

GREEN

Safety Assessment of
Hydrolyzed Source Proteins
as Used in Cosmetics

CIR EXPERT PANEL MEETING
DECEMBER 10-11, 2012

Cosmetic Ingredient Review

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Memorandum

To: CIR Expert Panel Members and Liaisons
From: Christina L. Burnett
Scientific Writer/Analyst
Date: November 16, 2012
Subject: Draft Report on Hydrolyzed Proteins

In May 2012, CIR issued the Scientific Literature Review (SLR) for hydrolyzed proteins. The ingredients in this group are interrelated because they each are prepared from proteins by partial hydrolysis to yield cosmetically acceptable raw materials. The definitions exclude the complete hydrolysis products of proteins, the amino acids, which have been reviewed separately in the source amino acid report.

The research strategy followed the approach taken with the safety assessment of α -amino acids, which was to rely heavily on the GRAS direct food additive status of the common amino acids and to demonstrate the absence of significant oral toxicity. That is the approach we have taken; however, only a small amount of information was found in the published literature, especially information on dermal exposures. In the SLR, data were requested on impurities and safety test data specific to dermal exposures.

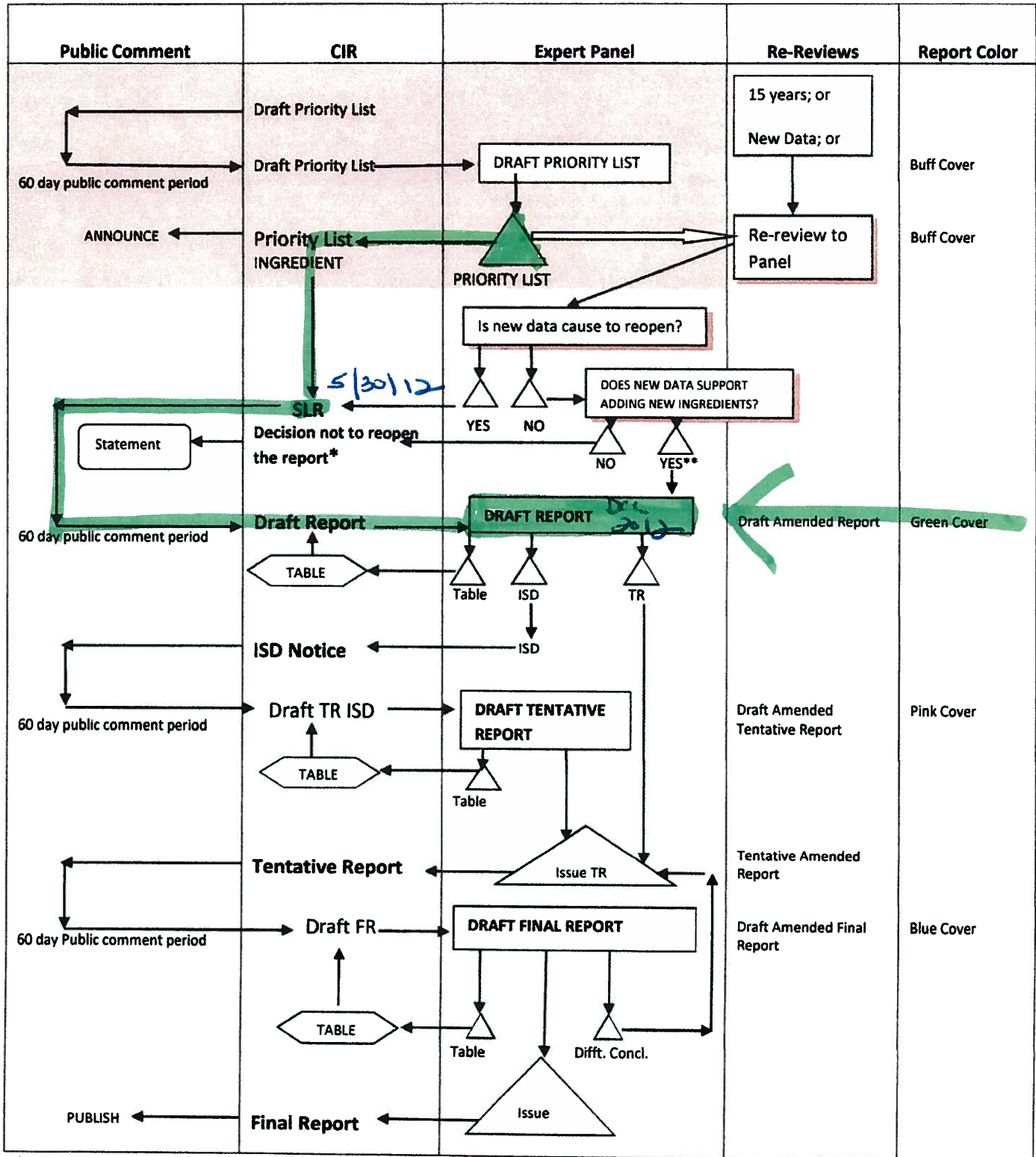
Since the May announcement, the Personal Care Products Council (Council) has provided comments on the report, which have been considered, method of manufacturing, impurities, molecular weight, concentration of use, irritation and sensitization, genotoxicity, and phototoxicity data. These data have been incorporated into the report. The comments can be found in this panel book and the unpublished data has been posted on-line at <http://www.cir-safety.org/supplemental/doc/data-safety-assessment-hydrolyzed-source-proteins-used-cosmetics>.

According to the FDA's VCRP database, hydrolyzed wheat protein has the most reported uses in cosmetic and personal care products, with a total of 1055; half of those uses are in leave-on formulations. Hydrolyzed soy protein has the second greatest number of overall uses reported, with a total of 681; 419 of those uses are in leave-on formulations.

In the Council's use concentration survey, hydrolyzed wheat protein had a wide maximum use concentration range of 2.0×10^{-5} to 1.7%, with the 1.7% reported in rinse-off non-coloring hair products. Hydrolyzed soy protein had a wide maximum use concentration range of 2.0×10^{-5} to 4%, with the 4% reported in face and neck skin care preparations. All other use concentrations that were reported had similar ranges. No uses were reported to the VCRP and/or the Council's use concentration survey for ammonium hydrolyzed collagen, calcium hydrolyzed collagen, hydrolyzed albumin, hydrolyzed avocado protein, hydrolyzed casein, hydrolyzed conalbumin, hydrolyzed egg protein, hydrolyzed fibronectin, hydrolyzed gadidae protein, hydrolyzed gelatin, hydrolyzed hemoglobin, hydrolyzed hemp seed protein, hydrolyzed honey protein, hydrolyzed lactalbumin, hydrolyzed maple sycamore protein, hydrolyzed pea protein, hydrolyzed reticulon, hydrolyzed royal jelly protein, hydrolyzed serum protein, hydrolyzed sesame protein, hydrolyzed spinal protein, hydrolyzed spongin, hydrolyzed whey protein, hydrolyzed zein, MEA-hydrolyzed collagen, MEA-hydrolyzed silk, sodium hydrolyzed casein, and zinc hydrolyzed collagen.

If no further data are needed, the Panel should issue a Tentative Report.

SAFETY ASSESSMENT FLOW CHART



*The CIR Staff notifies of the public of the decision not to re-open the report and prepares a draft statement for review by the Panel. After Panel review, the statement is issued to the Public.

**If Draft Amended Report (DAR) is available, the Panel may choose to review; if not, CIR staff prepares DAR for Panel Review.



Hydrolyzed Proteins History

May 2012 – Scientific Literature Review announced.

Hydrolyzed Proteins Data Profile* – December 2012 – Writer, Christina Burnett						
	Reported Use	Composition/ Impurities	Method of Manufacturing	Irritation/ Sensitization - Animal	Irritation/ Sensitization - Clinical	Ocular/ Mucousal
Ammonium Hydrolyzed Collagen						
Calcium Hydrolyzed Collagen						
Hydrolyzed Actin	X					
Hydrolyzed Albumen	X					
Hydrolyzed Amaranth Protein	X		X	X		X
Hydrolyzed Avocado Protein		X	X	X	X	X
Hydrolyzed Barley Protein	X					
Hydrolyzed Brazil Nut Protein	X		X			
Hydrolyzed Casein	X					
Hydrolyzed Conalbumin						
Hydrolyzed Conchiolin Protein	X					
Hydrolyzed Cottonseed Protein	X					
Hydrolyzed Egg Protein						
Hydrolyzed Elastin	X		X	X		X
Hydrolyzed Extensin	X					
Hydrolyzed Fibroin	X					
Hydrolyzed Fibronectin	X					
Hydrolyzed Gadidae Protein	X					
Hydrolyzed Gelatin						
Hydrolyzed Hair Keratin	X		X			
Hydrolyzed Hazelnut Protein	X	X	X	X		X
Hydrolyzed Hemoglobin						
Hydrolyzed Hemp Seed Protein						
Hydrolyzed Honey Protein	X					
Hydrolyzed Jojoba Protein	X					
Hydrolyzed Keratin	X		X	X		X
Hydrolyzed Lactalbumin						
Hydrolyzed Lupine Protein	X	X	X	X	X	X
Hydrolyzed Maple Sycamore Protein						
Hydrolyzed Milk Protein	X			X	X	X
Hydrolyzed Oat Protein	X					
Hydrolyzed Pea Protein	X					
Hydrolyzed Potato Protein	X			X		
Hydrolyzed Reticulin						
Hydrolyzed Royal Jelly Protein						
Hydrolyzed Sericin	X					
Hydrolyzed Serum Protein			X			
Hydrolyzed Sesame Protein						
Hydrolyzed Silk	X	X	X	X	X	X
Hydrolyzed Soy Protein	X	X	X	X	X	X
Hydrolyzed Soymilk Protein	X					
Hydrolyzed Spinal Protein						
Hydrolyzed Spongin						
Hydrolyzed Sweet Almond Protein	X		X	X		
Hydrolyzed Vegetable Protein	X			X		X
Hydrolyzed Wheat Gluten	X					
Hydrolyzed Wheat Protein	X	X	X	X	X	X
Hydrolyzed Whey Protein	X					
Hydrolyzed Yeast Protein	X			X		
Hydrolyzed Yogurt Protein	X					
Hydrolyzed Zein						
MEA-Hydrolyzed Collagen	X					
MEA-Hydrolyzed Silk						
Sodium Hydrolyzed Casein						
Zinc Hydrolyzed Collagen						

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

SEARCH STRATEGY FOR HYDROLYZED PROTEINS

(Performed by Christina Burnett and Ivan Boyer)

February-April: SCIFINDER search for Hydrolyzed Proteins (55 substances, searched under INCI names and CAS No.):

- Initial search for “**adverse effect, including toxicity**” yielded **18** references.
- Also performed searches using the following search terms (no limits for reference type):
 - “**Hydrolyzed Proteins in Cosmetics**” (yield = **30** references);
 - “**Skin Sensitization – Hydrolyzed Proteins**” (yield = **1** reference);
 - “**Bioactive Peptides - Cosmetics**” (yield = **18** references);
 - “**Skin Irritation – Polypeptides**” (yield = **23** references);
 - “**Skin Sensitization – Polypeptides**” (yield = **11** references);
 - “**Biogenic Peptides**” (yield = **1** reference);
 - “**Bioactive Peptides – Toxicity**” (yield = **28** references);
 - “**Bioactive Peptides – Skin Irritation**” (yield = **1** reference);
 - “**Hydrolyzed Proteins Chicken Cells in vitro**” (yield = **1** reference); and
 - “**Hydrolyzed Protein Irritation**” (yield = **7** references).

Many of the references were patents or efficacy reports.

13 references were ordered.

Safety Assessment of Hydrolyzed Proteins as Used in Cosmetics

Status: Draft Report for CIR Expert Panel Review
Release Date: November 16, 2012
Panel Meeting Date: December 10-11, 2012

The 2012 Cosmetic Ingredient Review Expert Panel members are: Chairman, Wilma F. Bergfeld, M.D., F.A.C.P.; Donald V. Belsito, M.D.; Ronald A. Hill, Ph.D.; Curtis D. Klaassen, Ph.D.; Daniel C. Liebler, Ph.D.; James G. Marks, Jr., M.D.; Ronald C. Shank, Ph.D.; Thomas J. Slaga, Ph.D.; and Paul W. Snyder, D.V.M., Ph.D. The CIR Director is F. Alan Andersen, Ph.D. This report was prepared by Christina Burnett, Scientific Analyst/Writer, and Bart Heldreth, Ph.D., Chemist CIR.

Cosmetic Ingredient Review

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INTRODUCTION

This safety assessment summarizes available data relevant to 55 hydrolyzed proteins and related salts from plant and animal sources as used in cosmetics. These ingredients function as skin and hair conditioning agents in personal care products. The list of ingredients in this report is found in Table 1.

A concurrent review of the safety of plant and animal derived amino acid ingredients as they are used in cosmetics is being performed by the Cosmetic Ingredient Review (CIR) Expert Panel. The Expert Panel previously has reviewed the safety of α -amino acids, hydrolyzed collagen, and hydrolyzed corn protein, and concluded that these ingredients are safe for use in cosmetic ingredients.¹⁻⁴

CHEMISTRY

The definitions of these ingredients are presented in Table 1.

The ingredients in this group are interrelated because they each are prepared from proteins by partial hydrolysis to yield cosmetically acceptable raw materials. The definitions exclude the complete hydrolysis products of proteins, the amino acids. These protein derivatives are prepared by subjecting animal or vegetable proteins to enzymatic (e.g., papain hydrolysis) or other chemical hydrolyses (e.g., acid or steam hydrolysis). The resulting polypeptide-, oligopeptide-, and peptide-containing products are used as conditioning agents in hair and skin products. Methods used to manufacture protein hydrolysates typically yield broad molecular weight distributions of peptides, 500-30,000 daltons (Da).^{5,6} However, certain enzymes, such as papain, can routinely yield narrower distributions, 500-10,000 Da.⁵ For example, if the average molecular weight of an amino acid is 135 Da, then, under the broader distribution figures, these ingredients are approximately 4 to 220 amino acids in length (and approximately 4 to 74 amino acids in length under the narrower distribution).⁷ Molecular weights provided for some of the specific ingredients in this report are presented in Table 2, and histograms showing the approximate distribution of molecular weights for hydrolyzed actin, hydrolyzed hazelnut protein, hydrolyzed soy protein, and hydrolyzed milk protein are shown in Figure 1.

Method of Manufacturing

Hydrolyzed Amaranth Protein

A supplier reported that hydrolyzed amaranth protein [molecular weight (MW) = 1500] is produced by filtering a solution of finely ground amaranth powder in water and then reacting the resultant colloidal protein solution with acid for a prescribed period of time and temperature until the hydrolyzed protein solution is obtained.⁸

Hydrolyzed Avocado Protein

Hydrolyzed avocado protein is reported to be prepared from sliced and dried avocado fruits.⁹ Cold pressure is used to extract lipids from the fruits and then the proteins are hydrolyzed by enzymatic reactions with a cellulose and a protease. Following centrifugation, the solution is purified by ultrafiltration to remove residual proteins and enzymes. The solution is further purified by nanofiltration to remove salts. The resulting solution is comprised of 20%-50% peptides (w/w) and 20%-30% carbohydrates (w/w).

Hydrolyzed Collagen

As given in the CIR safety assessment and re-review of this ingredient, hydrolyzed collagen may be prepared by alkaline hydrolysis of bovine or fish collagen, followed by enzymatic hydrolysis to the desired molecular weight.^{3,4}

Hydrolyzed Elastin

Hydrolyzed elastin is reported to be prepared from the skin of codfish or from bovine neck tendons.^{10,11} The fibrous tissue is washed and purified to remove soil and other residual materials and then dried. The dried elastin fibers are then hydrolyzed for several hours until the target molecular weight is reached. The final product is a solution, with the bovine source material being concentrated to a 30% active content.

Hydrolyzed Hazelnut Protein

A supplier reported that hydrolyzed hazelnut protein is produced by enzymatic hydrolysis.¹²

Hydrolyzed Keratin

Hydrolyzed keratin may be prepared from sheep's wool.¹³ The wool is first washed to remove soil and debris and then boiled to remove residual oils. Next, the wool is enzyme-hydrolyzed under mild conditions for 4-6 hours. When the target molecular weight is reached, the pH is adjusted to neutralize the enzyme. The resultant solution is a mixture for hydrolyzed keratin fractions with a molecular weight of ~ 1000 Da. The solution may be diluted to produce a 30% active material.

Hydrolyzed Lupine Protein

A supplier reported that hydrolyzed lupine protein is prepared by hydrolyzing lupine proteins in water through an enzymatic reaction with a protease.¹⁴ The solution is then centrifuged and purified by ultrafiltration to remove residual proteins and protease. The solution is further purified by nanofiltration to remove salts. The resulting solution is comprised of >90% peptides (w/w) and <4% carbohydrates (w/w).

Hydrolyzed Milk Protein

A supplier reported that hydrolyzed milk protein is produced from milk intended for human consumption.¹⁵ The milk solids are separated and hydrolyzed with a protease for 2 hours. When the target molecular weight is achieved, the enzyme is inactivated by heating the solution to 140°C for 30 minutes. The inactivation step is repeated if gelatin mixed with a sample loses viscosity, indicating the presence of active protease.

Hydrolyzed Corn and Hydrolyzed Soy Protein

The proteins of corn and soy may be combined and hydrolyzed with enzyme under mild conditions for several hours until the target molecular weight is achieved.¹⁶ The resultant hydrolyzed proteins may then be concentrated.

Hydrolyzed Serum Protein

Hydrolyzed serum protein can be derived from the enzymatic hydrolysis of defibrinated bovine blood plasma by food-grade microbial proteases and aided with heat denaturation.¹⁷ The maximum degree of hydrolysis was 43% at an enzyme concentration of 110 LAPU/g protein after 15.5 h. The resultant substrate consists of small peptides (molecular masses were less than 6.5 kDa and most were less than 1.04 kDa at maximum hydrolysis) and free amino acids, which including lysine, leucine, arginine, serine, and phenylalanine.

Hydrolyzed Silk

Hydrolyzed silk has been reported to be prepared from the cocoon of the silkworm moth (*Bombyx mori*).¹⁸ The silk thread is isolated from the cocoon and the fibers are cleaned and degummed. The individual silk fiber is then wound with other silk fibers to create one long thread. The threads are then combed to remove noils, which are short fibers considered to be by-products of the textile industry. The noils are used in the production of hydrolyzed silk proteins through carefully controlled hydrolysis. The resultant material is a 5% solution of a water soluble silk protein.

Another supplier reported that hydrolyzed silk protein (MW = 300 or 650) may be prepared by both alkaline and enzyme hydrolysis.^{19,20} These processes occur for several hours until the desired molecular weight is reached. The final product are a 20% water solution of hydrolyzed silk protein (MW = 300) or a 6.5% water solution of hydrolyzed silk protein (MW = 650).

Hydrolyzed Soy Protein

Soy hydrolysate may be dephosphorylated, deglycosylated and digested by a variety of endoproteases to generate oligopeptides.²¹ A supplier has reported that hydrolyzed soy protein is produced from isolated soy proteins that are hydrolyzed with a protease enzyme for 2 hours.²² The enzyme is inactivated by heat once the target molecular weight is achieved. The resultant solution may then be concentrated.

Another supplier reported that hydrolyzed soy protein (MW = 300) may be prepared by both alkaline and enzyme hydrolysis.²³ These processes occur for several hours until the desired molecular weight is reached. The final product is a 25% water solution of hydrolyzed soy protein.

Hydrolyzed Wheat Protein

A supplier reported that hydrolyzed wheat protein (MW = 350) may be prepared by both alkaline and enzyme hydrolysis.²⁴ These processes occur for several hours until the desired molecular weight is reached. The final product is a 25% water solution of hydrolyzed wheat protein.

Multiple Hydrolyzed Proteins

A supplier reported that hydrolyzed brazil nut protein, hydrolyzed hair keratin, and hydrolyzed keratin are obtained through acid hydrolysis.²⁵ Hydrolyzed sweet almond protein is obtained through enzyme hydrolysis.

Impurities

Hydrolyzed Avocado Protein

In data supplied by the Personal Care Products Council, hydrolyzed avocado protein has < 0.042 µg/kg benzo[a]pyrene; < 0.10 ppm arsenic; 0.70 ppm barium; < 0.10 ppm cadmium; < 0.75 ppm chromium; 0.40 ppm cobalt; < 0.05 ppm lead; and < 0.10 ppm mercury.⁹ Aflatoxins B&G combined were < 1.86 µg/kg. The absence of organochlorinated, organophosphorylated, , and organosulfur compounds from pesticides was noted.

Hydrolyzed Hazelnut Protein

A supplier reported that hydrolyzed hazelnut protein has less than 20 ppm heavy metals and less than 2 ppm arsenic.¹²

Hydrolyzed Lupine Protein

In data supplied by the Personal Care Products Council, hydrolyzed lupine protein has < 0.042 µg/kg benzo[a]pyrene; < 0.10 ppm arsenic; 0.70 ppm barium; < 0.10 ppm cadmium; < 0.20 ppm chromium; < 0.20 ppm cobalt; < 0.05 ppm lead; and < 0.10 ppm mercury.¹⁴ Aflatoxins B&G combined were < 1.86 µg/kg. The absence of organochlorinated, organophosphorated, and organosulfur compounds from pesticides was noted.

Hydrolyzed Silk Protein

A supplier reported that hydrolyzed silk protein (MW = 300) may contain heavy metals and arsenic at levels ≤ 4 ppm and 0.4 ppm, respectively.¹⁹ Hydrolyzed silk protein with MW = 650 may contain heavy metals and arsenic at ≤ 10 ppm and 1 ppm, respectively.²⁰

Hydrolyzed Soy Protein

A supplier reported that hydrolyzed soy protein (MW = 300) may contain heavy metals, arsenic, and iron at levels ≤ 10 ppm, 1 ppm, and 10 ppm, respectively.²³

Hydrolyzed Wheat Protein

A supplier reported that hydrolyzed wheat protein (MW = 350) may contain heavy metals and arsenic at ≤ 5 ppm and 0.5 ppm, respectively.²⁴

USE

Cosmetic

The hydrolyzed plant and animal proteins and related salts discussed in this safety assessment function primarily as hair conditioning agents and skin conditioning agents (miscellaneous) in cosmetic formulations.²⁶ Additional functions may include nail conditioning agents (calcium hydrolyzed collagen and hydrolyzed keratin), light stabilizers (hydrolyzed lupine protein), and film formers (hydrolyzed gadidae protein and hydrolyzed wheat protein).

Table 3 presents the current product-formulation data for hydrolyzed proteins. According to information supplied to the Food and Drug Administration (FDA) by industry as part of the Voluntary Cosmetic Registration Program (VCRP), hydrolyzed wheat protein has the most reported uses in cosmetic and personal care products, with a total of 1055; approximately half of those uses are in non-coloring hair products.²⁷ Hydrolyzed soy protein has the second greatest number of overall uses reported, with a total of 681; again, approximately half of those uses are in non-coloring hair products.

In the Personal Care Products Council's use concentration survey, hydrolyzed wheat protein had a wide maximum use concentration range of 2.0×10^{-5} to 1.7%, with the 1.7% reported in rinse-off non-coloring hair products.²⁸ Hydrolyzed soy protein had a wide maximum use concentration range of 2.0×10^{-5} to 4%, with the 4% reported in face and neck skin care preparations. All other use concentrations that were reported had similar ranges.

In some cases, reports of uses were received from the VCRP, but no concentration of use data were available. For example, hydrolyzed fibronectin was reported to be used in 10 formulations, but no use concentration data were available. In other cases, no reported uses were received from the VCRP, but a use concentration was provided in the industry survey. For example, hydrolyzed casein was not reported in the VCRP database to be in use, but the industry survey indicated that it is used in formulations at maximum concentrations ranging from 0.0001% to 0.02%. It should be presumed that hydrolyzed casein is used in at least one cosmetic formulation.

Non-Cosmetic

The FDA determined that the use of peptones as direct food substances is generally recognized as safe (GRAS). These GRAS peptones are defined as “the variable mixture of polypeptides, oligopeptides, and amino acids that are produced by partial hydrolysis of casein, animal tissue, soy protein isolate, gelatin, defatted fatty tissue, egg albumin, or lactalbumin (whey protein) (21 CFR §184.1553).

Conversely, the FDA prohibits the use of cattle spinal cord in human food (21 CFR §189.5).

The FDA defines the term “protein” to mean any α -amino acid polymer with a specific defined sequence that is greater than 40 amino acids in size.²⁹ The FDA considers a “peptide” to be any polymer composed of 40 or fewer amino acids.

The FDA requires allergen labeling when major allergens, such as milk and soy, are included in food.³⁰

TOXICOKINETICS

Hydrolyzed Milk Protein and Hydrolyzed Soy Protein

While no experimental data were available for the dermal absorption of hydrolyzed milk protein and hydrolyzed soy protein, it was noted that gastro-intestinal absorption allows for significantly higher bioavailability than dermal absorption.^{31,32} In worst-case scenarios of oral exposures greater than 2000 mg/kg, no signs of systemic toxicity were observed and therefore it was concluded that no systemic toxicity would occur for cutaneous exposure. It was also noted that hydrolyzed soy protein has a high molecular weight and polarity, which would allow for limited skin penetration and consequent systemic availability.

Hydrolyzed Soy Protein

Biologically active peptides were derived from the soy protein, by treatment with various proteases, including glycinin.²¹ These biological activities included angiotensin converting enzyme (ACE) inhibitory, anti-thrombotic, surface tension, and antioxidant properties.

TOXICOLOGICAL STUDIES

Most of the hydrolyzed proteins in this assessment are found in the foods we consume daily, and the daily exposure from food use would result in a much larger systemic dose than that resulting from use in cosmetic products. These ingredients would be safe if absorbed into the body. Consequently, single dose toxicity, repeated dose toxicity, or other systemic toxicity studies are not addressed in this report.

The safety focus of use of these hydrolyzed proteins as cosmetic ingredients is on the potential for irritation and sensitization.

GENOTOXICITY

Hydrolyzed Keratin

The potential of hydrolyzed keratin (MW = 3000; 14% peptide content) to induce gene mutation was studied in *Salmonella typhimurium* strains TA 98, TA 100, TA 1535, and TA 1537 and in *Escherichia coli* strain WP2 uvrA using the reverse mutation assay.³³ The assay was performed with and without S9 metabolic activation at concentrations up to 5000 μ g/plate. No positive mutagenic responses were observed with or without S9. It was concluded that hydrolyzed keratin was not mutagenic with or without metabolic activation.

Hydrolyzed Milk Protein

The potential of hydrolyzed milk protein to induce gene mutation was studied in *Salmonella typhimurium* strains TA 98, TA 100, TA 1535, and TA 1537 with and without S9 metabolic activation.³¹ Concentrations were tested up to 5000 μ g/plate. The test material did not induce reverse mutations with and without S9. It was concluded that hydrolyzed milk protein was not mutagenic.

Hydrolyzed Soy Protein

Hydrolyzed soy protein was analyzed for mutagenic potential in an assay using *Salmonella typhimurium* TA 1535/pSK1002 with and without S9 metabolic activation.³² Concentrations tested were 625, 1250, 2500, or 5000 µg/ml. No sign of mutagenicity was observed with or without S9. It was concluded that hydrolyzed soy protein was not mutagenic.

IRRITATION AND SENSITIZATION

Irritation

Dermal

Non-human and human irritation studies are presented in Table 4.

Ocular

Non-human and human ocular irritation studies are presented in Table 5.

Sensitization

Non-human and human sensitization studies are presented in Table 6.

Phototoxicity

Phototoxicity studies are presented in Table 7.

CASE STUDIES

Hydrolyzed Keratin

A 22-year-old woman was reported to have a severe allergic reaction that included marked periorbital edema and swollen, sore, and itchy eyes and hands following use of a hair conditioner.³⁴ Prick testing elicited a strong positive (10 mm) wheal-and-flare response to the hair conditioner, which contained stearyltrimonim hydrolyzed animal protein. Further prick testing showed further reactions to the quaternary hydrolyzed protein as well as to shampoos and conditioners that contained gelatin keratin amino acids, hydrolyzed keratin, and/or hydrolyzed collagen. Patch tests using the European standard series and a series of 15 common bases of medicines and cosmetics were negative.

Hydrolyzed Wheat Protein

Three cases of wheat-dependent exercise-induced anaphylaxis (WDEIA) were reported in Japan.³⁵ The 3 female patients had used the same brand of soap that contained hydrolyzed wheat protein. Skin prick tests reveal positive reactions to 0.1% of the soap solution in physiological saline solution and to 100 µg/mL hydrolyzed wheat protein in physiological saline solution. Western blotting of the patients' sera IgE found serum positive reaction to the hydrolyzed wheat protein. The researchers concluded that the WDEIA was caused by cross reaction to wheat protein.

In another case study, a 42-year-old woman reported an intense burning sensation over her face, neck, and scalp several hours after applying a moisturizing cream that contained hydrolyzed wheat protein.³⁶ Patch testing with the diluted ingredients from the moisturizing cream resulted in a positive reaction (D2+, D4+) to 50% aq. hydrolyzed wheat protein. No reactions were observed from skin prick testing to standardized wheat extract or contact urticaria testing with hydrolyzed wheat protein.

Contact urticaria was reported in a 46-year-old woman.³⁷ The patient developed the symptoms 3 months prior to consulting her physician after applying an eyelid cream and a body moisturizer that contained hydrolyzed wheat proteins. Strong positive reactions were observed from the preserved food, wheat gluten that was in the food, the cosmetic creams, and hydrolyzed wheat protein. Further investigation revealed that the hydrolyzed wheat proteins in the cosmetic creams were from the same manufacturer as the gluten in the preserved food.

A 27-year-old woman was reported to have a pruritic, erythematous, urticarial rash that became increasingly more intense after subsequent use of a moisturizing body cream that contained hydrolyzed wheat protein.³⁸ Skin prick tests with common inhalant allergens, natural rubber latex, and cereal grains including wheat were negative. Also negative were the results of prick tests to a series of 21 protein allergens from plant and animal sources that included hen's egg, cow's milk, milk casein, almond, silk protein, aloe gel, papaya fruit, and hydrolyzed collagen. Total serum immunoglobulin (Ig)E was slightly elevated. The individual components of the body cream tested negative in an open application test, but a skin prick test was positive (8 mm) to hydrolyzed wheat protein. Further IgE testing found that specific binding occurred to wheat hydrolysate.

In another case study, a 64-year-old woman was reported to have itchy, erythematous, edematous lesions on the eyelids, face, and neck following use of a moisturizing cosmetic cream.³⁹ The patient was patch tested with

the (GEIDC) standard and cosmetics series, the cosmetic cream, and the cream's individual ingredients. Positive reactions (++) were observed to nickel sulfate, the cosmetic cream (tested neat), and to an ingredient of the cream, hydrolyzed wheat protein (10% aq.). Open testing with hydrolyzed wheat protein (10% aq.) was negative at 30 min.

The sensitization of 9 female patients to hydrolyzed wheat protein were studied with skin reactivity and IgE testing.⁴⁰ The patients all experienced contact urticaria after using cosmetic products containing hydrolyzed wheat protein. Six of the patients had also experienced generalized urticaria or anaphylaxis to food containing hydrolyzed wheat protein, but all of the patients could tolerate traditional wheat products, bread, pastries, etc. All of the patients had low to moderate levels of IgE specific to wheat flour or gluten. Skin tests (open or prick) confirmed the sensitivity to hydrolyzed wheat protein and tolerance to unmodified wheat proteins. Immunoblotting analyses found that all of the patients reacted with almost all of the hydrolyzed wheat protein preparations tested, with most reactions occurring with large random peptide aggregates. Unlike the skin tests, IgE reactions were observed in the immunoblotting analyses with unmodified wheat proteins. Reactions in the immunoblotting analyses were observed always with salt soluble proteins, but varied with gluten proteins and were not observed with gliadins in patients without associated immediate reactions to food containing hydrolyzed wheat protein. The researchers noted that IgE reacted mostly with large aggregates, which suggests that hydrolysis of the proteins did not destroy pre-existing epitopes and may have created multiepitopic entities, some with charged groups, that allows for bridging of IgE on mastocytes and basophiles and increased solubility and transport.

SUMMARY

Hydrolyzed proteins derived from plant and animal sources function primarily as skin and hair conditioning agents in personal care products. These protein derivatives are prepared by subjecting animal or vegetable proteins to enzymatic or other chemical, partial hydrolyses.

Hydrolyzed wheat protein has the most reported uses in cosmetic and personal care products, with a total of 1055; approximately half of those uses are in non-coloring hair products. Hydrolyzed soy protein has the second greatest number of overall uses reported, with a total of 681; again, approximately half of those uses are in non-coloring hair products

In the Personal Care Products Council's use concentration survey, hydrolyzed wheat protein had a wide maximum use concentration range of 2.0×10^{-5} to 1.7%, with the 1.7% reported in rinse-off non-coloring hair products. Hydrolyzed soy protein had a wide maximum use concentration range of 2.0×10^{-5} to 4%, with the 4% reported in face and neck skin care preparations. All other use concentrations that were reported had similar ranges.

The FDA determined the use of peptones as direct food substances are GRAS, but prohibited use of cattle spinal cord from use in human food.

Biologically active peptides may be derived from hydrolysis of soy protein.

In dermal irritation studies, hydrolyzed amaranth protein, hydrolyzed avocado protein, hydrolyzed elastin, hydrolyzed hazelnut protein, hydrolyzed keratin, hydrolyzed lupine protein, hydrolyzed milk protein, hydrolyzed silk, hydrolyzed soy protein, hydrolyzed vegetable protein, and hydrolyzed wheat protein were not irritants.

Ocular irritation studies of many of the hydrolyzed proteins found these ingredients to be mostly non-irritating.

HRIPT studies of hydrolyzed protein ingredients concluded that these ingredients were not dermal irritants or sensitizers.

Hydrolyzed avocado protein, hydrolyzed hazelnut protein, hydrolyzed lupine protein, hydrolyzed milk protein, and hydrolyzed silk were not phototoxins and/or photosensitizers.

Several cases of allergic reactions were reported in women who had used personal care products that contained hydrolyzed wheat protein. One case study reported a positive reaction a hair conditioner that contained hydrolyzed keratin in a prick test.

TABLES AND FIGURES

Table 1. Definitions and functions of the ingredients in this safety assessment.²⁶
(The italicized text below represents additions made by CIR staff.)

Ingredient CAS No.	Definition	Function
Ammonium Hydrolyzed Collagen 68951-88-2	Ammonium Hydrolyzed Collagen is the ammonium salt of Hydrolyzed Collagen. <i>Wherein Hydrolyzed Collagen is defined as the hydrolysate of animal or fish collagen derived by acid, enzyme or other method of hydrolysis. Hydrolyzed Collagen is characterized by a significant level of hydroxyproline residues.</i>	Hair conditioning Agent; Skin-Conditioning Agent - Misc.
Calcium Hydrolyzed Collagen	Calcium Hydrolyzed Collagen is the calcium salt of Hydrolyzed Collagen. <i>Wherein Hydrolyzed Collagen is the partial hydrolysate of animal or fish collagen derived by acid, enzyme or other method of hydrolysis. Hydrolyzed Collagen is characterized by a significant level of hydroxyproline residues.</i>	Nail Conditioning Agent; Skin-Conditioning Agent - Misc
Hydrolyzed Actin 73049-73-7	Hydrolyzed Actin is the <i>partial</i> hydrolysate of actin derived by acid, enzyme or other method of hydrolysis.	Hair conditioning Agent; Skin-Conditioning Agent - Misc.
Hydrolyzed Albumen 73049-73-7	Hydrolyzed Albumen is the <i>partial</i> hydrolysate of Albumen derived by acid, enzyme or other method of hydrolysis. <i>Wherein Albumen is defined as the dried whites of chicken eggs.</i>	Skin-Conditioning Agent - Misc.
Hydrolyzed Amaranth Protein	Hydrolyzed Amaranth Protein is the <i>partial</i> hydrolysate of amaranth protein derived by acid, enzyme or other method of hydrolysis.	Skin-Conditioning Agent - Misc.
Hydrolyzed Avocado Protein	Hydrolyzed Avocado Protein is the <i>partial</i> hydrolysate of avocado protein derived by acid, enzyme or other method of hydrolysis.	Skin-Conditioning Agent - Misc.
Hydrolyzed Barley Protein	Hydrolyzed Barley Protein is the <i>partial</i> hydrolysate of barley protein derived by acid, enzyme or other method of hydrolysis.	Hair conditioning Agent; Skin-Conditioning Agent - Misc.
Hydrolyzed Brazil Nut Protein	Hydrolyzed Brazil Nut Protein is the <i>partial</i> hydrolysate of brazil nut protein derived by acid, enzyme or other method of hydrolysis.	Hair conditioning Agent; Skin-Conditioning Agent - Misc.
Hydrolyzed Casein 65072-00-6 73049-73-7 [generic to peptides obtained by the hydrolysis of animal tissue proteins]	Hydrolyzed Casein is the <i>partial</i> hydrolysate of Casein derived by acid, enzyme or other method of hydrolysis. <i>Wherein, Casein is a mixture of phosphoproteins obtained from cow's milk.</i>	Hair conditioning Agent; Skin-Conditioning Agent - Misc.
Hydrolyzed Conalbumin	Hydrolyzed Conalbumin is the <i>partial</i> hydrolysate of conalbumin derived by acid, enzyme or other method of hydrolysis. <i>Wherein, conalbumin is an avian egg protein, also known as ovotransferrin.</i>	Skin-Conditioning Agent - Misc.
Hydrolyzed Conchiolin Protein 73049-73-7	Hydrolyzed Conchiolin Protein is the <i>partial</i> hydrolysate of conchiolin, a peptide covering the inorganic portion of the pearl shell, derived by acid, enzyme or other method of hydrolysis.	Hair conditioning Agent; Skin-Conditioning Agent - Misc.
Hydrolyzed Cottonseed Protein	Hydrolyzed Cottonseed Protein is the <i>partial</i> hydrolysate of cottonseed protein derived by acid, enzyme or other method of hydrolysis.	Hair conditioning Agent; Skin-Conditioning Agent - Misc.
Hydrolyzed Egg Protein 73049-73-7	Hydrolyzed Egg Protein is the <i>partial</i> hydrolysate of egg protein derived by acid, enzyme or other method of hydrolysis.	Hair conditioning Agent; Skin-Conditioning Agent - Misc.
Hydrolyzed Elastin 73049-73-7 91080-18-1 100085-10-7	Hydrolyzed Elastin is the <i>partial</i> hydrolysate of elastin derived by acid, enzyme or other method of hydrolysis. <i>Wherein, Elastin is a fibrous protein found in the connective tissue of animals.</i>	Hair conditioning Agent; Skin-Conditioning Agent - Misc.
Hydrolyzed Extensin 73049-73-7	Hydrolyzed Extensin is the <i>partial</i> hydrolysate of extensin protein derived by acid, enzyme or other method of hydrolysis. <i>Wherein, extensins are defined as wall-located, basic, hydroxyproline rich structural glycoproteins with alternating hydrophilic and hydrophobic motifs.⁴¹</i>	Hair conditioning Agent; Skin-Conditioning Agent - Misc.
Hydrolyzed Fibroin	Hydrolyzed Fibroin is the <i>partial</i> hydrolysate of Fibroin derived by acid, enzyme or other method of hydrolysis. <i>Wherein, Fibroin is a protein filament produced by the silkworm, Bombyx mori which together with Sericin composes Silk.</i>	Hair conditioning Agent; Skin-Conditioning Agent - Misc.
Hydrolyzed Fibronectin 73049-73-7 100085-35-6	Hydrolyzed Fibronectin is the <i>partial</i> hydrolysate of Fibronectin derived by acid, enzyme or other method of hydrolysis. <i>Wherein, Fibronectin is a glycoprotein found in connective tissues, basement membranes, in plasma and other body fluids.</i>	Hair conditioning Agent; Skin-Conditioning Agent - Misc.

Table 1. Definitions and functions of the ingredients in this safety assessment.²⁶
(The italicized text below represents additions made by CIR staff.)

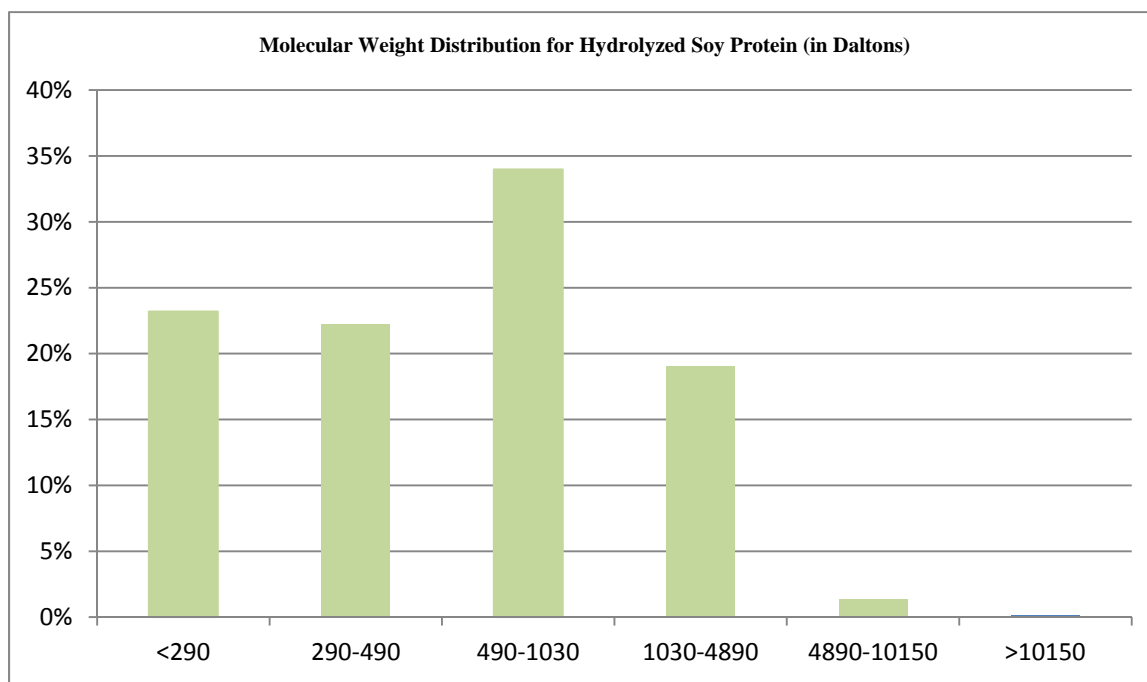
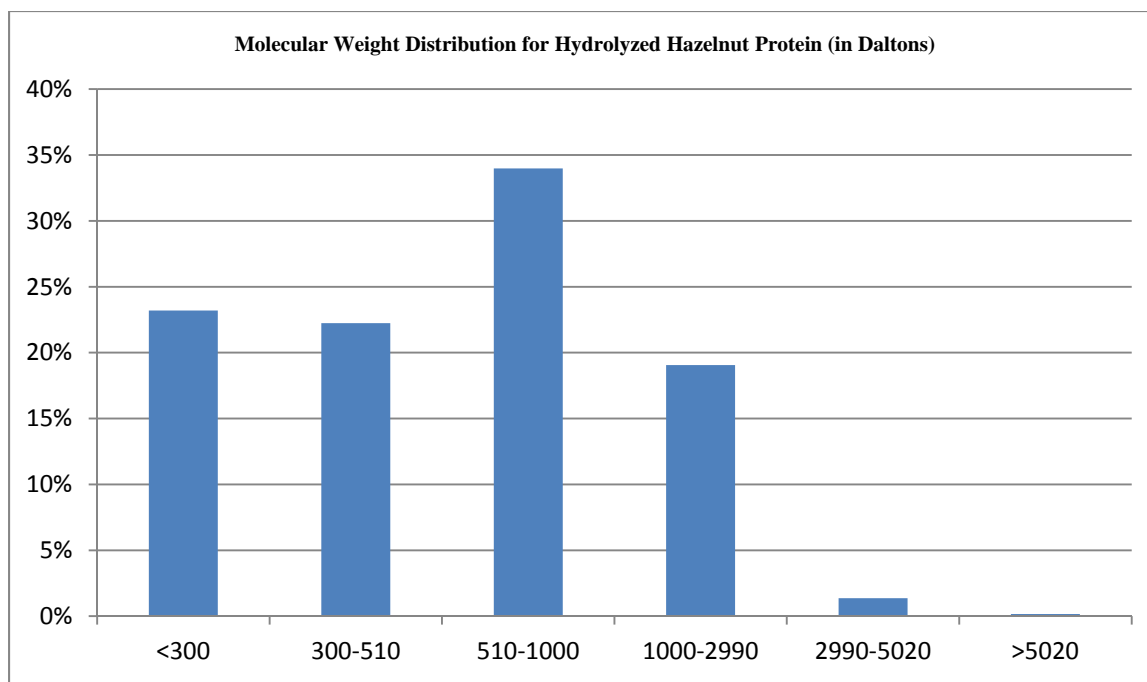
Ingredient CAS No.	Definition	Function
Hydrolyzed Gadidae Protein 73049-73-7	Hydrolyzed Gadidae Protein is the <i>partial</i> hydrolysate of the skin and flesh of the fish of the family, Gadidae, derived by acid, enzyme or other method of hydrolysis. <i>Wherein, the family Gadidae is a family of fish in the order Gadiformes, including cod, haddock, pollock, and hake.</i>	Film formers; Skin-Conditioning Agent - Emollient; Skin-Conditioning Agent - Misc.
Hydrolyzed Gelatin [68410-45-7]	Hydrolyzed Gelatin is the <i>partial</i> hydrolysate of Gelatin derived by acid, enzyme or other method of hydrolysis.	Skin-Conditioning Agent - Misc.
Hydrolyzed Hair Keratin 65997-21-9 73049-73-7	Hydrolyzed Hair Keratin is the <i>partial</i> hydrolysate of human hair keratin derived by acid, enzyme or other method of hydrolysis.	Hair conditioning Agent; Skin-Conditioning Agent - Misc.
Hydrolyzed Hazelnut Protein	Hydrolyzed Hazelnut Protein is the <i>partial</i> hydrolysate of hazelnut protein derived by acid, enzyme, or other method of hydrolysis.	Skin-Conditioning Agent - Misc.
Hydrolyzed Hemoglobin 73049-73-7	Hydrolyzed Hemoglobin is the <i>partial</i> hydrolysate of hemoglobin obtained by acid, enzyme or other method of hydrolysis.	Hair conditioning Agent; Skin-Conditioning Agent - Misc.
Hydrolyzed Hemp Seed Protein	Hydrolyzed Hemp Seed Protein is the <i>partial</i> hydrolysate of hemp seed protein derived by acid, enzyme or other method of hydrolysis.	Hair conditioning Agent; Skin-Conditioning Agent - Misc.
Hydrolyzed Honey Protein	Hydrolyzed Honey Protein is the <i>partial</i> hydrolysate of honey protein derived by acid, enzyme or other method of hydrolysis.	Hair conditioning Agent; Skin-Conditioning Agent - Misc.
Hydrolyzed Jojoba Protein 100684-35-3	Hydrolyzed Jojoba Protein is the <i>partial</i> hydrolysate of jojoba seed protein derived by acid, enzyme or other method of hydrolysis.	Hair conditioning Agent; Skin-Conditioning Agent - Emollient
Hydrolyzed Keratin 69430-36-0 73049-73-7	Hydrolyzed Keratin is the <i>partial</i> hydrolysate of keratin derived by acid, enzyme or other method of hydrolysis.	Hair Conditioning Agent; Nail Conditioning Agent; Skin-Conditioning Agent-Misc.
Hydrolyzed Lactalbumin 68458-87-7 73049-73-7	Hydrolyzed Lactalbumin is the <i>partial</i> hydrolysate of milk albumins derived by acid, enzyme, or other method of hydrolysis.	Skin-Conditioning Agent - Misc.
Hydrolyzed Lupine Protein 73049-73-7	Hydrolyzed Lupine Protein is the <i>partial</i> hydrolysate of lupine protein derived by acid, enzyme or other method of hydrolysis.	Hair Conditioning Agent; Light Stabilizer; Skin-Conditioning Agent - Misc.
Hydrolyzed Maple Sycamore Protein 73049-73-7	Hydrolyzed Maple Sycamore Protein is the <i>partial</i> hydrolysate of the protein derived from the maple sycamore tree, <i>Acer pseudoplatanus</i> , obtained by acid, enzyme, or other method of hydrolysis.	Hair Conditioning Agent; Skin-Conditioning Agent-Humectant; Skin-Conditioning Agent-Misc.
Hydrolyzed Milk Protein 92797-39-2	Hydrolyzed Milk Protein is the <i>partial</i> hydrolysate of milk protein derived by acid, enzyme or other method of hydrolysis.	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
Hydrolyzed Oat Protein 151661-87-9	Hydrolyzed Oat Protein is the <i>partial</i> hydrolysate of oat protein derived by acid, enzyme or other method of hydrolysis.	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
Hydrolyzed Pea Protein 222400-29-5 227024-36-4	Hydrolyzed Pea Protein is the <i>partial</i> hydrolysate of pea protein derived by acid, enzyme or other method of hydrolysis.	Hair Conditioning Agent; Skin-Conditioning Agent-Emollient; Skin-Conditioning Agent-Misc.
Hydrolyzed Potato Protein 169590-59-4	Hydrolyzed Potato Protein is the <i>partial</i> hydrolysate of potato protein derived by acid, enzyme or other method of hydrolysis.	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
Hydrolyzed Reticulin 73049-73-7 99924-37-5	Hydrolyzed Reticulin is the <i>partial</i> hydrolysate of the reticulin portion of animal connective tissue derived by acid, enzyme or other method of hydrolysis.	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
Hydrolyzed Royal Jelly Protein	Hydrolyzed Royal Jelly Protein is the <i>partial</i> hydrolysate of the proteins obtained from Royal Jelly derived by acid, enzyme or other method of hydrolysis. <i>Wherein, Royal Jelly is the pharyngeal secretion of worker bees.</i>	Skin-Conditioning Agent - Misc.
Hydrolyzed Sericin 73049-73-7	Hydrolyzed Sericin is the <i>partial</i> hydrolysate of Sericin derived by acid, enzyme or other method of hydrolysis. <i>Wherein, Sericin is a protein isolated from the silk produced by the silk worm, Bombyx mori.</i>	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
Hydrolyzed Serum Protein 73049-73-7	Hydrolyzed Serum Protein is the <i>partial</i> hydrolysate of Serum Protein derived by acid, enzyme or other method of hydrolysis. <i>Wherein, Serum Protein is the protein or protein fraction obtained from blood plasma.</i>	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
Hydrolyzed Sesame Protein	Hydrolyzed Sesame Protein is the <i>partial</i> hydrolysate of sesame protein derived by acid, enzyme or other method of hydrolysis.	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.

Table 1. Definitions and functions of the ingredients in this safety assessment.²⁶
(The italicized text below represents additions made by CIR staff.)

Ingredient CAS No.	Definition	Function
Hydrolyzed Silk 73049-73-7 96690-41-4	Hydrolyzed Silk is the <i>partial</i> hydrolysate of silk protein derived by acid, enzyme or other method of hydrolysis.	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
Hydrolyzed Soy Protein 68607-88-5	Hydrolyzed Soy Protein is the <i>partial</i> hydrolysate of soy protein derived by acid, enzyme or other method of hydrolysis.	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
Hydrolyzed Soymilk Protein	Hydrolyzed Soymilk Protein is the <i>partial</i> hydrolysate of the proteins obtained from Soymilk derived by acid, enzyme or other method of hydrolysis.	Skin-Conditioning Agent - Misc.
Hydrolyzed Spinal Protein 73049-73-7	Hydrolyzed Spinal Protein is the <i>partial</i> hydrolysate of animal spinal cord protein derived by acid, enzyme or other method of hydrolysis.	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
Hydrolyzed Spongin	Hydrolyzed Spongin is the <i>partial</i> hydrolysate of spongin derived by acid, enzyme or other method of hydrolysis. <i>Wherein, spongin is a protein related to keratin that forms the skeletal structure of certain sponges.</i>	Skin-Conditioning Agent - Misc.
Hydrolyzed Sweet Almond Protein 100209-19-6	Hydrolyzed Sweet Almond Protein is the <i>partial</i> hydrolysate of sweet almond protein derived by acid, enzyme or other method of hydrolysis.	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
Hydrolyzed Vegetable Protein 73049-73-7 100209-45-8	Hydrolyzed Vegetable Protein is the <i>partial</i> hydrolysate of vegetable protein derived by acid, enzyme or other method of hydrolysis.	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
Hydrolyzed Wheat Gluten 100684-25-1	Hydrolyzed Wheat Gluten is the <i>partial</i> hydrolysate of Triticum Vulgare (Wheat) Gluten derived by acid, enzyme or other method of hydrolysis.	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
Hydrolyzed Wheat Protein 70084-87-6 100209-50-5 222400-28-4	Hydrolyzed Wheat Protein is the <i>partial</i> hydrolysate of wheat protein derived by acid, enzyme or other method of hydrolysis.	Film formers; Hair Conditioning Agent; Skin-Conditioning Agent - Misc.
Hydrolyzed Whey Protein	Hydrolyzed Whey Protein is the <i>partial</i> hydrolysate of Whey Protein derived by acid, enzyme or other method of hydrolysis.	Skin-Conditioning Agent - Misc.
Hydrolyzed Yeast Protein 100684-36-4 227025-31-2	Hydrolyzed Yeast Protein is the <i>partial</i> hydrolysate of yeast protein derived by acid, enzyme or other method of hydrolysis.	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
Hydrolyzed Yogurt Protein	Hydrolyzed Yogurt Protein is the <i>partial</i> hydrolysate of yogurt protein derived by acid, enzyme or other method of hydrolysis.	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
Hydrolyzed Zein	Hydrolyzed Zein is the <i>partial</i> hydrolysate of Zein derived by acid, enzyme or other method of hydrolysis. <i>Wherein, Zein is an alcohol-soluble protein obtained from corn, Zea mays.</i>	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
MEA-Hydrolyzed Collagen	MEA-Hydrolyzed Collagen is the monoethanolamine salt of Hydrolyzed Collagen. <i>Wherein Hydrolyzed Collagen is defined as the partial hydrolysate of animal or fish collagen derived by acid, enzyme or other method of hydrolysis. Hydrolyzed Collagen is characterized by a significant level of hydroxyproline residues.</i>	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
MEA-Hydrolyzed Silk	MEA-Hydrolyzed Silk is the monoethanolamine salt of Hydrolyzed Silk. <i>Wherein Hydrolyzed Silk is defined as the partial hydrolysate of silk protein derived by acid, enzyme or other method of hydrolysis.</i>	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
Sodium Hydrolyzed Casein	Sodium Hydrolyzed Casein is the sodium salt of Hydrolyzed Casein. <i>Wherein Hydrolyzed Collagen is defined as the partial hydrolysate of animal or fish collagen derived by acid, enzyme or other method of hydrolysis. Hydrolyzed Collagen is characterized by a significant level of hydroxyproline residues.</i>	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.
Zinc Hydrolyzed Collagen	Zinc Hydrolyzed Collagen is the zinc salt of Hydrolyzed Collagen. <i>Wherein Hydrolyzed Collagen is defined as the partial hydrolysate of animal or fish collagen derived by acid, enzyme or other method of hydrolysis. Hydrolyzed Collagen is characterized by a significant level of hydroxyproline residues.</i>	Hair Conditioning Agent; Skin-Conditioning Agent-Misc.

Table 2. Physical and chemical properties.

Property	Value	Reference
<i>Hydrolyzed Amaranth Protein</i>		
Molecular Weight (Da)	~1500	8
<i>Hydrolyzed Avocado Protein</i>		
Molecular Weight (Da)	300-1200 (50-70% w/w)	9
<i>Hydrolyzed Brazil Nut Protein</i>		
Molecular Weight (Da)	~150	42
<i>Hydrolyzed Hair Keratin</i>		
Molecular Weight (Da)	~400	42
<i>Hydrolyzed Hazelnut Protein</i>		
Molecular Weight (Da)	510-1000 (mode value)	12
<i>Hydrolyzed Keratin</i>		
Molecular Weight (Da)	~1000; 150; ~3000	25,42,43
<i>Hydrolyzed Lupine Protein</i>		
Molecular Weight (Da)	300-1200 (40-50% w/w)	14
<i>Hydrolyzed Milk Protein</i>		
Molecular Weight (Da)	~1000	43
<i>Hydrolyzed Soy Protein</i>		
Molecular Weight (Da)	300 (average); ~1000; 490-1030 (mode value)	12,23,43
<i>Hydrolyzed Corn and Soy Protein</i>		
Molecular Weight (Da)	~1000	43
<i>Hydrolyzed Silk</i>		
Molecular Weight (Da)	~1000; 300 (average)	19,43
<i>Hydrolyzed Sweet Almond Protein</i>		
Molecular Weight (Da)	~3000	25



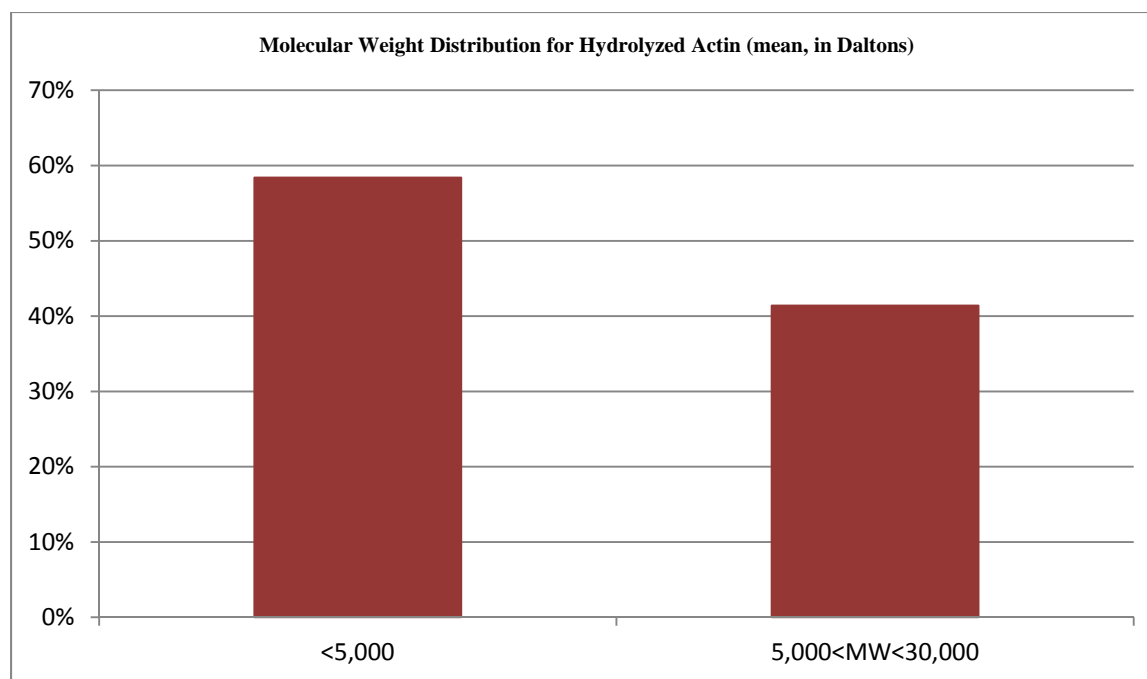
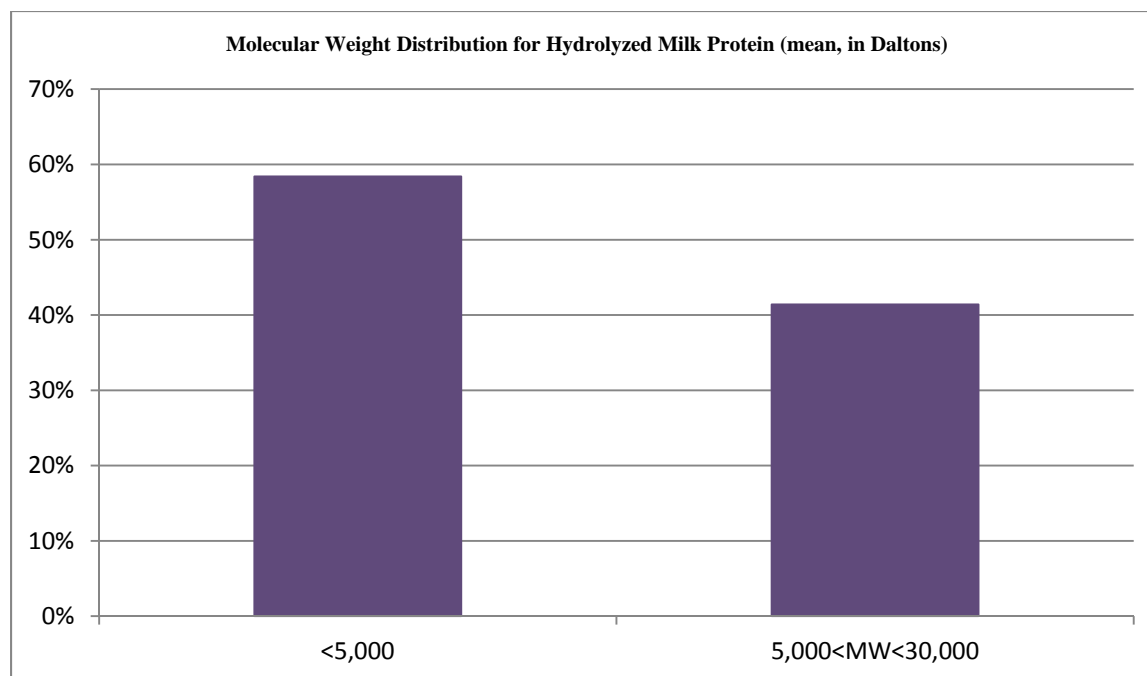


Figure 1. Molecular weight distribution of hydrolyzed proteins.^{12,44,45}

Table 3a. Frequency and concentration of use according to duration and type of exposure.^{27,28}

	Hydrolyzed Actin		Hydrolyzed Albumen		Hydrolyzed Amaranth Protein	
	# of Uses	Conc. of Use	# of Uses	Conc. of Use	# of Uses	Conc. of Use
Totals*	9	2	NR	0.002	1	0.02
<i>Leave-On</i>	9	2	NR	0.002	NR	0.02
<i>Rinse Off</i>	NR	NR	NR	NR	1	NR
<i>Diluted for (Bath) Use</i>	NR	NR	NR	NR	NR	NR
Eye Area	4	2	NR	NR	NR	NR
Incidental Ingestion	NR	NR	NR	NR	NR	NR
Incidental Inhalation-Sprays	NR	NR	NR	NR	NR	0.02 ^a
Incidental Inhalation-Powder	NR	NR	NR	NR	NR	NR
Dermal Contact	5	NR	NR	0.002	NR	NR
Deodorant (underarm)	NR	NR	NR	NR	NR	NR
Hair - Non-Coloring	2	NR	NR	NR	1	0.02
Hair-Coloring	NR	NR	NR	NR	NR	NR
Nail	NR	NR	NR	NR	NR	NR
Mucous Membrane	NR	NR	NR	NR	NR	NR
Baby Products	NR	NR	NR	NR	NR	NR

	Hydrolyzed Barley Protein		Hydrolyzed Brazil Nut Protein		Hydrolyzed Casein	
	# of Uses	Conc. of Use	# of Uses	Conc. of Use	# of Uses	Conc. of Use
Totals*	17	0.0002-0.005	10	0.00002-0.03	NR	0.0001-0.02
<i>Leave-On</i>	7	0.0002-0.003	5	0.002	NR	0.0001-0.02
<i>Rinse Off</i>	10	0.003-0.005	5	0.00002-0.03	NR	0.003-0.02
<i>Diluted for (Bath) Use</i>	NR	NR	NR	NR	NR	NR
Eye Area	NR	NR	NR	NR	NR	0.003
Incidental Ingestion	NR	NR	NR	NR	NR	NR
Incidental Inhalation-Spray	1	0.0002-0.003 ^b	NR	0.002 ^c	NR	0.0008 ^d
Incidental Inhalation-Powder	NR	NR	NR	NR	NR	NR
Dermal Contact	NR	NR	NR	NR	NR	0.0001-0.02
Deodorant (underarm)	NR	NR	NR	NR	NR	NR
Hair - Non-Coloring	17	0.0002-0.005	10	0.00002-0.03	NR	0.0001-0.02
Hair-Coloring	NR	NR	NR	NR	NR	NR
Nail	NR	NR	NR	NR	NR	NR
Mucous Membrane	NR	NR	NR	NR	NR	NR
Baby Products	NR	NR	NR	NR	NR	NR

	Hydrolyzed Conchiolin Protein		Hydrolyzed Cottonseed Protein		Hydrolyzed Elastin	
	# of Uses	Conc. of Use	# of Uses	Conc. of Use	# of Uses	Conc. of Use
Totals*	73	0.0000003-0.04	32	0.01-0.02	176	0.0000004-2.5
<i>Leave-On</i>	64	0.0000003-0.04	27	0.02	143	0.0000004-2.5
<i>Rinse Off</i>	9	0.000001-0.0009	5	0.01-0.02	33	0.000004-0.005
<i>Diluted for (Bath) Use</i>	NR	NR	NR	NR	NR	NR
Eye Area	8	0.000001-0.005	NR	NR	7	0.0004
Incidental Ingestion	NR	0.0001-0.0003	NR	NR	1	0.0004
Incidental Inhalation-Spray	NR	0.0000003-0.003 ^e	11	0.02 ^f	4	0.0004-0.05 ^g
Incidental Inhalation-Powder	3	0.00003-0.0005	NR	NR	2	0.000004
Dermal Contact	59	0.0000003-0.007	32	0.01-0.02	154	0.0000004-0.01
Deodorant (underarm)	NR	NR	NR	NR	NR	NR
Hair - Non-Coloring	10	0.00002-0.0009	NR	NR	21	0.0001-2.5
Hair-Coloring	NR	0.0003	NR	NR	NR	NR
Nail	4	0.04	NR	NR	NR	NR
Mucous Membrane	NR	0.0001-0.0003	3	0.02	6	0.0004
Baby Products	NR	NR	NR	NR	4	NR

Table 3a. Frequency and concentration of use according to duration and type of exposure.^{27,28}

	Hydrolyzed Extensin		Hydrolyzed Fibroin		Hydrolyzed Fibronectin	
	# of Uses	Conc. of Use	# of Uses	Conc. of Use	# of Uses	Conc. of Use
Totals*	30	0.2	1	0.001	10	NR
<i>Leave-On</i>	25	0.2	1	0.001	9	NR
<i>Rinse Off</i>	5	NR	NR	NR	1	NR
<i>Diluted for (Bath) Use</i>	NR	NR	NR	NR	NR	NR
Eye Area	2	NR	NR	NR	1	NR
Incidental Ingestion	NR	NR	NR	NR	NR	NR
Incidental Inhalation-Sprays	NR	NR	NR	NR	NR	NR
Incidental Inhalation-Powder	NR	NR	NR	NR	NR	NR
Dermal Contact	27	0.2	1	0.001	10	NR
Deodorant (underarm)	NR	NR	NR	NR	NR	NR
Hair - Non-Coloring	3	NR	NR	NR	NR	NR
Hair-Coloring	NR	NR	NR	NR	NR	NR
Nail	NR	NR	NR	NR	NR	NR
Mucous Membrane	NR	NR	NR	NR	NR	NR
Baby Products	NR	NR	NR	NR	NR	NR

	Hydrolyzed Gadidae Protein		Hydrolyzed Hair Keratin		Hydrolyzed Hazelnut Protein	
	# of Uses	Conc. of Use	# of Uses	Conc. of Use	# of Uses	Conc. of Use
Totals*	4	NR	32	0.005-0.05	24	0.3-1
<i>Leave-On</i>	3	NR	3	0.03-0.05	23	0.3
<i>Rinse Off</i>	1	NR	29	0.005	1	1
<i>Diluted for (Bath) Use</i>	NR	NR	NR	NR	NR	NR
Eye Area	1	NR	NR	NR	4	NR
Incidental Ingestion	NR	NR	NR	NR	NR	NR
Incidental Inhalation-Spray	NR	NR	NR	0.05 ^h	NR	NR
Incidental Inhalation-Powder	NR	NR	NR	NR	NR	NR
Dermal Contact	4	NR	NR	0.6	24	0.3
Deodorant (underarm)	NR	NR	NR	NR	NR	NR
Hair - Non-Coloring	NR	NR	32	0.005-0.05	NR	NR
Hair-Coloring	NR	NR	NR	NR	NR	1
Nail	NR	NR	NR	NR	NR	NR
Mucous Membrane	NR	NR	NR	NR	NR	NR
Baby Products	NR	NR	NR	NR	NR	NR

	Hydrolyzed Honey Protein		Hydrolyzed Jojoba Protein		Hydrolyzed Keratin	
	# of Uses	Conc. of Use	# of Uses	Conc. of Use	# of Uses	Conc. of Use
Totals*	NR	0.003-0.05	35	0.0003-0.03	399	0.00001-2.5
<i>Leave-On</i>	NR	0.003	15	0.003-0.03	181	0.00003-0.9
<i>Rinse Off</i>	NR	0.05	20	0.0003-0.02	217	0.00001-2.5
<i>Diluted for (Bath) Use</i>	NR	NR	NR	NR	1	NR
Eye Area	NR	0.003	1	NR	27	0.001-0.2
Incidental Ingestion	NR	NR	NR	NR	NR	NR
Incidental Inhalation-Spray	NR	NR	NR	0.003 ⁱ	17	0.00003-0.003 ^j
Incidental Inhalation-Powder	NR	NR	NR	NR	NR	NR
Dermal Contact	NR	0.003-0.05	9	NR	56	0.001-0.03
Deodorant (underarm)	NR	NR	NR	NR	1	NR
Hair - Non-Coloring	NR	NR	25	0.0003-0.03	281	0.00001-2.5
Hair-Coloring	NR	NR	NR	0.003	33	0.03-0.5
Nail	NR	NR	NR	NR	6	0.002-0.01
Mucous Membrane	NR	0.05	1	NR	20	0.002-0.03
Baby Products	NR	NR	NR	NR	NR	NR

Table 3a. Frequency and concentration of use according to duration and type of exposure.^{27,28}

	Hydrolyzed Lupine Protein		Hydrolyzed Milk Protein		Hydrolyzed Oat Protein	
	# of Uses	Conc. of Use	# of Uses	Conc. of Use	# of Uses	Conc. of Use
Totals*	79	0.005-7	123	0.0001-0.3	78	0.0001-0.3
<i>Leave-On</i>	73	0.005-7	71	0.02-0.3	31	0.0001-0.3
<i>Rinse Off</i>	6	0.005-0.2	45	0.0001-0.1	47	0.0001-0.2
<i>Diluted for (Bath) Use</i>	NR	NR	7	NR	NR	NR
Eye Area	11	0.02-0.2	2	NR	1	NR
Incidental Ingestion	NR	NR	3	NR	NR	NR
Incidental Inhalation-Sprays	NR	0.1	5	0.02 ^k	2	0.0001-0.01 ^l
Incidental Inhalation-Powder	NR	NR	NR	NR	NR	NR
Dermal Contact	72	0.005-0.3	96	0.0003-0.3	32	0.01-0.3
Deodorant (underarm)	NR	NR	2	NR	NR	NR
Hair - Non-Coloring	5	0.2-7	24	0.0001-0.02	42	0.0001-0.1
Hair-Coloring	NR	NR	NR	NR	NR	0.006
Nail	NR	NR	NR	NR	4	0.0001
Mucous Membrane	NR	NR	30	0.0003-0.02	20	0.2
Baby Products	NR	NR	NR	NR	NR	NR

	Hydrolyzed Pea Protein		Hydrolyzed Potato Protein		Hydrolyzed Sericin	
	# of Uses	Conc. of Use	# of Uses	Conc. of Use	# of Uses	Conc. of Use
Totals*	2	NR	15	0.000008-2.4	2	0.002
<i>Leave-On</i>	2	NR	15	0.00008-2.4	2	NR
<i>Rinse Off</i>	NR	NR	NR	0.000008-0.5	NR	0.002
<i>Diluted for (Bath) Use</i>	NR	NR	NR	NR	NR	NR
Eye Area	NR	NR	NR	NR	1	NR
Incidental Ingestion	NR	NR	NR	NR	NR	NR
Incidental Inhalation-Spray	NR	NR	2	NR	NR	NR
Incidental Inhalation-Powder	NR	NR	NR	NR	NR	NR
Dermal Contact	2	NR	15	0.000008-2.4	1	NR
Deodorant (underarm)	NR	NR	NR	NR	NR	NR
Hair - Non-Coloring	NR	NR	NR	NR	1	NR
Hair-Coloring	NR	NR	NR	NR	NR	0.002
Nail	NR	NR	NR	NR	NR	NR
Mucous Membrane	NR	NR	NR	NR	NR	NR
Baby Products	NR	NR	NR	NR	NR	NR

	Hydrolyzed Silk		Hydrolyzed Soy Protein		Hydrolyzed Soymilk Protein	
	# of Uses	Conc. of Use	# of Uses	Conc. of Use	# of Uses	Conc. of Use
Totals*	577	0.0000007-1	681	0.00002-4	7	0.0002
<i>Leave-On</i>	283	0.0002-1	419	0.0002-4	3	NR
<i>Rinse Off</i>	289	0.0000007-0.5	262	0.00002-1.8	4	0.0002
<i>Diluted for (Bath) Use</i>	5	0.002-0.004	NR	NR	NR	NR
Eye Area	33	0.0000007-0.5	46	0.07-3.5	NR	NR
Incidental Ingestion	6	0.004	1	1.1	NR	NR
Incidental Inhalation-Spray	33	0.0007-0.05 ^m	13	0.0002-0.1 ⁿ	NR	NR
Incidental Inhalation-Powder	8	0.004	2	NR	NR	NR
Dermal Contact	291	0.0000007-0.5	283	0.001-4	2	NR
Deodorant (underarm)	1	NR	1	0.4	NR	NR
Hair - Non-Coloring	251	0.00005-1	314	0.00002-1	5	0.0002
Hair-Coloring	14	0.0002-0.05	32	0.005-1.8	NR	NR
Nail	1	0.0002-0.09	32	0.01-0.03	NR	NR
Mucous Membrane	82	0.0003-0.1	9	1.1	NR	NR
Baby Products	6	NR	NR	NR	NR	NR

Table 3a. Frequency and concentration of use according to duration and type of exposure.^{27,28}

	Hydrolyzed Sweet Almond Protein		Hydrolyzed Vegetable Protein		Hydrolyzed Wheat Gluten	
	# of Uses	Conc. of Use	# of Uses	Conc. of Use	# of Uses	Conc. of Use
Totals*	77	0.00002-1.5	153	0.0004-0.2	49	0.005-0.09
<i>Leave-On</i>	42	0.00002-1.5	55	0.03-0.1	12	0.005-0.09
<i>Rinse Off</i>	35	0.0003-0.5	98	0.0004-0.2	34	0.005-0.01
<i>Diluted for (Bath) Use</i>	NR	NR	NR	NR	3	NR
Eye Area	6	0.3	4	0.05	1	0.09
Incidental Ingestion	NR	NR	NR	NR	NR	NR
Incidental Inhalation-Sprays	1	0.00002 ^p	5	NR	NR	NR
Incidental Inhalation-Powder	NR	NR	2	NR	1	NR
Dermal Contact	38	0.0002-0.5	50	0.03-0.1	29	0.01-0.09
Deodorant (underarm)	NR	NR	NR	NR	NR	NR
Hair - Non-Coloring	35	0.00002-1.5	49	0.04-0.2	19	0.005
Hair-Coloring	1	NR	54	0.0004-0.2	1	NR
Nail	NR	NR	NR	NR	NR	NR
Mucous Membrane	2	NR	2	NR	18	NR
Baby Products	NR	NR	NR	NR	3	NR

	Hydrolyzed Wheat Protein		Hydrolyzed Whey Protein		Hydrolyzed Yeast Protein	
	# of Uses	Conc. of Use	# of Uses	Conc. of Use	# of Uses	Conc. of Use
Totals*	1055	0.00002-1.7	NR	0.02	57	0.001-2
<i>Leave-On</i>	524	0.00006-1	NR	0.02	39	0.008-1
<i>Rinse Off</i>	524	0.00002-1.7	NR	NR	18	0.001-2
<i>Diluted for (Bath) Use</i>	7	0.00002	NR	NR	NR	NR
Eye Area	66	0.01-0.9	NR	NR	5	0.3-1
Incidental Ingestion	12	0.008-0.03	NR	NR	NR	NR
Incidental Inhalation-Spray	19	0.0003-0.5 ^p	NR	NR	1	1 ^q
Incidental Inhalation-Powder	6	0.05	NR	NR	NR	NR
Dermal Contact	377	0.00002-1	NR	NR	40	NR
Deodorant (underarm)	NR	NR	NR	NR	NR	NR
Hair - Non-Coloring	533	0.0003-1.7	NR	0.02	17	NR
Hair-Coloring	80	0.002-0.3	NR	NR	NR	NR
Nail	28	0.002-0.04	NR	NR	NR	NR
Mucous Membrane	96	0.00002-0.1	NR	NR	NR	NR
Baby Products	2	NR	NR	NR	NR	NR

	Hydrolyzed Yogurt Protein		MEA-Hydrolyzed Collagen		Ammonium Hydrolyzed Animal Protein**	
	# of Uses	Conc. of Use	# of Uses	Conc. of Use	# of Uses	Conc. of Use
Totals*	8	0.1-1	NR	0.03-0.2	1	NR
<i>Leave-On</i>	NR	NR	NR	0.1-0.2	1	NR
<i>Rinse Off</i>	8	0.1-1	NR	0.03-0.06	NR	NR
<i>Diluted for (Bath) Use</i>	NR	NR	NR	NR	NR	NR
Eye Area	NR	NR	NR	NR	NR	NR
Incidental Ingestion	NR	NR	NR	NR	NR	NR
Incidental Inhalation-Spray	NR	NR	NR	NR	NR	NR
Incidental Inhalation-Powder	NR	NR	NR	NR	NR	NR
Dermal Contact	8	0.1-1	NR	0.03-0.2	NR	NR
Deodorant (underarm)	NR	NR	NR	NR	NR	NR
Hair - Non-Coloring	NR	NR	NR	0.06	1	NR
Hair-Coloring	NR	NR	NR	NR	NR	NR
Nail	NR	NR	NR	NR	NR	NR
Mucous Membrane	8	0.1-1	NR	NR	NR	NR
Baby Products	NR	NR	NR	NR	NR	NR

Table 3a. Frequency and concentration of use according to duration and type of exposure.^{27,28}

	Hydrolyzed Animal Protein**		Hydrolyzed Fish Protein**	
	# of Uses	Conc. of Use	# of Uses	Conc. of Use
Totals*	143	NR	1	NR
<i>Leave-On</i>	72	NR	1	NR
<i>Rinse Off</i>	71	NR	NR	NR
<i>Diluted for (Bath) Use</i>	NR	NR	NR	NR
Eye Area	4	NR	1	NR
Incidental Ingestion	NR	NR	NR	NR
Incidental Inhalation-Sprays	1	NR	NR	NR
Incidental Inhalation-Powder	NR	NR	NR	NR
Dermal Contact	57	NR	1	NR
Deodorant (underarm)	NR	NR	NR	NR
Hair - Non-Coloring	85	NR	NR	NR
Hair-Coloring	NR	NR	NR	NR
Nail	1	NR	NR	NR
Mucous Membrane	NR	NR	NR	NR
Baby Products	NR	NR	NR	NR

* 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.

**Not an INCI ingredient. Listed in the FDA VCRP.

NR = none reported

^a0.02% in an aerosol hair spray.

^b0.0002% in an aerosol hair spray, 0.003% in a pump hair spray.

^c0.002% in a pump hair spray.

^d0.0008 in a pump hairspray.

^e0.0002-0.0003% in an aerosol hair spray, 0.0003% in a pump hair spray, 0.003% in a pump spray tonics, dressings, and other hair grooming aid, 0.0001% in a body and hand spray, 0.0000003% in a moisturizing spray.

^f0.02% in a body and hand spray.

^g0.0004-0.05% in a pump hair spray.

^h0.05% in an aerosol hair spray.

ⁱ0.003% in a pump hair spray.

^j0.00003-0.003% in aerosol hair sprays.

^k0.02% in pump hair spray.

^l0.0001% in a pump spray tonic, dressing, and other hair grooming aid; 0.01% in a body and hand spray.

^m0.001-0.007% in pump hair sprays; 0.003-0.02% in spray tonics, dressings, and other hair grooming aids; 0.005% in other hair preparation spray; 0.007-0.01% in body and hand sprays; 0.05% in indoor tanning preparations.

ⁿ0.0002-0.02% in aerosol hair sprays; 0.002-0.04% in pump hair sprays; and 0.1% in tonics, dressings and other hair grooming aid spray.

^o0.000002% in a pump hair spray.

^p0.03-0.05% in aerosol hair sprays; 0.0003-0.5% in pump hair sprays; and 0.002-0.02% in spray tonics, dressings, and other hair grooming aids.

^q1% in an aftershave lotion spray.

Table 3b. Ingredients not reported to be in use.^{27,28}

Ammonium Hydrolyzed Collagen
Calcium Hydrolyzed Collagen
Hydrolyzed Avocado Protein
Hydrolyzed Conalbumin
Hydrolyzed Egg Protein
Hydrolyzed Gelatin
Hydrolyzed Hemoglobin
Hydrolyzed Hemp Seed Protein
Hydrolyzed Lactalbumin
Hydrolyzed Maple Sycamore Protein
Hydrolyzed Reticulin
Hydrolyzed Royal Jelly Protein
Hydrolyzed Serum Protein
Hydrolyzed Sesame Protein
Hydrolyzed Spinal Protein
Hydrolyzed Spongin
Hydrolyzed Zein
MEA-Hydrolyzed Silk
Sodium Hydrolyzed Casein
Zinc Hydrolyzed Collagen

Table 4. Dermal irritation studies.

Ingredient	Concentration	Method	Results	Reference
Non-Human				
Hydrolyzed Amaranth Protein	20% in water	EpiDerm MTT Viability assay	Non-irritating	46
Hydrolyzed Elastin	Neat (MW = 3000)	Draize primary dermal irritation study in 6 New Zealand white rabbits; occluded for 24 h	PII = 0.38. Not a primary irritant.	47
Hydrolyzed Hazelnut Protein	100%	Dermal irritation study performed under OECD Guideline 404	Non-irritating	12
Hydrolyzed Keratin	100% (MW = 3000)	EpiDerm skin model	Non-irritating	48
Hydrolyzed Keratin	Neat (MW = 500)	Draize primary dermal irritation study in 6 New Zealand white rabbits; occluded for 24 h	PII = 2.15. Not a primary irritant.	49
Hydrolyzed Keratin	Neat (MW = 125,000)	Draize primary dermal irritation study in 6 female New Zealand White rabbits; occluded for 24 h	PII = 2.0. Not a primary irritant.	50
Hydrolyzed Keratin	Neat (MW = 600)	Draize primary dermal irritation study in 6 New Zealand white rabbits; occluded for 24 h	PII = 0.0. Not a primary irritant.	51
Hydrolyzed Milk Protein	10% (v/v) aqueous dilution, pH 6.7	Dermal irritation study performed under OECD Guideline 404 in 6 White New Zealand rabbits; semi-occluded for 24 h	Non-irritating	31
Hydrolyzed Milk Protein	25% w/v in water (MW = 1500)	Primary skin irritation study in 6 female New Zealand White rabbits, occluded for 24 h	PII = 1.3. Not a primary irritant.	52
Hydrolyzed Silk	Neat (MW = 300)	Draize primary skin irritation study in 6 female New Zealand White rabbits; occluded for 24 h	PII = 1.1. Not a primary irritant.	53
Hydrolyzed Silk	Neat (MW = 1000)	Draize primary dermal irritation study in 6 New Zealand white rabbits; occluded for 24 h	PII = 0.65. Not a primary irritant.	54
Hydrolyzed Silk	10% dilution (MW = 300)	Red Blood Cell Assay (RBCA)	Non-irritating	55
Hydrolyzed Silk	Not reported (MW = 650)	Primary skin irritation test with 6 New Zealand white rabbits; occluded for 24 h	PII = 0.05. Not a primary irritant	56
Hydrolyzed Silk	Not reported (MW = 650)	Cumulative application skin test with 8 Hartley guinea pigs	Non-irritating	57
Hydrolyzed Soy Protein	Not reported	Dermal irritation study performed under OECD Guideline 404	Non-irritating	12
Hydrolyzed Soy Protein	20% in distilled water	Draize test in 6 male White New Zealand rabbits; occluded	Non-irritating	32
Hydrolyzed Soy Protein	Neat (MW = 2000)	Draize primary dermal irritation in 6 New Zealand white rabbits; occluded for 24 h	PII = 0.33. Not a primary irritant.	58
Hydrolyzed Soy Protein	Neat (MW = 300)	Primary dermal irritation in 6 New Zealand white rabbits; occluded for 24 h	PII = 0.46. Not a primary irritant	59
Hydrolyzed Vegetable Protein	100% (MW = 750)	EpiDerm skin model	Non-irritating	60
Hydrolyzed Wheat Protein	Neat (MW = 350)	Primary dermal irritation study in 6 New Zealand white rabbits; occluded for 24 h	PII = 0.50. Not a primary irritant	61
Human				
Hydrolyzed Avocado Protein	Not reported	3D human skin model (MTT + IL1 α) performed under OECD draft guidelines and ECVAM protocol; 10 volunteers	Well tolerated	9
Hydrolyzed Lupine Protein	Not reported	3D human skin model (MTT + IL1 α) performed under OECD draft guidelines and ECVAM protocol; 20 volunteers	Well tolerated	14
Hydrolyzed Milk Protein	5% aq. dilution	Human phototoxicity study in 10 volunteers with single topical application	No skin irritation	31
Hydrolyzed Silk	Neat (MW= 300)	20 subjects received a single	3/20 had slight erythema	62

Table 4. Dermal irritation studies.

Ingredient	Concentration	Method	Results	Reference
		dermal dose under occlusive conditions for 24 h	after 24 h, 1/20 had slight erythema after 48 h. Not a dermal irritant	
Hydrolyzed Silk	Not reported (MW = 650)	24 subjects received a single dermal dose under occlusive conditions for 24 h	Non-irritating	⁶³
Hydrolyzed Soy Protein	20% in distilled water	50 subjects received 9 topical applications over 3 weeks; 24 h in duration; occluded	Non-irritating	³²
Hydrolyzed Soy Protein	Neat (MW = 300)	20 female subjects received a single dermal dose under occlusive conditions for 24 h	Not a dermal irritant	⁶⁴
Hydrolyzed Wheat Protein	Not reported (MW = 350)	42 subjects received a single dermal dose under occlusive conditions for 48 h	Non-irritating	⁶⁵

Table 5. Ocular irritation studies.

Ingredient	Concentration	Method	Results	Reference
Non-Human				
Hydrolyzed Amaranth Protein	20% dilutions	EpiOcular MTT viability irritation study	Non-irritating	66
Hydrolyzed Avocado Protein	Not reported	HET-CAM method	Moderately irritating	9
Hydrolyzed Avocado Protein	Not reported	BCOP ocular irritation study	Not severely irritating	9
Hydrolyzed Avocado Protein	Not reported	Neutral red release assay	Negligible cytotoxicity	9
Hydrolyzed Elastin	Neat (MW = 3000)	Draize ocular irritation study in 6 New Zealand white rabbits; unrinsed eyes	Non-irritating	47
Hydrolyzed Hazelnut Protein	Neat	Ocular irritation study performed under OECD guideline 405	Non-irritating	12
Hydrolyzed Keratin	Neat (MW = 500)	Draize ocular irritation study in 6 New Zealand white rabbits; unrinsed eyes	Minimal ocular irritant	49
Hydrolyzed Keratin	Neat (MW = 125,000)	Draize ocular irritation study in 6 female New Zealand White rabbits; unrinsed eyes	Non-irritating	67
Hydrolyzed Keratin	Neat (MW = 600)	Draize ocular irritation study in 6 New Zealand white rabbits; unrinsed eyes	Non-irritating	51
Hydrolyzed Keratin	1%, 5% and 10% (MW = 3000)	HET-CAM method	Practically no irritation potential at 1% and 5%. Slight irritation potential at 10%.	68
Hydrolyzed Lupine Protein	Not reported	HET-CAM method	Weakly irritating	14
Hydrolyzed Lupine Protein	Not reported	BCOP ocular irritation study	Weakly irritating	14
Hydrolyzed Lupine Protein	Not reported	Neutral red release assay	Negligible cytotoxicity	14
Hydrolyzed Milk Protein	10% aq. dilution at pH 6.7	Ocular irritation study performed under OECD guideline 405 using 6 albino White New Zealand rabbits	Not irritating	31
Hydrolyzed Milk Protein	25% in distilled water (MW = 1500)	Ocular irritation study in 6 female New Zealand White rabbits; unrinsed eyes	Not irritating	69
Hydrolyzed Silk	Neat (MW = 300)	Ocular irritation study in 6 female New Zealand White rabbits; unrinsed eyes	Not irritating	70
Hydrolyzed Silk	Neat (MW = 1000)	Ocular irritation study in 6 New Zealand white rabbits; unrinsed eyes	Not irritating	54
Hydrolyzed Silk	2% dilution in deionized water (MW = 300)	HET-CAM method	Not irritating	71
Hydrolyzed Silk	Not reported (MW = 650)	Primary ocular irritation study in 6 New Zealand white rabbits	Not irritating	72
Hydrolyzed Silk	Not reported (MW = 650)	Cumulative application ocular test with 6 New Zealand white rabbits	Practically non-irritating	73
Hydrolyzed Soy Protein	20% dilution, w/v	HET-CAM method	Slightly irritating	32
Hydrolyzed Soy Protein	20% active matter in distilled water	Ocular irritation study performed under OECD guideline 405 in 3 albino White New Zealand rabbits	Very slight irritant reactions to the conjunctiva that appeared reversible in less than 72 h	32
Hydrolyzed Soy Protein	Neat	Ocular irritation study performed under OECD guideline 405	Very slight irritant	12
Hydrolyzed Soy Protein	Neat (MW = 2000)	Ocular irritation study in 6 New Zealand white rabbits; unrinsed eyes	Not irritating	58
Hydrolyzed Soy Protein	Not reported (MW = 300)	Ocular irritation study in 6 albino rabbits; unrinsed	Not a primary eye irritant	74
Hydrolyzed Vegetable Protein	25%, 50%, and 100% (MW = 750)	HET-CAM method	Practically no irritation potential	75
Hydrolyzed Wheat Protein	Neat (MW = 350)	Ocular irritation study in 6 albino rabbits	Not a primary eye irritant	76

Table 6. Dermal sensitization studies.

Ingredient	Concentration	Method	Results	Reference
Non-Human				
Hydrolyzed Avocado Protein	Not reported	LLNA	Non-sensitizing	9
Hydrolyzed Hazelnut Protein	100%	Guinea pig dermal sensitization study performed according to OECD guideline 406	Non-sensitizing	12
Hydrolyzed Hazelnut Protein	5%	Sensitization study using the Marzulli-Maibach method	Non-irritating and non-sensitizing	12
Hydrolyzed Lupine Protein	Not reported	LLNA	Non-sensitizing	14
Hydrolyzed Milk Protein	5% dilution in water, v/v, with 5% in adjuvant and water for intracutaneous induction and 100% for epicutaneous induction and challenge	Maximization test in male and female Pirbright white guinea pigs	No irritation or sensitization	31
Hydrolyzed Silk	25%, 50% or 100% in dimethyl formamide (MW =300)	LLNA	Non-sensitizing	77
Hydrolyzed Silk	Not reported (MW = 650)	Maximization test in male and female Hartley guinea pigs	Non-sensitizing	78
Hydrolyzed Soy Protein	20% for the intracutaneous and epicutaneous induction, 10% and 20% solutions for challenge	Maximization test in male and female albino Dunkin Hartley guinea pigs	No skin reactions	32
Human				
Hydrolyzed Amaranth Protein	Not reported	HRIPT with 108 subjects; semi-occlusive	No dermal irritation or sensitization	79
Hydrolyzed Avocado Protein	Not reported	HRIPT that utilized the Marzulli-Maibach method in 50 subjects	Non-sensitizing	9
Hydrolyzed Elastin	25% w/v in corn oil (MW = 3000)	HRIPT with 52 subjects; occlusive	No dermal irritation or sensitization	80
Hydrolyzed Keratin	Not reported (MW = 3000)	HRIPT with 51 subjects; semi-occlusive	No dermal irritation or sensitization	81
Hydrolyzed Lupine Protein	Not reported	HRIPT that utilized the Marzulli-Maibach method in 100 subjects	Non-sensitizing	14
Hydrolyzed Potato Protein	1.5% in a face cream	HRIPT with 100 subjects; occlusive	No dermal irritation or sensitization	82
Hydrolyzed Potato Protein	2.4% in a night cream	HRIPT with 100 subjects; occlusive	No dermal irritation or sensitization	82
Hydrolyzed Silk	10% dilution in distilled water (MW = 1000)	HRIPT with 57 subjects; semi-occlusive	No dermal irritation or sensitization	83
Hydrolyzed Silk	Not reported (MW = 1000)	HRIPT with 49 subjects; semi-occlusive	No dermal irritation or sensitization	84
Hydrolyzed Silk	20% in water (MW =300)	HRIPT with 48 subjects; occlusive	No dermal irritation or sensitization	85
Hydrolyzed Soy Protein	Not reported (MW = 300)	HRIPT in 50 subjects; occlusive	No dermal irritation or sensitization	86
Hydrolyzed Soy Protein	20% dilution	HRIPT in 41subjects; occlusive	No dermal irritation or sensitization	32
Hydrolyzed Wheat Protein	Not reported (MW = 350)	HRIPT in 52 subjects; occlusive	No dermal irritation or sensitization	87
Hydrolyzed Yeast Protein	0.0143% in a face cream	HRIPT in 100 subjects; occlusive	No dermal irritation or sensitization	82
Multiple Hydrolyzed Proteins including Hydrolyzed Collagen, Hydrolyzed Elastin, Hydrolyzed Keratin, Hydrolyzed Milk Protein, Hydrolyzed Wheat Protein, Hydrolyzed Sweet Almond Protein, and Quaternized Hydrolyzed Proteins	Not reported	Sensitization study of protein hydrolysates in hair care products in 3 groups of patients. Group 1 was comprised of 11 hairdressers with hand dermatitis, group2 was comprised of 2160 consecutive adults with suspected allergic respiratory disease, and group 3 was comprised of 28 adults with atopic dermatitis. Subjects submitted to scratch and/or prick tests.	Positive reactions observed in 12 female patients with atopic dermatitis from 3 of 22 protein hydrolysates. Reactions were to quaternized hydrolyzed collagen, quaternized hydrolyzed milk protein, and hydrolyzed collagen. No adverse reactions to hydrolyzed elastin, hydrolyzed keratin, hydrolyzed milk protein, hydrolyzed wheat protein, or hydrolyzed sweet almond protein were observed.	88

Table 7. Phototoxicity studies.				
Ingredient	Concentration	Method	Results	Reference
Hydrolyzed Avocado Protein	Not reported	3T3 NRU method	Not phototoxic	⁹
Hydrolyzed Hazelnut Protein	Not reported	3T3 NRU method	Not phototoxic	¹²
Hydrolyzed Lupine Protein	Not reported	3T3 NRU method	Not phototoxic	¹⁴
Hydrolyzed Milk Protein	5% aq. dilution, v/v	Photoirritation study in 10 subjects; occluded. After 24 h exposure, 1 treated site irradiated with UVA (320-400 nm) for 15 min, other site was control.	Not a photoirritant	³¹
Hydrolyzed Milk Protein	5% dilution in water, v/v	Photosensitization study in 29 subjects; 3 weeks of 6 induction patches in duplicate. After 24 h exposure, 1 treated site irradiated with UV (260-400 nm for 15 min, other site was control. After 2 week rest, challenge on virgin irradiated and non-irradiated sites.	Not a photosensitizer	³¹
Hydrolyzed Silk	Not reported (MW = 650)	Phototoxicity study in groups of 6 Hartley guinea pigs	Not phototoxic	⁸⁹
Hydrolyzed Silk	Not reported (MW = 650)	Photoallergenicity test in groups of 6 Hartley guinea pigs	Not photosensitizing	⁹⁰

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05G - Tonics, Dressings, and Other Hair Grooming Aids	977092855	AMMONIUM HYDROLYZED ANIMAL PROTEIN	1
03D - Eye Lotion	977172147	HYDROLYZED ACTIN	1
03F - Mascara	977172147	HYDROLYZED ACTIN	2
03G - Other Eye Makeup Preparations	977172147	HYDROLYZED ACTIN	1
05G - Tonics, Dressings, and Other Hair Grooming Aids	977172147	HYDROLYZED ACTIN	2
12C - Face and Neck (exc shave)	977172147	HYDROLYZED ACTIN	3
05F - Shampoos (non-coloring)	999003529	HYDROLYZED AMARANTH PROTEIN	1
03C - Eye Shadow	100085618	HYDROLYZED ANIMAL PROTEIN	1
03F - Mascara	100085618	HYDROLYZED ANIMAL PROTEIN	3
05A - Hair Conditioner	100085618	HYDROLYZED ANIMAL PROTEIN	36
05D - Permanent Waves	100085618	HYDROLYZED ANIMAL PROTEIN	5
05F - Shampoos (non-coloring)	100085618	HYDROLYZED ANIMAL PROTEIN	26
05G - Tonics, Dressings, and Other Hair Grooming Aids	100085618	HYDROLYZED ANIMAL PROTEIN	14
05I - Other Hair Preparations	100085618	HYDROLYZED ANIMAL PROTEIN	4
07A - Blushers (all types)	100085618	HYDROLYZED ANIMAL PROTEIN	1
07C - Foundations	100085618	HYDROLYZED ANIMAL PROTEIN	1
08C - Nail Creams and Lotions	100085618	HYDROLYZED ANIMAL PROTEIN	1
12A - Cleansing	100085618	HYDROLYZED ANIMAL PROTEIN	3
12C - Face and Neck (exc shave)	100085618	HYDROLYZED ANIMAL PROTEIN	10
12D - Body and Hand (exc shave)	100085618	HYDROLYZED ANIMAL PROTEIN	13
12F - Moisturizing	100085618	HYDROLYZED ANIMAL PROTEIN	7
12G - Night	100085618	HYDROLYZED ANIMAL PROTEIN	6
12H - Paste Masks (mud packs)	100085618	HYDROLYZED ANIMAL PROTEIN	1
12J - Other Skin Care Preps	100085618	HYDROLYZED ANIMAL PROTEIN	10
13A - Suntan Gels, Creams, and Liquids	100085618	HYDROLYZED ANIMAL PROTEIN	1
05A - Hair Conditioner	999003488	HYDROLYZED BARLEY PROTEIN	5
05B - Hair Spray (aerosol fixatives)	999003488	HYDROLYZED BARLEY PROTEIN	1
05F - Shampoos (non-coloring)	999003488	HYDROLYZED BARLEY PROTEIN	3
05G - Tonics, Dressings, and Other Hair Grooming Aids	999003488	HYDROLYZED BARLEY PROTEIN	6
05H - Wave Sets	999003488	HYDROLYZED BARLEY PROTEIN	2
05A - Hair Conditioner	999002399	HYDROLYZED BRAZIL NUT PROTEIN	4
05F - Shampoos (non-coloring)	999002399	HYDROLYZED BRAZIL NUT PROTEIN	1
05G - Tonics, Dressings, and Other Hair Grooming Aids	999002399	HYDROLYZED BRAZIL NUT PROTEIN	4
05I - Other Hair Preparations	999002399	HYDROLYZED BRAZIL NUT PROTEIN	1
03C - Eye Shadow	169590572	HYDROLYZED CONCHIOLIN PROTEIN	1
03D - Eye Lotion	169590572	HYDROLYZED CONCHIOLIN PROTEIN	5

03G - Other Eye Makeup Preparations	169590572	HYDROLYZED CONCHIOLIN PROTEIN	2
05A - Hair Conditioner	169590572	HYDROLYZED CONCHIOLIN PROTEIN	2
05F - Shampoos (non-coloring)	169590572	HYDROLYZED CONCHIOLIN PROTEIN	3
05G - Tonics, Dressings, and Other Hair Grooming Aids	169590572	HYDROLYZED CONCHIOLIN PROTEIN	2
05I - Other Hair Preparations	169590572	HYDROLYZED CONCHIOLIN PROTEIN	3
07B - Face Powders	169590572	HYDROLYZED CONCHIOLIN PROTEIN	3
07F - Makeup Bases	169590572	HYDROLYZED CONCHIOLIN PROTEIN	3
07I - Other Makeup Preparations	169590572	HYDROLYZED CONCHIOLIN PROTEIN	1
08A - Basecoats and Undercoats	169590572	HYDROLYZED CONCHIOLIN PROTEIN	1
08E - Nail Polish and Enamel	169590572	HYDROLYZED CONCHIOLIN PROTEIN	2
08G - Other Manicuring Preparations	169590572	HYDROLYZED CONCHIOLIN PROTEIN	1
12A - Cleansing	169590572	HYDROLYZED CONCHIOLIN PROTEIN	2
12C - Face and Neck (exc shave)	169590572	HYDROLYZED CONCHIOLIN PROTEIN	11
12D - Body and Hand (exc shave)	169590572	HYDROLYZED CONCHIOLIN PROTEIN	5
12F - Moisturizing	169590572	HYDROLYZED CONCHIOLIN PROTEIN	20
12H - Paste Masks (mud packs)	169590572	HYDROLYZED CONCHIOLIN PROTEIN	2
12I - Skin Fresheners	169590572	HYDROLYZED CONCHIOLIN PROTEIN	1
12J - Other Skin Care Preps	169590572	HYDROLYZED CONCHIOLIN PROTEIN	3
04E - Other Fragrance Preparation	999002431	HYDROLYZED COTTONSEED PROTEIN	11
10A - Bath Soaps and Detergents	999002431	HYDROLYZED COTTONSEED PROTEIN	1
10E - Other Personal Cleanliness Products	999002431	HYDROLYZED COTTONSEED PROTEIN	2
12A - Cleansing	999002431	HYDROLYZED COTTONSEED PROTEIN	2
12D - Body and Hand (exc shave)	999002431	HYDROLYZED COTTONSEED PROTEIN	3
12F - Moisturizing	999002431	HYDROLYZED COTTONSEED PROTEIN	13
01B - Baby Lotions, Oils, Powders, and Creams	91080181	HYDROLYZED ELASTIN	2
01C - Other Baby Products	91080181	HYDROLYZED ELASTIN	2
03D - Eye Lotion	91080181	HYDROLYZED ELASTIN	2

03G - Other Eye Makeup Preparations	91080181	HYDROLYZED ELASTIN	5
05A - Hair Conditioner	91080181	HYDROLYZED ELASTIN	7
05B - Hair Spray (aerosol fixatives)	91080181	HYDROLYZED ELASTIN	1
05F - Shampoos (non-coloring)	91080181	HYDROLYZED ELASTIN	4
05G - Tonics, Dressings, and Other Hair Grooming Aids	91080181	HYDROLYZED ELASTIN	9
07C - Foundations	91080181	HYDROLYZED ELASTIN	3
07E - Lipstick	91080181	HYDROLYZED ELASTIN	1
07F - Makeup Bases	91080181	HYDROLYZED ELASTIN	2
07I - Other Makeup Preparations	91080181	HYDROLYZED ELASTIN	1
10A - Bath Soaps and Detergents	91080181	HYDROLYZED ELASTIN	1
10E - Other Personal Cleanliness Products	91080181	HYDROLYZED ELASTIN	4
11A - Aftershave Lotion	91080181	HYDROLYZED ELASTIN	1
12A - Cleansing	91080181	HYDROLYZED ELASTIN	13
12C - Face and Neck (exc shave)	91080181	HYDROLYZED ELASTIN	41
12D - Body and Hand (exc shave)	91080181	HYDROLYZED ELASTIN	12
12F - Moisturizing	91080181	HYDROLYZED ELASTIN	29
12G - Night	91080181	HYDROLYZED ELASTIN	10
12H - Paste Masks (mud packs)	91080181	HYDROLYZED ELASTIN	4
12J - Other Skin Care Preps	91080181	HYDROLYZED ELASTIN	19
13B - Indoor Tanning Preparations	91080181	HYDROLYZED ELASTIN	3
03D - Eye Lotion	999001145	HYDROLYZED EXTENSIN	2
05A - Hair Conditioner	999001145	HYDROLYZED EXTENSIN	1
05C - Hair Straighteners	999001145	HYDROLYZED EXTENSIN	2
07C - Foundations	999001145	HYDROLYZED EXTENSIN	1
07I - Other Makeup Preparations	999001145	HYDROLYZED EXTENSIN	1
11G - Other Shaving Preparation Products	999001145	HYDROLYZED EXTENSIN	1
12C - Face and Neck (exc shave)	999001145	HYDROLYZED EXTENSIN	5
12D - Body and Hand (exc shave)	999001145	HYDROLYZED EXTENSIN	3
12F - Moisturizing	999001145	HYDROLYZED EXTENSIN	8
12G - Night	999001145	HYDROLYZED EXTENSIN	3
12H - Paste Masks (mud packs)	999001145	HYDROLYZED EXTENSIN	1
12I - Skin Fresheners	999001145	HYDROLYZED EXTENSIN	1
12J - Other Skin Care Preps	999001145	HYDROLYZED EXTENSIN	1
12C - Face and Neck (exc shave)	999003774	HYDROLYZED FIBROIN	1
03G - Other Eye Makeup Preparations	100085356	HYDROLYZED FIBRONECTIN	1
12C - Face and Neck (exc shave)	100085356	HYDROLYZED FIBRONECTIN	2
12D - Body and Hand (exc shave)	100085356	HYDROLYZED FIBRONECTIN	1
12F - Moisturizing	100085356	HYDROLYZED FIBRONECTIN	2
12G - Night	100085356	HYDROLYZED FIBRONECTIN	2
12H - Paste Masks (mud packs)	100085356	HYDROLYZED FIBRONECTIN	1
12J - Other Skin Care Preps	100085356	HYDROLYZED FIBRONECTIN	1
03D - Eye Lotion	91079913	HYDROLYZED FISH PROTEIN	1

03G - Other Eye Makeup Preparations	999002326	HYDROLYZED GADIDAE PROTEIN	1
12A - Cleansing	999002326	HYDROLYZED GADIDAE PROTEIN	1
12C - Face and Neck (exc shave)	999002326	HYDROLYZED GADIDAE PROTEIN	2
05A - Hair Conditioner	977153039	HYDROLYZED HAIR KERATIN	18
05F - Shampoos (non-coloring)	977153039	HYDROLYZED HAIR KERATIN	11
05G - Tonics, Dressings, and Other Hair Grooming Aids	977153039	HYDROLYZED HAIR KERATIN	1
05I - Other Hair Preparations	977153039	HYDROLYZED HAIR KERATIN	2
03D - Eye Lotion	999001271	HYDROLYZED HAZELNUT PROTEIN	2
03G - Other Eye Makeup Preparations	999001271	HYDROLYZED HAZELNUT PROTEIN	2
12C - Face and Neck (exc shave)	999001271	HYDROLYZED HAZELNUT PROTEIN	13
12D - Body and Hand (exc shave)	999001271	HYDROLYZED HAZELNUT PROTEIN	4
12F - Moisturizing	999001271	HYDROLYZED HAZELNUT PROTEIN	2
12H - Paste Masks (mud packs)	999001271	HYDROLYZED HAZELNUT PROTEIN	1
03F - Mascara	100684353	HYDROLYZED JOJOBA PROTEIN	1
05A - Hair Conditioner	100684353	HYDROLYZED JOJOBA PROTEIN	9
05E - Rinses (non-coloring)	100684353	HYDROLYZED JOJOBA PROTEIN	2
05F - Shampoos (non-coloring)	100684353	HYDROLYZED JOJOBA PROTEIN	4
05G - Tonics, Dressings, and Other Hair Grooming Aids	100684353	HYDROLYZED JOJOBA PROTEIN	10
10A - Bath Soaps and Detergents	100684353	HYDROLYZED JOJOBA PROTEIN	1
11E - Shaving Cream	100684353	HYDROLYZED JOJOBA PROTEIN	2
11G - Other Shaving Preparation Products	100684353	HYDROLYZED JOJOBA PROTEIN	1
12A - Cleansing	100684353	HYDROLYZED JOJOBA PROTEIN	1
12C - Face and Neck (exc shave)	100684353	HYDROLYZED JOJOBA PROTEIN	1
12F - Moisturizing	100684353	HYDROLYZED JOJOBA PROTEIN	3
02B - Bubble Baths	69430360	HYDROLYZED KERATIN	1
03A - Eyebrow Pencil	69430360	HYDROLYZED KERATIN	1
03D - Eye Lotion	69430360	HYDROLYZED KERATIN	1
03F - Mascara	69430360	HYDROLYZED KERATIN	23
03G - Other Eye Makeup Preparations	69430360	HYDROLYZED KERATIN	2
04E - Other Fragrance Preparation	69430360	HYDROLYZED KERATIN	1
05A - Hair Conditioner	69430360	HYDROLYZED KERATIN	88
05B - Hair Spray (aerosol fixatives)	69430360	HYDROLYZED KERATIN	15
05C - Hair Straighteners	69430360	HYDROLYZED KERATIN	6
05D - Permanent Waves	69430360	HYDROLYZED KERATIN	3
05E - Rinses (non-coloring)	69430360	HYDROLYZED KERATIN	6
05F - Shampoos (non-coloring)	69430360	HYDROLYZED KERATIN	60
05G - Tonics, Dressings, and Other Hair Grooming Aids	69430360	HYDROLYZED KERATIN	76
05H - Wave Sets	69430360	HYDROLYZED KERATIN	1
05I - Other Hair Preparations	69430360	HYDROLYZED KERATIN	26
06A - Hair Dyes and Colors (all types requiring caution statements and patch tests)	69430360	HYDROLYZED KERATIN	30

06D - Hair Shampoos (coloring)	69430360	HYDROLYZED KERATIN	1
06H - Other Hair Coloring Preparation	69430360	HYDROLYZED KERATIN	2
08B - Cuticle Softeners	69430360	HYDROLYZED KERATIN	2
08C - Nail Creams and Lotions	69430360	HYDROLYZED KERATIN	1
08E - Nail Polish and Enamel	69430360	HYDROLYZED KERATIN	1
08G - Other Manicuring Preparations	69430360	HYDROLYZED KERATIN	2
10A - Bath Soaps and Detergents	69430360	HYDROLYZED KERATIN	17
10B - Deodorants (underarm)	69430360	HYDROLYZED KERATIN	1
10E - Other Personal Cleanliness Products	69430360	HYDROLYZED KERATIN	2
12A - Cleansing	69430360	HYDROLYZED KERATIN	1
12D - Body and Hand (exc shave)	69430360	HYDROLYZED KERATIN	16
12F - Moisturizing	69430360	HYDROLYZED KERATIN	10
12J - Other Skin Care Preps	69430360	HYDROLYZED KERATIN	3
03D - Eye Lotion	977177119	HYDROLYZED LUPINE PROTEIN	9
03F - Mascara	977177119	HYDROLYZED LUPINE PROTEIN	2
05A - Hair Conditioner	977177119	HYDROLYZED LUPINE PROTEIN	3
05F - Shampoos (non-coloring)	977177119	HYDROLYZED LUPINE PROTEIN	1
05G - Tonics, Dressings, and Other Hair Grooming Aids	977177119	HYDROLYZED LUPINE PROTEIN	1
07C - Foundations	977177119	HYDROLYZED LUPINE PROTEIN	1
12C - Face and Neck (exc shave)	977177119	HYDROLYZED LUPINE PROTEIN	13
12D - Body and Hand (exc shave)	977177119	HYDROLYZED LUPINE PROTEIN	13
12F - Moisturizing	977177119	HYDROLYZED LUPINE PROTEIN	19
12G - Night	977177119	HYDROLYZED LUPINE PROTEIN	7
12H - Paste Masks (mud packs)	977177119	HYDROLYZED LUPINE PROTEIN	2
12I - Skin Fresheners	977177119	HYDROLYZED LUPINE PROTEIN	1
12J - Other Skin Care Preps	977177119	HYDROLYZED LUPINE PROTEIN	7
02A - Bath Oils, Tablets, and Salts	92797392	HYDROLYZED MILK PROTEIN	1
02B - Bubble Baths	92797392	HYDROLYZED MILK PROTEIN	2
02D - Other Bath Preparations	92797392	HYDROLYZED MILK PROTEIN	4
03D - Eye Lotion	92797392	HYDROLYZED MILK PROTEIN	1
03G - Other Eye Makeup Preparations	92797392	HYDROLYZED MILK PROTEIN	1
04A - Cologne and Toilet waters	92797392	HYDROLYZED MILK PROTEIN	1
05A - Hair Conditioner	92797392	HYDROLYZED MILK PROTEIN	9
05F - Shampoos (non-coloring)	92797392	HYDROLYZED MILK PROTEIN	11
05G - Tonics, Dressings, and Other Hair Grooming Aids	92797392	HYDROLYZED MILK PROTEIN	4
07E - Lipstick	92797392	HYDROLYZED MILK PROTEIN	3
10A - Bath Soaps and Detergents	92797392	HYDROLYZED MILK PROTEIN	9
10B - Deodorants (underarm)	92797392	HYDROLYZED MILK PROTEIN	2
10E - Other Personal Cleanliness Products	92797392	HYDROLYZED MILK PROTEIN	11
12A - Cleansing	92797392	HYDROLYZED MILK PROTEIN	3
12C - Face and Neck (exc shave)	92797392	HYDROLYZED MILK PROTEIN	12
12D - Body and Hand (exc shave)	92797392	HYDROLYZED MILK PROTEIN	28
12F - Moisturizing	92797392	HYDROLYZED MILK PROTEIN	9

12G - Night	92797392	HYDROLYZED MILK PROTEIN	6
12H - Paste Masks (mud packs)	92797392	HYDROLYZED MILK PROTEIN	2
12J - Other Skin Care Preps	92797392	HYDROLYZED MILK PROTEIN	2
13A - Suntan Gels, Creams, and Liquids	92797392	HYDROLYZED MILK PROTEIN	1
13B - Indoor Tanning Preparations	92797392	HYDROLYZED MILK PROTEIN	1
03G - Other Eye Makeup Preparations	977165722	HYDROLYZED OAT PROTEIN	1
05A - Hair Conditioner	977165722	HYDROLYZED OAT PROTEIN	14
05B - Hair Spray (aerosol fixatives)	977165722	HYDROLYZED OAT PROTEIN	2
05F - Shampoos (non-coloring)	977165722	HYDROLYZED OAT PROTEIN	10
05G - Tonics, Dressings, and Other Hair Grooming Aids	977165722	HYDROLYZED OAT PROTEIN	12
05I - Other Hair Preparations	977165722	HYDROLYZED OAT PROTEIN	4
07I - Other Makeup Preparations	977165722	HYDROLYZED OAT PROTEIN	1
08E - Nail Polish and Enamel	977165722	HYDROLYZED OAT PROTEIN	2
08G - Other Manicuring Preparations	977165722	HYDROLYZED OAT PROTEIN	2
10A - Bath Soaps and Detergents	977165722	HYDROLYZED OAT PROTEIN	5
10E - Other Personal Cleanliness Products	977165722	HYDROLYZED OAT PROTEIN	15
11A - Aftershave Lotion	977165722	HYDROLYZED OAT PROTEIN	1
11G - Other Shaving Preparation Products	977165722	HYDROLYZED OAT PROTEIN	1
12A - Cleansing	977165722	HYDROLYZED OAT PROTEIN	2
12C - Face and Neck (exc shave)	977165722	HYDROLYZED OAT PROTEIN	1
12F - Moisturizing	977165722	HYDROLYZED OAT PROTEIN	4
12I - Skin Fresheners	977165722	HYDROLYZED OAT PROTEIN	1
12C - Face and Neck (exc shave)	977177108	HYDROLYZED PEA PROTEIN	2
12C - Face and Neck (exc shave)	999001864	HYDROLYZED POTATO PROTEIN	3
12F - Moisturizing	999001864	HYDROLYZED POTATO PROTEIN	6
12G - Night	999001864	HYDROLYZED POTATO PROTEIN	3
12J - Other Skin Care Preps	999001864	HYDROLYZED POTATO PROTEIN	1
13A - Suntan Gels, Creams, and Liquids	999001864	HYDROLYZED POTATO PROTEIN	2
03G - Other Eye Makeup Preparations	999003277	HYDROLYZED SERICIN	1
05I - Other Hair Preparations	999003277	HYDROLYZED SERICIN	1
01A - Baby Shampoos	96690414	HYDROLYZED SILK	2
01B - Baby Lotions, Oils, Powders, and Creams	96690414	HYDROLYZED SILK	2
01C - Other Baby Products	96690414	HYDROLYZED SILK	2
02B - Bubble Baths	96690414	HYDROLYZED SILK	1
02D - Other Bath Preparations	96690414	HYDROLYZED SILK	4
03A - Eyebrow Pencil	96690414	HYDROLYZED SILK	1
03B - Eyeliner	96690414	HYDROLYZED SILK	2
03C - Eye Shadow	96690414	HYDROLYZED SILK	1
03D - Eye Lotion	96690414	HYDROLYZED SILK	10
03E - Eye Makeup Remover	96690414	HYDROLYZED SILK	1

03F - Mascara	96690414	HYDROLYZED SILK	14
03G - Other Eye Makeup Preparations	96690414	HYDROLYZED SILK	4
04E - Other Fragrance Preparation	96690414	HYDROLYZED SILK	1
05A - Hair Conditioner	96690414	HYDROLYZED SILK	74
05B - Hair Spray (aerosol fixatives)	96690414	HYDROLYZED SILK	8
05C - Hair Straighteners	96690414	HYDROLYZED SILK	13
05D - Permanent Waves	96690414	HYDROLYZED SILK	3
05E - Rinses (non-coloring)	96690414	HYDROLYZED SILK	1
05F - Shampoos (non-coloring)	96690414	HYDROLYZED SILK	80
05G - Tonics, Dressings, and Other Hair Grooming Aids	96690414	HYDROLYZED SILK	39
05H - Wave Sets	96690414	HYDROLYZED SILK	1
05I - Other Hair Preparations	96690414	HYDROLYZED SILK	30
06A - Hair Dyes and Colors (all types requiring caution statements and patch tests)	96690414	HYDROLYZED SILK	13
06H - Other Hair Coloring Preparation	96690414	HYDROLYZED SILK	1
07B - Face Powders	96690414	HYDROLYZED SILK	6
07C - Foundations	96690414	HYDROLYZED SILK	2
07D - Leg and Body Paints	96690414	HYDROLYZED SILK	3
07E - Lipstick	96690414	HYDROLYZED SILK	6
07F - Makeup Bases	96690414	HYDROLYZED SILK	1
07G - Rouges	96690414	HYDROLYZED SILK	5
07I - Other Makeup Preparations	96690414	HYDROLYZED SILK	4
08E - Nail Polish and Enamel	96690414	HYDROLYZED SILK	1
10A - Bath Soaps and Detergents	96690414	HYDROLYZED SILK	54
10B - Deodorants (underarm)	96690414	HYDROLYZED SILK	1
10E - Other Personal Cleanliness Products	96690414	HYDROLYZED SILK	17
11E - Shaving Cream	96690414	HYDROLYZED SILK	1
11G - Other Shaving Preparation Products	96690414	HYDROLYZED SILK	8
12A - Cleansing	96690414	HYDROLYZED SILK	15
12B - Depilatories	96690414	HYDROLYZED SILK	2
12C - Face and Neck (exc shave)	96690414	HYDROLYZED SILK	21
12D - Body and Hand (exc shave)	96690414	HYDROLYZED SILK	42
12F - Moisturizing	96690414	HYDROLYZED SILK	39
12G - Night	96690414	HYDROLYZED SILK	4
12H - Paste Masks (mud packs)	96690414	HYDROLYZED SILK	3
12I - Skin Fresheners	96690414	HYDROLYZED SILK	1
12J - Other Skin Care Preps	96690414	HYDROLYZED SILK	10
13B - Indoor Tanning Preparations	96690414	HYDROLYZED SILK	22
13C - Other Suntan Preparations	96690414	HYDROLYZED SILK	1
03A - Eyebrow Pencil	68607885	HYDROLYZED SOY PROTEIN	1
03D - Eye Lotion	68607885	HYDROLYZED SOY PROTEIN	12
03E - Eye Makeup Remover	68607885	HYDROLYZED SOY PROTEIN	1
03F - Mascara	68607885	HYDROLYZED SOY PROTEIN	19
03G - Other Eye Makeup Preparations	68607885	HYDROLYZED SOY PROTEIN	13

05A - Hair Conditioner	68607885	HYDROLYZED SOY PROTEIN	85
05B - Hair Spray (aerosol fixatives)	68607885	HYDROLYZED SOY PROTEIN	9
05C - Hair Straighteners	68607885	HYDROLYZED SOY PROTEIN	4
05D - Permanent Waves	68607885	HYDROLYZED SOY PROTEIN	2
05E - Rinses (non-coloring)	68607885	HYDROLYZED SOY PROTEIN	1
05F - Shampoos (non-coloring)	68607885	HYDROLYZED SOY PROTEIN	90
05G - Tonics, Dressings, and Other Hair Grooming Aids	68607885	HYDROLYZED SOY PROTEIN	66
05H - Wave Sets	68607885	HYDROLYZED SOY PROTEIN	6
05I - Other Hair Preparations	68607885	HYDROLYZED SOY PROTEIN	51
06A - Hair Dyes and Colors (all types requiring caution statements and patch tests)	68607885	HYDROLYZED SOY PROTEIN	27
06D - Hair Shampoos (coloring)	68607885	HYDROLYZED SOY PROTEIN	1
06H - Other Hair Coloring Preparation	68607885	HYDROLYZED SOY PROTEIN	4
07B - Face Powders	68607885	HYDROLYZED SOY PROTEIN	2
07C - Foundations	68607885	HYDROLYZED SOY PROTEIN	16
07E - Lipstick	68607885	HYDROLYZED SOY PROTEIN	1
07F - Makeup Bases	68607885	HYDROLYZED SOY PROTEIN	1
07I - Other Makeup Preparations	68607885	HYDROLYZED SOY PROTEIN	6
08A - Basecoats and Undercoats	68607885	HYDROLYZED SOY PROTEIN	5
08B - Cuticle Softeners	68607885	HYDROLYZED SOY PROTEIN	1
08C - Nail Creams and Lotions	68607885	HYDROLYZED SOY PROTEIN	1
08E - Nail Polish and Enamel	68607885	HYDROLYZED SOY PROTEIN	17
08G - Other Manicuring Preparations	68607885	HYDROLYZED SOY PROTEIN	8
10A - Bath Soaps and Detergents	68607885	HYDROLYZED SOY PROTEIN	3
10B - Deodorants (underarm)	68607885	HYDROLYZED SOY PROTEIN	1
10E - Other Personal Cleanliness Products	68607885	HYDROLYZED SOY PROTEIN	5
11E - Shaving Cream	68607885	HYDROLYZED SOY PROTEIN	3
11F - Shaving Soap	68607885	HYDROLYZED SOY PROTEIN	3
11G - Other Shaving Preparation Products	68607885	HYDROLYZED SOY PROTEIN	3
12A - Cleansing	68607885	HYDROLYZED SOY PROTEIN	11
12B - Depilatories	68607885	HYDROLYZED SOY PROTEIN	10
12C - Face and Neck (exc shave)	68607885	HYDROLYZED SOY PROTEIN	68
12D - Body and Hand (exc shave)	68607885	HYDROLYZED SOY PROTEIN	23
12F - Moisturizing	68607885	HYDROLYZED SOY PROTEIN	55
12G - Night	68607885	HYDROLYZED SOY PROTEIN	14
12H - Paste Masks (mud packs)	68607885	HYDROLYZED SOY PROTEIN	3
12I - Skin Fresheners	68607885	HYDROLYZED SOY PROTEIN	5
12J - Other Skin Care Preps	68607885	HYDROLYZED SOY PROTEIN	21
13B - Indoor Tanning Preparations	68607885	HYDROLYZED SOY PROTEIN	1
13C - Other Suntan Preparations	68607885	HYDROLYZED SOY PROTEIN	2
05A - Hair Conditioner	999002352	HYDROLYZED SOYMILK PROTEIN	2
05F - Shampoos (non-coloring)	999002352	HYDROLYZED SOYMILK PROTEIN	1
05G - Tonics, Dressings, and Other Hair Grooming Aids	999002352	HYDROLYZED SOYMILK PROTEIN	1
05I - Other Hair Preparations	999002352	HYDROLYZED SOYMILK PROTEIN	1

12A - Cleansing	999002352	HYDROLYZED SOYMILK PROTEIN	1
12J - Other Skin Care Preps	999002352	HYDROLYZED SOYMILK PROTEIN	1
03E - Eye Makeup Remover	977182641	HYDROLYZED SWEET ALMOND PROTEIN	1
03F - Mascara	977182641	HYDROLYZED SWEET ALMOND PROTEIN	3
03G - Other Eye Makeup Preparations	977182641	HYDROLYZED SWEET ALMOND PROTEIN	2
05A - Hair Conditioner	977182641	HYDROLYZED SWEET ALMOND PROTEIN	19
05E - Rinses (non-coloring)	977182641	HYDROLYZED SWEET ALMOND PROTEIN	1
05F - Shampoos (non-coloring)	977182641	HYDROLYZED SWEET ALMOND PROTEIN	7
05G - Tonics, Dressings, and Other Hair Grooming Aids	977182641	HYDROLYZED SWEET ALMOND PROTEIN	5
05I - Other Hair Preparations	977182641	HYDROLYZED SWEET ALMOND PROTEIN	3
06H - Other Hair Coloring Preparation	977182641	HYDROLYZED SWEET ALMOND PROTEIN	1
10A - Bath Soaps and Detergents	977182641	HYDROLYZED SWEET ALMOND PROTEIN	1
10E - Other Personal Cleanliness Products	977182641	HYDROLYZED SWEET ALMOND PROTEIN	1
12A - Cleansing	977182641	HYDROLYZED SWEET ALMOND PROTEIN	3
12C - Face and Neck (exc shave)	977182641	HYDROLYZED SWEET ALMOND PROTEIN	16
12D - Body and Hand (exc shave)	977182641	HYDROLYZED SWEET ALMOND PROTEIN	2
12F - Moisturizing	977182641	HYDROLYZED SWEET ALMOND PROTEIN	6
12G - Night	977182641	HYDROLYZED SWEET ALMOND PROTEIN	2
12H - Paste Masks (mud packs)	977182641	HYDROLYZED SWEET ALMOND PROTEIN	1
12J - Other Skin Care Preps	977182641	HYDROLYZED SWEET ALMOND PROTEIN	2
13C - Other Suntan Preparations	977182641	HYDROLYZED SWEET ALMOND PROTEIN	1
01B - Baby Lotions, Oils, Powders, and Creams	100209458	HYDROLYZED VEGETABLE PROTEIN	1
01C - Other Baby Products	100209458	HYDROLYZED VEGETABLE PROTEIN	1
03G - Other Eye Makeup Preparations	100209458	HYDROLYZED VEGETABLE PROTEIN	4
04E - Other Fragrance Preparation	100209458	HYDROLYZED VEGETABLE PROTEIN	1
05A - Hair Conditioner	100209458	HYDROLYZED VEGETABLE PROTEIN	18
05B - Hair Spray (aerosol fixatives)	100209458	HYDROLYZED VEGETABLE PROTEIN	2
05F - Shampoos (non-coloring)	100209458	HYDROLYZED VEGETABLE PROTEIN	15
05G - Tonics, Dressings, and Other Hair Grooming Aids	100209458	HYDROLYZED VEGETABLE PROTEIN	9
05I - Other Hair Preparations	100209458	HYDROLYZED VEGETABLE PROTEIN	5

06A - Hair Dyes and Colors (all types requiring caution statements and patch tests)	100209458	HYDROLYZED VEGETABLE PROTEIN	48
06B - Hair Tints	100209458	HYDROLYZED VEGETABLE PROTEIN	1
06D - Hair Shampoos (coloring)	100209458	HYDROLYZED VEGETABLE PROTEIN	1
06F - Hair Lighteners with Color	100209458	HYDROLYZED VEGETABLE PROTEIN	2
06H - Other Hair Coloring Preparation	100209458	HYDROLYZED VEGETABLE PROTEIN	2
07B - Face Powders	100209458	HYDROLYZED VEGETABLE PROTEIN	1
07C - Foundations	100209458	HYDROLYZED VEGETABLE PROTEIN	3
07H - Makeup Fixatives	100209458	HYDROLYZED VEGETABLE PROTEIN	1
10E - Other Personal Cleanliness Products	100209458	HYDROLYZED VEGETABLE PROTEIN	2
12A - Cleansing	100209458	HYDROLYZED VEGETABLE PROTEIN	6
12C - Face and Neck (exc shave)	100209458	HYDROLYZED VEGETABLE PROTEIN	4
12D - Body and Hand (exc shave)	100209458	HYDROLYZED VEGETABLE PROTEIN	2
12F - Moisturizing	100209458	HYDROLYZED VEGETABLE PROTEIN	14
12G - Night	100209458	HYDROLYZED VEGETABLE PROTEIN	5
12H - Paste Masks (mud packs)	100209458	HYDROLYZED VEGETABLE PROTEIN	3
13B - Indoor Tanning Preparations	100209458	HYDROLYZED VEGETABLE PROTEIN	1
13C - Other Suntan Preparations	100209458	HYDROLYZED VEGETABLE PROTEIN	1
01A - Baby Shampoos	977016526	HYDROLYZED WHEAT GLUTEN	2
01B - Baby Lotions, Oils, Powders, and Creams	977016526	HYDROLYZED WHEAT GLUTEN	1
02B - Bubble Baths	977016526	HYDROLYZED WHEAT GLUTEN	2
02D - Other Bath Preparations	977016526	HYDROLYZED WHEAT GLUTEN	1
03G - Other Eye Makeup Preparations	977016526	HYDROLYZED WHEAT GLUTEN	1
05A - Hair Conditioner	977016526	HYDROLYZED WHEAT GLUTEN	6
05F - Shampoos (non-coloring)	977016526	HYDROLYZED WHEAT GLUTEN	9
05I - Other Hair Preparations	977016526	HYDROLYZED WHEAT GLUTEN	2
06D - Hair Shampoos (coloring)	977016526	HYDROLYZED WHEAT GLUTEN	1
10A - Bath Soaps and Detergents	977016526	HYDROLYZED WHEAT GLUTEN	8
10E - Other Personal Cleanliness Products	977016526	HYDROLYZED WHEAT GLUTEN	7
12A - Cleansing	977016526	HYDROLYZED WHEAT GLUTEN	1
12C - Face and Neck (exc shave)	977016526	HYDROLYZED WHEAT GLUTEN	7
12J - Other Skin Care Preps	977016526	HYDROLYZED WHEAT GLUTEN	1
01C - Other Baby Products	977117728	HYDROLYZED WHEAT PROTEIN	2
02B - Bubble Baths	977117728	HYDROLYZED WHEAT PROTEIN	5
02D - Other Bath Preparations	977117728	HYDROLYZED WHEAT PROTEIN	2
03A - Eyebrow Pencil	977117728	HYDROLYZED WHEAT PROTEIN	1
03B - Eyeliner	977117728	HYDROLYZED WHEAT PROTEIN	2
03C - Eye Shadow	977117728	HYDROLYZED WHEAT PROTEIN	1
03D - Eye Lotion	977117728	HYDROLYZED WHEAT PROTEIN	11
03E - Eye Makeup Remover	977117728	HYDROLYZED WHEAT PROTEIN	2
03F - Mascara	977117728	HYDROLYZED WHEAT PROTEIN	25
03G - Other Eye Makeup Preparations	977117728	HYDROLYZED WHEAT PROTEIN	24
04C - Powders (dusting and talcum, excluding aftershave talc)	977117728	HYDROLYZED WHEAT PROTEIN	1

05A - Hair Conditioner	977117728	HYDROLYZED WHEAT PROTEIN	160
05B - Hair Spray (aerosol fixatives)	977117728	HYDROLYZED WHEAT PROTEIN	17
05C - Hair Straighteners	977117728	HYDROLYZED WHEAT PROTEIN	4
05D - Permanent Waves	977117728	HYDROLYZED WHEAT PROTEIN	5
05E - Rinses (non-coloring)	977117728	HYDROLYZED WHEAT PROTEIN	2
05F - Shampoos (non-coloring)	977117728	HYDROLYZED WHEAT PROTEIN	150
05G - Tonics, Dressings, and Other Hair Grooming Aids	977117728	HYDROLYZED WHEAT PROTEIN	122
05H - Wave Sets	977117728	HYDROLYZED WHEAT PROTEIN	7
05I - Other Hair Preparations	977117728	HYDROLYZED WHEAT PROTEIN	66
06A - Hair Dyes and Colors (all types requiring caution statements and patch tests)	977117728	HYDROLYZED WHEAT PROTEIN	63
06B - Hair Tints	977117728	HYDROLYZED WHEAT PROTEIN	1
06C - Hair Rinses (coloring)	977117728	HYDROLYZED WHEAT PROTEIN	10
06D - Hair Shampoos (coloring)	977117728	HYDROLYZED WHEAT PROTEIN	5
06G - Hair Bleaches	977117728	HYDROLYZED WHEAT PROTEIN	1
07A - Blushers (all types)	977117728	HYDROLYZED WHEAT PROTEIN	1
07B - Face Powders	977117728	HYDROLYZED WHEAT PROTEIN	5
07C - Foundations	977117728	HYDROLYZED WHEAT PROTEIN	15
07E - Lipstick	977117728	HYDROLYZED WHEAT PROTEIN	12
07I - Other Makeup Preparations	977117728	HYDROLYZED WHEAT PROTEIN	13
08A - Basecoats and Undercoats	977117728	HYDROLYZED WHEAT PROTEIN	5
08B - Cuticle Softeners	977117728	HYDROLYZED WHEAT PROTEIN	2
08C - Nail Creams and Lotions	977117728	HYDROLYZED WHEAT PROTEIN	1
08E - Nail Polish and Enamel	977117728	HYDROLYZED WHEAT PROTEIN	7
08F - Nail Polish and Enamel Removers	977117728	HYDROLYZED WHEAT PROTEIN	3
08G - Other Manicuring Preparations	977117728	HYDROLYZED WHEAT PROTEIN	10
10A - Bath Soaps and Detergents	977117728	HYDROLYZED WHEAT PROTEIN	59
10E - Other Personal Cleanliness Products	977117728	HYDROLYZED WHEAT PROTEIN	18
11A - Aftershave Lotion	977117728	HYDROLYZED WHEAT PROTEIN	2
11G - Other Shaving Preparation Products	977117728	HYDROLYZED WHEAT PROTEIN	2
12A - Cleansing	977117728	HYDROLYZED WHEAT PROTEIN	20
12C - Face and Neck (exc shave)	977117728	HYDROLYZED WHEAT PROTEIN	64
12D - Body and Hand (exc shave)	977117728	HYDROLYZED WHEAT PROTEIN	30
12F - Moisturizing	977117728	HYDROLYZED WHEAT PROTEIN	36
12G - Night	977117728	HYDROLYZED WHEAT PROTEIN	16
12H - Paste Masks (mud packs)	977117728	HYDROLYZED WHEAT PROTEIN	12
12I - Skin Fresheners	977117728	HYDROLYZED WHEAT PROTEIN	2
12J - Other Skin Care Preps	977117728	HYDROLYZED WHEAT PROTEIN	29
13B - Indoor Tanning Preparations	977117728	HYDROLYZED WHEAT PROTEIN	1
13C - Other Suntan Preparations	977117728	HYDROLYZED WHEAT PROTEIN	1
03D - Eye Lotion	100684364	HYDROLYZED YEAST PROTEIN	4
03G - Other Eye Makeup Preparations	100684364	HYDROLYZED YEAST PROTEIN	1
05A - Hair Conditioner	100684364	HYDROLYZED YEAST PROTEIN	2
05B - Hair Spray (aerosol fixatives)	100684364	HYDROLYZED YEAST PROTEIN	1

05D - Permanent Waves	100684364	HYDROLYZED YEAST PROTEIN	4
05F - Shampoos (non-coloring)	100684364	HYDROLYZED YEAST PROTEIN	4
05G - Tonics, Dressings, and Other Hair Grooming Aids	100684364	HYDROLYZED YEAST PROTEIN	1
05H - Wave Sets	100684364	HYDROLYZED YEAST PROTEIN	1
05I - Other Hair Preparations	100684364	HYDROLYZED YEAST PROTEIN	4
07C - Foundations	100684364	HYDROLYZED YEAST PROTEIN	1
07I - Other Makeup Preparations	100684364	HYDROLYZED YEAST PROTEIN	1
11G - Other Shaving Preparation Products	100684364	HYDROLYZED YEAST PROTEIN	1
12A - Cleansing	100684364	HYDROLYZED YEAST PROTEIN	4
12C - Face and Neck (exc shave)	100684364	HYDROLYZED YEAST PROTEIN	5
12D - Body and Hand (exc shave)	100684364	HYDROLYZED YEAST PROTEIN	1
12F - Moisturizing	100684364	HYDROLYZED YEAST PROTEIN	9
12G - Night	100684364	HYDROLYZED YEAST PROTEIN	3
12H - Paste Masks (mud packs)	100684364	HYDROLYZED YEAST PROTEIN	2
12I - Skin Fresheners	100684364	HYDROLYZED YEAST PROTEIN	1
12J - Other Skin Care Preps	100684364	HYDROLYZED YEAST PROTEIN	7
10A - Bath Soaps and Detergents	999002282	HYDROLYZED YOGURT PROTEIN	8

7

Final Report on the Safety Assessment of Hydrolyzed Collagen

Hydrolyzed Collagen is a hydrolysate derived from animal byproducts. It is used in cosmetic products as a conditioner or moisturizer at concentrations less than 5 percent.

Hydrolyzed Collagen was practically nontoxic when administered orally or dermally in acute animal toxicity studies. This ingredient was minimally irritating to rabbit eyes when tested full-strength. Primary skin irritation tests in rabbits indicated that Hydrolyzed Collagen was nonirritating or minimally irritating when tested full-strength. Subchronic dermal studies on 2 cosmetic formulations containing 2 percent Hydrolyzed Collagen were negative for systemic toxicity. Hydrolyzed Collagen was nonsensitizing in guinea pigs.

In clinical studies, Hydrolyzed Collagen produced no skin irritation, sensitization, or indication of phototoxicity.

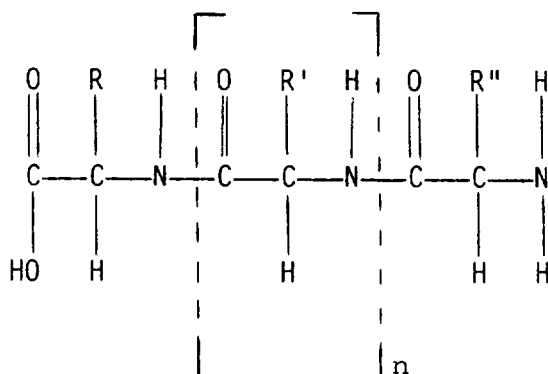
On the basis of the available animal and clinical data, it is concluded that Hydrolyzed Collagen is safe as a cosmetic ingredient in the present practices of use and concentration.

INTRODUCTION

This report presents the available information published between 1940 and the present and the unpublished cosmetic industry data on Hydrolyzed Collagen (formerly Hydrolyzed Animal Protein).

CHEMICAL AND PHYSICAL PROPERTIES

Hydrolyzed Collagen is defined as a collagen hydrolysate derived by acid, enzyme, or other method of hydrolysis.⁽¹⁾ It is a polypeptide of molecular weight 1000 to 10,000 with the following generalized structure:⁽²⁾



The typical amino acid composition is given in Table 1 along with the corresponding structures of R, R', and R''.

Hydrolyzed Collagen is an off-white to white hygroscopic powder. It is also available as a viscous, amber aqueous solution.⁽²⁾ The physicochemical properties of Hydrolyzed Collagen and its solution are given in Table 2.

Hydrolyzed Collagen can be prepared by any 1 of 3 methods: alkaline hydrolysis of bovine skin products followed by enzymatic hydrolysis to the desired molecular weight, enzymatic hydrolysis of fresh animal byproducts or bovine-derived leather, or acid or alkaline hydrolysis of chrome leather fold splinters with inorganic acids or lyes to a defined molecular weight. The hydrolysate produced by the latter method is purified in an aqueous solution and then by precipitation and filtration to effectively remove the heavy ions.⁽²⁾ Acid hydrolysis tends to split the polypeptide bond between proline (or hydroxyproline) and other amino acids, with very little specificity for which amino acid donates its amino group to the peptide bond.⁽³⁾

The spectrum of amino acids resulting from the hydrolysis of collagen differs substantially from that of other proteins by its high content of glycine and proline and low content of histidine, tryptophan, and cystine (Table 1). Collagen also contains 2 amino acids, hydroxyproline and hydroxylysine, not found in other proteins.⁽⁴⁾

Hydrolyzed Collagen is analyzed primarily by column chromatography. The literature cites numerous chromatographic methods.⁽⁵⁻⁹⁾ Its solution can be positively identified by comparison to a standard infrared spectrum.⁽¹⁰⁾ The 2 amino acids found only in collagen, hydroxyproline and hydroxylysine, allow for differentiation between collagen hydrolysates and other protein hydrolysates.⁽⁴⁾

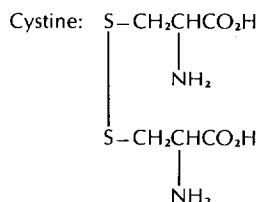
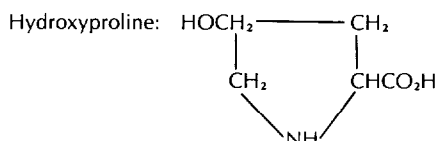
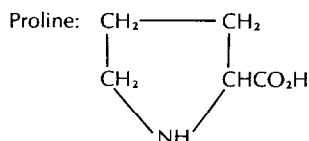
Hydrolyzed Collagen is stable under the conditions of normal cosmetic use. However, the addition of perfumes high in aldehyde content may cause color reactions and odor changes due to the reaction of active carbonyl groups with the amino group of the hydrolysate.^(2,11) Burnett⁽¹¹⁾ has found that cosmetics containing proteins are better preserved at successively lower pH values, whereas Frech et al.⁽¹²⁾ found that sodium acetate is an effective preservative in protein hydrolysate solutions. Hydrolyzed Collagen has shown a 15 to 45 percent reduction in viscosity after storage for 1 month at room temperature.⁽¹³⁾

TABLE 1. Typical Amino Acid Composition of Hydrolyzed Collagen⁽²⁾

Amino Acid	R, R', or R''	Typical Composition* (%)
Glycine	– H	20.0–30.5
Alanine	– CH ₃	8.0–11.0
Serine	– CH ₂ OH	2.9–4.1
Threonine	– CHOHCH ₃	1.8–2.6
Proline	CYCLIC [†]	13.7–18.0
Hydroxyproline	CYCLIC [†]	12.1–14.5
Valine	– CHCH ₃ CH ₃	2.1–3.4
Isoleucine	– CHCH ₃ CH ₂ CH ₃	1.3–1.8
Leucine	– CH ₂ CHCH ₃ CH ₃	2.8–3.5
Phenylalanine	– CH ₂ C ₆ H ₅	1.1–2.6
Tyrosine	– CH ₂ C ₆ H ₄ OH	0.2–1.0
Cystine/cysteine	– [†] /– CH ₂ – SH	0.0–0.9
Methionine	– CH ₂ CH ₂ SCH ₃	0.7–0.9
Aspartic acid	– CH ₂ CO ₂ H	5.7–9.0
Glutamic acid	– CH ₂ CH ₂ CO ₂ H	10.0–11.7
Arginine	– CH ₂ CH ₂ CH ₂ C(NH) ₂ NH ₂	7.8–9.0
Histidine	NHCHNCHCCH ₂ –	0.7–1.0
Lysine	– CH ₂ CH ₂ CH ₂ CH ₂ NH ₂	3.9–5.2
Hydroxylysine	– (CH ₂) ₂ CHOHCH ₂ NH ₂	0.7–1.2

*Prepared by alkaline hydrolysis of bovine skin products to form a food-grade gelatin, followed by enzyme hydrolysis to the desired molecular weight.

[†]These do not conform to the generalized formula.



USE

Cosmetic Use

Hydrolyzed Collagen is used in cosmetics, primarily hair and skin care products, because of its conditioning and moisturizing properties. It is generally used

TABLE 2. Physicochemical Properties of Hydrolyzed Collagen⁽²⁾

Property	Value	
	Powder	Solution
Molecular weight	1000 to 10,000	—
Solids content (105°C/16 hours)	—	35% min.
Moisture content (vacuum oven 90°C/6 hours)	8.0% max.	—
pH	—	4.0–6.5 (10% aqueous solution)
Nitrogen	12.0% min.	8.0% min.
Ash content	12.0% max.	5.0% max.
Iron	—	3 ppm max.
Heavy metals	—	25 ppm max.

at concentrations <5 percent in the following product categories: baby shampoos, bath, eye makeup, hair, hair coloring, makeup, manicuring, personal cleanliness, shaving, skin care, and tanning preparations.^(2,13–16)

Table 3 presents the FDA product formulation data for Hydrolyzed Collagen.⁽¹⁵⁾ The cosmetic product formulation computer printout that is made available by the Food and Drug Administration (FDA) is compiled through voluntary filing of such data in accordance with Title 21 part 720.4 of the Code of Federal Regulations. Ingredients are listed in prescribed concentration ranges under specific product type categories. Since certain cosmetic ingredients are supplied by the manufacturer at <100 percent concentration, the value reported by the cosmetic formulator may not necessarily reflect the actual concentration found in the finished product; the actual concentration in such a case would be a fraction of that reported to the FDA. The fact that data are only submitted within the framework of preset concentration ranges also provides the opportunity for overestimation of the actual concentration of an ingredient in a particular product. An entry at the lowest end of a concentration range is considered the same as one entered at the highest end of that range, thus introducing the possibility of a 2- to 10-fold error in the assumed ingredient concentration.

In 1981, approximately 95 percent of the 923 formulations containing Hydrolyzed Collagen incorporated the hydrolysate at concentrations of <5 percent: 23 percent of these at concentrations ≤0.1 percent, 50 percent at concentrations >0.1 to 1 percent, and 22 percent at concentrations >1 to 5 percent. Hair preparations accounted for 66 percent of the total product listings of Hydrolyzed Collagen, with the second highest listing (16 percent) found in skin care products.⁽¹⁵⁾

The formulation data presented in Table 3 indicate that cosmetic products containing Hydrolyzed Collagen may contact all external body surfaces and hair, as well as the eyes. These products may be used daily or occasionally over a period of up to several years. The frequency and length of application could result in continuous exposure.

Noncosmetic Use

Hydrolyzed Collagen is used in soaps to impart a soft texture to skin.⁽¹¹⁾ It is also used in a treatment for duodenal and gastric ulcers and gastritis.⁽¹⁷⁾ A protective film-forming composition containing Hydrolyzed Collagen is also applied to cow teats.⁽¹⁸⁾

GENERAL BIOLOGY

Substantivity

The literature contains many studies documenting the substantivity of Hydrolyzed Collagen to human hair and skin. Hydrolyzed Collagen generally is adsorbed on hair; the amount of protein adsorbed is measured by hydroxyproline analysis.^(13,19) Kim et al. found that adsorbed Hydrolyzed Collagen increased the tensile strength and elongation of hair. Increasing concentrations of Hydrolyzed Collagen increased adsorption.^(19,20) Hydrolyzed Collagens of average molecular weight 1000 and 2000 have greater substantivity to hair and skin, respectively, than those of average molecular weight 10,000.^(13,21) Brooks⁽²²⁾ stated that Hydrolyzed Collagen substantivity is due to the distribution of terminal amino groups along (primarily at the lysine and arginine residues) and at the ends of the polypeptide.

Cooperman and Johnsen⁽²³⁾ studied the penetration of Hydrolyzed Collagen into both unbleached and bleached hair strands. In the hair strands treated with the lower molecular weight Hydrolyzed Collagens, the cuticle contained the highest percentage of protein. However, the cortex, by virtue of its greater bulk, contained the greater amount of protein. Hair strands treated with the higher molecular weight Hydrolyzed Collagens had equal quantities of protein in the cuticle and cortex. Protein penetration increased with increasing damage to hair.

As a hygroscopic compound, Hydrolyzed Collagen helps bind water to the hair and skin surfaces.^(11,13,24,25) Brooks⁽²²⁾ found that Hydrolyzed Collagen binds water better at higher relative humidities and at pH 5 rather than pH 7 or 9. Hydrolyzed Collagen potentiates epidermal metabolism by providing a suitable, moist environment on the surface of the epidermis for healthy skin and hair.⁽¹¹⁾

The amphoteric nature of Hydrolyzed Collagen makes it an acceptable buffering agent for alkali in permanent waving products. It is incorporated in waving formulations to avoid or minimize damage to hair. A protective application of Hydrolyzed Collagen is sometimes made prior to waving. Hydrolyzed Collagen is also used in hair dyes to insure uniformity in dyeing.⁽¹¹⁾

General Effects

Various enzymes will hydrolyze collagen: trypsin,⁽²⁶⁾ intracellular proteolytic enzymes of *Oidiodendron kalari*,⁽²⁷⁾ *Streptomyces griseus* protease,^(28,29) collagenases of genera *Bacteroides*, *Clostridium*, and *Peptostreptococcus*,^(30,31) rat hepatic lysosomal extracts,⁽³²⁾ collagenases from rabbit synovial fibroblasts,⁽³³⁾ and cathepsin B and collagenolytic cathepsin from human placenta.⁽³⁴⁾

TABLE 3. Product Formulation Data⁽¹⁵⁾

Product Category	Total No. of Formulations in Category	Total No. Containing Ingredient	No. of Product Formulations Within Each Concentration Range (%)						
			>50	>25-50	>10-25	>5-10	>1-5	>0.1-1	≤0.1
Hydrolyzed Collagen									
Baby shampoos	35	1	—	—	—	—	—	—	1
Bath oils, tablets, and salts	237	2	—	—	—	—	2	—	—
Bubble baths	475	2	—	—	—	—	—	2	—
Other bath preparations	132	2	—	—	—	—	—	2	—
Eyeliner	396	1	—	—	—	—	—	—	1
Eye shadow	2582	6	—	—	—	—	—	—	1
Mascara	397	28	—	—	—	—	—	5	1
Other eye makeup preparations	230	5	—	—	—	—	1	15	13
Hair conditioners	478	174	—	—	—	—	—	—	2
Hair sprays (aerosol fixatives)	265	7	2	1	2	13	60	79	17
Hair straighteners	64	7	—	—	—	—	—	1	6
Permanent waves	474	70	—	—	—	—	—	7	—
Hair rinses (noncoloring)	158	34	—	—	1	8	24	21	16
Hair shampoos (noncoloring)	909	224	—	—	—	1	10	17	6
Tonics, dressings, and other hair grooming aids	290	35	—	—	—	3	36	133	52
Wave sets	180	39	1	—	1	—	11	17	5
Other hair preparations (noncoloring)	177	18	—	—	2	—	4	19	14
Hair tints	15	14	—	—	—	1	6	7	4
Hair rinses (coloring)	76	24	—	—	—	—	13	1	—
			—	—	—	—	—	—	24

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Hair bleaches	111	7	-	-	-	-	3	3	1
Other hair coloring preparations	49	1	-	-	-	-	-	1	-
Blushers (all types)	819	5	-	-	-	-	-	5	-
Face powders	555	5	-	-	-	-	-	2	3
Makeup foundations	740	10	-	-	-	-	-	8	2
Lipstick	3319	15	-	-	-	-	-	9	6
Makeup bases	831	15	-	-	-	-	-	10	5
Cuticle softeners	32	3	-	-	-	-	-	1	2
Nail creams and lotions	25	6	-	-	-	-	2	1	1
Nail polish and enamel	767	1	-	-	-	-	1	-	-
Nail polish and enamel remover	41	2	-	-	-	-	-	-	2
Other manicuring preparations	50	6	-	-	-	-	3	1	2
Bath soaps and detergents	148	3	-	-	-	-	1	2	-
Aftershave lotions	282	3	-	-	-	-	-	3	-
Skin cleansing preparations (cold creams, lotions, liquids, and pads)	680	27	-	-	-	-	4	14	9
Face, body, and hand skin care preparations (excluding shaving preparations)	832	46	-	-	-	-	4	37	4
Moisturizing skin care preparations	747	43	-	-	1	1	11	23	7
Night skin care preparations	219	11	-	-	-	1	5	5	-
Paste masks (mudpacks)	171	6	-	-	-	-	3	1	2
Skin fresheners	260	7	-	-	-	-	1	5	1
Wrinkle smoothers (removers)	38	1	-	-	-	-	1	-	-
Other skin care preparations	349	7	-	-	-	1	2	2	2
1981 TOTALS		923	3	3	7	30	208	461	211

In other studies, a proteinase from *Pseudomonas aeruginosa* catalyzed the hydrolysis of collagen,⁽³⁵⁾ whereas anthocyanosides isolated from *Vaccinium myrtillis* decreased collagen hydrolysis.⁽³⁶⁾ Hydrolyzed Collagen induced the activity of an extracellular collagenase produced by a marine *Vibrio*,⁽³⁷⁾ and a secondary vitamin K deficiency in rats increased the hydrolysis of collagen.⁽³⁸⁾ The epimerization of trans-4-hydroxy-L-proline to cis-4-hydroxy-D-proline during acid hydrolysis of collagen has also been documented.⁽³⁹⁾

Collagen hydrolysates have been studied for their stimulatory effect on the healing of open wounds.⁽⁴⁰⁾ The ears of 1 rabbit were incised: 1 was kept as a control and 1 was treated with Hydrolyzed Collagen. The same rate of healing was observed for the first few days; thereafter, the treated ear healed more rapidly.⁽¹⁶⁾

Animal Toxicology

Acute Toxicity

Oral

Hydrolyzed Collagen (100 percent) was analyzed for acute oral toxicity in rats in 2 tests. LD₅₀s were > 10 g/kg and > 15 g/kg, respectively. In each study, investigators concluded that Hydrolyzed Collagen was nontoxic by ingestion^(41,42) (Table 4).

Two shampoo formulations, each containing 2 percent Hydrolyzed Collagen, were tested for acute oral toxicity in mice and rats. LD₅₀s were > 15 ml/kg in both mice and rats. The investigators concluded that each formulation was practically nontoxic^(43,44) (Table 4).

A hair conditioner containing 0.5 percent Hydrolyzed Collagen had an oral LD₅₀ of > 40.0 ml/kg in rats, estimated by interpolation from the probit response curve. The investigators' observations included decreased activity, rales, diarrhea, salivation, and an increase in body weight⁽⁴⁶⁾ (Table 4).

Ocular Irritation

Four lots of Hydrolyzed Collagen (100 percent) were evaluated for ocular irritation by a modified Draize eye test. A 0.1 ml sample of Hydrolyzed Collagen

TABLE 4. Acute Oral Toxicity of Hydrolyzed Collagen

Compound	Species	LD ₅₀	Results/Comments	Reference
Hydrolyzed Collagen, 100%	Rat	> 10 g/kg	Nontoxic	41
Hydrolyzed Collagen, 100%	Rat	> 15 g/kg	Nontoxic	42
Hydrolyzed Collagen, 2% in a shampoo	Mouse	> 15 g/kg	Practically nontoxic	43
Hydrolyzed Collagen, 2% in a shampoo	Rat	> 15 g/kg	Practically nontoxic	44
Hydrolyzed Collagen 0.5% in a hair conditioner	Rat	> 40.0 ml/kg	—	45

was instilled into the conjunctival sac of 1 eye of each of 6 albino rabbits. Eyes were scored by Draize classification (max, 110) at 1, 2, 3, 4, and 7 days or until negative. The 4 lots had average irritation scores of 5, 4, 3, 0; 6, 3, 1, 0; 5, 3, 3, 0; and 6, 3, 3, 0 on Days 1, 2, 3, and 4, respectively. As judged by the Draize classification of eye irritation, Hydrolyzed Collagen was mildly irritating⁽⁴⁷⁾ (Table 5).

Hydrolyzed Collagen was evaluated as a 50 percent aqueous solution in a modified Draize eye irritation test. A 0.1 ml sample of the solution was instilled into the conjunctival sac of 1 eye of each of 6 albino rabbits; the other eye served as a control. The eyes were examined for 7 days or until negative. Average irritation scores were 1 and 0 (max, 110) on Days 1 and 2, respectively. As judged by the Draize classification of eye irritation, the test solution was minimally irritating⁽⁴⁸⁾ (Table 5).

A shampoo formulation containing 2 percent Hydrolyzed Collagen was tested for ocular irritation in 9 albino rabbits. The shampoo was diluted to a concentration of 20 percent (w/v) with distilled water, and a 0.1 ml sample was instilled into the conjunctival sac of the left eye. Each right eye served as a control.

TABLE 5. Ocular Irritation of Hydrolyzed Collagen

Compound	Number of Rabbits/Test Groups	Results/Comments	Reference
Hydrolyzed Collagen, 100% (4 lots)	6	Average irritation scores (max, 110) on Days 1, 2, 3, and 4, respectively were: Lot 1–5, 4, 3, 0 Lot 2–6, 3, 1, 0 Lot 3–5, 3, 3, 0 Lot 4–6, 3, 3, 0; Hydrolyzed Collagen was concluded to be mildly irritating	47
Hydrolyzed Collagen, 50% aqueous solution	6	Average irritation scores (max, 110) were 1 and 0 on Days 1 and 2, respectively; test solution concluded to be minimally irritating	48
Hydrolyzed Collagen, 0.2% in a shampoo, tested as a 20% (w/v) aqueous dilution	9/ 3 rinsed 6 unrinsed	No irritation observed in rinsed eyes; unrinsed eyes had mean total scores (max, 130) of 3.33, 0.67, 0.0, and 0.0 at 24, 48, and 72 hours and 7 days, respectively; concluded that shampoo was a nonirritant but could cause minimal irritation under test conditions	49
Hydrolyzed Collagen, 0.5% in a hair conditioner	9/ 3 unrinsed 3 rinsed 3 received 1:9 dilution with water	Conjunctival scores of 2 or less (max, 110) seen in all unrinsed eyes, 1 rinsed, and 1 dilution eye; effects cleared by 72, 72, and 48 hours, respectively; concluded that hair conditioner was a mild, transient irritant under all test conditions	50

The eyes of 3 rabbits were rinsed with water 2 seconds after application; the other 6 were not rinsed. Eyes were scored at 24, 48, and 72 hours and 7 days. The unrinsed treated eyes had conjunctival irritation at 24 hours consisting of slight erythema (6 rabbits), very slight edema (3 rabbits), and slight discharge (1 rabbit). Irritation decreased progressively, and all unrinsed eyes were normal at 72 hours. No irritation was observed in the rinsed eyes throughout the 7-day period. Mean total scores for the unrinsed eyes were 3.33, 0.67, 0.0, and 0.0 (max, 130) at 24, 48, and 72 hours and 7 days, respectively. The shampoo was found to be a nonirritant. However, under test conditions, it could cause minimal eye irritation⁽⁴⁹⁾ (Table 5).

A hair conditioner containing 0.5 percent Hydrolyzed Collagen was tested for ocular irritation in 9 albino rabbits. A 0.1 ml sample was applied to the right eye of each rabbit. The rabbits were divided into groups of 3: Group I received the product full-strength, Group II received a full-strength application followed by rinsing 4 seconds later, and Group III received a 1:9 dilution of the conditioner with water. Ocular reactions were recorded 24, 48, and 72 hours and 7 days after instillation of the test material. Conjunctival effects (score of 2 or less, max, 110) were seen in all 3 rabbits of Group I and in 1 rabbit of Groups II and III. These effects had disappeared by 72 hours in Groups I and II and by 48 hours in Group III. The investigators concluded that the hair conditioner was a mild, transient irritant when applied full-strength with or without rinsing, or diluted 1:9 with water⁽⁵⁰⁾ (Table 5).

Dermal/Primary Skin Irritation

Hydrolyzed Collagen (100 percent) was tested for primary skin irritation using 6 rabbits. The backs of the rabbits were clipped and divided into 2 sections, of which 1 was abraded. Hydrolyzed Collagen was applied to both sections under gauze pads for 24 hours. Sites were scored upon patch removal and 48 hours later. No reactions were noted. The investigators concluded that Hydrolyzed Collagen was nonirritating⁽⁴¹⁾ (Table 6).

Hydrolyzed Collagen was applied full-strength to the shaved backs of 9 rabbits in a modified Draize primary skin irritation test. A 0.1 ml sample was applied to each rabbit by occlusive filter disc for 24 hours. Four lots of Hydrolyzed Collagen were evaluated. Sites were graded upon disc removal and 48 hours later. Average irritation scores (max, 4) for the 4 lots were 0.25, 0.20, 0.16, and 0.26. The investigators concluded the Hydrolyzed Collagen was minimally irritating⁽⁵¹⁾ (Table 6).

Hydrolyzed Collagen was evaluated as a 50 percent aqueous solution for primary skin irritation using 9 albino rabbits. Samples of 0.1 ml were applied by occlusive filter disc to the shaved skin of the rabbits for 24 hours. Applications were made for 3 consecutive days. Sites were scored for irritation 24 hours after each application. The maximum average irritation response was 1.33 (max, 4); the test solution was considered mildly irritating⁽⁵²⁾ (Table 6).

Various concentrated solutions of a soap containing 26 to 28 percent Hydrolyzed Collagen were brushed onto the skin of guinea pigs (number unspecified). Skin changes were monitored clinically and microscopically. Body weights were recorded and necropsies performed on a number of the animals. No internal in-

juries were noted. The soap was considered significantly less irritating than a lauryl sulfate solution of the equivalent concentration⁽⁵³⁾ (Table 6).

A shampoo formulation containing 2 percent Hydrolyzed Collagen was evaluated for skin irritation and acute dermal toxicity. Three groups of 4 rabbits each received doses of 1.5, 3, and 6 ml/kg of the test shampoo, and a fourth group received 6 ml/kg of a regular shampoo as a comparative control. Backs of all rabbits were clipped; 2 in each group also were abraded. The shampoos were applied full-strength under an occlusive wrap for 24 hours. Sites were evaluated according to Draize at 24, 48, and 72 hours and 7 and 14 days. At 24 and 48 hours, moderate erythema (score of 2 on 0 to 4 scale) was seen in all control rabbits and 1 high-dose nonabraded rabbit. Slight erythema (score of 1) was noted in all others, and no edema was noted. The skin appeared darkened at 48 hours in 1 control rabbit and in the high-dose rabbit with moderate erythema. At 72 hours, no erythema or edema was noted. However, the same 2 rabbits still had darkened skin, whereas peeling and scaling of the skin was observed in the others. At 7 and 14 days, treated skin sites appeared normal except for areas of pustular dermatitis due to a secondary infection; 1 high-dose rabbit had darkened, thickened skin (Day 7), and 1 had dry, cracked, and peeling skin (Day 14). The investigators reported that application of the shampoo caused erythema and burning of the skin of the rabbit. Its application, under exaggerated conditions, was responsible for the degeneration and necrosis of the skin of some rabbits and was followed by secondary bacterial infection in others as manifested by a pustular dermatitis. There were no significant differences in hematological and urinalysis values. A dose-related decrease in feed consumption and body weight gain was observed. However, there were no other signs of systemic toxicity. Necropsy of the 3 rabbits that died during the study (2 control, 1 high dose) indicated that acute pneumonia was the prime contributing factor⁽⁵⁴⁾ (Table 6).

Two other shampoo formulations, each containing 2 percent Hydrolyzed Collagen, were evaluated for skin irritation using identical tests. The first shampoo was administered full-strength, whereas the second was diluted to 20 percent (w/v) in distilled water. Samples of 0.5 ml of the shampoo or dilution were administered to the intact and abraded skin of 6 rabbits and occluded for 24 hours. Sites were scored according to Draize at 24, 48 (shampoo only), and 72 hours. Moderate to severe irritation was observed in the rabbits treated with the full-strength shampoo; very slight to mild irritation was seen in the rabbits receiving the dilution treatments (Table 6). The shampoo and dilution had primary irritation scores (PIS) of 4.67 and 1.46, respectively, where a score of 5 (max, 8) is considered to be a primary skin irritant. The investigators concluded that the shampoo was a dermal irritant to the intact and abraded skin of rabbits, whereas the dilution was classified as a nonirritant. However, it was noted that mild irritation can occur under these test conditions.^(55,56)

A hair conditioner containing 0.5 percent Hydrolyzed Collagen was evaluated for dermal irritation in a combined irritation and phototoxicity test. Occlusive patches containing 0.20 ml of the conditioner were applied to the backs of 6 clipped rabbits for 48 hours. Sites were scored for erythema and edema at 49, 72, and 96 hours; all rabbits had a score of 0 (max, 8). It was concluded that the hair conditioner was not a primary dermal irritant⁽⁵⁷⁾ (Table 6).

TABLE 6. Primary Skin Irritation/Dermal Toxicity of Hydrolyzed Collagen

Compound	Test Method	Number and Species of Animals	Results/Comments	Reference
Hydrolyzed Collagen, 100%	Applied under gauze pads for 24 hours, I, A*	6 rabbits	No reactions PII [†] of 0; nonirritating	41
Hydrolyzed Collagen, 100% (4 lots)	Occlusive disc for 24 hours—I	9 rabbits	Average irritation scores (max, 4) for 4 lots were 0.25, 0.20, 0.16, and 0.26; concluded to be minimally irritating	51
Hydrolyzed Collagen, 50% aqueous solution	Occlusive disc for 24 hours, 3 consecutive applications—I	9 rabbits	Maximum average irritation response was 1.33 (max, 4); test solution considered mildly irritating	52
Hydrolyzed Collagen, 26–28% in a soap	Various concentrations brushed on—I	Guinea pigs, unspecified number	Considered significantly less irritating than a lauryl sulfate solution of the equivalent concentration; no internal injuries noted	53
Hydrolyzed Collagen, 2% in a shampoo	Occlusive patch for 24 hours—I, A—doses of 1.5, 3, and 6 ml/kg	12 rabbits	Moderate erythema observed in all control rabbits and 1 high-dose rabbit (score of 2, max, 4), slight erythema (score of 1) in all others, clearing by 72 hours; no edema noted; several rabbits had darkened or cracked and peeling skin, pustular dermatitis due to secondary infection in others; dose-related decrease in feed con-	54

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Hydrolyzed Collagen, 2% in a shampoo	Occlusive patch for 24 hours—I, A	6 rabbits	sumption and body weight gain; no other signs of systemic toxicity; conclusion: under exaggerated conditions, caused erythema and burning of rabbit skin and was responsible for the degeneration and necrosis of skin, allowing secondary infection	55
Hydrolyzed Collagen, 2% in a shampoo – diluted to 20% (w/v) in distilled water	Occlusive patch for 24 hours—I, A	6 rabbits	Moderate to severe and well-defined erythema was observed in 5 and 1 rabbits, respectively, at 24 hours, with similar results at 72 hours; very slight, slight, and moderate edema was observed in 1, 3, and 2 rabbits, respectively, at 24 hours, decreasing minimally by 72 hours; PIS [†] of 4.67 (max, 8); concluded to be dermal irritant on I and A skin	56
Hydrolyzed Collagen, 0.5% in a hair conditioner	Occlusive patch for 48 hours—I	6 rabbits	Well-defined and very slight erythema was observed in 3 and 3 rabbits, respectively, at 24 hours, diminishing to only very slight scores in 5 rabbits at 72 hours; slight and very slight edema was observed in 1 and 3 rabbits, respectively, at 24 hours, clearing totally by 72 hours; PIS of 1.46 (max, 8); concluded to be a nonirritant on I and A skin but can cause mild irritation under study conditions	57
			No erythema or edema; all rabbits had individual score of 0 (max, 8); concluded to be not a primary dermal irritant	

*I, intact; A, abraded.

[†]P_{II}, primary irritation index.[‡]PIS, primary irritation scores.

Subchronic Toxicity

Dermal

A hair preparation containing 2 percent Hydrolyzed Collagen was tested for subchronic dermal toxicity. Three groups of 2 male and 2 female rabbits received 100, 1000, or 3200 mg/kg of the test shampoo. Control groups received 2000 mg/kg of a marketed antidandruff shampoo, 3220 mg/kg of the test shampoo without the active drug ingredient, and 1 ml water/kg. All test and control applications were made daily for 30 days, were left on the skin for 15 minutes, and were then removed with water. The skin of 1 male and 1 female in each group was abraded weekly. The only treatment-related finding was local skin irritation. No deaths, abnormal behavior, gross or microscopic lesions were associated with treatment.⁽⁵⁸⁾

Another shampoo containing 2 percent Hydrolyzed Collagen was tested for dermal toxicity using Yorkshire pigs (white). Three groups of 2 male and 2 female pigs (1 of each sex abraded twice weekly) received applications of 0.5, 1.0, and 2.0 ml/kg. Control groups received 2.0 ml/kg saline and 2.0 ml/kg of another marketed shampoo. Applications were made to the clipped back of each pig twice daily for 4 weeks. Treated sites were rinsed with warm water 1 hour after each application. All pigs were given a general physical examination before and at 4 weeks observation. Body weights were recorded weekly, and blood samples were obtained for routine hematological and serum chemistry evaluations. All animals were necropsied. The abraded skin of 1 high-dose female was slightly irritated; the skin of all others appeared unremarkable. No dose-related effects were determined by physical examination, hematological and serum chemistry evaluations, necropsy examination, and histopathological evaluation. A statistically significant dose-regression relationship for male gonad weights was not considered indicative of systemic toxicity due to the immaturity and variation in size of the testes of these young pigs. Minimal focal inflammatory cellular infiltration was noted in the treated dermis of 1 middle-dose and 1 high-dose pig. However, this same condition was observed in the untreated skin of 1 saline control and 1 high-dose pig.⁽⁵⁹⁾

Sensitization

Hydrolyzed Collagen was tested for sensitization using 2 male white guinea pigs. A 0.1 percent solution of Hydrolyzed Collagen in physiological saline was injected intracutaneously into the clipped back or flank of each guinea pig every other day or 3 times weekly for a total of 10 injections. The first injection consisted of 0.05 ml of the test solution; each succeeding injection consisted of 0.1 ml. Sites were scored for diameter, height, and color 24 hours after each injection. After a 2-week rest period, a challenge injection of 0.05 ml was administered into a different site. Induction injections gave average diameter scores of 7.6 and 8.9 mm, average heights of 0.3 and 0.2 mm, and an average color of pink for the 2 guinea pigs. On challenge, 1 animal had no reaction, whereas the second had a reaction of diameter 5 mm, no height, and a pink color. Both animals had a sensitization score of 0 (9 or above is severely sensitizing), classifying Hydrolyzed Collagen as a nonsensitizer.⁽⁴¹⁾

A shampoo formulation containing 2 percent Hydrolyzed Collagen was

tested for sensitization using a modification of the Buehler and Griffith⁽⁶⁰⁾ method. A total of 3 inductive applications, 1 per week, were made to the clipped back of 10 guinea pigs. The first inductive application consisted of 0.5 ml of a 1, 5, and 10 percent dilution (distilled water v/v) occlusively patched on 3 separate sites on the right side of the animal for 24 hours. The second and third inductions consisted of 0.5 ml of a 10 percent aqueous dilution occlusively patched on the left side of the animal for 6 hours. A 24-hour challenge patch was applied 2 weeks later to an untreated site on the animal's right side. Sites were scored for erythema 24 hours after each application. Dinitrochlorobenzene (DNCB) was tested as a positive control. The first and third inductive patches produced no erythema, and the second produced very slight erythema in 2 guinea pigs. No erythema was observed at challenge. The investigators concluded that the shampoo formulation did not cause sensitization.⁽⁴⁵⁾

Phototoxicity

A shampoo formulation containing 2 percent Hydrolyzed Collagen was tested for phototoxicity using 2 guinea pigs. The shampoo was tested as a 20 percent (w/v) mixture in distilled water, and 8-methoxypsoralen was tested undiluted as a positive control. The back of each animal was clipped and divided into 4 sites; 2 received 0.1 ml of the shampoo and 2 received 0.05 ml of 8-methoxypsoralen. Fifteen to twenty minutes later, the right side of each animal was shielded with cardboard while the animals were irradiated for 1 hour with UVA light (320 to 400 nm) using a No. F40 BL 40W Westinghouse Blacklight. Sites were graded for erythema (max, 4) 24 hours after exposure. All exposed and unexposed sites treated with the shampoo dilution had scores of 0. 8-Methoxypsoralen gave a mean score of 3.5 for the irradiated sites and a score of 0 for the non-irradiated sites. The investigators concluded that the shampoo formulation was not phototoxic in guinea pigs.⁽⁶¹⁾

A hair conditioner containing 0.5 percent Hydrolyzed Collagen was also analyzed for phototoxicity using 6 rabbits. One rabbit received 8-methoxypsoralen as a positive control. A 0.20 ml sample of the conditioner was applied to a gauze patch, evaporated for 5 minutes, and then placed on the clipped back of the rabbit and occluded. Two patches were applied to each animal. Two hours later, 1 patch was removed and the other protected with aluminum foil while the animals were irradiated for 15 minutes with Sylvania lights No. F-40-BLB. The patches were then replaced until 48 hours posttreatment, at which time all patches were removed. Sites were scored for erythema and edema (max, 8) 1 hour later and at 72 and 96 hours. Each rabbit had an individual score of 0 for both irradiated and nonirradiated sites. The hair conditioner was neither a primary dermal irritant nor a phototoxic irritant to rabbit skin.⁽⁵⁷⁾

CLINICAL ASSESSMENT OF SAFETY

Irritation and Sensitization

Hydrolyzed Collagen (100 percent) was tested for skin irritation on 20 humans. A single patch containing 0.1 ml of Hydrolyzed Collagen was applied to the volar forearm or the inner aspect of the arm of each subject. A standard con-

trol was also tested. Reactions were recorded 2 and 24 hours after patch removal. The irritation score for Hydrolyzed Collagen and the controls in all 20 subjects was 0 (max, 4). No significant difference in irritancy potential existed between Hydrolyzed Collagen and the control⁽⁶²⁾ (Table 7).

Patch tests were performed on 33 subjects (18 men and 15 women) using Hydrolyzed Collagen at concentrations of 2 and 20 percent. Occlusive patches containing Hydrolyzed Collagen at each concentration were applied to the breast or arm for 24 hours. Sites were scored at 24, 48, and 72 hours; no reactions were observed⁽⁶³⁾ (Table 7).

A 21-day cumulative irritation test was conducted on a hair conditioner containing 0.5 percent Hydrolyzed Collagen. Semiocclusive patches with 0.5 ml of the conditioner were applied to the upper part of the back of 20 subjects for 24 hours. Patches were then removed, evaluated 30 minutes later, and a new patch was applied. These procedures were repeated for 15 applications, allowing for 21-day continuous exposure. Mineral oil was used as the standard control. Of the 17 subjects who completed the study, only 1 had any reaction, giving a cumulative irritation score of 0.5 (max, 84). The mean cumulative irritation score of 0.03 was exactly comparable to that of mineral oil (0.03). The investigators concluded that the product as used by label directions would not present any medical hazard to the consumer⁽⁶⁴⁾ (Table 7).

Various compositions of a soap containing 26 to 28 percent Hydrolyzed Collagen were applied daily to the skin as a 5 percent solution for 10 to 48 days. A large number of healthy subjects and people with dermatitis were used. A low degree of irritation was seen even at high concentrations of the least irritating composition. No sensitization was observed. The treated skin area was examined microscopically; those with acute dermatitis had moderate irritation⁽⁵³⁾ (Table 7).

Five cosmetic formulations were evaluated for irritation and sensitization in repeated insult patch tests (RIPT). Three of these, a morning cream, a suntan lotion, and a night cream, containing 3.0, 2.2, and 3.0 percent Hydrolyzed Collagen, respectively, were tested in the same manner. A series of 10 48-hour occlusive patches containing the undiluted formulation was applied to the back of each subject. Sites were graded after each removal and 24 hours after removal of the tenth patch (morning cream and suntan lotion only). After approximately an 11-day rest period, challenge patches were applied, occluded for 48 hours, and scored upon removal and 24 hours later. Scattered irritant responses after the third application were seen in the 103 subjects who completed the induction phase for the morning cream and suntan lotion. The maximum number of responses seen at any 1 reading for the suntan lotion was 11 with erythema and 8 with very mild erythema. Maximum response to the morning cream was 5 with erythema and 10 with very mild erythema. Of the 96 subjects who completed the challenge phase, 1 had erythema and 1 had very mild erythema to the suntan lotion at 24 hours. However, the panelist with erythema had a negative reaction on rechallenge. No reactions were observed on challenge with the morning cream. Two of the 113 panelists completing the induction phase testing of the night cream had irritant responses: 1 had very mild erythema and 1 had erythema. One of the 103 subjects completing the challenge phase had erythema. The investigators concluded that the morning cream, suntan lotion, and night cream were mildly irritating, definitely irritating, and nonirritating, respectively, whereas none of the formulations gave significant evidence of sensitization^(65,66) (Table 7).

TABLE 7. Clinical Irritation and Sensitization

Ingredient	Type of Test	Number of Humans	Results/Comments	Reference
Hydrolyzed Collagen, 100%	Single patch-type and duration not specified	20	Average irritation score of 0 for all 20 subjects (max, 4); no significant difference between test material and control	62
Hydrolyzed Collagen, 2 and 20%	Single, 24-hour occlusive patch	33	No reactions were observed	63
Hydrolyzed Collagen, 0.5% in a hair conditioner	21-day cumulative irritation test	17	One subject had a reaction, giving a cumulative irritation score of 0.5 (max, 84); mean cumulative irritation score of 0.03 was exactly comparable to control; product should not present any medical hazard; nonirritant	64
Hydrolyzed Collagen, 26–28% in a soap – tested as 5% solution of the soap	Applied daily to skin for 10–48 days	Large number (unspecified), both healthy subjects and some with dermatitis	Low degree of irritation, no sensitization was observed; those with acute dermatitis showed moderate irritation	53
Hydrolyzed Collagen, 3.0% in a morning cream	RIPT*	103–I† 96–C	Scattered irritant responses, maximum response at any one reading was 10 very mild erythemas and 5 erythemas; no reactions on challenge; mildly irritating and nonsensitizing	65
Hydrolyzed Collagen, 2.2% in a suntan lotion	RIPT	103–I 96–C	Scattered irritant responses, maximum response at any one reading was 8 very mild erythemas and 11 erythemas; 2 reactions to the challenge, 1 very mild erythema and 1 erythema; negative reaction on rechallenge; definitely irritating and nonsensitizing	65
Hydrolyzed Collagen, 3.0% in a night cream	RIPT	113–I 103–C	Two irritant responses: 1 very mild erythema and 1 erythema; 1 erythema reaction on challenge; nonirritating and nonsensitizing	66
Hydrolyzed Collagen, 0.5% in a mascara	RIPT	205	One subject exhibited faint erythema during the induction phase; no reactions at challenge; nonirritating and non-sensitizing	67

TABLE 7. (Continued)

<i>Ingredient</i>	<i>Type of Test</i>	<i>Number of Humans</i>	<i>Results/Comments</i>	<i>Reference</i>
Hydrolyzed Collagen, 0.5% in a hair con- ditioner tested as a 0.1% dilution	RIPT	207 – I 201 – C	Mean cumulative irritation scores (max, 50) as follows: 167 subjects had score of 0 22 subjects had score of 1 12 subjects had score of 2 1 subject had score of 3 1 subject had score of 4 3 subjects had score of 5 1 subject had score of 6; one subject exhibited erythema at challenge; however, this panelist reacted to 10 of the other 13 products and had no reaction on rechallenge No reactions were observed	68
Hydrolyzed Collagen, 0.5% in a mascara	Controlled use test, 4 weeks of daily use	27		69

*Repeated insult patch test.

†I, induction; C, challenge.

The fourth formulation tested by RIPT was a mascara containing 0.5 percent Hydrolyzed Collagen. Occlusive patches containing the undiluted mascara were applied to the upper backs of 205 subjects on Monday, Wednesday, and Friday for 3 consecutive weeks. Patches were removed and sites graded just prior to the next scheduled patch application. Following a 2-week rest, 2 consecutive 48-hour challenge patches were applied to adjacent sites on the back. Sites were graded at 48 and 96 hours. One subject had faint erythema during the induction phase; no reactions were observed at challenge. The mascara was found to be neither an irritant nor a sensitizer⁽⁶⁷⁾ (Table 7).

The fifth formulation evaluated by RIPT was a hair conditioner containing 0.5 percent Hydrolyzed Collagen. A 0.1 percent dilution of the conditioner was applied using semiocluded patches to the upper backs of the subjects for 48 hours. Patches were then removed, sites evaluated, and new patches applied for a total of 10 applications. Following a 2-week rest period, challenge patches were applied to the subjects' thighs. Mean cumulative irritation scores (max, 50) were as follows: 167 subjects had a score of 0, 22 subjects had a score of 1, 12 subjects had a score of 2, 3 subjects had scores of 3, 4, and 6 (each), and 3 subjects had a score of 5. Of the 201 subjects completing the challenge phase of the study, 1 had erythema. However, this panelist also reacted to 10 of the other 13 substances being tested. A rechallenge on the other thigh produced no reaction⁽⁶⁸⁾ (Table 7).

A mascara formulation containing 0.5 percent Hydrolyzed Collagen was evaluated by a 4-week controlled use test. Twenty-seven women used the product daily as per normal instructions. No reactions were observed⁽⁶⁹⁾ (Table 7).

A prospective study of cosmetic-induced dermatitis by 11 dermatologists of the North American Contact Dermatitis Group (NACDG) identified 1 case of dermatitis associated with use of Hydrolyzed Collagen from among a total of 487 cases.⁽⁷⁰⁾

Phototoxicity/Photosensitization

A mascara formulation containing 0.5 percent Hydrolyzed Collagen was tested for phototoxicity/photosensitization on a panel of 23 humans. Occlusive patches containing 0.1 g/cm² of the mascara were applied to the backs of the subjects for 24 hours. Patches were then removed, evaluated, and irradiated with 3 times the individual's MED using a xenon arc solar simulator (150 W) filtered to produce a continuous UVA-UVB emission spectrum (290 to 400 nm). Sites were evaluated 48 hours later, and the procedures of application, patching, and irradiation were repeated for a total of 7 applications. No reactions were observed. The investigators concluded that the mascara was neither phototoxic nor a photosensitizer.⁽⁷¹⁾

Domsch et al.⁽⁷²⁾ have found that UV-induced erythema was decreased by rubbing Hydrolyzed Collagen (mean molecular weight of 1500) into the skin. A 10 percent solution of Hydrolyzed Collagen applied immediately and 24 hours after irradiation decreased erythema by 20 percent at 24 hours and 25 percent at 48 hours.

SUMMARY

Hydrolyzed Collagen is a collagen hydrolysate derived by acid, enzyme, or other method of hydrolysis. It is a white to off-white hygroscopic powder of molecular weight 1000 to 10,000 and is also available as a viscous, amber aqueous solution.

Hydrolyzed Collagen can be prepared by any 1 of 3 methods: alkaline hydrolysis of bovine skin products followed by enzymatic hydrolysis to the desired molecular weight, enzymatic hydrolysis of fresh animal byproducts or bovine-derived leather, or acid or alkaline hydrolysis of chrome leather fold splinters with inorganic acids or lyes to a defined molecular weight. The hydrolysis of collagen yields a high content of glycine and proline compared to other proteins, as well as 2 unique amino acids, hydroxyproline and hydroxylysine. Hydrolyzed Collagen is usually analyzed by column chromatography.

Hydrolyzed Collagen is used in cosmetics, primarily hair and skin care products, because of its conditioning and moisturizing properties. It is usually incorporated at concentrations <5 percent and was in 923 formulations reported in 1981. Cosmetic products containing Hydrolyzed Collagen may contact all external body surfaces and hair, as well as the eyes. Frequency and length of application could result in continuous exposure.

Many studies have documented the substantivity of Hydrolyzed Collagen to human hair and skin. Hydrolyzed Collagen generally is adsorbed on hair, although it has been shown to penetrate the cuticle and cortex. Increasing concentrations of Hydrolyzed Collagen increased adsorption, as did increasing damage to hair. Hydrolyzed Collagen also binds water to the hair and skin surfaces and is used as a buffering agent for alkali in permanent waving preparations.

Acute toxicity studies found Hydrolyzed Collagen and formulations containing Hydrolyzed Collagen practically nontoxic when administered orally to mice and rats. Dermal studies gave no indication of systemic toxicity when formulations containing Hydrolyzed Collagen were applied to rabbits and guinea pigs. However, a shampoo formulation (2 percent Hydrolyzed Collagen) administered to rabbits under exaggerated conditions did cause erythema and burning, leading to degeneration and necrosis of the skin.

Hydrolyzed Collagen was minimally irritating to rabbit eyes when tested full-strength and in formulation. Primary skin irritation tests in rabbits indicated that Hydrolyzed Collagen was nonirritating or minimally irritating when tested full-strength, whereas a 50 percent aqueous solution of Hydrolyzed Collagen was mildly irritating. Shampoo formulations containing Hydrolyzed Collagen (2 percent) were generally nonirritating when tested as dilutions. However, these were irritating under the exaggerated conditions of a full-strength application. A soap (26 to 28 percent Hydrolyzed Collagen) and hair conditioner (0.5 percent Hydrolyzed Collagen) produced no dermal irritation in guinea pigs and rabbits, respectively.

Subchronic dermal studies on 2 cosmetic formulations containing 2 percent Hydrolyzed Collagen were negative for systemic toxicity in rabbits and Yorkshire pigs.

Hydrolyzed Collagen was nonsensitizing in guinea pigs. Cosmetic formulations containing Hydrolyzed Collagen (2, 2, and 0.5 percent) were also nonsensitizing and nonphototoxic in guinea pigs and rabbits.

In clinical studies, Hydrolyzed Collagen produced no skin irritation. Formulations containing Hydrolyzed Collagen at concentrations ranging from 0.5 to 28 percent produced some irritation. However, no significant evidence of sensitization was observed in any study. No phototoxicity or photosensitization was evident in a study of a mascara containing 0.5 percent Hydrolyzed Collagen. It has been reported that UV-induced erythema was decreased by rubbing Hydrolyzed Collagen into the skin after irradiation.

CONCLUSION

On the basis of the available animal and clinical data presented in this report, the Panel concludes that Hydrolyzed Collagen is safe as a cosmetic ingredient in the present practices of use and concentration.

ACKNOWLEDGMENT

Elizabeth Meerman Santos, Scientific Analyst and writer, prepared the literature review and technical analysis used by the Expert Panel in developing this report.

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Final Report of the Safety Assessment of Cosmetic Ingredients Derived From Zea Mays (Corn)

International Journal of Toxicology
30(Supplement 1) 17S-39S
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DOI: 10.1177/1091581811403832
http://ijt.sagepub.com



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Abstract

Many cosmetic ingredients are derived from *Zea mays* (corn). While safety test data were not available for most ingredients, similarities in preparation and the resulting similar composition allowed extrapolation of safety data to all listed ingredients. Animal studies included acute toxicity, ocular and dermal irritation studies, and dermal sensitization studies. Clinical studies included dermal irritation and sensitization. Case reports were available for the starch as used as a donning agent in medical gloves. Studies of many other endpoints, including reproductive and developmental toxicity, use corn oil as a vehicle control with no reported adverse effects at levels used in cosmetics. While industry should continue limiting ingredient impurities such as pesticide residues before blending into a cosmetic formulation, the CIR Expert Panel determined that corn-derived ingredients are safe for use in cosmetics in the practices of use and concentration described in the assessment.

Keywords

cosmetics, safety, *Zea mays* (corn)

Introduction

Various cosmetic ingredients are derived from *Zea mays* (Corn) plants. These include oils, cob meal, cob powder, corn acid (fatty acids) and its potassium salt, corn glycerides and oil unsaponifiables, fruit, germ extract and oil, gluten protein, hydrolyzed starch and protein, kernel extract and meal, seed flour, starch, and silk extract.

While safety test data that directly examine toxicity are available for several of these ingredients, they are not available for all. Extensive data are available for corn oil as a result of its use as a vehicle in studies that examine the toxicity of another entity.

In addition, corn oil has been used as a component of diet in studies that examine the effects of high fat content in the diet; for example, the effect of starch in the diet on growth of foci of dysplastic crypts in the colon. Representative dietary studies are provided.

- Zea Mays (Corn) Cob Meal (no CAS number available) is the milled powder prepared from the cobs of *Zea mays* that functions as an abrasive and bulking agent.
- Zea Mays (Corn) Cob Powder (no CAS number available) is the powder obtained from the dried ground cobs of *Zea mays* that functions as an abrasive.
- Zea Mays (Corn) Fruit (no CAS number available) is the fruit of *Zea mays* that has no defined function in cosmetics.
- Zea Mays (Corn) Germ Extract (no CAS number available) is an extract of the germ of the corn, *Zea mays* that has no defined function in cosmetics.
- Zea Mays (Corn) Germ Oil (no CAS number available) is the oil obtained from the germ of Zea Mays that functions as an occlusive skin conditioning agent.
- Zea Mays (Corn) Oil (CAS #8001-30-7) is the refined fixed oil obtained from the wet milling of corn *Zea mays* that functions as a fragrance ingredient, hair conditioning agent,

Chemistry

Definition

The *International Cosmetic Ingredient Dictionary and Handbook* defines these ingredients and gives their functions in cosmetics as follows¹:

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occlusive skin conditioning agent, surfactant-emulsifying agent.

- Zea Mays (Corn) Oil Unsaponifiables (no CAS number available) is the fraction of Zea Mays (Corn) Oil (qv), which is not saponified in the refining recovery of corn oil fatty acids, that functions as a hair conditioning agent and skin conditioning agent—miscellaneous.
- Zea Mays (Corn) Gluten Protein (CAS #66071-96-3) is a protein obtained from the starchy endosperm of corn, *Zea mays* that functions as a bulking agent, hair conditioning agent, and skin conditioning agent—miscellaneous.
- Zea Mays (Corn) Kernel Extract (no CAS number available) is an extract of the kernels of corn, *Zea mays* that has no defined function.
- Zea Mays (Corn) Kernel Meal (CAS #66071-96-3) is the coarse flour prepared by milling the kernels of *Zea mays* that functions as an abrasive and bulking agent.
- Zea Mays (Corn) Seed Flour (CAS #68525-86-0) is the powder prepared from the fine grinding of the inner portion of the seeds of *Zea mays* that functions as an abrasive, absorbent, binder, and bulking agent.
- Zea Mays (Corn) Silk Extract (no CAS number available) is an extract of the stigmas of the corn, *Zea mays* that has no defined function.
- Zea Mays (Corn) Starch (CAS #9005-25-8) is a starch obtained from corn, *Zea mays*, that functions as an abrasive, absorbent, skin protectant, and aqueous viscosity increasing agent.
- Hydrolyzed corn starch (no CAS number available) is the hydrolysate of Zea Mays (Corn) Starch (qv), derived by acid, enzyme, or other method of hydrolysis, that functions as a skin conditioning agent—humectant.
- Hydrolyzed Corn Protein (CAS #73049-73-7) is a hydrolysate of corn protein, derived by acid, enzyme, or other method of hydrolysis that functions as a hair conditioning agent and skin conditioning agent—miscellaneous.
- Corn Acid (CAS #68308-50-9) is a mixture of fatty acids derived from Zea Mays (Corn) Oil that functions as an opacifying agent and surfactant—cleansing agent.
- Potassium Cornate (CAS #61789-23-9) is the potassium salt of Corn Acid that functions as a surfactant—cleansing agent.
- Corn Glycerides (no CAS number available) is a mixture of mono, di, and triglycerides derived from Zea Mays (Corn) oil which functions as a skin conditioning agent—humectant and surfactant—emulsifying agent.

Zea Mays (Corn) Fruit can be considered as all edible crop tissues (fruit) of *Zea mays*.²

Zea Mays (Corn) oil or corn oil is a mixture consisting of glycerides of the following fatty acids: myristic 0.1% to 1.7%, palmitic 8% to 12%, stearic 2.5% to 4.5%, hexadecenoic 0.2% to 1.6%, oleic 19% to 49%, and linoleic 34% to 62%. The unsaponifiable fraction comprises 1% to 3% of the mixture. The crude oil may contain up to 2% phospholipids.³

Zea Mays (Corn) Starch is a carbohydrate polymer derived from corn of various types, composed of 25% amylase and 75% amylopectin.^{4,5}

Table 1. Physical and Chemical Properties of Corn Oil

Property	Description/Value
Color	Yellow ^a Pale-yellow ^b
Odor and taste	Faint ^a Characteristic ^b
Density	0.916-0.921 ^a 0.914-0.921 ^b
Melting point	-18°C to -10°C ^a -10°C ^b
Flash point	321°C ^a 254°C ^b
Ignition point	393°C ^{a,c}
Acid value	2-6 ^a
Saponification value	187-196 ^a
Iodine value	109-133 ^a 102-128 ^b

^a Budavari 1989³

^b Lewis 1997⁵

^c Lewis 1993¹⁴

Hydrolyzed corn starch can be considered the end product of starch-water treatment with dilute mineral acid. The acid modification reduces chain length but does not substantially change the molecular configuration.⁶

Although Zea Mays (Corn) Germ Oil and Zea Mays (Corn) Oil are listed in the *International Cosmetic Ingredient Dictionary and Handbook* as separate ingredients, the Corn Refiners Association reports that all Corn oil is made from corn germ (Corn Refiners Association, personal communication 2006). The additional term "Germ" is only to denote that a specific part of the plant was used in the manufacture of the cosmetic ingredient in the *International Cosmetic Ingredient Dictionary and Handbook* (Gottschalck, personal communication 2006).

For the sake of brevity, this report will use Corn instead of repeatedly spelling out Zea Mays (Corn). And corn oil will be used for both corn oil and corn germ oil.

Physical and Chemical Properties

Physical and chemical property information found on corn oil is given in Table 1.

Corn Silk Extract is a slightly viscous brown liquid with a pH of 4.5 to 6.5.⁷

Physical and chemical property information on Corn Cob Meal was available for a trade name ingredient, Grit-o'-cob, which is made from the woody ring of the corncob. This product is 84.4% structural polysaccharides and 7% water. Grit-o'-cob includes 36.9% crude fiber and 53.3% nitrogen-free extract (carbohydrates). Cellulose makes up 47.1% of the total, along with 37.3% hemicellulose. Of the total, 39.9% are hexosan, 36.5% are pentosan, and 31.6% are xylan. Physical and chemical properties and trace components for Grit-o'-cob appear in Table 2.

Table 2. Physical and Chemical Properties and Composition of a Tradename Corn Cob Meal, Grit-o'-cob Granules⁸

Property	Value ^a
Water absorption	133%
Bulk density	28.0 lbs/ft ³
Specific gravity	1.3
Surface area	5.85 m ² /g
Solubility in:	
Acetone	2.5%
Ethanol	1.6% (2 h)
Isopropyl alcohol	0.42%
1% potassium hydroxide	18.6%
10% sulfuric acid	2.5%
Hot water	9.5%
Room temperature Water	6.3%
Cation exchange Capacity	0.85 meq/g
Gross energy	4113 kcal/kg ^b
Total microorganism plate count	10,500/g
Coliform	Negative
Salmonella	Negative
Mold and yeast	80/g
Average particle diameter	0.00849-0.1785
Trace elements ^c	
Nitrogen	0.21%
Phosphorus	0.021%
Sulfur	0.013%
Potassium	0.93%
Sodium	0.14%
Magnesium	0.11%
Silicon	0.089%
Iron	0.013%
Calcium	0.011%
Alumnium	0.0053%
Selenium	<5ppb
Fat	0.2%
Protein	1.4%
Ash	1.2%
Vitamin A equiv	0.9 IU/g ^b
Carotene	0.5 mg/kg ^b
Biotin	0.02 µg/g
Niacin	6.8 µg/g
Pantothenic acid	2.5 µg/g
Pyridoxine	3.1 µg/g
Riboflavin	1.1 µg/g

^a Normalized to a 7% moisture basis.^b Estimated.^c Barium, chromium, copper, lead, manganese, nickel, vanadium, and zinc all <0.0001%.

A phytochemical and ethnobotanical database lists the chemicals expected to be present in various plant materials (Table 3).

Methods of Manufacture

Information on methods of manufacture are included where available:

Corn gluten protein (listed as corn gluten in the Code of Federal Regulations [CFR]) is produced as a byproduct

during the wet milling of corn for starch in whole or various fractions of dry milled corn to corn syrups (21CFR 184.1321).

Corn oil is usually extracted from the clean, dried corn germ by wet milling. Corn oil is produced by a combination of expelling in continuous screw presses and solvent extraction of the press cake. The initial expeller can recover a little more than half of the oil and subsequent solvent extraction with hexane, the preferred solvent, brings total yield to about 95%.

Corn silk extract is produced by dilute ethanol extraction of corn silk, the fresh styles and stigmas of *Zea mays* collected when the corn is in milk (21CFR 184.1262).

The raw herbal material is made into an extract using the percolation method with heated water. After soaking for approximately 12 hours, the liquid is extracted and filtered to remove insoluble material. Corn Silk Extract obtained and distilled by this method is blended with alcohol and water.¹⁰ This mixture is then heated and allowed to cool.

Corn starch is produced by softening cleansed corn with warm water containing dissolved sulfur dioxide to soften the kernel.⁶ Sulfur dioxide and water react to form sulfurous acid, which assists in separation of starch and protein. Softened corn kernels next pass through mild attrition mills to loosen the hull and free the germ from the starch-rich endosperm. Water is added to the attrition mills, and a thick slurry of macerated kernels and whole germ results. Because the germ at this stage contains 40% to 50% oil, it is lighter than the endosperm and hull. Density centrifugation is used to isolate the germ. The remaining mixture of hull and endosperm is ground and screened. Large hull particles are retained on screens and removed, while finer protein and starch particles pass through. The water slurry of starch and gluten protein is separated by centrifugation—nearly complete separation is achieved. Typical yields include a gluten stream containing over 60% protein, while the starch stream is greater than 99% starch. The white, nearly pure starch slurry is further washed to remove small quantities of solubles.

Hydrolyzed corn starch is produced by treating the slurry of washed starch (described above) with dilute mineral acid at temperature elevated but below the starch gelatinized temperature, for varying periods of time. The primary reaction taking place is hydrolysis of glucosidic bonds in starch molecules. At the desired viscosity, the acid is neutralized with sodium carbonate and the starch is filtered, washed, and dried.⁶

Analytical Methods

Adulteration of corn oil can be detected by the principal component scores of the $\delta^{13}\text{C}$ values of major and minor vegetable oil components.¹¹ This analytical method is based on the premise that each plant species has a unique pattern of naturally occurring stable isotopes of carbon, hydrogen, and oxygen and that variations between isotopic compositions of different

Table 3. List of Chemicals Present in Corn Plant (Duke 2007)⁹

Component	Chemicals Expected to be Present				
Anther	Peroxidase	Lignin	pantothenic-acid	pyridoxine	syringaldehyde
Cob	4-hydroxybenzaldehyde	Niacin	p-hydroxybenzaldehyde	riboflavin	vanillin
Cotyledon	Biotin				
Embryo	Hemicellulose	Tryptophan			
Endosperm	Indole-3-acetic-acid				
Essential Oil	Histidine				
Fruit	Protease inhibitor C1-4-A	Bisabolol	heptan-2-ol		
	2-ethyl-1-cyclohexen-1-yl	Campestanol ferulate	Fructose		
	4-acetyl-benzoxazolin-2-one	Campestanol-p-coumarate	Glucose	Oxalic-acid	Salicylates
	α -tocopherol	Chlorine	Glutathione	Palmitoleic-acid	Sucrose
	Amino acids	Cobalt	Glutelin	Phylloquinone	Sulfur
	Campestanol ferulate	Copper	Lignoceric-acid	Raffinose	Thiamin
	Campestanol-p-coumarate	Folacin		Riboflavin	Protease inhibitor
Oil	Chlorine	Gamma-tocopherol	Phylloquinone		
Pericarp	Alpha-tocopherol				
Plant	Chrysanthemin	Amino-adipic-acid	Galactose	Myo-inositol	Sd
	2-(2,4-dihydroxy-7-methoxy-1,4(2h)-benzoxazin-3(4h)				
	24-methylene-cycloartenol	Apigenin-glycoside	Galactoxylase	Myrecitin-glucoside	Serotin
	3-o-caffeoylquinic-acid	Arabinose	Gamma-sitosterol	Neocryptoxanthin	Subaphyllin
	3-o-coumarylquinic-acid	Arachidic-acid	Glutamic-acid-decarboxylase	Pectins	Succinic-acid
	4-vinyl-4-deethyl-chlorophyll-a	Beta-amylase	Glycerol	Pelargonidin-3-glucoside	Trans-ferulic-acid
	5-hydroxy-trans-ferulic-acid	Beta-zeacarotene	Guanidine	Phenylalanine-ammonia-lyase	Trans-sinapic-acid
	5-o-caffeoylquinic-acid	Beta-zein	Inositol	Phosphatidyl-choline	Tricarballoyl-acid
	5-o-coumarylquinic-acid	Cadaverine	Isoquercitrin	Phosphatidyl-ethanolamine	Tricin-glycoside
	5-o-feruloylquinic-acid	Cellulose	Lactic-acid	Phosphatidyl-inositol	Tryptamine
	6-methoxy-benzoxazolin-2-one	Colxol	Lanosterol	Phytin	Urease
	6, 7-dimethoxy-benzoxazolin-2-one	Cryptoxanthin	Leucocyanidin	Phytofluene	Uronic-acid
	Acetoin	Cyanidin-3-galactoside-p-coumaric-acid-ester	Leucopelargonidin	Quercetin-diglucoside	Xyloarabinose
	Aflatoxin B-1	Cyanidin-3-monoside	Linalool	Quinic-acid	Xylose
	Aflatoxin	Cystathione	Lipoxidase	Ricinoleic-acid	Zearalenone
	Aldobiouronic-acid	Cytochrome-c	Maleic-acid	Rubisco	Zeatin
	Alpha-amylase	D-glucuronic-acid	Malic-acid-dehydrogenase		Zeinoxanthin
	Alpha-amyrin	Dec-trans-2-cis-4-dien-1-al	Malonic-acid		
	Alpha-glucosidase	Dihydrostosterol	Maltosyl-transferase		
	Alpha-sitosterol	Dioxyannimic-acid	Mannose		
	Alpha-zeacarotene	Furfural	Monogalactosyl-diglyceride		
Seed	1-p-hydroxy-trans-cinnamoyl-glycerol	Calcium	Indole-3-acetic-acid-myoinositol	Pantothenic-acid	Vit-b-6
	28-norcastasterone	Calmodulin	Indole-3-butyric-acid	Phenylalanine	Water
	3-7-dihydroxy-indolin-2-one-3-acetic Acid	Campesterol	Iodine	Phospholipids	Ytterbium
	7-o-beta-glucoside				

(continued)

Table 3. (continued)

Component	Chemicals Expected to be Present	Carvacrol	Iron	Phosphorus	Zea Mays Alpha-amylase Inhibitor
	4-ethyl-guaiacol				
	4-ethylphenol	Castasterone	Isobehenic-acid	Phytate	
	4-methyl-guaiacol	Chitinase B	Isoleucine	Phytic-acid	Zeamatin
	4-vinyl-guaiacol	Chitinase a	Isopropyl-amine	Phytosterols	Zeanin
	4-vinyl-phenol	Cholesterol	Lead	Potassium	Zeanoside a
	6-deoxocastasterone	Choline	Leucine	Prolamine	Zeanoside-b
	8-hydroxy-quinol-2-one-4-carboxylic- acid-8'-o-beta-d-glucoside	Chromium	Linoleic-acid	Proline	Zeanoside-c
	Abcissin-ii	Chrysanthemin	Lithium	Protein	Zeaxanthin
	Adenine	Cystine	Lutein+Zeaxanthin	Pyrrolidine	Zein
	Alanine	Diethyl-amine	Lysine	Selenium	Zeta-carotene
	Allantoin	Digalactosyl-diglyceride	Magnesium	Silver	Zinc
	Alpha-carotene	Dimethyl-amine	Manganese	Sodium	Zirconium
	Alpha-linolenic-acid	Ethyl-amine	Melatonin	Spermidine	
	Alpha-tocopherol	Ethyl-methyl-amine	Mercury	Spermine	
	Alpha-zein	Eugenol	Methionine	Starch	
	Aluminum	Fat	Methyl-amine	Stearic-acid	
	Ammonia	Ferulic-acid	Mevalonic-acid	Stigmasterol	
	Aniline	Fixed-oil	Molybdenum	Strontium	
	Arginine	Folic-acid	Myristic-acid	Teasterone	
	Arsenic	Gamma-carotene	N-demethyl-diazepam	Threonine	
	Ascorbic-acid	Globulin	N-methyl-beta-phenethylamine	Tin	
	Ash	Glutamic-acid	N-methyl-phenethylamine	Titanium	
	Aspartic-acid	Glutathione	N-propyl-gallate	Tocopherols	
	Barium	Gluten	Niacin	Trans-ferulic-acid	
	Benzyl-amine	Glycine	Nickel	Trigonelline	
	Beta-carotene	Guaiaacol	Octadecadienoic-acid	Tryptophan	
	Beta-cryptoxanthin	Hexadecenoic-acid	Octadecatrienoic-acid	Tyrosine	
	Beta-sitosterol	Hexenyl-isothiocyanate	Octadecenoic-acid	Uridine	
	Boron	Histidine	Oleic-acid	Valine	
	Butenyl-isothiocyanate	Indole-3-acetic-acid-cellulosiglucon	P-coumaric-acid	Vanadium	
	Cadmium	Indole-3-acetic-acid-methyl-ester	Palmitic-acid	Vanillin	
	2-methyl-naphthalene	Decan-2-one	Heptan-2-one	Non-cis-3-en-1-ol	Octan-2-ol
	2-pentyl-furan	Geosmin	Hexan-1-al	Nonan-1-al	Octan-2-one
	Alpha-yangene	Geranyl-acetone	Hexan-1-ol	Nonan-1-ol	Phenyl-acetaldehyde
	Carvacrol	Hept-4-en-2-ol	Hexan-2-one	Nonan-2-ol	Thymol
	Deca-trans-2-cis-4-dien-1-al	Hept-4-en-2-one	Limonene	Nonan-2-one	Undecan-2-ol
	Deca-trans-2,trans-4-dien-1-al	Heptan-1-ol	Myrcene	Oct-1-en-3-ol	
	Decan-2-ol	Heptan-2-ol	Naphthalene	Octan-1-ol	
	24-methyl-23-dehydro-cholesterol	Alpha-tocopherol	Cyclosadol	Squalene	
	5-dehydro-avenasterol	Beta-sitosterol	Estrone	Stigmasterol	

(continued)

Table 3. (continued)

Component	Chemicals Expected to be Present	Campesterol	Protosan	Trans-24-methyl-23-dehydro-	
Shoot	7-dehydro-avenasterol	Campesterol	Protosan	lphenol	Lophenol
	2-o-p-coumaroyl-hydroxy-citric-acid	6,10,14-trimethyl-pentadecan-2-one	Campesterol	Ethyl-acetate	Obtusifolol
	24-ethylidene-lophenol	Aconitic-acid	Cholesterol	Friedelin	P-coumaric-acid
	24-methylene-cycloartanol	Benzaldehyde	Choline	Furfural	Pectic polysaccharide
	24-methylene-lophenol	Beta-amyrin	Cycloartenol	Hex-cis-3-en-1-ol	Proline
	6-methoxy-2-(3)-benzoxazolinone	Beta-sitosterol	Cycloeucalenol	Hexan-1-al	Stigmasteryl
	6-methoxy-benzoxazolin-2-one	Betaine	Ent-17-hydroxy-kaur-15-en-19-oic acid	Indole	
Silk essential oil	10-oxo-octadec-trans-11-en-13-olide seed 0.3 20426	Caffeic-acid	Ent-kaur-15-en-17-beta-ol	Isovaleraldehyde	Vanillin
	1,2-dimethyl-4-ethyl-benzene	Beta-ionone	Glycolic-acid	Maysin	Phenyl-acetaldehyde
	1,2,3-trimethyl-benzene	Beta-pinene	Gum	Menthol	Phosphorus
	1,2,4-trimethyl-benzene	Beta-sitosterol	Hept-4-en-2-ol	Methyl-phenylacetate	Phytohemagglutinin
	1,3-dimethyl-4-ethyl-benzene	Betaine	Hept-cis-4-en-2-ol	Naphthalene	Potassium
	1,8-cineol	Biphenyl	Hept-trans-2-en-1-al	Naphthalene	Propan-1-ol
	2"-o-alpha-rhamnosyl-6c-	Butan-1-ol	Hepta-trans-2-cis-4-dien-1-al	Niacin	Protein
	(6-deoxo-xylo-hexos-4-ulose)-apigenin	Calcium	Hepta-trans-2-en-1-al	Non-trans-2-en-1-al	Pyrrole
	2"-o-alpha-rhamnosyl-6c-	Carbohydrate	Hepta-trans-2-trans-4-dien-1-al	Nona-trans-2-trans-4-dien-1-al	Resin
	(6-deoxo-xylo-hexos-4-ulose)-chrysoeriol	Carvacrol	Heptan-1-al	Nonal-n-2-ol	Rhamnose
	2"-o-alpha-l-rhamnosyl-6-c-fucosyl-3'-methoxy-luteolin	Chlorogenic-acid	Heptan-1-ol	Nonan-1-al	Riboflavin
	2"-o-alpha-l-rhamnosyl-6-c-fucosyl-luteolin	Chromium	Heptan-2-ol	Nonan-1-ol	Saponin
	2"-o-alpha-l-rhamnosyl-6-c-quinovosyl-luteolin	Cinnamic-acid-ethyl-ester	Heptan-2-ol	Nonan-2-ol	Selenium
	2-ethyl-hex-an-1-ol	Cobalt	Hex-1-en-3-ol	Nonan-2-one	Silicon
	2-methyl-butan-1-al	Daucosterol	Hex-cis-3-en-1-ol	O-diethyl-phthalate	Silk stigma style
	2-methyl-butan-1-ol	Dec-trans-2-cis-4-dien-1-al	Hex-cis-3-en-1-ol	Oct-1-en-3-ol	Sodium
	2-methyl-naphthalene	Dec-trans-2-en-1-al	Hex-trans-2-en-1-al	Oct-1-en-3-ol	Stearic-acid
	2-methyl-pentan-3-one	Dec-trans-2-en-1-al	Hex-trans-2-en-1-ol	Oct-trans-2-en-1-al	Stigmasteryl
	2-methyl-propan-1-ol	Dec-trans-2-trans-4-dien-1-al	Hex-trans-2-trans-4-dien-1-al	Oct-trans-2-en-1-ol	Tartaric-acid
	2-pentyl-furan	Deca-trans-2-cis-4-dien-1-al	Hex-trans-3-en-1-ol	Octa-3-5-dien-2-one	Thiamin
	2-pentyl-furan	Deca-trans-2,trans-4-dien-1-al	Hexa-trans-2-trans-4-dien-1-al	Octa-trans-2-trans-5-dien-2-one	Thymol
	3'-methoxymaysin	Decan-1-al	Hexan-1-al	Octan-1-ol	Tin
	3-o-methyl-maysin	Decan-1-ol	Hexan-1-ol	Octan-1-ol	Undec-trans-2-en-1-al
	3-methyl-butan-1-ol	Decan-2-ol	Hexan-2-ol	Octan-2-ol	Undecan-2-ol
Alkaloids	Alpha-terpineol	Ergosterol	Hordenine	Oleanolic-acid	Vit-k
Aluminum	Aluminum	Ethanol	Iron	Orientin	Vitexin
Apiforol	Ethyl-acetate			Palmitic-acid	Water

(continued)

Table 3. (continued)

Component	Chemicals Expected to be Present				
Sprout seedling	Apigenidin	Ethyl-phenylacetate	Limonene	Pelargonidin	Zea glycoprotein u-cse-100
	Ascorbic-acid	Fat	Limonene	Pent-1-en-2-ol	Zea glycoprotein u-cse-300
	Ash	Fiber	Luteoforol	Pentan-1-ol	
	Benzaldehyde	Fluorene	Luteolinidin	Pentan-2-ol	
	Beta-carotene	Gamma-nonalactone	Magnesium	Pentan-3-one	
	Beta-ionone	Geosmin	Malic-acid	Phenethyl-alcohol	
		Geraniol	Manganese		
	1,3-amino-propyl-pyrrolinium	Delta-amino-levulinic-acid	Feruloylquinic-acid	Obtusifolol	Putrescine
	6-methoxy-benzoxazolin-2-one	Diamino-propane	Glucose	P-coumaric-acid	Pyruvic-acid
	Benzoxazone	Ethyl-amine	Isoamyl-amine	Phosphoenol-pyruvate	Vanillic-acid
Stem	Caffeic-acid	Ferulic-acid			
	Lignin				

species or different sources of the same species are due to isotopic fractionations arising from a variety of biochemical and environmental factors. However, before this technique can be used, it is necessary to determine the fatty acid compositions and $\delta^{13}\text{C}$ values of the major fatty acids of more than 150 vegetable oils in order to provide a database of mixing curves showing how the $\delta^{13}\text{C}$ value of a pure oil is altered as a particular adulterant oil is added.

Methods for characterization of corn starch properties including heavy metals, moisture, protein, etc and Hydrolyzed corn starch properties including pH, hydroxyethyl substitution level, etc have been reported.⁶

Impurities

Corn oil commands a premium price and is therefore a target of adulteration with cheaper vegetable oils. Detection of this activity is challenging because of the natural variability in the fatty acid composition and because of the high sterol and tocopherol contents.¹¹

Less than 0.0001% of barium, chromium, copper, lead, manganese, nickel, vanadium, and zinc has been reported in Corn Cob Meal.⁸

Use

Cosmetic

Corn-derived ingredients are defined in the *International Cosmetic Ingredient Dictionary and Handbook*, which gives the functions for these ingredients described earlier.¹ Under a voluntary reporting program (VCRP) industry provides information to the US Food and Drug Administration (FDA) on the types of products in which individual ingredients are used. An industry survey conducted by the Cosmetic, Toiletry, and Fragrance Association (CTFA), which is now the Personal Care Products Council (Council), collected current use concentration data. These data are summarized in Table 4.

Certain ingredients in this group are reportedly used in a given product category, but the concentration of use is not available. For other ingredients in this group, information regarding use concentration for specific product categories is provided, but the number of such products is not known. In other instances, an ingredient is not in current use, but may be used in the future.

For example, Corn Cob Meal was reportedly used in 1 hair straightener out of the 61 hair-straightener products reported to FDA, but no information on the concentration at which the ingredient is used was available.

If an ingredient is not included in Table 5, there were no reported uses to FDA or CTFA.

Noncosmetic

Food. Corn Fruit is consumed by humans and livestock. The CFR defines corn as grain that consists of 50% or more of whole kernels of shelled dent corn and/or shelled flint corn

(*Zea mays*) and not more than 10% of other grains (7CFR 810.401).

Corn Gluten Protein is used in food with no limitations and is listed as a Generally Recognized As Safe (GRAS) substance (21CFR 184.1)

White and Yellow Corn Meal are defined as food (21CFR 137.250, 21CFR 137.275) although they are not listed as GRAS substances.

The Food and Drug Administration (FDA), as presented in the Code of Federal Regulations (CFR), divides Corn Meal into the following groups: White, Yellow, Self-rising White, Self-rising Yellow, Degerminated White, Degerminated Yellow, Bolted White, and Bolted Yellow (21CFR 137.250, 137.275, 137.270, 137.290, 137.265, 137.285, 137.255, and 137.280).

Corn oil is used in food preparation. It is also used as a defoaming agent in the manufacture of paper and paperboard (21CFR 176.210) and as a resinous and polymeric coating (21CFR 175.300).

Corn seed flour is divided into White and Yellow Corn Flour by the CFR. Both are defined as food (21CFR 137.211, 21CFR 137.215).

Corn silk and corn silk extract are both listed as GRAS direct food substances (21CFR 184-1262).

Medical. In the United States, corn starch may be used as an active ingredient in OTC drug products, known as Topical Starch. It is used as an abrasive, absorbent, skin protectant, and a viscosity increasing agent.

In Canada, corn starch is approved for medicated skin care products in levels of less than or equal to 10%.

Toxicity

Acute Toxicity

The oral toxicity of corn kernel extract was evaluated in rats.¹⁵ Ten OFA Sprague-Dawley rats about 2 months old were used. The rats were not fed 16 hour prior to administration. A single dose of 20 mL/kg was administered via an esophageal probe. One hour after administration, there were no signs of toxicity. The animals were weighed 4, 7, and 14 days later. No signs of toxicity were observed.

Ocular Irritation

Eye irritation of Corn Kernel Extract in rabbits was evaluated.¹⁶ Three New Zealand albino rabbits were used, which were approximately 12-week-old at the beginning of the study and weighed approximately 2 kg. One tenth milliliter of pure corn kernel extract was applied directly into the lower conjunctival sinus of the right eye of each rabbit. Eye examinations were performed 1, 24, 48, and 72 hours after application, using the left eye as a control. Within 24 hours of application, slight redness of the conjunctiva and major watering of the eyes was observed in all of the animals, as well as a very slight epithelial attack of the cornea in a single animal. All of these reactions fully disappeared after 24 hours.

Table 4. Current Cosmetic Product Uses and Concentrations for Corn Ingredients

Product Category (FDA 2008 Totals)	2007 Uses (FDA 2007) ¹²	Use Concentration %(2006 ^a) ¹³
Zea Mays (Corn) Cob Meal		
Noncoloring hair care products		
Straighteners (144)	1	—
Skin care products		
Cleansing creams, lotions, liquids, and pads (1368)	—	3
Face and neck creams, lotions, powders, and sprays (1195)	—	3
Other (1244)	3	—
Total uses/ranges for corn cob meal	4	3
Zea Mays (Corn) cob powder		
Bath products		
Soaps and detergents (1329)	1	2
Skin care products		
Cleansing creams, lotions, liquids, and pads (1368)	2	0.7-10
Other (1244)	2	—
Total uses/ranges for Corn Cob Powder	—	0.7-10
Zea Mays (Corn) Germ Extract		
Skin care products		
Cleansing creams, lotions, liquids, and pads (1368)	2	—
Face and neck creams, lotions, powders, and sprays (1195)	1	—
Body and hand creams, lotions, powders, and sprays (1513)	1	—
Paste masks/mud packs (418)	1	—
Total uses/ranges for corn germ extract	5	—
Zea Mays (Corn) Germ oil		
Baby products		
Lotions, oils, powders, and creams (132)	1	—
Other (138)	1	—
Bath products		
Soaps and detergents (1329)	3	3
Fragrance products		
Perfumes (569)	—	20
Noncoloring hair care products		
Conditioners (1249)	1	0.2
Rinses (47)	1	—
Shampoos (1403)	1	—
Tonics, dressings, etc (1097)	1	—
Makeup		
Other (406)	1	—
Personal hygiene products		
Other (514)	1	—
Skin care products		
Cleansing creams, lotions, liquids, and pads (1368)	6	3
Face and neck creams, lotions, powders, and sprays (1195)	5	7
Body and hand creams, lotions, powders, and sprays (1513)	2	25
Moisturizers (2039)	10	—
Other (1244)	2	—
Suntan products		
Indoor tanning preparations (200)	1	—
Total uses/ranges for Corn Germ Oil	37	0.2-25
Zea Mays (Corn) Oil		
Baby products		
Lotions, oils, powders, and creams (132)	5	0.004
Other (138)	3	—
Bath products		
Soaps and detergents (1329)	5	0.005
Bubble bath (262)	—	0.01
Other (239)	—	0.001
Eye makeup		
Eyebrow pencils (147)	1	—

(continued)

Table 4. (continued)

Product Category (FDA 2008 Totals)	2007 Uses (FDA 2007) ¹²	Use Concentration % (2006 ^a) ¹³
Eyeliners (684)	14	0.2
Eye shadow (1196)	6	0.07-0.1
Eye lotions (177)	8	0.09
Eye makeup remover (131)	4	0.07
Mascara (463)	1	0.0008
Other (288)	5	0.02
Fragrance products		
Colognes and toilet waters (1288)	—	0.001-0.01
Powders (278)	—	0.1
Noncoloring hair care products		
Conditioners (1249)	10	0.0003-0.02
Sprays/aerosol fixatives (371)	1	0.005
Rinses (47)	1	—
Shampoos (1403)	11	0.0001-0.04
Tonics, dressings, etc (1097)	10	0.004
Other (716)	5	0.02
Hair coloring products		
Dyes and colors (2481)	178	0.007
Tints (58)	1	—
Coloring rinses (43)	1	—
Lighteners with color (22)	1	—
Other (166)	1	0.004
Makeup		
Blushers (539)	7	0.01-0.1
Face powders (613)	12	0.02-0.1
Foundations (635)	6	0.0004-0.3
Lipsticks (1912)	29	0.003-10
Makeup bases (164)	1	0.03-0.1
Makeup fixatives (38)	—	0.1
Other (406)	4	0.01-0.2
Nail care products		
Cuticle softeners (18)	—	0.001-5
Creams and lotions (17)	—	0.02
Nail polishes and enamels (419)	—	0.04
Other (124)	1	—
Personal hygiene products		
Underarm deodorants (540)	1	—
Other (514)	2	0.004-0.01
Shaving products		
Aftershave lotions (395)	2	0.02
Mens talcum (7)	—	10
Preshave lotions (27)	—	10
Other (107)	1	10
Skin care products		
Cleansing creams, lotions, liquids, and pads (1368)	17	0.008-14
Depilatories (62)	—	0.1-14
Face and neck creams, lotions, powders (1195)	26	0.002-14 ^b
Body and hand creams, lotions, powders (1513)	31	0.00003-14 ^b
Foot powders and sprays (48)	—	0.0001-14
Moisturizers (2039)	46	0.00003-14 ^b
Night creams, lotion, and powders (343)	5	0.001-14 ^b
Paste masks/mud packs (418)	5	0.1-14
Fresheners (285)	—	0.1-14
Other (1244)	24	0.01-14
Suntan products		
Suntan gels, creams, liquids, and sprays (156)	3	0.003-0.01
Indoor tanning preparations (200)	2	0.002-0.04
Other (62)	1	0.01
Total uses/ranges for Corn Oil	498	0.00003-14

(continued)

Table 4. (continued)

Product Category (FDA 2008 Totals)	2007 Uses (FDA 2007) ¹²	Use Concentration %(2006 ^a) ¹³
Zea Mays (Corn) Oil Unsaponifiables		
Skin care products		
Cleansing creams, lotions, liquids, and pads (1009)	1	—
Body and hand creams, lotions, powders, and sprays (992)	1	—
Moisturizers (1200)	1	—
Night creams, lotions, powders, and sprays ()	1	—
Other (1244)	3	—
Total uses/ranges for Corn Oil Unsaponifiables	7	—
Zea Mays (Corn) Gluten Protein		
Eye makeup		
Eye Shadow (1196)	1	0.1
Makeup		
Blushers (539)	—	0.03
Noncoloring hair care products		
Shampoos (1403)	3	—
Total uses/ranges for Corn Gluten Protein	4	0.03-0.1
Zea Mays (Corn) Kernel Extract		
Bath products		
Other (239)	1	0.00001
Eye Makeup		
Eye Lotions (177)	—	0.01
Makeup		
Face powders 613()	—	0.3
Foundations (635)	1	0.03-0.05
Makeup Bases (164)	—	0.005
Other (406)	1	—
Noncoloring hair care products		
Conditioners (1249)	1	—
Shampoos (1403)	1	—
Shaving products		
Other (107)	2	—
Skin care products		
Cleansing creams, lotions, liquids, and pads (1368)	—	0.0001
Face and neck creams, lotions, and powders (1195)	151	0.01-0.1
Body and hand creams, lotions, and powders (1513)		0.00001-0.5
Moisturizers (2039)	3	0.006-0.01
Night creams, lotions, and powders (343)	3	0.03-0.07
Paste masks/mud packs (418)	1	0.001
Lip cream	—	0.08
Other (1244)	5	0.02-0.1
Suntan products		
Suntan gels, creams, liquids and sprays(156)	—	0.05
Total uses/ranges for corn Kernel extract	35	0.00001-0.5
Hydrolyzed corn starch		
Bath products		
Bubble bath (262)	1	—
Eye makeup		
Mascara (463)	4	1
Lipstick (1912)	—	0.8
Other (288)	1	—
Noncoloring hair care products		
Tonics, dressings, etc (1097)	2	—
Personal hygiene products		
Underarm deodorants (540)	—	0.8
Shaving products		
Aftershave lotions (395)	1	—
Skin care products		

(continued)

Table 4. (continued)

Product Category (FDA 2008 Totals)	2007 Uses (FDA 2007) ¹²	Use Concentration %(2006 ^a) ¹³
Face and neck creams, lotions, and powders (1195)	1	—
Moisturizers (2039)	1	—
Fresheners (285)	1	—
Total uses/ranges for hydrolyzed corn starch	12	—
Hydrolyzed corn protein		
Eye makeup		
Eyeliner (684)	—	0.1
Eye lotion (177)	—	0.004
Mascara (463)	2	0.1
Noncoloring hair care products		
Conditioners (1249)	6	0.4
Sprays/aerosol fixatives (371)	2	—
Shampoos (1403)	5	0.09
Tonics, dressings, etc (1097)	1	0.008
Hair coloring products		
Tints (58)	—	2
Skin care products		
Face and neck creams, lotions, and powders (1195)	—	0.004
Moisturizers (2039)	—	0.01
Other (1244)	1	0.04
Total uses/ranges for hydrolyzed corn protein	17	—
Zea Mays (Corn) seed flour		
Noncoloring hair care products		
Tonics, dressings, etc (1097)	1	—
Skin care products		
Paste masks/mud packs (418)	2	—
Total uses/ranges for Corn Seed Flour	3	—
Zea Mays (Corn) Kernel Meal		
Skin care products		
Cleansing creams, lotions, liquids, and pads (1368)	9	—
Other (1244)	2	—
Total uses/ranges for corn Kernal meal	11	—
Zea Mays (Corn) silk extract		
Makeup		
Face powders (613)	—	0.1
Foundations (635)	—	0.1
Skin care products		
Face and neck creams, lotions, and powders (1195)	—	0.1
Total uses/ranges for corn silk extract	—	0.1
Zea Mays (Corn) starch		
Baby products		
Lotions, oils, powders, and creams (132)	12	98-99
Bath products		
Oils, tablets, salts, etc (257)	3	—
Soaps and detergents (1329)	2	0.01-0.4
Other (239)	2	—
Eye makeup		
Eyeliner (684)	1	3
Eye shadow (1196)	29	1-8
Eye lotion (177)	1	—
Mascara (463)	—	0.1
Other (288)	3	0.2
Fragrances		
Colognes and toilet waters (1288)	—	14
Perfumes (569)	—	14-34
Powders (278)	23	14-80
Sachets (28)	—	14
Other (399)	—	14

(continued)

Table 4. (continued)

Product Category (FDA 2008 Totals)	2007 Uses (FDA 2007) ^{1,2}	Use Concentration %(2006 ^a) ^{1,3}
Noncoloring hair care products		
Conditioners (1249)	3	2
Sprays/aerosol fixatives (371)	—	0.00002
Tonics, dresings, etc (1097)	—	0.00002-0.5
Hair coloring products		
Bleaches (152)	—	6 ^c
Makeup		
Blushers (539)	28	0.005-14
Face powders (613)	43	0.9-27
Foundations (635)	3	0.7-14
Lipsticks (1912)	19	1-5
Makeup bases (164)	—	14
Rouges (99)	—	12
Makeup fixatives (38)	—	14
Other (406)	8	2-15
Oral hygiene products		
Dentifrices (59)	—	25
Personal hygiene products		
Underarm deodorants (540)	6	0.01-2
Douches (12)	—	0.01
Feminine deodorants (21)	5	0.01-25
Other (514)	7	0.01-97
Shaving products		
Aftershave lotions (395)	—	10
Beard softeners (0)	—	10
Preshave lotions (27)	1	—
Shaving creams, gels, etc (162)	1	0.8
Other (107)	2	—
Skin care products		
Cleansing creams, lotions, liquids, and pads (1368)	8	5-20
Depilatories (62)	—	5
Face and neck creams, lotions, powders (1195)	18	8
Body and hand creams, lotions, powders (1513)	9	1-45
Foot powders and sprays (48)	6	25-65
Moisturizers (2039)	21	0.7-2
Night creams, lotion, and powders (343)	2	1
Paste masks/mud packs (418)	6	0.7-5
Other (1244)	5	2-98 ^d
Suntan products		
Indoor tanning preparations (200)	1	5
Total uses/ranges for corn starch	278	0.00002-99

^a No changes in use concentration were reported in 2007 or 2008.

^b The concentration range specifically for sprays in this category was 0.1% to 14%

^c 2% After dilution

^d 98% In a wax epilation product

Dermal Sensitization

Magnusson and Kligman Maximization tests using corn kernel extract were conducted on 30 albino guinea pigs (300–350 g).¹⁷ The scapular area of 15 animals was injected with 0.1 mL 50% Freund adjuvant in 0.9% isotonic NaCl solution, 0.1 mL of corn kernel extract, and a 0.1 mL mixture of equal parts Freund adjuvant and corn kernel extract. Six days later, 1 mL of a 10% lauryl sodium sulfate solution was applied. Twenty-four hours later, 0.5 mL of pure corn kernel extract was applied under a semi-occlusive dressing for 48 hours. The remaining 15 animals

underwent the same procedure, replacing corn kernel extract with physiological serum. All of the animals were allowed to rest for 2 weeks. On all the animals, 0.5 mL of corn kernel extract was then applied under a semi-occlusive dressing for 24 hours on the dorsal regions and 0.5 mL on the rear area of the back. No macroscopic reactions were observed in any animals.

Corn Oil as a Vehicle/Control

Corn oil frequently has been used as a vehicle/control in studies of a wide range of endpoints in which another chemical (or

several other chemicals) was actually the subject of the study. These endpoints include neuron apoptosis behavior, blood pressure, metabolism, body core temperature, enzyme activity, flash evoked potentials hair growth, hormone levels, immune response, seizures, learning ability, pulmonary damage, motor functions, and T cell effects.¹⁸⁻⁶⁶

Other studies addressed acute, short-term, subchronic, and chronic animal toxicity endpoints.⁶⁷⁻⁸²

Other studies particularly addressed hepatotoxicity, nephrotoxicity, neurotoxicity, Clara cell cytotoxicity, and ototoxicity.^{74,77,83-112}

Corn oil also has been used as a delivery vehicle and control in developmental toxicity studies, reproductive toxicity studies, sperm toxicity studies.¹¹³⁻¹⁵⁶

Corn oil has been used as a vehicle in gene expression studies, a DNA adduct study, a mutagenicity study, and antigenotoxicity studies.^{95,157-163}

These studies were carried out using rats, mice, hamsters, guinea pigs, rabbits, ferrets, sheep, pigs, cows, and monkeys. The bulk of the information in these studies concerns the tested chemicals, but, despite the wide range of endpoints studied and animal species used, in none of the studies was corn oil found to have an effect when evaluated as a vehicle/control.

Reproductive and Developmental Toxicity

The reproductive toxicity of corn oil was examined.¹⁶⁴ Seven-week-old female Sprague-Dawley rats were fed either an animal protein-based diet (CA-1) or a plant-based protein diet (CE-2). These animals were mated with a male on the same diet. The rats received 2 or 10 mL corn oil/kg body weight daily by gavage during a 2-week pre-mating period. After mating, pregnant females were weighed and their food consumption reduced on days 0, 7, 14, and 20 of gestation. Dams were weighed and food consumption recorded on days 0 and 4 of lactation. All females were exsanguinated under pentobarbital anesthesia on day 4 of lactation, and the uteri, ovaries, kidneys, livers, and thymuses examined. All pups were euthanized and necropsied on day 4 of lactation.

Dams fed the CA-1 diet and receiving 10 mL/kg corn oil demonstrated soiled fur, hunchback position, piloerection, hypothermia, and decrease in movement. Untreated dams and those on 2 mL/kg corn oil did not show any clinical abnormalities nor did any rats on the CE-2 diet. Both CA-1 and CE-2 rats receiving 10 mL/kg corn oil had significant decreases in food consumption and body weight gain. On day 4 of lactation, there was a significant decrease in the number of pups surviving from the CA-1 dams on 10 mL/kg corn oil. Autopsy of adult females fed the CA-1 diet and 10 mL/kg corn oil revealed some pale and dark-colored kidneys, small thymus, small spleen, erosion in the stomach, and soiled fur. These rats also showed severe epithelial necrosis and fatty degeneration of the proximal tubule.¹⁶⁴ The author suggested 10 mL/kg of corn oil gavage as a vehicle can be a confounding effect in pregnant rats.

Genotoxicity

The effect of corn oil in the diet on mammary gland mutations was examined using female Big Blue rats treated with either 75 mg/kg of 2-amino-1-methyl-6-phenylimidazo[4,5-b]pyridine (PhIP) or 125 mg/kg of 7,12-dimethylbenz[a]anthracene (DMBA).¹⁶⁵ Rats were assigned to either a high-fat (23.5% corn oil) or low-fat (5% corn oil) diet. For each carcinogen, a control group was given the identical volume (5 mL/kg) and dosage regimen of corn oil vehicle. PhIP was administered by oral gavage at 75 mg/kg mixed in corn oil. Ten doses were administered for 5 consecutive days, followed by 2 days without dosing, followed by another 5 days of dosing. DMBA was given as a single gavage dose of 124 mg/kg in corn oil at 50 days of age. The experiment duration was 6 weeks, which was sufficient for the expression of mammary gland mutations. Rats were killed and abdominal mammary glands and liver were examined. The level of corn oil in the diet had no effect.

Carcinogenicity

The link of a high-fat diet to cancer has been studied for mammary; colon; lung; liver; pancreatic; prostate; and intestinal cancer.¹⁶⁶⁻¹⁷² While a link to increased cancer incidence was found for some sites and not others, these studies were not considered relevant to the assessment of corn-derived ingredients in concentrations as used in cosmetics.

NTP studied the use of corn oil, safflower oil, and tricaprillin as gavage vehicles.¹⁷³ Groups of 50 male F344/N rats were administered 2.5, 5, or 10 mL/kg of each tested vehicle, or 10 mL/kg saline, by gavage for 5 d/week for 2y. Saline-treated animals served as controls. Corn oil, safflower oil, and tricaprillin each caused hyperplasia and adenoma of the exocrine pancreas, decreased incidences of mononuclear cell leukemia (MNCL), and reduced incidences/severity of nephropathy. The authors concluded that the use of corn oil as a gavage vehicle may have a confounding effect on the interpretation chemical induced proliferative lesions of the exocrine pancreas and MNCL.

Clinical Assessment of Safety

Dermal Sensitization/Irritation

Corn kernel extract. The contact-sensitization potential of a face cream containing 1% of a 3% corn kernel extract was evaluated in human skin by maximization assay on 27 adult volunteers (25 females, 2 males; ages 19-57).¹⁷⁴ Following pretreatment by 0.05 mL sodium laurel sulfate (SLS; 0.25%) for 24 hours, 0.05 mL of the test material was applied and occluded for 48 to 72 hours in 5 separate induction exposures. After a 10-day rest period, SLS pretreatment occurred, followed by challenge with 0.05mL of the test material.

One female participant dropped out of the study for unknown reasons. There were no instances of allergic reaction or sensitization 48 or 72 hours after application of challenge patches.

The skin tolerance of corn kernel extract, diluted at 10%, was tested on 21 participants (15 females, 6 males) using a single patch test.¹⁷⁵ The patch test utilized Finn Chambers on Scanpor tape with an area of 50 mm² and a 20 µL capacity. These were applied to the scapular area of the volunteers and covered with an adhesive; control patch tests with no product were conducted concurrently. The patches were removed 48 hours after application and evaluated for irritation after 30 minutes. Light erythema was observed in 1 male volunteer. There was no reaction in any other participants.

Corn starch. A repeated insult patch test (RIPT) was done on 99 participants (26 male 73 female, ages 18-70) using feminine powder containing 97% corn starch.¹⁷⁶ A patch was applied for 24 hours, after which it was removed. The same area was then repatched either 24 or 48 hours after the removal. This was repeated until 9 induction patchings were completed. Approximately 2 weeks after induction patching, a challenge patch was placed on a new site and then removed after 24 hours. The induction and challenge sites were observed at 48, 72, and 96 hours after the removal of the challenge patch. Four cases of faint, minimal erythema were observed during the induction phase, and there was no observed irritation during the challenge phase. This same methodology was repeated with 109 participants (35 male 74 female, age 18-68) completing the study. There was no observed irritation in the participants.

Corn gluten protein. A RIPT was done on 51 volunteers (43 female, 8 male) of phototypes I to IV using corn gluten protein.¹⁷⁷ A mixture of mineral oil (95%) and corn gluten protein (5%) was prepared and 0.05 g/cm² was applied to a patch. The patch was then applied to the right or left side of the backs of the participants and covered with semi-occlusive hypoallergenic tape. The patch was removed 48 hours later and reactions recorded after 30 minutes and 24 hours. Applications were performed daily to the same site over the course of 3 weeks, after the first application had been in place for 48 or 72 hours. After 14 consecutive applications, none were applied for a period of 2 weeks. Afterward, a single patch with the sample was applied to a virgin area. After 48 hours, the patch was removed and reactions recorded after 30 minutes and 24 hours. No adverse reactions were detected at the test sites for either primary or cumulative irritation and/or sensitization.

A study was done in which 94 participants (85 female, 9 male) completed an allergic contact sensitization test to eye shadow containing 12.6% of 1% corn gluten protein.¹⁷⁸ Ten patches were applied to the upper backs of the participants after 0.1 g of the test material was added. The patches were applied for 24 hours, 3 days/week for 3 wks to the same sites. During week 6, a single patch of the material was applied to a virgin site for 24 hours and then scored 24 and 48 hours after removal. There was no evidence of any effect from the patches.

Using the same method as above, an eye shadow containing 10.0% of 1% corn gluten protein was tested for allergic contact sensitization in 85 participants (82 female, 3 male).¹⁷⁹ Three participants exhibited minimal faint uniform or spotty

erythema and 1 exhibited bright red erythema with accompanying edema, petechiae, or papules. There were no other reported effects.

Corn silk extract. A RIPT was done on 56 participants (ages 20-79; 44 female, 12 male) using a powder containing 0.1% corn silk extract. Five participants did not complete the study for reasons unrelated to the test materials.¹⁸⁰ Approximately 0.2 g of the test material was applied as an occluded patch and applied to the upper back between the scapulae. Patches were applied 3 times a week for a total of 9 applications lasting 24 hours each with 24 or 48 hours between them. If any site exhibited a moderate reaction during this phase, the application was moved to an adjacent area. If the new site exhibited a moderate reaction, the applications were discontinued. Two weeks after the initial application, a challenge patch was applied to a site adjacent to the induction patch site. After the patch was removed, the site was scored 24 and 72 hours after removal. None of the participants exhibited any visible skin reaction.

An RIPT was done on 112 participants (ages 19-79; 83 female, 29 male) using a powder containing 0.1% corn silk extract.¹⁸¹ Six participants did not complete the study for reasons unrelated to the test materials. The same procedure as above was used. There were no visible skin reactions among the participants.

An RIPT was done on 57 participants (ages 16-79; 46 female, 16 male) using a powder containing 0.1% Corn Silk Extract.¹⁸² Six subjects did not complete the study for reasons unrelated to the test materials. The same procedure as above was used. There were no visible skin reactions among the subjects.

Twenty subjects (ages 21-45; 17 female, 3 male) with an active case or a history of acne vulgaris to a human comedogenicity test were tested with a powder composite containing 0.1% corn silk extract.¹⁸³ Sites on the upper back were wiped with 70% isopropyl alcohol, and approximately 0.2 g of the test material was applied onto an occlusive patch and affixed to the skin. An untreated patch was also applied as a negative control. Three days a week for 4 consecutive weeks, patches were removed and the sites evaluated for irritation. Fresh patches were applied afterward. At the final session, sites were cleansed with 70% isopropyl alcohol and patted dry. Follicular biopsies were taken from these sites and the specimens were evaluated by the ratio of microcomedones to follicles per visual field. There were no cases of dermal irritation. The follicular biopsy evaluation resulted in a microcomedones: follicle ratio of 0 for 18 participants, 0.6 in 1 participant, and 0.7 in 1 participant.

An RIPT was done on 55 participants (ages 22-69; 44 female, 11 male) using a powder containing 0.1% corn silk extract.¹⁸⁴ Two participants did not complete the study for reasons unrelated to the test materials. Before application to the upper back between the scapulae, the area was wiped with 70% isopropyl alcohol and allowed to dry. Once applied, a semi-occlusive patch with test material was in direct contact with the skin for 24 hours. Patches were applied 3 times a week for a total of 9 applications and evaluated 24 or 48 hours after

removal. After a 2-week rest period, challenge patches were applied to untreated sites on the back and left for 24 hours. The sites were evaluated at removal and reevaluated at 48 and 72 hours. Four participants exhibited a barely perceptible erythema at the first induction. There were no other visible skin reactions.

An RIPTS was done on 55 participants (ages 22-69; 44 female, 11 male) using a powder containing 0.1% corn silk extract.¹⁸⁵ Two participants did not complete the study for reasons unrelated to the test materials. The RIPT was performed as above. Three participants exhibited a barely perceptible erythema at the first induction. There were no other visible skin reactions.

Case Reports

Corn starch. Several reports of hypersensitivity to cornstarch used as a powder in medical gloves were reported.¹⁸⁶⁻¹⁹¹ One case report of a granuloma following laparoscopic surgery in which powered gloves were used.¹⁸⁹ Silver et al reported an instance of respiratory failure in an infant who had aspirated corn starch during a diaper change.¹⁹²

Summary

Corn Cob Meal is the milled powder prepared from the cobs of *Zea mays*. In cosmetics, it functions as an abrasive and bulking agent and is used at 3%.

Corn cob powder is the powder obtained from the dried ground cobs of *Zea mays*. It functions as an abrasive and is used at 0.7% to 10%.

Corn fruit is the fruit of *Zea mays*. This ingredient is currently not in use.

Corn germ extract is an extract of the germ of the corn, *Zea mays*. It is currently reported to be used, but at an unknown concentration.

Corn germ oil is the oil obtained from the germ of *Zea mays*. It functions as an occlusive skin conditioning agent and is used at 25%.

Corn gluten protein is a protein obtained from the starchy endosperm of corn, *Zea mays*. It functions as a bulking agent, hair conditioning agent, and skin conditioning agent and is used at up to 0.1%.

Corn kernel extract is an extract of the kernels of corn, *Zea mays*. It is used at 0.5%.

Corn kernel meal is the coarse flour prepared by milling the kernels of *Zea mays*. It is used as an abrasive and a bulking agent used at 3%.

Corn oil is the refined fixed oil obtained from the wet milling of corn *Zea mays*. It is a mixture of the following fatty acids: myristic, palmitic, stearic, hexadecenoic, oleic, and linoleic. Corn oil functions as a fragrance ingredient, hair conditioning agent, occlusive skin conditioning agent, surfactant, and emulsifying agent and is used at concentrations up to 14%.

Corn oil unsaponifiables is the fraction of corn oil (qv) which is not saponified in the refining recovery of corn oil fatty

acids. It functions as a hair conditioning agent, and skin conditioning agent. Although reportedly in current use, the concentration is unknown.

Corn seed flour is the powder prepared from the fine grinding of the inner portion of the seeds of *Zea mays*. It is used as an abrasive, absorbent, binder, and bulking agent. Although reportedly in current use, the concentration is unknown.

Corn silk extract is an extract of the stigmas of the corn, *Zea mays* used at 0.1%.

Corn starch is a starch obtained from corn, *Zea mays*. Corn starch is composed of amylose and amylopectin. It functions as an abrasive, absorbent, skin protectant, and a viscosity increasing agent and is used at concentrations up to 99%.

Hydrolyzed corn starch is the hydrolysate of corn starch (qv) derived by acid, enzyme, or other method of hydrolysis. It functions as a skin conditioning agent and is used at concentrations of 0.8% to 1%.

Hydrolyzed corn protein is hydrolysate of corn protein derived by acid, enzyme, or other method of hydrolysis. It functions as a hair conditioning agent and skin conditioning agent and is used at concentrations up to 2%.

Corn kernel extract was tested for ocular irritation, oral toxicity, and dermal sensitization. Slight eye irritation in rabbits occurred within a day of application, but all symptoms disappeared after 24 hours. Maximization tests performed on guinea pigs did not produce any observed dermal irritation. Oral toxicity in rats was tested, producing no signs of toxicity. One clinical test of corn kernel extract as a skin irritant produced light erythema in a single volunteer (n = 21), but no other signs of irritation. Corn kernel extract was also negative in 27 individuals in a maximization test.

There were no safety test data available for corn acid, potassium cornate, or corn glycerides.

Corn oil is commonly used as a vehicle in studies of lipid-soluble substances. These studies were carried out using rats, mice, hamsters, guinea pigs, rabbits, ferrets, sheep, pigs, cows, and monkeys. The bulk of the information in these studies concerns the tested chemicals, but, despite the wide range of endpoints studied and animal species used, in none of the studies was corn oil found to have an effect when evaluated as a vehicle/control.

When rats were fed different types of diets, those fed hydrolyzed corn starch had greater carbohydrate intake, total food intake, and body weight gain compared to those fed amylopectin. The same was true with the gel forms of the different carbohydrates as diets. The life spans of rats fed high-phytosterol oils, including corn oil, were significantly shorter than those fed a fat blend or soybean oil. Carcinogenesis studies in which the effect of corn oil in high-fat diets was examined and found to increase the likelihood of several, but not all, cancer types. These dietary studies, however, were not considered relevant to assessing the safety of corn oil at concentrations used in cosmetics.

There was 1 case report of respiratory distress in an infant that inhaled corn starch during a diaper change. There are several cases on granulomas and urticaria caused by the corn starch on latex gloves. Cases were usually of patients being treated by gloved personnel or hospital personnel themselves developing reactions. Eye shadow formulations tested in repeat insult patch tests containing corn gluten protein produced irritation in 3 of 85 participants, but no allergic reactions.

Discussion

The CIR Expert Panel found that the information provided is sufficient to identify the following ingredients as safe as used: Zea Mays (Corn) oil; Zea Mays (Corn) cob meal, Zea Mays (Corn) cob powder, Zea Mays (Corn) fruit, Zea Mays (Corn) germ extract, Zea Mays (Corn) germ oil, Zea Mays (Corn) gluten protein, Zea Mays (Corn) kernel extract, Zea Mays (Corn) kernel meal, Zea Mays (Corn) oil unsaponifiables, Zea Mays (Corn) seed flour, Zea Mays (Corn) silk extract, Zea Mays (Corn) starch; corn acid, corn glycerides, hydrolyzed corn protein, hydrolyzed corn starch, and potassium cornate.

Although there are not safety test data available on all of these ingredients, the Panel has considered that the chemical characterization of each ingredient for which data are available is sufficient to apply those available data to ingredients with similar composition.

Also relevant is the long history of use of corn oil as a vehicle/control in studies of other chemicals. While it is the intent that a vehicle will itself have no effect, the Panel reviewed studies that addressed a large number of different endpoints that were unaffected by corn oil as a vehicle. These included behavioral studies; acute, short-term, subchronic, and chronic toxicity studies; reproductive and developmental toxicity studies; and genotoxicity studies.

There were numerous carcinogenicity studies in which the effect of corn oil in high-fat diets was examined, but these data were not considered relevant to assessing the safety of these ingredients at concentrations used in cosmetics.

The CIR Expert Panel recognizes that there are data gaps regarding use and concentration of these ingredients. However, the overall information available on the types of products in which these ingredients are used and at what concentrations indicate a pattern of use, which was considered by the Expert Panel in assessing safety.

The Expert Panel expressed concern regarding pesticide residues and heavy metals that may be present in botanical ingredients. They stressed that the cosmetics industry should continue to use the necessary procedures to limit these impurities in the ingredient before blending into cosmetic formulation.

Concern was expressed about alternative approaches to extraction that might not produce material with the same safety profile described in this safety assessment, especially if pesticides were used on the plants. While extracts from pesticide-free plants were not genotoxic and there do not appear to be any

components that could be carcinogenic, pesticide residues could raise this issue. The Panel urged that manufacturers limit pesticide residues to the limit previously used for lanolin of not more than 40 ppm (with not more than 10 ppm for any 1 residue). Extracts not prepared in a manner that produces a similar chemical profile would be considered safe if they have a similar safety test profile.

Conclusion

The CIR Expert Panel concludes that the following ingredients are safe as used as described in this report: Zea Mays (corn) oil; and Zea Mays (corn) cob meal, Zea Mays (corn) cob powder, Zea Mays (Corn) fruit, Zea Mays (corn) germ extract, Zea Mays (corn) germ oil, Zea Mays (corn) gluten protein, Zea Mays (corn) kernel extract, Zea Mays (corn) kernel meal, Zea Mays (corn) oil unsaponifiables, Zea Mays (corn) seed flour, Zea Mays (corn) silk extract, and Zea Mays (corn) starch; and corn acid, corn glycerides, hydrolyzed corn protein, hydrolyzed corn starch, and potassium cornate. 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 the group.

Author's Note

Unpublished sources cited in this report are available from the Director, Cosmetic Ingredient Review, 1101 17th St., Suite 412, Washington, DC 20036, USA.

Acknowledgment

The Cosmetic Ingredient Review Program is financially supported by the Personal Care Products Council. No potential conflict of interest relevant to this article was reported. F. Alan Andersen, PhD, is employed by Cosmetic Ingredient Review.

Declaration of Conflicting Interests

The author(s) declared no conflicts of interest with respect to the authorship and/or publication of this article.

Funding

The author(s) disclosed receipt of the following financial support for the research and/or authorship of this article: The articles in this supplement were sponsored by the Cosmetic Ingredient Review.

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Memorandum

TO: F. Alan Andersen, Ph.D.
Director - COSMETIC INGREDIENT REVIEW (CIR)

FROM: Halyna Breslawec, Ph.D.
Industry Liaison to the CIR Expert Panel *HBreslawec*

DATE: July 18, 2012

SUBJECT: Comments on the Scientific Literature Review: Hydrolyzed Proteins as Used in Cosmetics

Key Issue

If available, some indication of the molecular weight of the peptides used in each study should be stated.

Additional Comments

The Council does not have any suppliers listed for the following ingredients included in this Scientific Literature Review:

Hydrolyzed Avocado Protein

Hydrolyzed Extensin

Hydrolyzed Fibronectin

Hydrolyzed Reticulin

Hydrolyzed Sesame Protein

Hydrolyzed Spinal Protein

Hydrolyzed Hemp Seed Protein (the supplier indicates that this product has been discontinued)

Hydrolyzed Zein

Sodium Hydrolyzed Casein

Somewhere in this report, please note the sources for which FDA requires allergen labeling when they are included in food. Information about FDA food allergen labeling requirements can be found at:

<http://www.fda.gov/Food/GuidanceComplianceRegulatoryInformation/GuidanceDocuments/FoodLabelingNutrition/ucm059116.htm>

p.2 - If available, please indicate the use for the Hydrolyzed Serum Protein described in reference 15.

p.3 - The study describing the biologically active proteins from soy (reference 16) is not appropriate for the Toxicokinetics section.

p.12-15, Table 3a - NR (not reported) in the concentration of use column is not appropriate as the concentration of use survey for these ingredients is not yet complete.