Safety Assessment of Myristoyl Pentapeptide-4, Palmitoyl Pentapeptide-4, and Pentapeptide-4 as Used in Cosmetics

Status:	Draft Tentative Report for Panel Review
Release Date:	March 4, 2024
Panel Meeting Date:	March 28 - 29, 2024

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.; Allan E. Rettie, Ph.D.; David Ross, 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., and the Senior Director is Monice Fiume. This safety assessment was prepared by Preethi Raj, M.Sc., Senior Scientific Analyst/Writer, CIR.

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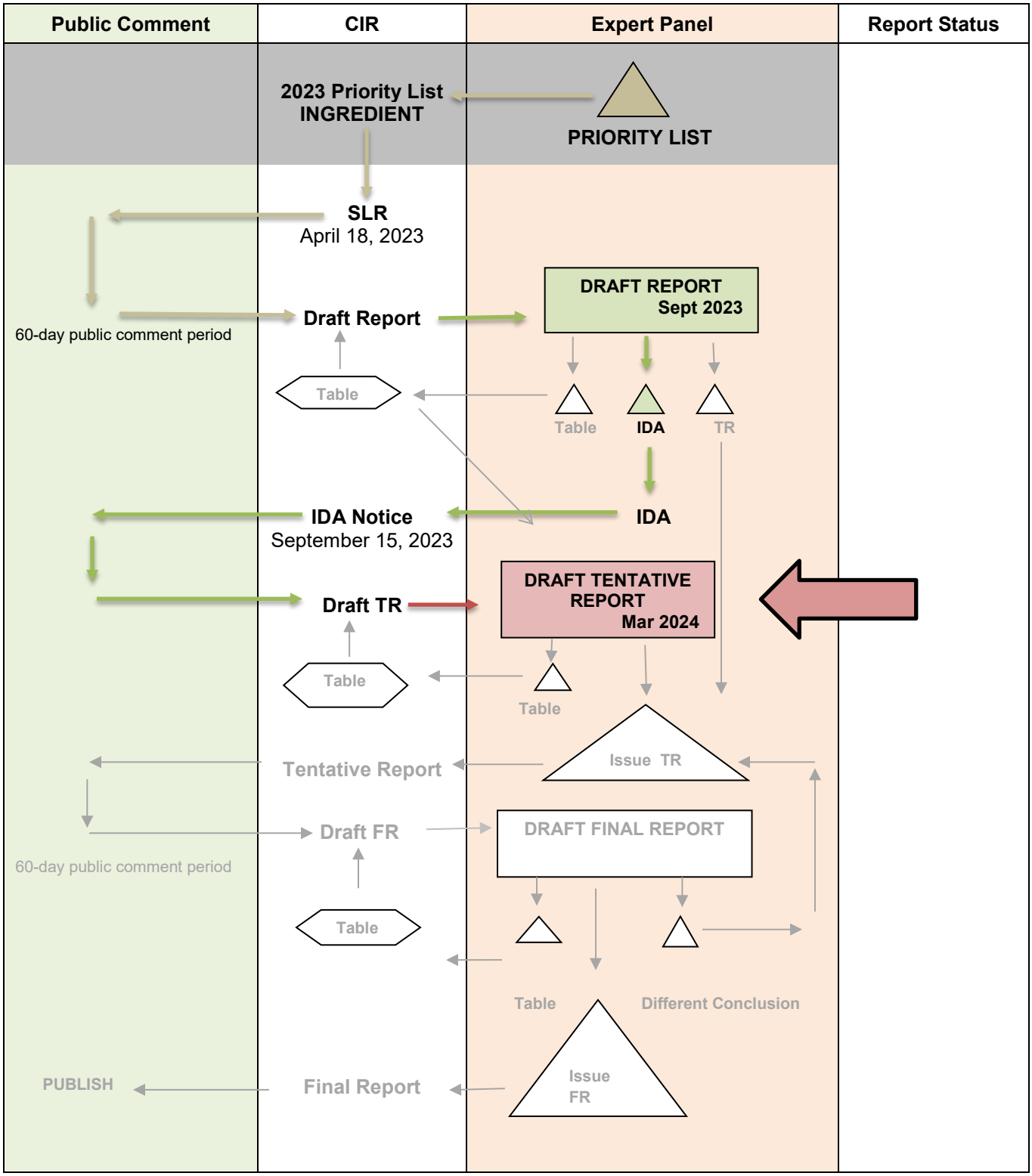
SAFETY ASSESSMENT FLOW CHART

INGREDIENT/FAMILY Pentapeptide Ingredients

MEETING March 2024



Cosmetic Ingredient Review

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Memorandum

To: Expert Panel for Cosmetic Ingredient Safety Members and Liaisons
From: Preethi S. Raj, M.Sc.
Senior Scientific Analyst/Writer, CIR
Date: March 4, 2024
Subject: Safety Assessment of Myristoyl Pentapeptide-4, Palmitoyl Pentapeptide-4, and Pentapeptide-4 as Used in Cosmetics

Enclosed is a Draft Tentative Report of the Safety Assessment of Myristoyl Pentapeptide-4, Palmitoyl Pentapeptide-4, and Pentapeptide-4 as Used in Cosmetics (identified as *report_Pentapeptides_032024* in the pdf). This is the second time the Panel is seeing a safety assessment of these 3 cosmetic ingredients. As noted during the last review, data for two amino acid sequences of Pentapeptide-4 have been included, namely lysine-threonine-threonine-lysine-serine (KTTKS) and lysine-threonine-serine-lysine-serine (KTSKS). Test article sequences have been indicated throughout the report. At the September 2023 meeting, a Draft Report was presented and the Panel issued an Insufficient Data Announcement (IDA) for the following data needs:

- Dermal irritation and sensitization data for the lysine-threonine-serine-lysine-serine (KTSKS) amino acid sequence
- Skin penetration and degradation data for Myristoyl Pentapeptide-4 (KTSKS sequence)
- Clarification of the concentration of use tested in the HRIPT study currently summarized in the report on Palmitoyl Pentapeptide-4 (Pal-lysine-threonine-threonine-lysine-serine; Pal-KTTKS) sequence

Subsequently, the following data were received in response to the IDA. Notably, the previously reported maximum use concentration of 0.05% Myristoyl Pentapeptide-4 in other eye makeup products was verified to be for an experimental product that was never developed (*data1_Pentapeptides_032024)*. Thus, the corrected maximum reported concentration of use for these ingredients is 0.0035% Palmitoyl Pentapeptide-4 in hair conditioners and the highest reported leave-on concentration of use is 0.012% Palmitoyl Pentapeptide-4 in face and neck preparations.

data1_Pentapeptides_032024

- Personal Care Products Council. 2023. Concentration of Use by FDA Product Category: Pentapeptide-4 Ingredients Revised. (Unpublished data submitted by the Personal Care Products Council on September 21, 2023.)

data2_Pentapeptides_032024

- Anonymous. 2023. ACD Percepts Version 2022.2.3. Predicted log P values for Pentapeptide-4 ingredients. (Unpublished data submitted by Personal Care Products Council on September 18, 2023.)

data3_Pentapeptides_032024

- Sederma. 2023. Product information and summary of HRIPT conducted by Consumer Product Testing Co. nº C99-0567.02. (Unpublished data submitted by Sederma on October 9, 2023.)

Also, included in this package, for your review, are a flow chart (*flow_Pentapeptides_032024*), literature search strategy (*search_Pentapeptides_032024*), ingredient data profile (*dataprofile_Pentapeptides_032024*), ingredient history (*history_Pentapeptides_032024*), transcripts from the previous meeting (*transcripts_Pentapeptides_032024*), and meeting minutes associated with the previous review of Palmitoyl Pentapeptide-4 (*originalminutes_Pentapeptides_032024*).

The Panel should carefully consider and discuss the data (or lack thereof), and the draft Abstract and draft Discussion presented in this report. A Tentative Report with a safe as used, safe with qualifications, insufficient, split, or unsafe conclusion should then be issued.

CIR History of:

Myristoyl Pentapeptide-4, Palmitoyl Pentapeptide-4, and Pentapeptide-4

March 2013

The Panel initially reviewed a large family of ingredients called Palmitoyl Oligopeptides. Since, this grouping was quite broad, the Panel decided to table this report to regroup the ingredients. Palmitoyl Petnapeptide-4 was one of said ingredients.

Previously received (unpublished) data for Palmitoyl Pentapeptide-4 includes:

- January 2013: concentration of use info

- November 2012: data from industry:

- Palmitoyl Pentapeptide-4 tested at 0.01% (vehicle and other contents not specified):
  -acute dermal irritation and acute eye irritation in rabbits, acute oral toxicity in rats, 2-wk dermal irritation in guinea pigs, HET-CAM assay, acute dermal irritation in 10 subjects, HRIPT in 51 subjects
- GPMT (0.0075% in saline and 0.01% during induction; 0.0025% in saline during challenge)
- Ames test (0.5% Palmitoyl Pentapeptide, in ethanol and water)

July 2022; February 2023

-Concentration of use data submitted by Council**;** Updated frequency of use data received from the VCRP program

April 2023

-SLR posted on CIR website; comments on SLR received from Council

- A memo was received from the Council stating that in a study summarized in the report, Palmitoyl Pentapeptide-4 did not exceed the concentration of this ingredient in face and neck products reported to the PCPC concentration of use survey

May - July 2023

In response to the SLR, the following data were received:

- Summary Information on Palmitoyl Pentapeptide-4 (provides an overview of the individual data files listed below)
- EpiSKIN® test with MTT assay (formulation containing 0.12% Palmitoyl Pentapeptide-4)
- Human patch test (formulation containing 0.12% Palmitoyl Pentapeptide-4)
- HET-CAM assay (formulation containing 0.12% Palmitoyl Pentapeptide-4)
- In vitro ocular irritation: SkinEthic™ model (formulation containing 0.12% Palmitoyl Pentapeptide-4)
- HRIPT (formulation containing 0.12% Palmitoyl Pentapeptide-4)
- XenoScreen YES/YAS endocrine disruptor testing (formulation containing 0.12% Palmitoyl Pentapeptide-4)
- XenoScreen XL YES endocrine disruptor testing (formulation containing 0.12% Palmitoyl Pentapeptide-4)
- DPRA (81.6% Palmitoyl Pentapeptide-4)
- In vitro sensitization test using KeratinoSens™ cell line (81.6% Palmitoyl Pentapeptide-4)
- Ames test (81.6% Palmitoyl Pentapeptide-4)
- In vitro mammalian cell micronucleus test (81.6% Palmitoyl Pentapeptide-4)
- Phototoxicity test (Palmitoyl Pentapeptide-4, tested at 0.0015%)

September 2023

A Draft Report was presented to the Panel. After reviewing the available data, the Panel issued an IDA with the following data needs:

- Dermal irritation and sensitization data for the lysine-threonine-serine-lysine-serine (KTSKS) amino acid sequence
- Skin penetration and degradation data for Myristoyl Pentapeptide-4 (KTSKS sequence)
- Clarification of the concentration of use tested in the HRIPT study currently summarized in the report on Palmitoyl Pentapeptide-4 (Pal-lysine-threonine-threonine-lysine-serine; Pal-KTTKS) sequence

The following data were received:

- revised concentration of use data, clarifying that the previously reported max concentration of use was for an experimental product
- predicted log P values for all 3 ingredients, and both amino acid sequences
- Clarification of the concentration of use and other details in the HRIPT study on Palmitoyl Pentapeptide-4

March 2024

A Draft Tentative Report is being presented to the Panel for review.

Pentapeptides Data Profile* – March 28 - 29, 2024 – Writer, Preethi Raj																														
				Toxicokinetics			Acute Tox			Repeated Dose Tox			DART		Genotox		Carci		Dermal Irritation			Dermal Sensitization				Ocular Irritation		Clinical Studies	
	Reported Use	Method of Mfg	Impurities	log P/log K_{ow}	Dermal Penetration	ADME	Dermal	Oral	Inhalation	Dermal	Oral	Inhalation	Dermal	Oral	In Vitro	In Vivo	Dermal	Oral	In Vitro	Animal	Human	In Vitro	Animal	Human	Phototoxicity	In Vitro	Animal	Retrospective/ Multicenter	Case Reports
Myristoyl Pentapeptide-4	X																													
• Pal-KTTKS				X																										
• Pal-KTSKS				X																										
Palmitoyl Pentapeptide-4	X																													
• Pal -KTTKS		X	X	X	X	X		X		X					X					X	X		X	X		X	X		
• Pal-KTSKS		X	X	X											X				X		X	X		X	X		X			
Pentapeptide-4	X																													
• Pal-KTTKS				X	X	X																								
• Pal-KTSKS				X																										

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

[Pentapeptides]

Ingredient	CAS #	PubMed	FDA	HPVIS	NIOSH	NTIS	NTP	FEMA	EU	ECHA	ECETOC	SIDS	SCCS	AICIS	FAO	WHO	Web
Palmitoyl Pentapeptide-4	521091-64-5 214047-00-4	✓	NR	NR	NR	NR	NR	NR	✓*	NR	NR	NR	NR	NR	NR	NR	✓
Pentapeptide-4	NA	✓	NR	NR	NR	NR	NR	NR	✓*	NR	NR	NR	NR	NR	NR	NR	✓
Myristoyl Pentapeptide-4	NA	NR	NR	NR	NR	NR	NR	NR	✓*	NR	NR	NR	NR	NR	NR	NR	✓

NR- not reported; ✓ - data available; ✓*- data available, but not relevant

Search Strategy

PubMed

[*total # of hits / # hits that were useful*] – search last performed: 01/15/2024

((((((((((((((myristoyl pentapeptide-4) OR (Myristoyl Pentapeptide-3)) OR (Collasyn 514KS)) OR (Palmitoyl Pentapeptide-3)) OR (Palmitoyl Pentapeptide-4)) OR (521091-64-5)) OR (214047-00-4)) OR (1392416-25-9)) OR (149128-48-3)) OR (N2-(1-oxohexadecyl)-L-lysyl-L-threonyl-L-seryl-L-lysyl-L-serine)) OR (N2-(1-oxohexadecyl)-L-lysyl-L-threonyl-L-threonyl-L-lysyl-L-serine)) OR (Palmitoyl Pentapeptide-3)) OR (Lipopentapeptide 3)) OR (OriStar POPP)) OR (SpecPed SC-PP4)) OR (ApepPPP-5)) OR (BsPep-5)) OR (Matrixyl)
- 4,366/6 results

AND

DPRA (Direct Peptide Reactivity Assay) – 1 hit/0 useful
ADRA (Amino acid Derivative Reactivity Assay)- 0 hits
kDPRA (Kinetic DPRA) – 0 hits
IL-8-Luc (Interleukin8 Reporter Gene Assay) - 0 hits
GARD skin – 0 hits
SenCeeTox – 0 hits
VITOSens – 0 hits
PBMDC – 0 hits
SensiDerm – 0 hits
mMUSST – 0 hits

General Search

palmitoyl pentapeptide-4 cosmetic toxicity – 403,000/3

oligopeptide toxicity pentapeptide-4 – 264,000/4

LINKS

Search Engines

- Pubmed - http://www.ncbi.nlm.nih.gov/pubmed
  - appropriate qualifiers are used as necessary
  - search results are reviewed to identify relevant documents
- Connected Papers - https://www.connectedpapers.com/

Pertinent Websites

- wINCI - https://incipedia.personalcarecouncil.org/winci/ingredient-custom-search/
- FDA databases http://www.ecfr.gov/cgi-bin/ECFR?page=browse
- FDA search databases: http://www.fda.gov/ForIndustry/FDABasicsforIndustry/ucm234631.htm;,
- Substances Added to Food (formerly, EAFUS): https://www.fda.gov/food/food-additives-petitions/substances-added-food-formerly-eafus
- GRAS listing: http://www.fda.gov/food/ingredientspackaginglabeling/gras/default.htm
- SCOGS database: http://www.fda.gov/food/ingredientspackaginglabeling/gras/scogs/ucm2006852.htm
- Indirect Food Additives: http://www.accessdata.fda.gov/scripts/fdcc/?set=IndirectAdditives
- Drug Approvals and Database: http://www.fda.gov/Drugs/InformationOnDrugs/default.htm
- FDA Orange Book: https://www.fda.gov/Drugs/InformationOnDrugs/ucm129662.htm
- (inactive ingredients approved for drugs: http://www.accessdata.fda.gov/scripts/cder/iig/
- HPVIS (EPA High-Production Volume Info Systems) - https://iaspub.epa.gov/oppthpv/public_search.html_page
- NIOSH (National Institute for Occupational Safety and Health) - http://www.cdc.gov/niosh/
- NTIS (National Technical Information Service) - http://www.ntis.gov/
  - technical reports search page: https://ntrl.ntgis.gov/NTRL/
- NTP (National Toxicology Program) - http://ntp.niehs.nih.gov/
- Office of Dietary Supplements https://ods.od.nih.gov/
- FEMA (Flavor & Extract Manufacturers Association) GRAS: https://www.femaflavor.org/fema-gras
- EU CosIng database: http://ec.europa.eu/growth/tools-databases/cosing/
- ECHA (European Chemicals Agency – REACH dossiers) – http://echa.europa.eu/information-on-chemicals;jsessionid=A978100B4E4CC39C78C93A851EB3E3C7.live1
- ECETOC (European Centre for Ecotoxicology and Toxicology of Chemicals) - http://www.ecetoc.org
- European Medicines Agency (EMA) - http://www.ema.europa.eu/ema/
- OECD SIDS (Organisation for Economic Co-operation and Development Screening Info Data Sets)- http://webnet.oecd.org/hpv/ui/Search.aspx
- SCCS (Scientific Committee for Consumer Safety) opinions: http://ec.europa.eu/health/scientific_committees/consumer_safety/opinions/index_en.htm
- AICIS (Australian Industrial Chemicals Introduction Scheme)- https://www.industrialchemicals.gov.au/
- International Programme on Chemical Safety http://www.inchem.org/
- FAO (Food and Agriculture Organization of the United Nations) - http://www.fao.org/food/food-safety-quality/scientific-advice/jecfa/jecfa-additives/en/
- WHO (World Health Organization) technical reports - http://www.who.int/biologicals/technical_report_series/en/
- www.google.com - a general Google search should be performed for additional background information, to identify references that are available, and for other general information

Botanical Websites, if applicable

- Dr. Duke's - https://phytochem.nal.usda.gov/phytochem/search
- Taxonomy database - http://www.ncbi.nlm.nih.gov/taxonomy
- GRIN (U.S. National Plant Germplasm System) - https://npgsweb.ars-grin.gov/gringlobal/taxon/taxonomysimple.aspx
- Sigma Aldrich plant profiler- http://www.sigmaaldrich.com/life-science/nutrition-research/learning-center/plant-profiler.html
- American Herbal Products Association Botanical Safety Handbook (database) - http://www.ahpa.org/Resources/BotanicalSafetyHandbook.aspx
- National Agricultural Library NAL Catalog (AGRICOLA) https://agricola.nal.usda.gov/
- The Seasoning and Spice Association List of Culinary Herbs and Spices
- http://www.seasoningandspice.org.uk/ssa/background_culinary-herbs-spices.aspx

Fragrance Websites, if applicable

- IFRA (International Fragrance Association) – https://ifrafragrance.org/
- Research Institute for Fragrance Materials (RIFM) - https://www.rifm.org/#gsc.tab=0 http://fragrancematerialsafetyresource.elsevier.com/

SEPTEMBER 2023 PANEL MEETING – INITIAL REVIEW/DRAFT REPORT

Belsito Team – September 11, 2023

DR. BELSITO: Okay. Pentapeptides.

MS. RAJ: Before you start, I just have some printouts for everyone. It's the data profile by sequence, because there are two sequences in the report.

DR. KLAASSEN: Right.

MS. RAJ: As well as the paper that you mentioned.

DR. BELSITO: I think everyone got the food paper, right?

DR. KLAASSEN: Mm-Hmm.

MS. RAJ: It's in there.

DR. BELSITO: Right. But everyone should have gotten it ahead of time because Bart sent it out. I mean, that was my major issue with this report, is that it sort of raised my antenna because it's used as anti-wrinkle, which can be used as a marketing claim. And the other study that was done in Asia just showed some improvementm in crow's lines.

But in the Fu study, I mean, the interesting thing is they didn't see any changes in trans-epidermal water loss and other endpoints where there was biological activity, except in the keratin profile. And so my question is, do we consider that kind of change that they noted to be biologic and therefore not a cosmetic or not?

DR. RETTIE: Is it up to us to decide whether it's biological or not? Isn't it -- doesn't it have to have something to do with the claims of the manufacturer?

DR. BELSITO: Well, no, the claims they're making are purely marketing. You know, it's being marketed as an anti-wrinkle. So their marketing claims keep it within the level of cosmetics and what dermatologists incorrectly called cosmeceuticals.

The dilemma I have is that we have the Fu paper and there are changes in the keratin profile. And then we have a manufacturer telling us that the Fu levels do not exceed the levels that we use in our product. So sometimes, Allan, we've had materials where they can cause -- the most common thing is a bleaching effect. And then we'll say, well, that would not be appropriate for a cosmetic.

But here we have a reference to this paper and then a reference below it, personal communication, to the PCPC, that the Fu study was using a material that does not exceed the levels that we're reporting here. So then it becomes up to us to say, okay, it didn't cause any changes in trans-epidermal water loss, blah, blah, blah. You know, there were some changes in the subtypes of keratin but we don't consider this a significant biological effect. Or we do, in which case we can't rule on the safety of that concentration.

DR. RETTIE: My reading was that it was a difference in percentages of the different subtypes of keratin.

DR. BELSITO: Right.

DR. RETTIE: And going back to you, is that -- I don't know how to evaluate that.

DR. BELSITO: I don't either. I'm a dermatologist, I'm not a keratin person. And you would think that if it changed -- if there was biological significance there would've been a change in trans-epidermal water loss, which wasn't seen in that report, right?

DR. RETTIE: So that's a more gross effect. And could we refer back to that transepidermal --

DR. BELSITO: What do you think?

DR. SNYDER: It's a biological effect, period.

DR. BELSITO: Yeah.

DR. SNYDER: It changed the profile.

DR. BELSITO: It's a biological --

DR. BELSITO: If the profile didn't have an apparent effect, other than a wrinkle --

DR. SNYDER: In toxicology, we talk about the fact that it has to alter the function or something. But we don't know. I mean, it clearly altered the function if it decreased wrinkles.

DR. RETTIE: Well, it didn't alter the function in terms of trans-epidermal water loss.

DR. SNYDER: That's only one function of it, though.

DR. BELSITO: Right. I mean, it's a conundrum.

DR. SNYDER: You weren't listening to Don this morning about the KeratinoSens.

MS. RAJ: Dr. Belsito, a very, I guess, simplified understanding of this paper, isn't it basically saying that the formulation that they tested, that included this peptide, was tolerable compared to, say, the tretinoin?

DR. BELSITO: Well, that was what they were saying. But also what they're reporting is a change in keratin profile.

DR. SNYDER: To me, that's a biological effect. That's a drug, right?

DR. BELSITO: Yeah. I mean, what they're showing is that, okay, there's -- you know, the retinoid increases erythema, increases dryness; this pentapeptide does not. The changes in baseline, the trans-epidermal water loss, we're seeing with the retinoid, but we're not seeing with the pentapeptide. But then there were changes in the keratin profile that were seen with the pentapeptide.

DR. RETTIE: So that's the only change.

DR. BELSITO: It's the only change reported in this paper.

DR. SNYDER: Don has a comment.

DR. BJERKE: Yeah, if I may. The way I kind of wrap my mind around this, is to look at whether it's a NOEL, no-observable-effect-level, or it's a no-observable-adverse-effect-level. Because there's no restriction on cosmetic ingredients that they can't have any biological activity. They all do. Look at salicylates, look at retinoids.

Almost everything that we do in parts of benefit to the appearance of the skin. And so, I think what we're talking about here is a -- we don't have an inert ingredient. We don't have something that we can say has no effects. But what we can say is it has no adverse effects. Because if you think about these, these peptides are present at, let's say, 5 to maybe 100, maybe max 200 part per million.

DR. BELSITO: Yeah, they're small.

DR. BJERKE: They don't get across the skin very well either, typically.

DR. BELSITO: I mean, yeah, if you look at Oil of Olay, Regenerist Micro-sculpting cream --

DR. BJERKE: Exactly.

DR. BELSITO: -- it has this pentapeptide in it.

DR. BJERKE: Right.

DR. BELSITO: It's the ingredients, da-da-da-da, and then in small print -- then the pentapeptide comes in at the end.

DR. BJERKE: Absolutely. I mean, based on this study, to try to ascribe something specifically to the pentapeptide as opposed to the overall formulation, I think it's somewhat difficult as well.

DR. BELSITO: I'm not arguing with you. I'm just pointing out that I saw this change and I didn't know how to deal with it, particularly because the next reference after was that the Fu study was what we have in our product possibly.

DR. BJERKE: Yeah, exactly. And then I think the way that we position this is it helps to improve the appearance of fine lines and wrinkles. If we were to say that this decreases wrinkles or removes wrinkles --

DR. BELSITO: No, I know. I know the marketing claim.

DR. BJERKE: Yeah. Yeah. And you pointed that out quite clearly, which is exactly how we approach these.

DR. BELSITO: I mean, I'm fine. I just want to point it out and have this discussion because it's the first time we've ever dealt with something where there's been any kind of effect that we've seen, that we haven't been able to simply say should not occur.

DR. BJERKE: Right. Right.

DR. BELSITO: Because as used, this change in keratin profile seems to be occurring, but there's no redness, there's no irritation, there's no increased trans-epidermal water loss. We have no other evidence of an adverse effect. And if everyone else is comfortable with that, I'll forget I ever mentioned it.

DR. RETTIE: I'm comfortable. I had a couple of comments.

DR. BELSITO: Paul?

DR. SNYDER: I'm fine.

DR. BELSITO: Okay. Go ahead, Allan, with your comments.

DR. RETTIE: I'm trying to find it. Well, one's just a note. I was curious about the reported log p(s) for these things. The Myristoyl Pentapeptide at a log p of (negative) -0.3?

DR. BELSITO: I had the same question.

DR. RETTIE: And then you jump up for the Palmitoyl to like 3.

DR. BELSITO: Yeah. Is this true for the others as well or just the Palmitoyl? Yeah.

DR. RETTIE: I mean, if you look at Palmitoyl versus Myristoyl, log p(s) is like -- the difference is one right, for the acids. So this just seemed -- I mean, they're calculated, so we just don't know. So I just let it go because it was estimated, but I was surprised by it. And the only other comment I had was under method of manufacture.

DR. BELSITO: Yeah, I have the same question.

DR. RETTIE: Yeah.

DR. BELSITO: In impurities, can the palmitoyl serve as a read across for the Myristoyl and the Pentapeptide-4?

DR. RETTIE: That was part of it. I was more specifically thinking about deleting the second paragraph under methods of manufacture. Seemed to me they were both provided by Sederma. And the first one came years ago and it was like a very condensed version. And the newer one, which is the first paragraph, is lots of information about peptide synthesis.

So, I thought you could probably strike the second one since they were both from the same manufacturer. And one was very generic, the other was quite specific.

DR. BELSITO: Okay. What page are you on, Allan, PDF what?

DR. SNYDER: Page 55.

DR. RETTIE: 55, is it?

DR. SNYDER: Yeah.

DR. BELSITO: So, you're saying?

DR. RETTIE: I'm saying -- Page 55, if I ever get there? There's a redundancy there is basically what I'm saying.

DR. BELSITO: So, the PCPC also raised, they are general to the production of peptide synthesis. It's unknown whether it's specific ingredients of cosmetics. But then one of the methods is exactly what you described.

DR. RETTIE: Yep. So the last paragraph in this section, on PDF Page 56.

DR. BELSITO: But first we should address the PCPC comment. Are we fine with dropping method --

DR. RETTIE: Yes, I think so.

DR. BELSITO: It says methods of manufacturing detailed are general to the production of peptide synthesis. Unknown whether they're specific to the ingredients that are used in cosmetics. But then the description that you are giving,

, is essentially the description for one of the cosmetics. So do we get rid of that whole paragraph?

DR. EISENMANN: I don't care. I just don't like that -- it's saying it's unknown.

DR. BELSITO: Right.

DR. EISENMANN: I mean, I don't care how you resolve it, but it's known, and it agrees with what you -- the general --

DR. BELSITO: Right. I mean, I agree. So what do we do with that paragraph? Get rid of it and then just discuss the synthesis for Palmitoyl Pentapeptide-4?

DR. KLAASSEN: That's what I would think.

DR. EISENMANN: That's fine.

DR. RETTIE: So, (inaudible) after -- general to the production of peptide synthesis?

DR. BELSITO: No, we're talking about getting rid of the entire paragraph because then it's described again under Palmitoyl. And then the other question is, can that method of manufacturing and impurities data for Palmitoyl Pentapeptide cover Pentapeptide-4 and Myristoyl, because we don't have manufacturing impurities for those two.

DR. RETTIE: But you would imagine it's not a long, big stretch to the synthesis of just Pentapeptide-4 or the Myristoylated version of it.

DR. BELSITO: I agree. I'm just pointing that out.

DR. RETTIE: Yep, I agree.

DR. BELSITO: So, we're fine with using the Palmitoyl to support the safety?

DR. RETTIE: Yes.

DR. SNYDER: Dan Liebler previously had raised the issue of clarification of the nomenclature of the composition. And it was his opinion they should be reviewed independently.

DR. RETTIE: The Palmitoyl versus the Myristoyl?

DR. SNYDER: Correct.

DR. BELSITO: Who did?

DR. SNYDER: Dan.

DR. KLAASSEN: Why?

DR. SNYDER: It was in our old notes.

DR. RETTIE: Was that because of potential biological effects being so different?

DR. SNYDER: Because this is essentially a Pentapeptide-4 report.

DR. RETTIE: Mm-Hmm.

DR. SNYDER: Yeah. So I don't know why -- what -- I don't know.

DR. BELSITO: Well, he's not here to answer it, so we have to rely on Allan.

DR. SNYDER: Well, no, I was trying to get Allan's take on that.

DR. BELSITO: Okay. You're trying to prompt Allan, huh?

DR. SNYDER: Well, yeah, a little bit.

DR. RETTIE: I didn't have awful lot of problems with it.

DR. SNYDER: Okay.

DR. RETTIE: I mean, for the Myristoylated and Palmitoylated compounds, those are going get cleaned. You're going to get Pentapeptide-4 from it.

DR. BELSITO: Okay.

DR. RETTIE: I think it's a decent group.

DR. BELSITO: You're sure, as opposed to Diglycerin?

DR. RETTIE: This one I'm pretty sure.

DR. BELSITO: Okay. Okay.

DR. SNYDER: All right. Okay.

DR. BELSITO: Okie doke.

DR. RETTIE: I had fun with this. My son is a peptide chemist. Got his input.

DR. SNYDER: Must be pretty cool for you. Yeah. That's pretty cool.

DR. BELSITO: Preethi, I had a question. That's the same question that Allan had about the numbers. Why minus three for Myristoyl and plus for the Palmitoyl?

MS. RAJ: So those two values come from a paper, if you see Reference Number 2, it's Abu Samah.

DR. BELSITO: Yeah.

MS. RAJ: So that's where I got them from. I could possibly -- I think there were PubChem pages, too, which had log p values if those would be more suitable to you. But I figured since they're comparing both of the ingredients in the same paper, that you could have confidence that they would be not just random, but.

DR. KLAASSEN: Maybe one was a typing error.

MS. RAJ: Um-hmm. Maybe.

DR. KLAASSEN: Those things do happen.

DR. BELSITO: Then in the introduction, you say the Palmitoyl Pentapeptide, and then the Pentapeptide has different amino acid groups. Is that true for the Myristoyl as well or just for the Palmitoyl?

MS. RAJ: Well I think the sequences are referring more to the Pentapeptide-4 molecule, which is the same for all three ingredients reviewed in this report, right? So there could potentially even be more sequences. But I think the reason why we added that language is because we have data, two sequences. And just to let you know, in the wINCI monograph for Palmitoyl Pentapeptide-4, both of the sequences for which we have data, on the report are listed.

DR. BELSITO: So then should we say other sequences are possible?

MS. RAJ: Yes, we could. I mean, if the chemists agreed.

DR. BELSITO: I mean, I'm just saying because as I read this now we're saying that --

DR. EISENMANN: They would name it different. Usually now they're naming each sequence separately. This was an early peptide and they kind of grandfathered in that there's two sequences.

DR. BELSITO: So there are only two sequences?

DR. EISENMANN: Right?

DR. BELSITO: But Myristoyl Pentapeptide could have one of these two sequences as well? And Pentapeptide-4 could as well?

MS. RAJ: I would think so.

DR. RETTIE: So is it six sequences we're dealing with or three is the question.

DR. BELSITO: We're dealing with two.

DR. EISENMANN: No, it's two sequences that might be attached. And it's very unlikely that Pentapeptide is used on its own because it wouldn't get into the skin is my understanding. It may be one of these things that they put a name in the dictionary to name something else.

DR. BELSITO: Right. Okay.

DR. EISENMANN: So, reference material.

DR. BELSITO: I'm just trying to clarify the two sequences that we're seeing. Could those also be for Myristoyl and for Pentapeptide-4? Or is it only the Palmitoyl that has those two sequences?

MS. RAJ: We did not find data with those sequences associated the other ingredients.

DR. BELSITO: But they could be?

MS. RAJ: I would think so, but I'll let the chemists speak on that. If they --

DR. BELSITO: Well, I mean the chemists aren't going to know what the cosmetic companies are doing.

DR. EISENMANN: I have to go back to the supplier and ask them again. And ask.

DR. BELSITO: Well, I mean, I just think we need clarification in the language, whether, you know -- because right now the way it's written, Myristoyl Pentapeptide and Palmitoyl have an additional saturated da-da-da-da. The amino acid sequence of the ingredients can vary.

So instead of just saying Palmitoyl, you say the amino acid sequence of the Pentapeptide portion of these ingredients can vary. The two variations are KTTKS and KTSKS, rather than linking them to Palmitoyl. Is that fair?

MS. RAJ: I just wonder though -- I mean, I'm just thinking out loud. Because these two sequences were on the Palmitoyl Pentapeptide monograph and not on the Pentapeptide-4 monograph, would that make any difference? I don't know.

DR. BELSITO: I mean, I'm just trying to be as accurate as possible. Because as I read this, do we say what the sequence is for Myristoyl Pentapeptide?

MS. RAJ: No.

DR. BELSITO: Fine.

DR. RETTIE: We don't associate it with either the KTTKS or the other one.

DR. BELSITO: Okay. So just leave it as it is. It's fine.

DR. RETTIE: But I'm wondering if some clarification is needed, that when you go back in time and you read all the information, when it first came up, there was a laundry list yay big and a lot of questions about nomenclature which I wasn't necessarily part of. But when I just read the Chemistry section here, Pentapeptide-4 is a synthetic peptide comprised of lysine, serine, and threonine linked in varied 5-amino sequences, two of which. So that doesn't seem to preclude other sequences to me.

DR. BELSITO: No, but what Carol's saying is those two have come under -- grandfathered as Pentapeptide-4.

DR. EISENMANN: I'll double check with Joanne, but I'm quite certain (inaudible) now.

DR. BELSITO: Pentapeptide-4 is that one or the other?

DR. RETTIE: Okay.

DR. BELSITO: But not any of the other combinations.

DR. RETTIE: That's doesn't come through in the way it's written to me at least, right now. Can we clarify that?

DR. BELSITO: So, what we can say is Pentapeptide-4, da-da-da-da. The two --

DR. SNYDER: Or varied 5-amino acid sequences (forming a pentapeptide) -- take out the two of which -- are lysine-threonine-threonine-lysine-serine. Just take out that two of which are. Just say what they are.

DR. BELSITO: The two referred to as Pentapeptide-4 are.

DR. SNYDER: Right.

DR. RETTIE: You could take out varied from 5-amino acid sequences. Just say which are linked, and 5-amino acid sequences, e.g.

MS. RAJ: Okay.

DR. BELSITO: Okay. So Pentapeptide-4 is a synthetic peptide comprised of -- what are you doing here, Allan?

DR. SNYDER: Comprised of either.

DR. BELSITO: Comprised of either.

DR. SNYDER: Or.

DR. RETTIE: Yeah.

DR. SNYDER: Just list the two, either or. The KTTKS and the KTSKS.

DR. EISENMANN: Is it either or, or is it a mix?

DR. BJERKE: I think we have to go back to the INCI dictionary, right? Because it's the same issue that we've been dealing with historically.

DR. EISENMANN: Yeah, I'm not sure.

DR. BJERKE: Yeah.

DR. SNYDER: All the rest of it is not important. We're defining what it is we're looking at.

DR. BELSITO: Is either serine-lysine-threonine -- okay. I got it.

DR. SNYDER: Yeah. Yep. Or, then the other.

DR. RETTIE: There might be more discussion with the other group tomorrow, particularly around biological activities of myristoylated and palmitoylated peptides, but I'm not sure that those are really relevant to what -- we'll see tomorrow. I'm expecting some --

DR. SNYDER: We've already reviewed myristic acid and palmitic acid.

DR. RETTIE: Very specifically when they're attached to proteins they have different biological functions.

DR. SNYDER: Okay. Fair enough.

DR. RETTIE: And in my opinion should not be treated together. But for peptides, these things are very specifically on a position that's not that relevant to their biological activity.

DR. SNYDER: Okay.

DR. RETTIE: It is my reading.

DR. SNYDER: Okay.

DR. BELSITO: Okay, Carol, are we going to get updates? Because right now, concentration of use is from 2013 from one of these.

MS. RAJ: No, that's just because data on Palmitoyl Pentapeptide-4 had previously been reviewed, but was not published. So that's why that data (inaudible).

DR. BELSITO: But we don't have 2022 or 2023 data for it.

MS. RAJ: We do. We do. It's in Table --

MS. FIUME: PDF Page 64.

MS. RAJ: Yes. Oh, you're saying it's --

MS. FIUME: So it's not recorded for the Pentapeptide-4. There was nothing (inaudible).

MS. RAJ: It's just for Palmitoyl Pentapeptide. And there's one concentration piece for (inaudible). That's actually the highest, if I'm not mistaken.

DR. EISENMANN: Yes, it is.

DR. BELSITO: So, the data in Table 3 is from 2023 -- the concentrations are from 2022 for all product categories?

MS. RAJ: Yes.

DR. BELSITO: Okay. Then I misunderstood. So, then in the cosmetics, you -- so this is PDF Page 56 under the Cosmetic, the third paragraph down. Historical concentration of use, where you say that Palmitoyl Pentapeptide-4 in 2013, I would get rid of that because it's misleading. We're looking at data for the current, we don't care about historical.

MS. RAJ: Okay.

DR. BELSITO: At least I don't care. Because that's where I thought you were giving me data from 2013.

MS. RAJ: But obviously we're still keeping the tox data?

DR. BELSITO: Yeah, the tox data is fine. But the concentration of use, I mean, I thought that it was old.

MS. RAJ: Okay.

DR. BELSITO: Under dermal penetration, Preethi, your second paragraph.

DR. SNYDER: It says Dermal Permeation, not Penetration.

DR. BELSITO: It's really more akin to dermal metabolism rather than penetration.

DR. SNYDER: You got Permeation instead of Penetration.

DR. BELSITO: Yeah. But the second paragraph is really more metabolism, no?

MS. RAJ: I think that's how they described it in the paper, but --

DR. BELSITO: Well, it says the dermal stability was evaluated in vitro.

DR. SNYDER: But it's okay if you look at -- if we would of have the normal category ADME instead of dermal permeation. So I'd change that dermal permeation to ADME absorption, distribution, metabolism.

MS. FIUME: Actually, we do break out the dermal penetration versus ADME.

DR. SNYDER: Okay.

MS. FIUME: So, it's typically broken out.

DR. BELSITO: But the second paragraph is not really penetration, it's more metabolism. Right?

DR. RETTIE: Which page are we on?

DR. SNYDER: Page 57.

DR. BELSITO: Page 57, the second paragraph. At predetermined times, the amount of Palmitoyl Pentapeptide-4 and Pentapeptide-4 present incubated mixtures was sampled and analyzed. Pentapeptide-4 was almost fully degraded in the dermal skin extract and whole skin homogenate. I mean, it's really metabolism.

DR. KLAASSEN: Yeah, just give it a new heading.

MS. FIUME: So you would want that as ADME?

DR. BELSITO: Yeah.

DR. KLAASSEN: Yeah. That's fine.

MS. RAJ: Just the second paragraph?

DR. KLAASSEN: Correct?

DR. BELSITO: Yeah. The first paragraph is penetration, not permeation. And the second paragraph is ADME.

DR. RETTIE: This supports all the expectations that (inaudible) will slightly impede metabolism, which is what you'd expect.

DR. BELSITO: So, um, we have no DART data here, so I just have a question. Assuming in concentrations maximum 0.05 percent limited percutaneous absorption in vitro; are we okay with the lack of systemic data?

DR. SNYDER: I think so. There was an endocrine study that was negative at 0.12 percent. So there's no -- I think so.

DR. BELSITO: So that would go in the Discussion?

DR. SNYDER: Yeah.

DR. BELSITO: Okay. So low maximum use concentration, limited percutaneous absorption data, in vitro, the absence of endocrine activity at certain concentration -- 0.1 percent obviated the Panel's need for DART data.

MS. RAJ: And I guess the genotox takes care of carci?

DR. BELSITO: Yeah. The dermal, as Don reviewed this morning, two out three in vitro means non-sensitizing. This is PDF Page 59, the second paragraph. So we have in vitro data supporting non-sensitization as well.

DR. RETTIE: I wondered about the estrogenic activity that was reported. The concentrations of some kind of context in the YAS agonist assay. Is there any concern about that?

DR. SNYDER: I didn't pick up on that.

DR. RETTIE: So it's at the bottom of Page 60, last paragraph?

DR. SNYDER: Yeah, that's nothing I don't think.

DR. RETTIE: So, that's 6.9 micromolar. Is that what it's talking about?

DR. SNYDER: Yeah.

DR. RETTIE: So that's pretty bad. Yeah. Okay.

DR. SNYDER: You have more, Don?

DR. BELSITO: I'm just trying to look here. Yeah. Under phototoxicity studies. So, basically, they were just doing a UV absorbance and it didn't absorb. So, the test article was predicted to be non-photo toxic and also non-photo allergenic, both endpoints.

Henry's law, less than a thousand, you're fine. And then, I just had a question on the HET CAM study for ocular toxicity. It says published in the *Journal Official Republique Francaise*. Do we know what protocol that was? Because there's no OECD test guidelines for HET CAM. There's an ECVAM protocol, was that the one that was used?

MS. RAJ: I believe so. I mean, whatever was provided in the data is what we put.

DR. BELSITO: Yeah. Rather than putting as published in whatever journal, if they say what protocol they used, I would get rid of the journal and just put the protocol.

MS. RAJ: Just in case the actual protocol numbers isn't found, what would you want then, Dr. Belsito?

DR. BELSITO: Then I think you just reference --

DR. EISENMANN: I'm pretty sure that's France's, like, federal register. That's not a journal. That's like a regulatory.

DR. BELSITO: Okay. So they're probably using an ECVAM criteria?

DR. EISENMANN: I don't know what they were using. But that's like the regular requirement of --

DR. BELSITO: If you could just see if they state what protocol they used for the HET CAM, because there are different protocols. I think the Japanese protocol differs from the European.

DR. KLAASSEN: I'm thinking this is a French CAM.

DR. BELSITO: I think we're getting to a safe as used, but then I just want to point out that in -- that's why I'm getting to the ocular irritation studies -- that one of them had a test article was classified as moderately irritating and this is max uses in an eye area formulation. So, and our safe is used, do we do formulated to be non-irritating?

MS. FIUME: You can. I mean, a lot of times -- the panel has gone both ways on it.

DR. BELSITO: It doesn't hurt.

MS. FIUME: So, yeah.

DR. BELSITO: Because once I got past the issue of biological effect, I thought we could go safe as used when formulated not to be irritating, based upon that ocular irritation. Which I think is probably not real, but there's certainly -- well we don't know

in the Fu study whether it was 0.05 percent. But there was no erythema, no dryness, so there was no evidence of irritation in that study. But we don't know the concentration. So is everyone okay with that conclusion?

DR. SNYDER: Yes.

DR. BELSITO: Okay. So the discussion, Preethi, would be we noted some changes in the keratin profile, but they do not appear to be adverse effects based upon lack of erythema, dryness, trans-epidermal water loss. That the molecule -- low concentration, not likely to be absorbed, lack of endocrine disruption obviated the needs for DART. Anything else in the discussion?

DR. RETTIE: I don't think not observed is appropriate, not for the palmitoylated one anyway.

DR. BELSITO: I'm sorry. Not observed what?

DR. RETTIE: I think you included not absorbed in the list of things that would mitigate concerns. But the palmitoylated peptide is going to be absorbed.

DR. EISENMANN: Into the skin, but not beyond the --

DR. RETTIE: Not beyond it? Is that what it said? Little to no permeation. You're correct. All good.

MS. FIUME: Can I ask for clarification for the Discussion just so it doesn't come back in the future? Does anything need to be discussed regarding the different sequences being that these have been broken up?

DR. BELSITO: No. I wouldn't go that far. Can we take a five minute bio break? Back at 3:15. And then we got to start boogying.

DR. SNYDER: Re-reviews, we're in good shape. We'll be fine.

DR. BELSITO: Yeah. That's what I hear. I don't know we're going to argue about read across.

[BREAK]

Cohen Team – September 11, 2023

DR. COHEN: Myristoyl Pentapeptide-4, Palmitoyl Pentapeptide-4, and Pentapeptide-4. This is a draft report of the safety assessment of these. This is the first time we're seeing these three ingredients, which comprised of five amino acid sequence of a lysine-serine and threonine. And these are sub-fragments of Type 1 collagen propeptide.

The Panel previously reviewed the safety of individual amino acids comprising of these ingredients as well as myristic acid and palmitic acid. And in 2013, the alpha amino acids were considered safe.

In 2019, the panel issued a final report on myristic and palmitic acid with the conclusions they were safe. Apropos to our last conversation, the hydrophilic and charged nature of Pentapeptide-4 makes it difficult for it to pass through an intact stratum corneum. These function as skin conditioning agents.

We have method of manufacturing and impurity. We have a frequency of use of 239 formulations, of which 223 are leave on with a maximum concentration of Palmitoyl Pentapeptide of 0.0035 percent. And Myristoyl Pentapeptide has four uses at 0.05 percent in an eye makeup preparation. So, we have these reported near the eye. It's reported to be in a face powder, which could possibly be inhaled. We have HRIPT in excess of the max use.

We received an article from Don which was referring to a cosmetic product that had retinyl propionate in it as well. So I know it's a little hard to pull out any adverse issues from the chemicals at hand, so I'll open it up for discussion.

DR. HELDRETH: I have one additional thing that Preethi wanted me to hand along. Was that we were looking at this report carefully and the ingredients therein, and realizing that there's actually two different amino acid sequences represented by the same ingredient name. So, Preethi prepared a data table, broken out by the two different amino acid sequences, with the thought that maybe you wouldn't think that read across would be appropriate between the two.

DR. ROSS: Yeah, I mean, I struggled with that. And I haven't seen this new table, but because they are different peptide structures I didn't think you could read across. And you know we only have the penetration and degradation and masking from one of those forms, and that's the KTTKS. The KTSKS -- this is going to get real confusing -- we don't have that. And so, I thought that probably should be done. The Palmitoyl KTSKS and masking.

Due to the lack of penetration stability, which David referred to, in the skin, I wasn't too concerned with systemic tox but there was no dermal effects on the, again, the KTSKS. So, I thought that needed to be done.

DR. COHEN: Can you repeat so I have that again?

DR. ROSS: Yeah. I mean, if we can get some sort of dermal toxicity on the KTSKS --

DR. COHEN: KTSKS.

DR. ROSS: But, I mean, the first thing, I think, would be -- the first point of departure here would be do we see penetration and degradation with the KTSKS. The Palmitoyl KTSKS? Because as, I think, Preethi has split this out here -- yeah. So, you see here, David, there's very little data with the KTSKS.

DR. COHEN: Right. And so, can you read-across with different peptide sequences?

DR. ROSS: I don't think so. There might be some discussion of that tomorrow, but I wouldn't have thought so, no. I'm sure Susan has an opinion on that.

DR. COHEN: Susan? Susan, what are your thoughts on read-across?

DR. TILTON: Yeah, I mean, I had the same questions, actually, about whether or not we could read across for the different peptides. I do think, especially, since we're at the draft report stage that we could request data on absorption or penetration and degradation dermally.

DR. COHEN: For the same one David mentioned, the KTSKS?

DR. TILTON: Yes. KTTKS?

DR. COHEN: Wait, K --

DR. ROSS: KTSKS.

DR. COHEN: KTS- -- well, let me see.

DR. TILTON: Oh.

DR. COHEN: We don't have acute tox on the KTSKS. We don't have that. Okay. I think that makes sense because we don't know what the biological signal will be by switching the two peptides around, right?

DR. ROSS: We don't --

DR. HELDRETH: I'll say in a previous safety assessment by CIR -- or by the Expert Panel I should say -- this had to be at least five or six years ago, the Panel did a report on so-called oligopeptides. And we ran into very similar situation where one ingredient name resulted in two different amino acid sequences that could fit under that name.

So, the Panel drafted the Discussion and their Conclusion at those sequences. So, it was something like oligopeptide X was safe as used when the amino acid sequences KTSKS or something like that.

And so, in the same report, you're looking at one ingredient, per se, but you're really looking at two different chemicals.

DR. SLAGA: Right.

DR. COHEN: I think we're all in agreement on that. So, what other data needs might we have? I just don't want to leave it hanging with just one and not have everything now.

DR. ROSS: What have you got there, David? What's your list so far?

DR. COHEN: Dermal tox on KTSKS. We have -- oh, I got to look back on this one. We have human sensitization on both, and I think they're at good concentrations.

DR. ROSS: The human HRIPT -- was the concentration specified for KTTKS? I have in my notes that it wasn't specified.

DR. HELDRETH: Yeah. I don't think it was specified. If you see on the list there you'll see sometimes there's an X next to, say like Palmitoyl Pentapeptide-4, you see there's an X under reported use. Having an X there in that category means that we have data, but it didn't specify if it's -- what amino acid sequence.

DR. ROSS: Could we try and get that concentration or, I mean, it would be impossible to get?

DR. HELDRETH: We could ask for it.

DR. ROSS: What's -- yeah.

DR. COHEN: Why would it be impossible?

DR. ROSS: I mean, it must be there somewhere.

DR. COHEN: Yeah. K- -- well.

DR. ROSS: The Palmitoyl KTTKS.

DR. COHEN: Well, we have KTTKS at 0.01 percent.

DR. ROSS: What page are you on?

DR. COHEN: I'm on 66. This is for irritation. And for sensitization for KTTKS, on the HRIPT, we don't have concentration so we need that.

DR. ROSS: Yeah. That's right.

DR. COHEN: Concentration --

DR. ROSS: Wasn't specified.

DR. COHEN: -- of the HRI- -- I mean, they just need to pull a report for that one. It's referenced. Now, we're going to read across for Pentapeptide-4 and Myristoyl Pentapeptide-4.

DR. ROSS: I don't know. Are we?

DR. COHEN: That's why I'm asking.

DR. ROSS: I mean, there are very few uses with the Myristoyl, but it's mainly ocular. But yeah, I mean, those groups are going to govern penetration. In a different way, they're going to get to different spots. I mean, you could make an argument for reading across, but I think being conservative, maybe we shouldn't read across them.

DR. COHEN: So, then we need the full dossier for the Myristoyl Pentapeptide-4.

DR. ROSS: You would. I think you'd need to start out with those skin penetration studies, because there's not much point in doing a full dossier of toxicity if it doesn't get in, right?

DR. COHEN: So, we need dermal tox for not only KTSKS but for the --

DR. ROSS: First and foremost, you need the skin penetration and degradation study. And I think that should be number one for the Palmitoyl as well.

DR. SLAGA: That was the only need that I found.

DR. COHEN: Which one?

DR. SLAGA: Was the penetration and --

DR. ROSS: Yeah.

DR. COHEN: For the Myristoyl?

DR. SLAGA: Yeah.

DR. COHEN: And you mentioned one other thing right after that. I didn't catch it. Skin penetration for the Myristoyl and --

DR. ROSS: And degradation.

DR. COHEN: And degradation. Yeah, penetration and degradation for Myristoyl.

DR. ROSS: Now the Myristoyl is used mainly ocular, four uses. So, we've got no ocular data with the Myristoyl.

DR. COHEN: Ocular tox?

DR. ROSS: Well, yeah, some sort of molecular test. A HET-CAM or something like that. Because the maximum concentration of that one, I think, is a lot higher than the Palmitoyl.

DR. COHEN: Hold on, that's -- the Palmitoyl goes by the eye, right, at point 0.05 percent right?

DR. ROSS: Myristoyl right?

DR. COHEN: I have Palmitoyl Pentapeptide-4 is used to up to 0.05 percent in eye makeup preparations.

DR. ROSS: Yeah, I question whether that was correct or not.

DR. COHEN: Because in another place it says Myristoyl Pentapeptide-4, right?

DR. ROSS: Exactly. So, I think the max use that I got from --

DR. COHEN: We've got to go to the table and make sure did I pull this out correctly.

DR. ROSS: Yeah, let's look at the table. The max use on Palmitoyl is 0.0035 percent.

DR. COHEN: It's 0.0035 percent for which?

DR. ROSS: For the Palmitoyl.

DR. COHEN: Where did this come from?

DR. ROSS: And the max use on the Myristoyl is 0.05 percent, it's a lot higher. So even if you were doing read across, you don't have a high enough concentration so you're going to need it in those ocular tests of some sort. But yeah, I had a comment in here that maybe that was a misstatement.

DR. COHEN: Oh, you saw the same thing?

DR. ROSS: Yeah. I can find the sticky note.

DR. COHEN: I'm just putting one more thing down before I hit that.

DR. ROSS: Yeah. That was on Page 56 of the PDF. And it says that Palmitoyl Pentapeptide-4 is used at up to 0.05 percent in eye makeup. And so, I asked the question should that be Myristoyl?

DR. COHEN: Yeah, because in Table 3 there's no max use at that concentration listed.

DR. ROSS: Correct. Yeah. So, I think it just --

DR. COHEN: It's a typo.

DR. ROSS: It's a typo, yeah.

DR. HELDRETH: Where is that, again?

DR. ROSS: I just had it there. So, I think I said --

DR. COHEN: I'll find it --

DR. ROSS: -- 56 of the PDF that I have.

DR. COHEN: It's in Cosmetic Use. Use Cosmetic. One, two, three, fourth paragraph, second part of the first line.

DR. ROSS: The paragraph starts, "Some of the ingredients are reported to be used in products that are applied near the eye."

DR. COHEN: And it says Palmitoyl Pentapeptide-4 is used up to 0.05 percent in eye makeup preparations. We don't think is correct, but just need to double check that. Because either the sentence is wrong, or the table is wrong.

DR. ROSS: Yeah. So, we've got penetration and degradation on the different Palmitoyl.

DR. COHEN: Wait, not Myristoyl?

DR. ROSS: And Myristoyl.

DR. COHEN: Palmitoyl and Myristoyl?

DR. ROSS: Yeah.

DR. COHEN: So, the KTSKS?

DR. ROSS: Yeah.

DR. COHEN: All right, is it --

DR. ROSS: KTSKS.

DR. COHEN: Pal-KTSKS.

DR. ROSS: Yeah.

DR. COHEN: Concentration of the HRIPT at KTTKS. Ocular data tox.

DR. ROSS: Yeah, some sort of molecular test for the Myristoyl.

DR. COHEN: You mean molecular or in vitro test?

DR. ROSS: Anything. I'll take anything that shows some sort of safety at 0.05 percent on ocular cells.

DR. COHEN: We can't read across. Okay.

DR. ROSS: But, you know, if for example, Myristoyl doesn't get in, or the other form of Palmitoyl doesn't get in, do we need any of the other tests if these things don't get in and they're rapidly degraded.

DR. COHEN: No.

DR. BERGFELD: Eye is different than skin.

DR. COHEN: No, no.

DR. ROSS: Even with skin, yeah.

DR. COHEN: No, no, he's saying if the Palmitoyl and the -- if the Myristoyl doesn't get in, do we need the whole repertoire of the whole HRIPT and everything else if the sequence is the same? Right? The peptide sequence is going to be the same.

DR. ROSS: The peptide sequence would be the same, yeah.

DR. TILTON: Yeah, I think we could use sort of an if/then situation.

DR. ROSS: Yeah.

DR. COHEN: And by the way, does anyone remember the Myristoyl Pentapeptide uses which sequence? Is it KTTKS? Do we know, do we remember?

DR. HELDRETH: So, the reason that it's listed this way without either sequence written, is because we don't -- the study that has the X there didn't determine which amino acid sequence.

DR. ROSS: Oh, really?

DR. HELDRETH: So, you think about it as Pentapeptide-4 without any fatty acid attached to it. So, it could be either sequence, but many times the research papers don't tell you. I think, a lot of times the authors didn't even know that they were looking at potentially two different sequences.

DR. COHEN: So, if we have the full dossier of both sequence -- but we don't even know if those two sequences are the ones used in the Myristoyl, do we?

DR. HELDRETH: Yes, we do. So, think about there's Pentapeptide-4, it's got the two amino acid sequences. So, then Palmitoyl Pentapeptide can be a palmitoylated version of either one, as can Myristoyl be a myristoylated version of either sequence there.

DR. COHEN: Yeah.

DR. HELDRETH: But we found data on the two different amino acid sequences for Palmitoyl Pentapeptide-4, but not for the Myristoyl and not for the unesterified Pentapeptide-4.

DR. COHEN: So, the anchor to read across is going to be on the Palmitoyl Pentapeptide because we know the two sequences there. Right? So, we can assume it's one or the other based on the other two, but we need all the data on both of them. Does that make sense or no?

DR. ROSS: But if it doesn't get in and it's degraded in the skin, and you're looking at many skin effects, specifically, rather than systemic effects.

DR. COHEN: So, we have irritation and sensitization. We don't have any carci data on it. But we don't know if there's any other biologic signals from this in the skin.

DR. ROSS: Yeah. I think that was part of sort of what Don was getting at with his paper. Are these things having other effects on the skin, specifically?

DR. COHEN: They are very complicated with the other ingredients though, because it had a retinoid in it.

DR. ROSS: Yeah, but the cellular effects in skin, though, I think the last time this was discussed there was a lot of discussion about angiogenesis. I didn't see that cropping up at all in this document, which is fine.

DR. COHEN: So, Tom, do we need carcinogenicity data or other -- we have in vitro genotox, right?

DR. SLAGA: Yeah. We have sufficient genotox and it's not an irritant, so I don't think we need any carcinogenicity.

DR. COHEN: Okay.

DR. ROSS: I don't think so either.

DR. COHEN: Okay.

DR. ROSS: I agree with Tom there. One thing I would say, one of these things was estrogenic in the saccharomyces study. We have no DART data.

But if you actually go back into that saccharomyces yeast study, I went back into it actually. The max concentration we have of the Palmitoyl Pentapeptide is 0.0035 percent, okay. And when I did the cross calculation on molarity that comes out to 4.3 times 10 to the minus 5. And the reason I'm saying that, is that if you go to the actual data produced from industry -- I think that's PDF Page 231 -- there was no agonist activity at that concentration. So, I think we're okay there.

So even though it had some estrogen antagonist activity --

DR. COHEN: 231?

DR. ROSS: Yeah. It's a long way down, I know.

DR. COHEN: That's a lot of thumb action on the little pad here.

DR. ROSS: You'll see a graph. You've actually got to go into the real data.

DR. COHEN: Inactive.

DR. ROSS: So, I think we're okay there.

DR. COHEN: Was the concentration appropriate, you said?

DR. ROSS: Yeah.

DR. BERGFELD: Concentration is no effect.

DR. ROSS: Yeah.

DR. COHEN: Yeah. No. I just want to make sure we weren't dealing with an order or two orders of magnitude lower a concentration that we're dealing with here, because then it wouldn't be fungible to this discussion, right? Okay.

DR. ROSS: So, I think we're okay.

DR. COHEN: All right, that'll be good discussion for tomorrow.

DR. BERGFELD: Summarize what you're going to ask for, please.

DR. COHEN: Okay, so we do not feel we can read across with the different peptide sequences. We'd like dermal tox on KTSKS, skin penetration and degradation from Myristoyl and palmitic KTSKS. Ocular tox or in vitro ocular data on Myristoyl pentapeptide-4, and the concentration of the HRIPT for KTTKS.

DR. ROSS: Can you lead with the degradation and penetration of the skin? I believe that's the first.

DR. COHEN: Should we lead with not read-across, though?

DR. ROSS: Oh, yeah. You could lead with not read-across, but then ask for the penetration and degradation in the skin. I think that's the first issue.

DR. COHEN: Will do.

DR. ROSS: And then if we need additional tox after that -- I think Susan commented earlier we could do this as an if/then kind of thing.

DR. COHEN: So, the dermal tox for KTSKS is part of the if/then statement?

DR. ROSS: Yeah, if it gets in. Yeah.

DR. COHEN: Got it. Are we satisfied with that insufficiency list? Yeah.

DR. BERGFELD: You felt that human studies were fine? I forgot if they had them.

DR. COHEN: The human studies, we're okay. We just don't have a concentration on the HRIPT.

DR. BERGFELD: Okay.

DR. COHEN: I mean, I think that's very doable to get that. I mean, it's a cited report. It probably was just left off when it came in. Okay.

DR. BERGFELD: Going out as an IDA?

DR. COHEN: Yes. That was a complicated one.

DR. ROSS: Yeah.

Full Panel – September 12, 2023

DR. BELSITO: Yep. Okay, so this is a draft report on the safety assessment of the Myristoyl Pentapeptide-4, Palmitoyl Pentapeptide-4, and Pentapeptide-4 as used in cosmetics. It's the first time that we're looking at this material.

I had a little concern about whether there was biological activity. Because in the Fu study, although there was no increase in trans-epidermal water loss, there was no erythema, there was no dryness. There was a slight change in the keratin profile. We discussed this somewhat extensively in our group, and we felt that these effects were not adverse effects and didn't appear to alter biological function. And so, we were fine with going ahead with a safe as use conclusion for these.

DR. SNYDER: No.

DR. BELSITO: Oh wait a minute. I'm sorry.

DR. SNYDER: With the caveat, non-irritating due to --

DR. BELSITO: Right. Non-irritating because of some irritation studies around the eye. Correct.

DR. BERGFELD: You're putting that in the Conclusion or the Discussion?

DR. BELSITO: When formulated to be non-irritating.

DR. BERGFELD: Okay. Dr. Cohen?

DR. COHEN: So we contemplated this quite a bit as well. We felt, primarily, that because the peptide sequences were different that we couldn't read across. I think if we read-across we might have come to similar conclusions, but we didn't have a comfort level that we could say the biologic activity was going to be the same if you switch the sequence of the amino acids.

So, we proposed an IDA asking for HRIPT for the KTSKS, skin penetration and degradation for Myristoyl and Palmitoyl KTSKS, and if it gets in, dermal tox, and some ocular tox or in vivo data on Myristoyl Pentapeptide. And the concentration of the HRIPT in the KTTKS report because it was listed as blank.

DR. BERGFELD: Any other comments? We have a motion and we have a counter opinion.

DR. RETTIE: Could I just make a comment on the read across. Sure they're different pentapeptides, but the difference is switching a serine at a three position to a threonine at the three position, which is the most conservative change you can make. And so, on the basis of that, at least, I felt more comfort that we could read across. I think that was maybe our basis for coming to a different conclusion on the read across than you.

DR. COHEN: We certainly didn't have a level of confidence, perhaps, based on -- I certainly couldn't say that that substitution was going to be biologically similar. You've made a comment about the conservativeness of that substitution, but you want to comment on it, David?

DR. RETTIE: I mean there's just both alcohols, serine and threonine. Very similar physical chemical properties?

DR. ROSS: No, it is -- I mean, it's a reasonable discussion to have because it is a relatively minor change. But since this was the first time we were seeing this, we wanted to ask for, and we felt it would be valuable for a second or third time we see it, to look at the other peptide and look at penetration -- from Palmitoyl -- look at penetration and degradation in the skin to have similar data with that.

It's a judgment call whether they're going to be suitable for read across or not. We often get into this discussion, is it a read across or is it not a read across? And we came down on the side that it wasn't in this case, and you came down on the other side of that it sounds like. So, I guess we have to come to some resolution of that.

DR. BERGFELD: You have proposed --

DR. BELSITO: This is not in my area of expertise, but if we think that the substitution is minor and would not result in any significant changes -- again, I'm very much aware that we should not be asking for data that the next go around if we don't get it, we go, oh yeah, okay, we can read across. So, if you really believe that we can't do that, that's one thing. If it's like, oh, it's the first time and it would be nice to have it, but I'm going to say it's okay if I don't get it, then I don't think that's reasonable.

DR. ROSS: I think I said that if it was the first time, the second time or the third time, it would be valuable to have. So, it's not just asking for it because it's the first time. I think we were in the position that there wasn't a read across here. And, I guess, as I pointed out, you've come down on the other side of that.

DR. BELSITO: So, you felt -- because you said it was a minor substitution. But you still felt that it could not be read across?

DR. ROSS: Yeah, it's peptides. I'm not sure what the activity of different peptides are going to be.

DR. COHEN: Yeah, we just didn't have comfort.

DR. RETTIE: That's a very conservative approach and it's very difficult to argue with. Like I said, I was moved by the similarity in the two amino acid structures. Any time I made a serine to threonine mutation in any of my CYP enzymes it didn't make any difference, but that's a whole different kettle of fish. I mean I can acquiesce to your cautious approach since it is the first time we're looking at this.

DR. BELSITO: Okay, fine. The only other question that I then have is, since we're saying formulated to be non-irritating, why do you want additional ocular data on Myristoyl Pentapeptide?

DR. COHEN: Don, I think that would cover that, right? We weren't creating our IDA based on your conclusion.

DR. ROSS: Yeah, we didn't have the non-irritating in our conclusion.

DR. COHEN: We didn't have a conclusion.

DR. ROSS: Yeah, I mean the Myristoyl, there was four uses and I think they were all essentially ocular.

DR. BELSITO: Right. It's .05.

DR. ROSS: And so, that's why we went with some molecular tests for ocular irritation.

DR. BELSITO: Well, we have ocular irritation, it's just on Palmitoyl.

DR. ROSS: Yeah, exactly.

DR. BERGFELD: So I gather, Don, you're rescinding your motion?

DR. BELSITO: I believe it was David's motion, no?

DR. BERGFELD: Yours.

DR. COHEN: No, it's yours.

DR. BELSITO: Oh, my motion. Yeah, fine. So, insufficient for -- David repeat your needs.

DR. COHEN: We wanted an HRIPT at 0.12 percent for KTSKS.

DR. SNYDER: HRIPT or just sensitization data?

DR. COHEN: Thank you. Irritation and sensitization. Thank you. That's right, especially after the last lecture we had. irritation and sensitization for KTSKS. Skin penetration and degradation for Myristoyl and Palmitoyl, KTSKS, and if it gets in, dermal tox. Concentration of the HRIPT report for KTTKS.

DR. BELSITO: Since we're saying formulated to be nonirritating, do you still need the irritation data?

DR. COHEN: Well it was sensitization as well.

DR. BELSITO: Well, but they could do sensitization in vitro. It'd be a lot cheaper.

DR. COHEN: No, but the report's done. The report is in there, it just didn't list the concentration. So, if the old report can be just pulled up and we see what the concentration is, we don't need to go invent new data.

DR. BELSITO: Okay, so what you're asking for is that they clarify the data that's already existing?

DR. COHEN: Exactly, exactly. We weren't asking for new data.

DR. BELSITO: Because it sounded like you were asking for new data.

DR. ROSS: It did sound a bit like that, actually, I agree with you. But, I think, clarification was what we were after. And it's on PDF Page -- for the writers -- it's on PDF Page 67. And it's the second entry in the Human Table.

DR. COHEN: It's just blank.

DR. ROSS: Just says not specified. Test concentration.

DR. BELSITO: Okay, fine.

DR. BERGFELD: All right, so we have a second?

DR. BELSITO: Yes.

DR. BERGFELD: And we understand what we need?

DR. BELSITO: Yep.

DR. BERGFELD: All right.

DR. BELSITO: I do.

DR. BERGFELD: It's all been clarified, I'll call for the vote. All those in favor of an IDA? Thank you. I saw all the raised hands. Thank you. Unanimous. Moving on to our next ingredient, Dr. Cohen, Charcoal ingredients.

MARCH 2013 PANEL MEETING – INITIAL REVIEW/DRAFT REPORT

Belsito Team – March 18, 2013

DR. BELSITO: Well, I thought that -- I mean, those are case reports. I thought that the sensitization data was fine in terms of covering the range of use. So I really didn't have an issue with that.

I mean, you're occasionally going to get a case of a report of something causing a problem in somebody. I thought your data was sufficient enough to support the concentrations of use.

Anything else? Okay, palmitoyl oligopeptides. The day has come. I think that this is the first time we're looking at this. We do have some data -- safety test data on a few ingredients.

But the issue I had was that these peptide residues vary in chain length. We're told that the order of the amino acids within the residue can be very different despite having the same name. Clearly, some of these are biologically active, not only in terms of things that might not give me concern, like collagen synthesis in getting rid of wrinkles, but muting proinflammatory cytokines like IL6.

So one of the questions I would have is, what happens if you put this over a skin cancer, like a melanoma, and you reduce immune response, do you enhance the risk of metastases?

My safety issues with this group went on and on, and I really thought that sort of at the end of the day maybe -- let me see if I can see all the comments.

It's really difficult, and I almost felt that there were only two products -- the nanofiber gel and the biocide, nanofiber gel and the biopeptide CL -- that maybe had enough data, but even then you had to go set up with trade name because the other ones might have totally different amino acid sequences as far as my understanding.

So I had a real hard time wrapping myself around this family of ingredients and even thinking that we could do palmitoyl oligopeptides as a family.

So, with that as an overview, I'll turn it over to -- I had a lot of comments throughout the document, but I'll turn that over to Dan and Paul.

DR. LIEBLER: Okay. So, this one -- I had a lot of comments on it.

It seems more complicated than it really is, just from a chemistry perspective. But these products are all peptides, and the end terminus is modified by palmitic acid. So it's an N-palmitoyl version of each of these peptides.

The ingredients have a nomenclature that's confusing at first because a palmitoyl dipeptide, for example, could have a number of different ingredients. But the number after the dipeptide is apparently a code for which amino acids are in the dipeptide or in the tripeptide or in the tetrapeptide and so forth.

And I think table 1 provides some of that information, but it's confusing to me because when I read the method of manufacturer it indicates that most of these and almost all of these are produced by solid-phase peptide synthesis, which generally means you put on specifically one amino acid; then you put on another amino acid; then you put on another and another. And so, that will produce peptide sequences of high purity.

And of course, the last step -- you put on the palmitic acid on the end terminus, and you're done. That generally produces peptides of high purity in a very well defined composition sequence.

Now the table 1 is confusing because it has entries that indicate this tripeptide, for example, may contain lysine and glycine, for example, or two other amino acids. At least three amino acids with at least two of these -- I mean -- I'm sorry. A tripeptide with at least two of these amino acids in any order.

The in-any-order part, which appears in the table, seems to be inconsistent with the way solid-phase peptide synthesis works unless the synthesis actually took as the first amino acid a pool of the possible amino acids, added that, and then in the second step used a pool instead of a pure amino acid and added that.

And I can't tell -- I mean, I don't know how it's done. I could imagine that that could be done.

I'm not sure why you would do it except to have more diversity in the structures and only have to do one batch process to make the compound.

So I think we need to have more information about that because the method of synthesis suggests logically that these should be defined sequences whereas the table information suggests that these are semi-randomized sequences of a least defined composition in terms of the amino acids.

DR. BELSITO: Well, we're talking about table 1, Wilbur. There were three ingredients where you say monograph development in progress. Is this a REACH dossier? What is this monograph that's in process?

DR. ANDERSEN: Something new.

DR. BRESLAWEC: It's an INCI monograph.

DR. BELSITO: INCI monograph.

DR. BRESLAWEC: Nomenclature.

DR. BELSITO: Nomenclature, okay.

DR. ANDERSEN: Something new is being added to the dictionary.

DR. BELSITO: Okay.

DR. ANDERSEN: Dan, what's the counterpart methodology that would yield a gemisch of ordering?

DR. LIEBLER: Well, if you did pools, if you used the solid-phase method and you did pools instead of -- for example, let's say you were making -- I'm trying to find an example.

Here's palmitoyl peptide-4, which is page 33 of the PDF file. It's got lysine-threonine, lysine-serine as the four amino acids. You could start with a pool of all of serine, lysine, threonine, and then the first residue would be a mixture of those, proportional to what's in the pool. And then in the next you could use another pool. But then you wouldn't get a defined sequence that is portrayed here.

It seems to me that something is not correct in the way that this is described. It could be that these are actually randomized sequences that contain the named amino acids, but it might be that they're not really randomized. So I don't know.

That information should be available. It's just a matter of asking the precise of the manufacturers about how it's actually done and what these do contain.

DR. EISENMANN: I think they've had difficult naming these ingredients over the years as I think you've noticed. So they've started -- one of the first ones that was ever named is this palmitoyl oligopeptide.

DR. LIEBLER: Right.

DR. EISENMANN: And when they gave it a name, I think they gave a name to cover more than one peptide.

DR. LIEBLER: Yes, right. That sounds like a catch-all name.

DR. EISENMANN: Right, right. It's a catch-all name, and now -- right now -- one company is using that name for two sequences.

And then there are other names in there where they -- and then they've gone to this naming, you know, palmitoyl peptide, dipeptide-2. So the next peptide will get the next number.

DR. LIEBLER: Right.

DR. EISENMANN: And, unfortunately, the name is not sequence-specific, and I'm not sure why the INCI committee decided to do it this way. But it seems like the ingredients are sequence-specific based on the information that's been coming in.

DR. LIEBLER: Yes. So, I mean, if the decision has been taken not to make the name sequence-specific, that's out of our hands. We can still deal with these. It just makes it a little trickier.

But what we need is the table 1, essentially, to be a more accurate look-up representation. So, if we look at palmitoyl oligopeptide-6 and we look to table 1, we can see that that's, you know, valine-lysine, valine-histamine, or something like that. We can find it.

DR. EISENMANN: Right. They're just putting them in alphabetical order in the definition, but the information that's been coming in made them telling the sequence of what they actually are.

DR. LIEBLER: So, if these are defined sequence, table 1 currently suggests that they're semi-random.

DR. EISENMANN: Correct.

DR. LIEBLER: So that probably isn't correct. So we need to fix table 1 and then just add the defined sequence.

DR. EISENMANN: Unfortunately, that's the definition.

And I don't know if you've read the memo -- that we're suggesting that you cut back on these ingredients, not to do all the peptides together, that you should pick a peptide in addition to like palmitoyl.

DR. BELSITO: When did we get that memo?

DR. LIEBLER: I might not have gotten that memo.

DR. EISENMANN: I put it on your (inaudible) this morning.

DR. LIEBLER: Oh, I didn't look at that.

DR. EISENMANN: From CIR SSC.

DR. LIEBLER: So what do you want to do?

DR. BRESLAWEC: Well, our proposal is --

DR. EISENMANN: Do a peptide instead of basing it on palmitoyl because, unfortunately, the two peptides that are in palmitoyl oligopeptide also have other names. They're also tripeptide-1 and hexapeptide-12.

DR. LIEBLER: Okay. So we respectfully suggest that CIR Expert Panel table as before so that the panel and staff can consider the following suggestions and develop reasonable science-based strategy for grouping.

So I definitely agree with the table again at this point.

DR. EISENMANN: So then the focus would be for this report just to do tripeptide, which is this three sequence. It's lysine-lysine and the other one is valine- glysine, valine-alanine-lysine. That's the two --

DR. BELSITO: So do a specific amino acid sequence.

DR. EISENMANN: Correct.

DR. BELSITO: And say that specific sequence --

DR. EISENMANN: Correct.

DR. BELSITO: -- is okay.

DR. EISENMANN: Right. That sequence would be sold under these names, under more than one name, unfortunately, currently.

DR. LIEBLER: Well, that would the least of our problems.

DR. BELSITO: But would there be -- I guess I'm less concerned about the same thing having different names than different things having the same name.

DR. BRESLAWEC: But that's a situation that exists with the current naming.

DR. BELSITO: How is that going to be rectified?

DR. EISENMANN: You would have to say that your judgment is on this sequence, and if something else is being sold under this name --

DR. BELSITO: So, in other words, the report could not be titled The Safety of Ingredient X. It would have to be The Safety of Amino Acid ABCD Palmitoyl and Amino Acid.

I mean, how are you going to do that?

DR. EISENMANN: In some ways it's similar to when you do botanicals. For some botanicals, you say the safety you're assessing is of the extract that was tested.

So that also would be true here. It would be the safety of the sequence for which you have data.

DR. BRESLAWEC: What this does is it takes it out of the basic review on the palmitoyl component and focusing on the peptide.

DR. BELSITO: Yes, which is where we need to focus.

I mean, I don't have a problem with that. I don't have a problem looking over the data again. I think that would certainly make it much easier -- to look at a specific amino acid sequence and see what data are out there for it.

I guess I just want to go on record; if one of them is the one that down-regulates IL6, I'm very concerned about a cosmetic product that now is having a potentially significant biological effect in terms of immune responses.

That might be very beneficial for the rosacea patient who has erythema, but I'm concerned about the patient who has a skin cancer who's throwing something on that's going to down-regulate proinflammatory immunity. Just, my big concern with these.

DR. LIEBLER: Yes.

DR. BELSITO: I'm not concerned about sensitization. I'm not concerned about irritation.

DR. LIEBLER: I do think that those claims are being somewhat oversold when I look at the references that supposedly support these kinds of biospecific effects based on these peptides.

I mean, we'll have to come back to that when we have specific ingredients in the table and consider that. It's a potentially important consideration, but I do believe from what I saw that some of that stuff is hype. For example, in a couple of spots in the report, some of these sequences were referred to as being part of a certain collagen or a certain antibody, but with a di or tripeptide sequence that doesn't mean squat, you know, when it comes down to biological activity.

And those sequences are also parts of many other proteins, especially when you're down to a tri or tetrapeptide sequence. They're highly -- appear in many other proteins.

DR. BELSITO: Okay. I mean, I guess the only other comment -- I know this is probably meaningless given the fact that we're going to table this, and I certainly agree with it.

You know, since these are small peptides, when you look at the skin irritation and sensitization studies in table 3, page 41 of the Panel Book and you start looking, early on you see a lot of reports of slight erythema, slight erythema, slight erythema, erythema -- all of which were considered to be nonirritant. But it just makes me a little bit concerned were these urticarial reactions to these peptides.

So, I mean, just as we look at it again I would just like everyone to keep that in mind.

Those are my two major issues.

DR. LIEBLER: So I also was going to suggest removing the palmitoylated hydrolyzed plant and animal proteins as being a bridge too far, and that's, I guess, the last paragraph of this suggestion from the council.

And then these potentially other derivatives, like the acetyl tripeptide, azole tripeptide and the copper complexes and manganese complexes and so forth -- I think we do need to have some further discussion and consideration of how to make this grouping more rationale.

DR. SNYDER: So how easy is it going to be -- if we go for the peptide, how easy is it going to be to then know exactly what in the dictionary is encompassed under the peptide designation rather than the lead ingredient, palmitoyl?

So, I mean, how are we going to capture -- are we going to be able to capture all the ingredients?

DR. BRESLAWEC: First of all, I would be glad to bring the editor of the INCI dictionary to provide a briefing about the nomenclature and how it's applied to peptides. I'm not sure it's going to clear everything up, but I think it might provide a better framework for the discussion.

We're not suggesting that from now on every single protein, or peptide, be looked at separately. Our suggestion is based on the fact that this is a new type of ingredient for the panel and work through a more limited sort of report, identify the issues that are critical and then consider, or reconsider, different grouping mechanisms.

So we're not suggesting every peptide should be reviewed on its own from now on.

DR. BELSITO: I totally agree. My thought as I tried to wrap my arms around this family -- I just didn't see it as a family.

So I applaud PCPC for coming up with that approach and also having us look at defined amino acid groups rather than saying, well, this is four amino acids that can be arranged any way you want.

DR. BRESLAWEC: Then again, I don't know. Maybe at the end of your discussion you'll determine that, gee, that's okay.

DR. BELSITO: Right.

DR. BRESLAWEC: But I'm not sure that you can reach that conclusion now.

DR. BELSITO: Right.

DR. BRESLAWEC: And we certainly can't.

DR. BELSITO: Mm-hmm.

DR. LIEBLER: So the idea would be that we would do a focused report with a couple of ingredients and then once we've got our bearings either reopen the report or do another report with more ingredients?

DR. BRESLAWEC: I think that's an administrative question --

DR. LIEBLER: Okay.

DR. BRESLAWEC: -- and can be handled pretty much a lot of different ways.

DR. LIEBLER: Okay, but I agree with that strategy.

DR. BELSITO: But don't you think the order of the amino acid is going to have very significant effects on their biological activities, or you think these are so small and they're linked to this palmitoyl group that it really doesn't matter?

DR. LIEBLER: I favor the latter. I mean, in other words, I don't think that these -- I don't think that we're going to be running into magic sequences that have profound biological activities with these very small peptides.

My only concern is when we actually get into larger peptides from hydrolyzed proteins. That's when we might actually get into antigenic epitopes that would be likely to produce allergic responses. I'm not sure that we would be getting, you know, profound mimicry of biological signaling molecules with these peptides, for example, certainly not with dipeptides, tetrapeptides, those kinds of things here.

DR. BELSITO: Anything else? Okay, if not, tabled.

DR. ANDERSEN: Tabling it is easy. Thinking about what the right groupings should be is a step that I'm not comfortable that I understand what's being proposed by the council.

So I think we've got a lot of work ahead of us to figure out just what such a grouping would look like, strategically. I wish it was clear to me now, but it's not.

DR. BRESLAWEC: We also wish we could propose a clear path forward here, and I'm not sure that we can. We just think it deserves, you know, a little more time and consideration.

MR. JOHNSON: I just have one question regarding one of the comments provided, you know, stating that the substances in the report do not have INCI names and they should be deleted from the safety assessment.

DR. BELSITO: They don't have cosmetic uses. I don't know that they should be deleted. I think it's the purview of the panel, you know, as to whether we think it contributes to the understanding of the safety of the ingredients that are actually used as cosmetics.

DR. EISENMANN: Well, if the report now is going to be on a specific sequence, then data on other sequences are not necessary.

DR. BELSITO: Right. That would be --

DR. EISENMANN: So that's those other -- some sequences they're developing just to simulate the immune system, and those aren't relevant to --

DR. BELSITO: No, no, no. We're going to look at two or three specific amino acid sequences, and all of this other -- any data on other sequences should not be included in the report.

DR. BRESLAWEC: I guess my question is, did you decide then to go ahead with this study but limited to the one ingredient which has two different specific peptides in it and table the rest?

Are you planning on moving ahead --

DR. BELSITO: No, we're going to table the whole thing.

DR. BRESLAWEC: The whole thing, okay.

DR. BELSITO: Yes. I mean, I think it will be easier to look at the data again --

DR. LIEBLER: Right.

DR. BELSITO: -- when all the extraneous stuff has been removed and we know what we're looking at.

DR. BRESLAWEC: Okay.

DR. ANDERSEN: My inclination at this point -- until we better understand the proposal on the flipside here of keeping palmitoyl oligopeptide and adding a whole bunch of things that weren't previously in the report, it's hard to assess that without doing some homework. I could see just backing the report off to the oligopeptide with its two incarnations.

DR. EISENMANN: See, but those two incarnations have other names. That's the thing. The peptide part of it is tripeptide-1 or hexapeptide-12.

DR. ANDERSEN: Okay.

DR. EISENMANN: So that's why I suggested some of these other components.

DR. LIEBLER: Yes, I think having, you know, palmitoyl oligopeptide as sort of the blanket name and then there are other ingredients that are chemically defined, with different names, doesn't really pose a big problem for us for reviewing those.

There could be a problem more on the end of the definitions in the dictionary and how council deals with it. That's another issue really.

But I think particularly for this prototype, if we end up dealing with two or three chemically well-defined substances, then I think we're back in business.

DR. ANDERSEN: Okay. So let me feed it back to you, Carol, and see if I understand the rationale.

Palmitoyl oligopeptide is an old INCI name that is, in truth, two more specific INCY names -- palmitoyl tripeptide-1 and palmitoyl hexapeptide-12.

DR. EISENMANN: Correct.

DR. ANDERSEN: Okay. So you would add those two. And then taking off on that, any safety assessment of palmitoyl tripeptide-1 would legitimately address tripeptide-1 by itself. Any review of palmitoyl hexapeptide-12 would naturally address hexapeptide-12. And as long as you're going to focus on tripeptide-1, why not look at manganese tripeptide-1?

DR. EISENMANN: Right, and I don't know where I cut it, or we cut it -- you know, these other -- I didn't think these other ones in the bottom group were necessarily appropriate to look at, but I just wanted to be inclusive and include every tripeptide-1 that was in the dictionary at this point.

DR. ANDERSEN: I understand, but in a limited fashion --

DR. EISENMANN: Correct.

DR. ANDERSEN: -- the logic is oligopeptide is actually, again, an old definition that includes two specific newly identified names.

So, in a formulation, a company could use either palmitoyl tripeptide-1 or palmitoyl oligopeptide and be kosher as it now stands?

DR. EISENMANN: Probably both. If one company has the name palmitoyl oligopeptide and if you probably bought the ingredient from them, you would use -- so if you bought it from another company, you would probably use palmitoyl tripeptide-1.

DR. ANDERSEN: But for purposes of constructing a family, I think get the logic represented in this first grouping.

DR. LIEBLER: So, Alan, are you suggesting that this construction include the palmitoyl versions of these peptides as well as the non-palmitoyl versions of these peptides?

DR. BELSITO: That would be more reasonable, yes.

DR. ANDERSEN: I think so because I can't picture that the palmitoyl moiety is going to be the issue.

DR. LIEBLER: Well, it's going to change the properties of these a lot.

DR. EISENMANN: Frequently, the peptides -- as I understand it, the peptides themselves aren't necessarily used. They've all been added to the dictionary as part of the naming process. So, if somebody just comes in and wants the palmitoyl peptide, they will also name the peptide itself now, so whether or not the peptide itself is used.

DR. ANDERSEN: It's a building block.

DR. LIEBLER: So that's my concern. If the -- because I think the palmitoyl versions of these short peptides are going to have very different properties than the nonmodified versions and these short peptides are going to really fall into our short peptide analysis family. I'm wondering if they could be included in the other reports we're doing on short peptides or hydrolyzed proteins.

DR. BELSITO: Well, why don't we see what's there?

DR. LIEBLER: Yes.

DR. BELSITO: We don't even know what's in the dictionary. It may turn out that there's only palmitoyl for these, I mean.

DR. EISENMANN: No, they're listed --

DR. LIEBLER: They're listed at the bottom.

DR. BELSITO: So, I mean, let's look at them. We can always delete the ingredients, but I'm certainly more comfortable using the framework of the amino acid sequence and then look at what's added to it rather than palmitoyl with any old amino acid sequence.

DR. LIEBLER: Okay. I mean, we're going to go through another kind of --

DR. BELSITO: We're going to table it again anyway.

DR. LIEBLER: Yes, we're going to go through another round of thinking about this and another round of discussion, and we may end up backtracking a little bit on some things that we come up with today. That's fine.

DR. BELSITO: Okay. Anything else?

MR. JOHNSON: Dr. Belsito, I have one question.

DR. BELSITO: Yes.

MR. JOHNSON: Carol mentioned two palmitoyl oligopeptides. I know on Panel Book page 14 under Composition and Impurities there are two different CAS numbers listed for palmitoyl oligopeptide. And I was wondering, are those indicative of the two different names that you were referring to earlier?

Those two, okay.

DR. BELSITO: We will also see structures next time and have very specific molecules to work with.

DR. EISENMANN: They provided sequences.

DR. BELSITO: Yes.

DR. EISENMANN: All right.

DR. BELSITO: Okay, Christina, you're up -- 6- hydroxyindole.

Marks Team – March 18, 2013

DR. MARKS: Are there any other comments? If not, we'll move on to the next ingredient. Wilbur, you're up again. This is the palmitoyl oligopeptides, and this is the first time we're looking at this group of ingredients. We received a memo dated March 18 on paper. This morning I found it on my desk here from the CIR Science and Support Committee of the Personal Care Products Council suggesting that this group of ingredients should be tabled. There are some issues in terms of nomenclature and also a suggestion as to what should be included. Do you want to address that memo? I think that's important. Have you seen this memo, Tom, Ron, Ron? Are you reading it now?

DR. JOHNSON: I think our primary objection is that the family is generated by the palmitoyl moiety when we don't think the palmitoyl moiety is going to drive the physiologic activity. We think it would be more appropriate from a toxicologic standpoint to generate the families based on the protein loyalty which is more likely to be the driver. We're not suggesting that the family is wholly inappropriate or that it needs to be done material by material, but we would slice and dice this group quite differently and not based on alphabetization but, rather, generated on chemistry and physiology and therefore are not really prepared to suggest how the grouping should be formed today but that it be tabled and reexamined.

DR. MARKS: I see Ron Hill shaking his head yes, nonverbally communicating he likes that idea. Ron Shank, Tom?

DR. SHANK: I have other concerns. There are several pages in the report on cellular activity. These compounds are capable of stimulating collagen synthesis, angiogenesis and others, and I wonder does that not make them drugs? And if the answer is, no, it does not, could the Council please address that for us? And if it does, should these be reviewed as a cosmetic ingredient? Also I think there is a strong data need because of this that we need reproductive and developmental toxicity data. In Wave 2 we got some information on penetration of these compounds and it shows that they penetrate into the dermis. But then the author concluded it would not therefore to into lymph and blood and I don't quite understand that. If it gets into the dermis I think it would get into the lymph and blood. I have several concerns about the safety not just the grouping.

DR. MARKS: Thank you, Ron. I was going to ask for a preview of where we would be going with these ingredients. You had also endorsed tabling it, but I raise these concerns at this point so that the next time we see these ingredients these issues may be addressed.

DR. SHANK: Yes. It's going to a scientific committee of the Council? If they could consider not only how to group these chemically but also are these drugs as well as cosmetic ingredients?

DR. ANSELL: I think the CIR Support Committee would be more than happy to reply suggesting a more appropriate grouping. We would be happy to address any questions. As it relates to the physiologic activity as it relates to its designation as a drug or cosmetic, I would suggest that would be directed to the FDA liaison as to its regulatory status.

DR. SLAGA: Do these have a type of effect in vivo if the doses are used in cosmetics? I agree with Ron that the cellular effects are quite pronounced in some cases, but what I'm suggesting is does this really occur in vivo as a cosmetic?

DR. SHANK: I could see advantages in cosmetics to stimulate collagen synthesis.

DR. SLAGA: But not angiogenesis.

DR. SHANK: Not angiogenesis, no, but certainly collagen synthesis for removing wrinkles.

DR. BERGFELD: That's very significant. Was there significance in the paper?

DR. SHANK: Yes.

MS. LORENTZ: May I comment that that gets to the whole grouping question too, again the driver not being the palmitoyl.

DR. SHANK: Using the driver is maybe not the right term because it's the palmitoyl that drives the peptide into the skin. Just the peptide itself would be absorbed I would think rather slowly, but when you add a fatty acid to it, that makes penetration much more likely.

DR. HILL: Not just into the skin, but into cells once it gets inside. The other thing I picked up on this was that it also increases the interest in impurities if you have for example a palmitoyl pentapeptide which perhaps in and of itself isn't inactive, you would want to know if there was a tripeptide or tetrapeptide impurity for example based on the ways that peptide synthesis is done. It's challenging to get 100 percent pure peptides. Then you need information about those impurity levels and some biological activity, so that would be another related issue to this in my mind. I was looking for ingredient-by-ingredient information if we were going to go forward with these as they were.

DR. ANSELL: We're not suggesting what the conclusion would be. We're certainly not suggesting ingredient-by-ingredient reviews. But we do think that if we pulled the family apart and reassemble it into smaller groups, perhaps they would lend themselves to building new and larger families but not based solely on the derivation but, rather, a look at both sides.

MS. WEINTRAUB: I also noted the lack of reproductive and developmental toxicity data, also no carcinogenicity data and absorption, distribution, metabolism and excretion wasn't found either. So there seems to be a lot of data needed in addition to other concerns.

DR. MARKS: Tom, were you concerned about carcinogenicity of these compounds?

DR. SLAGA: No.

DR. MARKS: With the lack of data, the reason you weren't, again going forward? Or you thought there was enough in here in answering Rachel's concern?

DR. SLAGA: On carcinogenicity? I didn't have a concern about carcinogenicity. I do believe that we had some genotoxicity data and that was sufficient.

DR. SHANK: But there is a data need for reproductive and developmental.

DR. SLAGA: Yes.

DR. MARKS: That was clear. Rachel was anticipating my next question which was were there any other obvious data needs? Obviously since we're tabling it we're not going to sort through the compounds. That was one of the things I had printed out, the long list of compounds or ingredients.

DR. HELDRETH: I wanted to add that if we're going to look at these in a different grouping fashion based on the protein portion of the molecule, you'll note in the definition that there's a great amount of possible variability of what the amino acids are and what order they're in. So if we're going to base it on that part of the molecule, it would be nice if the support committee could provide us with which actual compounds are designated, say, palmitoyl tetrapeptides because that can be a number of different actual ingredients that if we're looking at the protein function or protein side of it are quite different.

DR. MARKS: Is there any comment about that, Jay?

DR. ANSELL: Yes. There was a change in nomenclature from the parent ingredient to nomenclature of other naming conventions later on in the INCI process that are perhaps better and would certainly try to point out the correct INCI nomenclature.

DR. HILL: One thing I wanted to follow-up on to Dr. Slaga's comment is while there is nothing indicative of carcinogenicity, I don't remember if it's in the first submission or in Wave 2, there is language to suggest that there might be changes in invasiveness of cells depending on the particular peptide so that's something that needs some attention. It's different than tumorigenicity and any of that, but it's changing the invasive character of the cells, then that's important.

DR. SLAGA: That relates to the angiogenesis that Ron has concern about.

DR. MARKS: Tom, going back to Rachel's question, it doesn't change your concern.

DR. SLAGA: No, it doesn't change anything about the genesis.

DR. MARKS: But the question is could it enhance the invasion if were metastatic?

DR. SLAGA: Angiogenesis inhibitors are used a lot in treatment of malignancy.

DR. HILL: Angiogenesis and cell invasion are related, but distinct phenomena as well.

DR. MARKS: We have some significant issues to resolve with these ingredients in terms of its biologic activity and back to originally is this a drug or a cosmetic. Perhaps we could dodge it if we say it's a drug, but I don't think we can do that. We have to address this. Rachel?

MS. WEINTRAUB: I have one question for Tom about genotoxicity that nanofiber gel CS was found to be genotoxic but not without metabolic activation in certain strains.

DR. SLAGA: What you have to do is take the total data for all of genotoxicity and in general it's more predominantly negative. That was the only positive and that happens now and again. I can't argue that that's not true, but generally since the majority are negative, I don't have a concern.

DR. MARKS: Anticipating when this is resubmitted, the different ingredients, was there any concern of several of these ingredients having hydrolyzed collagen? You were talking about the peptide variability. Is there any concern that these have collagen? You, Ron Shank, mentioned right in the beginning that they can increase collagen synthesis, but if you look at sodium palmitoyl hydrolyzed collagen, there is palmitoyl hydrolyzed collagen and then there are these proteins also. If we hadn't looked at this group I was going to say did you want to split any of these things out because of the difference there. But what's your sense with those? Did that raise any concerns or not, different than the peptide?

DR. SHANK: No concerns other than the other report which is on hydrolyzed amino acids and proteins.

DR. MARKS: Presumably tomorrow I'll be seconding a motion that these ingredients be tabled and I'll raise the concerns that were raised here, the need for repro and developmental toxicity. And I'll raise the thorny question whether this is a drug or a cosmetic or at least mention it because of the activity of increased collagen synthesis, the angiogenesis, and obviously the grouping is the main driver.

DR. BERGFELD: I want to add a comment that in many cosmetic products that have antioxidants in them there is increased collagen being formed and there is a biological activity of the epidermis and the upper portions of the dermis and that is still considered a cosmetic. It's dependent on concentration of the antioxidant and there are some that are released only to physicians which are higher concentrations and to my knowledge there is no prescription item in the antioxidant group.

DR. HILL: Are you talking about something like kojic acid?

DR. BERGFELD: We have a lot of antioxidants that fall into the fruit acid groups and the retinoids in a whole line of cosmetics now, but they all have biological activity and we can even see it on histology as well as clinical improvement of skin texture and wrinkles.

DR. HILL: I brought up sunscreens at the last meeting in a moment of lapse, but I had read just a few weeks before that the very extended version of what's a drug, a medical device and a cosmetic and there are clearly gray areas and maybe this is a case where we need some further clarification, and it may not be forthcoming anytime soon I guess.

DR. MARKS: I'll mention it tomorrow, but as Wilma you've said, we have other ingredients that have had biologic effects like alteration of the immune system and perhaps one of them that we're discussing today can have an impact. It should be raised and then we'll see where we go with it as we review the ingredient.

DR. BERGFELD: Ron, as to localized effects versus a systemic effect, would that make a difference for you between cosmetic and drug?

DR. SHANK: Certainly if it's systemic, yes. As to local, I'm not so much concerned.

DR. BERGFELD: I think these are mainly local.

DR. ANSELL: At very low-use concentrations with some products as well at ten-thousandth of a percent.

DR. MARKS: Are there any other comments about these palmitoyl oligopeptides?

DR. JOHNSON: I have one question, Dr. Marks, with respect to the Wave 2 data. Is it agreeable that all of those data will be incorporated into the safety assessment?

DR. MARKS: I guess if it's applicable to the ingredients in this new presentation. We'll see in the next draft report. Is there anything else about these ingredients?

DR. HILL: Wilbur, I assume you would like whatever commentary, thoughts or feedback on that, you would like that along with the actual report.

DR. JOHNSON: Yes.

DR. HILL: It will be forthcoming. I just wanted to be sure.

DR. MARKS: Thanks for bringing that, Ron. For the writers, is a better way as we work through going paperless to have a flash drive and at the end of the meeting tomorrow give you the flash drive with our changes? Some of us will have it in paper. Some of us will be on either a Word document or PDF. Which is the best way to give those back to you? In the past we got the books, and I assume what you did, Wilbur, was go page by page.

DR. JOHNSON: Yes.

DR. MARKS: That hasn't changed. We'll go page by page. It's just going to be on a screen. Lillian?

DR. GILL: I think we suggested in our memorandum to you that we would like to have the flash drive back and we can get it around to each of the writers and get your comments. Certainly you've had some team members that sent them by email. If that's better for you and you want to spend some more time, that's good as well. Either way, Jim, works for us as long as the reviewers or the writers get the comments.

DR. HILL: One thought I had on that subject was a full version of that group, that would allow lifting the pages which I don't have on this, but it would allow lifting out the pages from for example Wave 2 on the pertinent ingredient and you could tack them onto the end of the others. Maybe in the future we should aim for something along those lines.

DR. MARKS: I think we need to continue as we work though this certainly for the next few meetings as to which way to handle the data electronically and how that works and it may work one for one individual and a little differently for the other. I think sharing the way we do it is important. Jay just told me to download it from the flash drive and put it on my hard drive and showed me how to do that so that there is going to be a learning curve with varying steepness depending on the individual.

DR. ANSELL: May I also put in a request for the naming convention to use a little more of the title? I found it very difficult to find which file aligns with which report because you're only picking the first four letters. They're hard to find.

DR. BERGFELD: The memo that I reviewed said that we were to go in and rename it. I thought the responsibility was transferred to us to go in and fill in the whole name with our initials after it.

DR. GILL: I think the example allowed for renaming at the end with just the initials, but if you name it something and we can find it, Wilma, that will be fine.

DR. BERGFELD: Whatever we're supposed to do, I'd like to know that.

DR. GILL: The most critical issue was putting your initials so we would know from the comment.

DR. HILL: The only difficulty I had with that is the places where the alphabetization varied from the ingredient name and sometimes those change over time anyway, but when it's substantially different, so I marked up the agenda with what the names were so that I could quickly find them. That's a small, small thing in my opinion.

DR. BERGFELD: I would like to make another comment here. I think what was in the Buff Book before used to be in our transmission on this because we can go back and we can figure out the order of events and order up our ingredients in that way. With not having an agenda, I found that it was hard because it was free-flowing whatever compound came up.

DR. SHANKS: Wasn't that the admin Buff?

DR. BERGFELD: I didn't see it.

DR. HILL: It's there.

DR. SHANK: It's in the PDF for, but I don't think it's in the Word book.

DR. BERGFELD: I didn't see it.

DR. SHANK: The PDF files were much more complete than the Word.

DR. HILL: The first thing I did was go in and print the first 10 pages of the Buff Book that had the order of ingredients. If you were in the MS Word, you didn't see it. When I got it on the flash drive the name had changed from what was on the original website.

DR. MARKS: Obviously the data needs to be sent consistently. I gather there is among our members here a difference. You used Word and PDF, Ron?

DR. SHANK: Right. I appreciated having both forms available. I can do it in Word much faster than I can do it in PDF. I have Acrobat, but for me it's just faster in Word.

DR. MARKS: Do you use track change?

DR. SHANK: Yes, I do.

DR. MARKS: So that you can understand, Wilma, now why you were asking about the admin Buff because it is on the PDF. I guess that won't happen again, that if that were sent in Word there won't be inconsistency.

DR. ANSELL: If you use a tablet, you can't use the flash drive, you have to download it from the CIR site.

DR. HILL: The way you do that is you take the flash drive files, you put them on the computer, you upload them to Box.net and then you download them into the tablet. It's not hard, but it includes additional steps. It works slickly.

DR. ANSELL: I would suggest we recommend to staff that they pick a file name and stick to it because my file name is different that I got from the CIR from the file name for those people who use the flash drive to upload their documents.

DR. MARKS: I see this as part of growing pains, and when you make changes there are always going to be difficulties. It's going to be interesting to see how it works out over the next three or four meetings. We've obviously committed to that. I haven't heard anybody say I can't do this. I'm quitting. So that's good. Again I'll reiterate that it's a good thing we didn't have 2 hours of lectures this morning because we'll make up the difference in working through how to do this electronically. I appreciate everybody's willingness to share and in my case reveal my ignorance.

Next is 6-hydroxyindole. How many like using the down button, the little scroller or just dragging? My finger gets tired if I keep rolling the little whatever this thing is called.

Full Team – March 19, 2013

DR. BERGFELD: Thank you, unanimous. Okay, the next ingredient is Dr. Snyder, Palmitoyl Oligopeptides.

DR. SNYDER: Yes, the Palmitoyl Oligopeptide is a -- the document, the first time we've seen this, the document comprises of 45 ingredients based upon a scientific literature research that was conducted in August of 2014. We had quite a lengthy discussion about this ingredient, also, and our team came to the conclusion that we wanted to make a motion to table this ingredient to identify the correct groupings and have the groupings be based upon the peptide, not based upon what the peptide is bound to. We thought that would be a better way to look at these ingredients and maybe to bring other ingredients into the mix. And, so, we would make a motion to table this ingredient.

DR. MARKS: Second.

DR. BERGFELD: Motion's been made and a second. There's no comment on that. All those in favor at the table?

DR. MARKS: Well --

DR. BERGFELD: Well, you can comment afterwards.

(Hands raised)

DR. BERGFELD: Okay, it's approved. To table and comments now.

DR. MARKS: Yes, our team discussed some needs. Even though we tabled it, we wanted to alert interested parties that we were concerned about reproductive and developmental toxicity and we needed data on that. And then we actually had a fairly robust discussion about the potential drug effects of these particularly increased angiogenesis from these compounds. So, again, we wanted to delve into the cellular activity of these compounds and get some more --

DR. SLAGA: No, the cellular effects and cell culture, that's where this data came from being stimulating angiogenesis and really the critical thing is does this occur in vivo?

DR. BERGFELD: Ron Shank?

DR. SHANK: Well, and also it has many biological activities and I questioned whether these were actually drugs and should be reviewed as cosmetic ingredients at all. I think when there is a discussion, that has to be handled somehow. And that's why we felt a need for reproductive and developmental toxicity data.

DR. BERGFELD: Paul?

DR. SNYDER: We would agree with that assessment.

DR. BERGFELD: All right, any other discussion points that need to be put on the table for the minutes?

Dan?

DR. LIEBLER: One other point was just some clarification of the actual chemical composition of whichever of the Palmitoyl Oligopeptides are advanced for a consideration in the future. Most of these appear to be made by solid phased peptide synthesis, which should produce defined sequences, mixtures are pretty high purity, but the table one indicates that they're semi-random mixtures.

So, I mean, I suppose you could do a pooled approach to solid phase synthesis where you use a pool of amino acids for the first cycle, a pool for the second cycle. I'm not sure what's done, so, that needs to be clarified and the naming conventions, well, there isn't any, but it is completely unsatisfactory for these, so, it's really hard to tell what we're reviewing.

So, whether it's industry or whether it's the CIR staff or some interaction between those to come up with information to tell us exactly what it is that we're looking at. It's important and it's particularly going to be important if we're considering any biological activities of these compounds because if they are specific to sequences, then, obviously, we need to know the sequences involved.

DR. BERGFELD: Ron Hill?

DR. HILL: Yes, in follow-up to that, it's very well-known that when you do peptide synthesis of any kind, be it solid phase or liquid phase, certain steps are more problematic and sometimes they don't go to 100 completion. So, then what you have a fraction, however small, depending on how the analytical chemistry is done where there's a missing amino acid, and, so, then if it does on to the next step, suddenly, you've got a sequence that's different, it's shorter, and sometimes that can be 10 percent of the mixture and then that's typically handled at the end with purification.

So, that's the kind of information that would be needed particularly when we're looking at biological activity and I was trying to do a search here because they tried to use -- I didn't know whether this came from staff or from industry or who, but they tried to use DEREK to justify lack of hits and I think that's an entirely inappropriate use of that program in this particular case, and, so, I wouldn't really buy that without a lot more detail.

DR. SNYDER: I'll throw one more piece on the pile here.

DR. BERGFELD: Okay.

DR. SNYDER: The only acceptable method for characterizing compounds like this these days is mass spectrometry.

DR. BERGFELD: Thank you. All right, we'll move ahead then. This has been tabled.

Dr. Marks presenting on tromethamine.

Meeting Summary

Palmitoyl oligopeptides

The report was tabled pending reorganization of this document. These ingredients were preliminarily grouped together, as they are related structurally by an identical fatty, hydrophobic tail connected to a variable sequence of peptides.

The Panel noted, however, that the terminology used for these ingredients does not enable adequate evaluation.

Further information is sought to better understand the extent and manner in which solid-phase peptide synthesis is used to create the peptide portion of such fatty acid peptide ingredients. If additional information enables a better understanding of the amino acid sequences of the peptides of these ingredients than afforded by their definitions in the dictionary, then grouping them together in some fashion may be reasonable.

If there is a substantial degree of randomness associated with the peptides of these ingredients, then it would be important for the Panel to consider how that might influence the safety evaluation. For example, some small peptides are potent stimulators of angiogenesis. The potential for such an activity to promote tumor growth and metastasis in people with undiagnosed skin cancer might then be an issue. Given the present uncertainties, grouping a large number of these ingredients together might be inappropriate.

At the time the report was tabled, the following ingredients were included:

palmitoyl oligopeptide
palmitoyl dipeptide-7
palmitoyl dipeptide-10
palmitoyl dipeptide-13
palmitoyl dipeptide-17
palmitoyl dipeptide-18
palmitoyl tripeptide-1
palmitoyl tripeptide-4

palmitoyl tripeptide-5
palmitoyl tripeptide-8
palmitoyl tripeptide-28
palmitoyl tripeptide-29
palmitoyl tripeptide-31
palmitoyl tripeptide-36
palmitoyl tripeptide-37
palmitoyl tripeptide-38
palmitoyl tripeptide-40
palmitoyl tripeptide-42
palmitoyl tetrapeptide-7
palmitoyl tetrapeptide-10
palmitoyl tetrapeptide-20
palmitoyl pentapeptide-4
palmitoyl pentapeptide-5
palmitoyl hexapeptide-12
palmitoyl hexapeptide-14
palmitoyl hexapeptide-15
palmitoyl hexapeptide-19
palmitoyl hexapeptide-26
palmitoyl hexapeptide-32
palmitoyl hexapeptide-36
palmitoyl hexapeptide-27 acetate
palmitoyl heptapeptide-5
palmitoyl nonapeptide-6
palmitoyl decapeptide-21
palmitoyl oligopeptide-70
palmitoyl hydrolyzed collagen
palmitoyl hydrolyzed milk protein
palmitoyl hydrolyzed wheat protein
potassium palmitoyl hydrolyzed corn protein
potassium palmitoyl hydrolyzed oat protein
potassium palmitoyl hydrolyzed rice protein
potassium palmitoyl hydrolyzed sweet almond protein
potassium palmitoyl hydrolyzed wheat protein
sodium palmitoyl hydrolyzed collagen
sodium palmitoyl hydrolyzed wheat protein

MARCH 2014 PANEL MEETING – SECOND REVIEW/DRAFT TENTATIVE REPORT

Belsito Team – March 17, 2014

DR. SNYDER: You have palmitoyl oligopeptides?

DR. BELSITO: No, tripeptides.

MR. JOHNSON: Well, they have the whole name, palmitoyl oligopeptides. So that's the same report.

DR. BELSITO: Okay.

DR. KLAASSEN: You have to look under the palmitoyl.

MR. JOHNSON: Right.

DR. BELSITO: Okay, so we got these very convincing arguments from Lintner in Wave 2.

DR. EISENMANN: You know Dr. Lintner is here?

DR. BELSITO: Okay. Very convincing, Dr. Lintner, thank you.

DR. LINTNER: Thank you. I'm open to your questions. I shared some in the other room this morning, so I'll be happy to answer any further questions you have.

DR. BELSITO: Page 21; that was just my general comments. So we need to discuss with teams. Given the 1 percent report -- I'm not sure what that's --

DR. EISENMANN: Now, the 1 percent is definitely wrong.

DR. BELSITO: Okay.

DR. EISENMANN: I haven't provided new use data because I still have 0.5 and 0.25 percent, and those are probably also wrong because those are probably -- they're providing the concentration of the mixture rather than the ingredient itself.

DR. BELSITO: Okay. So assuming those are wrong and assuming -- you know, we were told the absorption with similar peptide was 3 percent. But even if you assume 100 percent, it's still within a safe limit --

DR. LINTNER: Absolutely.

DR. BELSITO: -- from your calculations. Then when I said well, 100 ppm seems to have some irritation and that would be an issue at 1 percent. But if 1 percent goes away, that's not an issue in terms of the irritation. So then if the 1 percent goes away and the 0.25 goes away, then I think we can go with a safe as used in those very low limits with the calculation even assuming 100 percent absorption.

DR. LINTNER: I can guarantee you that nobody uses these specific peptides any higher than 10 ppm, 0.0001 percent in a cosmetic cream. There is no reason to do it, and it's much too expensive. So the use levels of 0.1, 0.25, and 1 percent are absolutely wrong. They're based on, as Carol said, on the use level of the 100 ppm commercial solution, but not the true peptide content. Most people use it at 3, 5, maximum 10 ppm of peptide. The rest is just water glycerin and butylene glycol.

MR. JOHNSON: And that's for any peptide.

DR. LINTNER: I can only speak about the Sederma products, the ones under discussion here -- tripeptide-1, palmitoyl, hexapeptide-12. I don't speak for competitors' products. Most of them, indeed, are similarly formulated, but I don't have any data on those.

MR. JOHNSON: Well, let me ask you a question. In terms of --

DR. BELSITO: When you say similarly formulated, this is where we get back to my being upset with the INCI names and not being specific to a given chemical again. My assumption is that when we're saying tripeptide-1 and hexapeptide-12, we are now talking about a very specific amino acid sequence.

DR. LINTNER: Correct.

DR. BELSITO: And regardless of whether it's your specific amino acid sequence or another company's specific amino acid sequence, it would be the same, no?

DR. LINTNER: Well, well, unfortunately, that is more complicated historically. Twenty years ago our company introduced the first synthetic palmitoyl peptide to the cosmetic industry. We got the INCI name, CTFA name then, of palymitoyl oligopeptide. Why? I don't know. Strange, but that's the way it is. We supplied it. A couple of weeks later the second peptide, totally different chemical structure, it got the same INCI name, palmitoyl oligopeptide. Recently, this error was

corrected and the INCI Nomenclature Committee now gave the name tripeptide-1, palmitoyl tripeptide-1, to the tripeptide with glycine- histidine-lysine sequence, and palmitoyl and hexapeptide-12 to hexapeptide with six amino acids of a different sequence. That should have resolved the issue, to have one INCI name for one specific sequence. From what I've heard this morning in the other group, somebody -- I don't know, I've never heard of this and never run across it before -- some company seems to supply or propose a hexapeptide, palmitoyl hexapeptide, with the same amino acid composition with a different sequence. Now, this is, of course, a totally different chemical entity with totally different possible biological activity and/or, if ever, toxicity. But apparently -- Wilbur may correct or confirm -- this other competitor's product is also named palmitoyl hexapeptide-1, which just shouldn't be.

The Sederma Company also proposes and sells a palmitoyl tripeptide, but with a different number, which is composed of the same amino acids as the tripeptide-1, but again with a different sequence. Instead of glycine- histidine-lysine, it's glycine-lysine-histidine; two peptides in cell culture, but very different biological activities. So it is really up to the INCI Nomenclature Committee to clean up their act, if I may say so.

DR. BELSITO: Well, I agree, but we have a way around that. We can say that tripeptide-1 is defined as the trimer specifically composed of this sequence and that is safe as used, and the hexapeptide-12 is the hexapeptide specifically composed of this sequence, and any other resequencing of these would be insufficient.

DR. LINTNER: Correct. Yes, I agree with that.

DR. BELSITO: I mean that's easy for us to deal with.

DR. LINTNER: Good.

MR. JOHNSON: One question. In terms of the maximum use concentration, are we going to rely on industry or the Council for that information, or are we going to rely on Dr. Lintner's information?

DR. BELSITO: Well, I think you know, again -- I mean I didn't realize you were in the audience when I complimented you, but I'm glad you were here.

DR. LINTNER: Thank you for having me.

DR. BELSITO: I think that looking at what companies are trying to do with these products, looking at the activity -- I mean when I was looking at those very high levels, I reviewed this before we got Wave 2. On page 36 of the document, I said "not liking these and do we not really know what we're reviewing?" As I started seeing all of these rather potent biological effects on angiogenesis, et cetera, et cetera, and then getting your letter and putting this into perspective and the cost of manufacturing them and their solubility and the impossibility of getting them to be used at those concentrations, it sort of made sense. Companies are using these -- it's marketing tools.

DR. LINTNER: Of course.

DR. BELSITO: And they're using them because they have biological effects and they may or may not have some trivial biological effects in their cosmetic agents, but people are wanting to suck up and pay $100 for something that promises them a face lift without visiting a plastic surgeon, right?

DR. LIEBLER: But what to do about the concentration?

DR. EISENMANN: I plan to go to go back to the companies -- I've gone back to one -- and provide them with Dr. Lintner's information. They're likely to change by that simple fact and say oh, we made a mistake.

DR. BELSITO: Well, I think what we can do with it is go with Dr. Lintner's information about concentration and say that it is the assumption of the CIR -- we're making several assumptions here on the safety of these; that the ingredients are not used above the level that Dr. Lintner said and that what we call the tripeptide is this specific sequence. It's not any rearrangement of those various amino acids, and what we call the hexapeptide is this very sequence. It's not any rearrangement of those sequences. And anyone who -- and even if INCI calls it the same thing, if anyone wants to use a hexapeptide that falls under that nomenclature, but is a different sequence, the data is insufficient. And we can even put that in our conclusion. "The data are sufficient for the tripeptide of this specific sequence, but insufficient for other tripeptides that may be found under that INCI name. And it's sufficient for this sequence, but insufficient for any other hexapeptides that may fall under that INCI name." And I think we've covered our bases.

DR. LIEBLER: Well, I agree with all of that, and I also appreciate Dr. Lintner's comments. And I just think that we ought to be able to certify that industry has supplied us with the correct numbers, even if we have to prompt them with Dr. Lintner's information to tell them to check their calculations.

DR. SNYDER: So what would the title of this document be because it won't be linked to an INCI ingredient?

DR. BELSITO: It has to be linked to an INCI name, and I think that again it just points out that the dictionary has to get on mark and start giving specific names to chemicals that they group and are very different. It's very annoying.

DR. BRESLAWEC: Again, this was an attempt to do so. Obviously, some tweaks are needed in the system and Joanne Nikitakis, who is the new editor of the INCI Dictionary, will be here tomorrow and prepared to discuss this and answer any questions. That's certainly our intent.

DR. SNYDER: So, again, what are we going to title it?

DR. BELSITO: We're going to title it -- we can even put an asterisk to the title, or we can put in the title, which is going to be too long for the journal, a specific sequence that we're defining as the tripeptide and the hexapeptide.

DR. SNYDER: Excellent.

DR. LIEBLER: I think we could keep the title and add in the conclusion the clarification as to what specifically we're reviewing.

DR. BELSITO: And put it in the introduction that "there may be different hexapeptides and tripeptides with different sequences that would fall under this same INCI name; however, this review is strictly being limited to these specific sequences."

DR. SNYDER: But my only question is if you go to INCI, you're not going to find tripeptide-1, hexapeptide-12.

DR. BRESLAWEC: Yes, you will.

DR. BELSITO: Yes, they're existing INCI names, but the tripeptide may have a different sequence.

DR. SNYDER: Oh, okay. I'm sorry. I thought they were all under --

DR. EISENMANN: Actually, that name's been retired so they're working on transitioning everything over to the other name.

DR. SNYDER: Okay, I understand.

MR. STEINBERG: Yes, it's on Social Security now.

DR. BRESLAWEC: But you can't take a name and just throw it out of the dictionary because of legal and time to change the labeling in China and a lot of other things.

DR. LINTNER: If I may say, having read your preliminary draft for this meeting, it's a long list of many, many other acetyl and palmitoyl and biotinoyl and whatever peptides you have there. You're going to have a lot of work in investigating each and every one because of different chemical entities or just different substances. You can't group peptides together like you can maybe other ingredients. These peptides are each very different with their own data.

DR. LIEBLER: Well, it does raise the issue of whether those other peptide derivatives belong in this report or not.

DR. BELSITO: I think that that's --

DR. EISENMANN: Especially the ones where you haven't reviewed the other portion?

DR. LIEBLER: Right.

DR. HELDRETH: The ones that were recommended to us.

DR. SNYDER: So in this document, in the introduction, we'll have to have something that we really haven't had in specifying specifically -- so currently we don't -- I think we need to --

DR. BELSITO: Well, we're dealing with these small molecules that have biological activity and when you rearrange the amino acid sequences, you get very different activity. So that's the problem. The INCI name may not specify the sequence. So I mean I think that we are where we were at when we first started struggling with botanicals and decided to look at composition as a way of dealing with it. I think with these small peptide molecules in cosmetics, we're going to have to deal with specific sequences. And if there are several different chemicals that have the same INCI name with different sequences, we're going to have to create different reports on each of those chemicals. We're going to have to say that tripeptide-1 defined as this sequence is safe as used up to this concentration. Tripeptide-1 as defined by this different sequence is insufficient, is unsafe. I mean the conclusions may be very different.

DR. LINTNER: I think it's somewhat simpler. I think you have this situation only for the palmitoyl hexapeptide-12, which for historical reasons was only for peptide like the other tripeptides. I think today anybody who supplies or who asks for an INCI name for a hexapeptide, even if it's a palmitoyl hexapeptide, will have a new number, a chronological number. So this ambiguity in my understanding exists only on the term hexapeptide-12 because under the palmitoyl oligopeptide name, there was a tripeptide and we've taken care of that. But there were two hexapeptides, one supplied by Sederma with the leave-on, and somebody else's -- I don't know whose -- hexapeptide. They also swam in the pool under oligopeptides probably to get around the patent by Sederma and have a different sequence, but also was called oligo. And now with the change from oligo to hexapeptide-12, apparently also is called hexapeptide-12. But I don't think that you will have other instances where two

different sequences will have the same INCI name. So it shouldn't be too difficult in the future. But in your report, if you make sure that you declare the leave-on hexapeptide as safe and the other as insufficient, then that's fine.

DR. BRESLAWEC: I actually think it will be other such cases --

DR. LINTNER: Really?

DR. BRESLAWEC: Yeah. Yeah.

DR. LINTNER: Well that's a shame.

DR. BRESLAWEC: They have not been naming -- INCI has not been providing some of the names for another -- other peptide ingredients.

DR. LINTNER: Right.

DR. BRESLAWEC: So, in other categories there will be a similar sort of an issue.

DR. LINTNER: Absolutely, yeah. You should use the IUPAC Nomenclature after all, that's a group we have. Any chemist knows IUPAC, I don't know -- sorry.

DR. HELDRETH: Call it legal labeling.

DR. LINTNER: But anyway --

DR. BELSITO: Then the only other question that I had for this, looking at even -- with defining the tripeptide specific sequence is this Ursoloyl Tripeptide? Do you have any concern with that ursoloyl part of it? I mean, it looks so different from all the other animals that we looked at here?

DR. LIEBLER: I'm looking again.

DR. BELSITO: It's on page 45 of the document.

DR. LIEBLER: Ursoloyl? I guess I have the general peptide -- a general question. Are these peptides -- are we looking at the peptides or the other piece? I mean, we've got all these -- we even have it in the thioctoyl, the retinoyl ones, the quinoyl ones --

DR. BELSITO: It's my understanding that it's the -- I mean it's really the tripeptide sequence that is the driver here. Is it not? And then we are only concerned about what it's attached to, if we think that there could issues with it? But maybe I'm wrong.

DR. LIEBLER: Some of the -- some of the modifying groups could have at least as much biology as the peptides, so.

DR. BELSITO: So, then do we want to look at only the peptides with straight chains, and then look at the ones that are -- have separate chains. I mean, how do we want to split it? Because right now we are looking at -- so we are at the point where we said okay, the tripeptides with this specific amino acid sequence are fine. But now we have a whole list of tripeptides that may have that specific amino acid sequence. Are we saying those are all fine? I guess that's my question. When I looked at them, I just pulled out that last one that looked beastly as compared to the others.

DR. LIEBLER: Right. But you could make the same argument about the retinoyl one, for example.

DR. BELSITO: Okay. So then we need to -- we need --

DR. KLASSEN: I think that these -- these molecules that likely have very specific biological actions that we don't know of, we really can't put them together like -- you know, on Table 1 we've got a, you know, big steroid in here, and when that split off, what's that going to do? I mean, I think, you know we -- so I have great difficulty (inaudible) -- at least some of them, and you know, same with the retinol. I mean, you know, retinoic acid and its derivatives have tremendous biological effects just like, you know, these other more classical steroids. I think we have to be very careful here.

DR. BELSITO: So we are saying that -- we need to go back and look at our conclusion because we are going to say, safe as used with specific amino acid sequences within the limits that Dr. Lintner had told us these were being used at, and now I'm hearing from my experts that not all these tripeptides necessarily belong in this group. So are we prepared to carve these out? Or, do we want to say, insufficient, and we want industry to clarify the concentrations of use. And we want time to relook at the moieties that are attached to these tripeptides, to decide whether this is a correct grouping of the tri and hexapeptides, because I'm not the person to do this. This is not my area of expertise.

DR. SNYDER: We are talking about the single-digit parts per million for this range?

DR. BELSITO: Yeah.

SPEAKER: Right.

DR. SNYDER: I mean I don't think that's going to be (inaudible)

DR. BELSITO: I mean, that's fine, I'm just clarifying that --

DR. KLASSEN: But that just relies on argument that, well, nothing at that level could have any safety concerns. I understand the practical reaction to it, Paul, but I don't think it's a good rationale for us to be using. So, I'm trying to come up with --

DR. BELSITO: So you're prepared to slice and dice today? Or do you want to table it for a -- you know, let industry know, this is where we are going, we want concentration of use, because it doesn't seem to be what -- the reported usage seems exorbitantly high, from what we heard from industry experts. And in the meantime, give you and Curt, and other people who understand the chemistry, time to look through the list and get rid of ones that don't belong in this grouping, and create a new name of linear palmitoyl tripeptide ones?

DR. LIEBLER: If you were to ask me today, I would delete all the modified peptides with the exception of the fatty acyl derivatives, the palmitoyls.

DR. BELSITO: What's driving this report? What is the ingredient driving it?

DR. HELDRETH: It was originally palmitoyl and oligopeptide, which is now being retired so essentially, palmitoyl -- tripeptide-1, palmitoyl and hexapeptide-12, would be the driving wheel.

DR. BELSITO: Okay. So then why don't we -- so what's that family, what are we going to call that family?

DR. HELDRETH: The rest of the ingredients that the group is preparing were suggested by industry --

DR. LIEBLER: Yeah. Right. It doesn't look like there's any uses for almost all of them.

SPEAKER: Yeah. In that case it does --

DR. LIEBLER: Biotinoyl has a handful, and other than that, we've got the copper complex which I don't have as much problem with, and the palmitoyl, those are the only ones that really seem to have any uses.

DR. BELSITO: So, we'll call it palmitoyl and copper tripeptide-1?

DR. LIEBLER: I think we could call it oligopeptide -- or the tripeptide-1, and hexapeptide-12 --

DR. BELSITO: Hexapeptide-12 --

DR. LIEBLER: -- and relate it to amides, but just delete most of those other compounds.

DR. BELSITO: And related to amides. And the others aren't amides? They are, and that's --

DR. LIEBLER: They are. Yeah.

DR. BELSITO: So we can't call it --unless you can prove it.

DR. LIEBLER: But the copper complex isn't in amide.

DR. BELSITO: Then, so why don't we just --

DR. LIEBLER: But the others are all -- actually they are all attached as amides I believe.

DR. BELSITO: Right. So we can --

DR. LINTNER: No. No. No. No.

DR. LIEBLER: I'm scrolling to it.

DR. LINTNER: These are nothing but -- these are carboxyl groups --

SPEAKER: Carboxamide?

DR. LINTNER: No.

DR. HELDRETH: They are attached to the --

DR. LINTNER: COOH, they are acid --

DR. LIEBLER: They are attached to the N-terminus? That's what I was referring to.

DR. LINTNER: The palmitoyl is attached to N- terminal. Yeah.

DR. LIEBLER: Oh. The peptide -- the retinoyl is attached to the N-terminus of the peptide.

DR. LINTNER: Oh. Okay. Sorry.

DR. LIEBLER: That's what I was referring to. The quinoyl is attached to the N-terminus of the peptide, all of these are attached to the N-terminus?

DR. LINTNER: Right.

DR. BELSITO: Well, if we use a name that would include all these ingredients for excluding that doesn't make sense to me. If all you want to review is copper and palmitoyl tripeptide, then I would say, safety assessment of copper tripeptide-1, palmitoyl tripeptide-1, and then what hexapeptides do we want to review?

DR. HELDRETH: Just number 12.

DR. BELSITO: Just hexapeptide-12. So it is --

DR. HELDRETH: That's the essential (inaudible) --

DR. BELSITO: Safety assessment of copper tripeptide-1, palmitoyl tripeptide-1 and hexapeptide-12 is used in cosmetics.

DR. HELDRETH: That's just myristoyl hexapeptide- 12.

DR. BELSITO: That's fine. So then how do we --

DR. LIEBLER: And their fatty acyl amides.

DR. BELSITO: Okay. So, safety assessment of copper tripeptide-1, fatty acyl amide tripeptide-1 derivatives, related ingredients, and hexapeptide-12?

DR. LIEBLER: Safety assessment of tripeptide-1, hexapeptide-1, copper tripeptide-1, and their fatty acyl derivatives.

DR. BELSITO: Repeat that again. Relabel as?

DR. LIEBLER: Tripeptide-1, hexapeptide-12, copper tripeptide-1, and their fatty acyl derivatives.

DR. HELDRETH: Is the manganese tripeptide-1 an issue?

DR. KLASSEN: Tripeptide-1, hexapeptide-12, their metal salts and fatty acyl derivatives. How's that?

DR. BELSITO: Bart, how's that? Are we capturing everything?

DR. HELDRETH: Except they find another one.

DR. LIEBLER: And not the weird stuff.

DR. HELDRETH: As you think of -- No.

DR. BELSITO: Okay. So, it's going to be relabeled as tripeptide-1, hexapeptide-12, and their metal salts and fatty acid -- fatty acyl derivatives as used in cosmetics.

(Recess)

DR. SNYDER: So how much of Table 2 will go away?

DR. BELSITO: A lot.

DR. SNYDER: Yeah. And how much -- I mean --

DR. LIEBLER: Of Table 2 will go away? On Table 2 the only thing that gets deleted from Table 2 is biotinoyl tripeptide, Table 1.

DR. SNYDER: Well we are not -- we are referring to it as palmitoyl oligopeptide, GHK palmitoyl oligopeptide, VGVAPG levered.

MS. EISENMANN: You see, the problem is treasury survey was done before they decided to retire the name.

DR. SNYDER: So we have -- anyway, that has to be redone.

MS. EISENMANN: I have suggested that we will combine the GHK, and tripeptide-1 did in the palmitoyl VG, the other -- those two together, so you only have one person, but there's still a number of companies who could not tell me which sequence they were using under the name coumaroyl oligopeptide at that point.

DR. BELSITO: Okay. Well we are going to clarify it. We are going to clarify the sequence they can use. So, we don't really care. We are just going to clarify -- we want to go out to them Carol, and we don't want to say, "What concentration are you using in palmitoyl tripeptide- 1?" We want to say, "What concentration are you using palmitoyl tripeptide-1 as defined by this specific sequence in tripeptide?" And you need to let us know -- you need to know what sequence of tripeptide you're using, and if you are using another sequence of tripeptide, then your use is zero, and your concentration is zero.

Because basically what we are going to say is any other sequence is insufficient till we have biological data on that sequence.

MS. EISENMANN: So, was not planning on doing another complete concentration of ursoloyl, it was just planning on --

DR. BELSITO: No. just take the people who told you they are using it at --

MS. EISENMANN: High concentration.

DR. BELSITO: -- high concentration.

MS. EISENMANN: Right.

DR. BELSITO: You know, you don't need to reinvent the wheel. All the concentrations that are below what Dr. Lintner has told us are feasible to use. Don't go back and ask them.

MS. EISENMANN: Right.

DR. BELSITO: And, if they come back and say they are using it at that level, then our go-around to come to the safe as use concentration is to set the limits that Dr. Lintner told us are reasonably used in cosmetic products.

DR. LINTNER: If they use it at this level, I'm going to go after them for a patent infringement, because they can't do that. Sorry.

DR. BELSITO: Right -- well, I mean, no. If they use it at that level just as the reported levels for the cell tones, FDA should be going after them.

DR. LINTNER: Right.

DR. BELSITO: Right? And they can write all the nasty letters they want to us, in the end if they don't agree with our opinions, then let them formulate their own opinions because we are not doing it; and if they don't want to formulate their own opinions, take our opinions and enforce it, which they are also not doing, so.

MR. JOHNSON: Dr. Belsito, I'd like to call the Panel's attention to page 24.

DR. BELSITO: Yes.

MR. JOHNSON: Apparently on one of the trade names under which palmitoyl, oligopeptide is being marketed in Matrixyl 3000, and the Matrixyl 3000 consists of palmitoyl tripeptide-1 and palmitoyl tetrapeptide-7, and along with the -- and industry safety assessment are on Matrixyl 3000. Now, the amino acid sequence associated with the tetrapeptide-7, is included in the safety assessment, and we have a reference for that, but that sequence is not included in the dictionary. So, we actually have another sequence --

DR. BELSITO: Okay.

MR. JOHNSON: -- in the safety assessment.

DR. BELSITO: For tripeptide-1?

DR. SNYDER: No, for --

MR. JOHNSON: No. For palmitoyl tetrapeptide-7.

DR. SNYDER: Tetrapeptide-7 -- tripeptide, the new ones.

DR. BELSITO: But that's not in the safety assessment, we are just looking at tripeptide-1 --

DR. SNYDER: But that's he's saying, some of the data that is using that, a lot of the data we've got is on the Matrixyl 3000, which is containment --

DR. BELSITO: So they want to create tetrapeptide- in this report?

DR. LINTNER: That's right. I just sent by email to Dr. Gill, the safety information on this tetrapeptide -- on computer.

DR. BELSITO: Seven?

DR. LINTNER: So it's HRIPT, skin irritation, eye irritation, and Ames test.

DR. BELSITO: Okay. So then, I guess we are now changing what we are doing, and we are tabling it to include the tetrapeptide-7 --

DR. LINTNER: Palmitoyl and tetrapeptide-7.

DR. BELSITO: Palmitoyl -- No. It will be whatever the title --

SPEAKER: --

DR. BELSITO: -- we just created, metal, salts and fatty acyl derivatives of tripeptide-1, hexapeptide-6, and tetrapeptide-7.

DR. LINTNER: Right.

DR. BELSITO: And those peptides will be defined by your Company's specific sequences, and the data would be insufficient for biological activity on any other combination of those amino acid sequences.

DR. LINTNER: Right.

DR. BELSITO: So, I guess that's where we are at now.

MS. EISENMANN: Correct. So these tabled -- I think because --

DR. BELSITO: That's what I just said, we are changing what we are saying.

MS. EISENMANN: Because I --

DR. BELSITO: We are tabling it to include tetrapeptide-7 as defined by this specific amino acid sequence. The assumption is, we will get enough data from industry to support the safety, because we'll get the sensitization, irritation data. We already have the biological activity data. We'll clarify concentrations of use for that.

MS. EISENMANN: To actually do it while new --

DR. BELSITO: Yeah. I understand. I understand. But, you know, it gets to another thing off the place that we actually have data for, it gets a lot of stuff off the plate that we don't have data for. Good pick up, Wilbur. Thank you.

MR. JOHNSON: Okay.

DR. LINTNER: It says, the point of information to this -- this tetrapeptide-7 is sold as a solution, as such, the tetrapeptide -- under tetrapeptide-7, under the commercial name from rigin, R-I-G-I-N, and it's concentrated at 500 ppm in that solution. The same peptide is used at 50 ppm in the Matrixyl 3000 combination. So all the data that we have from Matrixyl 3000 include quite a combination; the safety of this combination, 100 ppm tripeptide-1, and 50 ppm of tetrapeptide-7. Right? So it's just in the -- 10 times concentrated version of this tetrapeptide-7 -- it is unique, you know, to Dr. Gill. Go ahead, Doc.

DR. LIEBLER: Just on one thing Dr. Belsito, I'd just like to be clear on the -- each ingredient that will be included in the revised safety assessment.

DR. BELSITO: Will be the metal salts and the fatty acyl derivatives of tripeptide-1, hexapeptide-2, and tetrapeptide-7. So this report will be labeled Safety Assessment of Tripeptide-1, Hexapeptide-12, Tetrapeptide-7 and their Metal Salts and Fatty Acyl Derivatives as Used in Cosmetics, will be the title of the new report.

And I don't think you have to do -- change much of the report, except get -- you know, get rid of the ingredients that we are not reviewing, that don't fall under that rubric. And I don't think we have data in the report on any of those other ingredients, so it's really just your tables and the list of ingredients we are reviewing that will change.

SPEAKER: Sure.

DR. BELSITO: Any other surprises that we didn't think of?

MR. JOHNSON: I think that's the last one.

DR. BELSITO: Okay. That was a good point Wilbur, we don't want to miss that. Thank you.

MR. JOHNSON: You're welcome.

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DR. MARKS: Yes. Actually we're removing sodium sulfate and the three per-sulfates that had been previously removed, or previously reviewed and concluded on. Okay. Any other comments about the inorganic sulfates? Thanks, Wilbur.

Next is the palmitoyl oligopeptides. So this is the second look at this group of -amides or -amides. It's a draft report. It was tabled to focus on knowing the peptide sequence at our last meeting. So we're back to Tom and Ron's -- are the ingredients okay? There were significant amount of comments about -- on Wave 2 from the Lintner letter. Ron Shank had raised concerns about the biologic activities, like angiogenesis, collagen census on repro development and I will open it up now. It's been retitled to the tripeptide-1, hexapeptide-12, and that was the focus on the Lintner letter. So Rons and Tom -- comments? And Hal, how do you want to proceed?

DR. SHANK: I still have the question, are these considered drugs? And I think we've had some response. Dr. Lintner says absolutely no. On the other hand I would like to hear what the FDA -- is there an official position? Oh -- okay, sorry --

DR. LINTNER: Dr. Lintner.

DR. SHANK: You are Dr. Lintner, all right. Good morning.

DR. ANSELL: Thank you for being here.

DR. SHANK: I very much appreciate --

DR. LINTNER: Thank you for letting me be here.

DR. SHANK: I very much appreciated all the material that you submitted, and I understand your position's saying no these are not drugs, but I would like to know what FDA says, so -- there is significant biological activity by these compounds, but what does FDA say? Are they drugs or are they not?

DR. GILL: We don't have any representatives from FDA present today, as far as I know.

DR. SHANK: And they didn't comment -- in the past?

DR. GILL: We got no comment from FDA on it.

DR. HILL: Yes, so I agree. I'm happy, since you're sitting three feet from me, to say I agree with everything you said. I still have the same question that Dr. Shank has, of, are these drugs or not, but it seems if FDA is not commenting, then we're reviewing them. But then you'll like me even less than with the metals report because for me, because we are seeing activity, it puts us, from where I sit, into a peptide review regime. One ingredient -- what do we know about the biology? Because if we are effecting things like keratinocyte proliferation, then that's going to be a very specific biological activity and not just something that is non-specific membrane effects, or like that, so -- I completely agreed. I was thrilled to see your letter. It pretty much put my thoughts nicely into writing, so that I didn't have to do it. So what he said is my response, and then that, to me, means we don't group. We put them in one ingredient -- one report. And nobody's going to like that -- that wants to have efficiency and contrasting and all of that, but, for me, mixing toxicology in this way, unless there's a specific read across, and then when you see a specific biological effect, with a dose response, then how do you read across? You can't, so that's how I felt about the whole thing.

DR. BERGFIELD: Could I respond on the collagen synthesis? We have a lot of cosmetic ingredients that do that. The angiogenesis is the new kind of statement. But with collagen synthesis you're going to have some angiogenesis in the skin. But we have the alpha-hydroxys, all of the fruit acids that are in that group -- all of them do that -- it's been well established.

DR. HILL: And those ought to be drugs, number one, and since they're not, I think I would contrast, in teaching -- I'm sorry, I know I shouldn't have come out and said that in open meeting, it will be captured on transcript -- but if not, fine. That's up for the FDA to decide and only the FDA to decide. But if not, then the mechanism matters. So when I began teaching medicinal chemistry to people who have never heard it before, I talk about specific versus non-specific actions. And we use the example of molecules that simply modify membrane characteristics in such a way that tadpoles stop swimming, just as an example. So the mechanism then matters, and mechanisms for compounds such as Wilma was talking about -- I think I have some sense of how those go. These I suspect aren't of that same kind of nature -- that there's some more specific mechanisms going on here. If we knew that that was not the case, then that changes things, but without knowing that that's the case, I don't know how to decide whether one can read across or not.

DR. SLAGA: Well I had a concern that there was many very strong biological activities. Most of these are done in cells in culture, which are very different from you applying on the stratum corneum of the whole skin. And first of all, I'd like to see if any of these angiogenesis really occurs when you put them on the skin. I really doubt it but we don't have that data to really do that. Our big concern last time was if you have some latent tumor cells related to either non-melanoma skin cancer or even melanoma, what these type of things may have on pushing those cells to a more active state. All of that's going to be very, very difficult to get at. So it's really, when it comes down to concentration, what effect they really have on the skin -- not what they do in vivo, or in cells in culture.

DR. MARKS: So I think it would be helpful. We keep referring to your letter, Dr. Lintner, and how good it is, but maybe you can summarize it, because what I took out of it was -- you really focused -- there were just two peptides being used -- the ones mentioned in this draft -- the tri-peptide, the hexapeptide-12 -- that it's in very small amounts -- like 10 parts per million I think was what you said.

DR. LINTNER: Even less, yes.

DR. MARKS: There's small size and low penetration, therefore these are -- these two are safe, but that -- I may have interpreted what you said in your letter incorrectly, so, why don't you embellish that, and I guess the Panel Members might say -- if you want to split out, maybe we should only do these two ingredients and only relate to them. But I don't know if that will answer the concerns about biological activity. But Dr. Lintner, please elucidate and correct what I may have misinterpreted.

DR. LINTNER: I don't think you misinterpreted anything. Thank you for all your comments. There are a number of things which I have to answer. First the question of FDA -- I've had some informal discussion with the person from FDA. I think his name is John, but I don't remember, was it at the SEC meeting recently, where he says the FDA has a difficulty defining what is a drug and what is a cosmetic, because they realize that almost everything that you put on the skin has some biological activity. J&J has recently published a small item showing that glycerin modulates keratin proliferation so shall we call glycerin a drug? I don't think so. The question is -- and the FDA actually -- CFR 21 clearly says what makes it a drug is the intent presented to the consumer, not what the biological activity of the substance is. So these products, as long as they are used in cosmetic pilots, in cosmetic presentation, with claims that are acceptable -- they are not to be considered as drugs. If they have systemic activity, that would be something different. But as I've tried to show in my paper, the amount of peptides that could even get into blood is so tiny, that there is just no concern. Caroline Eisenmann suggested that I focus on these two peptides, especially because of the fact that there is 50 or 60 in your list. Most of them are from companies I don't -- I've never worked for -- I don't know their product so I cannot say anything about peptides from other people, and so she has suggested I talk with

just on this two, which I did. As you said, these two peptides, they are being used in cosmetic products worldwide, in general at use levels of between 1 and maybe 10 ppm -- very, very unlikely that they are used at higher concentrations because there is no need, because these are the recommended concentrations by the supplier, and because they are also very expensive. So as I said, in one small comment -- the FDA usage showing that people have used it at one percent, is absurd. That cannot be imagined -- 5 ppm, 10 ppm is the usual use concentration in any cosmetic cream or other Galenic form. Most of these peptides are used just in eye creams, skin care creams, and not in large volume products. As an estimate, over the last 20 years since they have been introduced, there is not more than 50 kilograms, 80 kilograms -- that the total maximum of these peptides worldwide. So the amounts are very small. Amount doesn't mean anything. We know that there are some substances that are highly toxic, but this is certainly not the case with these peptides. One thing I tried also to show in my paper, is that if you take into account penetration, or even if you don't and you assume 100 percent penetration, you will still be at an extremely low concentration of these peptides in the tissue -- in the living tissue. And especially these two peptides being fragments of natural proteins, such as the elastin and collagen, I've tried to do a calculation and estimation of the ratio of the peptide being applied to the skin at the ratio of these fragments being present constantly in our body, in our skin, due to skin renewal, which is appearing all the time. So, if these peptides have a negative affect being a thousand times more concentrated in our tissue constantly, than those that we put on the skin, then these peptides would do us damage rather than help us, which is their purpose in the so-called matrikine concept.

DR. HILL: Could I interrupt briefly, since I have the luxury of you sitting here. The one place where I did part from your logic is palmitoylation does several specific things. One is -- it would considerably increase the likelihood that we at least, to get down to the viable epidermis. Two -- it will greatly increase the ability of a peptide to get inside a cell if there is any action there. And three -- palmitoylation specifically protects peptides and small proteins from enzymatic degradation. So if we have free peptides generated as fragments from collagen, for example, they can be enzymatically handled. But for example, maybe this is a bad example, but the drug Liraglutide uses the strategy of palmitoylation to greatly extend the half-life of that protein in the blood stream and in tissues, and as far as I understand it, although the manufacturers are unclear and there are no publications -- that palmitoyl group does not have to be removed in vivo for it to exert its anti-diabetic action -- so in cretin type effect. So I think we can see that they are doing something at very low concentrations. That argues for a specific rather than a non-specific activity. I think with the palmitoyl group in place, it's highly likely that they would persist for a substantially longer period of time and so to right off that -- well, we have a lot more of that naturally present because we're always turning over collagen which is true, doesn't completely put to bed in my mind, the fact that these things are doing something biologically specific, and since FDA has washed their hands and I don't -- that's okay -- I think our science is at least as good as theirs in some cases -- we're going to just review what goes on with these things under the conditions of use. But the palmitoylation was the one place where I think I parted ways with you in terms of all of the overall logic.

DR. LINTNER: Thank you Ron. This is of course a very pertinent comment, and I agree partly with you. Palmitoylation is a natural process in our body, but that means the body has enzymes to cut the palmitoyl and to put it there. So the natural mechanisms to take off the palmitoyl are in the skin. It is indeed extremely difficult to study what happens to peptides that you use at 3 ppm in a cell culture and to see, does it get cut down into smaller pieces? Two comments, nevertheless. One is -- the study that we published in the International Journal of Cosmetic Science, 2000, which has cited where, with the radioactive label study on it -- even smaller peptides. The smaller reduced, the higher per possibility of the peptide getting through the stratum corneum, and we compared the Lipo- carnosine dipeptide with just the carnosine without the palmitoyl. And the purpose was, indeed, to show that skin diffusion into the epidermis is improved by the palmitoyl. But, with this diffusion cell -- France diffusion cell -- we also looked at the radioactivity in the receptor fluid. And not only did we get a 100th of a percent of the applied radioactivity to the skin, in the receptor fluid, over six hours of study, 100th -- 0.01 percent, only, and secondly -- the small amount of radioactivity that we found in the receptor fluid was almost following the same identical curve for both the carnosine and the Lipo-carnosine, which I interpret, because the radioactivity was not the whole peptide -- it was just on one amino acid, the histadine residue. I interpret this as showing that once you are below or in the dermis, where you get the contact with receptor fluid, the peptide has been cut short, otherwise we would have a much more significant difference in the trans- dermal diffusion, or penetration of the peptide -- into the receptor fluid. That is one aspect. The other is, as I mentioned also, on a different, slightly longer but highly charge anhydrophilic peptide, KTPKS -- also palmitoyl -- we did a penetration study, again, with the radioactive label, and we found only three percent of the applied radioactivity in the dermis. And again, I don't know if the peptide or just the lysine residue is what we found. But only three percent was found in the dermis. So I think the two assume that negligible amount of entire or still active peptides are found in the blood stream, if it gets there.

DR. HILL: I want to be very clear. I wasn't the least bit concerned about any systemic toxicology -- only things going on in the skin.

DR. LINTNER: Yes, but in --

DR. HILL: In all the statements I said before, I wasn't concerned about anything systemic -- only things happening in the skin.

DR. LINTNER: Okay, but in the skin, it's perfectly fine if we stimulate collagen, like retinol does, or alpha-hydroxy acids and other things. These two peptides have never claimed angiogenesis, neither inhibition nor stimulation. They are simple collagen and tissue ENC extra cell ACM tissue modulating activities.

DR. MARKS: So let's get back to the ingredients. Because I, in my own mind, and not clear if we go on, however you want to -- which page you want to go to. We go on page 6 of this report. Wilbur has listed all the ingredients and right at the top is the palmitoyl oligopeptide which I understand now as basically a synonym or the INCI is for the tripeptid-1 and the -- or I mean for the palmitoyl tripeptide-1 and palmitoyl hexapeptide-12. So I kind of in my mind group those three different things together, or ingredients together, and then everything else is -- what do the Team Members want to do? Ron Shank, do you want to limit this report to just the tripeptide-1 and palmitoyl hexapeptide-12, since we seem to know the most about that? And again, that being the equivalent of the palmitoyl oligopeptide, is what I understand.

DR. LINTNER: This is a historical mistake by the nomenclature committed of the then CTFA, who, for the first time, were confronted with a synthetic peptide with a -- as a cosmetic ingredient. They named the first, and the second, and bunched it together as palmitoyl, which I thought, well -- strange. But recently this was corrected and now term INCI name palmitoyl oligopeptide disappears and is replaced by a specific name for each of the two peptides.

DR. HILL: Right.

DR. LINTNER: So you can -- I don't want to teach you but --

DR. HILL: You can teach us.

DR. LINTNER: You can write your report on tripeptide-1 and hexapeptide -- palmitoyl hexapeptide-12 -- and to keep the palmitoyl hexapeptide without the palmitoyl, is a different story, like the copper peptides and so on.

DR. MARKS: So team, what do you want to do on that, because the title is enro -- interrelated -- whatever chemicals -- I have to go back and look at it. Do you want to limit it to these two? Can you read across to all of the rest? I can see Ron Shank shaking his head yes, limit. So -- oh, he did it non-verbally. But we'll get his verbal comment for the recording, too. So we would limit to the tripeptide-1 and the hexapeptide-12 and not -- and eliminate the related -amides. Which is the correct name, -amides or - amides?

DR. LINTNER: Yes.

DR. MARKS: Or is it like either?

DR. LINTNER Yes.

DR. HILL: I was going to say potayto, potahto, but it was recently brought to my attention that nobody says potahto.

DR. MARKS: Yes, anymore. So let's --

DR. HILL: However, there's a remaining unresolved issue which is that hexapeptide-12 can apparently be one of two different molecules and we have a lot of biological data about one and zip on the other.

DR. MARKS: Well let's get back to -- well, we'll answer that question in a minute. Do you want to limit it to those two, or can you read across to the other ingredients that --

DR. HILL: I would very much -- I think you heard me earlier -- like to limit it to no more than those two.

DR. MARKS: Okay. Ron Shank?

DR. SHANK: Just the two, yes.

DR. MARKS: Okay, so we're limited to those two. And then, Ron Hill, you had a concern about the hexapeptide-12.

DR. HILL: I think Dr. Lintner was going to make a comment and the one he was -- there was one of the two that he was familiar with, but it appears that there's another vendor or another company out there who decided to do a sequence scrambling, I assume, so they could get around the patent, and it still fits within the INCI description. So if it is doing something specific, which the kinds of concentrations that are being applied suggest that there is -- there are biochemically specific effects in the cells and in the skin, then that would be presumably fairly highly dependent on the sequence, and we don't have any biology on that other one. And so if I were to draft a conclusion on my own, with no other input, I would say, evidence is sufficient for those two, and insufficient for the third.

DR. MARKS: Ron Shank? Or Dr. Lintner.

DR. LINTNER: Just a comment. I was not aware of this other hexapeptide until I read your draft report -- never heard of this before, and I have absolutely no idea who or what and why.

DR. MARKS: Wilbur?

MR. JOHNSON: So the safety assessment will just be on -- based upon your recommendation, tripeptide-1 and hexapeptide-12?

DR. MARKS: Correct.

MR. JOHNSON: What about the palmitoyl peptide-1 and palmitoyl hexapeptide-12?

DR. LINTNER: That's what I meant. The palmitoyl --

DR. MARKS: Yes.

MR. JOHNSON: So all four?

DR. LINTNER: No, no, no, no, no.

DR. MARKS: Two -- palmitoyl.

MR. JOHNSON: There are two palmitoyls.

DR. MARKS: Right.

MR. JOHNSON: But the tri-peptide-1 and hexapeptide-12 will not be included?

DR. MARKS: Correct. That's what Lintner referred to in his letter. So -- but, let's get back to this supposed other hexapeptide-12. Wilbur, do you want to comment on that, since Dr. Lintner wasn't aware of its existence? Where did that come from Ron Hill? You focused on that.

DR. HILL: So this derma molecule is apparently APGVGV -- which is alanine, proline, glycine, valine, glycine -- the other one -- it's the other one.

DR. LINTNER: It's the other one --

DR. HILL: Okay, no, you said the APGVGV sequence was never proposed by anyone in the capes but it appears pretty clearly that it is being made. We had some specific information to say that yes, this other sequence is out there in the market.

DR. LINTNER: I don't know everything.

DR. HILL: I know -- I know, I wasn't suggesting you should or did, but we have means of getting some of that information and the report suggests that other peptide is out there, and I can only presume that gets them around the patent, but we don't have any biology.

DR. LINTNER: Right.

DR. MARKS: Wilbur?

MR. JOHNSON: One other concern that I have is the fact that data on Matrixyl 3000 are included in the safety assessment but that trade name material consists of palmitoyl tripeptide-1 and palmitoyl tetrapeptide-7. So with that in mind, should data on Matrixyl 3000 be used in this safety assessment?

DR. HILL: Do we know if that hexapeptide's -- no, the peptide-7 -- is it hexapeptide-7?

MR. JOHNSON: A palmitoyl tetrapeptide-7.

DR. HILL: Tetrapeptide-7 -- do we know if that's being used separately and individually? Is that one of these ingredients? I'm not looking back. I should.

MR. JOHNSON: It is.

DR. HILL: Okay, so then we probably need to include that in the report and go ahead and use the data on that mixture. What do you think?

DR. SLAGA: Well, maybe, maybe not. That's most of the data, isn't it?

MR. JOHNSON: Yes, that's --

DR. SLAGA: If you eliminate that, you eliminate the report.

DR. MARKS: Ron Shank, comment?

DR. SHANK: So let's go back to your -- I want to get -- that's another issue. Let's resolve Ron Hill's issue of this other peptide sequence for the hexapeptide-12. Is that correct?

DR. HILL: Yes, I don't think we need to resolve it. I'm just saying, if I were to draft a conclusion, based on what's in there now, it's sufficient for -- but we have the issue that we have a dictionary name that's ambiguous. That's the catch.

DR. MARKS: Okay. So, and then, now Wilbur, so you're not concerned it can still say is the tripeptide-1, hexapeptide-12, and the Wilbur, you bring up the issue that the material that's being tested contains, besides the 12, it contains --

MR. JOHNSON: Palmitoyl tetrapeptide-7.

DR. HILL: So we ought to leave that ingredient in, as part of the review, and see if we get any more data on that one specifically. I'm guessing we already beat on industry, and this is what we're getting.

DR. MARKS: So actually, the ingredients will be three now. It will be the tetra -- the tripeptide-1, the hexapeptide-12 and the third ingredient will be the --

DR. LINTNER: The tetrapeptide-7. But I'm sure that Sedema will supply information on that one too. Because again, it's a Sedema product.

DR. MARKS: If not, it sounds like we might can get it -- tomorrow.

MR. JOHNSON: I think the data on that ingredient are included in the initial safety assessment on palmitoyl oligopeptide, so if that is the case, those data can be incorporated here.

DR. MARKS: That would be wonderful. So tomorrow, how do you want to proceed? Are we going to do a tentative report with safe? Are we reassured now with the biologic effects for these three ingredients? If not safe, then is there an insufficient data notice?

DR. HILL: I would like it to be insufficient with respect to that alternative peptide - palmitoyl peptide sequence -- the impersonator hexapeptide -- hexapeptide-

-- palmitoyl hexapeptide-12. For me, that's still insufficient. So I don't how we make a conclusion where we have one ingredient that's schizophrenic, but that's -- that's where it lands in my mind.

DR. MARKS: Ron Shank? Tom?

DR. SHANK: Do we have clear use concentration data? Because the report seems to imply that some of the products use too much. It's not realistic that they would use so much. The reason I ask that is because the skin sensitization data that we have is well below what we have use concentration. But certainly agrees with what you have said. The use concentrations would be much lower, and therefore, we have sensitization data. So I'm just confused as what data are we using for concentration of use?

DR. HILL: For me that's easy. We add to the conclusion and say, no more than x percent. And then if somebody's out there actually using it higher, leave them to support they can prove it's okay. That's how I would look at it.

DR. MARKS: Yes, I thought the sensitization was fine, but I was going on what Dr. Lintner's concentrations of the 0.001 percent, and we have HRIPT on .1 and .01 percent.

DR. ANSELL: Yes, Carol is chasing after this, but it's almost certain that they're using one percent of a solution that they bought.

DR. MARKS: Right.

DR. ANSELL: Which itself contains a few ppm of the material.

DR. LINTNER: So one hundredth --

DR. ANSELL: Yes, yes, and so that would be much more typical in reports like this, consistent with the entire marketplace. It's inconceivable that anyone would actually use it at these percentages. They'd essentially have to buy the worldwide supply of it for their one product.

DR. HILL: And if they were, then I retract what I said about systemic toxicology, because that, I think would put in a regime where we might be concerned.

DR. MARKS: Yes, I actually -- go to page 46 Ron Shank.

DR. SHANK: Forty-six?

DR. MARKS: Yes. So, if we look at the 1 -- the use concentration's.001, and if we look at the 12, it's.002, so in that use concentration, it's very low. It's consistent with what we heard. Where was the one that -- and then the other one that we're going to include is the tetra -- is that right? Is that even being used? Yes. What was the third ingredient? I have to go back and --

DR. LINTNER: Palmitoyl tetrapeptide-7.

DR. MARKS: Seven. Is that -- is that on the --

DR. HILL: It's not showing up on the creative use table though.

DR. MARKS: Yes, exactly. So, that's where I got confused, I think, Wilbur, when you brought that up. If it's in the material, why isn't it in the table?

DR. LINTNER: Maybe it's not much sold in the U.S.

MR. JOHNSON: Yes, with respect to the memo-

DR. MARKS: Yes.

MR. JOHNSON: We received from industry, that wasn't one of the ingredients that was recommended for inclusion in the revised safety assessment. So that's why it doesn't appear here.

DR. HILL: Well if we can find out that that's because it's not being sold as a separate ingredient in the United States, then that -- we just have to figure out how to put that in the report.

DR. LINTNER: We can't guarantee that, but perhaps so small amount that it doesn't show up in the FDA. I don't know.

DR. HILL: That's just a direct -- somebody can easily get that answer. Is it being sold separately in the United States, or only as a combination?

MR. JOHNSON: And one other thing -- the amino acid sequence was not included -- the specific amino acid sequence was not included in the dictionary. But I think that according to one of the publications included in the text, that amino acid sequence is stated.

DR. HILL: Well the point is with the hexapeptide, there are two alternative ingredients -- two different molecules that meet the criteria to be called that particular ingredient name, by INCI's name. The point is that --

DR. ANSELL: The point is, as Dr. Lintner pointed out, that's an historical artifact, it has been corrected and that name is retired.

DR. HILL: So hexapeptide --

DR. ANSELL: Yes. The palmitoyl peptide.

DR. HILL: I thought it was the palmitoyl oligopeptide that's been retired. So now we have hexapeptide, but the point is that there are two different sequences that both meet the hexapeptide name. So there are two different molecules, which, if they're not doing anything specific in the tissue, and we know that, fine. But all the evidence suggests to me that that's not the case, and that these two different sequences would be biologically disparate, and so, that's why I say, a conclusion for me would be -- that ingredient with this specific hexapeptide sequence -- safe as used, capped at.1 percent. The other one -- insufficient data.

MR. JOHNSON: On page 24 --

DR. HILL: I'm there, yes.

MR. JOHNSON: Yes. The amino acid sequence for the palmitoyl tetrapeptide-7 is included. And that paragraph immediately above -- physical and chemical properties --

DR. HILL: Yes, I wasn't talking about 7 right then, though. I was talking about the two different flavors of palmitoyl hexapeptide.

DR. MARKS: Which one is it?

DR. HILL: 12 -- hexapeptide-12. There are two different versions of palmitoyl -- two different molecules that are both called palmitoyl hexapeptide-12.

MR. JOHNSON: Right. Yes.

DR. HILL: All right, one -- I think we have enough information to conclude -- safe as used if, in reality the use is.1 percent. The other one, we have no data, for me, that one's insufficient. So if we say it's sufficient, meeting -- let's see -- which is the current -- the sequence we know a lot about --

DR. LINTNER: VGB.

DR. HILL: Yes, VGB. That one we know. The one that is VGV -- no, let me see, wait a minute, where is it --

DR. LINTNER: On the (inaudible), VGVAPG.

DR. GILL: I have it on page 23, Ron. I think it's -- Wilbur has it as also known as tripeptide-1. It's that second sequence for the hexapeptide-12.

DR. HILL: Yes. Okay. So tripeptide-1 is a different ingredient.

DR. LINTNER: Yes.

DR. HILL: And so -- so how do we deal with this? So if it's being sold under both names, that's a problem. I should say, if it's being labeled under both names -- the hexapeptide-12 is maybe actually two different molecules. That's a problem. Because

one of them we have data for and the other one we don't. I need to write down so I can use a cheat sheet and reference it, but -- am I losing you totally? There's two different molecules --

MR. JOHNSON: I know -- I know what you're saying.

DR. HILL: All right. There's two different molecules called hexapeptide -- palmitoyl hexapeptide-12. One of them we have plenty of data on. The other one, we have zero data on. So in my mind, we're sufficient -- safe,.1 percent or below and the other one --

DR. MARKS: I don't think we need the 1 percent limit, do we? Because it's not being used there.

DR. HILL: If we know for very certain that everybody is already in use at.1 percent --

DR. MARKS: Well, it's much less than 1 percent.

DR. HILL: Then we should make sure we state that in the discussion --

DR. LINTNER: .1 is fine.

DR. MARKS: But we normally have a conclusion, the present use and concentration, the present use of that, or the present concentration in the table was much lower than.1 percent.

DR. HILL: But we have tabulated data up to 10 percent, which we think is fictional, but we don't have information to suggest it's fictional.

DR. LINTNER: No, one percent is --

DR. HILL: Or one percent rather.

DR. LINTNER: But that is impossible.

DR. HILL: And he says it's impossible, but --

DR. GILL: And Carol is checking on that, correct?

DR. HILL: So if we know that that's impossible, I agree, we can take that out. And we still have insufficient for the second version of palmitoyl hexapeptide-12. If it has an alternative name, then the problem is just that it's being sold under the wrong name. Labeled under the wrong name -- I don't know.

DR. LINTNER: Probably, it may pop up with other peptides, because the INCI Nomenclature Committee has decided not to reveal the sequences -- just amino acid composition. And if different sequences may be even supplied under CDA -- if there's a supplier doesn't want to give the sequence, which I find absurd, but if that's the case, then that's okay, but at least different chemical entities should have different INCI name.

DR. HILL: Well, if, in terms of the safety review, from where I sit, if we have some sense that something specific's going on, and it's just nonspecific effects, then they need to -- I can't reach a conclusion if they don't reveal the sequence.

DR. MARKS: So which is the alt -- the other one you said -- what was that sequence, just so I have it noted here, Ron Hill?

DR. HILL: VGVAPG is the --

DR. LINTNER: The correct one, the safe one.

DR. HILL: VPG, or VGVAPG, which is valine, lysine, valine, alamine, proline, glycine. I'm going to put --

DR. MARKS: All right, I'm going to call on you tomorrow, Ron --

DR. HILL: Okay, I'll make sure I make myself cheat notes so that I don't have this problem referring to --

DR. MARKS: Ron Shank and Tom Slaga -- do you have the same concerns, with this different amino acid sequence, that that's going to make a big difference?

DR. SLAGA: Well, the amino acids will make a difference and I think we have to be specific to the sequence.

DR. MARKS: Okay.

MR. JOHNSON: One point -- the reason why I mentioned the amino acids sequence for palmitoyl tetrapeptide-7 is because, the reason for this particular grouping of ingredients in this report was based upon the known amino acid sequence. The known amino acid -- there was no known amino acid sequence in the dictionary with respect to palmitoyl tetrapeptide-7, so this if from --

DR. HILL: It's given in the report.

MR. JOHNSON: Yes, but this is from another reference. It's not from the dictionary.

DR. HILL: Ah. Well, then the deal would be the same. If it meets the sequence, we have data, and it supports the safety and we're find, and if it does not meet the sequence -- insufficient.

MR. JOHNSON: Um-hm.

DR. MARKS: And where, Wilbur, on this report, do you refer to the tetrapeptide-7 so --

MR. JOHNSON: It's on --

DR. MARKS: What page or pages?

MR. JOHNSON: Page 24, in the section immediately above physical and chemical properties.

DR. HILL: The sequence it's giving in the text is PAL, which is palmitoyl GQPR -- Gentleman's Quarterly Public Relations.

DR. MARKS: Wilbur, where is the testing where you say it contained the tetrapeptide-7? So which, and then the comment was made -- well, that's in everything that's being tested. Which product was that again, that you were talking --

MR. JOHNSON: That's Matrixyl 3000. That's a trade name.

DR. MARKS: And what page is that?

MR. JOHNSON: Well, it's on page 24 and I think I have it in the introduction.

DR. MARKS: Page 24.

DR. HILL: Well, it's in that same section where you just referred us to. It says, according to another source, let's see -- data on Matrixyl 3000 are included in the safe palmitoyl -- okay, somewhere you say, right there -- is one of two active ingredients in that -- right at the beginning of that paragraph.

DR. MARKS: Introduction, okay. Let me go back and -- I'm missing it. Where in the introduction is it?

DR. HILL: No, it's that same paragraph right above physical and chemical properties. I don't know if it's written again in the introduction or not. But that same paragraph, he just referred us to, where we had the GQPR -- on page 24.

DR. MARKS: So it's right above -- yes, okay. So that's the Matrixyl.

DR. HILL: And then it says the other active ingredient is tetrapept -- palmitoyl tetrapeptide-7.

MR. JOHNSON: Yes, that's the first occurrence of it, on page 24.

DR. MARKS: Okay.

MR. JOHNSON: Oh no, it is -- it is mentioned in the introduction, I'm sorry, it is. It is there, yes.

DR. SHANK: Yes, it is.

MR. JOHNSON: So would we need to -- would the Council need to confirm that that is the amino acid sequence for palmitoyl tetrapeptide-7, or is the Panel accepting this reference?

DR. HILL: Is there any reason to believe that reference is incorrect?

MR. JOHNSON: You'd have to ask the Council.

DR. HILL: Me? That's a rhetorical question at this point.

DR. LINTNER: Tetrapeptide -- palmitoyl tetrapeptide-7 is the sequence that you have announced. It is the main ingredient that is mixed with the palmitoyl tripeptide-1 in the commercially called blend Matrixyl 3000, but you have data on biopeptide CL which contains the 100 ppm of tripeptide -- palmitoyl tripeptide number 1, and then this Matrixyl -- it's just added another peptide. But it's still safe. So addition of the peptide, tetrapeptide-7 to the tripeptide-1, does not change the safety. So it does not --

DR. MARKS: The only problem I have when I'm looking at the Matrxyl under sensitization is, I don't know what concentration -- I don't know what amount of the seven is in there, to say this is a safe limit. Did I miss that?

So I would probably not include seven in the report, just because it's not under the use concentration, and I'm not sure it's --

DR. LINTNER: The amount is half of -- it's 50 ppm.

DR. MARKS: Fifty.

DR. LINTNER: Fifty. It's half of the tripeptide.

DR. MARKS: Okay.

DR. HILL: Well, then that poses the difficulty. If it was sold individually and separately, would they put it in there up to a hundred? In which case we don't have data to support it which I think is what you were trying to say.

DR. LINTNER: But we can send data -- specific data on the palmitoyl tetrapeptide-7. That is another product from Sederma. That is sold individually. Maybe not much in the U.S., but there are some sales, so, we have data on this like for the other peptide.

DR. MARKS: Okay.

DR. HILL: So if we could get that, that would be nice to keep that in, even if it's not sold in the U.S., because it rounds out this -- I think it nicely rounds out the evaluation.

DR. MARKS: So one of the things we could move forward, again an insufficient data notice to clarify the alternative -- hexapeptide-12 -- Ron Hill, you were asking for that -- that amino acid sequence and the other would be further data on the tetrapeptide-7, in terms of sensitization you said. So there could be an insufficient data notice or we could go forward with a safe. Which way would you prefer?

DR. SLAGA: Well it's early in the game, so insufficient --

DR. HILL: Yes, I think even if you just include that one insufficiency, that right now for me that's enough, and then we just ask for the other, and if we have it great, and if not, we limit to fifty -- I don't know. Matrixyl is okay and we don't know about the individual ingredient.

DR. MARKS: Yes, so the other would be an HRIPT of the tetrapeptide-7 -- even at use concentration, since we don't even have a use in the U.S., at least in the table. Yes, Rachel?

DR. WEINTRAUB: In my notes, I had there's still no reproductive or development toxicity and carcinogenicity data, so would that be a need, or is the Panel comfortable with not having that data based on other information?

DR. SHANK: We felt we didn't need because of the very low use concentration applied to the skin -- there wouldn't be sufficient systemic exposure.

DR. SLAGA: I agree with Ron Shank. It's so low -- genotoxicity and carcinogenicity is not an issue.

DR. WEINTRAUB: Thank you.

DR. HILL: However, I think it would be beneficial in the discussion to capture salient points made in Dr. Lintner's letter. It's not a published reference, but he provides references where we can capture that and reference it.

DR. GILL: You can reference a letter.

DR. HILL: Okay, personal communication.

DR. GILL: Right.

DR. MARKS: Okay, any other comments, Ron Hill, again, I'm looking at the paragraph down here on the 12 -- is it the one that is the PAL-alanine or the one that begins with PAL-valine, as the one that you need?

DR. HILL: PAL-alanine.

DR. MARKS: Okay.

MR. JOHNSON: What (inaudible)?

DR. HILL: Anything. We have nothing. Or it goes insufficient on that version of it, which is cumbersome, I know, because we have two different molecules, one ingredient but that's the deal, from where I sit. Dr. Liebler may have another take, but --

DR. MARKS: PAL-alanine was the one that you needed, right?

DR. HILL: Yes sir.

DR. MARKS: Okay. Anything else? So tomorrow I'll move that we issue an insufficient data notice. That's not uncommon. That doesn't mean that we're coming --

DR. LINTNER: That's just for the PAL-alanine?

DR. MARKS: Yes, for the PAL-alanine and then also an HRIPT on the tetrapeptide-7 or other information, but I particularly would like to see that.

DR. LINTNER: Oh sure. I'll send it tomorrow. I'll send it to you.

DR. MARKS: Yes, and Ron Shank, or Tom Slaga or Ron Hill, would you like to see anything more on the tetrapeptide-7? Are you concerned about the other toxicities with this, or not?

DR. HILL: The only other piece of missing information for me was in the summary of the manufacturing process, we -- not that I'm worried that much about it from a safety point of view, but we were not given any information about the palmitoylation

process. That's still a black box, in terms of what I see in the report. I can surmise how it would likely be done, but we don't have information.

DR. LINTNER: It is done exactly the same way as the other amino acids -- it is acid that is coupled to --

DR. HILL: Yes, but how is it coupled? We don't get that. So is it mixed anhydride, acid chloride? That's the point. And like I say, I don't really have any overwhelming concerns, because those processes are going to be proprietary.

DR. LINTNER: It is phase synthesis in --

DR. HILL: It's all with phase synthesis using what? Palmitoyl? How is it activated, I guess?

DR. MARKS: Okay, well that was a robust discussion. We're going to limit it to the one, twelve and seven ingredients -- seven meaning the number seven, not that there are seven ingredients. So the tetrapeptide-1, hexapeptide-12, and the tetrapeptide-7, so we're only going to have three ingredients in this report, and I'm going to suggest that we are moved that we have an insufficient data notice for what I said before, in terms of the hexapeptide- 12, the PAL-alanine, etcetera, amino acid sequence -- we need more information on that, and then the HRIPT and other information, if available on the tetrapeptide-7 -- use concentration.

DR. LINTNER: I'll send that tomorrow.

DR. MARKS: Ron, Ron, Tom -- does that summarize where we're at?

DR. SLAGA: Yes.

DR. MARKS: Wilbur?

DR. SLAGA: I think it's safe. But we'll get that next time.

DR. MARKS: Yes, okay. Yes, it's probably going to get there, but let's be sure. If we're going to err, we're going to err on being on the safe side. Any other comments? Okay. It's after twelve.

DR. LINTNER: Thank you ladies and gentlemen.

DR. SLAGA: Thank you.

DR. HILL: Thank you.

DR. MARKS: Thank you. That's okay. I would have kept on going, but I looked at the clock. I think we will take a break then for lunch.

Full Panel – March 18, 2014

DR. BERGFELD: And then the last item in our total list here is Dr. Marks. Palmitoyl oligopeptides.

DR. MARKS: So there was a draft report on these oligopeptides in March of last year, which was tabled. There was discussion as to what ingredients really we should be reviewing and there was a robust discussion in our team meeting on that. We felt that we would limit to three ingredients -- tripeptide 1 as in the draft report titles from Wilbur dated February 21st; the hexapeptide 12; and the only other amide would be the tetrapeptide 7. And so we would recommend limiting it to those three ingredients, that there be an insufficient data notice. And what we wanted to confirm is the hexapeptide. Ron Hill would clarify that. There are different amino acid sequences -- and if I remember of the hexapeptide 12 -- and the one I believe Ron Hill, you were concerned about, was the palmitoyl alanine. And then we need an HRIPT for the tetrapeptide 7 use concentration.

So the motion would be insufficient data notice for those three ingredients and those were the needs.

DR. BERGFELD: Ron Hill, did you want to comment before we ask the Belsito team?

DR. HILL: I just wanted to be clear that we were talking about the palmitoyliated species.

DR. MARKS: Correct.

DR. HILL: Which we have.

DR. MARKS: Thank you.

DR. HILL: And I don't know -- and further reflecting overnight, I'm not sure what we want to do with the unpalmitoyliated peptides that are the same peptides. So whether we want to keep that in this same report or not. So, I apologize. That was further reflection last night and this morning. Hopefully, the other team will have comment on that.

In terms of the hexapeptide 12, it's because there are two different versions on the market and all the biology we have is for the VGV APG and not the APG VGV. But I'm also told that we will probably be able to get the data on that alternative hexapeptide 12 and then the big question is what about the INCI nomenclature that's ambiguous in this case.

DR. BERGFELD: Dr. Belsito?

DR. BELSITO: Well, we took a similar but different approach.

So, first of all, we thought we needed to relabel this as tripeptide 1, hexapeptide 12. There are metal salts and there are fatty acyl derivatives, to address Ron's point that we are cutting out, other than the metal salts from the fatty acyl derivatives and tetrapeptide 7 as used in cosmetics.

We agree that we remain concerned that an INCI name can refer to different chemicals, and in this case it really does make a point because the amino acid sequence can have very different effects. In other discussion, we point out the specific amino acid sequences that we're referring to when we talk about the hexapeptide and say that only that specific amino acid sequence, hexapeptide, is safe as used and that any other hexapeptide under the INCI name that didn't have that exact sequence was not safe. Furthermore, we appreciated Dr. Lintner's comments that these ranges of ingredients that were used in products made absolutely no sense. We wanted some further clarification on that. Otherwise, we would restrict to the range that we're being told is the range that could be used, and therefore, we would not need the HRIPT that you're asking for.

So our group was recommending that with a relabel as tripeptide 1, hexapeptide 12 -- hexapeptide 6, right? Not 12 -- 12, and their metal salts and fatty acyl derivatives and tetrapeptide 7 as used in cosmetics is safe as used. And the as used will be the new defined range of limits that Dr. Lintner told us were used. And again, the discussion would clearly say the safe as used implies that the hexapeptide 12 has the exact same amino acid sequence as the one we reviewed, and any other hexapeptide 12 that had a different sequence would be insufficient.

DR. BERGFELD: Dr. Marks?

DR. MARKS: I'm going to ask Ron Hill and Ron Shank.

Okay.

DR. BERGFELD: So are you withdrawing your motion?

DR. MARKS: Yes, I'll withdraw.

DR. BERGFELD: And your motion?

DR. MARKS: My motion.

DR. BERGFELD: Do you want to make a motion?

DR. BELSITO: So my motion is that we go out and resurvey Industry because we're told the concentration of uses we've gotten are impossible. If we get them back, we will go back and say we found these ranges of concentration of use; however, we've been informed that the usual concentration of use is these extremely low uses and that that's the ones that we're reporting on safety where the manufacturer tells us is the concentrations that they recommend these be used as. And in the discussion, again point out that it's not any hexapeptide 12; it's the hexapeptide 12 with that specific amino acid sequence. With all of those aspects in the discussion and with the relabel of the report as tripeptide 1, hexapeptide 12, their metal salts and the fatty acyl derivatives, getting rid of all of the other ingredients that aren't metal salts or fatty acyl derivatives, we felt we could go with a safe as used conclusion.

DR. BERGFELD: Now, I have to ask you a question. You're asking for a search on the actual concentrations prior to this going out?

DR. BELSITO: I'm asking -- I'm asking that Carol go with the ingredients we are now including, which is a much smaller universe than the ingredients we looked at.

DR. BERGFELD: Okay.

DR. BELSITO: Whether any of those that fall outside of the bound that Dr. Lintner told us yesterday is the usual and customary use that she go back to those companies and say, "Are you really using it at that concentration?" If they say yes, you know, it will be reported. But then in the Cosmetic Use section I think there would be a paragraph that, you know, information that we've received from the manufacturer is that these are -- their recommendations are for concentrations of use within a given range.

Is that not correct, Dr. Lintner, that you have that?

DR. LINTNER: Yes.

DR. BELSITO: And that we say these are the ranges that we would consider safe as used.

DR. BERGFELD: Ron Hill and then --

DR. HILL: Yeah. Because out discussion yesterday was only the palmitoyl. So I want to be clear. When you say fatty acyl, are you restricting to myristoyl and palmitoyl?

DR. BELSITO: Yes.

DR. HILL: Okay. I'm okay with that.

DR. BELSITO: I think those were the only other two fatty acids.

DR. HILL: I think so. I just want to be clear.

DR. BELSITO: So we're eliminating all of the others. It's a very small group. And those are the ones that are used.

DR. JOHNSON: Dr. Lintner, just for the record, what is that use concentration range?

DR. LINTNER: It is in what is called a PPM range between one and let's say 20, 30 PPM, parts per million of peptide in a cosmetic product.

DR. JOHNSON: Between one and --

DR. LINTNER: One and 30.

DR. JOHNSON: But what is the customary concentration?

DR. LINTNER: It is below 10.

DR. JOHNSON: Below 10. Okay, thank you.

DR. MARKS: So Don, do you mind going on to page six and saying which specific ingredients going from that list? I know you grouped it.

DR. BELSITO: Basically -- I was moving on to the next one, so let me go back.

DR. MARKS: Oh, that can be clarified later. You can clarify that.

DR. BELSITO: I mean, basically, where the metal salts and palmitoyl and myristoyl.

DR. BERGFELD: Okay.

DR. MARKS: Okay.

DR. BERGFELD: All right. We understand what we're voting on now?

All right. No other discussion? I'm going to call the question then. All those in favor of this motion please indicate by raising your hand. Thank you. It is unanimous.

(Motion passed)

Meeting Summary

Tripeptide-1, Hexapeptide-12, their Metal Salts and Fatty Acyl Derivatives, and Palmitoyl Tetrapeptide-7

The Panel issued a tentative safety assessment for public comment with the conclusion that the following 10 ingredients identified as tripeptide-1, hexapapeptide-12, their metal salts and fatty acyl derivatives, and palmitoyl tetrapeptide-7, are safe in the present practices of use and concentration in cosmetics. This conclusion is applicable only to ingredients with peptide sequences that are defined as follows: tripeptide-1 (glycine-histidine-lysine), hexapeptide-12 (valine-glycine-valine-alanine-proline-glycine only), and tetrapeptide-7 (glycine-glutamine-proline-arginine).

tripeptide-1

palmitoyl tripeptide-1

myristoyl tripeptide-1*

hexapeptide-12*

palmitoyl hexapeptide-12

myristoyl hexapeptide-12*

copper tripeptide-1

bis(tripeptide-1) copper acetate*

manganese tripeptide-1*

palmitoyl tetrapeptide -7

*Not reported to be in current use. Were ingredients in this group not in current use to be used in the future, the expectation is that they would be used in product categories and at concentrations comparable to others in this group.

Palmitoyl hexapeptide-12 is reported to function as an antioxidant in cosmetic products; the remaining 9 ingredients reportedly function as skin conditioning agents.

These ingredients were initially included in the CIR safety assessment titled Palmitoyl Oligopeptides. This group was subsequently revised to include only ingredients with a defined peptide sequence (i.e., tripeptide-1[glycine-histidine-lysine] and hexapeptide-12 [valine-glycine-valine-alanine-proline-glycine]) bonded to a palmityl group or one of various other groups. The Panel specifically pointed out that this assessment does not apply to the other sequence listed in the INCI dictionary for hexapeptide-12 (i.e., ala-pro-gly-val-gly-val). Because of major differences in chemistry/biological activity between some of the more complex groups attached to the peptide, the Panel determined that the current safety assessment should include only tripeptide-1, hexapeptide-12 (valine-glycine-valine-alanine-proline-glycine only), their metal salts and fatty acyl derivatives, and palmitoyl tetrapeptide-7 (Pal-glycine-glutamine-proline-arginine). The latter ingredient was added because it is a component of one of the trade name mixtures containing palmitoyl tripeptide-1, for which safety test data are available.

During the Panel discussion, an expert research scientist in the field of cosmetic peptide chemistry commented that peptide ingredients are used in cosmetic products at concentrations between 1 ppm and 30 ppm, but concentrations < 10 ppm are customary. He also provided genotoxicity, ocular irritation, and human repeated insult patch test data on palmitoyl tetrapeptide-7. The Panel determined that, overall, the data in the safety assessment, were sufficient to support the safety of these ingredients in present practices of use and concentration in cosmetics.

JUNE 2014 PANEL MEETING – THIRD REVIEW/DRAFT FINAL REPORT

Belsito Team - June 9, 2014

DR. BELSITO: Good, so that takes care of that. Moving along. Okay, so tripeptide 1, hexapeptide 12, and then metal salts. At the March meeting we concluded that they -- tripeptide 1, hexapeptide 12, their metal salts and fatty acid derivatives and palmitoyl tetrapeptide 7 were safe in the present practice of use and concentration in cosmetics, and went on to further specify that the safe conclusion is applicable only to the named ingredients that have the following peptide sequences, and we defined them because as we learned, the INCI dictionary is not inadequate for ceramides, it's inadequate for these as well, so we felt the need to define the chemical sequence, and I thought it looked good. I don't know if I have any comments. Let me open the document after I save this. That's under peptides, right?

DR. BERGFELD: Right. Peptides.

DR. BELSITO: Page 36. Oh, I guess we don't do the inhalation boilerplate until the discussion. Is that correct? So, we don't need that there, so that I can delete. And then --

MR. JOHNSON: Excuse me, Dr. Belsito, generally we do include that information in the use section because we include the references --

DR. BERGFELD: Right.

MR. JOHNSON: -- in the use section. But I guess during the last review it wasn't requested that information be included in the use section. You just mentioned the discussion.

DR. BELSITO: Okay, so we normally do put it in?

MR. JOHNSON: We normally do put it in there.

DR. BELSITO: So, then it should be in.

MR. JOHNSON: Okay, so that will be added.

DR. BELSITO: And then just a comment here on page 54 on palmitoyl tetrapeptide 7, I'm just shocked that it has 249 uses and no reported concentration.

DR. EISENMANN: That's because you put it in last meeting there and they didn't want to delay it, so there wasn't time to do a concentration of use survey, so I'm assuming you're limiting it to --

DR. BELSITO: The same low levels --

DR. EISENMANN: -- same -- 30 ppm with a typical of 10 ppm. That's --

DR. BELSITO: Is there a request out to get ranges?

DR. EISENMANN: No, I have not sent out a request to include that in the -- because I knew you wanted to finalize it at this meeting, and there just wasn't going to be time between March and now, so if you want to delay it -- to ask, I—

DR. BELSITO: You know, from Dr. Lintner's presentation, it's quite clear that from a financial and chemical standpoint these molecules are going to be used in very low levels, so I presume that the palmitoyl tetrapeptide-7 will be in that same range? If I were wanting to be in a group that was negative of what the CIR does and our thought processes, I would look at that and go, "There are

250 uses of this product, and they don't know how much is in these 250 products." I know you don't like to delay things. I'd be fine going out as a final, but I mean that would be purely editorial to add that information unless it ended up changing our discussion and then we could go "oops," so I would like to see you get that and put it in before this paper gets published.

DR. LIEBLER: I agree. And it's one thing to sort of proceed provisionally with the expectation that the use levels would be very low so we can finalize our discussion and all of that. That's what we've done, but to not have it there when that's sort of a basic due diligence.

DR. BELSITO: Yeah, there are no reported uses for it if it's used in one product and we're not getting any information. It doesn't bother me, but when there are 250 reported uses and we don't have a single concentration range, I don't like my name being on a document like that.

DR. LIEBLER: (inaudible) some other changes.

DR. BELSITO: Yes, go ahead.

DR. LIEBLER: So, I think in terms of readability, with all these peptide names and then the three-letter abbreviations for the peptides which are really not very much used in the literature on proteins and peptides because they're not that much easier to read than reading the whole names, so I suggest that after you introduce these either full spelled out names, you abbreviate them with their single letter abbreviations and use those thereafter, and so I've indicated where you can do that. The other thing is that I think in the conclusion there's a nice layout of the ingredients and the sequences of how they relate to each other which is the two-column display in the conclusion. That would be so useful up front in the introduction for the reader to see, okay, this is what we're dealing with rather than the sort of gobbledygook of names in a paragraph. So, I suggest that that also be put into the introduction as a way to introduce the reader to what we're dealing with here because then they can see, oh, we've got essentially a group of peptides that are GHK and derivatives of GHK. And then we've got another group that are VGVAPG and derivatives of those, and then we've got the GQPR-oddball by itself, the palmitoyl tetrapeptide. And it just makes the report easier to understand just in terms of the ingredients just from the get-go.

DR. BELSITO: So you want this put into the introduction as well?

DR. LIEBLER: Yes, it's a two-column display, and in fact, put all the GHKs in the left-hand column, and then put all the VGVs in the right-hand column, and you'll have a nice lineup.

DR. KLAASEN: And GQ by itself.

DR. LIEBLER: At the bottom of the right.

DR. KLAASEN: Right, less space in-between.

DR. BELSITO: Mr. GQ.

DR. LIEBLER: Now, I think in several places and even in one of the figures in referring to hexapeptide 12 there's this bad history of hexapeptide 12 representing two distinct sequences, and then you were saying not alaprolivale that one. Instead of saying that not in italics everywhere in the report, just take that out and instead you have the sentence right up at the beginning that --

DR. BELSITO: Page?

DR. LIEBLER: Well, this is what I'm adding, but this would be in the introduction. It would be --

DR. BELSITO: Page 32?

DR. LIEBLER: Yeah, page 32.

DR. BELSITO: So, you would put your table after the first sentence, the safety of --

DR. LIEBLER: So, you put the table somewhere in that, either right after that introductory paragraph, right there, okay. And

then I think I suggest you have that first introductory paragraph is fine as it is. You can perhaps go -- let's see. The first one, two, three sentences and then splice in that two-column display; so, three sentences, then put in the two-column display.

DR. BELSITO: So, you're putting it where? After --

DR. LIEBLER: Right after the word "dictionary," halfway through that paragraph.

DR. BELSITO: Okay, so in the dictionary (inaudible) there.

DR. LIEBLER: Yeah, carriage return, couple carriage returns, put in those two columns, and then begin a new paragraph still under introduction with this safety assessment also includes data on trade name material, blah-blah. And all the way to the end of that sentence that ends with an oligo peptide component." Now at that point I suggest you splice in a sentence that says

"This safety assessment addresses only these specified sequences: The data or conclusions are not applicable to other peptide sequences."

MR. JOHNSON: Do you have that?

DR. LIEBLER: I have it right here.

MR. JOHNSON: Okay.

DR. LIEBLER: I'm just reading you what my edit is.

MR. JOHNSON: Okay.

DR. BELSITO: This safety assessment addresses --

DR. LIEBLER: -- only these specified sequences, and they're already laid out for you.

DR. BELSITO: Peptides with the specified sequences?

DR. LIEBLER: "This safety assessment addresses only peptides with these specified sequences: The data or conclusions are not applicable to other peptide sequences." And having said that you can scratch out everywhere where we have an italics not that other sequence so that saves you from having to throw those in all over the place. And then after the example structures, you have another paragraph on page 33 that starts the ingredient name palmitoyl oligopeptide in the INCI dictionary's been retired. You've actually just said that up above, so it's not necessary to have this entire paragraph, and I suggest leaving that paragraph (inaudible).

DR. BELSITO: Where are you?

DR. LIEBLER: It's on page 33 of the PDF right under Figure 1, Example Structures. That entire paragraph can be deleted, and then you resume with this short sentence: "The definition structures and functions of the ingredients in this report are included in Table 1." I think it will just make this report much more digestible for the reader.

DR. BELSITO: But this isn't a GRAS substance.

DR. LIEBLER: I have a couple other comments. On page 42, this is under the cell --it's on the cellular effects study. Page 42

there's a study on cell proliferation and another one, "Effect of Growth Factor Production." I

didn't look at those references, but I would delete -- those are probably of marginal relevance. I would delete them unless there is really strong evidence for a peptide sequence-specific effect on the biological endpoints described. In other words, do they have adequate controls?

DR. BELSITO: Which are you talking about now?

DR. LIEBLER: Cell proliferation and effect on growth factor production.

DR. BELSITO: And the reason for deleting them is?

DR. LIEBLER: Because the way it reads right now it doesn't look like this is going to be any evidence specific for that particular peptide, or in other words they might have thrown in that peptide, but how do they know that it's anything specific to that sequence? They have adequate controls to show that it's not a non- specific effect. I doubt that these are going to be relevant to our consideration anyway. And then on page 43 there's a section called, "Enzyme of Regulation Release Metallic Proteases," and that's all at very high concentrations that I think are irrelevant, relatively high concentrations that are irrelevant.

MR. JOHNSON: So, delete that entire --

DR. LIEBLER: Delete that section, and then there's a section on page 44 called, "Effect on Cell Adhesion." Same thing for the very high concentrations. Delete the "Effect on Cell Adhesion," and then under other –

DR. BELSITO: You're getting -- I'm not a typist, so you got rid of the Growth and then the next one before the Cell Adhesion was --

DR. LIEBLER: Enzyme of Regulation Release.

DR. BELSITO: That's page 44?

DR. LIEBLER: Correct. That's 43 actually.

DR. BELSITO: Forty-three, and then Cell Adhesion is 44.

DR. LIEBLER: Correct.

DR. BELSITO: And you're deleting all of those because of dose?

DR. LIEBLER: Right, and I'm not finished.

DR. BELSITO: Okay, but I mean that doesn't give you information that you want to consider and then the discussion saying it's not relevant because of dose, because otherwise doesn't it look like we're not looking at this data?

DR. LIEBLER: I thought this stuff was really of marginal relevance regardless of whether we looked at it. I mean it's -- the same thing as the effect of (inaudible).

DR. BELSITO: I mean I guess my only concern is that our approach has always been to be as transparent as possible and to look at all of the data that is out there. I mean, yeah, I don't have a problem with deleting the paragraph that some boy stuffed starring down his ear canal, but I guess these paragraphs do describe activity of the compounds that we're looking at, albeit not at levels that we're concerned about, and to delete them would be to the individual reading the report would (inaudible) weren't these guys and gals aware that there's data on cell adhesion and cell proliferation for these molecules? Did they not take that into account? And it sounds like you're saying, "Yeah, I took it into account, but it's not relevant to the concentrations that are used in cosmetics", and that would be something we would say in the discussion.

DR. LIEBLER: So, this takes up about two pages.

DR. BELSITO: I understand, but they're small little summaries. It's not like it's two pages on cell adhesion, two pages on growth promotion. They're saying there's a study. Here are the details, and then in the discussion we're saying we're aware that there are these biological effects of these compounds, but at concentrations used in cosmetics it's not relevant.

DR. LIEBLER: So, I think you could shrink this all down to about a paragraph.

DR. BELSITO: I don't have a problem with shrinking it as much as you want to shrink it. I mean you could even shrink it into a table and say there are reports that these peptide sequences can have biological effects, table, whatever, and you put in the reference. You put in the dose range. You put in the effects, and add the discussion –

DR. LIEBLER: (inaudible) way to deal with it is to use a table at the end and then have a brief section on cellular effects in various cellular models where you essentially indicate that these peptides have been studied and shown to have effects on A, B, C, D & E and various cellular models, see table 12. And then we can talk about that at the discussion that we noted this, that these were at levels of well above the use concentrations and probably not relevant to safety.

DR. BELSITO: And not relevant, not probably not relevant. Not relevant.

DR. LIEBLER: I say "probably" in discussion, but not in writing.

DR. BELSITO: Okay.

DR. LIEBLER: I've said my piece. I'm trying to save a tree or a byte, kilobyte.

DR. BELSITO: So instead of deleting everything we just talked about you were going to tabulate it, and then we'll just in the discussion point out that these various biological effects of these peptides are seen at concentrations that are well above what would be in a cosmetic product. Anything else? Okay. Well, let's save this puppy -- and styrene.

Marks Team – June 9, 2014

DR. MARKS: So, next are the peptides and we have before us -- Wilbur, you're really up here in the beginning.

MR. JOHNSON: Yeah.

DR. MARKS: We have Wilbur before us again. We have the draft final report on -- it's important that we notice that it's tripeptide-1, hexapeptide-12, their metal salts, fatty acyl derivatives, and palmitoyl tetrapeptide-7. The conclusion was safe. And are there any discussions to that? Let me see. That's the motion I'm going to make that we issue a final report where these ingredients are safe.

DR. SLAGA: Agreed.

DR. SHANK: I agree with the conclusion.

DR. MARKS: I guess -- and this is really minor, Wilbur, but when you look at the conclusion, my first look at that was I looked at the initials after and then I had to look up in the discussion to see what the initials stood for. I don't know whether we want to keep the initials the way they are in the conclusion. That's page 49. Very minor, but if somebody looked at the conclusion tripeptide-1 and then has GHK. Would you know what the GHK stands for under the, say, palmitoyl tetrapeptide-12 as VGVAPG? And that's minor. I mean, you could do it with an asterisk, as you've done with the others, or in this case maybe two asterisks, but you go right above in the same page, you have what they are.

MR. JOHNSON: Yes, that's what was mentioned. Yes.

DR. MARKS: So, I don't -- to me, I don't know whether Ron -- obviously, Ron Shank, Tom and Ron Hill, you didn't pick that out, but for me, when I first looked at the conclusion I said, what do these initials mean? Maybe not being a peptide chemist --

DR. HILL: There was a place where it became GHL instead of GHK, so I did flag that.

DR. MARKS: Okay, so --

DR. HILL: I mean, we --

DR. MARKS: Are you fine with the conclusion, with the initials in the way they are, guys? If you are, then --

DR. SHANK: I am.

DR. MARKS: You are? Okay, fine. So, ignore that comment.

MR. JOHNSON: Okay.

DR. HILL: I did have a couple of questions on page 38. These might be slightly rhetorical, but -- hang on a second. The comment I had in the document was, it's at PDF page 38, I need to be enlightened as to how 38 percent cell loss is declared not cytotoxic. They didn't do a dose response, so I was bugged by that, not in terms of changing the conclusion, and the other thing is --

MR. JOHNSON: Oh, excuse me, Dr. Hill, you mean the negligible cytotoxicity?

DR. HILL: Yeah, it said negligible cytotoxicity, I think, but then they said 38 percent cell loss, so I was puzzled how that could be declared not cytotoxic and maybe it relates to how they judge the (inaudible) controls. I'm not sure. But the way it's written sounds funny, so we might need to go back and look at the original reference and see.

MR. JOHNSON: Now, you said 38 percent cell loss, is that --

DR. HILL: That's why I'm trying to find out if I'm on the right page because --

MR. JOHNSON: Because I see 37.

DR. HILL: All right, well, 37 percent, maybe 38 -- page 38 and 37 percent? But the question is, how is that not cytotoxic? So, that was one question and then the other one was --

MR. JOHNSON: Because it said negligible cytotoxicity, is that --

DR. HILL: Yeah, how do you call 37 percent cell loss negligible cytotoxicity? I'm guessing it's because the controls also exhibit cell loss or something along those lines, but it just sounds really odd.

The other question I had in this was maybe more biologically significant. It talks about a beige color and I just wondered, from the dermatologists, was there any chance that that was indicative of increased melanin secretion.

MR. JOHNSON: What page are you on, doctor?

DR. HILL: It's also on page 38 and it is the second to last paragraph. So, it's the biopeptide CL study. It says, "A very slight beige coloration of the skin was observed in each animal." Which would not be irritation, of course, but my question was, is that indicative of an effect on melanin secretion? That's the only place I ran across that kind of effect in all of this.

DR. MARKS: I thought that was insignificant.

DR. HILL: Okay, well, I think everybody probably did, but I thought while I had a panel of dermatologists sitting here, I would ask.

DR. MARKS: Anything else?

MS. LORETZ: I have a question about the hexapeptide-12, so there's difference sequences but only the one was reviewed, and in the future, other sequences could be added that would fall under that name. Should that just be made really clear what CIR --

DR. HILL: I thought it was, isn't it?

DR. MARKS: I also thought it was clear, that's why the peptide sequence was identified.

DR. HILL: Somewhere there's language, but if it --

MS. LORETZ: I just wondered about, like, the naming, how they're named, because there is the potential for the future for more to be added.

DR. HILL: I think somewhere we said exactly this sequence and only this sequence the safety review pertains to and I think it was written in a way that -- did I get that somewhere?

DR. MARKS: If you look on page 49 --

DR. HILL: That's where I'm at.

DR. MARKS: -- at the conclusion in that paragraph at the top it says, "Noted, the safe conclusion applicable only to ingredient names associated with the following known peptide sequences." So, that's where it's being very specific. So, I guess if you came back with a different peptide sequence you would -- there better be safety to support that.

DR. HILL: And we know those are out there because we saw that in the last version of the document, but we didn't get safety data and it was asserted that there might be safety data, but we haven't seen it.

So, then we would have to reopen. Yes?

DR. MARKS: Yes. Absolutely. Does that answer your question?

MS. LORETZ: Yeah, I guess I was thinking more in the introduction it would be kind of nice to set that up rather than putting it at the end.

DR. MARKS: Again, that would be editorial, so, Wilbur, if you think that -- I mean, I have no problems with that. It's easy to repeat it again in the introduction, just it's, again, the sort of -- my -- in the conclusion, having the GHK and the VGBAPG -- it's, again, pretty obvious with that in there once you look at what those mean were very specific in terms of the sequencing.

DR. HELDRETH: It also is mentioned in the first sentence of the chemistry section. Specifically your hexapeptide-12 that we mean the sequence that's in the definition and not the other one.

DR. HILL: However, I mean, it would be editorial and almost inconsequential to add to that second sentence in the conclusion -- in the footnote. So, were ingredients in this group not -- seemed like what somebody said, add another footnote or something, I don't know.

It didn't jump out at her, it might not jump out at everybody that we're talking about exactly these sequences.

DR. MARKS: Okay. And then maybe add that in the introduction. Tom, Ron, Ron, do you feel strongly? Or do you think it's a good idea --

DR. HILL: I'll let everybody else hash it out at CIR staff.

DR. MARKS: Well, we need to give guidance. To me, repeating it causes no harm. It's like one more sentence.

MS. LORETZ: It's just an unusual naming convention, that's a little a different in that respect.

DR. SLAGA: That would be fine.

DR. MARKS: Yes. Okay, so, Wilbur, if you'd put it in the introduction and also to draw emphasis to this nuance.

Okay, so --

DR. SHANK: I have one comment -- comment on the discussion. Since the body of the report has some in vitro studies showing that some of the peptides induced angiogenesis, this is important in carcinogenesis, but we don't mention it in the discussion and I think it might be helpful if we add a sentence or two to the discussion saying that these peptides can induce angiogenesis in in vitro studies, but because these are low concentrations and are rapidly hydrolyzed in the plasma, there's little concern about promotion of skin tumors through cosmetic use.

DR. MARKS: That's sort of --

DR. SHANK: Just so it shows (inaudible) did consider it. I have the words (inaudible).

DR. MARKS: I think tomorrow, Ron, I'm going to ask you to make that comment.

DR. SHANK: Okay.

DR. MARKS: I think that's an important editorial comment. It doesn't change the conclusion, obviously, but --

DR. SHANK: It just shows we recognize that --

DR. SLAGA: Yeah, we had a lot of discussion about that.

DR. SHANK: Yes, we did.

DR. MARKS: Is that -- which do you prefer, Tom? Do you want to comment or Ron Shank, since Ron --

DR. SHANK: This is your --

DR. SLAGA: No, no, no.

DR. MARKS: Yeah, I know. That's why --

DR. SLAGA: We had extensive discussion last time about that.

DR. HILL: Well, and yeah, because I mean it's applicable in the skin, so your proviso about rapidly hydrolyzing the plasma, I don't know that that necessarily applies to things happening dermally, but then the flip side --

DR. SHANK: But the fact that it's used in such low concentration, we really didn't have any issues with the --

DR. HILL: That and also the fact that there are basically biochemical systems in place in the skin to keep that from becoming overactive. I mean, I think that is there in the report. So, I wasn't worried about it. I'm just saying, you raise it -- if you bring it up in the discussion, somebody could ask that question.

DR. SLAGA: It's already in the --

DR. HILL: I can give this to you.

DR. SLAGA: -- document, so -- I could go either way. It doesn't matter to me.

DR. SHANK: Okay. Well, if you don't need to add it.

DR. MARKS: What do you feel, Tom?

DR. SLAGA: I don't think we need it.

DR. MARKS: Okay.

DR. SLAGA: I mean, if we dismissed it because it's low concentration --

DR. MARKS: Okay. So, you don't feel we need it in the discussion and comment to that effect on angiogenesis. And that's fine. We haven't had it up to this point. I think your comments are straight on. The question is, does it need to be repeated or not?

DR. SHANK: I guess not. No.

DR. HILL: Well, you could bring it up tomorrow and see --

DR. MARKS: Well, no. If Tom and our team doesn't feel --

DR. SLAGA: Let me see -- presented?

DR. MARKS: I'm presenting. So, I'll be moving the final safe and then we'll ask for editorial comments. Tom and Ron, you'll have time to think about it more between now and tomorrow and Tom, if you feel the same, I'm not going to bring it up, Tom, since it sounds like now the way we've come to the conclusion it's adequately addressed in the document already, but we want to clarify it more tomorrow, Ron, we can get the reaction of the other team.

DR. SHANK: Fine.

DR. MARKS: Okay, any other comments? So, I will move tomorrow that a final report be issued with a safe conclusion for these peptides. So, we're down -- next ingredient is styrene. And this is the first time we've seen these ingredients.

Full Panel – June 10, 2014

DR. MARKS: So, we have the draft final report on these Peptides, Tripeptide-1, Heptapeptide-12, they are metal salts and fatty acid salt derivatives, and Palmitol Tetrapeptide-7, and we move a final report could be issued with these Peptides having a conclusion of safe in the present practice of use and concentration.

DR. BELSITO: Second.

DR. BERGFIELD: Any discussion or comment regarding this ingredient?

DR. BELSITO: We made a number of changes within the document, particularly Dan liked that table in the conclusion and recommended that it be brought up into the introduction so it was quite clear immediately that we're dealing with only specific Peptide sequences, but it was all editorial through the document and nothing major.

DR. BERGFIELD: Any other comments that you want to make.

DR. MARKS: Similar. We had editorial comments, but that's --

DR. BERGFIELD: Nothing changing anything.

DR. MARKS: No, nothing changing the conclusion.

DR. BERGFIELD: Dan, did you have a comment, or you just raised your hand out of fun? Okay. All right.

DR. MARKS: He's already voting.

DR. BERGFIELD: I'll call the question, all those in favor. Please indicate by raising your hands. Thank you, unanimous. Then moving on to the next ingredient, Dr. Belsito, the Pentaerythrityl.

Meeting Summary

Tripeptide-1, Hexapeptide-12, their Metal Salts and Fatty Acyl Derivatives, and Palmitoyl Tetrapeptide-7

The Panel issued a final safety assessment with the conclusion that the following 10 ingredients, identified as tripeptide-1, hexapapeptide-12, their metal salts and fatty acyl derivatives, and palmitoyl tetrapeptide-7, are safe in the present practices of use and concentration in cosmetics.

tripeptide-1
palmitoyl tripeptide-1
myristoyl tripeptide-1*
hexapeptide-12*
palmitoyl hexapeptide-12
myristoyl hexapeptide-12*
copper tripeptide-1
bis(tripeptide-1) copper acetate*
manganese tripeptide-1*
palmitoyl tetrapeptide-7

*Not reported to be in current use. Were ingredients in this group not in current use to be used in the future, the expectation is that they would be used in product categories and at concentrations comparable to others in this group.

This conclusion is applicable only to ingredients with peptide sequences that are defined as follows: tripeptide-1 (glycine-histidine-lysine), hexapeptide-12 (valine-glycine-valine-alanine-proline-glycine only), and tetrapeptide-7 (glycine-glutamine-proline-arginine). This assessment does not apply to the hexapeptide-12 (i.e., ala-pro-gly-val-gly-val) sequence listed in the INCI dictionary, because of the potential for major differences in chemistry and biological activity of some of the more complex groups attached to the peptide compared to those of the ingredients included in this safety assessment.

The peptides are used in cosmetic products at concentrations between 1 ppm and 30 ppm, and use at concentrations < 10 ppm is customary. However, data on the use concentrations of palmitoyl tetrapeptide-7 were not provided for this safety assessment. Given the high use frequency of use of palmitoyl tetrapeptide-7 reported to FDA, industry was urged to complete a use concentration survey for this ingredient.
Palmitoyl hexapeptide-12 is reported to function as an antioxidant in cosmetic products; the remaining 9 ingredients reportedly function as skin conditioning agents.

Safety Assessment of Myristoyl Pentapeptide-4, Palmitoyl Pentapeptide-4, and Pentapeptide-4 as Used in Cosmetics

Status: Draft Tentative Report for Panel Review
Release Date: March 4, 2024
Panel Meeting Date: March 28 - 29, 2024

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.; Allan E. Rettie, Ph.D.; David Ross, 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., and the Senior Director is Monice Fiume. This safety assessment was prepared by Preethi Raj, M.Sc., Senior Scientific Analyst/Writer, CIR.

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ABBREVIATIONS

ARE	antioxidant/electrophile response element
CAS	Chemical Abstracts Service
CIR	Cosmetic Ingredient Review
Council	Personal Care Products Council
CPSC	Consumer Product Safety Commission
DHT	5α-dihydrotestosterone
DMSO	dimethyl sulfoxide
DPBS	Dulbecco's phosphate buffer solution
DPRA	direct peptide reactivity assay
E2	17-β estradiol
EC_{10}	10% effect concentration
ECVAM DB-ALM	European Centre for Validation of Alternative Methods Database on Alternative Methods
FCA	Freund's complete adjuvant
Fmoc	fluorenylmethoxycarbonyl
Fmoc-Lys(Boc)-OH	N_α-fluorenylmethoxycarbonyl-N_ε-(*t*-butoxycarbonyl)-lysine
Fmoc-Ser(tBu)-OH	N_α-fluorenylmethoxycarbonyl-*O*-(*t*-butyl)-L-serine
Fmoc-Thr(tBu)-OH	N_α-fluorenylmethoxycarbonyl-*O*-(*t*-butyl)-L-threonine
FDA	Food and Drug Administration
GLP	good laboratory practices
HEPES	4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid
hERα	human estrogen receptor α
hAR	human androgen receptor
HET-CAM	hen's egg-chorioallantoic membrane
HRIPT	human repeated insult patch test
I_{max}	maximal response
ICH Q3C	International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use Guideline for Residual Solvents
KTTKS	lysine-threonine-threonine-lysine-serine; Pentapeptide-4
KTSKS	lysine-threonine-serine-lysine-serine; Pentapeptide-4
LC-MS/MS	liquid chromatography with tandem mass spectrophotometry
LoD	limit of detection
LOQ	limit of quantification
LPPS	liquid-phase peptide synthesis
MTT	3-[4,5-dimethylthiazol-2-yl]-2,5 diphenyl tetrazolium bromide
NR	none reported
OD	optical density
OECD	Organisation for Economic Cooperation and Development
Pal-KTTKS	Palmitoyl Pentapeptide-4
Panel	Expert Panel for Cosmetic Ingredient Safety
PBS	phosphate-buffered solution
PCI	primary cutaneous irritation
SDS	sodium dodecyl sulfate
SLS	sodium lauryl sulfate
SPPS	solid-phase peptide synthesis
TG	test guideline
US	United States
UVA/UVB	ultraviolet light A/ultraviolet light B
VCRP	Voluntary Cosmetic Registration Program
YAS	Yeast Androgen Screen
YES	Yeast Estrogen Screen
wINCI; *Dictionary*	web-based *International Cosmetic Ingredient Dictionary and Handbook*

DRAFT ABSTRACT

The Expert Panel for Cosmetic Ingredient Safety (Panel) assessed the safety of Myristoyl Pentapeptide-4, Palmitoyl Pentapeptide-4, and Pentapeptide-4, which are reported to function as skin-conditioning agents in cosmetic products. The Panel reviewed the available data to determine the safety of these ingredients and concluded…[to be determined.]

INTRODUCTION

This assessment reviews the safety of Myristoyl Pentapeptide-4, Palmitoyl Pentapeptide-4, and Pentapeptide-4 as used in cosmetic formulations. According to the web-based *International Cosmetic Ingredient Dictionary and Handbook* (wINCI *Dictionary*), these ingredients are reported to function in cosmetics as skin-conditioning agents (Table 1).[1]

The 3 ingredients included in this safety assessment are synthetic peptides which comprise a 5-amino-acid-sequence (pentapeptide) containing lysine, serine, and threonine. One such sequence is lysine-threonine-threonine-lysine-serine, also represented as Lys-Thr-Thr-Lys-Ser, or, KTTKS.[2] Myristoyl Pentapeptide-4 and Palmitoyl Pentapeptide-4 have an additional saturated fatty acid group attached to the peptide structure, namely myristic acid and palmitic acid, respectively. The amino acid sequence of the pentapeptide portion of these ingredients can vary; thus, data for two variations of Pentapeptide-4, namely, KTTKS and KTSKS (Lys-Thr-Ser-Lys-Ser), are included in this report.

The Panel has also previously reviewed the safety of the individual amino acids comprising these ingredients, as well as myristic acid and palmitic acid. In 2013, the Panel published a final report with the conclusion that α-amino acids are safe in the present practices of use and concentration in cosmetics as described in the safety assessment.[3] The safety of myristic acid and palmitic acid has been evaluated in several reviews.[4-7] Ultimately, in 2019, the Panel issued a final report on the safety of myristic acid and palmitic acid (as part of the safety assessment of fatty acids and fatty acid salts) with the conclusion that the ingredients are safe in cosmetics in the present practices of use and concentration described in the safety assessment when formulated to be non-irritating and non-sensitizing, which may be determined based on a quantitative risk assessment.[7]

This safety assessment includes relevant published and unpublished data that are available for each endpoint that is evaluated. Published data are identified by conducting an extensive search of the world's literature; a search was last conducted January 2024. A listing 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 provided on the Cosmetic Ingredient Review (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 are provided by the cosmetics industry, as well as by other interested parties.

CHEMISTRY

Definition and Structure

Pentapeptide-4 (CAS No. 149128-48-3) is a synthetic peptide comprised of the amino acids, lysine, serine, and threonine (forming a pentapeptide), in either the lysine-threonine-threonine-lysine-serine (also represented as Lys-Thr-Thr-Lys-Ser; i.e, KTTKS) or, lysine-threonine-serine-lysine-serine (also represented as Lys-Thr-Ser-Lys-Ser, i.e. KTSKS) sequence (Figure 1).[1,2] Myristoyl Pentapeptide-4 (CAS No. 1392416-25-9) and Palmitoyl Pentapeptide-4 (CAS No. 521091-64-5; 214047-00-4) each have a myristic acid or palmitic acid group, respectively, attached to the *N*-capped end of this sequence. The definitions and structures of the ingredients included in this review are provided in Table 1.

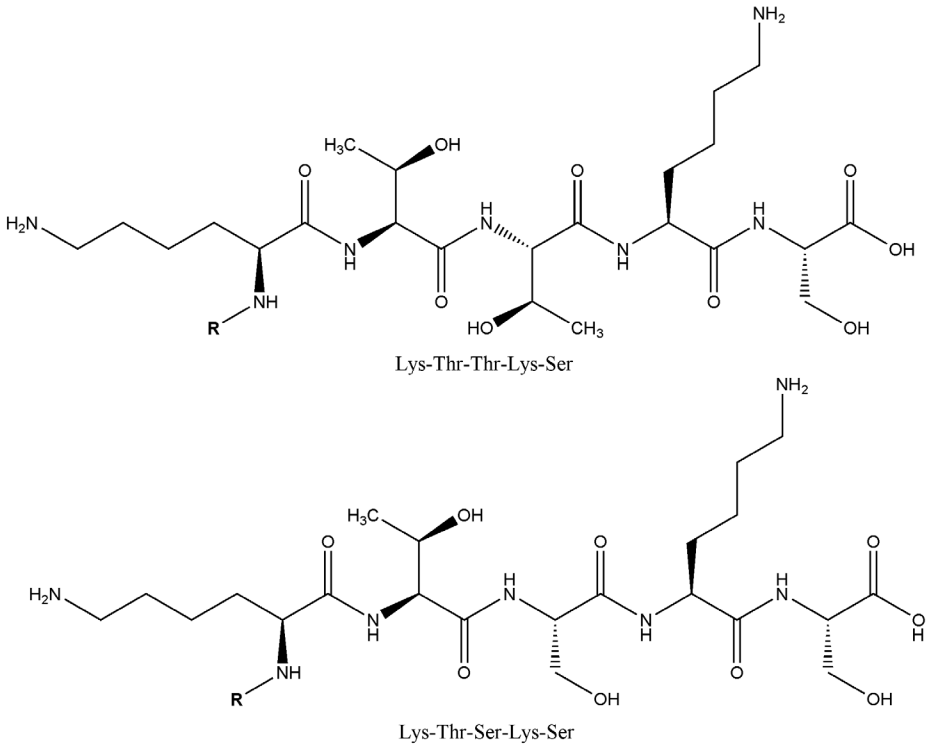


Figure 1. Pentapeptide-4 (when R is hydrogen) and *N*-capped derivatives (when R is the residue of myristic or palmitic acid)

Pentapeptide-4 is a subfragment of type I collagen propeptide, and is regarded as a signal peptide and a matrikine, which possesses the ability to enhance dermal remodeling by triggering cellular processes, such as inhibiting collagenase activity and increasing extracellular matrix production.[2,8-11] The hydrophilic and charged nature of Pentapeptide-4 makes it difficult for it to pass through the intact stratum corneum.[12] However, through the attachment of a fatty acid, such as palmitic acid, which has a 16-carbon chain, the peptide is rendered more lipophilic and is more easily able to penetrate into the skin.[13]

Chemical Properties

Myristoyl Pentapeptide-4, Palmitoyl Pentapeptide-4, and Pentapeptide-4 have molecular weights of 774 g/mol,[14] 802.1 g/mol,[15,16] and 563.6 g/mol,[17] respectively. Additionally, these ingredients have the following predicted log p values for the KTTKS and KTSKS sequences, respectively: Myristoyl Pentapeptide-4 (1.85; 1.6), Palmitoyl Pentapeptide-4 (2.72; 2.52) , and Pentapeptide-4 (-4.12; -4.39).[18] Chemical properties for ingredients in this report are further outlined in Table 2.

Method of Manufacture

Palmitoyl Pentapeptide-4

Two samples of Palmitoyl Pentapeptide-4 (Pal-KTTKS and Pal-KTSKS) are described by a supplier as being obtained via solid phase synthesis at room temperature using Fmoc-amino acid derivatives.[19] An N_{α}-fluorenylmethoxycarbonyl-N_{ε}-(*t*-butoxycarbonyl)-lysine (Fmoc-Lys(Boc)-OH) complex is first activated with a coupling agent and reacted on serine-protected resin. Deprotection of the Fmoc residue with a base produces a dipeptide on the resin. For the Pal-KTTKS sequence, both activation and coupling are achieved using the N_{α}-fluorenylmethoxycarbonyl-*O*-(*t*-butyl)-L-threonine (Fmoc-Thr(tBu)-OH) complex, and deprotection is achieved with the Fmoc-Lys(Boc)-OH group. For the Pal-KTSKS sequence, the N_{α}-fluorenyl-methoxycarbonyl-*O*-(*t*-butyl)-L-serine (Fmoc-Ser(tBu)-OH), Fmoc-Thr(tBu)-OH, and Fmoc-Lys(Boc)-OH groups are utilized for activation, coupling, and deprotection, respectively. After the last Fmoc-deprotection step, palmitic acid is reacted in the same manner in each process and the resulting products are fully deprotected and purified to yield the final amino acid sequences (Pal-Lys-Thr-Thr-Lys-Ser-OH and Pal-Lys-Thr-Ser-Lys-Ser-OH).

Impurities

Palmitoyl Pentapeptide-4

The impurities found in a sample of Palmitoyl Pentapeptide-4 (Pal-KTTKS), as described by a supplier, were: acetate (< 10%), palmitic acid (< 5%), water (< 5%), and residual solvents (in accordance with the International Council for Harmonisation Of Technical Requirements for Pharmaceuticals for Human Use Guideline for Residual Solvents (ICH Q3C)).[15] Two distinct samples of Palmitoyl Pentapeptide-4, each comprising the Pal-KTTKS or Pal-KTSKS sequence, were described by a supplier as having ≥ 90% purity at 210 nm.[19] The supplier described the impurities in the first sample of Palmitoyl Pentapeptide-4 (Pal-KTTKS) as stereoisomers of Pal-KTTKS-OH, myristine-lysine-threonine-threonine-lysine-serine-OH, and stearyl-lysine-threonine-threonine-lysine-serine-OH. The impurities in the second Palmitoyl Pentapeptide-4

sample (Pal-KTSKS) were described by the supplier as stereoisomers of Pal-KTSKS-OH, myristine-lysine-threonine-serine-lysine-serine-OH, and stearyl-lysine-threonine-serine-lysine-serine-OH.

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.

According to 2023 VCRP survey data, Palmitoyl Pentapeptide-4 has the greatest reported frequency of use; it is reported to be used in 239 formulations, 223 of which are leave-on products (Table 3).[20] Myristoyl Pentapeptide-4 is reported to have 4 uses, while Pentapeptide-4 has 1 reported use. The results of the concentration of use survey conducted by the Council in 2022, and revised in 2023, indicate Palmitoyl Pentapeptide-4 has the highest maximum reported concentration of use, at up to 0.0035% in hair conditioners.[21] The highest leave-on maximum concentration of use reported is 0.0012% Palmitoyl Pentapeptide-4 in face and neck preparations. Concentration of use data were not reported for the other 2 ingredients.

Some of these ingredients are reported to be used in products that are applied near the eye; Palmitoyl Pentapeptide-4 is used at up to 0.0012% in eye lotions. Palmitoyl Pentapeptide-4 is reported to be used in a face powder (concentration not provided) and could possibly be inhaled. 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 cosmetics 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. 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 Pentapeptide-4 ingredients named in the report are not restricted from use in any way under the rules governing cosmetic products in the European Union.[22]

Non-Cosmetic

Palmitoyl Pentapeptide-4 (Pal-KTTKS) has been tested for its wound-healing effects.[23] Palmitoyl Pentapeptide-4 applied in a patch (0.1 and 1 mg) and cream (1 mg) form had a larger impact on wound healing in animals, compared to negative controls (untreated) and positive controls (ready-to-wear dressing; $p < 0.05$).

TOXICOKINETIC STUDIES

Dermal Penetration

In Vitro

Palmitoyl Pentapeptide-4; Pentapeptide-4

The permeability of Palmitoyl Pentapeptide-4 (Pal-KTTKS) and Pentapeptide-4 (KTTKS) was evaluated in an in vitro study using 3 replicate skin samples of CrlOri: SKH1-hr strain hairless mice.[24] Intact hairless mouse skin was mounted on Franz diffusion cells with the epidermal side facing the donor compartment. In the receptor compartment, 4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid (HEPES) buffer was mixed with 15% ethanol containing phenylmethane-sulfonylfluoride and 1,10-phenanthroline at final concentrations of 5 and 1 mM, respectively, as proteolytic enzyme inhibitors. The donor compartment was loaded with a 1 ml of Palmitoyl Pentapeptide-4 or Pentapeptide-4 (100 µg/ml in 15% ethanol) solution. After 24-h incubation, the skin was removed from the diffusion cell and the remaining donor solution on the skin surface was washed 4 times with 1 ml of distilled water. Upon drying, separation, and mincing of the skin layers (stratum corneum, epidermis, and dermis), the amount of Palmitoyl Pentapeptide-4 or Pentapeptide-4 distributed in each skin layer was extracted using 1 ml of methanol for 24 h with continuous shaking. The extracted samples were centrifuged and

the supernatants were analyzed using liquid chromatography with tandem mass spectrometry (LC-MS/MS). No detectable level of Pentapeptide-4 was observed in the receptor solution over an observation period of 48 h. A trace amount of Palmitoyl Pentapeptide-4 was detected in the receptor solution after 24 h by LC-MS/MS; however, it was below the limit of quantification (LOQ; < 0.5 µg/ml). No amount of Pentapeptide-4 was detected in any of the skin layers over a period of 24 h. Palmitoyl Pentapeptide-4 was observed in every skin layer: 4.2 ± 0.7 µg/cm^2 in the stratum corneum, 2.8 ± 0.5 µg/cm^2 in the epidermis, and 0.3 ± 0.1 µg/cm^2 in the dermis. Overall, 14.6% of the applied Palmitoyl Pentapeptide-4 was retained in the skin: 8.3% in the stratum corneum, 5.6% in the epidermis, and 0.6% in the dermis. Therefore, the researchers concluded that neither Palmitoyl Pentapeptide-4 nor Pentapeptide-4 could permeate through full-thickness hairless mouse skin over the time period used in these experiments.

Absorption, Distribution, Metabolism, and Excretion (ADME)

In Vitro

Palmitoyl Pentapeptide-4; Pentapeptide-4

The dermal stability of Palmitoyl-Pentapeptide-4 (Pal-KTTKS) and Pentapeptide-4 (KTTKS) was evaluated in vitro in epidermal and dermal skin extracts and whole skin homogenate prepared from hairless mouse skin.[24] Pentapeptide-4 (200 µl) or Palmitoyl Pentapeptide-4 (40 µg/ml in 10 mM HEPES buffer, pH 7.4, as peptide concentration) was incubated with 200 µl of the epidermal skin extract, dermal skin extract, or whole skin homogenates at 37 °C for 120 min. At predetermined times, the amount of Palmitoyl Pentapeptide-4 and Pentapeptide-4 present in the incubated mixtures was sampled and analyzed by LC-MS/MS. Pentapeptide-4 was almost fully degraded in the dermal skin extract and whole skin homogenate, with 3.2% remaining in the dermal skin extract at 30 min and 1.5% remaining in the whole skin homogenate at 60 min. The degradation of Pentapeptide-4 in the epidermal skin extract was slower than that seen in the dermal skin extract and whole skin homogenate, which was potentially attributed to lower amounts of proteolytic enzymes. Palmitoyl Pentapeptide-4 was more stable in the skin extracts over time, compared to Pentapeptide-4. The concentration of Palmitoyl Pentapeptide-4 detected in the epidermal skin extract after 120 min was similar to the initial concentration. After 60 min, 11.2% Palmitoyl Pentapeptide-4 remained in the whole skin homogenate, and, after 120 min, 9.7% Palmitoyl Pentapeptide-4 remained in the dermal extract.

TOXICOLOGICAL STUDIES

Acute Toxicity Studies

Oral

Palmitoyl Pentapeptide-4

The acute oral toxicity of Palmitoyl Pentapeptide-4 (Pal-KTTKS), tested at 0.01% (vehicle not specified), was evaluated in Sprague-Dawley rats (5/sex), in accordance with Organisation for Economic Cooperation and Development (OECD) test guideline (TG) 401.[15,25] A single dose of the test substance (20 ml/kg) was administered via gavage. Mortality, clinical abnormalities, and body weight gain were monitored for a period of up to 14 d; all animals were killed at the end of the study. No deaths occurred during the study and no apparent changes or abnormalities were observed in general behavior, body weight gain, or upon necropsy.

Short-Term Toxicity Studies

Dermal

Palmitoyl Pentapeptide-4

Groups of guinea pigs (5/sex; strain not specified) were treated with 0.01% Palmitoyl Pentapeptide (0.05 ml; vehicle not specified; Pal-KTTKS) in a 2-wk dermal irritation study.[15,26] No deaths or clinical signs related to treatment were noted during the study; internal organs were not examined. No further details were provided.

Subchronic, and Chronic Toxicity Studies

No subchronic or chronic toxicity studies were found in the published literature, and unpublished data were not submitted.

DEVELOPMENTAL AND REPRODUCTIVE TOXICITY STUDIES

No developmental and reproductive toxicity studies were found in the published literature, and unpublished data were not submitted.

GENOTOXICITY STUDIES

Details of the in vitro genotoxicity studies summarized below are provided in Table 4.

A solution of 0.5% Palmitoyl Pentapeptide-4 (Pal-KTTKS) in distilled water and ethanol (75/25), tested at 2% in distilled water, was not mutagenic in an Ames test at concentrations up to 5000 µg/plate using *Salmonella typhimurium* strains TA98, TA100, TA1535, TA1537 and *Escherichia coli* WP2*uvr*A.[15,27] In another Ames test, performed in accordance with OECD TG 471, Palmitoyl Pentapeptide-4 (81.6% pure, Pal-KTSKS) in dimethyl sulfoxide (DMSO) was not mutagenic when tested at concentrations up to 5000 µg/plate using *S. typhimurium* strains TA98, TA100, TA102, TA1535, and TA1537,

with or without metabolic activation; signs of cytotoxic activity were observed under test conditions.[19,28] The genotoxic potential of Palmitoyl Pentapeptide-4 (> 96% pure, Pal-KTSKS) in water was evaluated in an in vitro mammalian cell micronucleus test in accordance with OECD TG 487 using cultured human lymphocytes.[19,29] Cells were treated with 250, 500, or 1000 µg/ml of the test article in the presence of metabolic activation for 4-h, followed by a 24-h recovery period; cells were also treated with 375, 500, or 750 µg/ml of the test article in the absence of metabolic activation for 4 h, followed by a 24-h recovery period (short treatment). In an additional assay, cells were treated with concentrations of 250, 320, or 400 µg/ml Palmitoyl Pentapeptide-4 for 24 h without a recovery period (continuous treatment). Neither statistically nor biologically significant increases in the number of micronucleated cells were observed with either treatment period; the test article was deemed not genotoxic.

CARCINOGENICITY STUDIES

No carcinogenicity studies were found in the published literature, and unpublished data were not submitted.

OTHER RELEVANT STUDIES

Endocrine Activity

Palmitoyl Pentapeptide-4

The estrogenic and androgenic activity of a formulation containing 0.12% Palmitoyl Pentapeptide-4 (other contents not specified; Pal-KTSKS) was evaluated in transformed yeast cells using the XenoScreen Yeast Estrogen Screen (YES) and Yeast Androgen Screen (YAS) assays.[19,30] *Saccharomyces cerevisiae* cells were genetically transformed with human estrogen receptor α (hERα) and human androgen receptors (hAR) and, additionally, had an expression plasmid carrying the reporter gene lacZ inserted. Binding of the test article with hERα or hAR receptors resulted in the interaction of these receptors with the corresponding response elements on the expression plasmid, in turn affecting β-galactosidase gene expression. Thus, the amount of secreted β-galactosidase, which was correlated with colorimetric quantification of the conversion of the yellow substrate, chlorophenol red-β-D-galactopyranoside, into a red product at 570 nm (corrected for unspecific absorption and light scattering at 690 nm), indicated the estrogenic or androgenic activity of the test article. The difference between these optical density (OD) absorbance values (OD_{690} - OD_{570}) was used to calculate growth factor values and induction ratios. Eight serial dilutions of the test article (half-log steps) in DMSO, resulting in final concentrations of 1 x 10^{-2} – 3.16 x 10^{-6} M, were added to yeast cells in the agonist assays. For the agonist YES assay, 17-β estradiol (E2) was used as the positive control at 7 final concentrations between 1 x 10^{-11} – 1 x 10^{-8} M; 5α-dihydrotestosterone (DHT) was used as the positive control for the agonist YAS assay at 7 final concentrations between 1 x 10^{-9} – 1 x 10^{-6} M, using half-log dilution steps. DMSO (1%) was used as the solvent control. The inhibitory activity of the test article dilutions were evaluated in the presence of E2 (1.3 x 10^{-9} M) in an antagonistic YES assay and in the presence of DHT (3 x 10^{-8} M) in an antagonistic YAS assay. Serial dilutions of 4-hydroxytamoxifen and flutamide were used as antagonist positive controls. The test article exhibited cellular toxicity (growth factors ≤ 0.5) at the two highest tested concentrations and estrogenic activity with a 10% effect concentration (EC_{10}) value of 6.9 x 10^{-3} M in the YES agonist assay. No estrogenic antagonist or androgenic agonist/antagonist activities were observed.

Similarly, the estrogen agonist effects of a formulation containing 0.12% Palmitoyl Pentapeptide-4 (other contents not specified; Pal-KTSKS), were assessed in a XenoScreen XL YES assay.[31] Lyticase and a detergent were used to facilitate the secretion of the intracellularly synthesized β-galactosidase. Test article samples were serially diluted in 8 steps (half-log steps) in water with 1% DMSO, with concentrations ranging from 5.21 x 10^{-5} – 6.7 x 10^{-3} M. E2 was used as the positive control in 8 final concentrations between 2.1 x 10^{-12} – 6.7 x 10^{-9} M, using half-log dilution steps; 1% DMSO served as the solvent control. The limit of detection (LoD) for estrogenic activity was 1.49 x 10^{-11} M E2. No inhibition of cellular growth or estrogenic agonist activity was observed at any concentration tested.

DERMAL IRRITATION AND SENSITIZATION STUDIES

Details on the dermal irritation and sensitization data summarized below can be found in Table 5.

A formulation containing 0.12% Palmitoyl Pentapeptide-4 (tested as supplied; Pal-KTSKS) did not cause irritation when applied to a reconstructed human epidermis model (EpiSkin®) in a cutaneous primary irritation test performed in accordance with OECD TG 439.[19,32] Palmitoyl Pentapeptide-4, tested at 0.01% (vehicle not specified; Pal-KTTKS), was not irritating in an acute dermal irritation test performed in accordance with OECD TG 404 using New Zealand white rabbits nor in a 2-wk dermal irritation study performed in accordance with OECD TG 404 using guinea pigs.[15,26,33] A trade name mixture containing 0.01% Palmitoyl Pentapeptide-4 (applied neat; Pal-KTTKS) was tested for acute skin irritation using 10 subjects.[15,34] Very slight erythema was observed in 1 of the subjects and the primary cutaneous irritation (PCI) score was determined to be 0.10. The test substance was considered to be well-tolerated. A formulation containing 0.12% Palmitoyl Pentapeptide-4 (tested at 15% in distilled water; Pal-KTSKS) was not irritating when applied for 48 h, under semi-occlusive conditions in a patch test using 11 subjects.[19,35]

Palmitoyl Pentapeptide-4 (81.6% pure, Pal-KTSKS) was predicted to be non-sensitizing when tested at 5 mM (5 µl) and 25 mM (250 µl) in water in a direct peptide reactivity assay (DPRA) performed in accordance with OECD TG 442C.[19,36]

Palmitoyl Pentapeptide-4 (81.6% pure; Pal-KTSKS) was tested at up to 200 µM (0.05 ml) in DMSO using the KeratinoSens™ cell line in an antioxidant/electrophile response element (ARE)-Nrf2 luciferase assay, performed in accordance with OECD TG 442D.[19,37] The test article yielded a maximal response value (I_{max}) of 1.35 compared to an I_{max} of 5.12 for the positive control, cinnamaldehyde; the test article was predicted to be non-sensitizing. A guinea pig maximization test was performed in accordance with OECD TG 406, to evaluate the sensitization potential of Palmitoyl Pentapeptide-4 (0.01%; Pal-KTTKS).[15,38] Thirty guinea pigs (test animals: 10/sex; controls: 5/sex), received the test substance at an effective concentration of 0.0075% (w/w; in saline) followed by an undiluted epicutaneous application during induction, and a dermal application of the test substance at an effective concentration of 0.0025%, in saline, during challenge. No skin reactions were observed during evaluation of the test sites 24 and 48 h after patch removal; the test substance was deemed non-sensitizing. A formulation containing 0.12% Palmitoyl Pentapeptide-4 (tested at 15% in distilled water; Pal-KTSKS) was not irritating or sensitizing when applied under semi-occlusive conditions in a human repeated insult patch test (HRIPT) using 106 subjects.[19,39] The undiluted application of a trade name mixture containing 0.01% Palmitoyl Pentapeptide-4 (Pal-KTTKS) to a 3.61 cm^2 area, resulting in 5.54 $\mu g/cm^2$ applied Palmitoyl Pentapeptide-4, did not cause irritation or sensitization in an occlusive HRIPT using 51 subjects.[15,40,41]

Phototoxicity Studies

Palmitoyl Pentapeptide-4

The potential for a sample of Palmitoyl Pentapeptide-4 (tested at 0.0015%; Pal-KTSKS), in water, to absorb ultraviolet light A (UVA) and ultraviolet light B (UVB) was evaluated, in accordance with OECD TG 101.[19,42] The diluted article (1 ml) was placed in a calibrated spectrophotometer in order to read UVA/UVB absorption. No absorbance peak was observed between 290 and 400 nm, which was suggestive of a molar extinction coefficient (ε; a measure of how strongly a chemical species or substance absorbs light at a particular wavelength; is an intrinsic property of chemical species that is dependent on structure) < 1000 M^{-1} cm^{-1}. The test article was predicted to be non-phototoxic.

OCULAR IRRITATION STUDIES

Details on the ocular irritation studies summarized below can be found in Table 6.

A formulation containing 0.12% Palmitoyl Pentapeptide-4 (300 µl dose; Pal-KTSKS) was tested in an in vitro hens egg-chorioallantoic membrane (HET-CAM) assay, performed in agreement with French Good Laboratory Practices (GLP) and the European Directive 2004/10/EC.[19,43] The mean score calculated for hyperemia, hemorrhage, and coagulation, opacity, and/or thrombosis was 4.25; the test article was classified as slightly irritating. In another HET-CAM assay, a trade name mixture containing 0.01% Palmitoyl Pentapeptide-4 (Pal-KTTKS), which was tested as supplied, produced a mean irritation index of 6.0; the mean irritation index of the positive control, sodium dodecyl sulfate, was 12.0.[15,34] The test article was classified as moderately irritating. The ocular irritation potential of a formulation containing 0.12% Palmitoyl Pentapeptide-4 (tested at 30% in glycerin and water; Pal-KTSKS) was tested in a SkinEthic™ human corneal epithelial model, in accordance with OECD TG 492.[19,44] Mean cell viability when tested with the test article was 104.3%; the test article was considered not irritating. Palmitoyl Pentapeptide-4 tested at 0.01% (vehicle not specified; Pal-KTTKS) was assessed for ocular irritation in 3 male New Zealand white rabbits, in accordance with OECD TG 405.[15,45] A single dose of 0.1 ml was instilled into the conjunctival sac of the left eye, and the eye was not rinsed. All mean values for chemosis, redness of the conjunctiva, iris lesions, and corneal opacity were 0 at each tested time interval. The test substance was deemed non-irritating to rabbit eyes under the conditions of this study.

CLINICAL STUDIES

Use Studies

Palmitoyl Pentapeptide-4 has been tested in several clinical studies for its use as an anti-wrinkle agent. A moisturizer containing 3 ppm Palmitoyl Pentapeptide-4 was well tolerated in a 12-wk, double-blind, placebo-controlled, split face, left-right randomized clinical study performed in 93 female subjects.[46] In an 8-wk, randomized parallel-group study conducted in 196 women, a cosmetic product regimen containing niacinamide, Palmitoyl Pentapeptide-4, palmitoyl-lysine-threonine, retinyl propionate, and carnosine in a moisturizing base was well tolerated compared to a moisturizer containing 0.02% tretinoin;[47] although the concentration of Palmitoyl Pentapeptide-4 in the moisturizing base is not provided, it was reported to not exceed the maximum reported concentration of use of this ingredient in non-spray face and neck products that was reported to the Council in response to the use survey (i.e., 0.0012%).[48] Palmitoyl Pentapeptide-4 was also well tolerated in another 8-wk, double-blind randomized trial evaluating the effectiveness of 3 cream formulations containing either acetylhexapeptide-3, Pentapeptide-4, or placebo (concentrations not provided).[49]

SUMMARY

This assessment reviews the safety of Myristoyl Pentapeptide-4, Palmitoyl Pentapeptide-4, and Pentapeptide-4 as used in cosmetic formulations. These 3 synthetic peptides are comprised of a varied 5-amino-acid-sequence containing lysine, threonine, and serine; this report reviews the safety of two sequences, namely Pal-KTTKS and Pal-KTSKS. According to the *Dictionary*, these ingredients are reported to function in cosmetics as skin-conditioning agents. As reported in 2023 VCRP data, Palmitoyl Pentapeptide-4 is used in 239 formulations. Palmitoyl Pentapeptide-4 had the highest maximum

concentration of use reported in response to a 2022 concentration of use survey; it is used at up to 0.0035% in hair conditioners.

The permeability of Palmitoyl Pentapeptide-4 (Pal-KTTKS) and Pentapeptide-4 (KTTKS) was evaluated in an in vitro study using hairless mice skin. Either 1 ml of Palmitoyl Pentapeptide-4 or Pentapeptide-4 was incubated with skin samples for 24 h; the amount of each substance distributed in each skin layer was extracted using methanol and analyzed using LC-MS/MS. Pentapeptide-4 was not detected in the receptor solution after an observation period of 48 h; a trace amount of Palmitoyl Pentapeptide-4 was detected after 24 h, but it was below the LOQ at < 0.5 µg/ml. No amount of Pentapeptide-4 was detected in any of the skin layers over a period of 24 h. Palmitoyl Pentapeptide-4 was observed in every skin layer at: 4.2 ± 0.7 µg/cm^2 in the stratum corneum, 2.8 ± 0.5 µg/cm^2 in the epidermis, and 0.3 ± 0.1 µg/cm^2 in the dermis. Overall, 14.6% of the applied Palmitoyl Pentapeptide-4 was retained in the skin: 8.3% in the stratum corneum, 5.6% in the epidermis, and 0.6% in the dermis. The researchers concluded that Palmitoyl Pentapeptide-4 and Pentapeptide-4 did not permeate through full-thickness mouse skin.

The in vitro dermal stability of Palmitoyl Pentapeptide-4 and Pentapeptide-4 was evaluated in several mouse skin extracts. Either 200 µl Pentapeptide-4 or 40 µg/ml Palmitoyl Pentapeptide-4 (in 10 mM HEPES buffer) was incubated with 200 µl of the epidermal skin extract, dermal skin extract, or whole skin homogenates at 37 °C for 120 min. The amounts of each substance present in the incubated mixtures were sampled and analyzed by LC-MS/MS. Pentapeptide-4 was almost fully degraded in the dermal skin extract and whole skin homogenate, with 3.2% remaining in the dermal skin extract at 30 min and 1.5% remaining in the whole skin homogenate at 60 min. Pentapeptide-4 degradation was slower in the epidermal skin extract which was attributed to lower amounts of proteolytic enzymes. Palmitoyl Pentapeptide-4 was more stable in the skin extracts over time; the amount detected in the epidermal skin extract after 120 min was similar to the initial concentration. After 60 min, 11.2% Palmitoyl Pentapeptide-4 remained in the whole skin homogenate and after 120 min, 9.7% Palmitoyl Pentapeptide-4 remained in the dermal extract.

In an acute oral toxicity study, performed in accordance with OECD TG 401, groups of Sprague-Dawley rats (5/sex) received a single dose of Palmitoyl Pentapeptide-4 (20 ml/kg; Pal-KTTKS), tested at 0.01%, via gavage. No deaths occurred during the study and no abnormalities were observed in the general behavior, body weight gain, or upon necropsy. No deaths or clinical signs related to treatment were noted in groups of guinea pigs (5/sex) treated with 0.01% Palmitoyl Pentapeptide (0.05 ml) in a 2-wk dermal irritation study.

A solution of 0.5% Palmitoyl Pentapeptide-4 (Pal-KTTKS) in distilled water and ethanol (75/25), tested at 2% in distilled water, was not mutagenic at up to 5000 µg/plate, with or without metabolic activation using *S. typhimurium* strains TA98, TA100, TA1535, TA1537 and *E. coli* WP2*uvr*A. Palmitoyl Pentapeptide-4 (Pal-KTSKS) in DMSO was not mutagenic to *S. typhimurium* strains TA98, TA100, TA1535, and TA1537, with or without metabolic activation, in another Ames test performed in accordance with OECD TG 471; signs of cytotoxic activity were observed under test conditions. In an in vitro mammalian cell micronucleus test, performed in accordance with OECD TG 487, cultured human lymphocytes were treated for 4 h with up to 1000 µg/ml Palmitoyl Pentapeptide-4 (Pal-KTSKS) in the presence of metabolic activation (24-h recovery), and for 4 h with up to 750 µg/ml Palmitoyl Pentapeptide-4 in the absence of metabolic activation (24-h recovery). Additionally, cells were treated continuously for 24 h (without a recovery period), in the absence of metabolic activation, with up to 400 µg/ml Palmitoyl Pentapeptide-4. Neither statistically nor biologically significant increases in the number of micronucleated cells were observed with the short-term or continuous treatments; the test article was deemed non-genotoxic.

When tested in XenoScreen YES and YAS agonist and antagonist assays, a formulation containing 0.12% Palmitoyl Pentapeptide-4 (Pal-KTSKS), exhibited cellular toxicity (growth factors ≤ 0.5) at the two highest concentrations tested and estrogenic activity with a EC_{10} value of 6.9×10^{-3} in the YES agonist assay; no estrogenic antagonist, or androgen agonist/antagonist activities were observed. The same test article did not exhibit inhibition of cellular growth or estrogen agonist activity at any concentration tested in another Xenoscreen XL YES assay; the LoD for estrogenic activity was 1.49×10^{-11} M E2 .

A formulation containing 0.12% Palmitoyl Pentapeptide-4 in glycerin and water (tested as supplied; Pal-KTSKS) was not irritating to an EpiSkin® model in a cutaneous primary irritation test performed in accordance with OECD TG 439. Palmitoyl Pentapeptide-4, tested at 0.01% (Pal-KTTKS), was not irritating to rabbit skin in an acute dermal irritation study, nor was it irritating to guinea pig skin in a 2-wk dermal irritation study. In a clinical acute irritation study using 10 subjects, a trade name mixture containing Palmitoyl Pentapeptide-4 (0.01%) was well tolerated; very slight erythema was seen in 1 of the subjects, and the PCI was 0.10. A formulation containing 0.12% Palmitoyl Pentapeptide-4 (in distilled water; Pal-KTSKS) was not irritating in a human patch test using 11 subjects.

Palmitoyl Pentapeptide-4 (81.6% pure; Pal-KTSKS) was predicted to be non-sensitizing when tested in a DPRA (OECD TG 442C) and a ARE-Nrf2 luciferase assay (OECD 442D). In a guinea pig maximization test, Palmitoyl Pentapeptide-4 (0.01%; Pal-KTTKS) was not sensitizing when injected at effective test concentrations of 0.0075% in saline during intradermal induction, applied at 0.01% during epicutaneous induction, and applied at 0.0025% in saline during challenge. A formulation containing 0.12% Palmitoyl Pentapeptide-4 (tested at 15% in distilled water; Pal-KTSKS) was not irritating or sensitizing when tested under semi-occlusive conditions in an HRIPT using 106 subjects. No irritation or

sensitization was observed in an occlusive HRIPT in which 51 subjects were treated with a trade name mixture containing 0.01% Palmitoyl Pentapeptide-4.

The potential for a sample of Palmitoyl Pentapeptide-4 (tested at 0.0015%; Pal-KTSKS) to cause phototoxicity was evaluated in an UVA/UVB spectrum test performed in accordance with OECD TG 101. No absorbance peak was observed between 290 and 400 nm, which was suggestive of a molar extinction coefficient < 1000 M^{-1} cm^{-1}; the test article was predicted to be non-phototoxic.

A formulation containing 0.12% Palmitoyl Pentapeptide-4 (300 µl dose; Pal-KTSKS) yielded a mean irritation score of 4.25 when tested in a HET-CAM assay and was classified as slightly irritating. Similarly, the ocular irritation potential of a trade name mixture containing 0.01% Palmitoyl Pentapeptide-4 (tested as supplied) was evaluated in another HET-CAM assay. The mean irritation index for the test substance, when tested as supplied, was 6.0, compared to a score of 12.0 for the positive control, sodium dodecyl sulfate. Thus, the test substance was classified as a moderate ocular irritant. Mean cell viability of a SkinEthic™ human corneal epithelial model when tested with a formulation containing 0.12% Palmitoyl Pentapeptide-4 (in glycerin and water; Pal-KTSKS) was 104.3%; the test article was considered non-irritating. In an acute ocular irritation study, a single, 0.1 ml dose of Palmitoyl Pentapeptide-4 tested at 0.01% (vehicle not specified; Pal-KTTKS) was not irritating to New Zealand white rabbit eyes.

Clinically, a moisturizer containing 3 ppm Palmitoyl Pentapeptide-4 was well-tolerated in a 12-wk, double blind placebo-controlled, split face, left-right randomized clinical study performed in 93 female subjects. Palmitoyl Pentapeptide-4 has also been shown to be well tolerated in other randomized trials where it was tested in cosmetic formulations (concentration did not exceed the maximum reported concentration of use in face and neck products).

DRAFT DISCUSSION

[Note: This Discussion is in the draft form, and changes will be made following the Panel meeting.]

This assessment reviews the safety of Myristoyl Pentapeptide-4, Palmitoyl Pentapeptide-4, and Pentapeptide-4 as used in cosmetic formulations. The Panel concluded [TBD].

The Panel considered their previous safety review of the individual amino acids comprising these ingredients, as well as myristic acid and palmitic acid; the Panel also considered the available method of manufacturing and impurities data for Palmitoyl Pentapeptide-4. The Panel sought clarification of the concentration at which a mixture containing Palmitoyl Pentapeptide-4 (Pal-KTTKS) was tested in an HRIPT; upon receiving information about the applied dose of Palmitoyl Pentapeptide-4 (5.54 µg/cm^2), the Panel noted the absence of dermal irritation and sensitization in the HRIPT and the other available data. Additionally, the Panel noted some changes in the keratin profile of subjects treated with a facial cream containing Palmitoyl Pentapeptide-4, suggesting a potential biologic effect; however, these were not considered adverse effects based upon the lack of erythema, dryness, and transepidermal water loss. The Panel also considered the low reported maximum concentration of use for these ingredients (Palmitoyl Pentapeptide-4 at 0.0035% in hair conditioners and 0.0012% in face and neck preparations) and that in vitro data evidenced limited percutaneous absorption in the skin. Furthermore, the Panel discussed the available genotoxicity data and the absence of endocrine disruption at a concentration of 0.12%, which obviated the need for carcinogenicity and developmental and reproductive toxicity data, respectively.

The Panel also discussed the issue of incidental inhalation exposure resulting from these ingredients; for example, Palmitoyl Pentapeptide-4 is reported to be used in a face powder (concentration not provided) and could be possibly inhaled. Inhalation toxicity data were not available. Coupled with the small actual exposure in the breathing zone and the low concentrations at which these ingredients are used (or expected to be used) in potentially inhaled products, the available information indicates that incidental inhalation would not be a significant route of exposure that might lead to local respiratory or systemic effects. A detailed discussion and summary of the Panel's approach to evaluating incidental inhalation exposures to ingredients in cosmetic products is available at https://www.cir-safety.org/cir-findings.

The Panel's respiratory exposure resource document (see link above) notes that airbrush technology presents a potential safety concern, and that no data are available for consumer habits and practices thereof. As a result of deficiencies in these critical data needs, the safety of cosmetic ingredients applied by airbrush delivery systems cannot be assessed by the Panel. Therefore, the Panel has found the data insufficient to support the safe use of cosmetic ingredients applied via an airbrush delivery system.

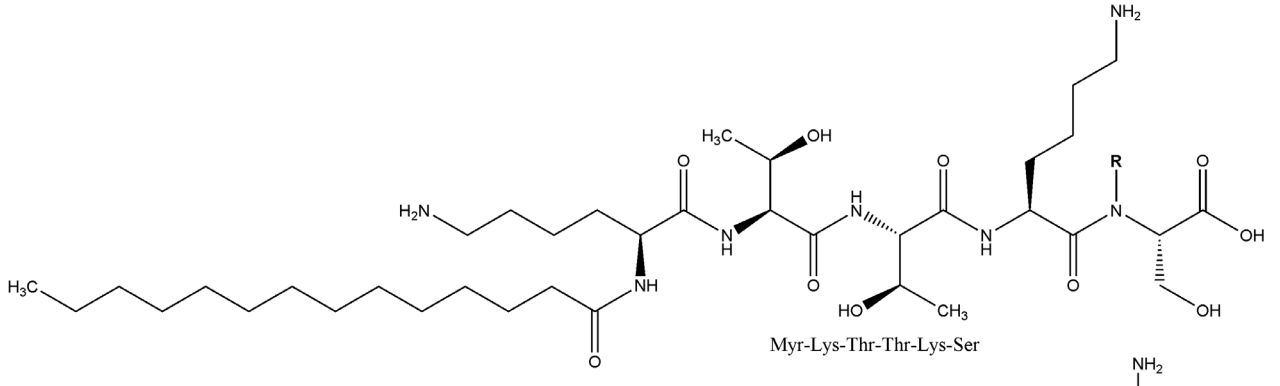
CONCLUSION

To be determined.

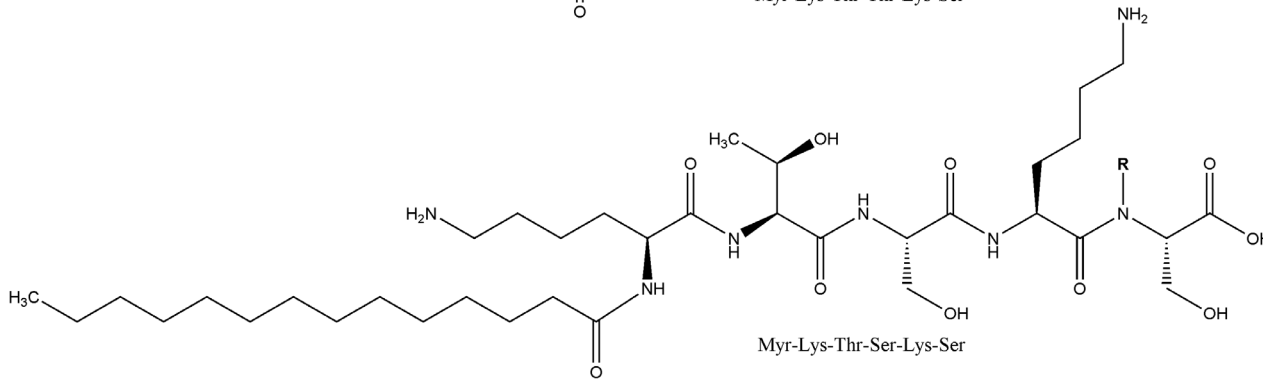
TABLES

Table 1. Definitions, structures, and functions of the ingredients in this assessment[1, CIR Staff]

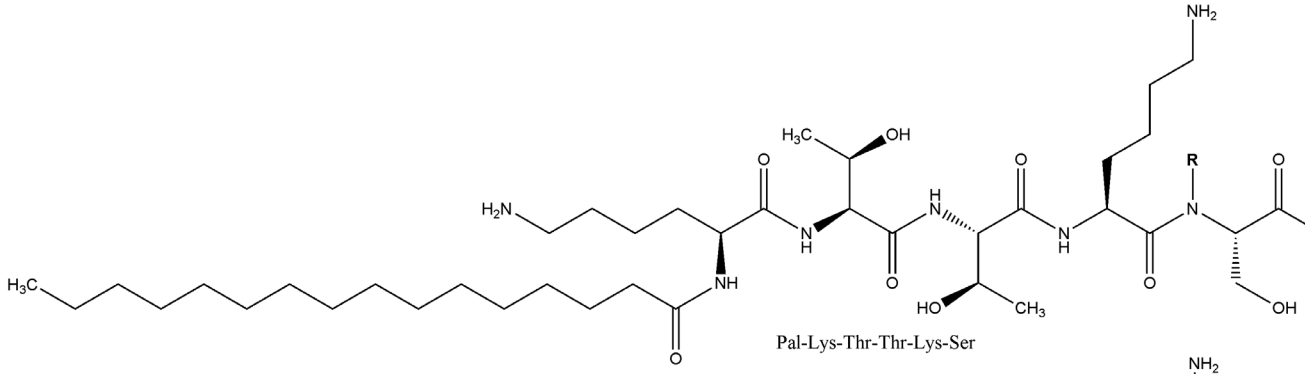
Ingredient	Definition	Function
Myristoyl Pentapeptide-4 1392416-25-9	Myristoyl Pentapeptide-4 is the reaction product of myristic acid and Pentapeptide-4.	Skin-conditioning agent - miscellaneous
Palmitoyl Pentapeptide-4 521091-64-5 214047-00-4	Palmitoyl Pentapeptide-4 is the reaction product of palmitic acid and Pentapeptide-4.	Skin-conditioning agent - miscellaneous



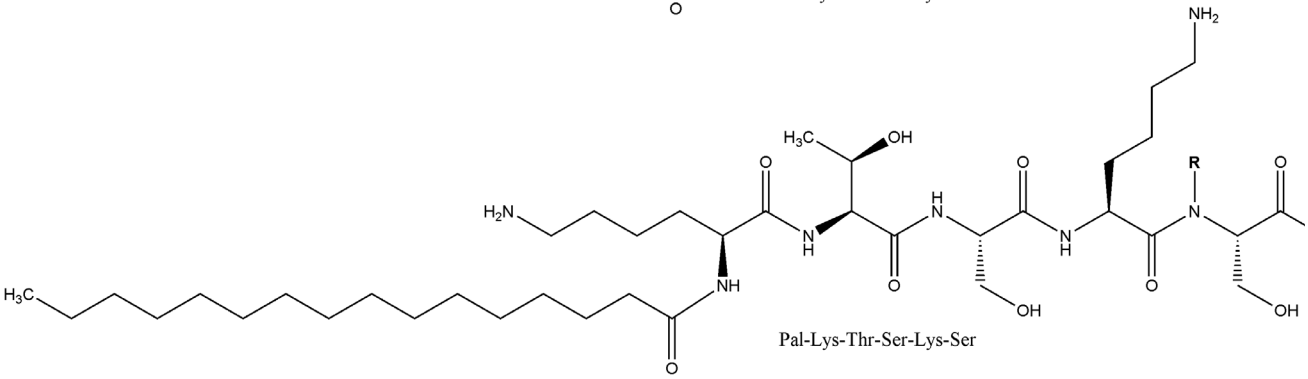
Myr-Lys-Thr-Thr-Lys-Ser



Myr-Lys-Thr-Ser-Lys-Ser



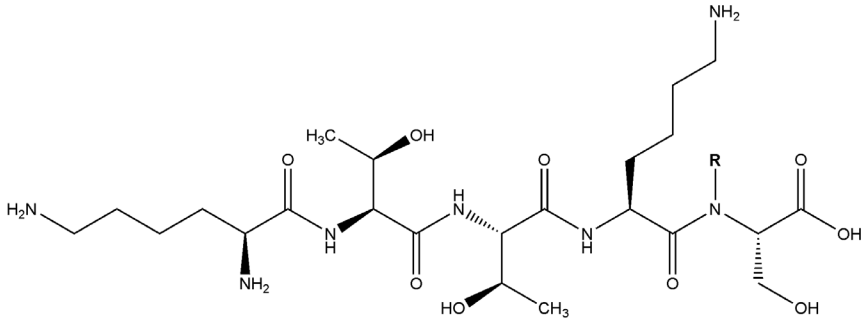
Pal-Lys-Thr-Thr-Lys-Ser



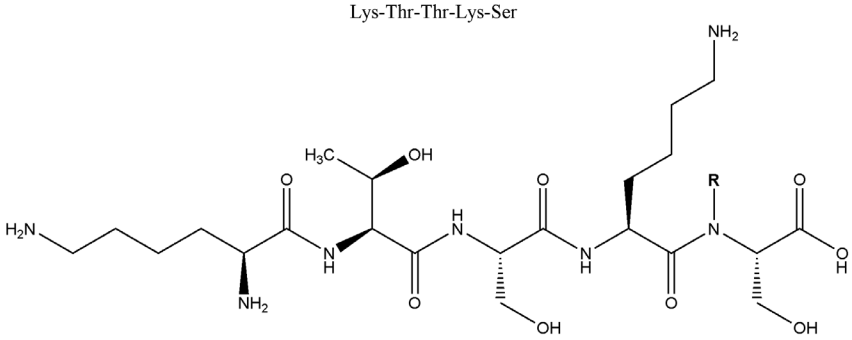
Pal-Lys-Thr-Ser-Lys-Ser

Table 1. Definitions, structures, and functions of the ingredients in this assessment[1, CIR Staff]

Ingredient	Definition	Function
Pentapeptide-4 149128-48-3	Pentapeptide-4 is a synthetic peptide containing lysine, serine, and threonine.	Skin-conditioning agent - miscellaneous



Lys-Thr-Thr-Lys-Ser



Lys-Thr-Ser-Lys-Ser

Table 2. Chemical properties

Property	Value	Reference
	Myristoyl Pentapeptide-4	
Molecular Weight (g/mol)	774 (Myr-Lys-Thr-Thr-Lys-Ser) 759.99 (Myr-Lys-Thr-Ser-Lys-Ser)	14
Topological Polar Surface Area (Å^2)	296 (estimated; Myr-Lys-Thr-Thr-Lys-Ser)	14
log p	1.85 (predicted; Myr-Lys-Thr-Thr-Lys-Ser) 1.6 (predicted; Myr-Lys-Thr-Ser-Lys-Ser)	18
	Palmitoyl Pentapeptide-4	
Physical Form	Powder	15
Color	White	15
Molecular Weight (g/mol)	802.1 (Pal-Lys-Thr-Thr-Lys-Ser) 788.04 (Pal-Lys-Thr-Ser-Lys-Ser)	15,16
Topological Surface Area (Å^2)	296 (estimated; Pal-Lys-Thr-Thr-Lys-Ser)	16
log p	2.72 (predicted; Pal-Lys-Thr-Thr-Lys-Ser) 2.52 (predicted; Pal-Lys-Thr-Ser-Lys-Ser)	18
	Pentapeptide-4	
Molecular Weight (g/mol)	563.65 (Lys-Thr-Thr-Lys-Ser) 549.63 (Lys-Thr-Ser-Lys-Ser)	17
Topological Polar Surface Area (Å^2)	292 (estimated; Lys-Thr-Thr-Lys-Ser)	17
log p	-4.12 (predicted; Lys-Thr-Thr-Lys-Ser) -4.39 (predicted; Lys-Thr-Ser-Lys-Ser)	18

Table 3. Frequency (2023)[20] and concentration (2022)[21] of use according to likely duration and exposure by product category

	Myristoyl Pentapeptide-4		Palmitoyl Pentapeptide-4		Pentapeptide-4	
	# of Uses	Max Conc of Use (%)	# of Uses	Max Conc of Use (%)	# of Uses	Max Conc of Use (%)
Totals*	**4**	**NR**	**239**	**0.000005-0.0035**	**1**	**NR**
summarized by likely duration and exposure**						
Duration of Use						
Leave-On	*4*	*NR*	*223*	*0.00036 – 0.0012*	*1*	*NR*
Rinse-Off	*NR*	*NR*	*16*	*0.000005 – 0.0035*	*NR*	*NR*
Diluted for (Bath) Use	*NR*	*NR*	*NR*	*NR*	*NR*	*NR*
Exposure Type						
Eye Area	4	NR	31	0.0012	NR	NR
Incidental Ingestion	NR	NR	NR	NR	NR	NR
Incidental Inhalation-Spray	NR	NR	117[a]; 64[b]	NR	1[a]	NR
Incidental Inhalation-Powder	NR	NR	1; 64[b]	0.00036 – 0.0012[c]	NR	NR
Dermal Contact	4	NR	236	0.000005 – 0.0012	1	NR
Deodorant (underarm)	NR	NR	NR	NR	NR	NR
Hair - Non-Coloring	NR	NR	3	0.00035 – 0.0035	NR	NR
Hair-Coloring	NR	NR	NR	NR	NR	NR
Nail	NR	NR	NR	NR	NR	NR
Mucous Membrane	NR	NR	2	0.000005	NR	NR
Baby Products	NR	NR	NR	NR	NR	NR
as reported by product category						
Eye Makeup Preparations						
Eye Lotion			21	0.0012		
Other Eye Makeup Preparations	4	NR	10	NR		
Hair Preparations (non-coloring)						
Hair Conditioner			1	0.0035		
Rinses (non-coloring)			1	NR		
Shampoos (non-coloring)			1	0.00035		
Makeup Preparations						
Face Powders			1	NR		
Foundations			4	NR		
Personal Cleanliness Products						
Bath Soaps and Detergents			1	0.000005		
Other Personal Cleanliness Products			1	NR		
Skin Care Preparations						
Cleansing			10	0.000005		
Face and Neck (exc shave)			59	0.0012 (not spray)		
Body and Hand (exc shave)			5	0.00036 (not spray)		
Moisturizing			101	0.00059 (not spray)	1	NR
Night			8	NR		
Paste Masks (mud packs)			1	NR		
Skin Fresheners			8	NR		
Other Skin Care Preparations			6	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.

**likely duration and exposure are derived based on product category (see Use Categorization https://www.cir-safety.org/cir-findings)

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

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

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

Table 4. Genotoxicity studies

Test Article	Vehicle	Concentration/Dose	Test System	Procedure	Results	Reference
IN VITRO						
0.5% Palmitoyl Pentapeptide-4 in distilled water/ethanol (75/25) Pal-KTTKS	distilled water	tested at 2% 312.5, 625, 1250, 2500, and 5000 µg/plate, with or without metabolic activation	*S. typhimurium* TA98, TA100, TA1535, TA1537, and *E. coli* WP2*uvr*A	Ames test. For positive controls, sodium azide, 9-aminoacridine, 2-nitrofluorene, and 4-nitroquinoline were tested in the absence of metabolic activation, while 2-anthramine was tested in the presence of metabolic activation. Revertant colonies were scored after 48 to 72 h of incubation at 37 °C.	Not mutagenic. Results for the vehicle and positive controls were as expected.	[15,27]
Palmitoyl Pentapeptide-4, 81.6% pure Pal-KTSKS	DMSO	1.6, 5, 16, 50, 160, 500, 1600, and 5000 µg/plate, with or without metabolic activation	*S. typhimurium* TA98, TA100, TA102, TA1535, and TA1537	Ames test. OECD TG471. In the absence of metabolic activation, sodium azide and mitomycin were tested in water, and 2-nitroflourene and 9-aminoacridine were tested in DMSO, for positive controls. In the presence of metabolic activation, 2-aminoanthracine was tested in DMSO as a positive control.	Not mutagenic. Signs of cytotoxic activity were observed under test conditions for the test article; controls produced expected results.	[19,28]
Palmitoyl Pentapeptide-4, > 96% pure Pal-KTSKS	sterile water	as supplied <u>with metabolic activation:</u> 4-h treatment, 24-h recovery: 250, 500, or 1000 µg/ml <u>without metabolic activation</u>: 4-h treatment, 24-h recovery: 375, 500, or 750 µg/ml 24-h, continuous treatment: 250, 320, or 400 µg/ml	Cultured human peripheral blood lymphocytes	Micronucleus test. OECD TG 487. Cells were treated for 4 h, with a 24-h recovery period, with and without metabolic activation (short treatment). In an additional assay, cells were treated for 24 h without a recovery period (continuous treatment). Cells treated were treated for 4 h followed by a 24-h recovery period, with cyclophosphamide in the presence of metabolic activation and with mitomycin in the absence of metabolic activation. Mitomycin and griseofulvin were used as positive controls in the 24-h, continuous assay.	Not genotoxic. Neither statistically or biologically significant increases in the number of micronucleated cells were observed with the short-term or continuous treatments.	[19,29]

DMSO – dimethyl sulfoxide; OECD – Organisation for Economic Cooperation and Development; TG – test guideline

Table 5. Dermal irritation and sensitization studies

Test Article	Vehicle	Test Concentration/Dose	Test Population/ System	Procedure	Results	Reference
IRRITATION						
IN VITRO						
Formulation containing 0.12% Palmitoyl Pentapeptide-4, glycerin, and water Pal-KTSKS	tested as supplied	10 µl; 100% (effective test concentration: 0.12% Palmitoyl Pentapeptide-4)	EpiSkin® reconstructed human epidermis model	Cutaneous primary irritation test. OECD TG 439. The test article, positive control (10 µl SDS), and negative control (10 µl PBS) were in contact with the epidermis model for 15 min, followed by a 42-h incubation period. Cell viability was evaluated via an MTT assay.	Predicted to be not irritating. The test article, as supplied did not stain the cells or interact with MTT.	[19,32]
ANIMAL						
Palmitoyl Pentapeptide-4 Pal-KTTKS	not specified	0.01%; 0.5 ml	3 male New Zealand white rabbits	Acute dermal irritation study. OECD TG 404. Semi-occlusive application of the test substance was made to shaved skin for 4 h. Skin reactions were observed 1, 24, 48, and 72 h after patch removal. Mean values for erythema and edema were calculated for each animal.	Not irritating. Very slight erythema was observed in 1 animal, only on day 1. All erythema and edema mean scores over 24, 48, and 72 h were 0.	[15,33]

Table 5. Dermal irritation and sensitization studies

Test Article	Vehicle	Test Concentration/Dose	Test Population/ System	Procedure	Results	Reference
Palmitoyl Pentapeptide-4 Pal-KTTKS	not specified	0.01%; 0.05 ml	Guinea pigs (5/sex; strain not specified)	2-wk dermal irritation study. Open application to a shaved, 2 cm^2 area of the left flank daily for 14 d; the site was not rinsed. Purified water applied to the right flank served as the control. Skin reactions were evaluated before and approximately 24 h after each application; these values were used to calculate daily irritation and weekly mean irritation indices.	Non-irritating. Very slight erythema was noted in 1 animal on days 12 and 13. According to the researchers, these reaction were not attributed to an irritant effect of the test substance because they were very slight and only occurred in 1 animal.	15,26
HUMAN						
Trade name mixture containing 0.01% Palmitoyl Pentapeptide-4 Pal-KTTKS	tested as supplied	0.02 ml (effective test concentration: 0.01% Palmitoyl Pentapeptide-4)	10 subjects	Acute skin irritation study. A single occlusive, neat application of the test substance was made to a 50 mm^2 area of the back for 48 h using Finn chambers. Untreated sites covered with an occlusive patch served as negative controls. Skin reactions were scored 30 min after patch removal.	Well-tolerated. Very slight erythema (hardly visible) in 1 of the subjects. PCI = 0.10.	15,34
Formulation containing 0.12% Palmitoyl Pentapeptide-4 Pal-KTSKS	distilled water	160 µl; 15% (effective test concentration: 0.018% Palmitoyl Pentapeptide-4)	11 subjects; phototype II - IV	Patch test; semi-occlusive application to 400 mm^2 for 48 h; test sites were scored before patching and 15 – 30 min after patch removal	Not irritating. No reactions were observed in either the test or control subjects.	19,35
SENSITIZATION						
IN CHEMICO/ IN VITRO						
Palmitoyl Pentapeptide-4, 81.6% pure Pal-KTSKS	water	5 (50 µl) and 25 mM (250 µl)	cysteine and leucine	DPRA; OECD TG 442C and ECVAM DB-ALM Protocol No 154; 24-h incubation period; each concentration was tested 3 times; mean percent depletion of cysteine and lysine was evaluated; positive control: cinnamaldehyde in acetonitrile; negative control: peptide in buffer	Prediction of non-sensitizing. Mean percent depletion of cysteine and lysine was 4.58%, reflecting no or minimal reactivity.	19,36
Palmitoyl Pentapeptide-4, 81.6% pure Pal-KTSKS	DMSO	0.98 – 2000 µM; 0.05 ml	KeratinoSens™ cell line	ARE-Nrf2 Luciferase test method; OECD TG 442D and ECVAM DB-ALM protocol 155; performed 2 times; positive control: cinnamaldehyde; negative controls: 1% DMSO in treatment medium	Prediction of non-sensitizing. I_{max} of 1.35, compared to 5.12 for positive control.	19,37
ANIMAL						
Palmitoyl Pentapeptide-4, 0.01% Pal-KTTKS	saline	Induction: 75% (effective concentration 0.0075%); topical induction: applied neat (effective concentration 0.01%) Challenge: 25% (effective concentration: 0.0025%)	Guinea pigs (strain not specified) test animals: 10/sex controls: 5/sex	OECD TG 406. Guinea pig maximization test. Saline solution and mercaptobenzothiazole in corn oil served as negative and positive controls, respectively. On day 1, the test substance was mixed with FCA and injected intradermally in the back. After pretreatment of the test site with 10% SLS (pet) on day 7, the test substance was applied on day 8 under occlusion to the same region for 48 h. After a non-treatment period of 12 d, both test and control animals received an occlusive challenge application of the test substance to the right flank, as well as an occlusive application of the vehicle control to the left flank, both for 24 h. Skin reactions were evaluated 24 and 48 h after patch removal.	Not sensitizing. Controls yielded expected results.	15,38

Table 5. Dermal irritation and sensitization studies

Test Article	Vehicle	Test Concentration/Dose	Test Population/ System	Procedure	Results	Reference
				HUMAN		
Formulation containing 0.12% Palmitoyl Pentapeptide-4 Pal-KTSKS	distilled water	160 µl; 15% (effective concentration: 0.018% Palmitoyl Pentapeptide-4)	106 subjects; phototype II - III	HRIPT; semi-occlusive conditions (400 mm^2); induction: 9 applications (48 – 72 h) were made to the upper back over a 3-wk period. Concurrent applications of distilled water under the same conditions served as control sites. Challenge: after a non-treatment period of 2 wk, a 48-h application was made to an induction site and an untreated site. Treated sites were scored before patching, 15 – 30 min after patch removal, and, additionally, 48 h after patch removal during the challenge phase	Not irritating or sensitizing. No reactions were induced during the induction or challenge phases.	[19,39]
Trade name mixture containing 0.01% Palmitoyl Pentapeptide-4 Pal-KTTKS	NA	applied undiluted: 55.4 mg/cm of the mixture this results in 5.54 µg/cm^2 of Palmitoyl Pentapeptide-4	51 subjects	HRIPT; The remaining ingredients in the mixture included: glycerin (qsp 100), water (25%), butylene glycol (20%), carbomer (1%), polysorbate 20 (0.5%), sodium lactate (max 1%); 2 g of the test material applied to a 3.61 cm^2 area under occlusive conditions.	Non-irritating and non-sensitizing	[15,40,41]

ARE – antioxidant/electrophile response element; DPRA – direct peptide reactivity assay; ECVAM DB-ALM - European Centre for Validation of Alternative Methods Database on Alternative Methods; FCA – Freund's Complete Adjuvant; HRIPT – human repeated insult patch test; MTT - 3-[4,5-dimethylthiazol-2-yl]-2,5 diphenyl tetrazolium bromide; NA – not applicable; OECD – Organisation for Economic Cooperation and Development; PBS – phosphate-buffered solution; PCI – primary cutaneous irritation; SDS – sodium dodecyl sulfate; SLS – sodium lauryl sulfate; TG – test guideline

Table 6. Ocular irritation studies

Test Article	Vehicle	Test Concentration/Dose	Test Population	Procedure	Results	Reference
IN VITRO						
Formulation containing 0.12% Palmitoyl Pentapeptide-4 Pal-KTSKS	water	300 µl; 10% (effective test concentration: 0.012% Palmitoyl Pentapeptide-4)	4 eggs (test article); 2 eggs (reference controls)	In vitro HET-CAM assay; performed in agreement with French GLP, the European Directive 2004/10/EC, and the August 2004 decree from the *Journal Officiel Republique Francaise*; positive control: 0.4 and 3.2% lauryl sulfobetaine in saline solution; negative control: 0.05% lauryl sulfobetaine in saline solution	Classified as slightly irritating. The mean score calculated for hyperemia, hemorrhage, and coagulation, opacity, and/or thrombosis was 4.25.	[19,43]
Trade name mixture containing 0.01% Palmitoyl Pentapeptide-4 Pal-KTTKS	tested as supplied	dose not specified (effective test concentration: 0.01% Palmitoyl Pentapeptide-4)	HET-CAM	In vitro HET-CAM assay; 1996 HET CAM protocol published in the *Journal Officiel Republique Francaise*; positive control: SDS (0.05% (w/v))	Classified as moderately irritating. The mean irritation index for the SDS was 12, while the mean irritation index for the test substance was 6	[15,34]
Formulation containing 0.12% Palmitoyl Pentapeptide-4; glycerin, and water Pal-KTSKS	water	30 µl; 30% (effective test concentration: 0.036% Palmitoyl Pentapeptide-4)	human immortalized comeal epithelial cells	SkinEthic™ human corneal epithelial model. OECD TG 492, in agreement with French GLP, European Directive 2004/10/CE, and 2004 decree published in the *Journal Officiel Republique Francaise*. 2 epithelia were used as replicates; 30 min incubation period; positive control: methyl acetate; negative control: DPBS; cell viability evaluated via MTT assay	Not irritating. Mean cell viability for the test article was 104.3%. Positive controls yielded expected results.	[19,44]
ANIMAL						
Palmitoyl Pentapeptide-4 Pal-KTTKS	not specified	0.01%; 0.1 ml	3 male New Zealand white rabbits	OECD TG 405. A single dose was instilled into the conjunctival sac of the left eye. Treated eyes were not rinsed; right eyes served as control Ocular reactions were evaluated 1, 24, 48 and 72 h. Mean values for chemosis, redness of the conjunctiva, iris lesions, and corneal opacity were calculated for each animal	Classified as non-irritant. All mean values were 0 at each time interval.	[15,45]

DPBS – Dulbecco's phosphate buffer solution; GLP – good laboratory practices; HET-CAM – hen's egg-chorioallantoic membrane test; MTT – 3-[4,5-dimethylthiazol-2-yl]-2,5 diphenyl tetrazolium bromide; OECD – Organisation for Economic Cooperation and Development; SDS – sodium dodecyl sulfate; TG – test guideline

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Concentration of Use by FDA Product Category – Pentapeptide-4*

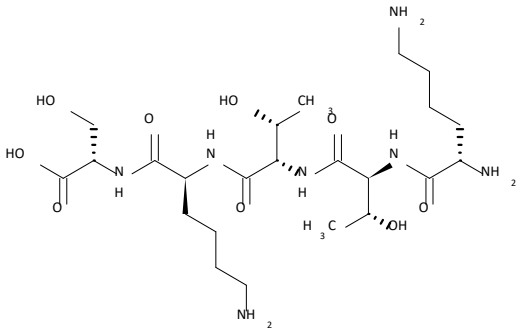
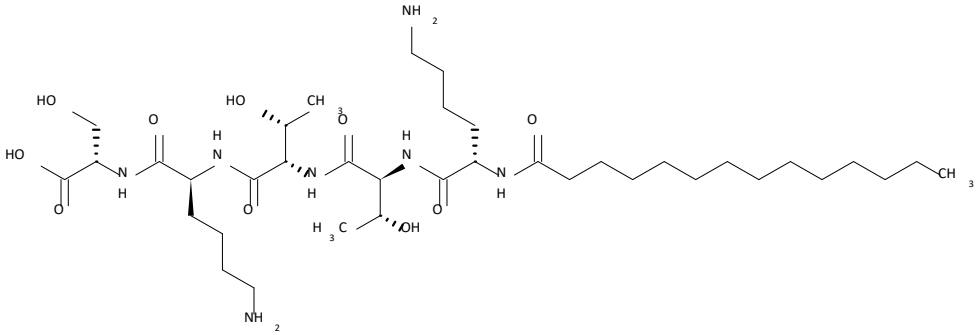
Palmitoyl Pentapeptide-4
Pentapeptide-4
Myristoyl Pentapeptide-4

Ingredient	FDA Product Category	Maximum Concentration of Use
Palmitoyl Pentapeptide-4	Eye lotions	0.0012%
Palmitoyl Pentapeptide-4	Hair conditioners	0.0035%
Palmitoyl Pentapeptide-4	Shampoos (noncoloring)	0.00035%
Palmitoyl Pentapeptide-4	Bath soaps and detergents	0.000005%
Palmitoyl Pentapeptide-4	Skin cleansing (cold creams, cleansing lotions, liquids, and pads)	0.000005%
Palmitoyl Pentapeptide-4	Face and neck products Not spray	0.0012%
Palmitoyl Pentapeptide-4	Body and hand products Not spray	0.00036%
Palmitoyl Pentapeptide-4	Moisturizing products Not spray	0.00059%

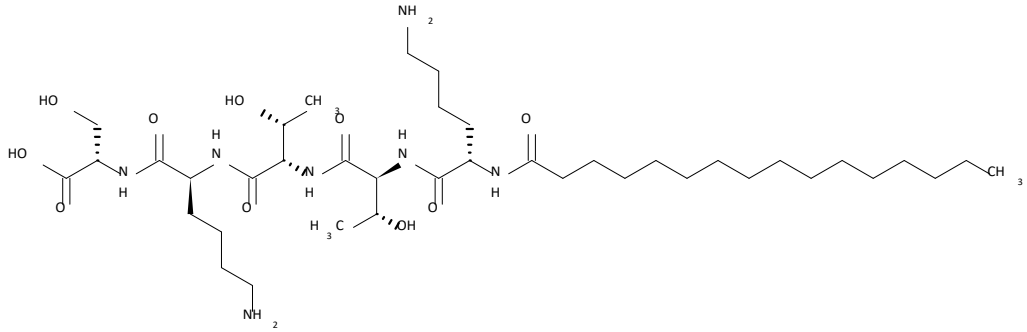
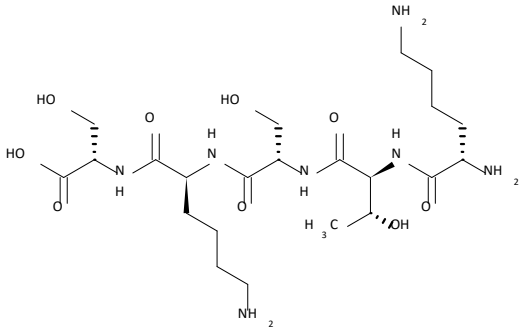
*Ingredients included in the title of the table but not found in the table were included in the concentration of use survey, but no uses were reported.

Information collected in 2022
Table prepared: July 6, 2022
Revised September 21, 2023: removed 0.05% Myristoyl Pentapeptide-4 other eye makeup preparations (this was an experimental product that was never developed)

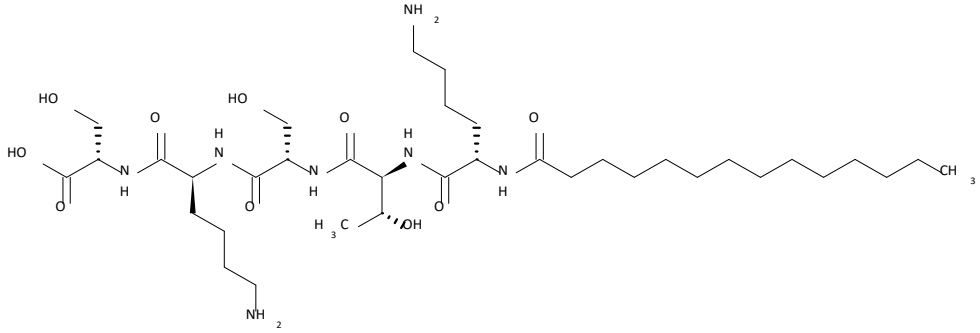
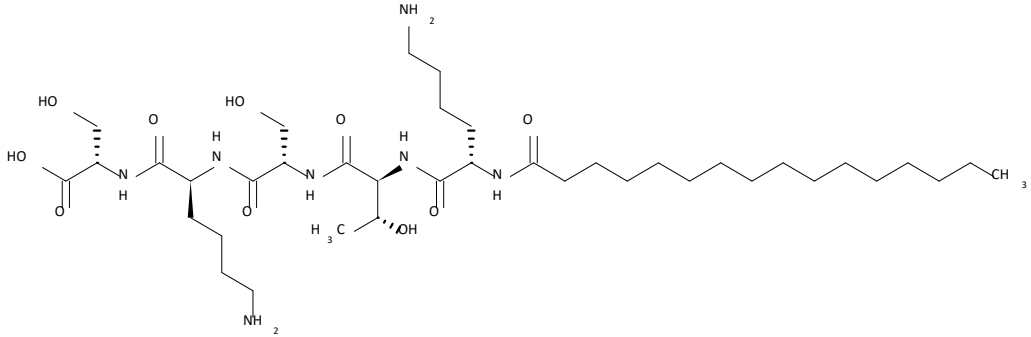
data2_Pentapeptides_032024.pdf -- Predicted log P values for Pentapeptide-4 ingredients. (Unpublished data submitted by Personal Care Products Council on September 18, 2023.)

Structure	**CAS RN**	**Chemical Identifier**	**LogP ACD Percepta Ver. 2022.2.3**
	149128-48-3	Pentapeptide-4 (KTTKS)	-4.12
	1392416-25-9	Myristoyl Pentapeptide-4 (KTTKS)	1.85

data2_Pentapeptides_032024.pdf -- Predicted log P values for Pentapeptide-4 ingredients. (Unpublished data submitted by Personal Care Products Council on September 18, 2023.)

Structure	**CAS RN**	**Chemical Identifier**	**LogP ACD Percepta Ver. 2022.2.3**
	214047-00-4	Palmitoyl Pentapeptide-4 (KTTKS)	2.72
	2758678-86-1	Pentapeptide-4 (KTSKS)	-4.39

data2_Pentapeptides_032024.pdf -- Predicted log P values for Pentapeptide-4 ingredients. (Unpublished data submitted by Personal Care Products Council on September 18, 2023.)

Structure	**CAS RN**	**Chemical Identifier**	**LogP ACD Percepta Ver. 2022.2.3**
	No CAS RN	Myristoyl Pentapeptide-4 (KTSKS)	1.6
	521091-64-5	Palmitoyl Pentapeptide-4 (KTSKS)	2.52

Product information

Our Ref: MATRIXYL®

Date: 5 October 2023

STATEMENT

In 1999, MATRIXYL® has been tested in HRIPT test at the concentration of 100% (report from Consumer Product Testing Co, n°C99-0567.02). The test was conducted on MATRIXYL® with the batch number MATRIXV1/001. According to this report, the active ingredient shows neither skin irritation nor skin sensitizing effect on human (n=59).

SEDERMA herbily confirms that the formula of MATRIXYL® did not change since 1999 and is available below:

INCI	%	CAS Nr	EINECS Nr
Glycerin	qsp 100	56-81-5	200-289-5
Water (Aqua)	≈ 25	7732-18-5	231-791-2
Butylene Glycol	≈ 20	107-88-0	203-529-7
Carbomer	≈ 1	9003-01-4	n/a
Polysorbate 20	≈ 0.5	9005-64-5	n/a
Palmitoyl Pentapeptide-4	≈ 0.01	214047-00-4	n/a
Preservatives - Antioxidants: / *Manufacturing additive*:			
Sodium Lactate	max. 1	72-17-3	200-772-0

Regarding the protocol, approximately 0.2 mL or 0.2g of the test material was applied on the upper back between the scapulae, with a surface area of 3/4" x 3/4" (or 19mm x 19mm), under occluded patch. This corresponds to a surface of 361 mm² or 3.61 cm².

MATRIXYL® was applied pure. Then, the amount applied is approximately 0.2g/3.61 cm² or 55.4 mg/cm². If the concentration of the peptide (100 ppm or 0.01%) is taken into account, the amount applied in this test is 5.54 µg/cm².

Dr. Philippe MONDON, *PhD*
Scientific Director

Vincent Vicedo, *MsC*
Toxicologist/Risk Assessor

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