ViriMASK, Ltd. Document Control

18 May 2020

Document:

Release of Animal Irritation Test - ISO (GLP) Nelson Laboratories & American Preclinical Services.

Purpose:

The purpose of this study was to evaluate the irritation potential of a test article. This test was used as a method for screening topical irritants in rabbits. The results are used as predictive measures for detecting potential irritants in humans.

Summary:

The study was conducted in compliance to the International Organization for Standardization (ISO), 10993-10: 2010 and British Standard European Norm ISO (BS EN ISO) 10993-10: 2013 (Biological Evaluation of Medical Devices, Part 10: Tests for Irritation and Skin Sensitization).

A total of 3 animals were used. 4-24 hours prior to testing, the hair on the back of the animals was closely clipped, allowing a sufficient distance on both sides of the spine for the application of test article and negative control. The test article was moistened and dosed neat by direct application to the skin, and the sites were covered with a 2.5 cm X 2.5 cm non-occlusive dressing (such as absorbent gauze). A negative control dressing was also applied to the skin after being moistened with water. The application sites were wrapped with a bandage (semi-occlusive) for a minimum of 4 hours. At the end of the contact duration, the dressings were removed and the sites where the test article and negative control patches were located were marked with permanent ink. When appropriate, residual test article was removed by washing with lukewarm water or other suitable non-irritating solvent, followed by careful drying. Observations were performed on each application site 1 ± 0.1, 24 ± 2,48 ± 2, and 72 ± 2 hours after unwrap. The tissue reactions were graded for erythema and edema for each application site at each time interval. The 1 ± 0.1 hour observation period was performed to ensure there were no adverse clinical signs for the animals, so the tissue grades each animal received during that time point was not included in the calculations of the primary irritation score or primary irritation index.

Results:

To obtain the primary irritation index, after the 72 ± 2 hours scoring, all erythema grades plus edema grades were totaled separately for each test article and negative control for each animal. The primary irritation score for an animal was calculated by dividing the sum of all the scores by 6 (two test/observation sites, three time points [24 ± 2,48 ± 2, and 72 ± hours]). The primary irritation index was obtained for the test article by adding all the primary irritation scores of the individual animals and dividing by the number of animals. The primary irritation score for the negative control(s) was calculated and subtracted from the test article score to obtain the primary irritation index.

The test article Primary Irritation Index was 0.0.

Nelson Labs™
A Sotera Health company

Sponsor:
Larry Murdock
OHK Medical Devices, Inc.
2885 Sanford Ave.
SW #14751
Grandville, MI 49418

Animal Irritation Test – ISO (GLP)

Test Article:	NZ-0100-PATCH
Purchase Order:	PO VM1886A
Study Number:	1289100-S01
Study Received Date:	15 Apr 2020
Testing Facility:	American Preclinical Services
Deviations:	None

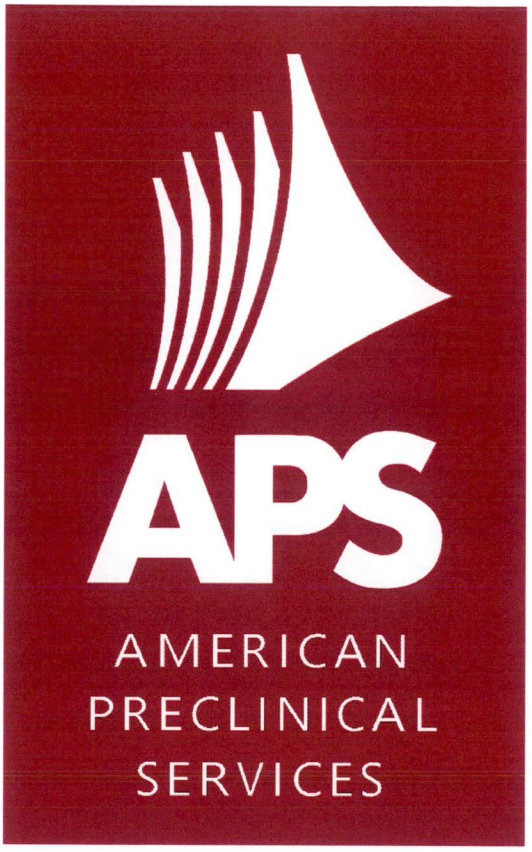
Summary: Enclosed is the final report for the testing we coordinated for you. The information is retained by the testing laboratory.

If you have any questions, please feel free to call or email any of our Subcontracting personnel at 801-290-7500 or subcontracting@nelsonlabs.com. Thank you for testing with Nelson Laboratories, LLC.

Toxicologist — Trevor Fish, M.S.

15 May 2020
Study Completion Date

1289100-S01



Animal Irritation Test (ISO) – Final Report

APS Study ID	PRF1127-IR11
APS Project ID	APS20-0897
Sponsor Study ID	1289100
Compliance	GLP (21 CFR Part 58)

1 SIGNATURES

Name, Study Role	Signature	Date
Benjamin Vos, BS APS Study Director		05-15-2020
Dr. Emily Drake, DVM Laboratory Management		05.15.2020

2 QUALITY ASSURANCE STATEMENT

Inspection Date	Phase Inspected	Date Findings Reported to Study Director and Management
28Apr2020	Sample Preparation	28Apr2020
12May2020	Final Report	14May2020

The above phases of this study were inspected in accordance with 21 CFR 58 by a representative of the American Preclinical Services Quality Assurance Unit. Findings were reported to the Study Director and Testing Facility Management on the dates indicated.

Name, Study Role	Signature	Date
Nick McCune, APS Lead Quality Assurance Auditor		15May2020

3 TABLE OF CONTENTS

3.1 TABLE OF FIGURES AND TABLES

3.2 TABLE OF APPENDICES

4 ABBREVIATIONS

Table 1. Standard Abbreviations

Abbreviation	Name
AAALAC	Association for Assessment and Accreditation of Laboratory Animal Care International
ANAB	ANSI National Accreditation Board
APS	American Preclinical Services, LLC
BID	Twice Daily
CM	Castrated Male
D or d	Day
EPA	Environmental Protection Agency
F	Intact Female
FDA	Food and Drug Administration
Freq	Frequency
GLP	Good Laboratory Practices
IA	Intra-arterial Administration
IACUC	Institutional Animal Care and Use Committee
ID	Intradermal
IM	Intramuscular
Inhal	Inhalation
IP	Intraperitoneal
IV	Intravenous
M	Intact Male
NA	Not Applicable
PHS	Public Health Service
PO	Oral
PRN	As Needed
Q or q	Every
QAU	Quality Assurance Unit
QID	Four Times Daily
QW	Once a Week
SID	Once Daily
SOP	Standard Operating Procedure
SQ	Subcutaneous
TID	Three Times Daily
USDA	United States Department of Agriculture

Table 2. Study Specific Abbreviations

Abbreviation	Name
BS EN	British Standard European Norm
E	Erythema (redness)
ISO	International Organization for Standardization
NS	Normal Saline
S	Edema (swelling)
SPA	Study Protocol Appendix
SSO	Sesame Seed Oil
Wt	Weight

5 EXECUTIVE SUMMARY

5.1 PURPOSE / OBJECTIVE

The purpose of this study was to evaluate the irritation potential of a test article. This test was used as a method for screening topical irritants in rabbits. The results are used as predictive measures for detecting potential irritants in humans.

5.2 ISO COMPLIANCE

The study was conducted in compliance to the International Organization for Standardization (ISO) 10993-10: 2010 and British Standard European Norm ISO (BS EN ISO) 10993-10: 2013 (Biological Evaluation of Medical Devices, Part 10: Tests for Irritation and Skin Sensitization). The study articles were prepared in compliance to the ISO 10993-12: 2012 and BS EN ISO 10993-12: 2012 (Biological Evaluation of Medical Devices, Part 12: Sample Preparation and Reference Materials).

5.3 TEST/CONTROL ARTICLE

NZ-0100-PATCH

5.4 STUDY OUTCOME

Completed (Negligible Irritant)

See section 9.2 Final Result for details.

6 GENERAL STUDY INFORMATION

6.1 STUDY INITIATION AND COMPLETION DATES

Date of Protocol Approval	See the Protocol (Appendix B)
Date of Study Completion	See Section 1 Signatures

6.2 STUDY TITLE

Animal Irritation Test (ISO)

6.3 STUDY COMPLIANCE

6.3.1 GLP Compliance Statement

This study was conducted in compliance to the FDA GLP Regulations, 21 CFR Part 58 (FDA).

6.3.2 ISO Compliance

The study was conducted in compliance to the International Organization for Standardization (ISO) 10993-10: 2010 and British Standard European Norm ISO (BS EN ISO) 10993-10: 2013 (Biological Evaluation of Medical Devices, Part 10: Tests for Irritation and Skin Sensitization). The study articles were prepared in compliance to the ISO 10993-12: 2012 and BS EN ISO 10993-12: 2012 (Biological Evaluation of Medical Devices, Part 12: Sample Preparation and Reference Materials).

6.3.3 Protocol Changes

Changes, if any, to the approved protocol are listed in Appendix C.

6.3.4 Deviations

There were no deviations that affected the quality or integrity of the data.

6.4 STUDY FACILITIES

Test Facility (Corporate Headquarters)	American Preclinical Services, LLC (APS) 8945 Evergreen Boulevard NW Minneapolis, MN 55433 763-717-7990 office 763-717-2042 fax
Study Sponsor	Nelson Laboratories 6820 S. Redwood Rd Salt Lake City, UT 84123 801-290-7500

6.5 STUDY PERSONNEL

Test Facility Management	Michael Conforti, DVM, MS, MBA 8945 Evergreen Boulevard NW Minneapolis, MN 55433 763-951-8030 phone mconforti@apsemail.com	
Study Sponsor Representative	Trevor Fish 6820 S. Redwood Rd. Salt Lake City, UT 84123 801-290-7500	
Study Director	See Section 1 Signatures	
Supervisory Personnel	Manager, In-Vitro Testing	Abby Beltrame, BS
	Supervisor, Sample Preparation	André Nelson, BS
	Director, Quality Assurance	Tammy Fossum, BA
	Director, Laboratory Services	Sarah Howard, BA
	Chief Scientific Officer	Jim Pomonis, PhD
	Senior Director, Scientific	Adam Blakstvedt, BS
	Senior Manager, Scientific	Heather Ackerson, MS
	Manager, Small Animal–Animal Care	Elizabeth Clark, BS, LAT
	Manager, Small Animal Operations	Jaimie McKenzie, BS, RLAT
	Manager, Pathology Services	Eric DoBrava
	Supervisor, Necropsy Operations	Rob Hanson, BS, CVT
	Chief Operations Officer	Erik Steinmetz, BA
	Chief Veterinary Officer	Emily Drake, DVM
	Senior Director, Biocompatibility	Yan Chen, PhD

Lead QAU

APS QAU
8945 Evergreen Blvd. NW
Minneapolis, MN 55433
763-717-7990

6.6 PURPOSE / OBJECTIVES

The purpose of this study was to evaluate the irritation potential of a test article. This test was used as a method for screening topical irritants in rabbits. The results are used as predictive measures for detecting potential irritants in humans.

6.7 ENDPOINTS

Endpoint 1 **Tissue reaction grades**
Erythema and edema were scored at 1 ± 0.1, 24 ± 2, 48 ± 2, and 72 ± 2 hours post test and control article contact duration. The grading system for intradermal reactions was based on a 0 to 4 scale.

Endpoint 2 **Primary irritation index**
Evaluation of results via tissue reaction grading at the 24 ± 2, 48 ± 2, and 72 ± 2 hours post test and control article contact duration. A primary irritation index was characterized by number (tissue grades) and description (response category) of the test article and negative control treatment sites.

Endpoint 3 **Overall animal health (moribundity)**
Clinical observations were conducted daily by trained personnel to assess the overall health of the animal.

6.8 ASSAY VALIDITY CRITERIA

Validity Criteria 1 The animals survived the in-life duration of the study and appeared outwardly healthy throughout the study duration.

Validity Criteria 2 No sign of infection associated with any of the application sites on the animals.

Validity Criteria 3 The negative control scores were within acceptable values.

6.9 STUDY ARTICLES

6.9.1 Characterization

Test Article characterization and stability testing under the conditions of administration will be the responsibility of and maintained by the Sponsor. The Sponsor will assure traceability of the test article characterization and stability data to a known identification number such as a lot, batch, or serial number. The control articles were used from a commercial lot and were characterized by the packaging label or serial number as appropriate.

6.9.2 Test Article 01

Test Article Name	NZ-0100-PATCH
Identification	Lot #: NZ-0100-Patch
Sterilization	Study Article not sterilized (non-sterile)
Expiration Date	Currently Undetermined
Storage Conditions	Room Temperature

6.9.3 Control Article 01

Control Article Name	Negative Control: Gauze
Manufacturer	McKesson
Identification	Lot #: CAD08-02
Sterilization	Not Applicable
Expiration Date	Not Applicable
Storage Conditions	Ambient

7 STUDY SUMMARY

7.1 STUDY DESIGN GENERAL INFORMATION

7.1.1 Summary

A total of 3 animals were used. 4 – 24 hours prior to testing, the hair on the back of the animals was closely clipped, allowing a sufficient distance on both sides of the spine for the application of test article and negative control. The test article was moistened and dosed neat by direct application to the skin, and the sites were covered with a 2.5 cm X 2.5 cm non-occlusive dressing (such as absorbent gauze). A negative control dressing was also applied to the skin after being moistened with water. The application sites were wrapped with a bandage (semi-occlusive) for a minimum of 4 hours. At the end of the contact duration, the dressings were removed and the sites where the test article and negative control patches were located were marked with permanent ink. When appropriate, residual test article was removed by washing with lukewarm water or other suitable non-irritating solvent, followed by careful drying. Observations were performed on each application site 1 ± 0.1, 24 ± 2, 48 ± 2, and 72 ± 2 hours after unwrap. The tissue reactions were graded for erythema and edema for each application site at each time interval. The 1 ± 0.1 hour observation period was performed to ensure there were no adverse clinical signs for the animals, so the tissue grades each animal received during that time point was not included in the calculations of the primary irritation score or primary irritation index.

7.1.2 Control of Bias

Animals were selected from a stock pool of Test Facility animals. All animals selected for this study were naïve and observed prior to entry into the study to ensure that the animals were not exhibiting any signs of clinically relevant abnormalities.

7.1.3 Adverse Events

There were no adverse events on this study.

7.1.4 Exclusions

There were no exclusions for this study.

8 STUDY RESULTS

8.1 SAMPLE PREPARATION

The test article was prepared per the SPA. The control articles were prepared per the protocol, and were tested in parallel with the test article. 2.5 cm x 2.5 cm pieces were prepared for testing.

8.2 SAMPLE EXTRACTION

The test article and negative vehicle controls were dosed neat. The study articles were stored at room temperature.

8.3 TEST SYSTEMS

Test System Number	01
Species	Rabbit
Breed	New Zealand White
Sex	Male
Test System ID	01: Rabbit – New Zealand White - Male
Animal Source	Robinson Services
Total # of Animals	3
Age at Initial Procedure	Age appropriate to weight
Wt. Range at Initial Procedure	2.6 – 2.8 kg
Identification	Per APS SOP.

8.4 GROUP DESIGNS

Test System ID: Species: Sex	Group ID: Study Article	# of Animals	Procedure Type(s)	Procedure Timing, if applicable, and/or Termination Timing, if applicable
TS01: Rabbit: Male	GRP01: Test and Control	3 per neat dose	Application	Day 0
			Scoring	1 ± 0.1 hour, 24 ± 2 hours, 48 ± 2 hours, and 72 ± 2 hours after completion of application duration
			Termination	After 72 ± 2 scoring

8.5 OBSERVATIONS

8.5.1 Clinical Observations

All animals survived the study. All animals had intact healthy skin at the start of the study and there were no signs of infection associated with any of the application sites. All animals were in overall good health over the course of the study.

8.5.2 Gross Pathology Observations

There were no post mortem observations from unscheduled deaths, as all animals survived the study.

8.6 BODY WEIGHTS

All animals weighed ≥ 2.0 kg at the start of the study.

Table 3. Bodyweights

Animal Number	Weight (kg)
612760	2.8
612761	2.6
612762	2.8

8.7 TREATMENT SITE GRADES

8.7.1 Evaluation

The skin reactions were graded for erythema and edema according to the grading system per protocol.

Table 4. Scoring System for Skin Reactions

Reaction	Numerical Grading
Erythema (redness) and Eschar (scab) formation	
No erythema	0
Very slight erythema (barely perceptible)	1
Well-defined erythema	2
Moderate erythema	3
Severe erythema (beet-redness) to eschar formation preventing grading of erythema	4
Edema (swelling) formation	
No edema	0
Very slight edema (barely perceptible)	1
Well-defined edema (edges of area well-defined by definite raising)	2
Moderate edema (raised approximately 1 mm)	3
Severe edema (raised more than 1 mm and extending beyond exposure area)	4
Maximum possible score for irritation	8
Other adverse changes at the injection sites shall be recorded and reported.	

8.8 PRIMARY IRRITATION INDEX

8.8.1 Evaluation

The final test article was described using the following scale.

Table 5. Primary Irritation Categories

Mean Score	Response Category
0 – 0.4	Negligible
0.5 – 1.9	Slight
2 – 4.9	Moderate
5 – 8	Severe

Table 6. Scoring Results

Animal Number		Animal ID: 612760				Animal ID: 612761				Animal ID: 612762			
		Control Article		Test Article		Control Article		Test Article		Control Article		Test Article	
		Edema	Erythema	Edema	Erythema	Edema	Erythema	Edema	Erythema	Edema	Erythema	Edema	Erythema
24 Hour Score	Left	0	0	0	0	0	0	0	0	0	0	0	0
	Right	0	0	0	0	0	0	0	0	0	0	0	0
48 Hour Score	Left	0	0	0	0	0	0	0	0	0	0	0	0
	Right	0	0	0	0	0	0	0	0	0	0	0	0
72 Hour Score	Left	0	0	0	0	0	0	0	0	0	0	0	0
	Right	0	0	0	0	0	0	0	0	0	0	0	0
Total Score		0		0		0		0		0		0	
Primary Irritation Score per Animal Treatment (Total Score/ 6 total observations per treatment)		0.0		0.0		0.0		0.0		0.0		0.0	
Primary Irritation Index per Treatment (Sum of Primary Irritation Scores per animal/Total number of animals (3))						**Control Article**		0.0		**Test Article**		0.0	
Primary Irritation Index (Test Article Irritation Index per Treatment – Negative Control Irritation Index per Treatment)						0.0							

8.9 STATISTICAL ANALYSIS

No statistical analysis was performed.

9 DISCUSSION / CONCLUSION

9.1 VALIDITY CRITERIA

All animals survived the in-life duration of the study and appeared outwardly healthy throughout the study duration. There were no signs of infection associated with any of the application sites and the negative control scores were within acceptable values.

9.2 FINAL RESULT

To obtain the primary irritation index, after the 72 ± 2 hours scoring, all erythema grades plus edema grades were totaled separately for each test article and negative control for each animal. The primary irritation score for an animal was calculated by dividing the sum of all the scores by 6 (two test/observation sites, three time points [24 ± 2, 48 ± 2, and 72 ± hours]). The primary irritation index was obtained for the test article by adding all the primary irritation scores of the individual animals and dividing by the number of animals. The primary irritation score for the negative control(s) was calculated and subtracted from the test article score to obtain the primary irritation index.

The test article Primary Irritation Index was 0.0.

9.3 STUDY OUTCOME

Completed (Negligible Irritant)

10 STUDY MATERIAL RETENTION/ARCHIVING

Item(s) Retained / Archived	Retention / Archiving Site
Study Article Characterization	The Sponsor is responsible for maintaining all test article characterization data as specified in 21 CFR Part 58.105. The control articles were commercial products and were characterized by the packaging label.
Testing Facility Raw Data	Data will be retained at the Test Facility according to GLP regulations for GLP studies, as specified in 21 CFR Part 58.195 for retention of records.
Study Articles	Test and control articles were not retained at the Test Facility
Final Report	APS

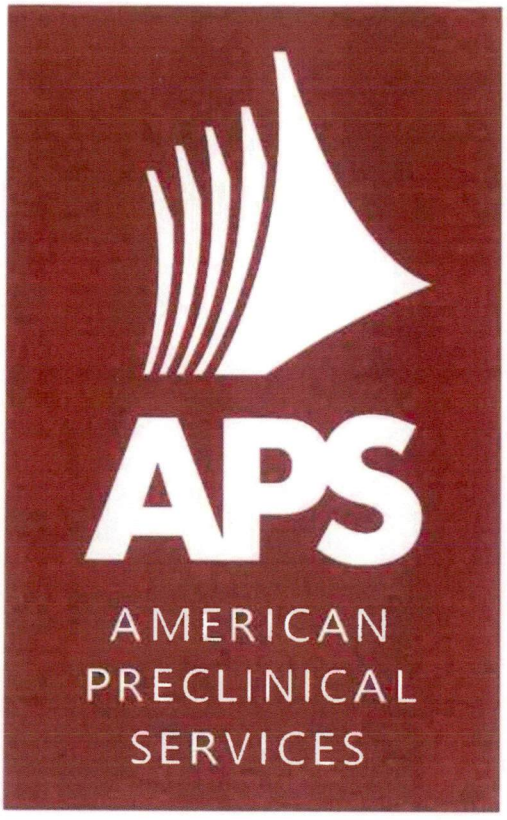
Appendix A POSITIVE CONTROL CERTIFICATE

Positive Control Validity					
Study ID		APS489-IR10			
Test Method Compliance		The study was conducted in compliance to FDA GLP Regulations, 21 CFR Part 58, and ISO 10993-10: 2010 and BS EN ISO 10993-10: 2013 (Biological Evaluation of Medical Devices, Part 10: Tests for Irritation and Skin Sensitization). The study articles will be prepared in compliance to the ISO 10993-12: 2012 and BS EN ISO 10993-12: 2012 (Biological Evaluation of Medical Devices, Part 12: Sample Preparation and Reference Materials). This positive control study demonstrated validity for APS standard protocol Animal Irritation Test (ISO).		
Control Material	**Control Type:**	Positive Control			
	Name:	Formaldehyde (12.33% solution in 0.9% NaCl)			
	Identification (lot number):	Formaldehyde: MKCJ7131 0.9% Normal Saline: J9A944			
	Expiration (if applicable):	Formaldehyde: 3/30/2021 0.9% Normal Saline: 01/2022			
Test System	**Animal Strain:**	New Zealand White			
	Animal Source:	Robinson Services			
Primary Irritation Index per Treatment (Sum of Primary Irritation Scores per animal/Total number of animals (3))		**Negative Control Article**	0.2	**Positive Control Article**	3.1
Primary Irritation Index (Positive Control Article Irritation Index per Treatment – Negative Control Article Irritation Index per Treatment)		2.9			
Study Result		Pass, Moderate Irritant			
Result Validity		Effective Date: 4/24/2020			
		Results are valid for three months before and after the effective date.[1]			

Certificate Issuance		
Issued By	**Name**	Benjamin Vos, BS
	Signature	Benj Vos
	Date	04.24.2020
Reviewed By	**Name**	Katie Jenkins
	Signature	Katie Jenkins
	Date	24 April 2020

[1] Use of International Standard ISO 10993-1, "Biological evaluation of medical devices – Part 1: Evaluation and testing within a risk management process", Guidance for Industry and Food and Drug Administration Staff, 2016.

Appendix B PROTOCOL



Animal Irritation Test (ISO) – Standard Protocol

APS Study ID		PRF1127-IR11
Study Director	Name	Benjamin Vos
	Email	bvos@apsemail.com
	Office Phone	763-951-8998
	Signature	Benjamin Vos
	Date	04.27.2020
Sponsor Approval		See the Study Protocol Appendix

1 TABLE OF CONTENTS

1.1 TABLE OF FIGURES AND TABLES

1.2 TABLE OF APPENDICES

2 ABBREVIATIONS

Table 1. Standard Abbreviations

Abbreviation	Name
AAALAC	Association for Assessment and Accreditation of Laboratory Animal Care International
ANAB	ANSI-ASQ National Accreditation Board
APS	American Preclinical Services, LLC
BID	Twice Daily
CM	Castrated Male
D or d	Day
EPA	Environmental Protection Agency
F	Intact Female
FDA	Food and Drug Administration
Freq	Frequency
GLP	Good Laboratory Practices
IA	Intra-arterial Administration
IACUC	Institutional Animal Care and Use Committee
ID	Intradermal
IM	Intramuscular
Inhal	Inhalation
IP	Intraperitoneal
IV	Intravenous
M	Intact Male
NA	Not Applicable
PHS	Public Health Service
PO	Oral
PRN	As Needed
Q or q	Every
QAU	Quality Assurance Unit
QID	Four Times Daily
QW	Once a Week
SID	Once Daily
SOP	Standard Operating Procedure
SQ	Subcutaneous
TID	Three Times Daily
USDA	United States Department of Agriculture

Table 2. Study-Specific Abbreviations

Abbreviation	Name
BS EN	British Standard European Norm
S	Edema (swelling)
E	Erythema (redness)
ISO	International Organization for Standardization
SPA	Study Protocol Appendix
Wt	Weight

3 GENERAL STUDY INFORMATION

3.1 STUDY TITLE AND IDENTIFICATION

Study Title	Animal Irritation Test (ISO)
APS Study ID	See the Study Protocol Appendix
Sponsor Project ID	See the Study Protocol Appendix, if applicable
Sponsor Study ID	See the Study Protocol Appendix, if applicable

3.2 STUDY COMPLIANCE

3.2.1 GLP Compliance

See the Study Protocol Appendix.

3.2.2 GLP Compliance Statement

For GLP Studies, this study will be conducted in compliance to the FDA GLP Regulations, 21 CFR Part 58 (FDA).

3.2.3 ISO Compliance

The study will be conducted in compliance to the International Organization for Standardization (ISO) 10993-10:2010 and British Standard European Norm ISO (BSEN ISO) 10993-10: 2013 (Biological Evaluation of Medical Devices, Part 10: Tests for Irritation and Skin Sensitization). The study articles will be prepared in compliance to the ISO 10993-12: 2012 and BS EN ISO 10993-12: 2012 (Biological Evaluation of Medical Devices, Part 12: Sample Preparation and Reference Materials).

3.2.4 Protocol Changes

Per APS SOP, protocol changes will be documented in writing via a protocol amendment. Amendments shall include, at a minimum, the following information: APS study ID, amendment number (sequentially numbered for each protocol starting with #1), the specific section of the protocol to be amended, description of the change and a reason for the change. The amendment will be signed and dated by the Study Director and the Sponsor Study Contact will be notified of the amendment. Protocol amendments affecting animal care or usage will also be reviewed and approved by the APS IACUC.

3.2.5 Deviations / Adverse Events

Test Facility Deviations: Per APS SOP, (1) Deviations will be documented in the raw data and the Study Director notified. (2) Documentation of deviations will be present in a summary document filed in the study records that includes: Communication ID, test system identification (if applicable), date of occurrence, description, impact assessment, and corrective action(s) within the study. All deviations that may have affected the quality or integrity of the data will be discussed in the final report, including an evaluation of whether or not the deviation impacts the study endpoint (s).

Adverse Events: Adverse Events as defined per APS SOP as "an unanticipated response to the Test System or an unforeseen circumstance that may affect the quality and integrity of the study" will be assessed by the Study Director for impact to study quality. All Adverse Events that may have affected the quality or integrity of the data will be discussed in the final report, including an evaluation of whether or not the Adverse Event impacts the study endpoint(s) and the corrective action(s) within the study.

3.2.6 IACUC Review

This Standard Protocol was reviewed and approved by APS' Institutional Animal Care and Use Committee (IACUC) as part of APS334-BCBLK, Blanket IACUC Protocol - Rabbits.

3.2.7 APS Registrations & Accreditations

APS has the following certifications and accreditations:

- USDA registration number 41-R0074.
- AAALAC accreditation number 001236.
- PHS assurance number A4586-01.
- ISO 17025 accreditation body and number: ANAB # L2348.

3.3 STUDY FACILITIES

Study Sponsor	See the Study Protocol Appendix
Test Facility (Corporate Headquarters)	American Preclinical Services, LLC (APS) 8945 Evergreen Boulevard NW Minneapolis, MN 55433 763-717-7990 office 763-717-2042 fax

3.4 STUDY PERSONNEL

Study Sponsor Representative	See the Study Protocol Appendix
Study Director	See cover page
Test Facility Management	Michael Conforti, DVM, MS, MBA 8945 Evergreen Boulevard NW Minneapolis, MN 55433 763-951-8030 phone mconforti@apsemail.com
Lead QAU (GLP only)	APS QAU 8945 Evergreen Boulevard NW Minneapolis, MN 55433 763-717-7990 phone

3.5 BACKGROUND

The Animal Irritation Test is a widely used method for evaluating the irritation potential of medical devices used topically (dermal placement) that will come in contact with humans. The Animal Irritation Test is designed for test articles that are placed directly in contact with the skin. An assessment is made for the potential of the test material to produce an irritation following topical application.

3.6 PURPOSE / OBJECTIVE

The purpose of this study is to evaluate the irritation potential of a test article. This test is used as a method for screening topical irritants in rabbits. The results are used as predictive measures for detecting potential irritants in humans.

3.7 ENDPOINTS

Endpoint 1	**Tissue reaction grades: Scoring 1 ± 0.1, 24 ± 2, 48 ± 2, 72 ± 2 hours post test and control article contact duration** The grading system for dermal reactions is based on a 0 to 4 scale.
Endpoint 2	**Primary irritation index: Evaluation of results via tissue reaction grading at 24 ± 2, 48 ± 2, and 72 ± 2 hours post test and control article contact duration** A primary irritation index is characterized by number (tissue grades) and description (response category) of the test article and negative control treatments sites.
Endpoint 3	**Overall animal health (moribundity): Daily** Clinical observations will be conducted by trained personnel to assess the overall health of the animal.

3.8 ASSAY VALIDITY

Validity Criteria 1	The animals survive the in-life duration of the study and appear outwardly healthy throughout the study duration.
Validity Criteria 2	There will be no signs of infections associated with any of the application sites on the animals.
Validity Criteria 3	The negative control scores are within acceptable values.

3.9 STUDY ARTICLES

3.9.1 Characterization

Test article characterization and stability testing will be the responsibility of and maintained by the Sponsor. The Sponsor will assure traceability of the test article characterization and stability data to a

known identification number such as a lot, batch, or serial number. The control articles will be used from a commercial lot and will be characterized by the packaging label or serial number as appropriate.

3.9.2 Disposition

Used and unused test article(s) will be disposed of unless indicated otherwise by the Sponsor in the Study Protocol Appendix. Used and unused, non-retained control articles will be disposed of following administration.

3.9.3 Test Article 01

Test Article Name	See the Study Protocol Appendix. The test article name will be included in the final report.
Identification	See the Study Protocol Appendix. Traceable identification such as a lot, batch or serial number will be recorded and included in the final report.
Storage Condition(s)	See the Study Protocol Appendix. Any special handling instructions will be arranged prior to shipping the test article to the Test Facility.
Tracking	Test article(s) will be supplied by the Sponsor with a completed Study Protocol Appendix. As per APS SOP, a record of all test articles received from the Sponsor will be maintained.
Labeling	Test article(s) will be individually labeled for identity and traceability.
Sterilization	See the Study Protocol Appendix. Test article(s) may be submitted sterile or non-sterile.
Preparation	See the Study Protocol Appendix. Standard medical device study articles will be prepared according to Table 3. Chemicals, combination devices, fluid pathways, or liquid test articles may be prepared according to the Sponsor's instructions, regulatory guidelines, and /or APS SOP. All test articles will be dosed neat (as is) unless instructed otherwise by the Sponsor. If the test article cannot be tested neat, the use of extraction methods may be deployed in accordance to ISO 10993-12 guidelines. Unless specified on the Study Protocol Appendix, test and control article dose amount will follow 0.

Table 3. Standard Surface Area and Extract Liquid Volumes

Thickness (mm)	Extract Ratio (Surface area or mass/volume ± 10%)	Examples of Materials
<0.5	6 cm^2/mL	Film, sheet, tubing wall
0.5-1.0	3 cm^2/mL	Tubing wall, slab, small molded items
>1.0	3 cm^2/mL	Larger molded items

>1.0	1.25 cm^2/mL	Elastomeric closures
Irregularly shaped solid devices	0.2 g sample/mL	Powder, pellets, foam, non-absorbent molded items
Irregularly shaped porous devices (low density material)	0.1 g/mL	Membranes

Per ISO 10993-12, there are currently no recognized standard methods for testing absorbents and hydrocolloids. APS has developed its own standardized methodology for assessing absorbency. Additional pre-study absorbency testing may be requested by the Sponsor and conducted separately from individual study protocols. In the event a test article is experimentally determined to be sufficiently absorbent to require a change to standard extraction ratio, the volume of extraction vehicle(s) will be adjusted to compensate accordingly. Absorbency testing results will be included in study files and actual extraction ratios and volumes used will be documented in the raw data.

3.9.4 Control Article 01

Control Article Name	Negative Control: Gauze or Vehicle without the test article. Unless specified on the Study Protocol Appendix, standard control article referenced in 0 will be used.
Description	Negative control is blank gauze or the vehicle only preparations subjected to the same conditions as the test article.
Identification	A traceable identification number such as a lot, batch, or serial number will be recorded.
Tracking	The negative control will be supplied by APS.
Labeling	The negative control will be labeled for identity and traceability.

Table 4. Standard Test and Control Dose Amount

Test Article Type	Control Article	Test/Control Dose Amount
Extract	Extract Vehicle	0.5 mL
Liquid (dose neat)	Vehicle without the active ingredient	0.5 mL
Gel or Cream (dose neat)	Vehicle without the active ingredient or 2.5 cm x 2.5 cm blank gauze (moistened with water during application)	0.5 mL, 0.5 g of test article; control article will be 2.5 cm x 2.5 cm blank gauze
Powder (dose neat)	2.5 cm x 2.5 cm blank gauze (moistened with water during application)	0.5 g of test article; control article will be 2.5 cm x 2.5 cm blank gauze
Solid material (dose neat)	Blank gauze (moistened with water during application)	Test article will be cut to size fit in the 2.5 cm x 2.5 cm patch (moistened with water during application); control article will be 2.5 cm x 2.5 cm blank gauze

4 METHODS

4.1 STUDY DESIGN GENERAL INFORMATION

4.1.1 Summary

A total of 3 animals per test article (extract or neat dose) will be used. 4 – 24 hours prior to testing, the hair on the back of the animals will be closely clipped, allowing a sufficient distance on both sides of the spine for the application of test article and negative control. The test and control article will be applied directly to the clipped skin. The application sites will be wrapped with a bandage (semi-occlusive) for a minimum of 4 hours. At the end of the contact duration, the dressings will be removed and the sites where the test article and negative control patches were located will be marked with permanent ink. Where appropriate, residual test article will be removed by washing with lukewarm water or other suitable non-irritating solvent, followed by careful drying. Observations will be performed on each application site 1 ± 0.1, 24 ± 2, 48 ± 2, and 72 ± 2 hours after unwrap. The tissue reactions will be graded for erythema and edema for each application site at each time interval. The 1 ± 0.1 hour observation period is performed to ensure there are no adverse clinical signs for the animals, so the tissue grades each animal receives during this time point is not included in the calculations of the primary irritation score or primary irritation index.

The positive control tests are conducted within three months (before or after) of the study article testing under a separate internal Test Facility protocol in accordance with GLP and ISO Regulations. The results of the most recent positive control test will be on file with the Test Facility and embedded in the final report. A copy of the report will be available for review at the request of the Sponsor.

The results of this study will be interpreted based upon the frequency of animals displaying irritation responses and the comparative result between the test article and negative control scores. By calculating a primary irritation score for the test and control article, a categorical result will be determined via the primary irritation index.

4.1.2 Control of Bias

Animals are selected from a stock pool of Test Facility animals. All animals selected for this study will be naïve, and observed prior to entry into the study to ensure that they are not exhibiting any signs of clinically relevant abnormalities.

4.1.3 Exclusion Criteria

Any animal may be excluded due to any occurrence of death, disease, injury, or inappropriate study article dosing that may affect study outcome or analysis and is determined to be unrelated to the test and/or control article with valid justification by the Study Director.

4.1.4 Clinical Risk Analysis

Topical dosing complications may include, but are not limited to: pain or swelling.

Animals are not expected to otherwise display clinical signs of acute distress (characterized by any or all of the following: chronic piloerection, chronic sedation, aggression, constant vocalization) at any time following injection of test/control articles.

4.1.5 Humane Endpoints

The veterinarian will be contacted if any animals on study are observed with abnormalities including, but not limited to, the following:

- Weight loss: An animal has not consumed an appreciable amount of food for a time resulting in substantial weight loss of more than 20% of its body weight.
- Moribund state: Depression, complete anorexia and hypothermia, comatose/pale/cold to the touch for an extended period of time. Inability or extreme reluctance to stand which persists for 24 hours (assuming that the animal has recovered from anesthesia). The animal is consistently unwilling / unable to stand. CNS disorders such as persistent head tilt, incoordination, ataxia, tremors, spasticity, seizures, circling, or paresis for longer than 1 hour.
- Severe irritation or uncontrollable pain/distress: Animals showing signs of pain and/or distress for an extended period of time.
- Miscellaneous Conditions: Diarrhea, especially if prolonged, leading to emaciation and/or debilitation. Prolonged or intense diuresis leading to severe dehydration.

If the animal is in a moribund state and is suffering, the Study Director must be contacted and the Sponsor will be notified; the animal will be euthanized and treated as a study death. In the event that the Study Director cannot be reached, the Veterinarian may authorize euthanasia of the animal.

4.2 TEST SYSTEMS

Test System Number	01
Species	Rabbit
Breed	New Zealand White
Sex	Male and/or Female; females will be nulliparous, not pregnant, and housed separately from males for the duration of the test.
Test System ID	01: Rabbit - New Zealand White - Male and/or Female
Animal Source Type	Single Source
Animal Source	Approved APS vendor(s) to be documented in the raw data.
Total # of Animals	3 animals per extract or neat dose
Total # of Back-up Animals	1 per extract or neat dose
Age at Initial Procedure	Age appropriate to weight
Min. Wt. at Initial Procedure	2.0 kg
Definition of a Back-Up Animal	A back-up animal is approved for use in the event of: (1) a death during dosing due to reasons not considered test article related or (2) animal does not receive full dose due to reasons not considered test article related. Note: if additional animals are required in order to complete the study design for any reason not stated above, a protocol amendment will need to be generated for IACUC review and approval.
Disposal of Animal Carcasses	Per APS SOP, animal carcasses will be disposed of via an approved, licensed APS vendor.
Animal Identification	Per APS SOP.

4.3 GROUP DESIGN

Test System ID: Species: Sex	Group ID: Study Article	# of Animals	Procedure Type(s)	Procedure Timing, if applicable and/or Termination Timing, if applicable
TS01: Rabbit: Male and/or Female	GRP01: Test and Control	3 per extract or neat dose	Application	Day 0
			Scoring	1 ± 0.1 hour, 24 ± 2 hours, 48 ± 2 hours, and 72± 2 hours after completion of application duration
			Termination	Post 72 hour scoring

4.4 ANIMAL HUSBANDRY

4.4.1 Quarantine

For all study Groups: Animals will be received and managed under an independent APS holding protocol where quarantine (as appropriate to species) and veterinary exams will be performed. Animals released to study will be considered suitable for enrollment in the study. Enrollment will be confirmed by administration of test article to test system.

4.4.2 Housing

For all study Groups: Per APS SOP, animals will be housed individually or in groups (as animal behavior allows) with like species.

4.4.3 Cage Cleaning

For all study Groups: Per APS SOP, cage pan liners will be replaced minimally two times per week. Animal cage racks will be changed out for cleaning minimally every two weeks for rabbits.

4.4.4 Feeding

For all study Groups: Per APS SOP, rabbits will be fed a fixed-formula diet certified by the manufacturer for nutritional components and environmental contaminants. Rabbits may be fed commercial pet food, human food, or fresh produce in order to stimulate appetite, administer medications, or ease handling.

4.4.5 Watering

For all study Groups: Per APS SOP, tap water (unfiltered) will be provided *ad libitum* unless otherwise noted. Per APS SOP, samples of animal care room water are analyzed annually for total coliform, lead, and copper. Municipal water testing is conducted annually for EPA specified microbiological content and selected environmental contaminants. Results of these analyses are filed at the Test Facility as they become available.

4.4.6 Contaminants

For all study Groups: No contaminants are known to be present in the water or feed at levels that would affect the outcome or integrity of this study.

4.4.7 Medical Treatments

For all study Groups: Per APS SOP, medical treatments and diagnostics not specified or contraindicated in this protocol may be administered by veterinarian order. The veterinarian will act in the best interest of the animal's welfare and, if deemed necessary in accordance with the Animal Welfare Act, the veterinarian may authorize treatment to mitigate pain/distress up to and including euthanasia. The reason for treatment, description of treatment, date(s) of treatment, and resolution will be documented in the animal's records, preferably in SOAP (subjective, objective, assessment, plan) format.

4.4.8 Emergency Treatments

For all study Groups: Per APS SOP, emergency treatments not specified or contraindicated in this protocol may be prescribed by veterinarian order. The description of emergency, activities, administered substances, and conclusion will be documented and reviewed by a Veterinarian.

Emergency treatments required in the operating room may be directed by a Surgeon or Interventionalist's per APS SOP, based on the condition of the animal during the procedure, if not contraindicated in this protocol. All medications will be documented on the Procedure Substance Administration Form and activities will be documented in the records.

4.4.9 Treatment Contraindications

For all study Groups: All attempts will be made to avoid treatments that may interfere with study endpoints. If treatment(s) have the potential to interfere with study endpoints, animals may be euthanized in lieu of treatments.

4.5 CLINICAL OBSERVATIONS

Per APS SOP, unless otherwise directed per (1) protocol specified postoperative clinical observations, (2) other protocol requirements, (3) by *ad hoc* Veterinarian order, or (4) *ad hoc* Study Director order, the animals will be observed at a minimum of SID throughout the duration of the study for physical and behavioral attributes including, but not limited to, the following:

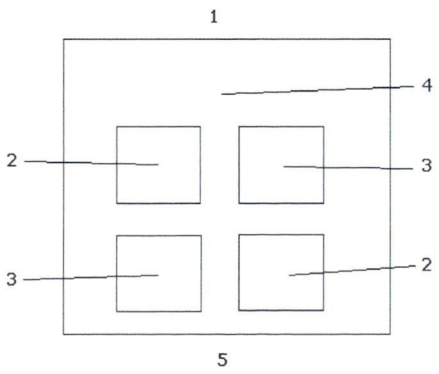
- Interaction with pen-mates (if applicable).
- Elimination of urine & feces, discolored urine (if applicable), diarrhea, absence of feces (constipation).
- Signs of illness or injury, lethargy, vomiting, excessive salivation, abnormal posture, pain, lameness, discomfort, unwillingness or inability to move.
- Additional assessments will be performed whenever warranted based on clinical observations.

4.6 PROCEDURE METHODS

4.6.1 Application (Topical Phase)

Activity	Time Point	Relative Time	Freq.	Method
Clipping	Not applicable	4-24 hours prior to topical administration	1x	Clip the back of the animal with the use of an electric clipper, providing an adequate region for topical application. The skin must be free of hair.
Weigh	Not applicable	Between Day -1 and Day 0, prior to topical administration	1x	Weigh the animal per APS SOP. Record the weight in the raw data.
Topical Administration	Not Applicable	Day 0	1x	Per the configuration in Figure 1, the test article will be applied directly to the skin via a 2.5 cm X 2.5 cm non-occlusive dressing (such as absorbent gauze) soaked with 0.5 mL of the test article, or if appropriate, dose neat by direct application of test article as listed on Table 4 or the Study Protocol Appendix. If solid test article is applied, moisten the study article sufficiently with water to ensure good contact with skin and cover with a non-occlusive dressing. Similarly, the negative control will also be applied to the skin (unless a physically similar, non-irritant control is available). The application site patches will be secured by wrapping the animals with a semi-occlusive bandage for a minimum of 4 hours.
Patch Removal	Not Applicable	≥ 4 hours but not to exceed 6 hours after completion of topical application	1x	The dressings will be removed and the test article and negative control sites marked with permanent ink. Where appropriate, residual test article will be removed by washing with lukewarm water or other suitable non-irritating solvent, and careful drying.

Figure 1. Location of Skin Application Sites



Key
1 Cranial end
2 Test site
3 Control site
4 Clipped dorsal region
5 Caudal end

4.6.2 Scoring

Activity	Time Point	Relative Time	Freq.	Method
Scoring	1 hr, 24 hr, 48 hr, and 72 hr Scoring	1 ± 0.1 hours, 24 ± 2 hours, 48 ± 2 hours, and 72 ± 2 hours after removal of dressing	3x	If applicable, clip the animals to allow for adequate grading of the treatment sites. Observe the appearance of the test article and negative control sites at 1 ± 0.1 hours, 24 ± 2 hours, 48 ± 2 hours, and 72 ± 2 hours after the removal of the dressings. At each time interval, the tissue reactions will be graded for erythema and edema according to the grading scale given in Table 5.

Table 5. Scoring System for Skin Reactions

Reaction	Numerical Grading
Erythema (redness) and Eschar (scab) formation	
No erythema	0
Very slight erythema (barely perceptible)	1
Well-defined erythema	2
Moderate erythema	3
Severe erythema (beet-redness) to eschar formation preventing grading of erythema	4
Edema (swelling) formation	
No edema	0
Very slight edema (barely perceptible)	1
Well-defined edema (edges of area well-defined by definite raising)	2
Moderate edema (raised approximately 1 mm)	3
Severe edema (raised more than 1 mm and extending beyond exposure area)	4
Maximum possible score for irritation	8

Other adverse changes at the application sites shall be recorded and reported.

4.6.3 Termination

4.6.3.1 Pre-Procedure Substance Administration

Substance, Dose, Route, Freq.	Drug Type, Reason	Relative Time Pre-procedure
Acepromazine, 0.25-2 mg/kg, IM, 1x	Phenothiazine, Sedative	At induction

4.6.3.2 Procedure Activities

Activity	Time Point	Relative Time	Freq.	Method
Euthanasia	Termination	Post 72 ± 2 hour score	1x	Euthanize per APS SOP.

4.7 CLINICAL PATHOLOGY

Not required.

4.8 UNSCHEDULED DEATH

4.8.1 Procedural Death (PD)

- The Study Director or designated APS staff will notify the Sponsor Representative of the procedural death.
- Document the details of the procedural death in the raw data.
- A pathologist or qualified prosector will perform a necropsy according to methods in the pathology section of this protocol.
- If the necropsy cannot be performed within 30 minutes, the animal should be refrigerated until necropsy.

4.8.2 Early Termination (ET)

- The Study Director or designated APS staff will notify the Sponsor Representative of the early termination.
- Document the reason for the early termination in the raw data.
- Euthanize according to the methods described in this protocol.
- A pathologist or qualified prosector will perform a necropsy according to methods in the pathology section of this protocol.
- If the necropsy cannot be performed within 30 minutes after euthanasia, the animal should be refrigerated until necropsy.

4.8.3 Early Death (ED)

Per APS SOP, APS staff discovering the dead animal will:

- Record the animal's condition and surroundings upon discovery, including discovery time and date.
- APS staff discovering the dead animal will notify the Study Director, Vet and/or APS management upon discovery.
- The Study Director or designated APS staff will notify the Sponsor Representative as soon as possible after the discovery.
- A pathologist or qualified prosector will perform a necropsy according to methods in the pathology section of this protocol.
- If the necropsy cannot be performed within 30 minutes after discovery, the animal should be refrigerated until necropsy.

4.9 GROSS PATHOLOGY

4.9.1 Scheduled (Sch) Termination

Pathologist / DVM Requirement:	Not Required
Necropsy Type:	NA

- A necropsy is not required for scheduled terminations.

4.9.2 Procedural Death (PD)

Pathologist / DVM Requirement:	Not Required
Necropsy Type:	Complete or Limited

- Per APS SOP, trained APS staff will perform a necropsy.
- A limited necropsy may be performed if the cause of death is identified by an APS Pathologist or APS Veterinarian, or when directed by the Study Director.
  - Limited necropsies include examination of the dose site tissues and collateral structures for lesions that may be attributed to the test article.
- A complete necropsy, with calvarium, may be performed to attempt to determine cause of death per APS SOP if the prosector is not directed to perform a limited necropsy as above.
- Tissues may be collected and sent for histological evaluation, or additional diagnostic techniques may be performed.
- Per APS SOP, photographs may be taken of any tissues to supplement written descriptions and prosector notes. The use of photography will be documented in the raw data.

4.9.3 Early / Unscheduled Death (ED)

Pathologist / DVM Requirement:	Not Required
Necropsy Type:	Complete

- Per APS SOP, trained APS staff will perform a necropsy.
- A complete necropsy, with calvarium, will be performed to attempt to determine cause of death per APS SOP.
- Tissues may be collected and sent for histological evaluation, or additional diagnostic techniques may be performed.
- Per APS SOP, photographs may be taken of any tissues to supplement written descriptions and prosector notes. The use of photography will be documented in the raw data.

4.9.4 Early Termination (ET)

Pathologist / DVM Requirement:	Not Required
Necropsy Type:	NA or Complete or Limited

- Per APS SOP, trained APS staff may perform a necropsy.
- Necropsies need not be performed or a limited necropsy may be performed if the reason for termination is identified by an APS Pathologist or APS Veterinarian, or when directed by the Study Director.
  - Limited necropsies include examination of the tissues determined to be related to the reason for termination.
  - Additional tissues may be examined.

- A complete necropsy, with calvarium, may be performed to attempt to determine the need for termination per APS SOP if the prosector is not directed to perform a limited necropsy as above.
- Tissues may be collected and sent for histological evaluation, or additional diagnostic techniques may be performed.
- Per APS SOP, photographs may be taken of any tissues to supplement written descriptions and prosector notes. The use of photography will be documented in the raw data.

4.10 STUDY SPECIFIC ANALYSIS

4.10.1 Primary Irritation Index

After the 72 ± 2 hours scoring, all erythema grades plus edema grades are totaled separately for each test article and negative control for each animal. The primary irritation score for an animal is calculated by dividing the sum of all the scores by 6 (two test/observation sites, three time points [24 ± 2, 48 ± 2, and 72 ± hours]). To obtain the primary irritation index for the test article, add all the primary irritation scores of the individual animals and divide by the number of animals. The primary irritation score for the negative controls is calculated and subtracted from the test article score to obtain the primary irritation index. Any negative difference will be recorded as zero.

Table 6. Primary Irritation Categories

Mean Score	Response Category
0 – 0.4	Negligible
0.5 – 1.9	Slight
2 – 4.9	Moderate
5 – 8	Severe

4.11 STATISTICAL ANALYSIS

4.11.1 Test Article and Negative Control

The results of this study will be interpreted based on the frequency of animals displaying irritation responses, by determining the mean test and control scores for each animal.

4.12 STUDY OUTCOMES

4.12.1 Completed

The test article will be characterized by the primary irritation score.

4.12.2 Expanded (Persistent Lesions)

Extended observation may be necessary if there are persistent lesions in order to evaluate the reversibility or irreversibility of the lesions, not exceeding a period of 14 days. If expanded testing is considered, a protocol amendment will be generated describing the methodology. Results from the initial test and the expanded test will be reported.

4.12.3 Expanded (Validity Criteria Failure)

If any validity criteria are not met, the results will be deemed invalid. The invalid portion of the study will be rerun under the same study identification. A protocol amendment will be generated describing the methodology. Invalid data will not be reported, but will be kept on file at the Test Facility.

4.13 REPORTING

Report Name	Author	Report Description
Final Study Report	APS Study Director	The final report will be written according to GLP regulations or at the Sponsor's request for non-GLP studies.

4.14 STUDY MATERIAL RETENTION/ARCHIVING

Item(s) Retained / Archived	Retention / Archiving Site
Study Article Characterization	The Sponsor is responsible for maintaining all test and control (if not supplied by APS) article characterization data as specified in 21 CFR Part 58.105. If APS supplies the control article, it will be characterized by the product label.
Testing Facility In-Life Raw Data	Data will be retained according to GLP regulations for GLP studies, as specified in 21 CFR Part 58.195 for retention of records, or at the Sponsor's request for non-GLP studies.
Study Articles	For studies of 4 week's duration or less, test and control (if applicable) articles will not be retained at the Test Facility.
Wet Specimens	APS, if applicable
Final Report	APS

Appendix A STUDY PROTOCOL APPENDIX (SPA)

Project Information			
Quote ID/APS Project ID	APS20-0897	**Compliance**	GLP
Sponsor Project ID			
APS Study ID(s)	PRF1127-IR11		

Sponsor Information			
Company Name	Nelson Laboratories	**Street Address**	6280 S. Redwood Rd.
First Name	Trevor	**City**	Salt Lake City
Last Name	Fish	**State**	UT
Primary Email	TFish@nelsonlabs.com	**Postal Code**	84123
Primary Phone	801-290-7500	**Country**	USA

Test Article Information		
Test Article Name	NZ-0100-PATCH	
Unique Identifier	Lot #	
Identification ID	NZ-0100-PATCH	
Storage Conditions	Room Temperature	
Sterilization and Sterility	Study Article not sterilized (non-sterile)	
Expiration Date	Currently Undetermined	
Material Type	Solid	
Amount of Test Article Initially Submitted for Testing	Units	8

Test Article Measurements	
Measurement Type (Surface Area is preferred by regulatory bodies)	Single Surface Area
Surface Area (cm^2/Test Article)	NA, Cut to size

Test Article and Vehicle Compatibility	
Is the Test Article compatible with proposed vehicle(s) and/or extraction condition temperature(s) selected in the Test Selection tab?	Yes

Test Article Disposition	
Test Article Disposition	Discard

Project Information			
Quote ID/APS Project ID	APS20-0897	**Compliance**	GLP
Sponsor Project ID			
APS Study ID(s)	PRF1127-IR11		

Control/Comparison Article Information	
Is a Predicate/Comparison Article being tested?	No

APS may cut the study article(s) unless 'Cutting and/or Disassembly' is selected in the Article Preparation Options field and instructions of DO NOT CUT are provided.

Test Selection #1	Irritation : Animal Irritation Test (ISO)
APS Study ID	PRF1127-IR11
Sponsor Study ID	1289100
Test Method Type	Standard
Extraction Ratio	2.5 cm x 2.5 cm x 2 per animal
Test Article Preparation Options	Preparation Instructions

Test Article Preparation Options	
Test Article Prep Instructions - Test Selection #1	Cut sample as needed.

Project Information			
Quote ID/APS Project ID	APS20-0897	**Compliance**	GLP
Sponsor Project ID			
APS Study ID(s)	PRF1127-IR11		

Terms and Conditions	
☒	I approve using the current version (at Study Director initiation date) of the APS Protocol for the selected test(s).
☒	I acknowledge the Study Article(s) Information provided in this document has traceability to manufacturer's internal records compliant to applicable regulations.
☒	I acknowledge that, if applicable, Study Article Measurements (surface area or mass) provided in this document WILL NOT be verified by APS.
☒	I acknowledge that, if applicable, APS will prepare macroscopically proportional representative components for testing when unable to include an entire Study Article.
☒	I authorize Study Article(s) preparation to be completed as directed in this form for use in the selected Test(s).
☒	I acknowledge that testing performed to characterize particulates present in extract(s) will be considered outside the scope of the biocompatibility testing performed by APS. APS will not be able to include, analyze, or otherwise discuss results of testing in APS reports beyond acknowledgement of the presence of particulates, their physical descriptions generated by APS, and that samples were supplied to Sponsor or a third party laboratory for testing purposes.
☒	I authorize APS to use the information provided in this document for final reporting.
☒	To the best of our knowledge, we have provided a complete and factual description of the information for the proposed biocompatibility test(s). For in life studies, the appropriate measures have been taken to ensure that the minimum number of animals required to achieve the experimental objectives and are not unnecessarily duplicating previous studies.
☒	I acknowledge the risk, if applicable, of test system contamination and/or particulate presence in extract(s) when submitting study articles that are non-sterile and/or not submitted in final commercial packaging.
☒	I acknowledge that Approver Name and Reviewer Name fields in the final SPA are not considered electronic signatures.
☒	I acknowledge that, if applicable, Study Articles with Sterilization = Sterilized, Submitted Sterile that do not contain sterilization indication on package label or indication within the packaging (e.g. indicator strip) are confirmed sterile.

Sponsor Approval	
Approval Date	04-24-2020
Approver Name	Trevor Fish
APS Review	
Review Date	04-24-2020
Reviewer Name	Nardina Nash

Appendix C AMENDMENTS

There were no amendments for this study.

Appendix D DEVIATIONS

There were no deviations that affected the quality or integrity of the data for this study.