Safety Assessment of Phytosteryl Glutamates as Used in Cosmetics

Status: Draft Report for Panel Review
Release Date: May 23, 2022
Panel Meeting Date: June 16-17, 2022

The Expert Panel for Cosmetic Ingredient Safety members are: Chair, Wilma F. Bergfeld, M.D., F.A.C.P.; Donald V. Belsito, M.D.; David E. Cohen, M.D.; Curtis D. Klaassen, Ph.D.; Daniel C. Liebler, Ph.D.; Allan E. Rettie, Ph.D.; David Ross, Ph.D..; Ronald C. Shank, Ph.D.; Thomas J. Slaga, Ph.D.; Paul W. Snyder, D.V.M., Ph.D.; and Susan C. Tilton, Ph.D. The Cosmetic Ingredient Review (CIR) Executive Director is Bart Heldreth, Ph.D. This report was prepared by Wilbur Johnson, Jr., M.S., former Senior Scientific Analyst/Writer, and Regina Tucker, M.S., Scientific Analyst/Writer, CIR.

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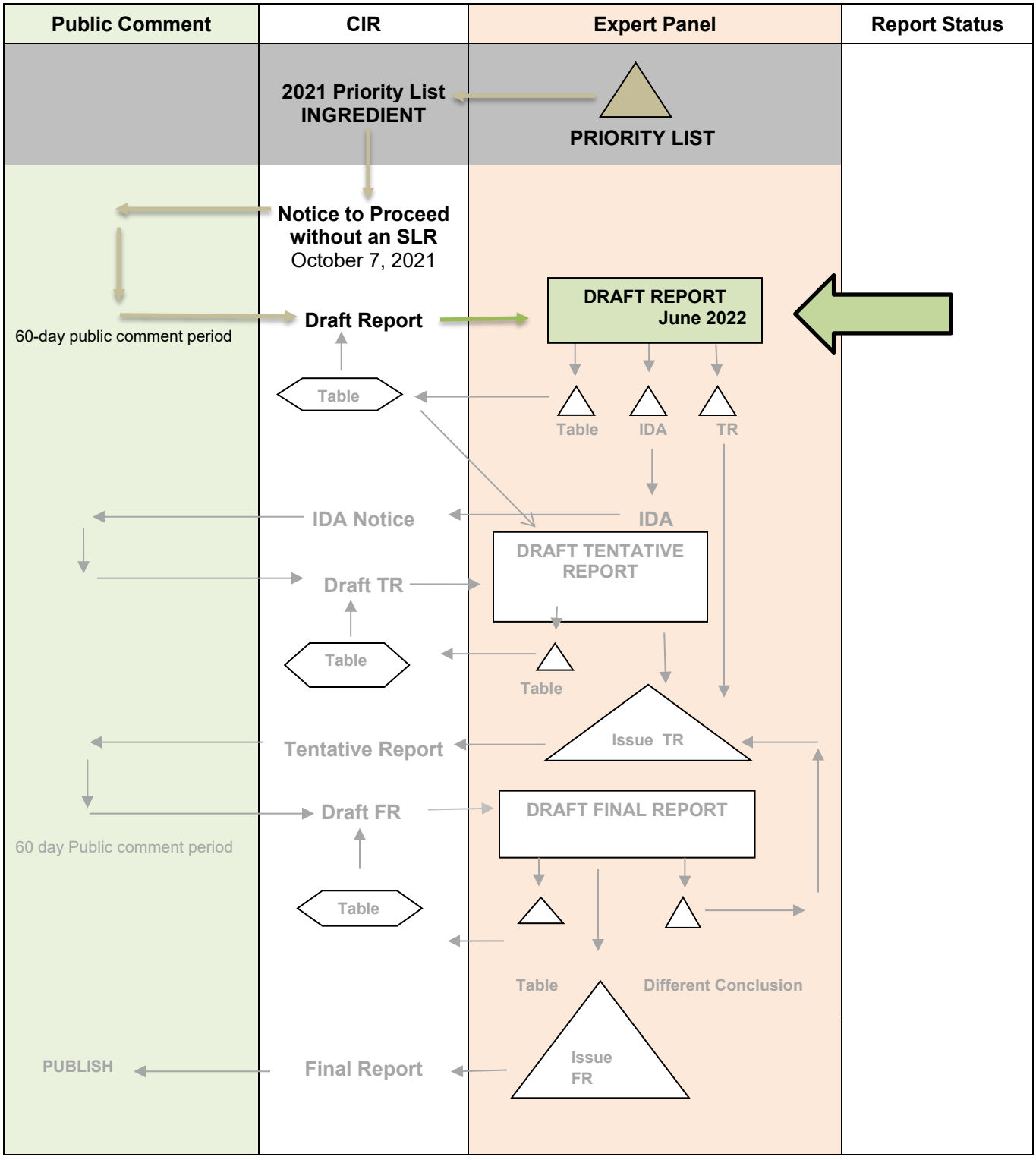
SAFETY ASSESSMENT FLOW CHART

INGREDIENT/FAMILY Phytosteryl Glutamates

MEETING June 2022



Cosmetic Ingredient Review

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Memorandum

To: Expert Panel for Cosmetic Ingredient Safety Members and Liaisons
From: Regina Tucker, M.S., Scientific Analyst/Writer, CIR
Date: May 23, 2022
Subject: Safety Assessment of Phytosteryl Glutamates as Used in Cosmetics

Enclosed is the Draft Report on the Safety Assessment of Phytosteryl Glutamates as Used in Cosmetics (*report_PhytosterylGlutamates_062022*). This ingredient group includes the following 3 phytosteryl glutamates: Phytosteryl/Octyldodecyl Lauroyl Glutamate, Phytosteryl/Behenyl/Octyldodecyl Lauroyl Glutamate, and Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate. The 3 phytosteryl glutamates are mixed esters that all comprise lauroyl glutamic acid esterified with a mixture of phytosterols and fatty alcohols. Following an intensive search of information in the published scientific literature, online databases, and other sources on this ingredient, there was insufficient information found to justify preparation of a formal Scientific Literature Review (SLR). Therefore, in October 2021, CIR issued a SLR Notice to Proceed (NTP) for Phytosteryl Glutamates to alert interested parties that a safety assessment is being prepared and to request information in multiple areas, including:

- Chemistry information, including composition and structure, method of manufacture, and impurity data
- Toxicokinetics data relevant to routes of exposure expected with cosmetic use
- General toxicity data
- Developmental and reproductive toxicity data
- Genotoxicity data
- Carcinogenicity data
- Dermal irritation and sensitization data
- Inhalation toxicity data
- Any other relevant safety information that may be available

Since the issuing of the NTP, the following unpublished data have been received, and are included in this packet:

- Repeated insult patch test on a mixture containing 5.999% Phytosteryl/Octyldodecyl Lauroyl Glutamate *(data2_PhytosterylGlutamates_062022)*
- Primary cutaneous tolerance: Cytotoxicity study performed on an EPISKIN® reconstructed human epidermis model (test mixture containing 1% Phytosteryl/Octyldodecyl Lauroyl Glutamate) *(data2_PhytosterylGlutamates_062022)*

Also included in this package for your review are the CIR report history (*history_PhtyosterylGlutamates_062022*), flow chart (*flow_PhytosterylGlutamates_062022*), literature search strategy (*search_PhytosterylGlutamates_062022*), ingredient data profile (*dataprofile_PhytosterylGlutamates_062022*), 2021 use concentration data (*data1_PhytosterylGlutamates_062022*), and 2022 FDA VCRP data (*VCRP_PhytosterylGlutamates_062022*).

After reviewing these documents, if the available data are deemed sufficient to make a determination of safety, the Panel should issue a Tentative Report with a safe as used, safe with qualifications, or unsafe conclusion, and Discussion items should be identified. If the available data are insufficient, the Panel should issue an Insufficient Data Announcement (IDA), specifying the data needs therein.

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Phytosteryl Glutamates-History

October 2021

A Scientific Literature Review (SLR) Notice to Proceed was issued and the following data was requested:

- Chemistry information, including composition and structure, method of manufacture, and impurities data (including residual monomer content)
- Toxicokinetic data relevant to routes of exposure expected with cosmetic use
- Short-term, subchronic, and chronic dermal/oral toxicity data
- Developmental and reproductive toxicity data
- Genotoxicity data
- Carcinogenicity data
- Dermal irritation and sensitization data at maximum reported use concentrations
- Inhalation toxicity data; and
- Any other relevant safety information that may be available

The following unpublished data was received:

Summary data received for Phytosteryl/Octyldodecyl Lauroyl Glutamate:

- Repeated insult patch test mixture containing 5.999% Phytosteryl/Octyldodecyl Lauroyl Glutamate.
- Primary cutaneous tolerance: Cytotoxicity study performed on an EPISKIN® reconstructed human epidermis model (test mixture containing 1% Phytosteryl/Octyldodecyl Lauroyl Glutamate).

April 2022

The following unpublished data, all received from the Council, have been added to the draft report and are included for the Panel's review:

- Updated (2022) VCRP data
- Human skin irritation study on an epidermis model containing 1% Phytosteryl/Octyldodecyl Lauroyl Glutamate)
- Skin sensitization study (HRIPT) on a test mixture containing 5.999% Phytosteryl/Octyldodecyl Lauroyl Glutamate)

Draft Report, Teams/Panel: June 17-18, 2022

***Phytosteryl Glutamates* Ingredients Data Profile* –June 16-17, 2022 – Wilbur Johnson/Regina Tucker**																															
						Toxico-kinetics		**Acute Tox**			**Repeated Dose Tox**			**DART**		**Genotox**		**Carci**		**Dermal Irritation**			**Dermal Sensitization**				**Ocular Irritation**		**Clinical Studies**	
	Reported Use	**GRAS**	**Method of Mfg**	**Constituents**	**Impurities**	**Dermal Penetration**	**ADME**	**Dermal**	**Oral**	**Inhalation**	**Dermal**	**Oral**	**Inhalation**	**Dermal**	**Oral**	**In Silico**	**In Vivo**	**Dermal**	**Oral**	**In Vitro**	**Animal**	**Human**	**In Vitro**	**Animal**	**Human**	**Phototoxicity**	**In Vitro**	**Animal**	**Case Report**	**Other Clinical Reports**
Phytosteryl/Octyldodecyl Lauroyl Glutamate	**325**																			**X**					**X**					
Phytosteryl/Behenyl/Octyldodecyl Lauroyl Glutamate	**25**																													
Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate	**1**																													

* “X” indicates that data were available in a category for the ingredient

Phytosteryl Glutamates

Ingredient	CAS #	InfoBase	SciFinder	PubMed		FDA	EU	ECHA	IUCLID	SIDS	HPVIS	NICNAS	NTIS	NTP	WHO	FAO	ECE-TOC	Web
Phytosteryl/Octyldodecyl Lauroyl Glutamate	220465-88-3	Yes		0/0		No	No	No	No	No	No	No	No	No	No	No	No	Yes
Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate	No CAS No.	Yes		0/0		No	No	No	No	No	No	No	No	No	No	No	No	Yes
Phytosteryl/Behenyl/Octyldodecyl Lauroyl Glutamate	No CAS No.	Yes		0/0		No	No	No	No	No	No	No	No	No	No	No	No	Yes

Search Strategy

[*document search strategy used for SciFinder, PubMed, and Toxnet*]

[*identify total # of hits /# hits that were useful or examined for usefulness*]

LINKS

InfoBase (self-reminder that this info has been accessed; not a public website) - http://www.personalcarecouncil.org/science-safety/line-infobase
ScfFinder (usually a combined search for all ingredients in report; list # of this/# useful) - https://scifinder.cas.org/scifinder
PubMed (usually a combined search for all ingredients in report; list # of this/# useful) - http://www.ncbi.nlm.nih.gov/pubmed
Toxnet databases (usually a combined search for all ingredients in report; list # of this/# useful) – https://toxnet.nlm.nih.gov/ (includes Toxline; HSDB; ChemIDPlus; DAR; IRIS; CCRIS; CPDB; GENE-TOX)

FDA databases – http://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfcfr/cfrsearch.cfm (CFR); then,
list of all databases: http://www.fda.gov/ForIndustry/FDABasicsforIndustry/ucm234631.htm; then,
https://www.fda.gov/food/food-additives-petitions/substances-added-food-formerly-eafus (Substances added to Food);
http://www.fda.gov/food/ingredientspackaginglabeling/gras/default.htm (GRAS);
https://www.fda.gov/food/generally-recognized-safe-gras/gras-substances-scogs-database (SCOGS database);
http://www.accessdata.fda.gov/scripts/fdcc/?set=IndirectAdditives (indirect food additives list);
http://www.fda.gov/Drugs/InformationOnDrugs/default.htm (drug approvals and database);
http://www.fda.gov/downloads/AboutFDA/CentersOffices/CDER/UCM135688.pdf (OTC ingredient list);
http://www.accessdata.fda.gov/scripts/cder/iig/ (inactive ingredients approved for drugs)

EU (European Union); check CosIng (cosmetic ingredient database) for restrictions and SCCS (Scientific Committee for Consumer Safety) opinions - http://ec.europa.eu/growth/tools-databases/cosing/
ECHA (European Chemicals Agency – REACH dossiers) – http://echa.europa.eu/information-on-chemicals;jsessionid=A978100B4E4CC39C78C93A851EB3E3C7.live1
IUCLID (International Uniform Chemical Information Database) - https://iuclid6.echa.europa.eu/search
OECD SIDS documents (Organisation for Economic Co-operation and Development Screening Info Data Sets)- http://webnet.oecd.org/hpv/ui/Search.aspx
HPVIS (EPA High-Production Volume Info Systems) - https://ofmext.epa.gov/hpvis/HPVISlogon
NICNAS (Australian National Industrial Chemical Notification and Assessment Scheme)- https://www.industrialchemicals.gov.au/chemical-information/search-assessments?assessmentcasnumber=39346-84-4

NTIS (National Technical Information Service) - http://www.ntis.gov/
NTP (National Toxicology Program) - http://ntp.niehs.nih.gov/
WHO (World Health Organization) technical reports - http://www.who.int/biologicals/technical_report_series/en/
FAO (Food and Agriculture Organization of the United Nations) - http://www.fao.org/food/food-safety-quality/scientific-advice/jecfa/jecfa-additives/en/ (FAO);
FEMA (Flavor & Extract Manufacturers Association) - http://www.femaflavor.org/search/apachesolr_search/
Web – perform general search; may find technical data sheets, published reports, etc
ECETOC (European Center for Ecotoxicology and Toxicology Database) - http://www.ecetoc.org/

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ABBREVIATIONS

CFR	Code of Federal Regulations
CIR	Cosmetic Ingredient Review
Council	Personal Care Products Council
CPSC	US Consumer Product Safety Commission
FDA	Food and Drug Administration
MTT	3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl-2H-tetrazolium bromide
HRIPT	human repeated insult patch test
NR	not reported
US	United States
VCRP	Voluntary Cosmetic Registration Program
WHO	World Health Organization
wINCI	web-based *International Cosmetic Ingredient Dictionary and Handbook*

INTRODUCTION

The safety of the following 3 phytosteryl glutamates as used in cosmetics is reviewed in this safety assessment.

Phytosteryl/Behenyl/Octyldodecyl Lauroyl Glutamate
Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate
Phytosteryl/Octyldodecyl Lauroyl Glutamate

According to the web-based *International Cosmetic Ingredient Dictionary and Handbook* (wINCI; *Dictionary*), all 3 phytosteryl glutamates are reported to function in cosmetics as skin conditioning agents, and Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate is also reported to function as a hair conditioning agent (Table 1)[1]. These ingredients are being reviewed together as they are all mixed esters of phytosterols, octyldodecanol (and other respective fatty alcohols), and lauroyl glutamic acid. The Expert Panel for Cosmetic Ingredient Safety (Panel) has previously reviewed the safety of several phytosterols and lauroyl glutamic acid. The phytosterols ingredient group were considered safe as used in the present practices of use and concentration (as described in that safety assessment).[2,3] Lauroyl glutamic acid was reviewed as part of the safety assessment of amino acid alkyl amides.[2] At the time of the assessment lauroyl glutamic acid was not in current use, but the Panel concluded it would be considered safe if used in product categories and at concentrations comparable to others in the group (as described in the safety assessment). The full reports on these ingredients can be accessed on the Cosmetic Ingredient Review (CIR) website (https://www.cir-safety.org/ingredients).

This safety assessment includes relevant published and unpublished data that are available for each endpoint that is evaluated. The published data in this document were identified by conducting an exhaustive search of the world's literature. A list of the search engines and websites that are used, and the sources that are typically explored, as well as the endpoints that the Panel typically evaluates, is available on the CIR website (https://www.cir-safety.org/supplementaldoc/preliminary-search-engines-and-websites; https://www.cir-safety.org/supplementaldoc/cir-report-format-outline). Unpublished data may be provided by the cosmetics industry, as well as by other interested parties.

CHEMISTRY

Definition and Structure

Each of these ingredients comprise 2 core chemical structural residues, phytosterols and lauroyl glutamate. These ingredients also comprise certain fatty alkyl chains. The "/" marks in the names of these ingredients signify mixtures. For example, Phytosteryl/Octyldodecyl Lauroyl Glutamate is a mixture of phytosteryl lauroyl glutamate and octyldodecyl lauroyl glutamate. The term "phytosteryl" also signifies a mixture, specifically of steroidal constituents derived from plants (i.e., "phyto"). The most common phytosterols (β-sitosterol, stigmasterol, avenasterol, campesterol, and campestanol) are illustrated in Figure 1, as is an example of connectivity with lauroyl glutamate.

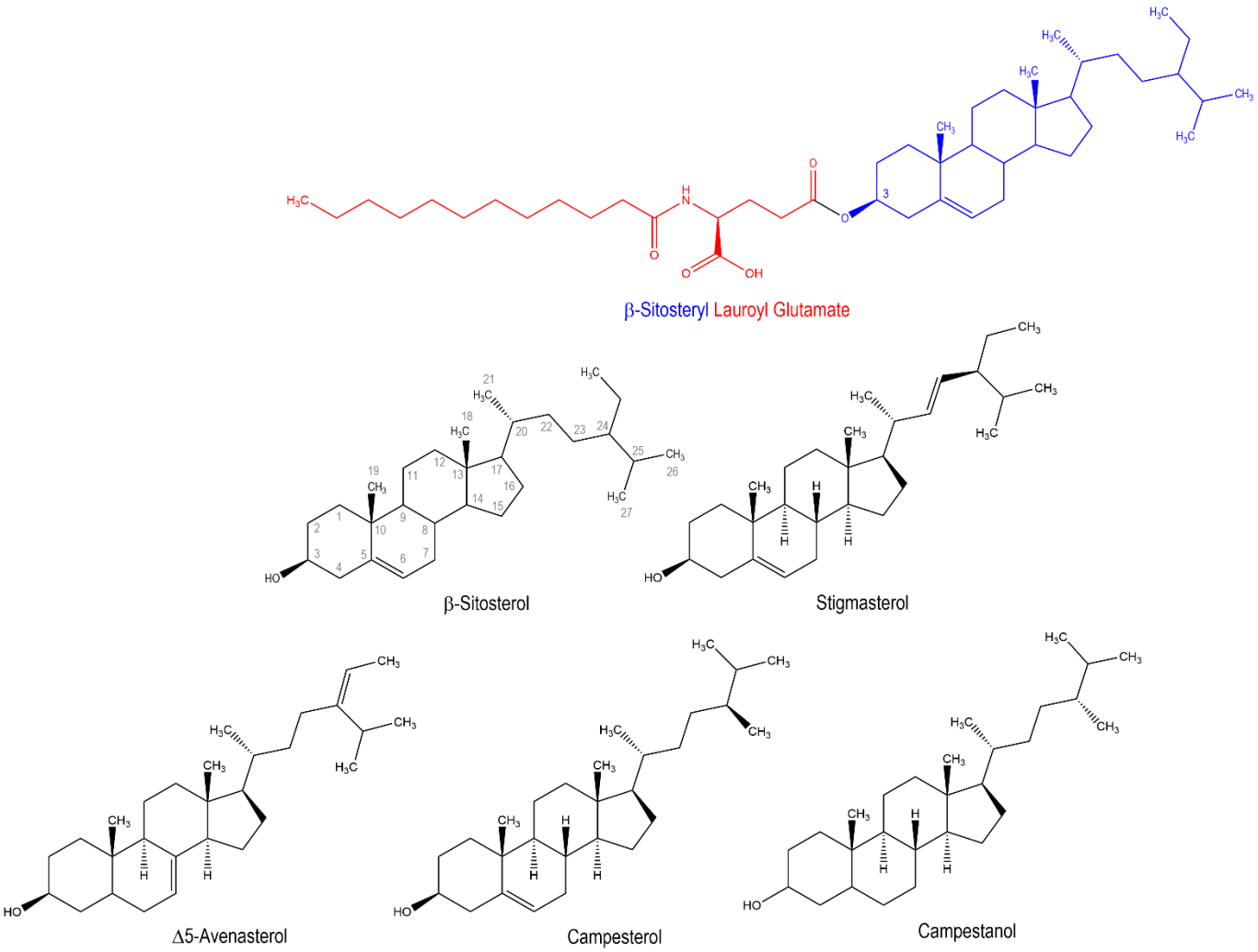


Figure 1. Phytosterols and Phytosteryl Connectivity

All such connectivities are the result of esterification via the 3-position alcohol functional group of one or more phytosterols. The connectivity of various fatty alkyl chains with lauroyl glutamate is similarly the result of esterification (e.g., octyldodecyl lauroyl glutamate (Figure 2)).

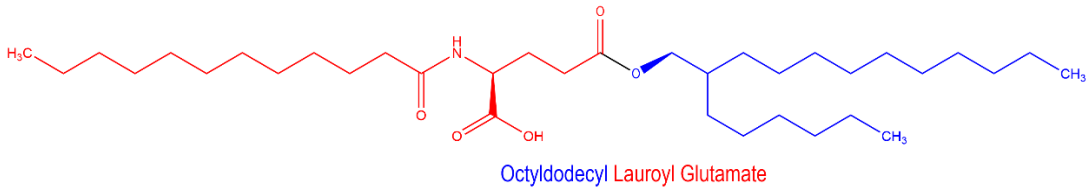


Figure 2. Octyldodecyl Lauroyl Glutamate

Accordingly, Phytosteryl/Octyldodecyl Lauroyl Glutamate is a mixture potentially comprising all of the above instances of esterified lauroyl glutamate. Likewise, Phytosteryl/Behenyl/Octyldodecyl Lauroyl Glutamate and Phytosteryl/Behenyl/ Octyldodecyl/Isostearyl Lauroyl Glutamate comprise similar such mixtures.

The definitions and reported functions of the phytosteryl glutamates included in this safety assessment are presented in Table 1.[1] The only ingredient with a CAS No. in this safety assessment is Phytosteryl/Octyldodecyl Lauroyl Glutamate (220465-88-33).

Chemical Properties

Chemical properties data for these ingredients were neither found in the available literature nor submitted as unpublished data.

Method of Manufacture

No ingredient-specific methods of manufacture were found in the literature or submitted as unpublished data.

Impurities

Impurities data for these ingredients were neither found in the available literature nor submitted as unpublished data.

USE

Cosmetic

The safety of the cosmetic ingredients addressed in this assessment is evaluated based on data received from the US Food and Drug Administration (FDA) and the cosmetics industry on the expected use of these ingredients in cosmetics, and does not cover their use in airbrush delivery systems. Data are submitted by the cosmetic industry via the FDA's Voluntary Cosmetic Registration Program (VCRP) database (frequency of use) and in response to a survey conducted by the Personal Care Products Council (Council) (maximum use concentrations). The data are provided by cosmetic product categories, based on 21CFR Part 720. For most cosmetic product categories, 21CFR Part 720 does not indicate type of application and, therefore, airbrush application is not considered. Airbrush delivery systems are within the purview of the US Consumer Product Safety Commission (CPSC), while ingredients, as used in airbrush delivery systems, are within the jurisdiction of the FDA. Airbrush delivery system use for cosmetic application has not been evaluated by the CPSC, nor has the use of cosmetic ingredients in airbrush technology been evaluated by the FDA. Moreover, no consumer habits and practices data or particle size data are publicly available to evaluate the exposure associated with this use type, thereby preempting the ability to evaluate risk or safety. Therefore, airbrush application of cosmetic products is not assessed by the Panel.

According to 2022 FDA VCRP data, Phytosteryl/Octyldodecyl Lauroyl Glutamate has the greatest frequency of use; it is reported to be used in 325 cosmetic products, 311 of which are leave-on products and over a third of which are in lipstick formulations (Table 2).[4] The results of the concentration of use survey conducted by the Council in 2021 indicate that Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate has the highest concentration of use; it is used at maximum use concentrations up to 25.6% in leave-on products (rouges).[5] The maximum concentration of use reported for Phytosteryl/Octyldodecyl Lauroyl Glutamate is very similar; it is reported to be used at up to 25% in rouges and in lipsticks.

Cosmetic products containing phytosteryl glutamates may incidentally come in contact with the eyes (e.g., Phytosteryl/Octyldodecyl Lauroyl Glutamate at concentrations up to 12% in eye shadow), and all 3 of these ingredients are used in products that are reported to be used in formulations that could be incidentally ingested and that come in contact with mucous membranes (e.g., Phytosteryl/Octyldodecyl Lauroyl Glutamate at concentrations up to 25% in lipstick). Use in baby products is also reported (e.g., Phytosteryl/Octyldodecyl Lauroyl Glutamate is used at up to 0.3% in baby lotions, oils, and creams).

Some of these ingredients are used in cosmetic products that could possibly be inhaled; for example, Phytosteryl/Octyldodecyl Lauroyl Glutamate is reported to be used in hairsprays (concentration not reported), aerosol deodorant (concentrations not reported), and in face powders at concentrations up to 5%. In practice, as stated in the Panel's respiratory exposure resource document (https://www.cir-safety.org/cir-findings), most droplets/particles incidentally inhaled from cosmetic sprays would be deposited in the nasopharyngeal and tracheobronchial regions and would not be respirable (i.e., they would not enter the lungs) to any appreciable amount. There is some evidence indicating that deodorant spray products can release substantially larger fractions of particulates having aerodynamic equivalent diameters in the range considered to be respirable. However, the information is not sufficient to determine whether significantly greater lung exposures result from the use of deodorant sprays, compared to other cosmetic sprays. Conservative estimates of inhalation exposures to respirable particles during the use of loose powder cosmetic products are 400-fold to 1000-fold less than protective regulatory and guidance limits for inert airborne respirable particles in the workplace.

Although products containing some of these ingredients may be marketed for use with airbrush delivery systems, this information is not available from the VCRP or the Council survey. Without information regarding the frequency and concentrations of use of these ingredients (and without consumer habits and practices data or particle size data related to this use technology), the data are insufficient to evaluate the exposure resulting from cosmetics applied via airbrush delivery systems.

The phytosteryl glutamates reviewed in this safety assessment are not restricted from use in any way under the rules governing cosmetic products in the European Union.[6]

TOXICOKINETIC STUDIES

Toxicokinetic data on phytosteryl glutamates were not found in the published literature, and unpublished data were not provided.

TOXICOLOGICAL STUDIES

Toxicological studies on the phytosteryl glutamates were not found in the published literature, and unpublished data were not submitted.

DEVELOPMENTAL AND REPRODUCTIVE TOXICITY STUDIES

Data on the developmental and reproductive toxicity of phytosteryl glutamates reviewed in this safety assessment were neither found in the published literature, nor were these data submitted.

GENOTOXICITY STUDIES

Data on the genotoxicity of phytosteryl glutamates reviewed in this safety assessment were neither found in the published literature, nor were these data submitted.

CARCINOGENICITY STUDIES

Data on the carcinogenicity of phytosteryl glutamates reviewed in this safety assessment were neither found in the published literature, nor were these data submitted.

DERMAL IRRITATION AND SENSITZATION STUDIES

Dermal irritation and sensitization studies are described in Table 3, and summarized below.

According to summary data, an in vitro cell viability assay was performed, using EpiSkin™ reconstituted human epidermis. The test substance, a product containing 1% Phytosteryl Octyldodecyl Lauroyl Glutamate, was predicted to be non-irritating.[7] In a human repeated insult patch test (HRIPT), Phytosteryl/Octyldodecyl Lauroyl Glutamate (5.999%, 219 subjects; tested neat, occlusive patch) was not an irritant or sensitizer.[8]

OCULAR IRRITATON STUDIES

Data on the ocular irritation of phytosteryl glutamates reviewed in this safety assessment were neither found in the published literature, nor were these data submitted.

SUMMARY

The safety of 3 phytosteryl glutamates as used in cosmetics is reviewed in this safety assessment. According to the *Dictionary*, Phytosteryl/Octyldodecyl Lauroyl Glutamate and Phytosteryl/Behenyl/Octyldodecyl Lauroyl Glutamate are reported to function in cosmetics as skin conditioning agents and Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate is reported to function as a hair conditioning agent and skin conditioning agent.

According to 2022 FDA VCRP data, Phytosteryl/Octyldodecyl Lauroyl Glutamate has the greatest frequency of use; it is reported to be used in 325 cosmetic products, (311 leave-on products and 14 rinse-off products). The results of a concentration of use survey conducted by the Council in 2021 indicate Phytosteryl/Octyldodecyl Lauroyl Glutamate has the highest concentration of use; it is used at maximum use concentrations up to 25.6% in leave-on products. The maximum concentration of use reported for Phytosteryl/Octyldodecyl Lauroyl Glutamate is very similar; it is reported to be used at up to 25% in rouges and in lipsticks.

An in vitro skin irritation assay was performed EpiSkin™ reconstituted human epidermis on a test substance containing1% Phytosteryl Octyldodecyl Lauroyl Glutamate. The test substance was predicted to be non-irritating. No sensitization was noted in an HRIPT performed in 219 subjects using a mixture of 5.999% Phytosteryl/ Octyldodecyl Lauroyl Glutamate.

DISCUSSION

To be developed.

CONCLUSION

To be determined.

TABLES

Table 1. Definitions and reported functions of the ingredients in this safety assessment[1]

Ingredient/CAS No.	Definition	Function(s)
Phytosteryl/Octyldodecyl Lauroyl Glutamate 220465-88-3	Phytosteryl/Octyldodecyl Lauroyl Glutamate is the mixed ester of phytosterol and octyldodecanol with lauroyl glutamic acid.	Skin-Conditioning Agents - Occlusive
Phytosteryl/Behenyl/Octyldodecyl Lauroyl Glutamate	Phytosteryl/Behenyl/Octyldodecyl Lauroyl Glutamate is the mixed ester of phytosterol, behenyl alcohol, and octyldodecanol with lauroyl glutamic acid.	Skin-Conditioning Agents - Occlusive
Phytosteryl/Behenyl/Octyldodecyl /Isostearyl Lauroyl Glutamate	Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate is the mixed ester of phytosterols, behenyl alcohol, octyldodecanol and isostearyl alcohol with lauroyl glutamic acid.	Hair Conditioning Agents; Skin-Conditioning Agents - Emollient

Table 2. Frequency (2022)[4] and concentration (2021)[2] of use according to duration and exposure

	# of Uses	Max Conc of Use (%)	# of Uses	Max Conc of Use (%)	# of Uses	Max Conc of Use (%)
	Phytosteryl/Octyldodecyl Lauroyl Glutamate		**Phytosteryl/Behenyl/Octyldodecyl Lauroyl Glutamate**		**Phytosteryl/Behenyl/Octyldodecyl/ Isostearyl Lauroyl Glutamate**	
Totals*	**325**	**0.005-25**	**25**	**NR**	**1**	**0.00028-25.6**
Duration of Use						
Leave-On	*311*	*0.01-25*	*25*	*NR*	*1*	*0.03-25.6*
Rinse-Off	*14*	*0.005-2*	*NR*	*NR*	*NR*	*0.00028-1*
Diluted for (Bath) Use	*NR*	*NR*	*NR*	*NR*	*NR*	*NR*
Exposure Type						
Eye Area	26	0.1-12	4	NR	NR	1-8.6
Incidental Ingestion	133	1-25	1	NR	1	0.1-7
Incidental Inhalation-Spray	1; 94[a]; 40[b]	0.1-2 [a]	4 [a];4 [b]	NR	NR	0.2 [a]
Incidental Inhalation-Powder	40[b]	5; 0.01-8[c]	4 [b]	NR	NR	1; 0.03-5[c]
Dermal Contact	178	0.005-25	24	NR	NR	0.00028-25.6
Deodorant (underarm)	NR	not spray: 0.1 spray: 0.1	NR	NR	NR	NR
Hair - Non-Coloring	13	0.1-2	NR	NR	NR	0.2
Hair-Coloring	NR	NR	NR	NR	NR	NR
Nail	1	NR	NR	NR	NR	NR
Mucous Membrane	133	0.005-25	1	NR	1	0.1-7
Baby Products	NR	0.3	NR	NR	NR	NR

NR = Not Reported

* Because each ingredient may be used in cosmetics with multiple exposure types, the sum of all exposure types may not equal the sum of total uses.

[a] It is possible that these products may be sprays, but it is not specified whether the reported uses are sprays

[b] Not specified these products are sprays or powders, but it is possible the use can be as a spray or powder, therefore the information is captured in both categories

[c] It is possible that these products may be powders, but it is not specified whether the reported uses are powders

Table 3. Irritation and sensitization studies

Test Article	Concentration/Dose	Test Population	Procedure	Results	Reference
IRRITATION					
In Vitro					
Mixture containing 1 % Phytosteryl/ Octyldodecyl Lauroyl Glutamate	150mg ± 5 mg, applied in duplicate	2 different lots of reconstructed human epidermis (EPISKIN)	Epidermis was prepared; a negative and positive control were tested in triplicate. At the end of incubation an MTT test was performed. Samples were plated, biopsied, and the epidermis was separated from the collagen and transferred to tubes. Cell viability was then determined. Acceptability and expression of results followed.	Mean viability greater than 50% is interpreted as being potentially non-irritant and in two samples the mean viability resulted in 81.1% and 72.4% thus this mixture is considered potentially non-irritant.	[7]
SENSITIZATION					
Human					
Mixture containing 5.999% Phytosteryl/ Octyldodecyl Lauroyl Glutamate	0.2 ml applied as supplied.	219 subjects	HRIPT evaluating sensitization potential. During induction, product was placed on an occlusive patch (2 cm x 2 cm), which was applied to the infrascapular area of the back (either to right or left of midline), or to the upper arm. Induction phase consisted of nine 24-h applications made over 4 consecutive weeks. After a 10-15 d non-treatment period, challenge patches were applied for 24 h to previously untreated sites Reactions were scored at 48 h and 72 h after patch removal.	During induction, no reactions were reported, and none were observed for any of the subjects at challenge. Under the conditions employed in this study, there was no evidence of sensitization to the product.	[8]

REFERENCES

1. Nikitakis J, Kowcz A. wINCI: International Cosmetic Ingredient Dictionary and Handbook. http://webdictionary.personalcarecouncil.org/jsp/Home.jsp. Washington, DC: Personal Care Products Coucil. Last Updated: 2020. Accessed: April 1, 2022.

2. Bergfield W, Belsito D, Hill R, et al. Final report on the safety assessment of amino acid alkyl amides as used in cosmetics. Washington, D.C.: Cosmetic Ingredient Review; 2017. https://www.cir-safety.org/ingredients.

3. Bergfield W, Belsito D, Hill RA, et al. Final report on the safety assessment of phytosterols as used in cosmetics. Washington, DC.: Cosmetic Ingredient Review; 2014. https://www.cir-safety.org/ingredients.

4. U.S. Food and Drug Administration (FDA) Center for Food Safety and Applied Nutrition (CFSAN). 2022. Voluntary Cosmetic Registration Program-Frequency of Use of Cosmetic Ingredients. (Obtained under the Freedom of Information Acts from CFSAN; requested as "Frequency of Use Data" January 4, 2022; recieved January 11, 2022). College Park, MD.

5. Personal Care Products Council. 2021. Concentration of use by FDA product category: Phytosteryl Glutamates. Unpublished data submitted byt the Personal Care Products Council on September 8, 2021.

6. European Commission. Cosing database; following Cosmetic Regulation Number 1223/2009. https://ec.europa.eu/growth/tools-databases/cosing/. Last Updated: 2020. Accessed: 9/1/2021.

7. Anonymous. 2008. Primary cutaneous tolerance: Cytoxicity study performed on an EPISKIN reconstructed human epidermis model (test mixture containing 1% Phytosteryl/Octyldodecyl Lauroyl Glutamate). Unpublished data submitted by the Personal Care Products Council on October 14, 2021.

8. Anonymous. 2016. Repeated insult patch test (mixture containing 5.999% Phytosteryl/Octyldodecyl Lauroyl Glutamate). Unpublished data submitted by the Personal Care Products Council on October 14, 2021.

Personal Care Products Council
Committed to Safety, Quality & Innovation

Memorandum

TO: Bart Heldreth, Ph.D.
Executive Director - Cosmetic Ingredient Review

FROM: Carol Eisenmann, Ph.D.
Personal Care Products Council

DATE: September 8, 2021

SUBJECT: Concentration of Use by FDA Product Category: Phytosteryl Glutamates

Concentration of Use by FDA Product Category*

Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate
Phytosteryl/Behenyl/Octyldodecyl Lauroyl Glutamate
Phytosteryl/Octyldodecyl Lauroyl Glutamate

Ingredient	Product Category	Maximum Concentration of Use
Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate	Eye shadows	8.6%
Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate	Eye lotions	1%
Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate	Tonics, dressings, and other hair grooming aids	0.2%
Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate	Blushers	7.3%
Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate	Face powders	1%
Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate	Foundations	1%
Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate	Lipstick	0.1-7%
Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate	Rouges	25.6%
Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate	Other makeup preparations	0.42%
Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate	Skin cleansing (cold creams, cleansing lotions, liquids, and pads)	0.00028-1%
Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate	Face and neck products Not spray	0.03-5%
Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate	Moisturizing products Not spray	0.5%
Phytosteryl/Behenyl/Octyldodecyl/Isostearyl Lauroyl Glutamate	Other skin care preparations	0.5%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Baby lotions, oils, and creams Not powder	0.3%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Eyeliners	7.5%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Eye shadows	12%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Eye lotions	2.5%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Other eye makeup preparations	0.1-4.2%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Hair conditioners	0.1-0.7%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Tonics, dressings, and other hair grooming aids	0.7-2%

Phytosteryl/Octyldodecyl Lauroyl Glutamate	Blushers	5.4%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Face powders	5%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Foundations	2.2-3.1%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Lipstick	1-25%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Makeup bases	1%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Rouges	25%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Other makeup preparations	1%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Bath soaps and detergents	0.005%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Deodorants Not spray Aerosol	 0.1% 0.1%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Skin cleansing (cold creams, cleansing lotions, liquids, and pads)	1-2%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Face and neck products Not spray	 0.3-8%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Body and hand products Not spray	 0.01-1%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Moisturizing products Not spray	 0.1-0.5%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Night products Not spray	1%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Paste masks and mud packs	0.1%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Skin fresheners	0.1-0.5%
Phytosteryl/Octyldodecyl Lauroyl Glutamate	Other skin care preparations	0.1-2%

*Ingredients included in the title of the table but not found in the table were included in the concentration of use survey, but no uses were reported

Information collected in 2021
Table prepared September 8, 2021

Personal Care Products Council
Committed to Safety, Quality & Innovation

Memorandum

TO: Bart Heldreth, Ph.D.
Executive Director - Cosmetic Ingredient Review

FROM: Carol Eisenmann, Ph.D.
Personal Care Products Council

DATE: October 14, 2021

SUBJECT: Phytosteryl/Octyldodecyl Lauroyl Glutamate

Anonymous. 2016. Repeated insult patch test mixture containing 5.999% Phytosteryl/Octyldodecyl Lauroyl Glutamate.

Anonymous. 2008. Primary cutaneous tolerance: Cytotoxicity study performed on an EPISKIN® reconstructed human epidermis model (test mixture containing 1% Phytosteryl/Octyldodecyl Lauroyl Glutamate).

REPEATED INSULT PATCH TEST

mixture containing 5.999% Phytosteryl/Octyldodecyl Lauroyl Glutamate

[redacted]

CONDUCTED FOR:

[redacted]

DATE OF ISSUE:

December 30, 2016

[redacted]

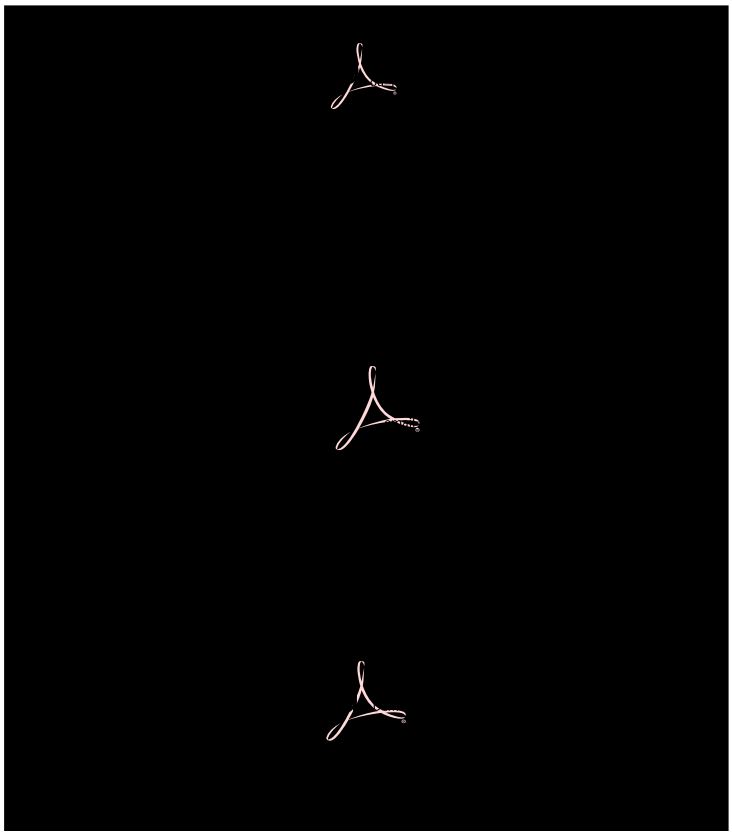
TABLE OF CONTENTS

APPENDICES

[redacted]

SIGNATURES

This study was conducted in compliance with the requirements of the protocol and [redacted] Standard Operating Procedures, and in the spirit of GCP ICH Topic E6.[1] The report accurately reflects the raw data for this study.



December 30, 2016
Date

December 30, 2016
Date

December 30, 2016
Date

STATEMENT OF QUALITY CONTROL

The Quality Control Unit of the Dermatological Safety Department conducted a 100% review of all study-related documents. The protocol was reviewed prior to the start of the study, and the medical screening forms and informed consent documents were reviewed in-process of the study. The regulatory binder and study data were reviewed post-study to ensure accuracy. The study report was reviewed and accurately reflects the data for this study.

[1] ICH Topic E6 "Note for guidance on Good Clinical Practices (CPMP/ICH/135/95)" – ICH Harmonised Tripartite Guideline for Good Clinical Practices having reached Step 5 of the ICH Process at the ICH Steering Committee meeting on 1 May 1996.

[redacted]

TITLE OF STUDY

Repeated Insult Patch Test

SPONSOR

[redacted]

STUDY MATERIAL

[redacted]

DATE STUDY INITIATED

October 24, 2016

DATE STUDY COMPLETED

December 9, 2016

DATE OF ISSUE

December 30, 2016

INVESTIGATIVE PERSONNEL

[redacted]

[redacted]

SUMMARY

One study material, [redacted], was evaluated neat to determine its ability to sensitize the skin of volunteer subjects with normal skin using an occlusive repeated insult patch study. Two hundred nineteen (219) subjects completed the study.

Under the conditions employed in this study, there was no evidence of sensitization to [redacted]

[redacted]

1.0 OBJECTIVE

The objective of this study was to determine the ability of the study material to cause sensitization by repeated topical applications to the skin of humans under controlled patch study conditions.

2.0 RATIONALE

Substances that come into contact with human skin need to be evaluated for their propensity to irritate and/or sensitize. Once an appropriate pre-clinical safety evaluation has been performed, a reproducible, standardized, quantitative patch evaluation procedure must be used to demonstrate that a particular material can be applied safely to human skin without significant risk of adverse reactions. The method herein employed is generally accepted for such a purpose.

Repeated insult patch evaluation is a modified predictive patch study that can detect weak sensitizers that require multiple applications to induce a cell-mediated (Type IV) immune response sufficient to cause an allergic reaction. Irritant reactions may also be detected using this evaluation method, although this is not the primary purpose of this procedure. Results are interpreted according to interpretive criteria based upon published works, as well as the clinical experience of [redacted] These interpretive criteria are periodically reviewed and amended as new information becomes available.

3.0 STUDY DESIGN

3.1 STUDY POPULATION

A sufficient number of subjects were enrolled to provide 200 completed subjects. In the absence of any sensitization reactions in this sample size (200 evaluable subjects), a 95% upper confidence bound on the population rate of sensitization would be 1.5%.

3.1.1 Inclusion Criteria

Individuals eligible for inclusion in the study were those who:

1. Were males or females, 18 to 70 years of age, in general good health;
2. Were free of any systemic or dermatologic disorder which, in the opinion of the investigative personnel, would have interfered with the study results or increased the risk of adverse events (AEs);
3. Were of any skin type or race, providing the skin pigmentation would allow discernment of erythema;
4. Had completed a medical screening procedure; and
5. Had read, understood, and signed an informed consent (IC) agreement.

3.1.2 Exclusion Criteria

Individuals excluded from participation in the study were those who:

1. Had any visible skin disease at the study site which, in the opinion of the investigative personnel, would have interfered with the evaluation;
2. Were receiving systemic or topical drugs or medication which, in the opinion of the investigative personnel, would have interfered with the study results;

[redacted]

3. Had psoriasis and/or active atopic dermatitis/eczema;
4. Were females who were pregnant, planning to become pregnant during the study, or breast-feeding;
5. Had a known sensitivity to cosmetics, skin care products, or topical drugs as related to the material being evaluated; and/or
6. Were participating in another study or had been recruited to participate in another study concurrently.

3.1.3 Informed Consent

A properly executed IC document was obtained from each subject prior to entering the study. The signed IC document is maintained in the study file. In addition, the subject was provided with a copy of the IC document (see Appendix III).

3.2 DESCRIPTION OF STUDY

3.2.1 Outline of Study Procedures

Subjects participated in the study over a 6-week period involving 3 phases: (1) Induction, (2) Rest, and (3) Challenge. Prior to study entry, the subjects were screened to assure that they met the inclusion/exclusion criteria. Informed consent was obtained. Each subject was provided with a schedule of the study activities. All subjects were told to avoid wetting the patches and were asked not to engage in activities that caused excessive perspiration. They were instructed to notify the staff if they experienced any discomfort beyond mild itching or observed any adverse changes at the patch sites, while on the study or within 2 weeks of completing the study.

The Induction Phase consisted of 9 applications of the study material and subsequent evaluations of the patch sites. Prior to application of the patches, the sites were outlined with a skin marker, eg, gentian violet. The subjects were required to remove the patches approximately 24 hours after application. They returned to the facility at 48-hour intervals to have the sites evaluated and identical patches applied to the same sites. Patches applied on Friday were removed by subjects after 24 hours. The sites were evaluated on the following Monday, ie, 72 hours after patch application.[2]

[2] A Monday or Friday holiday could result in evaluation at 96 hours after patch application.

[redacted]

Following the 9th evaluation, the subjects were dismissed for a Rest Period of approximately 10-15 days.

Subjects who were absent once during the Induction Phase received a make-up (MU) patch at the last induction visit. The MU applications were graded 48 hours later at the MU visit, or were recorded as N9G (no ninth grading).

The Challenge Phase was initiated during the 6th week of the study. Identical patches were applied to sites previously unexposed to the study material. The patches were removed by subjects after 24 hours and the sites graded after additional 24-hour and 48-hour periods (ie, 48 and 72 hours after application). Rechallenge was performed whenever there was evidence of possible sensitization.

To be considered a completed case, a subject must have had 9 applications and no fewer than 8 subsequent readings during Induction, and a single application and 2 readings at Challenge. Only completed cases were used to assess sensitization.

3.2.2 Study Flow Chart

WEEK 1

DAY	ACTIVITIES
1[3]	Staff obtained informed consent, reviewed completed medical screening form, applied patches
2	Subject removed patches
3	Staff graded sites, applied patches
4	Subject removed patches
5	Staff graded sites, applied patches
6	Subject removed patches

WEEK 2

DAY	ACTIVITIES
1	Staff graded sites, applied patches
2-6	Same as Week 1

WEEK 3

DAY	ACTIVITIES
1-6	Same as Week 2

WEEK 4

DAY	ACTIVITIES
1	Staff graded sites; applied make-up (MU) induction patches, if required
2	Subject removed MU patches

[3] Study flow starting with Week 1, Day 1, was altered when enrollment occurred on Wednesday or Friday. Study flow could be altered if a holiday occurred during the study.

[redacted]

3 Staff graded MU induction sites at MU visit

2-7 Rest Period

WEEK 5

DAY	ACTIVITIES
1-7	Rest Period

WEEK 6

DAY	ACTIVITIES
1	Staff applied patches
2	Subject removed patches
3	Staff graded sites
4	Staff graded sites

3.2.3 Definitions Used for Grading Responses

The symbols found in the scoring scales below were used to express the response observed at the time of examination:

SYMBOL		REACTION
-	=	No reaction
?	=	Minimal or doubtful response, slightly different from surrounding normal skin
+	=	Definite erythema, no edema
++	=	Definite erythema, definite edema
+++	=	Definite erythema, definite edema and vesiculation

SPECIAL NOTATIONS

E	=	Marked/severe erythema
S	=	Spreading of reaction beyond patch site (ie, reaction where material did not contact skin)
p	=	Papular response > 50%
pv	=	Papulovesicular response > 50%
D	=	Damage to epidermis: oozing, crusting and/or superficial erosions
I	=	Itching
X	=	Subject absent
PD	=	Patch dislodged
NA	=	Not applied
NP	=	Not patched (due to reaction achieved)
N9G	=	No ninth grading

[redacted]

3.2.4 Evaluation of Responses

All responses were graded by a trained dermatologic evaluator meeting [redacted] strict certification requirements to standardize the assignment of response grades.

4.0 NATURE OF STUDY MATERIAL

4.1 STUDY MATERIAL SPECIFICATIONS

Identification : [redacted]
Amount Applied : 0.2g

4.2 STORAGE, HANDLING, AND DOCUMENTATION OF STUDY MATERIAL

Receipt of the material used in this study was documented in a general logbook, which serves as a permanent record of the receipt, storage, and disposition of all study material received by [redacted] On the basis of information provided by the Sponsor, the study material was considered reasonably safe for evaluation on human subjects. A sample of the study material was reserved and will be stored for a period of 6 months. All study material was kept in a locked product storage room accessible to clinical staff members only. At the conclusion of the clinical study, the remaining study material was discarded or returned to the Sponsor and the disposition documented in the logbook.

4.3 APPLICATION OF STUDY MATERIAL

All study material was supplied by the Sponsor. Material was applied in an amount proportionate to the patch type or as requested by the Sponsor, generally 0.2 mL or g or an amount sufficient to cover the 2 cm x 2 cm patch. The patches were applied to the infrascapular area of the back, either to the right or left of the midline, or to the upper arm.

4.4 DESCRIPTION OF PATCH CONDITIONS

Material evaluated under occlusive patch conditions is applied to a 2 cm x 2 cm Webril™ pad attached to a non-porous, plastic film adhesive bandage (3M medical tape). The patches are secured with hypoallergenic tape (Micropore), as needed.

Material evaluated under semi-occlusive patch conditions is applied to a 2 cm x 2 cm Webril™ pad. The pads are affixed to the skin with hypoallergenic tape (Micropore).

5.0 INTERPRETATION

Sensitization is characterized by an acute allergic contact dermatitis. Typical sensitization reactions begin with an immunologic response in the dermis resulting in erythema, edema formation, and secondary epidermal damage (vesiculation), sometimes extending beyond the patch site and often accompanied by itching. Sensitization reactions tend to be delayed. The reaction typically becomes evident between 24 and 48 hours, peaks at 48-72 hours and subsequently subsides. The reaction is often greater at 72 hours than at 48 hours. The severity of the reaction is generally greater during the Challenge Phase of a Repeated Insult Patch Test (RIPT) than that seen during Induction.

Irritant reactions are characterized as a non-immunologic, localized, superficial, exudative, inflammatory response of the skin due to an externally applied material. The typical initial reaction does not develop much edema or vesiculation but results in scaling, drying, cracking, oozing,

[redacted]

crusting, and erosions. The reaction is usually sharply delineated, not spreading beyond the patch site. Irritant reactions are typically evident by 24 hours and diminish over the next 48-72 hours.

Removal of the offending agent results in gradual improvement of the epidermal damage. The reaction seen at 72 hours is, therefore, less severe than that seen at 48 hours. Finally, the severity of the reaction experienced in the Challenge Phase is generally similar to that seen during Induction.

If the results of the study indicate the likelihood of sensitization, the recommended practice is to rechallenge the subjects who have demonstrated sensitization-like reactions to confirm that these reactions are, indeed, associated with the product. Our preferred Rechallenge procedure involves the application of the product to naive sites, under both occlusive and semi-occlusive patch conditions. Use of the semi-occlusive patch condition helps to differentiate irritant and sensitization reactions. Generally speaking, if a product is a sensitizer it will produce a similar reaction under both occlusion and semi-occlusion. Whereas, if the product has caused an irritant reaction, the reactions will be less pronounced under the semi-occlusive condition.

6.0 DOCUMENTATION AND RETENTION OF DATA

The case report forms (CRFs) were designed to identify each subject by subject number and initials, and to record demographics, examination results, AEs, and end of study status. Originals or copies of all CRFs, correspondence, study reports, and all source data will be kept on hard-copy file for a minimum of 5 years from completion of the study. Storage was maintained either at a [redacted] in a secured room accessible only to [redacted] employees, or at an offsite location which provided a secure environment with burglar/fire alarm systems, camera detection and controlled temperature and humidity. Documentation will be available for the Sponsor's review on the premises of [redacted]

7.0 RESULTS AND DISCUSSION

Two hundred forty-four (244) subjects between the ages of 18 and 70 were enrolled and 219 subjects completed the study (see Tables 1 and 2 in Appendix I and Data Listings 1 and 2 in Appendix II). The following table summarizes subject enrollment and disposition.

Number enrolled:		244
Number discontinued:		25
Lost to follow-up:	22	
Voluntary withdrawal:	3	
Number completed:		219

Source: Table 1, Appendix I

There were no adverse events (AEs) reported during these studies.

A summary of response data is provided in Table 3, Appendix I. Individual dermatological response grades are provided in Data Listing 3, Appendix II.

[redacted]

8.0 CONCLUSION

Under the conditions employed in this study, there was no evidence of sensitization to [redacted]

9.0 REFERENCES

Schwartz L, Peck SM. The patch test in contact dermatitis. Publ Health Pep 1944; 59:2.

Draize JH, Woodward G, Calvary HO. Methods for the study of irritation and toxicology of substances applied topically to the skin and mucous membranes. J Pharmacol Exp Ther 1944; 82: 377-390.

Lanman BM, Elvers WB, Howard CS. The role of human patch testing in a product development program. Joint Conf Cosmet Sci Toilet Goods Assoc 1968; 135-145.

Marzulli FN, Maibach HI. Contact allergy: predictive testing in man. Contact Dermatitis 1976; 2:1.

Zhai H, Maibach HI. Dermatotoxicology. 6th ed. New York:Hemisphere, 1996.

Stotts J. Planning, conduct and interpretation of human predictive sensitization patch tests. In:Drill VA, Lazar P, eds. Current Concepts in Cutaneous Toxicity. New York: Academic Press, 1980:41-53.

Griffith JF. Predictive and diagnostic testing for contact sensitization. Toxicol Appl Pharmacol, Suppl 1969; 3:90.

Gerberick GF, Robinson MK, Stotts J. An approach to allergic contact sensitization risk assessment of new chemicals and product ingredients. American Journal of Contact Dermatitis 1993; 4(4): 205-211.

APPENDIX I

SUMMARY TABLES

[redacted]

Table 1: Summary of Subject Enrollment and Disposition

	N (%)
Subjects enrolled	126
Subjects completed induction phase	112 (88.9)
Subjects completed all phases	107 (84.9)
Total subjects discontinued	19 (15.1)
Lost to follow-up	19 (15.1)

Note: All percentages are relative to total subjects enrolled.

See data listing 1 for further detail.

[redacted]

[redacted]

Table 2: Summary of Subject Demographics
All Enrolled Subjects

Age	
N (%) 18 to 44	44 (34.9)
N (%) 45 to 65	78 (61.9)
N (%) 66 and up	4 (3.2)
Mean (SD)	47.6 (13.2)
Median	51.4
Range	18.1 to 70.5
Sex	
N (%) Male	32 (25.4)
N (%) Female	94 (74.6)
Race	
Asian	2 (1.6)
Black	41 (32.5)
Caucasian	80 (63.5)
Other	3 (2.4)
Ethnicity	
Hispanic/Latino	28 (22.2)
Not Hispanic/Not Latino	98 (77.8)

See data listing 2 for further detail.

[redacted]

[redacted]

Table 3: Summary of Dermatologic Response Grades
Number of Subjects by Product

[redacted]

	Induction Reading										Challenge Phase		
Response	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**Make Up**	**48hr**	**72hr**	**96hr(*)**
-	119	110	111	113	109	110	107	103	104	33	108	107	
?	0	0	0	0	0	1	1	0	0	0	0	0	
+	0	0	0	0	1	0	0	1	1	0	0	0	
Total evaluable	119	110	111	113	110	111	108	104	105	33	108	107	
Number absent	3	8	5	3	5	2	5	9	7		0	0	
Number discontinued	4	8	10	10	11	13	13	13	14		18	19	

Maximum Elicited Response During Induction
All Subjects Completing Induction (N=112)

Response	**n(%) Subjects**
-	111 (99.1%)
+	1 (0.9%)

(*) when required

See Table 3.1 for Key to Symbols and Scores

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[redacted]

Score or Symbol	Response or Description of Reaction
	Erythema Results
-	No reaction
?	Minimal or doubtful response, slightly different from surrounding normal skin
+	Definite erythema, no edema
++	Definite erythema, definite edema
+++	Definite erythema, definite edema and vesiculation
	Additional Comments
X	Reading not performed due to missed visit or subject discontinuation
D	Damage to epidermis: oozing, crusting and/or superficial erosions
E	Marked/severe erythema
I	Itching
p	Papular response >50%
pv	Papulovesicular response >50%
S	Spreading of reaction beyond patch site
NP	Not patched due to reaction achieved
PD	Patch dislodged
N9G	No ninth grading
NA	Not applied

[redacted]

Table 1: Summary of Subject Enrollment and Disposition

	N (%)
Subjects enrolled	118
Subjects completed induction phase	112 (94.9)
Subjects completed all phases	112 (94.9)
Total subjects discontinued	6 (5.1)
Lost to follow-up	3 (2.5)
Voluntary withdrawal	3 (2.5)

Note: All percentages are relative to total subjects enrolled.

See data listing 1 for further detail.

[redacted]

[redacted]

Table 2: Summary of Subject Demographics
All Enrolled Subjects

Age	
N (%) 18 to 44	19 (16.1)
N (%) 45 to 65	83 (70.3)
N (%) 66 and up	16 (13.6)
Mean (SD)	53.2 (11.6)
Median	54.2
Range	18.0 to 70.9
Sex	
N (%) Male	17 (14.4)
N (%) Female	101 (85.6)
Race	
Amer Ind	1 (0.8)
Asian	2 (1.7)
Black	2 (1.7)
Caucasian	112 (94.9)
Other	1 (0.8)
Ethnicity	
Hispanic/Latino	11 (9.3)
Not Hispanic/Not Latino	107 (90.7)

See data listing 2 for further detail.

[redacted]

[redacted]

Table 3: Summary of Dermatologic Response Grades
Number of Subjects by Product

[redacted]

	Induction Reading										Challenge Phase		
Response	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**Make Up**	**48hr**	**72hr**	**96hr(*)**
-	113	111	109	113	112	112	109	110	81	3	112	112	
Total evaluable	113	111	109	113	112	112	109	110	81	3	112	112	
Number absent	4	5	7	2	3	2	5	2	31		0	0	
Number discontinued	1	2	2	3	3	4	4	6	6		6	6	

Maximum Elicited Response During Induction
All Subjects Completing Induction (N=112)

Response	**n(%) Subjects**
-	112 (100.0%)

(*) when required

See Table 3.1 for Key to Symbols and Scores

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[redacted]

Table 3.1: Key To Symbols and Scores

Score or Symbol	Response or Description of Reaction
	Erythema Results
-	No reaction
?	Minimal or doubtful response, slightly different from surrounding normal skin
+	Definite erythema, no edema
++	Definite erythema, definite edema
+++	Definite erythema, definite edema and vesiculation
	Additional Comments
X	Reading not performed due to missed visit or subject discontinuation
D	Damage to epidermis: oozing, crusting and/or superficial erosions
E	Marked/severe erythema
I	Itching
p	Papular response >50%
pv	Papulovesicular response >50%
S	Spreading of reaction beyond patch site
NP	Not patched due to reaction achieved
PD	Patch dislodged
N9G	No ninth grading
NA	Not applied

APPENDIX II

DATA LISTINGS

[redacted]

Data Listing 1: Subject Enrollment and Disposition

	Study Dates						
Subject No.	Screened	1st Applic	Chall Applic	Ended	Last Reading #	Completion Status	Days in Study
001	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
002	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
003	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
004	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
005	10/24/16	10/24/16	--	10/31/16	I1	L	8
006	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
007	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
008	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
009	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
010	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
011	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
012	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
013	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
014	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
015	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
016	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
017	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
018	10/24/16	10/24/16	--	11/07/16	I4	L	15
019	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
020	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
021	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
022	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
023	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
024	10/24/16	10/24/16	--	11/29/16	I9	L	37
025	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
026	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
027	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
028	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
029	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
030	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
031	10/24/16	10/24/16	--	11/29/16	I9	L	37

Key:
Last Reading # (I=Induction Phase, C=Challenge Phase)
Completion Status (C=Completed, L=Lost to follow-up, S=Voluntary withdrawal, V=Protocol violation, AE=Adverse event, O=Other)

[redacted]

[redacted]

Data Listing 1: Subject Enrollment and Disposition

	Study Dates						
Subject No.	**Screened**	**1st Applic**	**Chall Applic**	**Ended**	**Last Reading #**	**Completion Status**	**Days in Study**
032	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
033	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
034	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
035	10/24/16	10/24/16	--	10/28/16	I0	L	5
036	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
037	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
038	10/24/16	10/24/16	--	11/14/16	I8	L	22
039	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
040	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
041	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
042	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
043	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
044	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
045	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
046	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
047	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
048	10/24/16	10/24/16	--	10/28/16	I0	L	5
049	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
050	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
051	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
052	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
053	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
054	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
055	10/24/16	10/24/16	--	11/07/16	I5	L	15
056	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
057	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
058	10/24/16	10/24/16	--	10/28/16	I0	L	5
059	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
060	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
061	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
062	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40

Key:
Last Reading # (I=Induction Phase, C=Challenge Phase)
Completion Status (C=Completed, L=Lost to follow-up, S=Voluntary withdrawal, V=Protocol violation, AE=Adverse event, O=Other)

[redacted]

[redacted]

Data Listing 1: Subject Enrollment and Disposition

	Study Dates						
Subject No.	Screened	1st Applic	Chall Applic	Ended	Last Reading #	Completion Status	Days in Study
063	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
064	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
065	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
066	10/24/16	10/24/16	--	10/31/16	I1	L	8
067	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
068	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
069	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
070	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
071	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
072	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
073	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
074	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
075	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
076	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
077	10/24/16	10/24/16	11/29/16	12/02/16	C1	L	40
078	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
079	10/24/16	10/24/16	--	10/31/16	I1	L	8
080	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
081	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
082	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
083	10/24/16	10/24/16	--	11/29/16	I9	L	37
084	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
085	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
086	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
087	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
088	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
089	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
090	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
091	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
092	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
093	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40

Key:
Last Reading # (I=Induction Phase, C=Challenge Phase)
Completion Status (C=Completed, L=Lost to follow-up, S=Voluntary withdrawal, V=Protocol violation, AE=Adverse event, O=Other)

[redacted]

[redacted]

Data Listing 1: Subject Enrollment and Disposition

	Study Dates						
Subject No.	**Screened**	**1st Applic**	**Chall Applic**	**Ended**	**Last Reading #**	**Completion Status**	**Days in Study**
094	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
095	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
096	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
097	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
098	10/24/16	10/24/16	--	11/02/16	I2	L	10
099	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
100	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
101	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
102	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
103	10/24/16	10/24/16	--	11/02/16	I2	L	10
104	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
105	10/24/16	10/24/16	11/29/16	12/02/16	C	C	40
106	10/28/16	10/28/16	11/29/16	12/02/16	C	C	36
107	10/28/16	10/28/16	11/29/16	12/02/16	C	C	36
108	10/28/16	10/28/16	11/29/16	12/01/16	I9	L	35
109	10/28/16	10/28/16	11/29/16	12/02/16	C	C	36
110	10/28/16	10/28/16	--	11/11/16	I5	L	15
111	10/28/16	10/28/16	11/29/16	12/02/16	C	C	36
112	10/28/16	10/28/16	11/29/16	12/02/16	C	C	36
113	10/28/16	10/28/16	--	11/02/16	I0	L	6
114	10/28/16	10/28/16	11/29/16	12/02/16	C	C	36
115	10/28/16	10/28/16	11/29/16	12/02/16	C	C	36
116	10/28/16	10/28/16	11/29/16	12/02/16	C	C	36
117	10/28/16	10/28/16	--	11/04/16	I1	L	8
118	10/28/16	10/28/16	11/29/16	12/02/16	C	C	36
119	10/28/16	10/28/16	11/29/16	12/02/16	C	C	36
120	10/28/16	10/28/16	11/29/16	12/02/16	C	C	36
121	10/28/16	10/28/16	11/29/16	12/02/16	C	C	36
122	10/28/16	10/28/16	11/29/16	12/02/16	C	C	36
123	10/28/16	10/28/16	11/29/16	12/02/16	C	C	36
124	10/28/16	10/28/16	11/29/16	12/02/16	C	C	36
125	10/28/16	10/28/16	11/29/16	12/02/16	C	C	36
126	10/28/16	10/28/16	11/29/16	12/02/16	C	C	36

Key:
Last Reading # (I=Induction Phase, C=Challenge Phase)
Completion Status (C=Completed, L=Lost to follow-up, S=Voluntary withdrawal, V=Protocol violation, AE=Adverse event, O=Other)

[redacted]

[redacted]

Data Listing 3: Dermatologic Response Grades
By Product and Subject

[redacted]

Subject No.	Induction Reading										Challenge Phase		
	1	2	3	4	5	6	7	8	9	MU	48hr	72hr	96hr(*)
001	-	-	-	-	-	-	-	-	-		-	-	
002	-	-	-	-	-	X	-	-	-	-	-	-	
003	-	-	-	-	-	-	-	-	-		-	-	
004	-	-	-	-	-	-	-	-	-		-	-	
005	-	X	X	X	X	X	X	X	X		X	X	
006	-	X	-	-	-	-	-	-	-	-	-	-	
007	-	-	-	-	-	-	-	-	-		-	-	
008	-	-	-	-	-	-	-	X	-	-	-	-	
009	-	-	-	-	-	-	-	-	-		-	-	
010	-	-	-	-	-	-	-	-	-		-	-	
011	-	-	-	-	-	-	-	-	-		-	-	
012	-	-	-	-	-	-	-	-	-		-	-	
013	-	-	-	-	-	-	-	-	-		-	-	
014	-	-	-	-	-	-	-	-	-		-	-	
015	-	-	-	-	-	-	-	-	-		-	-	
016	-	-	-	-	-	-	-	-	-		-	-	
017	-	-	-	-	-	-	-	-	-		-	-	
018	-	-	-	-	X	X	X	X	X		X	X	
019	-	-	-	-	X	-	-	-	-	-	-	-	
020	-	-	-	-	-	-	-	X	-	-	-	-	
021	-	-	-	-	-	-	-	-	-		-	-	
022	-	-	X	-	-	-	-	-	-	-	-	-	
023	-	-	-	-	-	-	-	-	-		-	-	

See Table 3.1 for Key to Symbols and Scores

MU = Make-up reading for missed induction visit

(*) When required

[redacted]

[redacted]

Data Listing 3: Dermatologic Response Grades
By Product and Subject

[redacted]

Subject No.	Induction Reading										Challenge Phase		
	1	2	3	4	5	6	7	8	9	MU	48hr	72hr	96hr(*)
024	-	-	-	-	-	-	-	-	-		X	X	
025	-	-	-	-	-	-	-	-	-		-	-	
026	-	-	-	-	-	-	-	-	-		-	-	
027	-	-	-	-	-	-	-	-	-		-	-	
028	-	-	-	-	-	-	-	-	-		-	-	
029	-	-	-	-	-	-	-	-	N9G		-	-	
030	-	-	-	X	-	-	-	-	-	-	-	-	
031	-	X	-	-	-	-	-	-	-	-	X	X	
032	-	X	-	-	-	-	-	-	-	-	-	-	
033	-	-	-	-	-	-	-	-	-		-	-	
034	-	-	-	-	-	-	-	-	-		-	-	
035	X	X	X	X	X	X	X	X	X		X	X	
036	-	-	-	-	-	-	-	-	-		-	-	
037	-	-	-	-	-	-	-	-	-		-	-	
038	-	X	-	-	-	-	-	-	X		X	X	
039	-	-	-	-	-	-	-	-	-		-	-	
040	-	-	-	-	-	-	-	-	-		-	-	
041	-	-	-	-	-	-	-	-	-		-	-	
042	-	-	-	-	-	-	-	-	-		-	-	
043	-	-	-	-	-	-	-	-	-		-	-	
044	-	-	-	-	-	-	-	-	-		-	-	
045	-	-	-	-	-	-	-	-	-		-	-	
046	-	-	-	X	-	-	-	-	-	-	-	-	

(*) When required

[redacted]

[redacted]

Data Listing 3: Dermatologic Response Grades
By Product and Subject

[redacted]

Subject No.	Induction Reading										Challenge Phase		
	1	2	3	4	5	6	7	8	9	MU	48hr	72hr	96hr(*)
047	-	-	-	-	-	-	-	-	-		-	-	
048	X	X	X	X	X	X	X	X	X		X	X	
049	-	-	-	-	-	-	-	-	-		-	-	
050	-	-	-	-	-	-	-	-	N9G		-	-	
051	-	-	-	-	-	-	-	-	-		-	-	
052	-	-	-	-	-	-	-	X	-	-	-	-	
053	-	-	-	-	-	-	-	-	-		-	-	
054	-	-	-	-	-	-	-	X	-	-	-	-	
055	X	-	-	-	-	X	X	X	X		X	X	
056	-	-	-	-	-	-	-	-	-		-	-	
057	-	-	-	-	-	-	-	-	-		-	-	
058	X	X	X	X	X	X	X	X	X		X	X	
059	-	-	-	-	-	-	-	-	-		-	-	
060	-	-	X	-	-	-	-	-	-	-	-	-	
061	-	-	-	-	-	-	-	-	-		-	-	
062	-	-	-	-	X	-	-	-	-	-	-	-	
063	-	-	-	-	X	-	-	-	-	-	-	-	
064	-	-	-	-	-	-	-	X	-	-	-	-	
065	-	-	-	-	-	-	-	X	-	-	-	-	
066	-	X	X	X	X	X	X	X	X		X	X	
067	-	-	-	-	-	-	-	X	-	-	-	-	
068	-	-	-	-	-	-	-	-	-		-	-	
069	-	-	-	X	-	-	-	-	-	-	-	-	

(*) When required

[redacted]

[redacted]

Data Listing 3: Dermatologic Response Grades
By Product and Subject

[redacted]

Subject No.	Induction Reading										Challenge Phase		
	1	2	3	4	5	6	7	8	9	MU	48hr	72hr	96hr(*)
070	-	-	-	-	-	-	-	-	-		-	-	
071	-	-	-	-	-	-	-	-	-		-	-	
072	-	-	X	-	-	-	-	-	-	-	-	-	
073	-	-	-	-	-	-	-	-	-		-	-	
074	-	-	-	-	-	-	-	-	-		-	-	
075	-	-	X	-	-	-	-	-	-	-	-	-	
076	-	-	-	-	-	-	X	-	-	-	-	-	
077	-	X	-	-	-	-	-	-	-	-	-	X	
078	-	-	-	-	-	-	X	-	-	-	-	-	
079	-	X	X	X	X	X	X	X	X		X	X	
080	-	-	-	-	-	-	-	-	-		-	-	
081	-	-	-	-	+	?	?	+	+		-	-	
082	-	-	-	-	-	-	-	-	-		-	-	
083	-	-	-	-	-	-	X	-	-	-	X	X	
084	-	-	-	-	-	-	-	-	-		-	-	
085	-	-	-	-	-	-	-	-	-		-	-	
086	-	-	-	-	-	-	-	-	-		-	-	
087	-	-	-	-	-	-	-	-	-		-	-	
088	-	-	-	-	-	-	-	-	-		-	-	
089	-	-	-	-	-	-	-	X	-	-	-	-	
090	-	-	-	-	-	-	-	-	-		-	-	
091	X	-	-	-	-	-	-	-	-	-	-	-	
092	-	-	-	-	-	-	-	-	N9G		-	-	

(*) When required

[redacted]

[redacted]

Data Listing 3: Dermatologic Response Grades
By Product and Subject

[redacted]

Subject No.	Induction Reading										Challenge Phase		
	1	2	3	4	5	6	7	8	9	MU	48hr	72hr	96hr(*)
093	-	-	-	-	-	-	-	-	N9G		-	-	
094	-	-	X	-	-	-	-	-	-	-	-	-	
095	-	-	-	-	-	-	-	-	-		-	-	
096	-	-	-	-	-	-	-	-	-		-	-	
097	-	-	-	-	X	-	-	-	-	-	-	-	
098	-	-	X	X	X	X	X	X	X		X	X	
099	-	-	-	-	-	-	-	-	N9G		-	-	
100	-	-	-	-	-	-	X	-	-	-	-	-	
101	-	-	-	-	-	-	-	-	-		-	-	
102	-	-	-	-	-	-	X	-	-	-	-	-	
103	-	-	X	X	X	X	X	X	X		X	X	
104	-	X	-	-	-	-	-	-	-	-	-	-	
105	-	X	-	-	-	-	-	-	-	-	-	-	
106	-	-	-	-	-	-	-	-	-		-	-	
107	-	-	-	-	-	-	-	-	-		-	-	
108	-	-	-	-	-	-	-	X	-	N9G	X	X	
109	X	-	-	-	-	-	-	-	-	N9G	-	-	
110	-	X	-	-	-	X	X	X	X		X	X	
111	-	-	-	-	-	-	-	-	-		-	-	
112	-	-	-	-	-	-	-	-	-		-	-	
113	X	X	X	X	X	X	X	X	X		X	X	
114	-	-	-	-	-	-	-	-	N9G		-	-	
115	-	-	-	-	-	-	-	-	-		-	-	
116	-	-	-	-	-	-	-	-	-		-	-	
117	-	X	X	X	X	X	X	X	X		X	X	
118	-	-	-	-	-	-	-	-	-		-	-	
119	-	-	-	-	-	-	-	-	-		-	-	
120	-	-	-	-	-	-	-	-	-		-	-	
121	-	-	-	-	-	-	-	-	-		-	-	
122	-	-	-	-	-	-	-	-	-		-	-	
123	-	-	-	-	-	-	-	-	-		-	-	
124	-	-	-	-	-	-	-	-	N9G		-	-	
125	-	-	-	-	X	-	-	-	-	N9G	-	-	
126	-	-	-	-	-	X	-	-	-	N9G	-	-	

(*) When required

[redacted]

[redacted]

Data Listing 1: Subject Enrollment and Disposition

	Study Dates						
Subject No.	Screened	1st Applic	Chall Applic	Ended	Last Reading #	Completion Status	Days in Study
001	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
002	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
003	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
004	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
005	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
006	10/31/16	10/31/16	--	11/09/16	I3	S	10
007	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
008	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
009	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
010	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
011	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
012	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
013	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
014	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
015	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
016	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
017	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
018	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
019	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
020	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
021	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
022	10/31/16	10/31/16	12/06/16	12/09/16	C	C	40
023	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
024	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
025	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
026	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
027	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
028	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
029	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
030	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
031	11/04/16	11/04/16	12/06/16	12/09/16	C	C	□□□

Key:
Last Reading # (I=Induction Phase, C=Challenge Phase)
Completion Status (C=Completed, L=Lost to follow-up, S=Voluntary withdrawal, V=Protocol violation, AE=Adverse event, O=Other)

[redacted]

[redacted]

Data Listing 1: Subject Enrollment and Disposition

	Study Dates						
Subject No.	Screened	1st Applic	Chall Applic	Ended	Last Reading #	Completion Status	Days in Study
032	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
033	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
034	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
035	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
036	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
037	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
038	11/04/16	11/04/16	--	11/23/16	I7	S	20
039	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
040	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
041	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
042	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
043	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
044	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
045	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
046	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
047	11/04/16	11/04/16	--	11/11/16	I1	L	8
048	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
049	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
050	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
051	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
052	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
053	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
054	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
055	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
056	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
057	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
058	11/04/16	11/04/16	--	11/23/16	I7	S	20
059	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
060	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
061	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
062	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36

Key:
Last Reading # (I=Induction Phase, C=Challenge Phase)
Completion Status (C=Completed, L=Lost to follow-up, S=Voluntary withdrawal, V=Protocol violation, AE=Adverse event, O=Other)

[redacted]

[redacted]

Data Listing 1: Subject Enrollment and Disposition

	Study Dates						
Subject No.	Screened	1st Applic	Chall Applic	Ended	Last Reading #	Completion Status	Days in Study
063	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
064	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
065	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
066	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
067	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
068	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
069	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
070	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
071	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
072	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
073	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
074	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
075	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
076	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
077	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
078	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
079	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
080	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
081	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
082	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
083	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
084	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
085	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
086	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
087	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
088	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
089	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
090	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
091	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
092	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
093	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36

Key:
Last Reading # (I=Induction Phase, C=Challenge Phase)
Completion Status (C=Completed, L=Lost to follow-up, S=Voluntary withdrawal, V=Protocol violation, AE=Adverse event, O=Other)

[redacted]

[redacted]

Data Listing 1: Subject Enrollment and Disposition

	Study Dates						
Subject No.	Screened	1st Applic	Chall Applic	Ended	Last Reading #	Completion Status	Days in Study
094	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
095	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
096	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
097	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
098	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
099	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
100	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
101	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
102	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
103	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
104	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
105	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
106	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
107	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
108	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
109	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
110	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
111	11/04/16	11/04/16	--	11/18/16	I5	L	15
112	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
113	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
114	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
115	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
116	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36
117	11/04/16	11/04/16	--	11/09/16	I0	L	6
118	11/04/16	11/04/16	12/06/16	12/09/16	C	C	36

Key:
Last Reading # (I=Induction Phase, C=Challenge Phase)
Completion Status (C=Completed, L=Lost to follow-up, S=Voluntary withdrawal, V=Protocol violation, AE=Adverse event, O=Other)

[redacted]

[redacted]

Data Listing 3: Dermatologic Response Grades
By Product and Subject

[redacted]

Subject No.	Induction Reading										Challenge Phase		
	1	2	3	4	5	6	7	8	9	MU	48hr	72hr	96hr(*)
001	-	-	-	-	-	-	-	-	N9G		-	-	
002	-	-	-	X	-	-	-	-	-	-	-	-	
003	X	-	-	-	-	-	-	-	-	N9G	-	-	
004	-	-	-	-	-	-	-	-	N9G		-	-	
005	-	-	-	-	-	-	-	-	-		-	-	
006	-	-	-	X	X	X	X	X	X		X	X	
007	-	-	-	-	-	-	-	-	-		-	-	
008	-	-	-	-	-	-	-	-	-		-	-	
009	-	-	-	-	-	-	-	-	-		-	-	
010	-	-	-	-	-	-	-	-	N9G		-	-	
011	-	-	-	-	-	-	-	X	-	-	-	-	
012	-	-	-	-	-	-	-	-	-		-	-	
013	-	-	-	-	-	-	-	-	-		-	-	
014	-	-	-	-	-	-	-	-	-		-	-	
015	-	-	-	-	X	-	-	-	-	-	-	-	
016	-	-	-	-	-	-	-	-	-		-	-	
017	-	-	-	-	-	-	-	-	-		-	-	
018	-	-	-	-	-	-	-	-	-		-	-	
019	-	-	-	-	-	-	-	-	N9G		-	-	
020	-	-	-	-	-	-	-	-	-		-	-	
021	-	-	-	-	-	-	-	-	N9G		-	-	
022	-	-	-	-	-	-	-	-	-		-	-	
023	-	-	-	-	-	-	-	-	-		-	-	

See Table 3.1 for Key to Symbols and Scores

MU = Make-up reading for missed induction visit

(*) When required

[redacted]

[redacted]

Data Listing 3: Dermatologic Response Grades
By Product and Subject

[redacted]

Subject No.	Induction Reading										Challenge Phase		
	1	2	3	4	5	6	7	8	9	MU	48hr	72hr	96hr(*)
024	-	-	-	-	-	-	-	-	N9G		-	-	
025	-	-	-	-	-	-	-	-	N9G		-	-	
026	-	-	-	-	-	-	-	-	N9G		-	-	
027	-	-	-	-	-	-	-	-	N9G		-	-	
028	-	-	-	-	-	-	-	-	-		-	-	
029	-	-	-	-	-	-	-	-	-		-	-	
030	-	-	X	-	-	-	-	-	-	N9G	-	-	
031	-	-	-	-	-	-	-	-	-		-	-	
032	-	-	-	-	-	-	-	-	N9G		-	-	
033	-	-	-	-	-	-	-	-	-		-	-	
034	-	-	-	-	-	-	-	-	-		-	-	
035	-	-	-	-	-	-	-	-	-		-	-	
036	-	-	X	-	-	-	-	-	-	N9G	-	-	
037	-	-	-	-	-	-	-	-	N9G		-	-	
038	X	-	-	-	-	-	-	X	X		X	X	
039	-	-	-	-	-	-	-	-	-		-	-	
040	-	-	-	-	-	-	-	-	-		-	-	
041	-	-	-	-	-	-	-	-	-		-	-	
042	-	-	-	-	-	-	-	-	-		-	-	
043	-	-	-	-	-	-	-	-	N9G		-	-	
044	-	-	-	-	-	-	-	-	-		-	-	
045	-	-	-	-	-	-	-	-	-		-	-	
046	-	-	-	-	-	-	-	-	N9G		-	-	

(*) When required

[redacted]

[redacted]

Data Listing 3: Dermatologic Response Grades
By Product and Subject

[redacted]

Subject No.	Induction Reading										Challenge Phase		
	1	2	3	4	5	6	7	8	9	MU	48hr	72hr	96hr(*)
047	-	X	X	X	X	X	X	X	X		X	X	
048	-	-	-	-	-	-	-	-	-		-	-	
049	-	-	-	-	-	-	-	-	-		-	-	
050	-	-	-	-	-	X	-	-	-	N9G	-	-	
051	-	-	-	-	-	-	-	-	N9G		-	-	
052	-	-	-	-	-	-	-	-	-		-	-	
053	-	-	-	-	-	-	-	-	-		-	-	
054	-	-	X	-	-	-	-	-	-	N9G	-	-	
055	-	-	-	-	-	-	-	-	-		-	-	
056	-	-	X	-	-	-	-	-	-	N9G	-	-	
057	-	-	-	-	-	-	-	-	-		-	-	
058	-	-	-	-	X	-	-	X	X		X	X	
059	-	X	-	-	-	-	-	-	-	N9G	-	-	
060	-	-	-	-	-	-	-	-	-		-	-	
061	-	-	-	-	-	-	-	-	-		-	-	
062	-	-	-	-	-	-	-	-	N9G		-	-	
063	-	-	-	X	-	-	-	-	-	N9G	-	-	
064	-	-	-	-	-	-	X	-	-	N9G	-	-	
065	-	-	-	-	-	-	-	-	-		-	-	
066	-	-	-	-	-	-	X	-	-	N9G	-	-	
067	-	-	-	-	-	-	-	-	-		-	-	
068	-	-	-	-	-	-	-	-	-		-	-	
069	-	-	-	-	-	-	-	-	-		-	-	

(*) When required

[redacted]

[redacted]

Data Listing 3: Dermatologic Response Grades
By Product and Subject

[redacted]

Subject No.	Induction Reading										Challenge Phase		
	1	2	3	4	5	6	7	8	9	MU	48hr	72hr	96hr(*)
070	-	-	-	-	-	-	-	-	-		-	-	
071	-	-	X	-	-	-	-	-	-	N9G	-	-	
072	-	-	-	-	-	-	-	-	-		-	-	
073	-	-	-	-	-	-	-	-	N9G		-	-	
074	-	-	-	-	-	-	-	-	N9G		-	-	
075	-	X	-	-	-	-	-	-	-	N9G	-	-	
076	-	-	-	-	-	-	X	-	-	N9G	-	-	
077	-	-	-	-	-	-	-	-	-		-	-	
078	-	-	X	-	-	-	-	-	-	N9G	-	-	
079	-	-	-	-	-	-	-	-	N9G		-	-	
080	-	-	X	-	-	-	-	-	-	N9G	-	-	
081	-	-	-	-	-	-	-	-	-		-	-	
082	-	-	-	-	-	-	-	-	N9G		-	-	
083	-	-	-	-	-	-	-	-	-		-	-	
084	-	-	-	-	-	X	-	-	-	N9G	-	-	
085	-	-	-	-	-	-	-	-	-		-	-	
086	-	-	-	-	-	-	-	-	N9G		-	-	
087	-	-	-	-	-	-	-	-	N9G		-	-	
088	X	-	-	-	-	-	-	-	-	N9G	-	-	
089	-	-	-	-	-	-	-	-	N9G		-	-	
090	-	X	-	-	-	-	-	-	-	N9G	-	-	
091	-	-	-	-	-	-	-	-	-		-	-	
092	-	-	-	-	-	-	-	-	N9G		-	-	

(*) When required

[redacted]

[redacted]

Data Listing 3: Dermatologic Response Grades
By Product and Subject

[redacted]

Subject No.	Induction Reading										Challenge Phase		
	1	2	3	4	5	6	7	8	9	MU	48hr	72hr	96hr(*)
093	-	-	-	-	-	-	-	-	-		-	-	
094	-	-	-	-	-	-	-	-	-		-	-	
095	-	-	-	-	-	-	-	X	-	N9G	-	-	
096	-	-	-	-	-	-	-	-	-		-	-	
097	-	X	-	-	-	-	-	-	-	N9G	-	-	
098	-	X	-	-	-	-	-	-	-	N9G	-	-	
099	-	-	-	-	-	-	-	-	-		-	-	
100	-	-	-	-	-	-	-	-	-		-	-	
101	-	-	-	-	-	-	-	-	N9G		-	-	
102	-	-	-	-	-	-	-	-	N9G		-	-	
103	-	-	-	-	-	-	-	-	-		-	-	
104	-	-	-	-	-	-	-	-	-		-	-	
105	-	-	-	-	-	-	-	-	N9G		-	-	
106	-	-	-	-	-	-	-	-	N9G		-	-	
107	-	-	-	-	-	-	-	-	N9G		-	-	
108	-	-	-	-	-	-	-	-	-		-	-	
109	-	-	-	-	-	-	-	-	N9G		-	-	
110	-	-	-	-	-	-	-	-	N9G		-	-	
111	X	-	-	-	-	X	X	X	X		X	X	
112	-	-	-	-	-	-	X	-	-	N9G	-	-	
113	-	-	-	-	-	-	-	-	-		-	-	
114	-	-	-	-	-	-	X	-	-	N9G	-	-	
115	-	-	-	-	-	-	-	-	N9G		-	-	
116	-	-	-	-	X	-	-	-	-	N9G	-	-	
117	X	X	X	X	X	X	X	X	X		X	X	
118	-	-	-	-	-	-	-	-	-		-	-	

(*) When required

[redacted]

[redacted]

IN VITRO STUDY REPORT

SPONSOR : [redacted]

IN VITRO STUDY : **PRIMARY CUTANEOUS TOLERANCE**

CYTOTOXICITY STUDY PERFORMED ON AN EPISKIN ®
RECONSTRUCTED HUMAN EPIDERMIS MODEL
(MTT conversion Assay)

PRODUCT : [redacted] mixture containing 1% Phytosteryl/ Octyldodecyl Lauroyl Glutamate

PROTOCOL : [redacted]

REPORT : [redacted]

Sponsor : [redacted]

Study Director and Test Facility : [redacted]

10 pages document (inc 2 pages of appendices)

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IN VITRO STUDY

PRIMARY CUTANEOUS TOLERANCE

CYTOTOXICITY STUDY PERFORMED ON AN EPISKIN® RECONSTRUCTED HUMAN EPIDERMIS MODEL

(Assay of the conversion of MTT)

I. AIM

The sponsor requested an evaluation of the primary cutaneous tolerance of the cosmetic product, [redacted] after application on reconstructed human epidermis and a cell viability assay using the MTT reduction test (Mosmann, T. , 1983).

II. RELEVANCE OF THE STUDY

The MTT conversion method, by cellular succinate dehydrogenases, is used to evaluate the irritancy potential of various products on monolayer cultures (Cornelis et al., 1992) and on three dimensional culture models (Gay et al., 1992 ; Roguet et al., 1992 ; Triglia et al., 1991).

Many studies (Roguet et al., 1994, 1998) on different classes of product show a good correlation between data from the Draize cutaneous test and the results obtained with the Episkin reconstructed epidermis model.

The units of reconstructed epidermis proposed by the model allow direct topical application of products of various consistencies (liquid, oil, cream, paste, powder, ...) to evaluate their effects on epidermal cells.

III. TEST SYSTEM

The evaluation of in vitro cutaneous tolerance was performed on a reconstructed human epidermis model (EPISKIN) supplied by EPISKIN SNC.

Every unit of reconstructed skin consists of :

- a type I collagen matrix, surfaced with a type IV collagen film, fixed to the bottom of a well using a toric ring,
- a stratified and differentiated epidermis obtained from human keratinocytes seeded on the collagen composite.

Reconstructed epidermis is maintained in agar medium for transportation.

The EPISKIN kits were delivered at D13 and used between D15 and D16.

IV. PROTOCOL

The reference protocol for the test is [redacted].

The product under test was qualified on 2 different lots of reconstructed epidermis.

1. Preparation of the epidermis

On D13, the kits were placed at room temperature with absence of light.
On D14, the color of the agar medium was verified.
The epidermis were transferred onto 2 ml of the maintenance medium (at room temperature), and incubated for 24 ± 3 hours (37 °C ± 3°C, 5% CO_2, 95 % humidity).
The lot release slip and control certificate were provided by EPISKIN SNC before application of the products.

2. Products application

The maintenance medium was removed and replaced with 2 ml of the same medium preheated to 37 °C.

A negative and a positive control (reference substance) were tested in triplicate :

- negative control : untreated epidermis
- positive control : 150 µL of an aqueous solution of Sodium Dodecyl Sulfate (Biorad, 1610301) at a concentration of 2 mg/ml.

150 mg ± 5 mg (double weighing) of the product were deposited in duplicate on the epidermis using a stainless steel curved micro-spatula.

The epidermis were incubated for 18 hours ± 1 hours (37 °C ± 3°C, 5% CO_2, 95 % humidity).

3. MTT test

At the end of the incubation period, the epidermis units were rinsed under a PBS spray (Gibco, 14040-091), removing any excess product adhering onto the epidermis with a cotton wool bud.

Each unit was then transferred to another well containing 2 ml dye solution (0,33 mg/ml MTT in the test medium) (Sigma, M2128).

The plates were incubated for 3 hours ± 15 minutes (37 °C ± 3°C, 5% CO_2, 95 % humidity).

At the end of incubation, a biopsy of epidermis was taken using a punch. The epidermis was separated from the collagen matrix using 2 curved tweezers and transferred into a tube containing 1250 µl acidified isopropanol (SDS, 0952716 and Prolabo, 20252290).

The extraction of formazan crystals was performed for 18 hours ± 2 hours at room temperature in the absence of light.

Then, each tube was stirred with a vortex mixer to homogenize the solution and 2 x 200 µL of each extract were transferred onto a plate containing 96 wells (4 lines maximum to avoid evaporation of the acidified isopropanol).

The optical density (O.D.) was measured at 570 nm vs. acidified isopropanol (blank).

4. Expression of cell viability

The percentage of cell viability of the product under test is :

$$\text{\% viability product under test} = \frac{\text{O.D. product under test} - \text{O.D. blank *}}{\text{O.D. negative control} - \text{O.D. blank *}} \times 100$$

** The O.D. of the blank (acidified isopropanol) is automatically substracted by the spectrophotometer.*

5. Acceptability and expression of results

- The O.D. of the negative control should be ≥ 0,600.

- The viability of the positive 2 mg/ml SDS control should be ≤ 35% with regard to the negative control.

VIABILITY LEVEL	Classification of the product under test
Mean viability value ≤ 50%	Potentially **Irritant** formula
Mean viability value > 50%	Potentially **Non-Irritant** formula

V. CONFORMITY TO PROTOCOL

No incidents were observed that could have affected the quality or interpretation of the results obtained.

VI. AUTHENTICATION

The study reported here was performed in conformity with the experimental protocol and laboratory procedures of [redacted] and in compliance with the principles of GLP (order of August 10th, 2004, published in the *"Journal Officiel de la République Française"* on September 18th, 2004).

The [redacted] general and technical procedures are designed to guarantee the quality, traceability and integrity of the results obtained, in particular for the in vitro tests performed and the study compliance with GLP.

Type of audit	Auditor	Date of audit	Date of reporting to the Study Director	Date of reporting to the Management
Study Plan	☒ Quality assistant ☐ Quality manager	28th oct 08		
Final report of the study	☒ Quality assistant ☐ Quality manager	19th nov 08		
Experimentation (in vitro testing) ☐ Audit of the present study ☒ Audit of a similar study	☒ Quality assistant ☐ Quality manager	23-29th sept 08	30th sept 08	30th sept 08

In addition to the above mentioned audits, the Quality Assurance department ensures that numerous audits are regularly performed relating to the following items :

- Standard Operating Procedures
- Laboratory facilities
- Quality systems used within the company in support of GLP activities.

Non GLP-related activities of the company are covered by the ISO 9001 certification of [redacted]

This report was reviewed by the [redacted] Quality Department and was found to be a faithful account of the protocol and the procedures followed and an exact reflection of the raw data generated during this study. [redacted]

Degree of conformity of the study to GLP requirements:

I, the undersigned Study Director, certify that :

- ☐ this study was performed in compliance with the principles of GLP (order of August 10th, 2004, published in the *"Journal Officiel de la République Française"* on September 18th, 2004) and in compliance with the study protocol and [redacted] procedures.
- ☒ this study does not conform to the principles of GLP (order of August 10th, 2004, published in the *"Journal Officiel de la République Française"* on September 18th, 2004) :
  The stability of the product is not available

[redacted]

VII. STORAGE OF THE PRODUCT UNDER TEST

Depending on the physicochemical characteristics, the product was stored at room temperature .
A sample of product will be stored in our laboratories for one year counting from the date of dispatch of the final report. From this date unless the Sponsor says otherwise, we will destroy the product.

VIII. RECORDING OF DATA AND ARCHIVING

The original documents and all raw data will be stored in the [redacted] archives for 10 years.
Detailed archiving rules are given in the corresponding [redacted] procedures.

IX. APPENDIX

The certificates of conformity of the lots of reconstructed epidermis used in the tests are appended.

X. RESULTS

See overleaf.

1. Details of results

Test 1 : lot Episkin 08-EPIS-036

		Quantity	OD			Mean	SD	% viability
			OD 1	OD 2	Mean			
Negative control	Epi. 1	/	1.685	1.695	1.690	1.611	0.072	**100.0**
	Epi. 2	/	1.559	1.539	1.549			
	Epi. 3	/	1.604	1.586	1.595			
Positive control	Epi. 1	150 µL	0.164	0.157	0.161	0.141	0.018	**8.8**
	Epi. 2	150 µL	0.182	0.088	0.135			
	Epi. 3	150 µL	0.133	0.120	0.127			
[redacted]	Epi. 1	150 mg	1.426	1.372	1.399	1.306	0.132	**81.1**
	Epi. 2	150 mg	1.216	1.209	1.213			

Test 2 : lot Episkin 08-EPIS-037

		Quantity	OD			Mean	SD	% viability
			OD 1	OD 2	Mean			
Negative control	Epi. 1	/	1.033	1.054	1.044	1.011	0.039	**100.0**
	Epi. 2	/	0.967	0.968	0.968			
	Epi. 3	/	1.021	1.019	1.020			
Positive control	Epi. 1	150 µL	0.101	0.087	0.094	0.077	0.015	**7.6**
	Epi. 2	150 µL	0.070	0.062	0.066			
	Epi. 3	150 µL	0.076	0.067	0.072			
[redacted]	Epi. 1	150 mg	0.728	0.773	0.751	0.732	0.027	**72.4**
	Epi. 2	150 mg	0.710	0.715	0.713			

2. Summary datasheet and conclusion

CYTOTOXICITY STUDY ON RECONSTRUCTED HUMAN EPIDERMIS (MTT reduction test)

CHARACTERISTICS OF THE PRODUCT UNDER TEST

- Code of the product under test : [redacted]
- Lot No :
- Galenic form and color : Very light orangish brown paste
- Nature : [redacted]
- Test request No. :
- Interaction with MTT test : Not required

PROTOCOL

- Experimental protocol No. : [redacted]
- Concentration tested : as is
- Quantity deposed : 150 mg
- Application time : 18h ± 1h
- Lots of reconstructed epidermis including the certificates of conformity are appended : test 1 : lot No. : 08-EPIS-036; test 2 : lot No. : 08-EPIS-037
- Special conditions : None

OBSERVATIONS

None

DATES

- Start of testing : 10 / 28 / 08
- End of testing : 11 / 07 / 08

ACCEPTABILITY

- O.D. of negative control ≥ 0,600
- Viability of 2 mg/ mL SDS ≤ 35%

} Test validated

RESULTS : % VIABILITY

Product	V1 Test 1	V2 Test 2	Mean	Standard deviation
700855 1	81.1	72.4	76.8	6.2

CONCLUSION

Depending on the experimental conditions used, the study to evaluate primary cutaneous tolerance on a reconstructed human epidermis model suggests that product [redacted] is **potentially non irritant**.

EXPERIMENTER (S) | STUDY DIRECTOR

[redacted]

2022 VCRP Data Phytosterol Glutamates

PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE

PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	03C	Eye Shadow	14
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	03D	Eye Lotion	5
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	03E	Eye Makeup Remover	1
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	03G	Other Eye Makeup Preparations	6
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	05A	Hair Conditioner	3
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	05B	Hair Spray (aerosol fixatives)	1
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	05E	Rinses (non-coloring)	1
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	05F	Shampoos (non-coloring)	2
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	05G	Tonics, Dressings, and Other Hair Grooming Aids	5
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	05I	Other Hair Preparations	1
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	07C	Foundations	12
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	07E	Lipstick	133
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	07G	Rouges	1
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	07I	Other Makeup Preparations	3
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	08G	Other Manicuring Preparations	1
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	12A	Cleansing	5
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	12C	Face and Neck (exc shave)	30
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	12D	Body and Hand (exc shave)	10
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	12F	Moisturizing	80
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	12G	Night	3
PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	12H	Paste Masks (mud packs)	2

PHYTOSTERYL/OCTYLDODECYL LAUROYL GLUTAMATE	12J	Other Skin Care Preps	6

Total 325

PHYTOSTERYL/BEHENYL/OCTYLDODECYL LAUROYL GLUTAMATE

PHYTOSTERYL/BEHENYL/OCTYLDODECYL LAUROYL GLUTAMATE	03D	Eye Lotion	4
PHYTOSTERYL/BEHENYL/OCTYLDODECYL LAUROYL GLUTAMATE	07E	Lipstick	1
PHYTOSTERYL/BEHENYL/OCTYLDODECYL LAUROYL GLUTAMATE	07G	Rouges	12
PHYTOSTERYL/BEHENYL/OCTYLDODECYL LAUROYL GLUTAMATE	12C	Face and Neck (exc shave)	4
PHYTOSTERYL/BEHENYL/OCTYLDODECYL LAUROYL GLUTAMATE	12F	Moisturizing	2
PHYTOSTERYL/BEHENYL/OCTYLDODECYL LAUROYL GLUTAMATE	12G	Night	2

Total 25

PHYTOSTERYL/BEHENYL/OCTYLDODECYL/ISOSTEARYL LAUROYL GLUTAMATE

PHYTOSTERYL/BEHENYL/OCTYLDODECYL/ISOSTEARYL LAUROYL GLUTAMATE	07E	Lipstick	1

Total 1