中国认可
国际互认
检测
TESTING
CNAS L13034

Oral Mucosa Irritation Test

Extraction Method

Final Report

Verification

Report Number: CSTBB2022110856

Article Name: Surgical guide resin

Method Standard: ISO 10993-23:2021

Sponsor

Shenzhen Yongchanghe Technology Co.,Ltd

1001, Huaide international building, No. 73, Fuyong section, Guangshen Road, Fuyong community, Fuyong street, Bao'an District, Shenzhen, China

Test Facility

CCIC Huatongwei International Inspection (Suzhou) Co., Ltd

Room 101, Building G, Ruoshui Road 388, Suzhou, Jiangsu, China

CONTENTS

Notices

1. Please apply for rechecking within 15 days of receiving the report if there is any objection.
2. Any erasure or without special testing seal renders the report null and void.
3. The report is only valid when signed by the persons who edited, checked and approved it.
4. The report is only responsible for the test results of the tested samples.
5. The report shall not be reproduced except in full without the written approval of the company.

Abstract

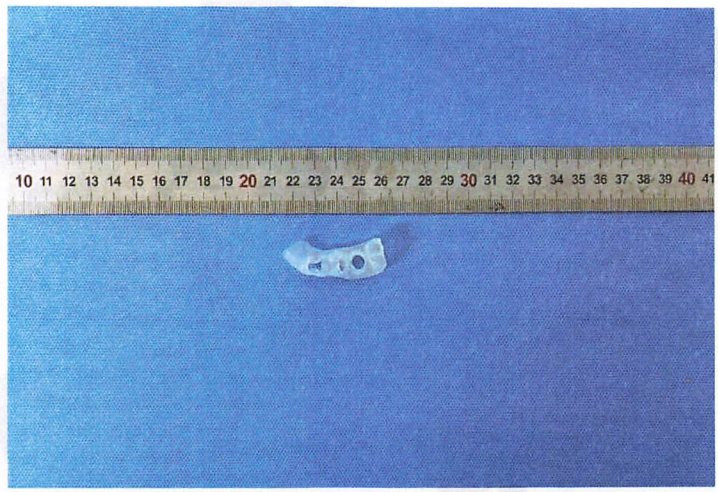
In this study, we took Syrian hamsters to observe the oral mucosa irritation of the test article according to ISO 10993-23:2021.

The test article were extracted by 0.9 % Sodium Chloride Injection and Sesame Oil. Soak a cotton-wool pellet in the sample, record the volume absorbed, and place a pellet in one pouch of each animal. Negative control sample is placed in the other cheek pouch. The duration of exposure shall be that expected for actual use of the material, but no shorter than 5 min. Following the exposure, remove and cotton-wool pellet and wash the pouch with physiological saline solution, taking care not to contaminate the other pouch. Repeat the above procedure every hour (±0.1 h) for 4 h.

The results showed that the Syrian hamsters retained a normal appearance throughout the test and showed no oral mucosa irritation. While in test article group, the response of oral mucosa irritation on testing side did not exceed that on the control side. The oral mucosa reactions for erythema and oedema were not observed in test article group.

Based on the above results, it can be concluded that under the experimental conditions, the test article has no potential oral mucosa irritation in the Syrian hamsters.

Study Verification and Signature



Protocol Number	SST2210041703BB
Protocol Effective Date	2022-11-02
Technical Initiation Date	2022-11-11
Technical Completion Date	2022-11-30
Final Report Completion Date	2023-03-09

Personnel — Betty Zhuang — 2023-03-09 (Date Completed)

Approved — Vicky Ji — Study Director — 2023-03-09 (Date Completed)

Supervisory — Test Facility Manager — Date Completed

CCIC Huatongwei international inspection (Suzhou) Co., Ltd.

Quality Assurance Statement and GLP Statement

Quality Assurance Statement

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

The final report was reviewed to assure that the report accurately describes the methods and standard operating procedures. The reported results accurately reflect the raw data of the nonclinical study conducted per the protocol.

Phase Inspected	Date	Study Director	Management
Experiment	2022-11-11	2022-11-11	2022-11-11
Raw Data	2022-11-30	2022-11-30	2022-11-30
Final Report	2023-03-09	2023-03-09	2023-03-09

The findings of these inspections have been reported to Management and the Study Director.

Hongxia Li
Quality Assurance

2023-03-09
Date

GLP Statement

This study was conducted in compliance with current 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 HTW, exempt from this Good Laboratory Practice Statement, included characterization and stability of the test article and its mixture with carriers, 21 CFR, Part 58.105 and 58.113.

Vicky Jin
Study Director

2023-03-09
Date

1.0 Purpose

The test was designed to evaluate the potential irritation caused by test article contact with the oral mucosa of Syrian hamsters and extrapolating the results to humans, but it does not establish the actual risk of irritation.

2.0 Reference

Biological evaluation of medical devices-Part 23: Tests for irritation (ISO 10993-23:2021)

Biological evaluation of medical devices-Part 2: Animal welfare requirements (ISO 10993-2:2006)

Biological evaluation of medical devices-Part 12: Sample preparation and reference materials (ISO 10993-12:2021)

3.0 Test and control articles

Groups	Test article	Negative Control Article(Polar)	Negative Control Article(Non-Polar)
Name	Surgical guide resin	0.9% Sodium Chloride Injection(SC)	Sesame Oil (SO)
Manufacturer	Shenzhen Yongchanghe Technology Co.,Ltd	Guangxi Yuyuan Pharmaceutical Co., Ltd	Ji'an Lv yuan natural flavor oil refinery
Size	PCS	500 ml	25kg
Model	DB-07	/	/
Lot Batch#	JH20220915	H21121903	2022.02.14
Test Article Material	3D PRINTER RESIN	/	/
Physical State	Solid	Liquid	Liquid
Color	clear	Colorless	Light yellow
Package material	PE BAG	/	/
Sterilized or Not	Not Sterilized	/	/
Concentration	/	0.9 %	/
Surface (cm^2)	Not Provided	/	/
Weight (g)	2.44	/	/
Storage Condition	Room Temp.	Room Temp.	Room Temp.
Note：The information about the test article was supplied by the sponsor wherever applicable.			

4.0 Identification of test system

4.1 Test animal

Species: Syrian hamsters

Number: 6

Sex: Either sex

Weight: 100~120 g

Health status: Healthy, not previously used in other experimental procedures. Female animals were nul liparous and not pregnant.

Animal identification: Ear tag

Cages: Plastic cage

Acclimation Period: 7 days under the same conditions as for the actual test

4.2 Justification of test system

The Syrian hamster is specified as an appropriate animal model for evaluating potential oral mucosa i rritants by the current testing standards. The species and number of animals as well as the route of admi nistration used, are recommended by Standard guidelines.

5.0 Animal management

Animal purchase: Hangzhou Qizhen Experimental Animal Technology Co., Ltd., SCXK (ZHE)2022-0005

Bedding: Corncob, Pizhou Xiaohe Technology Development Co., Ltd

Feed: Full-price pellets, Pizhou Xiaohe Technology Development Co., Ltd

Water: Drinking water met the Standards for Drinking Water Quality GB 5749-2006

Animal room temperature: 20-25 ℃

Animal room relative humidity: 30 %-70 %

Lights: 12 hours light/dark cycle, full-spectrum lighting

Personnel: Associates involved were appropriately qualified and trained

Selection: Only healthy, previously unused animals were selected

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

6.0 Equipment

Constant Temperature Vibrator (SHB007), Electronic scale (SHB017), Electron Microscope (SHB177)

7.0 Experiment design

7.1 Sample preparation

Before the test, the product shall be sterilized by hydrogen peroxide low-temperature plasma. After sterilization, observe that the physical state of the sample is normal and then start the biological test. The extracts of test article will be prepared according to the following steps:

<table>
<tr><td colspan="2">Aseptic Sampling</td><td colspan="5">Extraction in sterile vessels</td></tr>
<tr><td>Sampling Manner</td><td>Actually sampling</td><td>Ratio</td><td colspan="2">Reagent</td><td>Condition</td><td>pH</td></tr>
<tr><td rowspan="2">Whole</td><td>10.0 g</td><td rowspan="2">0.2g:1ml</td><td>SC</td><td>50.0 ml</td><td rowspan="2">50°C / 72 h/ 60rpm</td><td>5.5</td></tr>
<tr><td>10.0 g</td><td>SO</td><td>50.0 ml</td><td>/</td></tr>
</table>

The state of the leaching solution did not change visually after the leaching was advanced.The extraction solution is clear, and the pH value has not been adjusted,filtered,centrifuged,diluted and other processes. The control solution was prepared under the same conditions.The extraction of the test article could be stored at room temperature. for no more than 24 h.

The changes of the leaching solution was observed after extraction. No particulates or color changes were observed in pre- and post-extraction, the color and pH of the extraction solution did not change before and after use,and the pH value was 5.5, the status of the extract was shown in the table below.

Vehicle	Time Observed	Extracts	Condition of Final Extracts		
			Color	Clear or Not	Particulates
Polar	Before Extraction	Test article	Colorless	Clear	None
		Negative Control	Colorless	Clear	None
	After Extraction	Test article	Colorless	Clear	None
		Negative Control	Colorless	Clear	None
Non-Polar	Before Extraction	Test article	Light yellow	Clear	None
		Negative Control	Light yellow	Clear	None
	After Extraction	Test article	Light yellow	Clear	None
		Negative Control	Light yellow	Clear	None

7.2 Test method

Soak a cotton-wool pellet in the sample, record the volume absorbed, and place a pellet in one pouch of each animal. Negative control sample is placed in the other cheek pouch. The duration of exposure shall be that expected for actual use of the material, but no shorter than 5 min. Following the exposure, remove and cotton-wool pellet and wash the pouch with physiological saline solution, taking care not to contaminate the other pouch. Repeat the above procedure every hour (±0.1 h) for 4 h.

8.0 The results observed

Examine the pouches macroscopically following removal of the pellets and, if repeated applications are required, immediately prior to the next dosing. Describe the appearance of the cheek pouches for each animal and grade the pouch surface reactions for erythema according to the system given in Table 1 for each animal at each time interval. Record the results for the test report. At (24 ± 2) h after the final treatment, examine the cheek pouches macroscopically, and humanely sacrifice the hamsters and remove tissue samples from representative areas of the pouches. Place in an appropriate fixative prior to processing for histological examination.

Table 1 Grading system for oral reactions

Reaction	Numerical grading
Erythema and eschar 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
Other adverse changes of the tissues should be recorded and reported.	

9.0 Evaluation criteria

9.1 Macroscopic evaluation

Compare the treated cheek pouch with the cheek pouch on the contralateral side and, if a control group is included, with the pouches of animals in the control group.

The grades (see Table 1) for each observation are added and the sum is divided by the number of observations to determine the average grade per animal.

9.2 Histological evaluation

The irritant effects on oral tissue shall be evaluated microscopically by a pathologist. The pathologist may grade each tissue according to the system given in Table 2.

The grades for microscopic evaluation for all the animals in the test group are added and the sum is divided by the number of observations to obtain a test group average. Repeat for the control group(s). The maximum score is 16.

A total score greater than nine for the microscopic evaluation in the control cheek pouch can indicate underlying pathology or, in a control animal, it can indicate trauma at dosing. Either situation can require a retest if other test or control animals exhibit equivalent high scores.

Subtract the control group average from the test group average to obtain the irritation index (see Table 3).

Table 2 Grading system for microscopic examination for oral tissue reaction

Reaction	Numerical grading
Epithelium	
Normal,intact	0
Cell degeneration or flatting	1
Metaplasia	2
Focal erosion	3
Generalized erosion	4
Leucocyte infiltration (per high power field)	
Absent	0
Minimal (less than 25)	1
Mlid (26 to 50)	2
Moderate (51 to 100)	3
Marked (greater than 100)	4
Vascular congestion	
Absent	0
Minimal	1
Mild	2
Moderate	3
Marked, with disruption of vessels	4
Oedema	
Absent	0
Minimal	1
Mild	2
Moderate	3
Marked	4

Table 3 Irritation index

Average grade	Description of response
0	None
1 to 4	Minimal
5 to 8	Mild
9 to 11	Moderate
12 to 16	Severe
Other adverse changes of the tissues should be recorded and included in the assessment of the response.	

10.0 Results of the test

All animals were survived and no abnormal signs were observed during the study. According to what observed, the response of oral mucosa on testing side did not exceed that on the control side. See table 4~6.

11.0 Conclusion

The test article has no potential oral mucosa irritation in the Syrian hamsters.

12.0 Compliance

US FDA Good Laboratory Practice Regulations 21 CFR 58, effective June 20, 1979, as amended 52 FR 33780, Sept. 4, 1987, and subsequent amendments

Standard operating procedure of CCIC Huatongwei international inspection (Suzhou) Co., Ltd.

13.0 Record

All raw data pertaining to this study and a copy of the final report are retained in designated Huatongwei archive.

14.0 Confidentiality agreement

Statements of confidentiality were as agreed upon prior to study initiation.

15.0 Protocol amendment/deviations

There were no amendments or deviations that occurred during the course of this study.

Table 4 Oral mucosa irritation response observation

Reagent	Animal No.	Pretest weight(g)	Finished weight(g)	Group	Reaction	Interval (hours):score				
						0 h	1 h	2 h	3 h	24 h
SC	1	104.9	108.0	Test Article	Erythema and eschar formation	0	0	0	0	0
				Negative Control	Erythema and eschar formation	0	0	0	0	0
	2	114.5	119.6	Test Article	Erythema and eschar formation	0	0	0	0	0
				Negative Control	Erythema and eschar formation	0	0	0	0	0
	3	106.6	110.8	Test Article	Erythema and eschar formation	0	0	0	0	0
				Negative Control	Erythema and eschar formation	0	0	0	0	0
Primary irritation index：0										
SO	1	107.3	110.4	Test Article	Erythema and eschar formation	0	0	0	0	0
				Negative Control	Erythema and eschar formation	0	0	0	0	0
	2	108.5	113.1	Test Article	Erythema and eschar formation	0	0	0	0	0
				Negative Control	Erythema and eschar formation	0	0	0	0	0
	3	110.2	114.8	Test Article	Erythema and eschar formation	0	0	0	0	0
				Negative Control	Erythema and eschar formation	0	0	0	0	0
Primary irritation index: 0										

Table 5 The score of histological observation

Reagent	Group	Animal No.	Epithelium	Leucocyte infiltration (per high power field)	Vascular congestion	Oedema	Score
SC	Test Article	1	0	0	0	0	0
		2	0	0	1	0	1
		3	0	0	1	0	1
	Negative Control	1	0	0	1	0	1
		2	0	0	1	0	1
		3	0	0	1	0	1
SO	Test Article	1	0	0	1	0	1
		2	0	0	1	0	1
		3	0	0	1	0	1
	Negative Control	1	0	0	1	0	1
		2	0	0	1	0	1
		3	0	0	1	0	1

Table 6 The irritation index of the test articles

Reagent	Animal No.	Score		Irritation index
		Test Article	Negative Control	
SC	1	0	1	0.0
	2	1	1	
	3	1	1	
SO	1	1	1	0.0
	2	1	1	
	3	1	1	