TOXIKON FINAL GLP REPORT: 08-2140-G1

CLASS VI TEST – USP

Test Article
Watershed 11122XC

Author
Christopher Parker, M.S.

Final Report Date
June 18, 2008

COMPLIANCE
21 CFR, Part 58
Good Laboratory Practice for Non–Clinical Laboratory Studies

MANAGEMENT OF THE STUDY

Performing Laboratory
Toxikon Corporation
15 Wiggins Avenue
Bedford, MA 01730

Sponsor
DSM Somos
1122 Saint Charles Street
Elgin, IL 60120
TABLE OF CONTENTS

Title Page
Table of Contents
Study Summary
Quality Assurance Statement
Study Director Signature and Verification Dates

1.0 Purpose
2.0 References
3.0 Compliance
4.0 Identification of Test and Control Articles
5.0 Identification of Test System
6.0 Justification of Test System and Route of Administration
7.0 Experimental Design and Dosage
8.0 Evaluation Criteria
9.0 Results
10.0 Conclusion
11.0 Records
12.0 Confidentiality Agreement
13.0 Animal Welfare Statement

Table 1: Systemic Injection Test: Animal Weights and Clinical Observations
Table 2: Intracutaneous Injection and Implant Tests: Animal Weights and Clinical Observations
Table 3: Intracutaneous Test Skin Reaction Scores
Table 4: Implant Test: Macroscopic Observations

Appendix I: Evaluation of Skin Reactions
The USP 0.9% Sodium Chloride for Injection (NaCl), Cottonseed Oil (CSO), 1 in 20 Ethanol in NaCl (EtOH), and Polyethylene Glycol 400 (PEG) extracts of the test article and the test article, Watershed 11122XC, did not produce a biological response following intramuscular implantation in rabbits, intracutaneous injection in rabbits, or systemic injection in mice. Therefore, the test article meets the requirements of the USP guidelines, for Class VI Plastics – 50 °C.
QUALITY ASSURANCE STATEMENT

This study was conducted in compliance with U.S. Food and Drug Administration regulations set forth in 21 CFR, Part 58.

The sections of the regulations not performed by or under the direction of Toxikon Corporation, exempt from this Good Laboratory Practice Statement, included characterization and stability of the test article and its mixture with carriers, 21 CFR, Parts 58.105 and 58.113.

The Quality Assurance Unit conducted inspections on the following dates. The findings were reported to the Study Director and to Toxikon’s Management.

<table>
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<tr>
<th>INSPECTIONS</th>
<th>DATE OF INSPECTION</th>
<th>DATE REPORTED STUDY DIRECTOR</th>
<th>DATE REPORTED MANAGEMENT</th>
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</table>

Melissa Manning, B.S.  
Quality Assurance
STUDY DIRECTOR SIGNATURE AND VERIFICATION DATES

This study meets the technical requirements of the protocol. The study also meets with the requirements of the Good Laboratory Practice Regulations, 21 CFR, Part 58, with the exemptions as stated in the Quality Assurance Statement.

Protocol Number: DMS/VIVO/001-08/000
Study Director: Christopher Parker, M.S.
Company: Toxikon Corporation

Signature: [Signature]
Date: [Date]
Study Supervisor: Michael Fleming, B.A., ALAT

VERIFICATION DATES:
The Study Initiation Date is the date the protocol is signed by the Study Director.

Test Article Receipt: 05/09/08
Project Log Date: 05/09/08
Study Initiation Date: 05/07/08
Extraction Dates: 05/24/08 – 05/27/08
Technical Initiation: 05/27/08
Technical Completion: 06/03/08
1.0 PURPOSE

The purpose of the study was to determine the biological response of animals to direct and indirect contact with the test article or injection of the test article extract.

2.0 REFERENCES

The study was conducted based upon the following references:


3.0 COMPLIANCE

The study conformed to the current FDA 21 CFR, Part 58 – Good Laboratory Practice for Non–Clinical Laboratory Studies.

4.0 IDENTIFICATION OF TEST AND CONTROL ARTICLES

The Sponsor supplied the following information on a test requisition form or other correspondence, wherever applicable (excluding confidential or trade secret information). The Sponsor was responsible for all test article characterization data as specified in the GLP regulations.

4.1 Test Article:

Test Article Name: Watershed 11122XC  
CAS/Code #: Not Supplied by Sponsor (N/S)  
Lot/Batch #: 1300249  
Physical State: N/S  
Color: N/S  
Expiration Date: N/S  
Density: N/S  
Stability: N/S  
Solubility: N/S  
pH: N/S  
Storage Conditions: Room Temperature  
Safety Precautions: Standard Toxikon Laboratory Safety Precautions

4.2 Control Articles (Toxikon Supplied):

4.2.1 Negative Control Article Name: USP 0.9% Sodium Chloride for Injection (NaCl)  
Toxikon QC #: CSC-08-03-010-VV  
Physical State: Liquid
Color: Colorless  
Stability: Stable at Room Temperature  
Storage Conditions: Room Temperature  
Safety Precautions: Standard Laboratory Safety Precautions

4.2.2 Negative Control Article Name: Cottonseed Oil (CSO)  
Toxikon QC #: CSC-08-05-009-VV  
Physical State: Liquid  
Color: Yellow  
Stability: Stable at Room Temperature  
Storage Conditions: Room Temperature  
Safety Precautions: Standard Laboratory Safety Precautions

4.2.3 Negative Control Article Name: 1 in 20 Ethanol in NaCl (EtOH)  
Toxikon QC #: CSC-06-11-007-VV; CSC-08-03-010-VV  
Physical State: Liquid  
Color: Colorless  
Stability: Stable at Room Temperature  
Storage Conditions: Room Temperature  
Safety Precautions: Standard Laboratory Safety Precautions

4.2.4 Negative Control Article Name: Polyethylene Glycol 400 (PEG)  
Toxikon QC #: CSC-06-09-012-VV  
Physical State: Liquid  
Color: Colorless  
Stability: Stable at Room Temperature  
Storage Conditions: Room Temperature  
Safety Precautions: Standard Laboratory Safety Precautions

4.2.5 Negative Control Article Name: Negative Control High Density Polyethylene (Negative Control Plastic)  
Toxikon QC #: CSC-04-05-009-CC  
Physical State: Solid  
Color: White  
Storage Conditions: Room Temperature  
Safety Precautions: Standard Laboratory Safety Precautions

5.0 IDENTIFICATION OF TEST SYSTEM

5.1 Animals Used in the Study:

5.1.1 Systemic Injection Test:

Number and Species: 40 Albino Swiss Mice (*Mus musculus*)

Sex: female (females were non–pregnant and nulliparous)
Weight/Age Range: 17.1 – 22.9 grams / at least 34 days old (adult) 
weighed to the nearest 0.1 g

Health Status: healthy, not previously used in other experimental procedures

Animal Purchase: Harlan, Indianapolis, IN

Animal Identification: ear punch

Acclimation: minimum 3 days, under same conditions as for the actual test

Animal Selection: selected from larger pool and examined to ensure lack of adverse clinical signs

5.1.2 Intracutaneous Injection and Implant Tests:

Number and Species: 6 New Zealand White rabbits (Oryctolagus cuniculus)

Sex: 3 males and 3 females (females were non–pregnant and nulliparous)

Weight/Age Range: 2.36 – 2.70 kilograms for Intracutaneous 
2.78 – 3.09 kilograms for Implant Test 
at least 10 weeks old (young adult) 
weighed to nearest 10 g

Health Status: healthy, Intracutaneous animals not previously used in other experimental procedures and Implant animals previously used in other experimental procedures

Animal Purchase: Millbrook Breeding Labs, Amherst, MA

Animal Identification: ear marker

Acclimation: minimum 3 days, under same conditions as for the actual test

Animal Selection: selected from larger pool and examined to ensure lack of adverse clinical signs

5.2 Animal Care and Maintenance:

5.2.1 Systemic Injection Test:

Animal Room Temperature: 68 ± 5 °F

Animal Room Relative Humidity: 30 – 70%

Air Exchanges per Hour: a minimum of 10 changes per hour

Lights: 12–hour light/dark cycle, full spectrum fluorescent lights

Housing: group housed (5 per cage of same sex)

Cages: polycarbonate
Bedding: hardwood chips, P.W.I. Industries, St–Hyacinthe, Quebec, Canada (contact)

Animal Rations: TEK 7012 Rodent Diet, Harlan Teklad, Madison, WI, ad libitum

Water: tap water, ad libitum

There were no known contaminants present in the feed, water, or bedding expected to interfere with the test data.

The laboratory and animal rooms were maintained as limited-access facilities.

5.2.2 Intracutaneous Injection and Implant Tests:

Animal Room Temperature: 68 ± 5 °F

Animal Room Relative Humidity: 30 – 70%

Air Exchanges per Hour: a minimum of 10 changes per hour

Lights: 12–hour light/dark cycle, full spectrum fluorescent lights

Housing: individually housed

Cages: suspended stainless steel

Bedding: hardwood chips, P.W.I. Industries, St–Hyacinthe, Quebec, Canada (non-contact)

Animal Rations: TEK Hi–Fiber Rabbit Diet 2031, Harlan Teklad, Madison, WI, ad libitum

Water: tap water, ad libitum

There were no known contaminants present in the feed, water, or bedding expected to interfere with the test data.

The laboratory and animal rooms were maintained as limited-access facilities.

6.0 JUSTIFICATION OF TEST SYSTEM AND ROUTE OF ADMINISTRATION

6.1 Albino mice and rabbits were used in this study because they have historically been used in USP Class VI tests and the guidelines have no alternative (non–animal) methods. The species and number of animals used in this study were recommended by the USP guidelines.

6.2 Systemic injection in mice, intracutaneous injection, and intramuscular implantation in rabbits are recommended by the USP guidelines for Class VI tests.

6.3 The test article was exposed to the test system directly and through solvents compatible with the test system.
7.0 EXPERIMENTAL DESIGN AND DOSAGE

7.1 Preparation of Test and Control Articles:

7.1.1 Systemic and Intracutaneous Testing Preparation:

7.1.1.1 The test article (60 cm$^2$) was combined with 10 mL of vehicle at a ratio of 120 cm$^2$ per 20 mL per USP guidelines. The test article was separately extracted in NaCl, CSO, EtOH, and PEG at 50 ± 2 °C for 72 ± 2 hours.

7.1.1.2 Properly prepared test articles were placed in separate extraction bottles, and to each bottle the appropriate medium was added. The extraction medium completely covered the test article.

7.1.1.3 Each extracting medium (control article) was prepared for parallel treatments and comparisons. Each control article was prepared in the same manner as the test article.

7.1.1.4 The Systemic Injection and Intracutaneous tests were performed using the same extracts. The test article appeared unchanged by the extraction procedure. It was not degraded or deformed. The extract was clear and free from particulates. Each extract was agitated vigorously prior to administration. All other test article preparation was as specified by the Sponsor.

7.1.2 Implant Testing Preparation:

The test and control articles were cut into strips measuring 1 mm × 10 mm. The test and control article strips were sterilized by dipping in 70% ethanol prior to implantation.

7.2 Pre-Dose Procedure:

7.2.1 Systemic Injection Test:

7.2.1.1 Acclimated animals were weighed prior to dosing.

7.2.1.2 For the Systemic Injection Test, the PEG test article extract and the corresponding control were diluted with NaCl to obtain PEG concentration of approximately 200 mg/mL.

7.2.2 Intracutaneous Injection Test:

7.2.2.1 On the day of the test, the animals were weighed and clipped free of fur on the dorsal side.

7.2.2.2 For the Intracutaneous Test, the PEG test article extract and the corresponding control were diluted with NaCl to obtain PEG concentration of approximately 120 mg/mL.
7.2.3 Implant Test:
Two rabbits were used for the Implantation Test. On the day of the test, the animals were weighed and the skin on both sides of the spinal column was clipped free of fur. Each animal was anesthetized to prevent muscular movement.

7.3 Dose Administration:

7.3.1 Systemic Injection Test:
Groups of 5 animals were injected with either the test article extract or the corresponding control article extract in the same amounts and by the same routes set forth below:

<table>
<thead>
<tr>
<th>Extract</th>
<th>Route</th>
<th>Dose/kg</th>
<th>Injection Rate</th>
</tr>
</thead>
<tbody>
<tr>
<td>NaCl</td>
<td>Intravenous</td>
<td>50 mL</td>
<td>0.1 mL/second</td>
</tr>
<tr>
<td>CSO</td>
<td>Intraperitoneal</td>
<td>50 mL</td>
<td>___</td>
</tr>
<tr>
<td>EtOH</td>
<td>Intravenous</td>
<td>50 mL</td>
<td>0.1 mL/second</td>
</tr>
<tr>
<td>PEG</td>
<td>Intraperitoneal</td>
<td>10 g</td>
<td>___</td>
</tr>
</tbody>
</table>

7.3.2 Intracutaneous Injection Test:

7.3.2.1 A volume of 0.2 mL of each test article extract was injected intracutaneously at five sites on one side of each of two rabbits. More than one test article extract was used per rabbit.

7.3.2.2 At five sites on the other side of each rabbit, 0.2 mL of the corresponding control article was injected.

7.3.3 Implant Test:
Four samples of the test article were implanted into the paravertebral muscle on one side of the spine of each of two rabbits (2.5 to 5.0 cm from the midline, parallel to the spinal column and about 2.5 cm from each other). In a similar fashion, two strips of the Negative Control Plastic were implanted in the contralateral muscle of each animal.

7.4 Post–Dose Procedure:

7.4.1 Systemic Injection Test:

7.4.1.1 The animals were observed for clinical signs immediately after injection, 4 hours after injection, and at 24, 48, and 72 ± 2 hours after injection. Observations conducted included all clinical and toxicologic signs.

7.4.1.2 The animals were weighed at the end of the observation period.

7.4.1.3 Animals were sacrificed by carbon dioxide inhalation.
7.4.2 Intracutaneous Injection Test:

7.4.2.1 The injection sites on each animal were observed for signs of erythema and edema 24, 48, and 72 hours after injection of the test article. Observations were scored according to the Evaluation of Skin Reactions (Appendix I). Observations conducted also included all clinical signs.

7.4.2.2 All average erythema and edema scores for the test and control sites at 24, 48, and 72 hours were totaled separately and divided by 12 (2 animals × 3 scoring periods × 2 scoring categories) to determine the overall mean score for the test article versus the corresponding control article.

7.4.2.3 Animals were weighed at the end of the observation period.

7.4.2.4 The animals were returned to the general colony.

7.4.3 Implant Test:

7.4.3.1 The animals were maintained for a period of 7 days.

7.4.3.2 The animals were observed daily for this period to ensure proper healing of the implant sites and for clinical signs of toxicity. Observations included all clinical manifestations.

7.4.3.3 At the end of the observation period, the animals were weighed. Each animal was sacrificed by an injectable barbiturate.

7.4.3.4 Sufficient time was allowed to elapse for the tissue to be cut without bleeding.

7.4.3.5 The area of the tissue surrounding the center portion of each implant strip was examined macroscopically using a magnifying lens. Hemorrhaging, necrosis, discolorations, and infections were scored using the following scale:

<table>
<thead>
<tr>
<th>Capsule Width</th>
<th>Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>None</td>
<td>0</td>
</tr>
<tr>
<td>Up to 0.5 mm</td>
<td>1</td>
</tr>
<tr>
<td>0.6 to 1.0 mm</td>
<td>2</td>
</tr>
<tr>
<td>1.1 to 2.0 mm</td>
<td>3</td>
</tr>
<tr>
<td>Greater than 2.0 mm</td>
<td>4</td>
</tr>
</tbody>
</table>

Encapsulation, if present, was scored by first measuring the width of the capsule (the distance from the periphery of the implant to the periphery of the capsule) rounded to the nearest 0.1 mm. The encapsulation was scored as follows:

<table>
<thead>
<tr>
<th>Capsule Width</th>
<th>Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>None</td>
<td>0</td>
</tr>
<tr>
<td>Up to 0.5 mm</td>
<td>1</td>
</tr>
<tr>
<td>0.6 to 1.0 mm</td>
<td>2</td>
</tr>
<tr>
<td>1.1 to 2.0 mm</td>
<td>3</td>
</tr>
<tr>
<td>Greater than 2.0 mm</td>
<td>4</td>
</tr>
</tbody>
</table>
The differences between the average scores for the test article and control article implant sites were calculated.

8.0 EVALUATION CRITERIA

8.1 Systemic Injection Test:
The test is considered negative if none of the animals injected with the test article show a significantly greater biological reaction than the animals treated with the control article.

If two or more mice die, or show signs of toxicity such as convulsions or prostration, or if three or more mice lose more than 2 g of body weight, the test article does not meet the requirements of the test. If any animal treated with a test article shows only slight signs of biological reaction, and not more than one animal shows gross signs of biological reaction or dies, a repeat test is conducted using groups of 10 mice. On the repeat test, all 10 animals must not show a significantly greater biological reaction than the animals treated with the control article.

8.2 Intracutaneous Injection Test:
The requirements of the test are met if the difference between the test article and control article mean reaction scores (erythema/edema) is 1.0 or less.

If at any observation point, the average reaction to the test article sites is questionably greater than the corresponding control article sites, a repeat for the particular test article extract/solution is conducted using an additional 3 rabbits. On the repeat test, the requirements of the test is met if the difference between the test article and control article mean reaction scores (erythema/edema) is 1.0 or less.

8.3 Implant Test:
The test is considered negative if, in each rabbit, the difference between the average scores for each category of biological reaction for the test article and control article implant sites does not exceed 1.0; or if the difference between the mean scores for all categories of biological reaction for each test article and the average score for all categories for all the control implant sites does not exceed 1.0, for not more than one of four test article strips.

8.4 Class VI Requirements:
The test article satisfies the requirements of the USP Class VI test if the requirements described above are met.

8.5 The study and its design employ methodology to minimize uncertainty of measurement and control of bias for data collection and analysis.

9.0 RESULTS

9.1 Systemic Injection Test:

9.1.1 Animal Weights:
All of the test and control animals increased in weight (Table 1).
9.1.2 Clinical Observations:
None of the test or control animals exhibited overt signs of toxicity at any of the observation points (Table 1).

9.1.3 The test is considered negative because none of the animals injected with extracts of the test article showed a significantly greater biological reaction than the animals treated with the control articles.

9.2 Intracutaneous Injection Test:

9.2.1 Animal Weights:
All of the animals increased in weight (Table 2).

9.2.2 Clinical Observations:
There were no overt signs of toxicity observed in any test or control animals (Table 2).

9.2.3 The difference between the test article and control article mean reaction scores (erythema/edema) was less than 1.0. The test article meets the requirements of the Intracutaneous Test (Table 3).

9.3 Implant Test:

9.3.1 Animal Weights:
Both animals increased in weight (Table 2).

9.3.2 Clinical Observations:
There were no overt signs of toxicity noted in either animal. Macroscopic evaluation of the test and control article implant sites showed no significant infection, encapsulation, hemorrhage, necrosis, or discoloration (Tables 2 and 4).

9.3.3 The test is considered negative, since in each rabbit the difference between the average scores for all of the categories of biological reaction for the test article and control article implant sites did not exceed 1.0, and the difference between the mean scores for all categories of biological reaction for all of the test article implant sites and the average score for all categories for all the control implant sites did not exceed 1.0. The test article meets the requirements of the Implantation Test (Table 4).

10.0 CONCLUSION

The USP 0.9% Sodium Chloride for Injection (NaCl), Cottonseed Oil (CSO), 1 in 20 Ethanol in NaCl (EtOH), and Polyethylene Glycol 400 (PEG) extracts of the test article and the test article, Watershed 11122XC, did not produce a biological response following intramuscular implantation in rabbits, intracutaneous injection in rabbits, or systemic injection in mice. Therefore, the test article meets the requirements of the USP guidelines, for Class VI Plastics – 50 °C.
11.0 RECORDS

11.1 Original raw data are archived at Toxikon Corporation.

11.2 A copy of the final report and any report amendments is archived at Toxikon Corporation.

11.3 The original final report, and a copy of any protocol amendments or deviations, is forwarded to the Sponsor.

11.4 All used and unused test article shall be disposed of by Toxikon, per Sponsor’s request.

12.0 CONFIDENTIALITY AGREEMENT

Statements of confidentiality were not agreed upon prior to study initiation.

13.0 ANIMAL WELFARE STATEMENT

The Sponsor assured that, to the best of their knowledge, this study did not unnecessarily duplicate previous testing and that there were no non–animal alternatives acceptable for the evaluation of this test article as defined by the protocol.

No evidence of pain and suffering was reported to the Veterinarian and/or Study Director.

Toxikon strictly adhered to the following standards in maintaining the animal care and use program:

- Association for the Assessment and Accreditation of Laboratory Animal Care (AAALAC) International.
## TABLE 1
**Systemic Injection Test:**
*Animal Weights and Clinical Observations*

**Test Article:** Watershed 11122XC  
**Lot/Batch #:** 1300249

<table>
<thead>
<tr>
<th>Group</th>
<th>Animal #</th>
<th>Sex</th>
<th>Dose (mL)</th>
<th>Body Weight (g)</th>
<th>Signs of Toxicity*</th>
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<tr>
<td></td>
<td></td>
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<td>Day 0</td>
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<td>05/30/08</td>
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<td>Female</td>
<td>1.1</td>
<td>21.5</td>
<td>24.2</td>
</tr>
<tr>
<td>50 mL/kg</td>
<td>2</td>
<td>Female</td>
<td>1.0</td>
<td>19.0</td>
<td>21.4</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>Female</td>
<td>1.1</td>
<td>22.7</td>
<td>25.2</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>Female</td>
<td>1.1</td>
<td>22.8</td>
<td>24.3</td>
</tr>
<tr>
<td></td>
<td>5</td>
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<td>1.0</td>
<td>20.5</td>
<td>22.0</td>
</tr>
<tr>
<td>50 mL/kg</td>
<td>7</td>
<td>Female</td>
<td>1.1</td>
<td>21.1</td>
<td>22.5</td>
</tr>
<tr>
<td></td>
<td>8</td>
<td>Female</td>
<td>1.0</td>
<td>19.4</td>
<td>21.7</td>
</tr>
<tr>
<td></td>
<td>9</td>
<td>Female</td>
<td>1.1</td>
<td>22.6</td>
<td>25.3</td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>Female</td>
<td>1.1</td>
<td>21.9</td>
<td>24.4</td>
</tr>
<tr>
<td>CSO Test</td>
<td>11</td>
<td>Female</td>
<td>1.1</td>
<td>22.8</td>
<td>25.2</td>
</tr>
<tr>
<td>50 mL/kg</td>
<td>12</td>
<td>Female</td>
<td>1.1</td>
<td>21.1</td>
<td>23.5</td>
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<tr>
<td></td>
<td>13</td>
<td>Female</td>
<td>1.1</td>
<td>21.5</td>
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<td>14</td>
<td>Female</td>
<td>0.9</td>
<td>18.3</td>
<td>19.7</td>
</tr>
<tr>
<td></td>
<td>15</td>
<td>Female</td>
<td>0.9</td>
<td>18.7</td>
<td>20.3</td>
</tr>
<tr>
<td>CSO Control</td>
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<td>1.0</td>
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<td>21.2</td>
</tr>
<tr>
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<td>1.1</td>
<td>22.9</td>
<td>25.6</td>
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<td></td>
<td>18</td>
<td>Female</td>
<td>0.9</td>
<td>18.3</td>
<td>20.7</td>
</tr>
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<td></td>
<td>19</td>
<td>Female</td>
<td>1.0</td>
<td>19.3</td>
<td>21.7</td>
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<td></td>
<td>20</td>
<td>Female</td>
<td>0.9</td>
<td>18.8</td>
<td>21.4</td>
</tr>
<tr>
<td>EtOH Test</td>
<td>21</td>
<td>Female</td>
<td>0.9</td>
<td>17.1</td>
<td>19.8</td>
</tr>
<tr>
<td>50 mL/kg</td>
<td>22</td>
<td>Female</td>
<td>0.9</td>
<td>17.3</td>
<td>19.0</td>
</tr>
<tr>
<td></td>
<td>23</td>
<td>Female</td>
<td>1.0</td>
<td>19.3</td>
<td>22.0</td>
</tr>
<tr>
<td></td>
<td>24</td>
<td>Female</td>
<td>1.1</td>
<td>21.0</td>
<td>22.7</td>
</tr>
<tr>
<td></td>
<td>25</td>
<td>Female</td>
<td>1.1</td>
<td>21.3</td>
<td>23.5</td>
</tr>
<tr>
<td>EtOH Control</td>
<td>26</td>
<td>Female</td>
<td>1.0</td>
<td>20.9</td>
<td>23.3</td>
</tr>
<tr>
<td>50 mL/kg</td>
<td>27</td>
<td>Female</td>
<td>1.1</td>
<td>22.8</td>
<td>24.7</td>
</tr>
<tr>
<td></td>
<td>28</td>
<td>Female</td>
<td>1.1</td>
<td>21.3</td>
<td>23.8</td>
</tr>
<tr>
<td></td>
<td>29</td>
<td>Female</td>
<td>0.9</td>
<td>18.4</td>
<td>19.7</td>
</tr>
<tr>
<td></td>
<td>30</td>
<td>Female</td>
<td>1.0</td>
<td>20.3</td>
<td>22.5</td>
</tr>
<tr>
<td>PEG Test</td>
<td>31</td>
<td>Female</td>
<td>1.0</td>
<td>19.7</td>
<td>22.3</td>
</tr>
<tr>
<td>10 g/kg</td>
<td>32</td>
<td>Female</td>
<td>0.9</td>
<td>17.1</td>
<td>19.6</td>
</tr>
<tr>
<td></td>
<td>33</td>
<td>Female</td>
<td>1.0</td>
<td>19.5</td>
<td>21.6</td>
</tr>
<tr>
<td></td>
<td>34</td>
<td>Female</td>
<td>1.0</td>
<td>20.6</td>
<td>22.9</td>
</tr>
<tr>
<td></td>
<td>35</td>
<td>Female</td>
<td>1.1</td>
<td>22.8</td>
<td>25.3</td>
</tr>
<tr>
<td>PEG Control</td>
<td>36</td>
<td>Female</td>
<td>1.0</td>
<td>20.3</td>
<td>22.6</td>
</tr>
<tr>
<td>10 g/kg</td>
<td>37</td>
<td>Female</td>
<td>1.0</td>
<td>20.8</td>
<td>22.6</td>
</tr>
<tr>
<td></td>
<td>38</td>
<td>Female</td>
<td>1.1</td>
<td>22.2</td>
<td>24.8</td>
</tr>
<tr>
<td></td>
<td>39</td>
<td>Female</td>
<td>0.9</td>
<td>17.4</td>
<td>19.3</td>
</tr>
<tr>
<td></td>
<td>40</td>
<td>Female</td>
<td>1.0</td>
<td>20.2</td>
<td>21.6</td>
</tr>
</tbody>
</table>

* Summary of clinical observations - Immediately, 4, 24, 48, and 72 h after injection.
TABLE 2
Intracutaneous Injection and Implant Tests:
Animal Weights and Clinical Observations

Test Article: Watershed 11122XC
Lot/Batch #: 1300249

<table>
<thead>
<tr>
<th>Group</th>
<th>Animal #</th>
<th>Sex</th>
<th>Body Weight (kg) Day 0 05/27/08</th>
<th>Body Weight (kg) Day 3 05/30/08</th>
<th>Weight Change</th>
<th>Signs of Toxicity*</th>
</tr>
</thead>
<tbody>
<tr>
<td>NaCl &amp; CSO</td>
<td>80904</td>
<td>Female</td>
<td>2.42</td>
<td>2.66</td>
<td>0.24</td>
<td>None</td>
</tr>
<tr>
<td></td>
<td>80905</td>
<td>Male</td>
<td>2.61</td>
<td>2.76</td>
<td>0.15</td>
<td>None</td>
</tr>
<tr>
<td>EtOH &amp; PEG</td>
<td>80926</td>
<td>Female</td>
<td>2.36</td>
<td>2.51</td>
<td>0.15</td>
<td>None</td>
</tr>
<tr>
<td></td>
<td>80927</td>
<td>Male</td>
<td>2.70</td>
<td>2.87</td>
<td>0.17</td>
<td>None</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Group</th>
<th>Animal #</th>
<th>Sex</th>
<th>Body Weight (kg) Day 0 05/27/08</th>
<th>Body Weight (kg) Day 7 06/03/08</th>
<th>Weight Change</th>
<th>Signs of Toxicity*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Implant</td>
<td>80727</td>
<td>Male</td>
<td>2.78</td>
<td>2.93</td>
<td>0.15</td>
<td>None</td>
</tr>
<tr>
<td></td>
<td>80728</td>
<td>Female</td>
<td>3.09</td>
<td>3.34</td>
<td>0.25</td>
<td>None</td>
</tr>
</tbody>
</table>

* Summary of Clinical Observations, Day 0 through Day 3, excluding skin reactions for the Intracutaneous Injection Test and Day 0 through Day 7 for the Implant Test.
TABLE 3
Intracutaneous Test Skin Reaction Scores

Test Article: Watershed 11122XC

Lot/Batch #: 1300249

NaCl Extract

<table>
<thead>
<tr>
<th>Animal #</th>
<th>Vehicle</th>
<th>Time</th>
<th>Site Numbers Scoring (ER/ED)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>T−1</td>
</tr>
<tr>
<td>80904</td>
<td>NaCl</td>
<td>24 hours</td>
<td>0/0</td>
</tr>
<tr>
<td></td>
<td>NaCl</td>
<td>48 hours</td>
<td>0/0</td>
</tr>
<tr>
<td></td>
<td>NaCl</td>
<td>72 hours</td>
<td>0/0</td>
</tr>
<tr>
<td>80905</td>
<td>NaCl</td>
<td>24 hours</td>
<td>0/0</td>
</tr>
<tr>
<td></td>
<td>NaCl</td>
<td>48 hours</td>
<td>0/0</td>
</tr>
<tr>
<td></td>
<td>NaCl</td>
<td>72 hours</td>
<td>0/0</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td></td>
<td>0.0</td>
</tr>
</tbody>
</table>

Overall Mean Score* for Test Article = 0.0
Overall Mean Score* for Control Article = 0.0
Difference between Test Article and Control Article Overall Mean Score = 0.0–0.0 = 0.0

CSO Extract

<table>
<thead>
<tr>
<th>Animal #</th>
<th>Vehicle</th>
<th>Time</th>
<th>Site Numbers Scoring (ER/ED)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>T−1</td>
</tr>
<tr>
<td>80904</td>
<td>CSO</td>
<td>24 hours</td>
<td>0/0</td>
</tr>
<tr>
<td></td>
<td>CSO</td>
<td>48 hours</td>
<td>0/0</td>
</tr>
<tr>
<td></td>
<td>CSO</td>
<td>72 hours</td>
<td>0/0</td>
</tr>
<tr>
<td>80905</td>
<td>CSO</td>
<td>24 hours</td>
<td>0/0</td>
</tr>
<tr>
<td></td>
<td>CSO</td>
<td>48 hours</td>
<td>0/0</td>
</tr>
<tr>
<td></td>
<td>CSO</td>
<td>72 hours</td>
<td>0/0</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td></td>
<td>0.0</td>
</tr>
</tbody>
</table>

Overall Mean Score* for Test Article = 0.0
Overall Mean Score* for Control Article = 0.0
Difference between Test Article and Control Article Overall Mean Score = 0.0–0.0 = 0.0

ER = Erythema; ED = Edema; T = Test Sites; C = Control Sites
* Overall Mean Score = Total erythema plus edema scores divided by 12
  (2 animals × 3 scoring periods × 2 scoring categories)

Page 18 of 21
### TABLE 3
Intracutaneous Test Skin Reaction Scores (Cont.)

**Test Article: Watershed 11122XC**

**Lot/Batch #:** 1300249

<table>
<thead>
<tr>
<th>Vehicle</th>
<th>Time</th>
<th>Site Numbers Scoring (ER/ED)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>T−1</td>
</tr>
<tr>
<td>EtOH</td>
<td>24 hours</td>
<td>0/0</td>
</tr>
<tr>
<td></td>
<td>48 hours</td>
<td>0/0</td>
</tr>
<tr>
<td></td>
<td>72 hours</td>
<td>0/0</td>
</tr>
<tr>
<td>PEG</td>
<td>24 hours</td>
<td>0/0</td>
</tr>
<tr>
<td></td>
<td>48 hours</td>
<td>0/0</td>
</tr>
<tr>
<td></td>
<td>72 hours</td>
<td>0/0</td>
</tr>
</tbody>
</table>

Overall Mean Score* for Test Article = 0.0

Overall Mean Score* for Control Article = 0.0

Difference between Test Article and Control Article Overall Mean Score = 0.0–0.0 = 0.0

---

ER = Erythema; ED = Edema; T = Test Sites; C = Control Sites

* Overall Mean Score = Total erythema plus edema scores divided by 12
  (2 animals × 3 scoring periods × 2 scoring categories)
TABLE 4
Implant Test:
Macroscopic Observations

Test Article: Watershed 11122XC

Lot/Batch #: 1300249

Animal #: 80727

<table>
<thead>
<tr>
<th>Tissue Site</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>Test Average</th>
<th>C1</th>
<th>C2</th>
<th>Control Average</th>
</tr>
</thead>
<tbody>
<tr>
<td>Infection</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Encapsulation</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Hemorrhage</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Necrosis</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Discoloration</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Total</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Mean Score (total/5)</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

Animal #: 80728

<table>
<thead>
<tr>
<th>Tissue Site</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>Test Average</th>
<th>C1</th>
<th>C2</th>
<th>Control Average</th>
</tr>
</thead>
<tbody>
<tr>
<td>Infection</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Encapsulation</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Hemorrhage</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Necrosis</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Discoloration</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Total</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Mean Score (total/5)</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

T = Test
C = Control
APPENDIX I
Evaluation of Skin Reactions

**Erythema and Eschar Formation**

<table>
<thead>
<tr>
<th>Description</th>
<th>Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>No erythema</td>
<td>0</td>
</tr>
<tr>
<td>Very slight erythema (barely perceptible)</td>
<td>1</td>
</tr>
<tr>
<td>Well–defined erythema</td>
<td>2</td>
</tr>
<tr>
<td>Moderate to severe erythema</td>
<td>3</td>
</tr>
<tr>
<td>Severe erythema (beet redness) to slight eschar formation (injuries in depth)</td>
<td>4</td>
</tr>
</tbody>
</table>

Total possible erythema score = 4

---

**Edema Formation**

<table>
<thead>
<tr>
<th>Description</th>
<th>Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>No edema</td>
<td>0</td>
</tr>
<tr>
<td>Very slight edema (barely perceptible)</td>
<td>1</td>
</tr>
<tr>
<td>Slight edema (edges of area well–defined by definite raising)</td>
<td>2</td>
</tr>
<tr>
<td>Moderate edema (raised approximately 1 mm)</td>
<td>3</td>
</tr>
<tr>
<td>Severe edema (raised more than 1 mm and extending beyond area of exposure)</td>
<td>4</td>
</tr>
</tbody>
</table>

Total possible edema score = 4

* Excludes non–inflammatory (mechanical) edema from the blank or extract fluid.