-22B / light blue liquid dispersion

Species: 20 Sprague-Dawley derived, albino rats
Sex: 10 / sex (females were nulliparous and nonpregnant)
Age: Young adult (9 - 11 weeks)
Weight: Males: 291 - 381 grams at experimental start
Females: 200 - 248 grams at experimental start
Housing: Temperature Range: 19 - 23 °C
Relative Humidity: information not provided
Photoperiod: 12-hour light/dark cycle
Acclimation: 14 or 20 days
Source: Ace Animals, Inc., Boyertown, PA

Concentration:

Group	Gravimetric Exposure Concentration (mg/L)	Nominal Concentration (mg/L)
I	0.054	9.36
II	0.51	75.24

Summary:

1. **LC_{50} (mg/L) 4-hr exposure:** Males > 0.51 mg/L
Females between 0.054 and 0.51 mg/L
1. **The estimated LC_{50} is > 0.51 mg/L in males and between 0.054 and 0.51 mg/L in females**
3. **MMAD**: 3.4 µm
4. **Tox. Category**: II **Classification**: Acceptable

Procedure (Deviation From §81-3):
Due to a technician error, the clinical observations were inadvertently not recorded one hour after cage removal for Animal #7175 in the 0.5 mg/L exposure level group.

The oxygen content of chamber was not provided. The upper level of relative humidity of the chamber during the 0.054 mg/L exposure level was slightly above the 70% specified in the guidelines. At each level, the exposure period was extended beyond 4 hours to allow the chamber to reach equilibrium (T_{99}).

The relative humidity of caging conditions was not provided.

Results:

Reported Mortality

Exposure Concentration (mg/L)	Number of deaths / number tested		
	Males	Females	Combined
0.054	0 / 5	0 / 5	0 / 10
0.51	1 / 5	3 / 5	4 / 10

Chamber Atmosphere

Exposure conc. (mg/L)	No.	MMAD (µm)	GSD (µm)	% Particles at Effective Cutoff Diameter (Cumulative)							
				0.4 µm	0.7 µm	1.1 µm	2.1 µm	3.3 µm	4.7 µm	5.8 µm	9.0 µm
0.054	1	3.3	2.09	0.0	2.0	8.9	21.8	44.6	73.3	82.2	95.0
	2	3.4	2.31	1.1	3.2	9.5	21.1	43.2	71.6	80.0	92.6
0.51	1	3.4	1.73	0.0	0.3	3.4	11.8	44.6	74.3	85.1	95.6
	2	3.4	1.78	0.0	0.3	3.5	15.3	44.2	72.0	84.4	95.9

Chamber Environment During Exposure

Exposure Level (mg/L)	0.054	0.51
Chamber Volume (L)	6.7	6.7
Airflow (LPM)	25.7	25.7
Temperature (°C)	20 - 21	21
Relative Humidity (%)	57 - 72	58 - 67

Clinical Observations:
0.054 mg/L Exposure Level: All animals survived. Following exposure, one animal exhibited abnormal respiration, hunched posture, and hypoactivity. However, this animal recovered by Day 5, and along with the other animals, appeared active and healthy for the remainder of the 14-day observation period. One male exhibited a

minimal weight loss through Day 7, and one female lost one gram of body weight from Day 7 to Day 14. All animals gained body weight over the entire 14-day observation period.

0.51 mg/L Exposure Level: Two females were found dead following exposure (time after exposure was not specified in report). One male and one female died within two days of exposure to the test atmosphere. Toxic signs noted prior to death included abnormal respiration and posture, hypoactivity, facial staining, and reduced fecal volume. Following exposure (time after exposure was not specified), surviving animals exhibited similar clinical signs. However, these animals recovered from these symptoms by Day 12 and appeared active and healthy for the remainder of the study. Although the surviving animals lost body weight through Day 7, all survivors gained body weight over the 14-day observation period.

Gross Necropsy Findings: No gross abnormalities were noted for any of the animals at the 0.054 mg/L exposure level when necropsied at the conclusion of the 14-day observation period. At the 0.51 mg/L exposure level, gross necropsy of the four decedents revealed discoloration of the lungs, intestines, and/or liver, edema of the lungs, and/or rigor mortis. Gross necropsy of one euthanized animal revealed discoloration of the lungs. No gross abnormalities were noted for the remaining euthanized animals when necropsied at the conclusion of the 14-day observation period.

DATA REVIEW FOR PRIMARY SKIN IRRITATION TESTING (§81-5, 870.2500)

Product Manager: 34
MRID No.: 464381-06

Reviewer: Ian Blackwell
Study Completion Date: November 1, 2004
Report No.: 16105

Testing Laboratory: Product Safety Laboratories
Author: Daniel J. Merkel, B.S.

Quality Assurance (40 CFR §160.12): A Quality Assurance statement was provided. A statement of Good Laboratory Practice (GLP) compliance also was included, stating that the study meets the requirements of 40 CFR Part 160 and OECD specifications, with the following exceptions:

- Specific information related to the stability, characterization, identity and verification of the test substance concentration as received and tested was the responsibility of the study Sponsor.
- Due to the unexpected shortening of the evaluation period, no in-life inspections were performed by The Quality Assurance Unit. This exception did not have any impact on the conduct or interpretation of this study.

Test Material: ORD-X372 / Lot #FN1-22B / light blue, liquid dispersion
Dosage: 0.5 mL - administered as received

Species: 1 New Zealand albino rabbits
Sex: Male
Age: Young adult
Source: Robinson Services, Inc., Clemmons, NC
Housing: Temperature: 19 - 22 °C
Humidity: information not provided
Photoperiod: 12-hour light/dark cycle

Summary:

1. **Toxicity Category**: I / Corrosive
2. **Classification**: Acceptable

Procedure (Deviations From §81-4): No deviations were reported by the laboratory. Relative humidity of animal housing was not reported.

Results: One hour after the removal of the 3-minute, 1-hour, and 4-hour patches, the treated sites exhibited very faint to severe erythema and/or none to slight edema. By 24 hours, dermal corrosion developed at the 1-hour and 4-hour dose sites. Therefore, the study was terminated and the animal was euthanized for humane reasons. The animal appeared active and healthy. Apart from the dermal irritation noted below, there were no other signs of gross toxicity, adverse pharmacologic effects, or abnormal behavior.

Incidence of Irritation

Time after Patch Removal	Erythema	Edema
1 hour	1 / 1	1 / 1
24 hours	1 / 1	1 / 1

One hour after exposure/patch removal, the single treated animal displayed severe erythema, slight edema, and, "dermal corrosion".

DATA REVIEW FOR DERMAL SENSITIZATION TESTING (§81-6, 870.2600)
(BUEHLER METHOD)

Product Manager: 34
MRID No.: 464381-07

Reviewer: Ian Blackwell
Study Completion Date: November 11, 2004
Report No.: 16034

Testing Laboratory: Product Safety Laboratories
Author: Daniel J. Merkel, B.S.

Quality Assurance (40 CFR §160.12): A Quality Assurance statement was included. A statement of Good Laboratory Practice (GLP) compliance also was included stating that the study meets the requirements of 40 CFR Part 160 and OECD specifications, with the following exceptions:

- Specific information related to the stability, characterization identity and verification of the test substance concentration as received and tested is the responsibility of the study sponsor.
- The stability, uniformity of mixture and verification of concentration of alpha-Hexylcinnamaldehyde Technical (HCA) in its carriers during the historical positive control study were not determined.

Test Material: ORD-X372 / Lot #FN1-22B / light blue, liquid dispersion
Positive Control Material: alpha-Hexylcinnamaldehyde Technical (HCA)

Species: 36 Hartley albino guinea pigs
Sex: Male and Female; Females were nulliparous and nonpregnant
Age: Young adult
Weight: Test and naive control group males: 380 to 482 grams at experimental start
Source: Elm Hill Breeding Labs, Chelmsford, MA
Housing: Temperature Range: 18 - 23 °C
Relative Humidity: information not provided
Photoperiod: 12-hour light/dark cycle

Method: Buehler method

Summary:

1. **Based on the results of this study, the test substance is not considered to be a contact sensitizer.** (Study evaluated erythema only.)

2. **Classification**: Acceptable

Procedure (Deviation From §81-6): No deviations were reported by the laboratory. Relative humidity information of animal housing was not provided. Study evaluated erythema only. No information was provided regarding edema.

Procedure:
Preliminary Irritation: Fur was removed by clipping (Oster model #A5-small) the dorsal area and flanks of each guinea pig. This area was divided into four test sites (two sites on each side of the midline) on each animal. The test substance was

applied neat (100%) and also diluted with distilled water to yield w/w concentrations of 75%, 50%, 25%, 6%, 3%, 1%, 0.5%, 0.2%, 0.1%, and 0.05%. Each concentration was applied (0.4 mL each) using an occlusive 25-mm Hill Top Chamber. The sites were wrapped with non-allergenic Durapore adhesive tape. After 6 hours of exposure, the chambers were removed and the test sites were gently cleansed of any residual test substance. Approximately 24 hours after application, each site was evaluated for local reaction (erythema) according to the scoring system.

Induction Phase: Once each week for three weeks, 0.4 mL of a 1% w/w mixture of the test substance in distilled water was applied to the left side of each test animal using an occlusive 25-mm Hill Top Chamber. The chambers were secured in place and wrapped with non-allergenic Durapore adhesive tape. After the 6-hour exposure period, the chambers were removed and the test sites were gently cleansed of any residual test substance. Approximately 24 and 48 hours after each induction application, readings were made of local reactions (erythema only).

Challenge Phase: 27 days after the first induction dose, 0.4 mL of a 0.2% w/w mixture of the test substance in distilled water was applied to a naive site on the right side of each animal, using the procedures described above. These sites were evaluated for a sensitization response (erythema only) approximately 24 and 48 hours after the challenge application.

Results:
Based on the results of this study, the test substance is not considered to be a contact sensitizer.

	Sensitization Response Indices (Erythema)			
	Incidence of Positive Response[1]		**Severity[2]**	
	Hours		**Hours**	
	24	48	24	48
Test Animals	0 / 20	0 / 20	0.18	0.00
Naive Control Animals	0 / 10	0 / 10	0.20	0.00

[1] Animals with scores greater than 0.5
[2] Sum of erythema scores divided by the number of animals evaluated.