RCC PROJECT 651497

DETERMINATION OF PHOTOALLERGENICITY WITH

CGF-C-1607

IN ALBINO GUINEA PIGS
(INCLUDING INFORMATION ABOUT ALLERGENICITY,
PHOTOIRRITATION AND IRRITATION)

REPORT

Author: G. Arcelin

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Study Completion: 10-JUL-1997

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1. PREFACE

1.1. GENERAL

Title	Determination of Photoallergenicity with CGF-C-1607 in Albino Guinea Pigs (Including Information about Allergenicity, Photoirritation and Irritation).
Sponsor	Ciba Chemikalien GmbH Werk Grenzach Geb. 9001.6.23 Postfach 1266 D-79630 Grenzach/Wyhlen
Monitoring Scientist	Dr. U. Mentzel
Testing Facility	RCC, Research & Consulting Company Ltd. Zelgliweg 1, CH-4452 Itingen / Switzerland
RCC Project Number	651497
Test Article	CGF-C-1607
Test System	Albino Guinea Pigs

1.2. PROJECT STAFF

Study Director	G. Arcelin
Technical Coordinator	R. Sacher

1.3. SCHEDULE

Delivery of the animals	04-MAR-1997 12-MAR-1997
Acclimatization	12-MAR-1997 to 17-MAR-1997
Treatment / Observation	18-MAR-1997 to 11-APR-1997
Termination	11-APR-1997
Reported	10-JUL-1997

1.4. ARCHIVING

Research & Consulting Company Ltd., CH-4452 Itingen will archive the following data for at least 10 years: protocol, report, raw data and test article reference sample. No data will be discarded without the sponsor's consent.

1.5. PROJECT STAFF SIGNATURES

Study Director: G. Arcelin

Date: 10-JUL-1997

Management: for T.R. Allen

Date: 10-Jul-97

1.6. QUALITY ASSURANCE STATEMENT

RCC, Research & Consulting Company Ltd., CH-4452 Itingen / Switzerland

PROJECT NUMBER	:	651497
TEST ARTICLE	:	CGF-C-1607
STUDY DIRECTOR	:	G. Arcelin
TITLE	:	Determination of Photoallergenicity with CGF-C-1607 in Albino Guinea Pigs.

Study procedures were periodically inspected and this report was audited by the RCC Quality Assurance Unit. The dates are given below:

Dates of QAU Inspections / Audits	Dates of Reports to the Study Director and to Management
10-MAR-1997 21-MAR-1997 01-JUL-1997	10-MAR-1997 21-MAR-1997 01-JUL-1997

Manager, Quality Assurance Unit: (for) Dr. G. Menne

M. Döring

Date: 10 Jul 1997

GOOD LABORATORY PRACTICE

1.7. STATEMENT OF COMPLIANCE / GLP GUIDELINES

PROJECT NUMBER	:	651497
TEST ARTICLE	:	CGF-C-1607
STUDY DIRECTOR	:	G. Arcelin
TITLE	:	Determination of Photoallergenicity with CGF-C-1607 in Albino Guinea Pigs.

The study described in this report was conducted in compliance with the following Good Laboratory Practice Standards:

Good Laboratory Practice (GLP) in Switzerland, Procedures and Principles, March 1986.

OECD Principles of Good Laboratory Practice, Environment Monograph Number 45. OECD Series on Principles of Good Laboratory Practice and Compliance Monitoring - Number 1. Environment Directorate, Organisation for Economic Co-operation and Development, Paris 1992.

There were no circumstances that may have affected the quality or integrity of the data.

Study Director: G. Arcelin

Date: 10-JUL-1997

1.8. TEST GUIDELINES

The study procedures described in this report are based on the following guidelines:

CTFA Safety Testing Guidelines, The Cosmetic, Toiletry and Fragrance Association, Inc. Washington, D.C. 20036; "Guidelines for Evaluating Photodermatitis", 1991.

OECD Guidelines for Testing of Chemicals. Draft proposal for a New Guideline: "Acute Dermal Photoirritation Dose-Response Test", February 1995.

1.9. REFERENCES

Harber, L.C. and Shalita, A.R. (1975).
"The Guinea Pig as an Effective Model for the Demonstration of Immunologically Mediated Contact Photosensitivity". In Animal Models in Dermatology (H. Maibach, Ed.) Ch. 10, Churchill Livingstone, New York.

1.10. SUMMARY OF PROTOCOL DEVIATIONS

The test article is stable in PEG 400 for at least 48 hours.

The grading scale was used according to the CTFA Testing Guidelines.

The application sites were depilated approximately three hours prior to the challenge reading.

The viability/mortality and clinical signs were recorded on data sheet from acclimatization start to the end of the study.

2. SUMMARY

In order to assess the photoallergenic potential of CGF-C-1607 a photoallergenicity test according to the CTFA Safety Testing Guidelines was carried out in 30 female (20 test and 10 control) Dunkin Hartley guinea pigs.

For the induction of sensitization the CGF-C-1607 dissolved in polyethylene glycol 400 was applied epicutaneously at the concentration of 30% to a skin area of 8 cm^2 (marked previously with 4 intradermal injections of Freund's Complete Adjuvant). The test sites were then exposed to 1.8 J/cm^2 UV-B and 10 J/cm^2 UV-A irradiation. This procedure was repeated 4 times within 2 weeks of the induction phase. Control animals were treated with FCA/physiological saline only. Three weeks after beginning of the induction a challenge was carried out by treating the experimental animals (test and control) epicutaneously on both flanks with the test article at the concentrations of 30 %, 25 %, 15 % and 10 % (dilutions in polyethylene glycol 400). Treated sites were then either exposed to 10 J/cm^2 UV-A irradiation (left flank) or remained unirradiated (right flank). Cutaneous reactions, i.e. erythema and oedema formation were evaluated at 24, 48 and 72 hours after the challenge exposure.

3. SUMMARY OF RESULTS

The highest non-irritating concentration used for the challenge application was 30 % (determined under RCC project 651475).

ERYTHEMA REACTIONS AFTER THE CHALLENGE

	IRRADIATED			NON-IRRADIATED		
	positive / total			positive / total		
	% positive of total			% positive of total		
	24 hrs	48 hrs	72 hrs	24 hrs	48 hrs	72 hrs
TEST GROUP						
CGF-C-1607, 30 %	0/20 0	0/20 0	0/20 0	0/20 0	0/20 0	0/20 0
CGF-C-1607, 25 %	0/20 0	0/20 0	0/20 0	0/20 0	0/20 0	0/20 0
CGF-C-1607, 15 %	0/20 0	0/20 0	0/20 0	0/20 0	0/20 0	0/20 0
CGF-C-1607, 10 %	0/20 0	0/20 0	0/20 0	0/20 0	0/20 0	0/20 0
CONTROL GROUP						
CGF-C-1607, 30 %	0/10 0	0/10 0	0/10 0	0/10 0	0/10 0	0/10 0
CGF-C-1607, 25 %	0/10 0	0/10 0	0/10 0	0/10 0	0/10 0	0/10 0
CGF-C-1607, 15 %	0/10 0	0/10 0	0/10 0	0/10 0	0/10 0	0/10 0
CGF-C-1607, 10 %	0/10 0	0/10 0	0/10 0	0/10 0	0/10 0	0/10 0

No toxic symptoms were evident in the guinea pigs of the control or test group.

4. CONCLUSION

CGF-C-1607 possesses no photoallergenic and allergenic potential in the guinea pig under the study conditions. The results presented for the control group may lead to the conclusion that CGF-C-1607 showed no irritation potency (see non-irradiated site) respectively showed no phototoxic potency (see irradiated site).

5. OBJECTIVE

5.1. PURPOSE AND RATIONALE

The purpose of this photoallergenicity study was to assess the photoallergenic potential of CGF-C-1607 when applied to albino guinea pigs.

Statements were provided on the photoallergenicity, allergenicity, photoirritation and irritation potential of the test article.

Guinea pigs were selected as test species because in OECD Draft Proposal (02/1995) it is stated that "Although several mammalian species including albino rabbits or rats can be used, the guinea pig is the preferred species ...".

This study should provide a rational basis for risk assessment of the photoallergenic potential of the test article in man.

6. MATERIALS AND METHODS
Experimental Design

6.1. TEST SYSTEM

Test system	Albino Dunkin Hartley Guinea Pig, HsdPoc: DH, SPF
Rationale	Recognized by the international guidelines as a recommended test system (e.g. CTFA).
Source	Harlan Nederland B.V. Postbus 167 NL-3700 AD Zeist / The Netherlands Woundenbergseweg 55 NL-3707 HW Zeist / The Netherlands
Total number of animals	30 females, nulliparous and non-pregnant
Age at start of acclimatization	5 - 8 weeks
Body weight at start of acclimatization	303 - 451 g
Identification	By unique cage number and corresponding ear tags.
Randomization	Randomly selected at time of delivery.

Acclimatization — One week under test conditions, after health examination. Only animals without any visible signs of illness were used for the study.

The animals were distributed as follows:

	NUMBER OF ANIMALS PER GROUP	ANIMAL NUMBERS PER GROUP
1 Control Group	10	445 - 454
2 Test Group	20	455 - 474

The sensitivity and reliability of the experimental technique used was assessed with 3,3',4',5-TETRACHLOROSALICYLANILIDE which is known to have photoallergenic potential in the guinea pig strain. The most recent test was performed from 16-APR-1997 to 15-MAY-1997 (RCC project 901991). The results are described under the Appendix E.

6.2. HUSBANDRY

Room no. — E 22 / RCC

Conditions — **Standard Laboratory Conditions**
Air-conditioned with 10-15 air changes per hour and continuously monitored environment with a target range for room temperature of 22 ± 3 °C and for relative humidity of 40-70 % (values above 70 % during cleaning process possible). The animals were provided with a 12-hour light, 12-hour dark cycle. Fluorescent "Gold" lamps (Silvania Gold F40T1260) were used. Music was played during the daytime light period.

Accommodation — With the exception of the exposure period the animals were kept individually in type-3 wire mesh cages.
During the induction exposure with UV-B and UV-A irradiation: 5 animals / wire-mesh cage (25 x 35 cm).
During the challenge exposure with UV-A irradiation: 5 control animals and 10 test animals were positioned on the right flank in a plastic box (32 x 52 cm) and irradiated together. Successively the remaining 5 control and 10 test animals were irradiated.

Diet — Pelleted standard Nafag Ecosan 845 25W4, batch nos. 01/97 and 10/97 guinea pig breeding/maintenance diet ("Nafag", Nähr- und Futtermittel AG, CH-9202 Gossau), *ad libitum*. Results of analyses for contaminants are included in this report.

Water	Community tap water from Itingen, *ad libitum*. Once weekly additional supply of ascorbic acid (approx. 1 g/l) via the drinking water. Results of bacteriological, chemical and contaminant analyses are included in this report.

6.3. TEST ARTICLE (ACCORDING TO INFORMATION PROVIDED BY THE SPONSOR)

Identification	CGF-C-1607
Description	Yellow solid
Batch number	002
Purity	> 98 %
Stability of test article	Stable under specified storage conditions; expiration date: 31-AUG-1998
Stability of test article dilution	Stable in polyethylene glycol (PEG 400) for at least 48 hours.
Storage conditions	In the original container at room temperature away from direct sunlight at RCC.
Safety precautions	Gloves, goggles and face mask were obligatory to assure personnel health and safety.

6.4. TEST ARTICLE PREPARATION

The test article was placed into a glass beaker on a tared Mettler PM 460 balance and the vehicle (polyethylene glycol 400) was added. A weight by weight dilution was prepared using a glass rod, magnetic stirrer or Ultra-Turrax (Janke & Kunkel, D-79219 Staufen) as homogenizer. Homogeneity of the test article in the vehicle was maintained during treatment.

The preparation was made shortly before dosing.

6.5. LIGHT SOURCE

Induction

UV-A - 320 - 400 nm - 10 J/cm^2 - Philips Actinic "TLD" Lamps (36W/08)

UV-B - 280 - 320 nm - 1.8 J/cm^2 - with Philips UV-B-Sunlamp TL 20W/12

Challenge

UV-A - 320 - 400 nm - 10 J/cm^2 - Philips Actinic "TLD" Lamps (36W/08)

The duration of the exposure was regulated by a time control device.

6.6. RATIONALE

To determine the influence of UV-A and/or UV-B irradiation to the skin after test article treatment.

6.7. PRETEST

Previously a phototoxicity test was performed at RCC with the same test article (RCC 651475). The results from this study were used for challenge applications.

6.8. TEST PROCEDURE

INDUCTION (Test day 1 - 11)

On test day 1 the nuchal skin area of the test group was shaved and a test site of 6-8 cm^2 was defined by four 0.1 ml injections intradermally (i.d.) of Freund's Complete Adjuvant and physiological saline 1:1 into the corners (see Appendix B). 0.1 ml of the 30 % test article dilution in polyethylene glycol 400 were then epicutaneously applied to a skin area of 8 cm^2 (marked with a circular stamp). The test site was exposed to 1.8 J/cm^2 UV-B irradiation and 10 J/cm^2 UV-A. The mixture of Freund's Complete Adjuvant and physiological saline (1:1) was injected only once, i.e. at the time of the initial induction exposure. The remaining procedure (topical application of the test article followed by irradiation) was repeated four times within 2 weeks (days 3, 7, 9 and 11).

At day 1 of induction the control animals received 4 injections intradermally (i.d.) of Freund's Complete Adjuvant and physiological saline 1:1 (0.1 ml) into the corners of the test site of 6-8 cm^2 (nuchal skin areas, see Appendix B). No additional applications were performed during induction. The animals remained untreated.

CHALLENGE

Three weeks after beginning of the induction procedure the guinea pigs of the control and test groups were prepared for challenging. One day before application the animals were fasted but with free access to water and both flanks were shaved. One day later the animals were sedated with COMBELEN (0.2 ml/kg) and anesthetized with VETANARCOL (0.2 ml/kg) by intraperitoneal injection and the test sites of 2 cm^2 were marked with a stamp. Concentrations of 30 %, 25 %, 15 % and 10 % (in polyethylene glycol 400) of the test article were applied to the left flank at a dose of 0.025 ml/2 cm^2. The left flank was then exposed to 10 J/cm^2 UV-A irradiation only. After irradiation of the left flank, the right flank was treated with the test article accordingly without irradiation. The allocation of the different test dilutions to the sites on the animals was alternated in order to minimize the site to site variations in responsiveness. Which test site was used for the different concentrations is described in the tables attached (Appendix A).

The control animals were treated in the same way as described above.

Approximately three hours prior to the first reading the application sites were depilated with an approved depilatory cream (VEET Cream, Reckitt & Colman AG, CH-4123 Allschwil), so that possible erythema reactions were clearly visible at that time.

The depilatory cream was placed on the patch sites and surrounding areas, and left on for 3-5 minutes. It was then thoroughly washed off with a stream of warm, running water. The animals were then dried with a disposable towel, and returned to their cages.

Each animal was assessed for reactions at approximately 24, 48 and 72 hours after exposure (left flank) or test article application (right flank).

After treatment the animals were provided with the same diet as described previously.

6.9. READING AND SCORING

Each animal was assessed for reactions at 24, 48 and 72 hours after exposure for scoring.

The following grading scale was used (according to the CTFA Technical Guideline, Safety Testing Guidelines, 1981 - Guidelines for Evaluation Photodermatitis - Experimental Methods, Photoallergy):

0 = no reaction
1 = defined erythema
2 = erythema and defined edema
3 = marked erythema and edema
4 = severe erythema and edema with deep injuries

6.10. OBSERVATIONS

In addition to the sensitizing reactions the following observations and data were recorded during the test and observation period:

Viability / Mortality	Daily from acclimatization start to termination of the study.
Body weights	At acclimatization start, at day 1, prior to the narcosis procedure and at termination of test.
Clinical signs (local / systemic)	Daily from acclimatization start to termination of the study.
Skin reactions	At the time of reactions readings during induction and challenge period.

Records were maintained of all additional and standard observations.

6.11. PATHOLOGY

NECROPSY

No necropsies were performed in the animals sacrificed at termination of the test.

The animals were sacrificed by intraperitoneal injection of NARCOREN (Rhône Merieux GmbH, D-88471 Laupheim) at a dose of at least 5.1 ml/kg body weight (equivalent to 810 mg sodium pentobarbitone/kg body weight) and discarded.

6.12. STATISTICAL ANALYSIS

Mean values with standard deviations were calculated under the body weight tables.

6.13. DATA COMPILATION

The following data were recorded on data sheets and transcribed for compilation and analysis:

clinical signs (local / systemic),
mortality / viability,
skin reactions.

The following data were recorded on-line:

body weights at acclimatization start, at test day 1, prior to the narcosis procedure and at termination of test.

7. RESULTS

7.1. SKIN EFFECTS AFTER INTRADERMAL INDUCTION - PERFORMED ON TEST DAY 1/CONTROL AND TEST GROUP

The usual findings were observed after intradermal application of FCA in physiological saline; the intradermal applications were observed with erythema, oedema, necrotizing dermatitis and exfoliation of encrustation.

7.2. SKIN EFFECTS AFTER EPIDERMAL INDUCTION - PERFORMED ON TEST DAYS 1, 3, 7, 9 and 11 / TEST GROUP

Erythema/oedema were observed from test day 9 to 15 in relationship with the repeated application of the 30 % test article preparation.

7.3. PHOTOALLERGENICITY POTENTIAL AFTER THE CHALLENGE PROCEDURE - PERFORMED ON TEST DAY 22 / CONTROL AND TEST GROUP

No positive reactions were observed on the both irradiated and non-irradiated flanks of the control and test animals treated with the test article at 30 %, 25 %, 15 % and 10 % in polyethylene glycol 400.

See pp. 21-26

7.4. MORTALITY / VIABILITY / MACROSCOPIC FINDINGS

As there were no deaths during the course of the treatment period no necropsies were performed.

7.5. CLINICAL SIGNS, SYSTEMIC

No symptoms of systemic toxicity were observed in the animals.

7.6. BODY WEIGHTS

The body weight of the animals was within the range of physiological variability known for guinea pigs of this strain and age.

See pp. 30-32

APPENDIX A

TABLES OF REACTIONS IN THE CHALLENGE - PERFORMED ON TEST DAY 22

SKIN REACTION - MAIN STUDY / CHALLENGE PERIOD

Test Article: CGF-C-1607

Vehicle: Polyethylene glycol 400

CONTROL GROUP

	LEFT FLANK UV-A irradiated (10 J/cm^2)			RIGHT FLANK non-irradiated		
Hrs. after application	24	48	72	24	48	72
Animal No. / Sex						
445 / Female						
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
446 / Female						
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
447 / Female						
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
448 / Female						
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
449 / Female						
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0

SKIN REACTION - MAIN STUDY / CHALLENGE PERIOD

Test Article: CGF-C-1607

Vehicle: Polyethylene glycol 400

CONTROL GROUP

	LEFT FLANK UV-A irradiated (10 J/cm²)			RIGHT FLANK non-irradiated		
Hrs. after application	24	48	72	24	48	72
Animal No. / Sex						
450 / Female						
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
451 / Female						
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
452 / Female						
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
453 / Female						
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
454 / Female						
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0

SKIN REACTION - MAIN STUDY / CHALLENGE PERIOD

Test Article: CGF-C-1607

Vehicle: Polyethylene glycol 400

TEST GROUP

	LEFT FLANK UV-A irradiated (10 J/cm²)			RIGHT FLANK non-irradiated		
Hrs. after application	24	48	72	24	48	72
Animal No. / Sex						
455 / Female						
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
456 / Female						
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
457 / Female						
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
458 / Female						
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
459 / Female						
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0

SKIN REACTION - MAIN STUDY / CHALLENGE PERIOD

Test Article: CGF-C-1607

Vehicle: Polyethylene glycol 400

TEST GROUP

	LEFT FLANK UV-A irradiated (10 J/cm^2)			RIGHT FLANK non-irradiated		
Hrs. after application	24	48	72	24	48	72
Animal No. / Sex						
460 / Female						
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
461 / Female						
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
462 / Female						
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
463 / Female						
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
464 / Female						
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0

SKIN REACTION - MAIN STUDY / CHALLENGE PERIOD

Test Article: CGF-C-1607

Vehicle: Polyethylene glycol 400

TEST GROUP

	LEFT FLANK UV-A irradiated (10 J/cm²)			RIGHT FLANK non-irradiated		
Hrs. after application	24	48	72	24	48	72
Animal No. / Sex						
465 / Female						
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
466 / Female						
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
467 / Female						
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
468 / Female						
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
469 / Female						
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0

SKIN REACTION - MAIN STUDY / CHALLENGE PERIOD

Test Article: CGF-C-1607

Vehicle: Polyethylene glycol 400

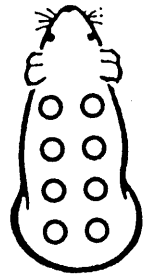
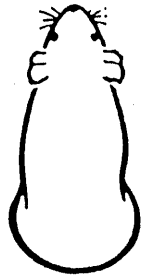
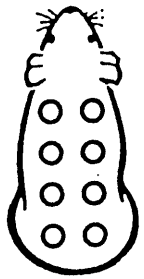
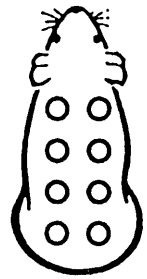
TEST GROUP

	LEFT FLANK UV-A irradiated (10 J/cm^2)			RIGHT FLANK non-irradiated		
Hrs. after application	24	48	72	24	48	72
Animal No. / Sex						
470 / Female						
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
471 / Female						
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
472 / Female						
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
473 / Female						
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0
B = 25 %	0	0	0	0	0	0
474 / Female						
B = 25 %	0	0	0	0	0	0
C = 15 %	0	0	0	0	0	0
D = 10 %	0	0	0	0	0	0
A = 30 %	0	0	0	0	0	0

APPENDIX B

SCHEMA OF THE PHOTOALLERGENICITY STUDY

DETERMINATION OF PHOTOALLERGENICITY IN GUINEA PIGS

ANIMAL-GROUPS	INDUCTION		CHALLENGE
	Day 1	**Days 3, 7, 9 and 11**	**Day 22**
TEST	id 4 x 0.1 ml FCA 50 % 1 x 0.1 ml / 8 cm^2 test substance + UV-A 10J + UV-B 1.8J / cm^2 	4 x 0.1 ml / 8 cm^2 test substance + UV-A 10J + UV-B 1.8J / cm^2 	0.025 ml / 2 cm^2 test substance Photoaller-genicity 10J UV-A/cm^2  Allergenicity no UV
CONTROL	id 4 x 0.1 ml FCA 50 % no UV-irradiation 	no pretreatment no UV-irradiation 	0.025 ml / 2 cm^2 test substance Photoirritation 10J UV-A/cm^2  Irritation no UV
POSITIVE CONTROL	id 4 x 0.1 ml FCA 50 % 1 x 0.1 ml / 8 cm^2 TCSA (3 %) + UV-A 10J + UV-B 1.8J / cm^2 	4 x 0.1 ml / 8 cm^2 TCSA (3 %) + UV-A 10J + UV-B 1.8J / cm^2 	0.025 ml / 2 cm^2 TCSA (0.1 %, 0.03 %, 0.01 % and 0.003 %) 10J UV-A/cm^2  no UV

APPENDIX C

BODY WEIGHTS

- SUMMARY

- INDIVIDUAL

RCC PROJECT 651497
CGF C-1607

BODY WEIGHTS (GRAM) SUMMARY
FEMALES

ACCLIMATIZATION			GROUP 1 CONTROL GROUP	GROUP 2 TEST GROUP
DAY	1	MEAN	373	374
WEEK	1	ST.DEV.	55.5	39.9
		MINIMUM	303	310
		MAXIMUM	436	451
		N	10	20

RCC PROJECT 651497
CGF C-1607

BODY WEIGHTS (GRAM) SUMMARY
FEMALES

TREATMENT			GROUP 1 CONTROL GROUP	GROUP 2 TEST GROUP
DAY	1	MEAN	421	429
WEEK	1	ST.DEV.	44.2	50.9
		MINIMUM	351	357
		MAXIMUM	495	531
		N	10	20
DAY	22	MEAN	567	560
WEEK	4	ST.DEV.	49.5	55.2
		MINIMUM	491	475
		MAXIMUM	625	716
		N	10	20
DAY	25	MEAN	541	539
WEEK	4	ST.DEV.	53.7	55.5
		MINIMUM	461	451
		MAXIMUM	610	671
		N	10	20

BODY WEIGHTS (GRAM)
FEMALES

	ACCLIMATIZATION	TREATMENT		
DAYS	1	1	22	25
WEEKS	1	1	4	4
ANIMAL				
GROUP 1 (CONTROL GROUP)				
445	395	425	491	463
446	428	463	535	506
447	427	495	596	579
448	436	351	511	520
449	433	465	600	552
450	303	366	517	461
451	327	418	618	610
452	324	406	625	574
453	332	423	610	608
454	322	402	563	533
GROUP 2 (TEST GROUP)				
455	350	386	475	477
456	342	397	568	558
457	329	395	532	543
458	450	531	653	624
459	451	511	599	566
460	377	424	567	567
461	356	415	586	586
462	429	469	555	517
463	373	424	573	556
464	411	522	716	671
465	385	429	564	562
466	339	392	551	471
467	355	404	610	490
468	347	396	507	489
469	417	475	543	546
470	351	398	523	501
471	350	377	497	451
472	310	357	509	485
473	356	401	532	523
474	395	482	546	593

APPENDIX D

WATER ANALYSES

- BACTERIOLOGICAL ASSAY OF DRINKING WATER

- CHEMICAL WATER ANALYSIS

- CONTAMINANT ASSAY OF DRINKING WATER

CONTAMINANT ANALYSIS OF FOOD

- ANALYTICAL TEST REPORT

RCC PROJECT 651497
CGF-C-1607

BACTERIOLOGICAL ASSAY OF DRINKING WATER, ITINGEN

Official Laboratory	Liestal, 23.10.96
Basel-Landschaft	Ref.no. 96100286
Sampling point:	59.99.N Net water RCC, Room No. 10
Sampled on:	23.10.96

Sample:

Time of sampling	08.45 a.m.
Water temperature (°C)	15.0

BACTERIOLOGICAL TEST:

Aerobic mesophilic bacteria (per ml)	9
E.coli (per 100 ml)	0
Enterococci (per 100 ml)	0

ASSESSMENT:

At the time of sampling, the tested bacteriological parameters met the requirements for drinking water according to article 260 of the "Eidg. Lebensmittelverordnung".

Official Laboratory
The Official Chemist

(signed Dr. W. Stutz)

RCC PROJECT 651497
CGF-C-1607

CHEMICAL WATER ANALYSIS, ITINGEN

Official Laboratory
Basel-Landschaft

Liestal, 23.10.96
Ref.no. 96100286

Sampling point: 59.99.N Net water RCC, Room No. 10

Sampled on: 23.10.96

Time of sampling 08.45 a.m.
Water temperature (°C) 15.0

CHEMICAL TEST:

Appearance			clear, colourless
Odor			not remarkable
Taste			not remarkable
Total hardness		fr.H°	38.7
Alkaline hardness		fr.H°	26.3
Non carbonate hardness		fr.H°	12.4
Conductivity		µS/cm	694.0
Oxygen demand	($KMnO_4$ cons.)	mg/l	2.4
Free ammonia	NH_4^+	mg/l	< 0.01
Nitrite	NO_2^-	mg/l	< 0.002
Nitrate	NO_3^-	mg/l	22.8
Chloride	Cl^-	mg/l	21.4
Calcium	Ca^{++}	mg/l	134.5
Magnesium	Mg^{++}	mg/l	12.5
Phosphate		mg/l	< 0.005

ASSESSMENT:

At the time of sampling, the tested chemical parameters met the requirements for drinking water according to article 260 of the "Eidg. Lebensmittelverordnung".

Official Laboratory
The Official Chemist

(signed Dr. W. Stutz)

RCC PROJECT 651497
CGF-C-1607

CONTAMINANT ASSAY OF DRINKING WATER, ITINGEN

RCC Project: 900720

Date of Sampling: 23.10.1996

Sample: H_2O I of room 10, 1st ground-floor, 8.45 h., 15 °C

PARAMETER	ASSAY LEVEL µg/l	LIMIT* µg/l
Lindane	< 0.05	0.1
Heptachlor	< 0.05	0.1
Malathion	< 0.05	0.1
DDT, total	< 0.05	0.1
Dieldrin	< 0.05	0.1
Cadmium	< 0.2	5
Arsenic	< 0.15	50
Lead	< 0.25	50
Mercury	< 0.05	1
Selenium	< 0.15	10
Copper	< 0.15	1500
PCBs (28, 52, 101, 138, 153, 180)	< 0.05	0.1
Nitrosamines, total (DMN, DEN, NPIP, NMORPH)	< 0.05	---

< 0.001 = less than 0.001 milligram per kilogram
* Schweizer Lebensmittelbuch

December 10, 1996

K. Biedermann

RCC PROJECT 651497
CGF-C-1607

ANALYTICAL TEST REPORT FOR FEED

RCC Project 647335
09.01.97

Prepared for	EBERLE NAFAG AG NAFAG ECOSAN Postfach 9201 Gossau
Attention of	Mr. F. Wetter
Authorized by	Letter of 07.01.1997
Materials tested	GLP No. 1/97 Nr. 845
Test performed	AAS, GC, GC-MS, HPLC
Test results	See attached Table 1
Submitted	J. Walker
Issued by	K. Biedermann

.....
January 27, 1997/waj

ATTACHMENT

RCC Project 647335
09.01.97

Table 1 - Test Results GLP No. 1/97 Nr. 845

PARAMETER	ASSAY LEVEL mg/kg	LIMIT* mg/kg
Aflatoxins (B1, B2, G1, G2), total	< 0.001	0.005
Estrogens (DES, Hexestrol, Dienestrol), total	< 0.001	0.001
Lindane	< 0.005	0.02
Heptachlor	< 0.005	0.02
Malathion	< 0.5	2.5
DDT, total	< 0.025	0.100
Dieldrin	< 0.005	0.02
Cadmium	0.04	0.160
Arsenic	< 0.15	1.0
Lead	0.50	1.5
Mercury	< 0.05	0.1
Selenium	< 0.15	0.6
Copper	15	---
PCBs	< 0.025	0.05
Nitrosamines (DMN, DEN, NPIP, NMORPH), total	< 0.01	0.010

< 0.001 = less than 0.001 milligram per kilogram
* = USP EPA, Federal Register, Vol. 44, No. 91, May 9, 1979

RCC PROJECT 651497
CGF-C-1607

ANALYTICAL TEST REPORT FOR FEED

RCC Project 649541
29.01.97

Prepared for	EBERLE NAFAG AG NAFAG ECOSAN Postfach 9201 Gossau
Attention of	Mr. F. Wetter
Authorized by	Letter of 28.01.1997
Materials tested	GLP No. 10/97 Nr. 845
Test performed	AAS, GC, GC-MS, HPLC
Test results	See attached Table 1
Submitted	J. Walker
Issued by	K. Biedermann

K. Biedermann

February 06, 1997/waj

ATTACHMENT

RCC Project 649541
29.01.97

Table 1 - Test Results GLP No. 10/97 Nr. 845

PARAMETER	ASSAY LEVEL mg/kg	LIMIT* mg/kg
Aflatoxins (B1, B2, G1, G2), total	< 0.001	0.005
Estrogens (DES, Hexestrol, Dienestrol), total	< 0.001	0.001
Lindane	< 0.005	0.02
Heptachlor	< 0.005	0.02
Malathion	< 0.5	2.5
DDT, total	< 0.025	0.100
Dieldrin	< 0.005	0.02
Cadmium	0.06	0.160
Arsenic	0.16	1.0
Lead	0.59	1.5
Mercury	< 0.05	0.1
Selenium	0.18	0.6
Copper	14	—
PCBs	< 0.025	0.05
Nitrosamines (DMN, DEN, NPIP, NMORPH), total	< 0.01	0.010

< 0.001 = less than 0.001 milligram per kilogram
* = USP EPA, Federal Register, Vol. 44, No. 91, May 9, 1979

APPENDIX E

RESULTS OF POSITIVE CONTROL

RCC PROJECT 651497
CGF-C-1607

RCC PROJECT 901991

DETERMINATION OF PHOTOALLERGENICITY WITH

3, 3', 4', 5-TETRACHLOROSALICYLANILIDE

IN ALBINO GUINEA PIGS

(INCLUDING INFORMATION ABOUT
ALLERGENICITY, PHOTOIRRITATION AND IRRITATION)

POSITIVE CONTROL

SUMMARY

For validation of sensitivity of test method and test system used, a known photosensitizer (3, 3', 4', 5-TETRACHLOROSALICYLANILIDE) described below was selected as a positive control. This was performed in accordance with CTFA Safety Testing Guidelines, The Cosmetic, Toiletry and Fragrance Association, Inc. Washington, D.C. 20036; "Guidelines for Evaluating Photodermatitis", 1991.

The raw data from this project are kept in a separate file at RCC. The test described was performed under GLP-conditions with a final QA-check.

Test article description:

Identification	3, 3', 4', 5-TETRACHLOROSALICYLANILIDE
Description	Beige crystalline powder
Batch number	A0096568
Purity	99 %
Stability of test article	Stable under storage conditions; expiration date: 20-JUN-2000
Stability of test article dilution	Unknown in ethanol.
Storage conditions	At room temperature
Safety precautions	Gloves, goggles and face mask were sufficient to assure personnel health and safety.

Test system:

Albino Dunkin Hartley Guinea Pig, HsdPoc: DH, SPF
delivered by

Harlan Nederland B.V.
Postbus 167
NL-3700 AD Zeist / The Netherlands

Woundenbergseweg 55
NL-3707 HW Zeist / The Netherlands

According to the procedures used in this experiment (run from 16-APR-1997 to 15-MAY-1997) positive results were observed in the treated animals after the epidermal challenge application.

ERYTHEMA REACTIONS AFTER THE CHALLENGE

	IRRADIATED			NON-IRRADIATED		
	positive / total			positive / total		
	% positive of total			% positive of total		
	24 hrs	48 hrs	72 hrs	24 hrs	48 hrs	72 hrs
TEST GROUP						
3,3',4',5-TETRACHLORO-SALICYLANILIDE, 0.1 %	16/20	16/20	16/20	4/20	4/20	2/20
	80	80	80	20	20	10
3,3',4',5-TETRACHLORO-SALICYLANILIDE, 0.03 %	12/20	13/20	13/20	0/20	0/20	0/20
	60	65	65	0	0	0
3,3',4',5-TETRACHLORO-SALICYLANILIDE, 0.01 %	10/20	12/20	12/20	0/20	0/20	0/20
	50	60	55	0	0	0
3,3',4',5-TETRACHLORO-SALICYLANILIDE, 0.003 %	6/20	8/20	6/20	0/20	0/20	0/20
	30	40	25	0	0	0
CONTROL GROUP						
3,3',4',5-TETRACHLORO-SALICYLANILIDE, 0.1 %	0/10	0/10	0/10	0/10	0/10	0/10
	0	0	0	0	0	0
3,3',4',5-TETRACHLORO-SALICYLANILIDE, 0.03 %	0/10	0/10	0/10	0/10	0/10	0/10
	0	0	0	0	0	0
3,3',4',5-TETRACHLORO-SALICYLANILIDE, 0.01 %	0/10	0/10	0/10	0/10	0/10	0/10
	0	0	0	0	0	0
3,3',4',5-TETRACHLORO-SALICYLANILIDE, 0.003 %	0/10	0/10	0/10	0/10	0/10	0/10
	0	0	0	0	0	0

No toxic symptoms were evident in the guinea pigs of the control or test group.

CONCLUSION

According to the results described above and communicated in the Appendix A a significant difference in the number of animals showing positive reactions within the test group was observed comparing the observations made on the non-irradiated and the irradiated skin sites. No positive skin reactions were observed in the animals of the control group treated with the same concentrations. It is concluded that the test article 3,3',4',5-TETRACHLORO-SALICYLANILIDE exhibits photoallergenic potential in the guinea pig under the study conditions. Looking at the results of the non-irradiated site of the test group animals it could be concluded that 3,3',4',5-TETRACHLOROSALICYLANILIDE showed allergenic potency at the concentrations of 0.1 %. The results presented for the control group may lead to the conclusion that 3,3',4',5-TETRACHLOROSALICYLANILIDE shows no irritation potency (see non-irradiated site) respectively showed no phototoxic potency (see irradiated site).

PHOTOALLERGENICITY POTENTIAL AFTER THE CHALLENGE / PERFORMED ON TEST DAY 22

Test Article: 3,3',4',5-TETRACHLOROSALICYLANILIDE

Vehicle: Ethanol

CONTROL GROUP

	LEFT FLANK UV-A irradiated (10 J/cm^2)			RIGHT FLANK non-irradiated		
Hrs. after application	24	48	72	24	48	72
Animal No. / Sex						
1 / Male						
A = 0.1 %	0	0	0	0	0	0
B = 0.03 %	0	0	0	0	0	0
C = 0.01 %	0	0	0	0	0	0
D = 0.003 %	0	0	0	0	0	0
2 / Male						
D = 0.003 %	0	0	0	0	0	0
A = 0.1 %	0	0	0	0	0	0
B = 0.03 %	0	0	0	0	0	0
C = 0.01 %	0	0	0	0	0	0
3 / Male						
C = 0.01 %	0	0	0	0	0	0
D = 0.003 %	0	0	0	0	0	0
A = 0.1 %	0	0	0	0	0	0
B = 0.03 %	0	0	0	0	0	0
4 / Male						
B = 0.03 %	0	0	0	0	0	0
C = 0.01 %	0	0	0	0	0	0
D = 0.003 %	0	0	0	0	0	0
A = 0.1 %	0	0	0	0	0	0
5 / Male						
A = 0.1 %	0	0	0	0	0	0
B = 0.03 %	0	0	0	0	0	0
C = 0.01 %	0	0	0	0	0	0
D = 0.003 %	0	0	0	0	0	0

PHOTOALLERGENICITY POTENTIAL AFTER THE CHALLENGE / PERFORMED ON TEST DAY 22

Test Article: 3,3',4',5-TETRACHLOROSALICYLANILIDE

Vehicle: Ethanol

CONTROL GROUP

	LEFT FLANK UV-A irradiated (10 J/cm^2)			RIGHT FLANK non-irradiated		
Hrs. after application	24	48	72	24	48	72
Animal No. / Sex						
6 / Male						
D = 0.003 %	0	0	0	0	0	0
A = 0.1 %	0	0	0	0	0	0
B = 0.03 %	0	0	0	0	0	0
C = 0.01 %	0	0	0	0	0	0
7 / Male						
C = 0.01 %	0	0	0	0	0	0
D = 0.003 %	0	0	0	0	0	0
A = 0.1 %	0	0	0	0	0	0
B = 0.03 %	0	0	0	0	0	0
8 / Male						
B = 0.03 %	0	0	0	0	0	0
C = 0.01 %	0	0	0	0	0	0
D = 0.003 %	0	0	0	0	0	0
A = 0.1 %	0	0	0	0	0	0
9 / Male						
A = 0.1 %	0	0	0	0	0	0
B = 0.03 %	0	0	0	0	0	0
C = 0.01 %	0	0	0	0	0	0
D = 0.003 %	0	0	0	0	0	0
10 / Male						
D = 0.003 %	0	0	0	0	0	0
A = 0.1 %	0	0	0	0	0	0
B = 0.03 %	0	0	0	0	0	0
C = 0.01 %	0	0	0	0	0	0

PHOTOALLERGENICITY POTENTIAL AFTER THE CHALLENGE / PERFORMED ON TEST DAY 22

Test Article: 3,3',4',5-TETRACHLOROSALICYLANILIDE

Vehicle: Ethanol

TEST GROUP

	LEFT FLANK UV-A irradiated (10 J/cm^2)			RIGHT FLANK non-irradiated		
Hrs. after application	24	48	72	24	48	72
Animal No. / Sex						
11 / Male						
A = 0.1 %	0	0	0	0	0	0
B = 0.03 %	0	0	0	0	0	0
C = 0.01 %	0	0	0	0	0	0
D = 0.003 %	0	0	0	0	0	0
12 / Male						
D = 0.003 %	0	0	0	0	0	0
A = 0.1 %	1	2	2	0	0	0
B = 0.03 %	1	1	1	0	0	0
C = 0.01 %	0	1	1	0	0	0
13 / Male						
C = 0.01 %	0	0	0	0	0	0
D = 0.003 %	0	0	0	0	0	0
A = 0.1 %	0	0	0	0	0	0
B = 0.03 %	0	0	0	0	0	0
14 / Male						
B = 0.03 %	0	0	0	0	0	0
C = 0.01 %	0	0	0	0	0	0
D = 0.003 %	0	0	0	0	0	0
A = 0.1 %	2	2	1	0	0	0
15 / Male						
A = 0.1 %	1	1	1	0	0	0
B = 0.03 %	0	0	0	0	0	0
C = 0.01 %	0	0	0	0	0	0
D = 0.003 %	0	0	0	0	0	0

PHOTOALLERGENICITY POTENTIAL AFTER THE CHALLENGE / PERFORMED ON TEST DAY 22

Test Article: 3,3',4',5-TETRACHLOROSALICYLANILIDE

Vehicle: Ethanol

TEST GROUP

	LEFT FLANK UV-A irradiated (10 J/cm^2)			RIGHT FLANK non-irradiated		
Hrs. after application	24	48	72	24	48	72
Animal No. / Sex						
16 / Male						
D = 0.003 %	0	0	0	0	0	0
A = 0.1 %	1	1	1	0	0	0
B = 0.03 %	1	1	1	0	0	0
C = 0.01 %	1	1	0	0	0	0
17 / Male						
C = 0.01 %	2	2	2	0	0	0
D = 0.003 %	0	1	1	0	0	0
A = 0.1 %	2	2	2	1	1	1
B = 0.03 %	2	2	1	0	0	0
18 / Male						
B = 0.03 %	0	0	0	0	0	0
C = 0.01 %	0	0	0	0	0	0
D = 0.003 %	0	0	0	0	0	0
A = 0.1 %	2	2	1	0	0	0
19 / Male						
A = 0.1 %	1	2	2	0	0	0
B = 0.03 %	0	1	1	0	0	0
C = 0.01 %	0	1	1	0	0	0
D = 0.003 %	0	0	0	0	0	0
20 / Male						
D = 0.003 %	1	1	0	0	0	0
A = 0.1 %	2	2	2	1	1	0
B = 0.03 %	1	1	1	0	0	0
C = 0.01 %	1	1	1	0	0	0

PHOTOALLERGENICITY POTENTIAL AFTER THE CHALLENGE / PERFORMED ON TEST DAY 22

Test Article: 3,3',4',5-TETRACHLOROSALICYLANILIDE

Vehicle: Ethanol

TEST GROUP

	LEFT FLANK UV-A irradiated (10 J/cm^2)			RIGHT FLANK non-irradiated		
Hrs. after application	24	48	72	24	48	72
Animal No. / Sex						
21 / Male						
C = 0.01 %	2	2	1	0	0	0
D = 0.003 %	2	1	1	0	0	0
A = 0.1 %	2	2	2	1	1	0
B = 0.03 %	2	2	2	0	0	0
22 / Male						
B = 0.03 %	1	2	2	0	0	0
C = 0.01 %	1	1	1	0	0	0
D = 0.003 %	0	1	0	0	0	0
A = 0.1 %	2	2	2	0	0	0
23 / Male						
A = 0.1 %	0	0	0	0	0	0
B = 0.03 %	0	0	0	0	0	0
C = 0.01 %	0	0	0	0	0	0
D = 0.003 %	0	0	0	0	0	0
24 / Male						
D = 0.003 %	1	2	2	0	0	0
A = 0.1 %	2	2	2	0	0	0
B = 0.03 %	2	2	2	0	0	0
C = 0.01 %	2	2	2	0	0	0
25 / Male						
C = 0.01 %	2	2	2	0	0	0
D = 0.003 %	1	1	1	0	0	0
A = 0.1 %	2	2	2	1	2	2
B = 0.03 %	2	2	2	0	0	0

PHOTOALLERGENICITY POTENTIAL AFTER THE CHALLENGE / PERFORMED ON TEST DAY 22

Test Article: 3,3',4',5-TETRACHLOROSALICYLANILIDE

Vehicle: Ethanol

TEST GROUP

	LEFT FLANK UV-A irradiated (10 J/cm^2)			RIGHT FLANK non-irradiated		
Hrs. after application	24	48	72	24	48	72
Animal No. / Sex						
26 / Male						
B = 0.03 %	1	1	1	0	0	0
C = 0.01 %	1	1	1	0	0	0
D = 0.003 %	0	0	0	0	0	0
A = 0.1 %	1	1	1	0	0	0
27 / Male						
A = 0.1 %	2	2	2	0	0	0
B = 0.03 %	1	1	1	0	0	0
C = 0.01 %	1	1	1	0	0	0
D = 0.003 %	1	1	1	0	0	0
28 / Male						
D = 0.003 %	0	0	0	0	0	0
A = 0.1 %	0	0	0	0	0	0
B = 0.03 %	0	0	0	0	0	0
C = 0.01 %	0	0	0	0	0	0
29 / Male						
C = 0.01 %	1	1	1	0	0	0
D = 0.003 %	1	1	0	0	0	0
A = 0.1 %	2	2	2	0	0	0
B = 0.03 %	2	1	1	0	0	0
30 / Male						
B = 0.03 %	1	1	1	0	0	0
C = 0.01 %	0	0	0	0	0	0
D = 0.003 %	0	0	0	0	0	0
A = 0.1 %	1	1	1	0	0	0

APPENDIX F

SUMMARY TABLE OF STUDY INFORMATION AND RESULTS

Test article identification: Name: CGF-C-1607				SUMMARY TABLE		
Batch No.: 002						
SKIN TOLERANCE STUDIES / IMMUNOSTIMULATION (photoallergenicity potential by epidermal administration)				Study No.: 651497		
Photoallergenicity Test				Report date: 10-JUL-1997		
Species/Strain: Himalayan white spotted GP				Number of exp. animals: 30		
Procedure		Administration route/site		Irradiation	Day	Vehicle
Induction (control and test)		intracutaneous/nuchal skin area		----	1	FCA/phys. saline 1:1
Induction (test group)		epicutaneous/nuchal skin area		1.8J/cm^2 UV-B +10J/cm^2 UV-A	1-10	PEG 400
Challenge		epicutaneous/left flank epicutaneous/right flank		10J/cm^2 UV-A none	22	PEG-400
Study group		Control group		Test group		
		Conc. of test art. in %	No. of appl. and dose	Conc. of test art. in %	No. of appl. and dose	
Induction (epicutaneous)	A	----	----	PEG 400	5x100µl/8cm^2	
Challenge	A	30	1x25µl/2cm^2	30	1x25µl/2cm^2	
	B	25		25		
	C	15	1x25µl/2cm^2	15	1x25µl/2cm^2	
	D	10		10		
Sex (f/m)		Female		Female		
Number of animals		10		20		
Test flank / irradiated		left / yes	right / no	left / yes	right / no	
Animals with positive irradiated/ non-irradiated sites	Challenge (24 hrs) A	0(10)	0(10)	0(20)	0(20)	
	Challenge (24 hrs) B	0(10)	0(10)	0(20)	0(20)	
	Challenge (24 hrs) C	0(10)	0(10)	0(20)	0(20)	
	Challenge (24 hrs) D	0(10)	0(10)	0(20)	0(20)	
	Challenge (48 hrs) A	0(10)	0(10)	0(20)	0(20)	
	Challenge (48 hrs) B	0(10)	0(10)	0(20)	0(20)	
	Challenge (48 hrs) C	0(10)	0(10)	0(20)	0(20)	
	Challenge (48 hrs) D	0(10)	0(10)	0(20)	0(20)	
	Challenge (72 hrs) A	0(10)	0(10)	0(20)	0(20)	
	Challenge (72 hrs) B	0(10)	0(10)	0(20)	0(20)	
	Challenge (72 hrs) C	0(10)	0(10)	0(20)	0(20)	
	Challenge (72 hrs) D	0(10)	0(10)	0(20)	0(20)	
Study conducted by the applicant: yes ☐ no ☒						
Study in compliance with GLP: yes ☒ no ☐				QAU inspected: Yes ☒ no ☐		

() = number of animals used

APPENDIX G

GLP-CERTIFICATE

EIDGENÖSSISCHES DEPARTEMENT DES INNERN
DÉPARTEMENT FÉDÉRAL DE L'INTÉRIEUR
DIPARTIMENTO FEDERALE DELL'INTERNO

GLP Compliance Statement

It is hereby certified that

on February 12-16, 1996
February 19-23, 1996
June 14, 1996

the testing facilities of RCC Holding Company Ltd
4414 Füllinsdorf
Switzerland

were inspected by the Federal Office of Public Health, the Federal Office of Environment, Forests and Landscape and the Intercantonal Office for the Control of Medicaments with respect to the compliance with the Swiss GLP Principles. The inspection was performed in agreement with the OECD Guidelines for National GLP Inspections and Audits and comprised the following testing facilities:

- RCC Research and Consulting Company Ltd, Itingen
- RCC Umweltchemie AG, Itingen
- RCC Pharmanalytics Ltd, Itingen
- BRL Biological Research Laboratories Ltd/Microbiology, Füllinsdorf

It was found that the aforementioned testing facilities were operating in compliance with the Swiss Principles of Good Laboratory Practice (Good Laboratory Practice [GLP] in Switzerland, Procedures and Principles, March 1986) at the time they were inspected.

FEDERAL DEPARTMENT OF THE INTERIOR

Bern, July 9, 1996

Ruth Dreifuss
Federal Councillor